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Perspective

Bruxism management during tooth wear rehabilitation: A perspective on the temporary use of contingent electrical stimulation

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Keywords: bruxism, tooth wear, electrical stimulation, resin composites

The temporary use of contingent electrical stimulation with the GrindCare® device may offer a practical, non-invasive solution to reduce bruxism and protect restorations during vulnerable phases of phased dental rehabilitation.

Bruxism, whether manifesting during wakefulness or sleep, is a commonly encountered phenomenon in clinical dental practice. It continues to stimulate discussion and research due to its multifactorial nature and diverse clinical implications. Notably, our current understanding suggests that bruxism may not always be a harmful behavior; instead, it can serve as either a risk and/or a protective factor, depending on the individual case and context.[1] This dual nature presents a unique challenge for clinicians tasked with discerning when, and how, to intervene.

Bruxism diagnostic tools are currently categorized into 3 diagnostic levels.[1,2] They are termed subject-based when they rely solely on patient self-reporting; clinically based when accompanied by clinical signs, such as the linea alba, impressions on the tongue or cheek, or tooth wear; and device-based when confirmed via electromyographic (EMG) or polysomnographic (PSG) recordings. While these definitions aid in structuring clinical evaluation, they do not always clarify whether bruxism is active or residual, further complicating treatment decisions.

Importantly, bruxism is no longer regarded as a disorder in itself. Instead, it is understood as a behavior that warrants treatment only when it results in negative outcomes (e.g., tooth wear, pain). One promising approach in such cases is contingent electrical stimulation (CES), where muscle activity is detected and interrupted via mild electrical impulses.[2] One of CES tools is Butler® GrindCare® (Sunstar Suisse SA, Etoy, Switzerland), a single-channel EMG-based device that exemplifies this approach.[2] It records activity from the temporalis muscle and, when in the therapeutic mode, responds to bruxism events with mild electrical stimulation intended to disrupt the event without causing discomfort. The device has utility for both diagnosis – by establishing a baseline level of activity – and therapy, by potentially reducing excessive muscle contractions during sleep. Following a standard protocol, patients wear the device for a two-week assessment period. If more than 15 episodes per hour are recorded, treatment is recommended.[2,3]

As one of the possible negative outcomes of bruxism, tooth wear is a multifactorial process in itself, and while bruxism plays a central role, other contributors must be acknowledged. Among these are opposing restorations, especially when constructed from hard materials, such as ceramics or metals. These can exacerbate wear patterns and pose additional risks during restorative treatment.

Tooth rehabilitation strategies are typically categorized into direct or indirect methods.[4] Direct composite restorations offer a conservative approach. They are minimally invasive, easier to repair, and generally exhibit mechanical properties, such as elasticity and wear resistance, more closely aligned with natural dentin.[5] On the other hand, they may be prone to discoloration over time, and because of placing multiple restorations over several appointments due to time constraints, patients might experience an open bite. In some cases, a Dahl plateau is created to establish a stable raised bite following the first appointment. However, this may not always be feasible due to the existing prosthodontic restorations or periodontal considerations. Between the 1st and 2nd session, there is an elevated risk of mechanical overloading with limited options for protecting the teeth during this time interval.

In this perspective, we illustrate the above notions with a case where the GrindCare was utilized temporarily during a dental rehabilitation procedure to reduce the risks posed by bruxism during a transitional, high-risk period between appointments.

A 67-year-old male patient was referred to our tertiary university clinic for the evaluation of severely worn lower anterior teeth. His primary concerns were both functional (lip biting during eating) and esthetic (dissatisfaction with the shortened appearance of the teeth) (Fig. 1A). His general medical and psychosocial histories were non-remarkable. Snoring, alcohol, caffeine and acidic beverage intake, obstructive sleep apnea (OSA), gastroesophageal reflux disease (GERD), eating disorders, medication use, depression, anxiety, stress, and dry mouth were assessed via structured interviews and validated self-report instruments, including Epworth Sleepiness Scale (ESS), GERD Questionnaire (GerdQ), eating habits questionnaires (Eating disorder Screen for Primary care (ESP) and the Sick-Control-One stone-Fat-Food (SCOFF)), Diagnostic Criteria for Temporomandibular Disorders (DC/TMD), Perceived Stress Scale 4 (PSS-4), Trauma Screening Questionnaire (TSQ), and Xerostomia Inventory (XI). All scores were zero except XI (score: 1). Sleep bruxism was assessed using the GrindCare and DC/TMD questionnaires, focusing on the past month. From a dietary standpoint, acidic intake was modest, with the patient consuming one piece of fruit daily and a small glass of fruit juice each morning.

Intraoral examination revealed healthy periodontal tissues with no probing depths exceeding 3 mm, and intact oral mucosa without signs of clenching. Overjet was 1 mm, overbite was 3 mm, and the patient had Angle class 1. Yet, there were visible bruxopositions (i.e., visible matching wear facets during laterotrusion and protrusion) on incisors and canines. There was a significant number of existing prosthetic restorations present, and their materials and occlusal design were contributing factors in the observed wear. Using the Tooth Wear Evaluation System (TWES),[6] we documented localized, extreme tooth wear in the lower anterior region, of a mechanical nature, influenced by the opposing ceramic restorations.

Cold sensitivity testing revealed delayed but positive pulp responses in lower incisors, while radiographic examination showed no underlying pathology. Based on these findings, bruxism was classified as clinically based wake or sleep bruxism, although clinical signs alone were insufficient to determine whether bruxism was ongoing.

To further investigate whether this process was active, we initiated a two-week assessment period, using the GrindCare.[2] The collected data revealed 246–582 bruxism episodes per night, translating to an hourly rate of 3–147 episodes – well above the clinical threshold of 18 events per hour.[2,7] This confirmed the presence of active sleep bruxism.

Our treatment strategy unfolded in 2 phases:

Phase 1 involved restoring teeth 15, 14, 11, 23, and 24, using direct composite restorations to raise the vertical dimension and create space for lower anterior reconstruction. We intentionally avoided pre-treatment mock-ups, wax-ups or guides, relying instead on direct intraoral assessment and real-time adaptation. The materials used included AP-X A3 (Kuraray Noritake Dental, Tokyo, Japan) for occlusal and palatal surfaces, and the Clearfil™ Photo Bright composite in shade UO (Kuraray Noritake Dental) for a veneer on tooth 11. Adhesion followed a standardized protocol with Clearfil Photo Bond (Kuraray Noritake Dental), 37% orthophosphoric acid etching and Clearfil SA Primer (Kuraray Noritake Dental).

Following the 1st session, the patient began using the GrindCare in the therapeutic mode, configured to deliver CES during episodes of increased temporalis activity. The device offers 10 intensity settings. Patients are guided to start at the lowest level, gradually increasing every night until the stimulation wakes them up, and then stepping down by one level. During this period, bruxism episodes dropped to 54–240 per night, indicating a substantial initial reduction.

Phase 2 addressed the lower anterior segment. The composite was applied to the lingual surfaces, using AP-X A3, and veneers were layered with Clearfil Photo Bright in multiple opacities (UO, YO, LO). The bonding protocol matched the one used in the upper arch. The posterior occlusion remained open, relying on passive eruption per the Dahl principle to close vertical gaps over time (Fig.1B,C).[8] During this phase, the patient continued using the GrindCare nightly, aiming to maintain reduced muscle activity.

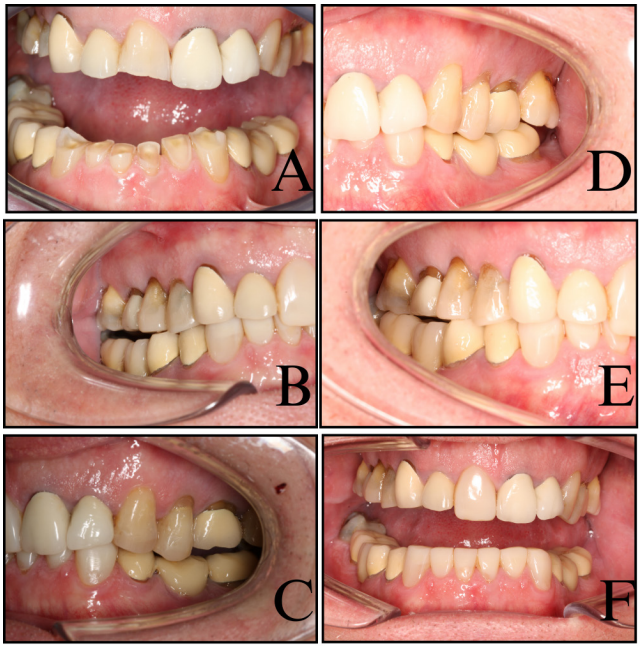

Fig. 1. A – initial situation; B, C – posterior space directly after the treatment; D, E – posterior space 24 months after the treatment; F – final restorations after 24 months

Over the next 12 months, follow-up assessment was performed at 3, 6 and 12 months. At 3 months, CES was discontinued, as the patient reported diminished responsiveness, and the bruxism index (BI) returned to baseline by week 8 (166–462 episodes/night). The posterior occlusion continued to re-establish, and the patient reported no functional limitations or eating restrictions.

At the 24-month review, molar contacts were restored through natural eruption, and all restorations remained intact. No fractures, debonding or chipping were noted. However, the crown on tooth 21 had begun to cause accelerated wear on the opposing restoration at tooth 32, warranting future monitoring (Fig. 1D–F).

This case highlights the pragmatic utility of the GrindCare during the transitional phases of tooth rehabilitation. When conventional occlusal splints are impractical – during occlusal instability or open-bite stages – devices like the GrindCare offer a non-invasive, patient-directed alternative for short-term bruxism management. While the mechanism behind CES remains only partially understood, its ability to temporarily modulate muscle activity is noteworthy. However, the observed decrease in muscle activity during GrindCare use between sessions may be attributed not only to the biofeedback mechanism of the device, but also to the transient occlusal changes induced by the interim composite restorations, which potentially altered proprioceptive input and reduced parafunctional triggers.[9]

Yet, our experience also underscores the temporary nature of CES efficacy. As in this case, initial reduction may not persist beyond 2 months, raising important questions regarding long-term viability. Further research is needed to explore adaptive mechanisms, such as sensory accommodation or neuroplasticity, that may limit prolonged effectiveness.

In terms of restorative choice, our decision to pursue direct composite restorations aligns with growing evidence that such materials offer repairability, biomimetic elasticity and acceptable esthetics in bruxism patients.[4] While ceramic options may provide superior esthetics, they pose a greater risk of total failure and contribute more aggressively to antagonist wear, especially in cases with the pre-existing ceramic restorations.

Ultimately, our experience affirms that the GrindCare can serve as a useful adjunct, particularly during vulnerable phases of treatment. However, randomized controlled trials are essential to validate its long-term role and clarify its mechanism of action. Until then, CES should be viewed as a complementary, rather than primary, modality in managing bruxism during complex restorative cases.

Patient consent

Written informed consent was obtained prior to the submission of this perspective. The patient authorized the use of clinical history, data and imagery for academic publication, under the condition that identifying features remain obscured.

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References

1. Verhoeff MC, Lobbezoo F, Ahlberg J, et al. Updating the bruxism definitions: Report of an international consensus meeting. *J Oral Rehabil*. 2025. doi:10.1111/joor.13985
2. Lobbezoo F, Aarab G, Ahlers MO, et al. Consensus-based clinical guidelines for ambulatory electromyography and contingent electrical stimulation in sleep bruxism. *J Oral Rehabil*. 2020;47(2):164–169. doi:10.1111/joor.12876
3. Conti PC, Stuginski-Barbosa J, Bonjardim LR, Soares S, Svensson P. Contingent electrical stimulation inhibits jaw muscle activity during sleep but not pain intensity or masticatory muscle pressure pain threshold in self-reported bruxers: A pilot study. *Oral Surg Oral Med Oral Pathol Oral Radiol*. 2014;117(1):45–52. doi:10.1016/j.oooo.2013.08.015
4. Shah Y, Shiraguppi V, Deosarkar BA, Shelke UR. Long-term survival and reasons for failure in direct anterior composite restorations: A systematic review. *J Conserv Dent*. 2021;24(5):415–420. doi:10.4103/jcd.jcd_527_21
5. Habib SR, Alotaibi A, Al Hazza N, Allam Y, AlGhazi M. Two-body wear behavior of human enamel versus monolithic zirconia, lithium disilicate, ceramometal and composite resin. *J Adv Prosthodont*. 2019;11(1):23–31. doi:10.4047/jap.2019.11.1.23
6. Wetselaar P, Wetselaar-Glas MJ, Katzer LD, Ahlers MO. Diagnosing tooth wear, a new taxonomy based on the revised version of the Tooth Wear Evaluation System (TWES 2.0). *J Oral Rehabil*. 2020;47(6):703–712. doi:10.1111/joor.12972
7. Stuginski-Barbosa J, Porporatti AL, Costa YM, Svensson P, Rodrigues Conti PC. Diagnostic validity of the use of a portable single-channel electromyography device for sleep bruxism. *Sleep Breath*. 2016;20(2):695–702. doi:10.1007/s11325-015-1283-y

8. Kim SJ, Kim KH, Yu HS, Baik HS. Dentoalveolar compensation according to skeletal discrepancy and overjet in skeletal Class III patients. *Am J Orthod Dentofacial Orthop*. 2014;145(3):317–324. doi:10.1016/j.ajodo.2013.11.014
9. Avivi-Arber L, Lee JC, Sessle BJ. Dental occlusal changes induce motor cortex neuroplasticity. *J Dent Res*. 2015;94(12):1757–1764. doi:10.1177/0022034515602478

Temporomandibular disorders: INfORM/IADR key points for good clinical practice based on standard of care: The Polish language version

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Keywords: recommendations, pain therapy, temporomandibular disorders, orofacial pain, good clinical practice

The article presents recommendations from the International Network for Orofacial Pain and Related Disorders Methodology (INfORM) for the management of temporomandibular disorders (TMD), following the article "Temporomandibular disorders: INfORM/IADR key points for good clinical practice based on standard of care" by Manfredini et al. The document was translated by Aleksandra Nitecka-Buchta, Karolina Walczyńska-Dragon and Justyna Sędkiewicz, and reviewed by Aleksandra Nitecka-Buchta, Stefan Baron, Małgorzata Pihut, Jolanta Kostrzewa-Janicka, Edward Kijak, Mieszko Więckiewicz, Magdalena Osiewicz, Małgorzata Gałczyńska-Rusin, Aneta Wieczorek, and Daniele Manfredini.

The Polish Association for Temporomandibular Disorders (PATMD) as the only organization in Poland focused on temporomandibular disorders (TMD), which has been involved in promoting knowledge and good clinical practice in the treatment of patients with TMD for 25 years already, has prepared the Polish language version of the 10 key points for good clinical practice based on standard of care.[1] The official position of the International Network for Orofacial Pain and Related Disorders Methodology (INfORM) was prepared during the March 2024 meeting in New Orleans, LA, USA, hosted by the International Association for Dental Research (IADR). The main elements the recommendations are based on are the etiology, diagnosis and therapy of TMD, as per the latest data obtained from evidence-based medicine (EBM). The present article summarizes and indicates the promising directions in the modern approach to TMD. As a scientific society, PATMD sees it as its responsibility to introduce modern ideas to the Polish medical community, where doctors,

in a conscious manner, strive to develop and follow current trends, thereby providing the highest level of medical care. The translation we are presenting has been prepared by a group of Polish TMD specialists who have been co-creating PATMD for many years. We have attempted to tailor this document to the cultural, social and healthcare system aspects of Poland.

Temporomandibular disorders have posed a challenge to clinicians worldwide for many years. The biopsychosocial factors, including the complex anatomy and physiology of the head and neck region, can contribute to the development of TMD, which is sometimes attributed to occlusion and the temporomandibular joint (TMJ) structures. These complex issues and the controversies surrounding their solutions have led to the emergence of numerous theories, not necessarily based on EBM. Currently, it is known that the biopsychosocial model provides the most probable explanation for the development of TMD. This publication compiles the most up-to-date information on diagnostic tools, including interviews, clinical examinations, psychosocial assessments, and imaging diagnostics, as well as the management of TMD patients, encompassing dental, orthopedic, neurological, psychological, and physical therapy approaches. The following key points also refer to the aspects of treatment, involving the use of neurological drugs and occlusal splints, and surgical interventions, which are reserved for selected cases. The PATMD translated and presented the current global concepts with regard to the treatment of TMD, which can prevent patients from inappropriate therapy, pain chronification and the iatrogenic effects of TMD management.

The list of the 10 key points of good clinical practice in the management of TMD, as stated in the current standards of treatment and patient needs, is presented in Fig. 1.

Knowledge about TMD etiology and diagnostics is essential for dental clinicians and physicians. The standards of good clinical practice in TMD management proposed by the INfORM group are necessary to complement basic dental and medical education. The 10 key points for good clinical practice will enable the establishment of a protocol for therapy and help prevent inappropriate treatment. They can also serve as recommendations for the future development of TMD diagnostics and treatment guidelines.

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References

1. Manfredini D, Häggman-Henrikson B, Al Jaghsi A, et al.; International Network for Orofacial Pain and Related Disorders Methodology. Temporomandibular disorders: INfORM/IADR key points for good clinical practice based on standard of care. *Cranio*. 2025;43(1):1–5. doi:10.1080/08869634.2024.2405298

TEMPOROMANDIBULAR DISORDERS: INfORM/IADR KEY POINTS FOR GOOD CLINICAL PRACTICE BASED ON STANDARD OF CARE: THE POLISH LANGUAGE TRANSLATION

No.	ENGLISH	POLISH
1	Patient-centered decision-making alongside patient engagement and perspective is critical to manage TMDs, with management being the process from history through examination into diagnosis and then treatment. Expectations should focus on learning to control and manage the symptoms and decrease their impact on the individual's everyday life.	Podejmowanie decyzji powinno być zorientowane na pacjenta, a jego zaangażowanie i perspektywa są kluczowe w leczeniu zaburzeń skroniowo-żuchwowych (ZSŻ); jest to proces trwający od zbierania wywiadu, przez badanie, po diagnozę, a następnie leczenie. Oczekiwania powinny koncentrować się na nauce kontroli i radzenia sobie z objawami ZSŻ oraz zmniejszeniem ich wpływu na codzienne życie chorego.
2	TMDs are a group of conditions that may cause signs and symptoms, such as orofacial pain and dysfunction of a musculoskeletal origin.	ZSŻ są grupą zaburzeń, mogącą wywoływać symptomy i objawy takie jak ból ustno-twarzowy czy dysfunkcje pochodzenia mięśniowo-szkieletowego.
3	The etiology of TMDs is biopsychosocial and multifactorial.	Etiologia ZSŻ jest biopsychospołeczna i wieloczynnikowa.
4	Diagnosis of TMDs is based on standardized and validated history taking and clinical assessment performed by a trained examiner and led by the patient perspective.	Diagnoza ZSŻ opiera się na wystandaryzowanym i zwalidowanym wywiadzie oraz ocenie klinicznej przeprowadzanej przez wykwalifikowanego specjalistę, z uwzględnieniem perspektywy pacjenta.
5	Imaging has been proven to have utility in selected cases but does not replace the need for careful execution of history taking and clinical examination. Magnetic resonance imaging is the current standard of care for soft tissues and cone-beam computed tomography for bone. Imaging should only be performed when it has the potential to impact the diagnosis or treatment. Timing of imaging is important and so is the cost–benefit–risk balance.	Udowodniono, że badania obrazowe są przydatne w wybranych przypadkach, jednak nie zastępują konieczności przeprowadzenia dokładnego wywiadu i badania klinicznego. Obrazowanie metodą rezonansu magnetycznego stanowi obecnie standard diagnostyki tkanek miękkich, natomiast CBCT jest standardem w przypadku tkanek twardych. Obrazowanie należy wykonywać wyłącznie wtedy, gdy może ono wpłynąć na diagnozę lub sposób leczenia. Warto zwrócić uwagę na odpowiedni moment przeprowadzenia badania oraz uwzględnić bilans kosztów, korzyści i potencjalnych zagrożeń.
6	The evidence base for all interventions or devices should be carefully considered before their implementation over and above normal standard of care. Knowledge on developments in the field should be kept up to date. Currently, technological devices to measure electromyographic activity at chairside, to track jaw motion, or to assess body sway, amongst others, are not supported.	Przed wprowadzeniem wszystkich interwencji lub stosowanych urządzeń należy starannie rozważyć podstawy naukowe, wychodząc poza standardową opiekę. Wiedza na temat postępów w tej dziedzinie powinna być stale aktualizowana. Obecnie nie ma dowodów popierających stosowanie urządzeń do pomiaru aktywności elektromiograficznej mięśni w gabinecie, do śledzenia ruchów żuchwy, oceny stabilności ciała bądź innych tego typu technologii.
7	TMD treatment should aim to reduce the impact of pain and decrease functional limitation. Outcomes should be evaluated also in relation with the reduction of exacerbations, education in how to manage exacerbations, and improvement in quality of life.	Leczenie ZSŻ powinno mieć na celu redukcję odczuwanego bólu oraz zmniejszenie ograniczeń czynnościowych. Wyniki leczenia powinny być oceniane również w odniesieniu do zmniejszenia liczby zaostrzeń, edukacji w zakresie radzenia sobie z nimi oraz poprawy jakości życia.
8	TMD treatment should primarily be based on encouraging supported self-management and conservative approaches, such as cognitive-behavioral treatments and physiotherapy. Second-line treatment to support self-management includes provisional, interim, and time-limited use of oral appliances. Only very infrequently, and in very selected cases, are surgical interventions indicated.	Leczenie ZSŻ powinno opierać się przede wszystkim na wspieraniu samodzielnego radzenia sobie z objawami przez pacjenta oraz na podejściu zachowawczym, takim jak terapia poznawczo-behawioralna i fizjoterapia. Leczenie drugiego rzutu, wspierające samodzielne radzenie sobie z dolegliwościami, obejmuje doraźne, tymczasowe i ograniczone czasowo stosowanie szyn okluzyjnych. Interwencje chirurgiczne są wskazane bardzo rzadko i tylko w wyselekcjonowanych przypadkach.
9	Irreversible restorative treatment or adjustments to the occlusion or condylar position are not indicated in management of the majority of TMDs. The exception to this may be an acute change in the occlusion, such as in the instance of a high filling or crown with TMD-like symptoms developing immediately following these procedures or a slowly progressing change in dental occlusion due to condylar diseases.	Nieodwracalne leczenie protetyczne, korekty zwarcia lub zmiany pozycji głowy żuchwy nie są zalecane w leczeniu większości przypadków ZSŻ. Wyjątek mogą stanowić nagłe zmiany w zwarciu, na przykład po wykonaniu zbyt wysokiego wypełnienia lub korony protetycznej (działanie jatrogenne), gdy bezpośrednio po tych zabiegach pojawiają się objawy przypominające ZSŻ, lub powoli postępujące zmiany w zwarciu spowodowane zmianami w obrębie stawów skroniowo-żuchwowych.
10	The presence of complex clinical presentations with uncertain prognosis, such as in the case of concurrent widespread pain or comorbidities, elements of central sensitization, long-lasting pain, or history of previous failed interventions, should lead to the suspicion of chronification of TMDs or non-TMD pain. Referral to an appropriate specialist is thus recommended; the specialty will be geographic-specific as not all countries have a specialty of orofacial pain.	Występowanie złożonych przypadków klinicznych z niepewnym rokowaniem, takich jak w przypadku współistniejącego rozległego bólu lub chorób współistniejących, elementów sensytyzacji ośrodkowego układu nerwowego, długotrwałego bólu lub historii poprzednich nieudanych interwencji terapeutycznych, powinno budzić podejrzenie chronifikacji ZSŻ lub bólu niezwiązanego z ZSŻ. Zaleca się wówczas skierowanie pacjenta do odpowiedniego specjalisty; wybór specjalizacji będzie uzależniony od lokalizacji, ponieważ nie wszystkie kraje mają specjalizację w zakresie bólu ustno-twarzowego.

Fig. 1. Polish translation of the 10 key points for good clinical practice, International Network for Orofacial Pain and Related Disorders Methodology (INfORM) 2025
TMDs – temporomandibular disorders; CBCT – cone-beam computed tomography.

Original paper

Assessment of oral hygiene knowledge and health-promoting behaviors among adults in the Masovian Voivodeship, Poland: A cross-sectional survey study

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Abstract

Background. The examination of pro-health behaviors is important in the assessment of factors that influence the health of the population. Despite a constant increase in pro-health awareness observed within Polish society, the epidemiological picture of adult oral health remains unsatisfactory.

Objectives. The aim of the study was to assess the level of knowledge of the study population regarding the prevention of caries, the role of fluoride compounds, and hygiene habits.

Material and methods. A quantitative cross-sectional methodology was employed in the study. A specially designed anonymous survey was used to ensure the confidentiality of participants and to encourage truthful responses. The data was collected over a 5-month period using computer-assisted web interviewing (CAWI) with a Google Forms survey. The link to the survey was shared in public groups and on social media platforms, ensuring the collection of a diverse sample. Submitted surveys were cataloged and de-identified prior to analysis.

Results. The final sample size of the study was 643 participants. In the present study, only 95 respondents (14.77%) demonstrated sufficient knowledge about oral hygiene and declared behaviors that align with recommended practices. Factors such as place of residence, education and income did not have a statistically significant effect on the level of health awareness in relation to the preferred health attitude of the "ideal patient". The study revealed a lack of knowledge regarding the role of fluoride in caries prevention, with some individuals considering it harmful and refraining from using fluoride toothpaste. Approximately 20% of the surveyed population lacked awareness of the presence of fluoride in their toothpaste. These observations were comparable across both rural and urban areas.

Conclusions. The present study indicates insufficient knowledge regarding oral health among the study population. It is necessary to introduce more comprehensive oral health education programs targeted at the adult population of Poland.

Keywords: surveys and questionnaires, health education, oral hygiene, health behavior, cross-sectional studies

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Highlights

- Knowledge of health-promoting behaviors related to oral health remains at an unsatisfactory level.
- Preventive programs focused on dental caries are needed for more effective oral health promotion in adults.
- Patient education and habit formation should involve dental professionals as the primary source of accurate oral health information.

Introduction

Maintaining proper oral hygiene is essential for ensuring optimal oral health, which in turn affects the overall health of the individual. Additionally, oral health has an influence on self-esteem, physical and mental well-being, as well as interpersonal contacts, thereby affecting the quality of life.[1,2] The World Health Organization (WHO) defines oral health as "the state of the mouth, teeth and orofacial structures that enables individuals to perform essential functions such as eating, breathing and speaking, and encompasses psychosocial dimensions such as self-confidence, well-being and the ability to socialize and work without pain, discomfort or embarrassment. Oral health varies over the life course from early life to old age, is integral to general health and supports individuals in participating in society and achieving their potential".[3]

The etiology of the majority of oral diseases, including caries and periodontal diseases, is determined by common etiological factors that are shared with many chronic diseases.[4,5] These factors include lifestyle and associated poor nutrition, smoking, alcohol, drugs, and poor hygiene practices.[6] The primary means of enhancing oral health are interdisciplinary health promotion and prevention strategies, targeting the needs of the individual and the population.[7,8] An individual's oral health behavior is influenced by a multitude of factors, including socioeconomic status, sex, level of education and knowledge, and attitudes toward oral health.[9,10] However, consistent adherence to recommended behaviors, such as brushing at least twice a day, cleaning interdental surfaces and using fluoride, has been proven to effectively reduce the frequency and severity of dental caries.[11] The international evidence base demonstrates that the use of fluoride reduces the susceptibility of teeth to caries and demineralizes early enamel damage, contributing to a significant and rapid reduction in the incidence of caries.[12–19] Despite the widespread availability of modern toothbrushes, toothpastes, rinses, and a range of other oral hygiene products used in home prophylaxis, the inadequate level of health education remains the limiting factor.[20,21]

A notable increase in pro-health awareness has been observed among the Polish population over time. However, epidemiological data indicates that the oral health of adult Poles remains unsatisfactory, as evidenced by the data presented. A notable decline in the prevalence of dental caries and a reduction in the number of tooth extractions due to caries among individuals aged 34–44 years has been observed in the 2019 study compared to the 2010 study.[22] However, the value of the decay-missing-filled (DMF) index remained relatively high.[22]

Studies have indicated that the pro-health behaviors exhibited by the population deviate from the established standard.[22] A significant proportion of respondents stated that their most recent dental visit was prompted by toothache and the necessity for immediate treatment, rather than routine check-ups and preventive care.[22] This phenomenon is associated with a lack of comprehension regarding the significance of prevention, which consequently results in inferior health outcomes. The allocation of resources toward oral health education has the potential to reduce the financial burden associated with the control and management of dental caries and its associated complications. Numerous studies have attested to the efficacy of fluoride; however, the WHO announcement regarding its potential neurotoxic effect has led to an increasing number of patients discontinuing its use.[23–25] Despite the clear stance of experts on the safety of fluoride compounds in oral care products, some individuals refrain from using them. Furthermore, there has been a notable increase in social media discourse addressing public concerns about healthy living. Consequently, natural and organic oral care products have become more popular.[26] This phenomenon may contribute to the dissemination of misinformation about fluoride on online platforms.[27] Further research is necessary to evaluate the extent and implications of this trend.

The objective of this study was to assess the level of knowledge about the etiology of dental caries, health-promoting behaviors, and the role of fluoride compounds in preventing dental caries among the adult population residing in the Masovian Voivodeship in Poland. Additionally, the study aimed to identify factors that influence appropriate health-promoting attitudes among the studied population, thereby providing insights into the effectiveness of current oral health education and practices.

Material and methods

The present study was conducted from December 30, 2022, to April 27, 2023. A quantitative cross-sectional

methodology was employed using a questionnaire. Eligibility for participation in the study was determined based on specific inclusion and exclusion criteria. Individuals who met the following criteria were included in the study: age >18 years; possessing at least 5 permanent teeth; having no professional affiliation with the field of dentistry; and residing in the Masovian Voivodeship. Conversely, individuals under the age of 18, with fewer than 5 permanent teeth, those professionally involved in dentistry (e.g., dentists, hygienists, dental assistants, or dental students), and those residing outside the Masovian Voivodeship were excluded from the study. All individuals who met the inclusion criteria were considered eligible to participate in the study.

The survey was designed to be anonymous and consisted of 21 questions (supplementary material – available on request from the corresponding author). The first 5 questions were designed to collect sociomedical data using a single-choice format. The subsequent 16 questions included both single- and multiple-choice options and focused on health awareness and attitudes, as well as the sources of information regarding health and hygiene habits of the respondents.

In addition, for the purpose of the survey, we have distinguished the attitude of the "ideal patient" based on current medical knowledge. To qualify as the "ideal patient", respondents were required to correctly answer 6 out of the 16 questions regarding health awareness, including 4 key questions. The key questions posed to the respondents were as follows:

1. How often do you brush your teeth? The correct answers were: "twice a day" or "three times a day or more";
2. Do you use toothpaste with fluoride? The correct answer was: "yes";
3. Do you floss your teeth? The correct answer was: "yes, regularly";
4. How long do you brush your teeth? The correct answers were: "2 minutes" or "more than 2 minutes".

The data was collected using the computer-assisted web interviewing (CAWI) method with a Google Forms (Google LCC, Moutain View, USA) survey. The survey was meticulously designed to ensure participant anonymity, guarantee data confidentiality, and encourage candid responses. The survey link was disseminated through local Facebook groups specifically targeted to the Mazovian Voivodeship. Additionally, the participants were encouraged to share the survey link with their acquaintances residing in the Mazovian Voivodeship. The invitation to participate in the survey explicitly stated that only residents of the Mazovian Voivodeship were eligible to take part in the study. To maintain the integrity of the data, the Google Forms platform was configured to block multiple submissions from the same user, ensuring that each participant completes the survey only once.

Ethics approval for the study was obtained from the Bioethics Committee of the Medical University of Warsaw (No. AKBE/143/2022). The participants were informed about the purpose of the study and the potential benefits through the survey platform. Completion and submission of the survey were considered consent for participation in the study.

Statistical analysis

Descriptive statistics were employed to summarize the basic characteristics of the study group, thereby providing the number and percentage of responses to each question in the survey. A subsequent comparison was made between the residents from rural and urban areas regarding their behaviors and knowledge levels using the χ^2 test. This analysis provided a foundational understanding of the general characteristics of the survey data.

Inferential analysis was conducted using a logistic regression model to explore the relationships between various independent variables (e.g., age, sex and socioeconomic status) and dependent variables related to health-promoting behaviors and knowledge levels. The model parameters were estimated using the maximum likelihood estimation (MLE) method. This analysis was instrumental in identifying the key factors that influence the adoption of health-promoting behaviors among the adult population of Warsaw and its surrounding areas. Odds ratios (*OR*s) were calculated for each predictor within the logistic regression model, along with their 95% confidence intervals (*CI*s), thus enabling the quantification of the strength and precision of the observed associations.

A significance level of 0.05 was adopted for all statistical analyses in this study. Statistical significance was assigned to results with a p-value of 0.05 or lower, indicating that the observed effects have less than a 5% probability of being attributable to random variation. All statistical analyses were performed using STATISTICA v. 13.3 (TIBCO Software, Inc., Palo Alto, USA) under a license from the Medical University of Warsaw.

Results

A total of 680 questionnaires were initially collected, with 37 subsequently excluded for failing to meet the established criteria, resulting in a final sample size of 643 participants. The age distribution among the respondents exhibited variability, with the largest group being those aged 18–25, constituting 29.86% of the sample. A significant majority of the participants were female, representing 73.56% of the study group. Regarding educational attainment, 60.19% of the respondents held higher education degrees, while 36.24% had completed secondary education and 3.57% had finished primary education. The majority of the participants resided in towns or cities (58.48%), with the remainder living in rural areas (41.52%). The respondents exhibited a wide range of income levels,

Table 1. Sociodemographic characteristics of the study group ($N = 643$)

Variable		Respondents, *n* (%)
Sex	female	473 (73.56)
	male	170 (26.44)
Age [years]	18–25	192 (29.86)
	26–35	125 (19.44)
	36–45	132 (20.53)
	46–65	147 (22.86)
	>65	47 (7.31)
Education	primary	23 (3.57)
	secondary	233 (36.24)
	higher	387 (60.19)
Place of residence	village	268 (41.68)
	town/city	375 (58.32)
Income	below average	106 (16.49)
	average	352 (54.74)
	above average	185 (28.77)

with 54.74% reporting average income, 28.77% earning above average, and 16.49% below average. A comprehensive overview of the sociodemographic characteristics of the study group is presented in Table 1.

The vast majority of respondents followed the principles of oral prophylaxis. It is important to note that nearly 80% of respondents indicated that their source of information was a dentist. Statistically significant differences were identified in relation to the place of residence of the respondents. A higher percentage of people living in cities used information from their family than those living in rural areas. The study population included individuals who had never received professional oral hygiene instruction.

The research results indicate that the role of fluoride in caries prevention is unsatisfactory. Some individuals consider it harmful and therefore do not use fluoride toothpaste. In the surveyed population, nearly 20% of respondents lacked awareness regarding the presence of fluoride in their toothpaste. In addition, 40% of respondents did

Table 2. Responses to the survey questions regarding the oral health habits and knowledge of the study group in relation to the place of residence

Question	Answer	Respondents	Area of residence		
			rural	urban	*p*-value
What factors regarding oral hygiene are important to you?	healthy teeth	596 (92.69)	249 (92.91)	347 (92.53)	0.856
	white teeth	249 (38.72)	91 (33.96)	158 (42.13)	0.036*
	no caries	411 (63.92)	162 (60.45)	249 (66.40)	0.121
	healthy gums	414 (64.39)	153 (57.09)	261 (69.60)	0.001*
Where do you get your knowledge about oral hygiene?	I have never received instructions on this subject	48 (7.47)	16 (5.97)	32 (8.53)	0.223
	from parents/family members	237 (36.86)	84 (31.34)	153 (40.80)	0.014*
	from the dentist	494 (76.83)	197 (73.51)	297 (79.20)	0.092
	from advertisements	83 (12.91)	36 (13.43)	47 (12.53)	0.737
	at school	91 (14.15)	41 (15.30)	50 (13.33)	0.481
In your opinion, what is the most important factor limiting the development of tooth decay?	brushing your teeth	196 (30.48)	75 (27.99)	121 (32.27)	0.210
	using fluoride toothpaste	21 (3.27)	12 (4.48)	9 (2.40)	
	brushing your teeth and using fluoride toothpaste are equally important	426 (66.25)	181 (67.54)	245 (65.33)	
How often do you brush your teeth?	less than once a day	17 (2.64)	8 (2.99)	9 (2.40)	<0.001*
	once a day	91 (14.15)	51 (19.03)	40 (10.67)	
	two times a day	448 (69.67)	188 (70.15)	260 (69.33)	
	three times a day or more	87 (13.53)	21 (7.84)	66 (17.60)	
What kind of toothbrush do you use?	manual	289 (44.95)	134 (50.00)	155 (41.33)	0.055
	electric	123 (19.13)	54 (20.15)	69 (18.40)	
	sonic	138 (21.46)	48 (17.91)	90 (24.00)	
	both manual and electric	93 (14.46)	32 (11.94)	61 (16.27)	
Do you use toothpaste with fluoride?	yes	415 (64.54)	186 (69.40)	229 (61.07)	0.090
	no	100 (15.55)	37 (13.81)	63 (16.80)	
	I don't know	128 (19.91)	45 (16.79)	83 (22.13)	
Do you floss your teeth?	yes, occasionally	280 (43.55)	109 (40.67)	171 (45.60)	0.048*
	yes, regularly	155 (24.11)	58 (21.64)	97 (25.87)	
	no	208 (32.35)	101 (37.69)	107 (28.53)	
Do you use oral rinses?	yes	173 (26.91)	76 (28.36)	97 (25.87)	0.769
	no	230 (35.77)	93 (34.70)	137 (36.53)	
	yes, occasionally	240 (37.33)	99 (36.94)	141 (37.60)	

Question	Answer	Respondents	Area of residence		
			rural	urban	*p*-value
What oral rinses do you use?	mouthwash containing fluoride	190 (29.55)	88 (32.84)	102 (27.20)	0.372
	chlorhexidine rinse	76 (11.82)	25 (9.33)	51 (13.60)	
	alcohol rinse	84 (13.06)	35 (13.06)	49 (13.07)	
	mouthwash with essential oils	25 (3.89)	11 (4.10)	14 (3.73)	
What influences your choice of toothpaste?	price	211 (32.81)	86 (32.09)	125 (33.33)	0.741
	taste	182 (28.30)	75 (28.00)	107 (28.53)	0.879
	advertisements	76 (11.82)	35 (13.06)	41 (10.93)	0.410
	I use the paste that I currently have at home	258 (40.12)	102 (38.06)	156 (41.60)	0.367
	I don't know	85 (13.22)	35 (13.06)	50 (13.33)	0.920
What is the effect of fluoride in toothpaste?	it cleans teeth	111 (17.26)	56 (20.90)	55 (14.67)	0.039*
	it protects against caries	485 (75.43)	200 (74.63)	285 (76.00)	0.690
	it refreshes the oral cavity	70 (10.89)	27 (10.07)	43 (11.47)	0.576
	it whitens teeth	39 (6.07)	20 (7.46)	19 (5.07)	0.209
	it has a negative impact on health	35 (5.44)	10 (3.73)	25 (6.67)	0.106
	I don't know	96 (14.93)	37 (13.81)	59 (15.73)	0.499
When do you brush your teeth?	before breakfast	246 (38.26)	102 (38.06)	144 (38.40)	0.930
	after breakfast	372 (57.85)	136 (50.75)	236 (62.93)	0.002*
	before dinner	14 (2.18)	6 (2.24)	8 (2.13)	0.928
	after dinner	240 (37.33)	97 (36.19)	143 (38.13)	0.616
	right before bed	434 (67.50)	167 (62.31)	267 (71.20)	0.018*
How long do you brush your teeth?	half a minute or less	23 (3.58)	9 (3.36)	14 (3.73)	0.939
	1 min	89 (13.84)	39 (14.55)	50 (13.33)	
	<2 min	124 (19.28)	54 (20.15)	70 (18.67)	
	2 min	239 (37.17)	95 (35.45)	144 (38.40)	
	>2 min	168 (26.13)	71 (26.49)	97 (25.87)	
How much toothpaste do you use?	size of a pea	131 (20.37)	58 (21.64)	73 (19.47)	0.108
	about 0.5 cm	111 (17.26)	36 (13.43)	75 (20.00)	
	about 1 cm	176 (27.37)	81 (30.22)	95 (25.33)	
	about 2 cm	67 (10.42)	33 (12.31)	34 (9.07)	
	I apply the toothpaste to the electric toothbrush once during a single brushing session	144 (22.40)	53 (19.78)	91 (24.27)	
	I apply the toothpaste to the electric toothbrush twice during a single brushing session	14 (2.18)	7 (2.61)	7 (1.87)	
Do you rinse your mouth with water after brushing your teeth?	I don't rinse	59 (9.18)	22 (8.21)	37 (9.87)	0.037*
	yes, occasionally	42 (6.53)	11 (4.10)	31 (8.27)	
	yes, often	56 (8.71)	18 (6.72)	38 (10.13)	
	yes, always	486 (75.58)	217 (80.97)	269 (71.73)	
How much water do you use to rinse your mouth after brushing your teeth?	a handful of water	217 (33.75)	81 (30.22)	136 (36.27)	0.091
	two handfuls of water	190 (29.55)	77 (28.73)	113 (30.13)	
	half a glass of water	137 (21.31)	70 (26.12)	67 (17.87)	
	a full glass of water	43 (6.69)	20 (7.46)	23 (6.13)	

* statistically significant ($p < 0.05$, χ^2 test). Data presented as frequency (percentage) (*n* (%)).

not know which toothpaste they were currently using, relying on the toothpaste available at home. More than a third of respondents acknowledged that their purchase decisions were influenced by factors such as price and advertising. These observations were consistent across both rural and urban areas.

The statistical analysis demonstrated that place of residence is a significant factor in attitudes toward oral health priorities. Individuals residing in urban areas were more likely to report that having white teeth and healthy gums is important to them, compared to those living in rural areas. Similar trends were identified in the frequency of tooth brushing, flossing and mouth washing after brushing. Table 2 presents the detailed responses to the questions included in the questionnaire.

Figure 1 provides a comprehensive overview of oral hygiene practices, focusing on daily routines that contribute to oral health. The analysis reveals varied adherence to recommended practices, which are critical for maintaining a healthy oral cavity. The majority of respondents (83.2%) brush their teeth twice a day or more often, adhering to dental recommendations, while 14.15% of the participants brush their teeth once a day. A mere 2.6% of the respondents brush less than once a day, indicating a strong general compliance with the recommended twice-daily brushing regimen. Furthermore, the analysis indicates a high prevalence of fluoride toothpaste usage, with 64.5% of the participants reporting its incorporation in their oral hygiene routines. Concerning the practice of flossing, the data reveals that less than half of the respondents (43.5%) floss occasionally, while 24.1% engage in regular flossing. Notably, 32.3% of the respondents do not floss at all. With regard to the duration of tooth brushing, a fundamental component of effective oral hygiene, 37.2% of the participants brush for the recommended 2 min. However, 26.1% brush their teeth for more than 2 min. In contrast, 19.3% of the individuals brush for less than 2 min, 13.8% for only 1 min, and a small minority (3.6%) brush for half a minute or less.

In the present study, only 95 respondents (14.8%) demonstrated sufficient knowledge regarding oral hygiene and declared behaviors that align with recommended practices. The analysis revealed that sex and age were significant predictors of being an "ideal patient" who adheres to recommended oral hygiene practices. Specifically, the likelihood of men being classified as "ideal patients" was significantly lower in comparison to women, with *OR* of 0.47 (95% *CI*: 0.25–0.85, $p = 0.013$), indicating that men were about 53% less likely to demonstrate ideal oral hygiene behaviors than women. Furthermore, respondents aged 46–65 showed a significantly higher probability of adhering to recommended oral hygiene practices, with *OR* of 2.90 (95% *CI*: 1.49–5.64, $p = 0.002$), suggesting that they were almost 3 times more likely to adhere to recommended practices compared to the youngest age group (18–25 years). Other age groups did not show significant differences when compared to the reference group. The analysis revealed that other sociodemographic variables such as education, place of residence and income level did not demonstrate a significant correlation with the likelihood of being an "ideal patient" (Table 3).

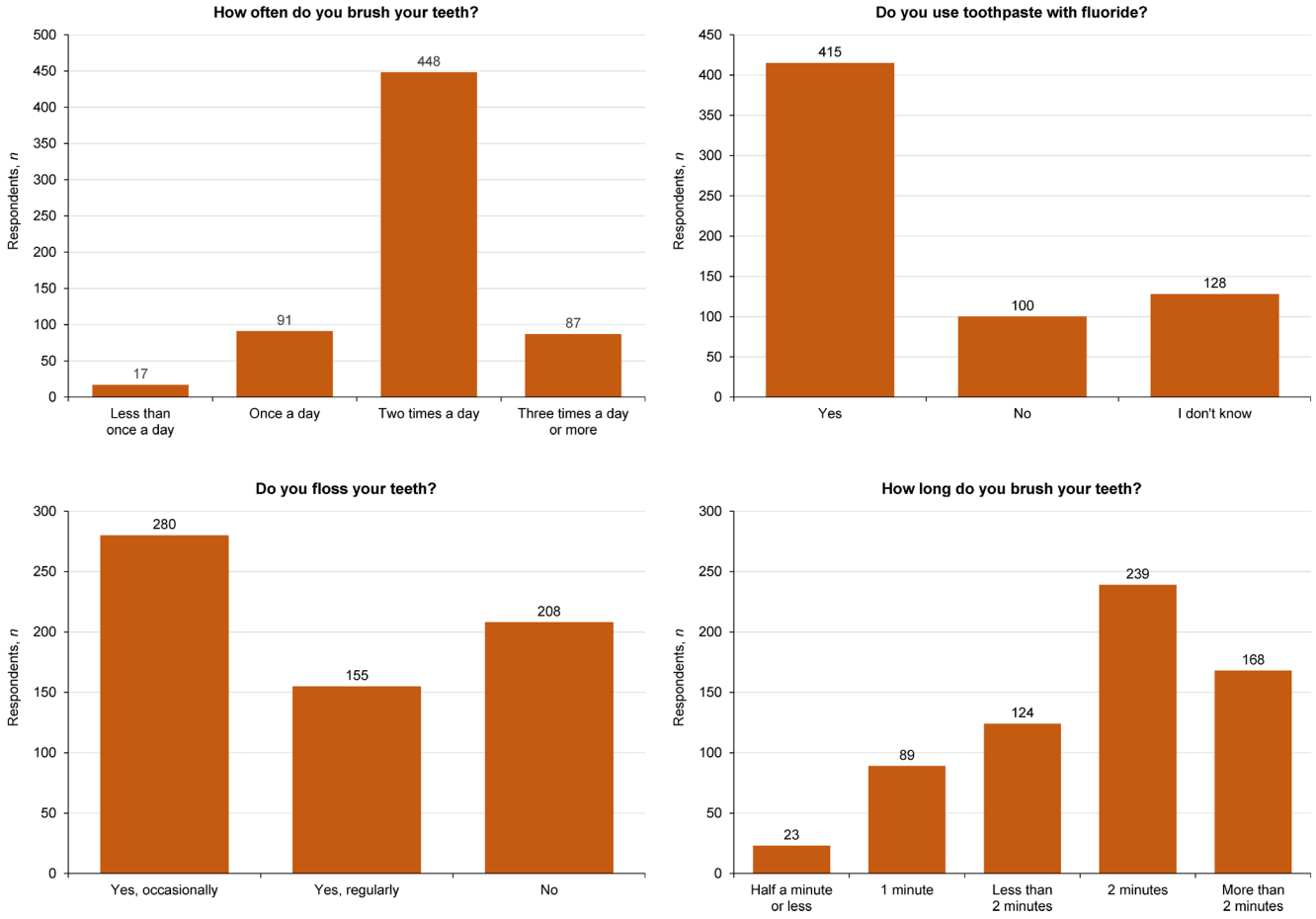

Fig. 1. Overview of oral hygiene practices in the study group ($N = 643$)

Table 3. Logistic regression analysis of factors influencing ideal oral hygiene behaviors in the study group

Predictor	Level	b	*OR*	95% *CI*	*Z*	*p*-value
Intercept	–	–1.85	0.16	0.03–0.78	–2.272	0.023*
Sex	female (ref.)	–	–	–	–	–
	male	–0.77	0.47	0.25–0.85	–2.494	0.013
Age [years]	18–25 (ref.)	–	–	–	–	–
	26–35	0.25	1.28	0.59–2.77	0.636	0.525
	36–45	0.47	1.60	0.76–3.38	1.228	0.219
	46–65	1.07	2.90	1.49–5.64	3.142	0.002*
	>65	–1.64	0.19	0.02–1.51	–1.564	0.118
Education	primary (ref.)	–	–	–	–	–
	secondary	–0.12	0.89	0.18–4.34	–0.151	0.880
	higher	0.29	1.34	0.28–6.41	0.369	0.712
Place of residence	village (ref.)	–	–	–	–	–
	town/city	0.17	1.18	0.73–1.91	0.675	0.500
Income	below average (ref.)	–	–	–	–	–
	average	–0.64	0.53	0.27–1.03	–1.883	0.060
	above average	–0.22	0.81	0.39–1.66	–0.588	0.557

* statistically significant ($p < 0.05$); b – unstandardized regression coefficient; *OR* – odds ratio; *CI* – confidence interval.

Discussion

The survey provided valuable information regarding the etiology of the high incidence of caries and periodontal diseases in the Polish population. The obtained results indicated that health awareness and related attitudes remain at a low level. The majority of respondents follow the basic rules of oral prevention, which is limited to brushing their teeth twice a day without regular flossing or using additional measures, e.g., fluoride rinses.

Socioeconomic status is an important determinant of health status, with its impact being manifested through dietary habits, hygiene practices and health awareness. The systemic changes that Poland has undergone have had a significant impact on the health status of the society. The general availability of a wide range of oral hygiene products, the development of the private sector, increased health awareness, but also the collapse of institutional dental care and relatively low expenditure on the medical sector create a situation that is difficult to assess unequivocally. Recent studies have indicated that individuals from families with a higher socioeconomic status exhibit a significantly better health status with respect to dental caries and periodontal diseases.[28–30] Similar trends have been observed in Polish society.[22] Despite the improvement in clinical parameters, a certain percentage of adult patients continue to experience the adverse effects of caries, gum and periodontal diseases in their daily lives.[31] In the present study, place of residence, reported income or education did not have a significant impact on the pro-health attitudes of respondents in the ideal patient model. However, the statistical analysis of the survey's questions sheds light on several additional aspects. The difference in attitudes between urban and rural residents was particularly interesting in relation to sources of dental information. Parents and family were statistically more frequently mentioned by urban residents. This finding underscores the important role of the dentist as a reliable source of knowledge and information for patients. Previous epidemiological studies have highlighted the lack of prevention and hygiene education in dental practices.[32] Routine dental visits present an opportunity for healthcare professionals to not only assess the risk of caries but also to motivate patients and update their knowledge. A study by Verploegen and Schuller revealed that patients receive insufficient information from the dental team during office visits.[33] Additionally, an increasing number of adults acquire health-related knowledge from the Internet.[33] While the Internet and social media may enhance health awareness, they do not necessarily motivate or increase self-efficacy in health promotion. This highlights the significance of providing evidence-based information in the dental office to modify patient behavior. The results of the study confirmed that dental professionals remain the most reliable source of information concerning prevention and oral health.[34]

The study respondents residing in urban areas were more likely to prioritize the importance of white teeth and healthy gums than respondents from rural areas. Similar trends were identified in relation to the frequency of brushing, flossing and mouth washing after brushing.

A total of 473 women and 170 men participated in the study, indicating that women are the more active and health-oriented group. At the same time, women exhibited

a higher level of knowledge and pro-health behavior. This phenomenon has been observed consistently over many years, not only in Polish society.[35–37] A notable finding was obtained in relation to the age of the respondents. Individuals over the age of 40 exhibited more favorable attitudes toward oral health maintenance. In contrast, an inverse relationship was described in the study by Jensen et al.[21] The authors observed that individuals over 35 years of age exhibited less favorable attitudes toward health promotion. These behaviors were particularly pronounced in the senior patient group.[21]

As individuals age, the importance of health increases due to the fact that the quality of life tends to deteriorate and there is an increase in limitations in independent functioning. While this relationship is evident in relation to general health, it remains insufficiently understood in relation to oral health. A growing number of patients prioritize the aesthetic appearance of their teeth, often neglecting their functional role.[38] The majority of respondents acknowledged the significance of healthy teeth, yet their responses indicated a lack of awareness regarding the progression of caries and the condition of the periodontium, suggesting a potential knowledge gap concerning these health concerns. Notably, individuals who considered the condition of their gums important and utilized chlorhexidine mouthwash, presumably due to underlying periodontal problems, presented more favorable pro-health attitudes. The hygienization phase of the treatment of patients with periodontal disease necessitates their adherence to recommendations. The prolonged nature of the treatment, which includes instruction, motivation and repeated professional dental cleaning procedures, contributes to patients' greater awareness and attention to maintaining good oral hygiene. The formation of proper health-promoting attitudes is implemented, but in practice, for a considerable number of individuals, this process ends during early childhood and school years. The situation is particularly problematic for adults, especially the senior population, for whom dental prophylaxis is often inadequate.

According to experts in the field, the theories regarding the adverse effects of fluoride are exaggerated and the concentration of this element in oral care products is sufficiently low to be considered safe.[39] Furthermore, there are no studies that associate fluoride in toothpaste with negative health effects. At the same time, there has been an increase in the number of patients who are aware of the contemporary world's demands and the transformations witnessed in recent years. Green dentistry, a concept linked to sustainable development, has emerged as a response to the pressing need for improvement in social, environmental and economic standards.[26] However, a paucity of studies has been conducted on the subject of organic toothpastes. Notably, none of the studies included comments on this topic. This may indicate that the promotion of these products is still insufficient. Price and advertising have been identified as the most common determinants of toothpaste purchase.

An effective oral hygiene routine includes brushing teeth twice a day, cleaning the interdental spaces, rinsing the mouth, using sugar-free gum, and maintaining an appropriate diet that limits the intake of products containing carbohydrates, including hidden sources. A study conducted in the late 1990s sought to assess the oral health of the Polish population and revealed a lack of awareness regarding health among patients.[34] At the time, the utilization of dental floss and the proper cleaning of interdental surfaces were the indicators of awareness. The authors found that while most respondents were aware of flossing, approx. 1 in 7 men and 1 in 4 young women actually practiced it.[34] Residential environment did not significantly differentiate either the awareness of the role of flossing or its use. In our research, improvements were noted in the issue at hand. After approx. 25 years, regular flossing has become a regular part of daily oral hygiene for a quarter of the respondents.

Limitations

The study's limitations include the selection of the population, which was focused on dwellings in Warsaw and its environs (Masovian Voivodeship). Therefore, the results obtained may be more favorable compared to the rest of the country, especially for those with lower incomes living in less developed areas.

To enhance the data collection process, it was decided to conduct the survey electronically. As a result, the study population consisted only of individuals with Internet access. Moreover, the nature of the survey design may have encouraged responses that were intended to improve the subjects' image, which could have resulted in the presentation of falsified data.

Conclusions

The outcomes of this research indicate that the population under study lack sufficient knowledge of oral health. The necessity for the implementation of effective domestic oral hygiene care measures is underscored. This study emphasizes the need to introduce comprehensive oral health education programs tailored to the adult Polish population, given the absence of dedicated programs for this demographic.

Ethics approval and consent to participate

Ethics approval for the study was obtained from the Bioethics Committee of the Medical University of Warsaw (No. AKBE/143/2022). Completion and submission of the survey were considered consent for participation in the study.

Data availability

The datasets generated and/or analyzed during the current study are available from the corresponding author on reasonable request.

Consent for publication

Not applicable.

Use of AI and AI-assisted technologies

Not applicable.

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References

1. Alshahrani S, Alshuaibi A, Alkhaldi M, Koppolu P. Perception and knowledge of patients from different regions in the kingdom of Saudi Arabia towards oral hygiene and oral hygiene aids. *Healthcare (Basel)*. 2021;9(5):592. doi:10.3390/healthcare9050592
2. Gerritsen AE, Allen PF, Witter DJ, Bronkhorst EM, Creugers NHJ. Tooth loss and oral health-related quality of life: A systematic review and meta-analysis. *Health Qual Life Outcomes*. 2010;8:126. doi:10.1186/1477-7525-8-126
3. World Health Organization. Oral Health. https://www.who.int/health-topics/oral-health#tab=tab_1. Accessed September 1, 2023.
4. Patel J, Wallace J, Doshi M, et al. Oral health for healthy ageing. *Lancet Healthy Longev*. 2021;2(8):e521–e527. doi:10.1016/S2666-7568(21)00142-2
5. Petersen PE, Bourgeois D, Ogawa H, Estupinan-Day S, Ndiaye C. The global burden of oral diseases and risks to oral health. *Bull World Health Organ*. 2005;83(9):661–669. PMID:16211157.
6. More AB, Rodrigues A, Sadhu BJ. Effect of smoking on oral health: Awareness among dental patients and their attitude towards its cessation. *Indian J Dent Res*. 2021;32(1):23–26. doi:10.4103/ijdr.IJDR_711_18
7. World Health Organization. Landmark global strategy on oral health adopted at World Health Assembly 75. https://www.who.int/news-room/feature-stories/detail/landmark-global-strategy-on-oral-health-adopted-at-world-health-assembly-75. Accessed September 1, 2023.
8. Ghaffari M, Rakhshanderou S, Ramezankhani A, Buunk-Werkhoven Y, Noroozi M, Armoon B. Are educating and promoting interventions effective in oral health? A systematic review. *Int J Dent Hyg*. 2018;16(1):48–58. doi:10.1111/idh.12305
9. Oyedele TA, Folayan MO, Chukwumah NM, Onyejaka NK. Social predictors of oral hygiene status in school children from suburban Nigeria. *Braz Oral Res*. 2019;33:e022. doi:10.1590/1807-3107bor-2019.vol33.0022
10. Lambert M, De Reu G, De Visschere L, Declerck D, Bottenberg P, Vanobbergen J. Social gradient in caries experience of Belgian adults 2010. *Community Dent Health*. 2018;35(3):160–166. doi:10.1922/CDH_4254Lambert07
11. Ganss C, Schlueter N, Preiss S, Klimek J. Tooth brushing habits in uninstructed adults – frequency, technique, duration and force. *Clin Oral Investig*. 2009;13(2):203–208. doi:10.1007/s00784-008-0230-8
12. Gao SS, Zhang S, Mei ML, Lo ECM, Chu CH. Caries remineralisation and arresting effect in children by professionally applied fluoride treatment: A systematic review. *BMC Oral Health*. 2016;16:12. doi:10.1186/s12903-016-0171-6
13. Amaechi BT, van Loveren C. Fluorides and non-fluoride remineralization systems. *Monogr Oral Sci*. 2013;23:15–26. doi:10.1159/000350458
14. Marinho VCC, Chong LY, Worthington HV, Walsh T. Fluoride mouthrinses for preventing dental caries in children and adolescents. *Cochrane Database Syst Rev*. 2016;7(7):CD002284. doi:10.1002/14651858.CD002284.pub2
15. Pessan JP, Toumba KJ, Buzalaf MAR. Topical use of fluorides for caries control. *Monogr Oral Sci*. 2011;22:115–132. doi:10.1159/000325154
16. Griffin SO, Regnier E, Griffin PM, Huntley V. Effectiveness of fluoride in preventing caries in adults. *J Dent Res*. 2007;86(5):410–415. doi:10.1177/154405910708600504
17. Schiffner U. Use of fluorides for caries prevention [in German]. *Bundesgesundheitsblatt Gesundheitsforschung Gesundheitsschutz*. 2021;64(7):830–837. doi:10.1007/s00103-021-03347-4
18. Satyarup D, Mohanty S, Nagarajappa R, Mahapatra I, Dalai RP. Comparison of the effectiveness of 38% silver diamine fluoride and atraumatic restorative treatment for treating dental caries in a school setting: A randomized clinical trial. *Dent Med Probl*. 2022;59(2):217–223. doi:10.17219/dmp/143547
19. Kooshki F, Fatemi SM, Darvishghaderi S, Vahedi P. Comparison of the effects of fluoride varnish containing silver nanoparticles and conventional fluoride varnish on the surface microhardness of tooth enamel. *Dent Med Probl*. 2024;61(2):241–247. doi:10.17219/dmp/151702
20. Hedman E, Ringberg C, Gabre P. Knowledge of and attitude to oral health and oral diseases among young adolescents in Sweden. *Swed Dent J*. 2006;30(4):147–154. PMID:17243442.
21. Jensen O, Gabre P, Sköld UM, Birkhed D. Is the use of fluoride toothpaste optimal? Knowledge, attitudes and behaviour concerning fluoride toothpaste and toothbrushing in different age groups in Sweden. *Community Dent Oral Epidemiol*. 2012;40(2):175–184. doi:10.1111/j.1600-0528.2011.00658.x
22. Olczak-Kowalczyk D, Mielczarek A, Kaczmarek U, Turska-Szybka A, Rusyan E, Adamczyk K. *Choroba Próchnicowa i Stan Tkanek Przyzębia Populacji Polskiej: Podsumowanie Wyników Badań z Lat 2016–2019*. Warsaw, Poland: Warszawski Uniwersytet Medyczny; 2021.
23. Guth S, Hüser S, Roth A, et al. Toxicity of fluoride: Critical evaluation of evidence for human developmental neurotoxicity in epidemiological studies, animal experiments and in vitro analyses. *Arch Toxicol*. 2020;94(5):1375–1415. doi:10.1007/s00204-020-02725-2
24. Miranda GHN, Alvarenga MOP, Ferreira MKM, et al. A systematic review and meta-analysis of the association between fluoride exposure and neurological disorders. *Sci Rep*. 2021;11(1):22659. doi:10.1038/s41598-021-99688-w
25. Grandjean P. Developmental fluoride neurotoxicity: An updated review. *Environ Health*. 2019;18(1):110. doi:10.1186/s12940-019-0551-x
26. Mazur M, Ndokaj A, Bietolini S, Nisii V, Duś-linicka I, Ottolenghi L. Green dentistry: Organic toothpaste formulations. A literature review. *Dent Med Probl*. 2022;59(3):461–474. doi:10.17219/dmp/146133
27. Lotto M, Zakir Hussain I, Kaur J, Butt ZA, Cruvinel T, Morita PP. Analysis of fluoride-free content on Twitter: Topic modeling study. *J Med Internet Res*. 2023;25:e44586. doi:10.2196/44586
28. Eustaquio MV, Montiel JM, Almerich JM. Oral health survey of the adult population of the Valencia region (Spain). *Med Oral Patol Oral Cir Bucal*. 2010;15(3):e538–e544. doi:10.4317/medoral.15.e538
29. Carvalho JC, Schiffner U. Dental caries in European adults and senior citizens 1996–2016: ORCA Saturday Afternoon Symposium in Greifswald, Germany – part II. *Caries Res*. 2019;53(3):242–252. doi:10.1159/000492676
30. Oscarson N, Espelid I, Jönsson B. Is caries equally distributed in adults? A population-based cross-sectional study in Norway – the TOHNN-study. *Acta Odontol Scand*. 2017;75(8):557–563. doi:10.1080/00016357.2017.1357080
31. White DA, Tsakos G, Pitts NB, et al. Adult Dental Health Survey 2009: Common oral health conditions and their impact on the population. *Br Dent J*. 2012;213(11):567–572. doi:10.1038/sj.bdj.2012.1088
32. Małkiewicz E, Wierzbicka M, Szatko F, et al. *Stan Zdrowia Jamy Ustnej i Jego Uwarunkowania oraz Potrzeby Profilaktyczno-Lecznicze Dzieci w Wieku 6 i 12 lat oraz Osób Dorosłych w Wieku 35–44 lat*. Warsaw, Poland: Warszawski Uniwersytet Medyczny; 2010.
33. Verploegen VJN, Schuller AA. Erosive tooth wear: Knowledge among young adults and their preferred information sources. *Int J Dent Hyg*. 2019;17(1):85–92. doi:10.1111/idh.12367

34. Wierzbicka M. *Intensyfikacja Profilaktyki Próchnicy Zębów i Chorób Przyzębia u Dzieci, Młodzieży i Kobiet Ciężarnych*. Warsaw, Poland: Państwowy Zakład Higieny; 1998:159–164.
35. Grzymisławska M, Puch EA, Zawada A, Grzymisławski M. Do nutritional behaviors depend on biological sex and cultural gender? *Adv Clin Exp Med*. 2020;29(1):165–172. doi:10.17219/acem/111817
36. Lipsky MS, Su S, Crespo CJ, Hung M. Men and oral health: A review of sex and gender differences. *Am J Mens Health*. 2021;15(3):15579883211016361. doi:10.1177/15579883211016361
37. Bukmir RP, Paljević E, Pezelj-Ribarić S, Pršo IB. Association of the self-reported socioeconomic and health status with untreated dental caries and the oral hygiene level in adult patients. *Dent Med Probl*. 2022;59(4):539–545. doi:10.17219/dmp/138908
38. Zavanelli AC, Sônego MV, Zavanelli RA, Quinelli Mazaro JV, Falcón-Antenucci RM. Perception and expectation: What do patients really want from the dental treatment? *Rev Gaúch Odontol*. 2017;65(3):243–248. doi:10.1590/1981-86372017000200093257
39. Toumba KJ, Twetman S, Splieth C, Parnell C, van Loveren C, Lygidakis NA. Guidelines on the use of fluoride for caries prevention in children: An updated EAPD policy document. *Eur Arch Paediatr Dent*. 2019;20(6):507–516. doi:10.1007/s40368-019-00464-2

Original paper

Treatment of deciduous teeth in children using the Er:YAG laser compared to the traditional method: A randomized clinical trial

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Abstract

Background. Erbium family lasers have been used as an alternative method for the treatment of dental caries in deciduous teeth, in comparison to traditional methods.

Objectives. The study aimed to determine the most optimal erbium-doped yttrium-aluminum-garnet (Er:YAG) laser settings for carious lesions in deciduous teeth on different surfaces with the evaluation of treatment time and pain level on a visual analog scale (VAS).

Material and methods. The randomized clinical trial involved 66 teeth with deep caries in 33 children aged 3–8 years, divided into 2 study groups: Er:YAG laser (test group); and conventional treatment with a dental turbine (control group). The time required for caries removal and cavity preparation using both methods was recorded. The inclusion criteria encompassed generally healthy pediatric patients, pain-free subjects, first-time dental appointment, and unicuspid primary teeth with caries. Dental caries treatment was performed without the administration of local anesthesia.

Results. The cavity preparation time was on average 2.5 times longer when the Er:YAG laser was used compared to the conventional method. The study demonstrated the effectiveness of the Er:YAG laser in hard tissue preparation of deciduous teeth with a pulse energy of 230 mJ for enamel preparation, mainly on occlusal and proximal surfaces, and with a pulse energy of 120 mJ and 150 mJ for dentin preparation. Optimum operating frequencies were 10 Hz and 20 Hz with corresponding water consumption. The level of discomfort among the laser-treated patients indicated 0 based on VAS, meaning no pain. Among patients treated with the traditional method, the pain level averaged 5.27.

Conclusions. The use of the Er:YAG laser offers substantial advancements in comparison to traditional treatment with dental turbines by enhancing the overall comfort during caries therapy in children. It fosters better patient cooperation and reduces the need for intraoperative anesthetics due to its minimally invasive nature and the fact that it is a relatively quiet operation. This approach minimizes anxiety and discomfort often associated with dental procedures, making them more amenable to children.

Keywords: pain, dental caries, laser, primary teeth, Er:YAG

Highlights

- The Er:YAG laser enables effective, minimally invasive cavity preparation in deciduous teeth of children aged 3–8 years, with reduced need for local anesthesia.
- Patients treated with the laser reported no pain (VAS: 0), compared to moderate pain experienced in the conventional treatment group (VAS: 5.27).
- Despite longer procedure time, children showed better cooperation and preferred laser treatment for future visits.
- The Er:YAG laser preserved pulp vitality, while conventional treatment caused significant increases in post-treatment readings, suggesting pulpal irritation.
- Standardized laser parameters such as the applicator type, angle, distance, and movement are essential for safe and effective pediatric dental procedures.

Introduction

Maintaining primary dentition until its natural replacement by permanent teeth is extremely important. Unfortunately, the treatment of primary teeth is still underappreciated by parents and dental practitioners. This phenomenon is also reflected in the limited number of scientific publications addressing this subject. If dental caries in primary teeth progresses to apical periodontitis, it can impact the tooth germ of the developing permanent teeth, increasing the risk of caries in newly erupted permanent teeth. The contemporary field of restorative dentistry for primary teeth places a significant emphasis on the utilization of minimally invasive techniques for the preparation and filling of carious lesions. Traditionally, turbine drills and micromotors were used. However, the loud sound and intense vibrations from rotary instruments often diminish children's cooperation during treatment and contribute to the development of dental anxiety in children. Therefore, with the advancement of technology, there is an increasing demand for contemporary methods of caries treatment that do not provoke negative experiences in children. One such method involves the use of the erbium-doped yttrium-aluminum-garnet (Er:YAG) laser.[1–4] High-power laser light is known due to its bactericidal effect.[5–18] This property of laser can reduce the risk of secondary infections and recurrent caries, contributing to long-lasting restorations. Erbium family lasers operate through a process known as thermo-mechanical ablation, as they are highly absorbed by the water chromophore within dental carious tissue. This interaction leads to the precise and minimally invasive removal of decayed tissue. The water content present in primary dental tissue is higher than in permanent teeth due to its lower degree of mineralization, which results in higher energy absorption. Lasers efficiently remove decayed and infected tissue without affecting the healthy parts of the tooth. Thus, lasers require different parameters for caries ablation in primary teeth compared to permanent teeth. However, current research has yet to establish definitive guidelines for the optimal physical parameters of the Er:YAG laser in the minimally invasive treatment of carious lesions in primary teeth. These teeth have a different structure from permanent teeth and thus require different physical parameters for proper cavity preparation.[19–21]

The null hypothesis of this study posits that the use of the Er:YAG laser for the preparation of carious lesions in primary teeth requires the application of a radiation beam perpendicular to the direction of enamel prisms and dentinal tubules, with appropriate low voltage delivered through a small-diameter optical fiber, along with significant water cooling. This approach is predicted to have no effect on pulp vitality and should not reduce pain sensations during the procedure.

The aim of the study was to determine the optimal settings for the Er:YAG laser in treating advanced lesions in the enamel and dentin of primary teeth on occlusal, interproximal and smooth surfaces. This investigation was conducted through the implementation of randomized clinical trials. The settings were compared to conventional methods for treating caries in primary teeth. The time required to prepare the cavity using the laser and conventional methods was examined. A pulp tester (Digitest® Pulp Vitality Tester; Parkell Inc., Edgewood, USA) was used to assess the impact of both methods on the pulp of the treated primary teeth before and immediately after cavity preparation. Additionally, the pain perception of young patients in both the laser and conventional groups was evaluated.[22–25]

Material and methods

Ethics approval

The study was designed as a randomized controlled trial. The Ethics Committee of Wroclaw Medical University, Poland approved the study (approval No. KB-547/2021), and informed consent was obtained from all participating subjects, as stipulated in the Declaration of Helsinki. This study was registered with the ISRCTN (International Standard Randomised Controlled Trial Number) Registry under the reference No. 45696-OH7CP3.

Study sample

Initially, a total of 47 subjects (94 teeth) were evaluated for eligibility. Of these, 14 patients (28 teeth with caries) were excluded from the study. This group included 11 children (22 teeth) who did not participate due to previous dental office visits, 2 individuals (4 teeth) who refused to participate, and 1 patient (2 teeth) who dropped out of the study due to an existing systemic disease. This randomized, split-mouth clinical trial involved 66 teeth affected by deep caries (33 subjects of both sexes; age: 3–8 years). The sample size was calculated to be 33 in each group using the G*Power v. 3.1 (Kiel University, Kiel, Germany) software, assuming 80% power, 95% confidence intervals, a level of significance of 0.05, and d = 0.62, based on preliminary studies conducted on a smaller number of subjects. Each tooth on the test side (n = 33) of the maxilla or mandible was treated with the Er:YAG laser, while the teeth on the opposite side (n = 33) served as a control group and received conventional treatment (Fig. 1).

The inclusion criteria encompassed children who had visited a dental office for the first time in their lives for a dental examination and possible conservative treatment, healthy children who were not taking any medication, subjects who did not report any pain when coming to the office, subjects aged 3–8 years, and patients with unicuspidal deciduous teeth with deep caries. The same dentist treated all patients.

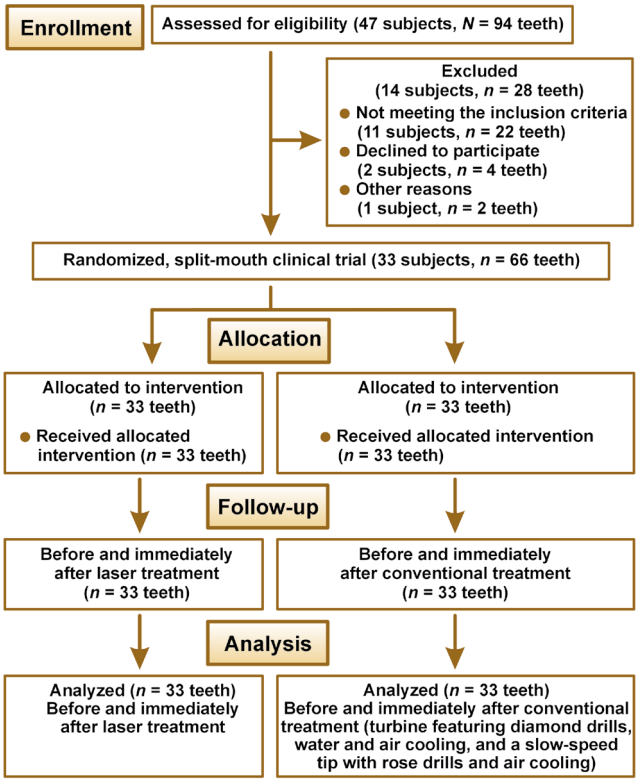

Fig. 1. CONSORT (Consolidated Standards of Reporting Trials) 2010 flowchart of the study

The initial visit did not include caries treatment. Instead, the children underwent a series of adaptive visits, tailored to their age and individual needs. This was followed by a comprehensive dental examination that included oral hygiene instruction, dietary guidelines, and a procedure to clean plaque from all teeth. The dental examination was conducted using the DIAGNOcam™ Vision Full HD diagnostic device (KaVo, Biberach, Germany) to identify occlusal and proximal caries in primary teeth. Patients presenting with carious lesions of similar severity in unicuspidal teeth were eligible for inclusion in the study. The decision as to which tooth would be treated with the conventional method and which with the laser method, as well as the method of the initial cavity preparation were selected through computerized randomization (blinding). The computer program determined whether the initial preparation would be conducted using conventional methods or the Er:YAG laser.

The conventional treatment was initiated in 12 patients, and the laser was employed as the initial treatment modality in 21 patients. The conservative management of deciduous teeth was conducted without the use of anesthetics (Fig. 1).

Treatment procedures

This study evaluated the efficacy of different treatment modalities for the removal of dental caries. One group of patients received treatment with the Er:YAG laser (AdvErL Evo; Morita, Kyoto, Japan), while the other group was treated with a turbine featuring diamond drills, water and air cooling, and a slow-speed tip with rose drills and air cooling.

Characteristics of the conventional method

In the traditional method, a turbine with blue-coated diamond drills (19 mm long, sizes 012–018), as well as water and air cooling were employed to create the cavities and work on enamel. For dentin preparation, a slow-speed carbide tip (22 mm long, sizes 018–021) with air cooling was used. The restorative material was glass ionomer.

Laser settings

A summary of the laser settings utilized for each group of cavities is presented in Tables 1–3. The following Er:YAG laser parameters were employed in the enamel on all surfaces: a frequency of 10 Hz for cavity opening and 20 Hz for enamel modification; a pulse energy of 160 mJ with a 400-µm diameter applicator and 230 mJ. The 600-µm diameter applicator was used to develop cavities on the proximal and occlusal surfaces. On smooth surfaces (palatal and labial) of anterior teeth, a pulse energy of 150 mJ with a 600-µm diameter and a pulse energy of 180 mJ with a 800-µm diameter were applied.

In the surface modification procedure, the pulse energy was set at 50 mJ and 70 mJ for 400-µm and 600-µm diameter applicators, respectively. The water content values oscillated between 2.5–3.0 mL/min, and the air cooling was set to 7 on a scale from 1 to 10.

The parameters employed in dentin across all cavity classes were as follows: a frequency of 10 Hz, a pulse energy of 120 mJ with a 600-µm diameter applicator, and 150 mJ with an 800-µm diameter applicator, respectively. The water content was used at a rate of 2.0–2.5 mL/min, and the air cooling was set to 7 on a scale from 1 to 10. The pulse duration was maintained at 300 µs throughout the enamel and dentin treatment (Table 1).

Laser applicators, type of primary teeth and working surfaces

The procedure was conducted in a non-contact mode, with the applicator positioned 1 mm from the target area in both enamel and dentin. The occlusal and proximal surfaces of the enamel in deciduous molars were treated with applicators that had a diameter of 400 µm and 600 µm. The smooth palatal and labial surfaces of the enamel in incisor and canine teeth were treated with a 800-µm diameter applicator. Regardless of the cavity class, dentin was treated with applicators having diameters of 600 µm and 800 µm.

During the operation, the applicator was positioned at a 90° angle to the tooth surface. This angle was selected as the ideal impact angle for laser ablation of enamel, as it is perpendicular to the orientation of the prisms. The procedure was performed with slow, overlapping movements, following the recommendations outlined in Table 2.

Pulp vitality

The Digitest® Pulp Vitality Tester was employed to assess the vitality of the dental pulp. The device quantifies pulp vitality on a scale ranging from 0 to 80 (0–40 – vital pulp; 40–80 – pulpitis; <80 – necrotic pulp). The pulp responses of deciduous teeth were studied using both traditional and laser methods.

Table 1. Characteristics of Er:YAG laser settings (wavelength: 2,940 nm)

Type of tissue		Frequency [Hz]	Pulse energy [mJ]	Type of applicator	Water content [mL/min]	Air cooling	Pulse duration [µs]
Enamel	occlusal surface	10	160 230	C400F C600F	2.5–3.0	7	300
	proximal surface	10	160 230	C400F C600F	2.5–3.0	7	300
	labial or palatal surface of anterior teeth	10	150 180	C600F C800F	2.5–3.0	7	300
Dentin	occlusal surface	10	120 150	C600F C800F	2.0–2.5	7	300
	proximal surface	10	120 150	C600F C800F	2.0–2.5	7	300
	labial or palatal surface of anterior teeth	10	120 150	C600F C800F	2.0–2.5	7	300
Surface modification	every surface	20	50 70	C400F C600F	2.5–3.0	7	300

Table 2. Characteristics of laser applicators, types of primary teeth treated and working surfaces

Type of applicator	Diameter of applicator [µm]	Setting angle	Contact/non-contact mode	Type of movement performed	Type of primary teeth	Working surface	Type of tissue
C400F/C600F C600F/C800F	400/600 600/800	perpendicular to the orientation of the prisms	non-contact mode, 1 mm away from the target area	slow, overlapping, successive movements	molars	occlusal	enamel and dentin
C400F/C600F C600F/C800F	400/600 600/800	perpendicular to the orientation of the prisms	non-contact mode, 1 mm away from the target area	slow, overlapping, successive movements	molars	proximal	enamel and dentin
C600F/C800F	600/800	perpendicular to the orientation of the prisms	non-contact mode, 1 mm away from the target area	slow, overlapping, successive movements	incisors, canines	palatal or labial	enamel and dentin

Working time

In the preparation of carious cavities in deciduous teeth, the working lengths were measured using conventional methods and the Er:YAG laser with the aid of a stopwatch.

VAS scale

Following the conventional and laser treatment visits, patients were asked to rate their pain sensations using emoticons according to the visual analog scale (VAS).[23,24] The results were collected and documented separately for the laser method and separately for the conventional method.

Statistical analysis

In order to answer the research questions and test the hypotheses, the statistical analysis was performed using the IBM SPSS Statistics for Windows software, v. 26.0 (IBM Corp., Armonk, USA). The Student's *t*-test was employed for both dependent samples, with $p < 0.05$ indicating the level of statistical significance.

Results

The study's participants included 33 subjects who underwent treatment with laser and conventional methods. The distributions for the treatment characteristics in both methods were examined sequentially. The laser method was divided into 3 stages, namely enamel treatment, dentin treatment and enamel modification. For each method, the teeth most frequently treated were molars (69.7%). A mere 18.2% of the treated teeth were incisors, while the remaining 12.1% comprised canines.

Characteristics of treatment methods

Further characterization was related solely to the laser method. The most frequently selected applicator type in the laser method for enamel work was the C600F applicator (54.5%). Over one-third (36.4%) of the enamel work settings were allocated to the C400F applicator. The remaining parameters (9.1%) pertained to the C800F applicator. In the context of dentin or enamel modification, the majority of settings (60.6%) were associated with the C600F applicator. In the remaining cases (39.4%), the applicator type was identified as C800F.

For enamel work, the pulse energy was predominantly 160 mJ (48.5%) and 230 mJ (33.3%). The remaining cavity preparations were based on pulse energies of 150 mJ (9.1%) and 180 mJ (9.1%). In the case of dentin, the pulse energy was lower. In the majority of cases, the energy level was set at 120 mJ (60.6%). The remaining studies on dentin employed a pulse energy of 150 mJ (39.4%), while enamel modification was treated with a weaker energy of 50 mJ (60.6%) and 70 mJ (39.4%).

The frequency for cavity preparation on enamel and dentin was 100.0% at 10 Hz, while for enamel modification, the frequency was 20 Hz (100.0%). During the process of cavity preparation in the enamel area or its

Table 3. Characteristics of the laser method and the conventional method ($N = 33$)

Variable		Laser method			Conventional method
		enamel	dentin	enamel modification	
Type of primary teeth	incisors	6 (18.2)	6 (18.2)	6 (18.2)	6 (18.2)
	canines	4 (12.1)	4 (12.1)	4 (12.1)	4 (12.1)
	molars	23 (69.7)	23 (69.7)	23 (69.7)	23 (69.7)
Type of applicator	C400F	12 (36.4)	0 (0.0)	0 (0.0)	–
	C600F	18 (54.5)	20 (60.6)	20 (60.6)	–
	C800F	3 (9.1)	13 (39.4)	13 (39.4)	–
Pulse energy [mJ]	50	0 (0.0)	0 (0.0)	20 (60.6)	–
	70	0 (0.0)	0 (0.0)	13 (39.4)	–
	120	0 (0.0)	20 (60.6)	0 (0.0)	–
	150	3 (9.1)	13 (39.4)	0 (0.0)	–
	160	16 (48.5)	0 (0.0)	0 (0.0)	–
	180	3 (9.1)	0 (0.0)	0 (0.0)	–
	230	11 (33.3)	0 (0.0)	0 (0.0)	–
Frequency [Hz]	10	33 (100.0)	33 (100.0)	0 (0.0)	–
	20	0 (0.0)	0 (0.0)	33 (100.0)	–
Water content [mL/min]	2.0–2.5	0 (0.0)	33 (100.0)	0 (0.0)	–
	2.5–3.0	33 (100.0)	0 (0.0)	33 (100.0)	–

Data presented as frequency (percentage) (*n* (%)).

modification, the water content was maintained at a level of 2.5–3.0 mL/min. In the dentin area, this value was lower (2.0–2.5 mL/min). The area setting for all laser preparations was set to 7, and in each case, the pulse duration was 300 µs. Table 3 presents the discussed data on the characteristics of both methods.

Subsequently, basic descriptive statistics were calculated for the variables of patient age, time to complete cavity treatment, pain as measured by the VAS, and pulp vitality before and after treatment. The calculations are presented separately for subjects treated with the laser method and separately for those treated with the conventional method (Table 4).

The analysis revealed that the average age of the children was 5.27 ±1.42 years. The youngest child was 3 years old, while the oldest was 8. The mean time to complete cavity preparation was 195.94 ±30.13 s among patients treated with the laser method. The shortest total cavity preparation time was recorded at 147 s, while the longest was 254 s. The mean time to complete cavity repair using the conventional method was 77.67 ±11.09 s. The shortest amount of time to complete cavity repair was 50 s, while the longest amount of time to complete cavity repair was 97 s. The level of discomfort measured by the VAS among the laser-treated patients was 0, indicating no pain. Among the subjects treated with the traditional method, pain levels exhibited a mean of 5.27 ±2.34, with the lowest recorded level at 2 and the highest indicating maximum pain intensity. In the group that received laser treatment, pulp vitality remained at the same level before and after the intervention (25.82 ±0.85). The lowest recorded value for pulp content in this group was 25, while the highest was 27. Among the children treated with the traditional method, differences were observed between the pre-operative and post-operative measurements. The pre-treatment mean pulp content was found to be analogous to that observed in the laser group, with a value of 25.88 ±0.82. The lowest recoded value for pulp compactness was 25, while the highest was 27. Following treatment, the value increased to 32.30 ±2.01. The lowest recorded value for pulp compactness was 29, while the highest was 35.

Descriptive statistics of the measured quantitative variables

In the subsequent phase of the analysis, the distribution of the quantitative variables was evaluated. This objective was achieved by calculating basic descriptive statistics and applying the Shapiro–Wilk test to determine the normality of the distribution. The results of this analysis are presented in Table 5.

Table 4. Descriptive statistics regarding the age of children, operative time, pain, and pulp vitality in conventional and laser-treated groups

Variable			*M*	*SD*	Min	Max
Age [years]			5.27	1.42	3.00	8.00
Laser method	time to complete cavity treatment [s]		195.94	30.13	147.00	254.00
	pain (VAS)		0	0	0	0
	pulp vitality	before treatment	25.82	0.85	25.00	27.00
		after treatment	25.82	0.85	25.00	27.00
Conventional method	time to complete cavity treatment [s]		77.67	11.09	50.00	97.00
	pain (VAS)		5.27	2.34	2.00	10.00
	pulp vitality	before treatment	25.88	0.82	25.00	27.00
		after treatment	32.30	2.01	29.00	35.00

M – mean; *SD* – standard deviation; Min – minimum value; Max – maximum value; VAS – visual analog scale.

Table 5. Descriptive statistics regarding pulp vitality and treatment time along with the results of the Shapiro–Wilk test

Variable			*M*	*Me*	*SD*	Sk	Kurt	Min	Max	W	*p*-value
Laser method	pulp vitality	before treatment	25.82	26.00	0.85	0.37	–1.52	25.00	27.00	0.77	<0.001*
		after treatment	25.82	26.00	0.85	0.37	–1.52	25.00	27.00	0.77	<0.001*
	time to complete cavity treatment [s]		195.94	199.00	30.13	0.12	–0.64	147.00	254.00	0.96	0.331
Conventional method	pulp vitality	before treatment	25.88	26.00	0.82	0.23	–1.47	25.00	27.00	0.79	<0.001*
		after treatment	32.30	33.00	2.01	–0.15	–1.28	29.00	35.00	0.91	0.013*
	time to complete cavity treatment [s]		77.67	79.00	11.09	–0.28	–0.15	50.00	97.00	0.98	0.704

* statistically significant ($p < 0.05$); Kurt – kurtosis; Sk – skewness.

The Shapiro–Wilk test results were found to be statistically insignificant for the time of complete cavity preparation, irrespective of whether a laser or traditional methods were employed. This finding suggests that, when categorized into 2 groups, this variable approached a normal distribution. In contrast, the results of the normality of distribution test suggest a distribution that does not align with the Gaussian curve for the other variables. Nevertheless, the absolute value of the skewness for each variable did not exceed 2, indicating a slight asymmetry in these distributions. Therefore, the analyses were based on parametric tests, provided that the remaining assumptions of these tests were met.

Differences in total cavity preparation time according to the method

In the subsequent stage of the statistical analysis, the total preparation time for 2 methods of cavity preparation, namely the laser method and the traditional method, was compared using the Student's *t*-test for dependent samples (Table 6).

The test results were found to be statistically significant, indicating the presence of a substantial observed effect. This finding suggests that there are differences between the methods in terms of the time required to complete the cavity, with the laser method requiring more time than the conventional method.

Pulp vitality before and after conventional treatment

Subsequently, the vitality of the pulp was evaluated before and after treatment using the Student's *t*-test for independent samples. This analysis was conducted exclusively in the conventional treatment group. In the group that underwent laser treatment, the pulp vitality values remained consistent before and after the procedure.

The results of the test were found to be statistically significant, indicating the presence of notable differences between the measurements of pulp vitality (Table 7). The observed effect was of high power. In the cohort of subjects who received the conventional treatment modality, the values for pulp vitality were found to be significantly higher after treatment in comparison to the pre-treatment values. This finding suggests that the procedure may have had an adverse effect on the vitality of the pulp.

Discussion

The utilization of laser technology in restorative dentistry remains a novel concept, not only in how it changes traditional treatment approaches but also in the manner in which it is perceived by dental professionals and their patients. The utilization of the Er:YAG laser for the treatment of carious enamel and dentin defects provides a more comfortable and safer experience for young patients when compared to conventional methods. The paucity of literature on the laser treatment of deciduous teeth underscores the importance of further research in this area, with the aim of developing a protocol for the appropriate use of the Er:YAG laser on the hard tissue of deciduous teeth. The objective of this study was to identify the most optimal physical settings of the Er:YAG laser for the treatment of carious lesions in the enamel and dentin of deciduous teeth on occlusal, tangential and smooth surfaces. A randomized clinical trial was conducted to compare the parameters of the Er:YAG laser settings for the conservative treatment of deciduous teeth in children with those of the conventional method.

The present study examines the reaction of children to the applied treatment, with a focus on the assessment of pain according to the VAS. The results demonstrated that pediatric patients who underwent laser treatment did not report any pain, despite the absence of local anesthesia. Conversely, the findings indicated that the patients participating in the study perceived the conventional method to be less favorable. The children reported pain, as evidenced by the VAS findings. Other researchers have evaluated the pain experienced during the use of the erbium, chromium:yttrium-scandium-gallium-garnet (Er,Cr:YSGG) laser on permanent teeth in children[24] and during Er:YAG laser treatment on deciduous teeth.[1,25–29]

Table 6. Comparison between the laser method and the traditional method based on the duration of cavity preparation

Variable	Laser method		Conventional method		*t*	*p*-value	95% *CI*		Cohen's *d*
	M	*SD*	*M*	*SD*			lower limit	upper limit	
Time to complete cavity treatment [s]	195.94	30.13	77.67	11.09	30.57	<0.001*	110.39	126.15	5.32

* statistically significant ($p < 0.05$, Student's *t*-test); *CI* – confidence interval.

Table 7. Comparison of the pulp vitality before and after conventional treatment

Variable	Before treatment		After treatment		*t*	*p*-value	95% *CI*		Cohen's *d*
	M	*SD*	*M*	*SD*			lower limit	upper limit	
Pulp vitality	25.88	0.82	32.30	2.01	−21.76	<0.001*	−7.03	−5.82	3.79

* statistically significant ($p < 0.05$, Student's *t*-test); *CI* – confidence interval.

The study also evaluated the time duration required for the laser treatment and the conventional treatment. The results indicated that operating the laser required approx. 2.5 more time than the conventional method. Nevertheless, the children aged 3–8 years who participated in the study found the laser method more acceptable.

In conclusion, in accordance with the results of preceding studies, the preparation time for laser treatment is approximately twice as long as that for rotary instruments. However, for pediatric patients, the duration of treatment is not the primary concern; rather, pain is the most important factor contributing to uncooperative behavior. Other studies have demonstrated that 70% of children felt more comfortable during laser cavity preparation. In comparison, a mere 30% of children expressed a similar level of comfort during conventional cavity preparation. The study demonstrated that the utilization of laser during tooth preparation requires more time compared to conventional methods. Nevertheless, children exhibit greater cooperation and calmness during laser treatment, leading to their preference for this method in subsequent visits.[27,30,31] The aforementioned advantages of lasers in pediatric dentistry result in a less traumatic experience for young patients, thereby enhancing their acceptance of the procedure.

Eren et al. evaluated the duration of laser cavity preparation and compared it to conventional methods in sixth molars in patients aged 7–10 years.[26] The laser preparation took an average of 7.4 min, while mechanical preparation took an average of 3.7 min. Liu et al. also investigated the duration of laser cavity preparation and compared it to conventional methods for deciduous teeth preparation.[29] The working time with the Er:YAG laser was approx. 2.35 times longer than that required for cavity preparation using conventional methods.

Laser tooth preparation entails the minimally invasive and selective removal of infected carious tissue from cavities. The utilization of laser techniques in hard tissues yields a minimal thermal effect or a complete lack of thermal effect in the pulp of teeth undergoing preparation.[31–34] This topic was also addressed in our work, wherein we employed the pulp tester to examine the pulp of the teeth under consideration. The results established that the laser technique does not have an influence on the condition of the pulp. In contrast, the utilization of rotary instruments has been demonstrated to enhance pulp viability following mechanical pulp treatment.

In the present study, a frequency of 10 Hz was employed for both enamel and dentin preparation, while 20 Hz was used exclusively for enamel modification. Upon analysis of other research papers and the frequency values cited therein, a significant discrepancy was observed, ranging from 2 Hz[35] to 20 Hz[8,36] and 25 Hz.[37] The researchers did not differentiate between the values of the applied frequency according to the work in enamel and dentin. In the context of laser applications, the frequency values of the Er,Cr:YSGG laser ranged from 20 Hz to 25 Hz,[22,30,36] while those of the Er:YAG laser ranged from 2 Hz to 10 Hz.[29,35,38] A review of the literature failed to reveal any rationale behind the significantly higher frequencies employed when using the Er,Cr:YSGG laser compared to the Er:YAG laser. Furthermore, none of the other studies analyzed such a relationship.

The frequency value appears to be important in the context of hard dental tissues. The operational speed of the Er:YAG laser is influenced not only by the laser beam parameters and the operator's expertise but also by the chemical composition of the hard dental substance to be treated (enamel, dentin). Enamel is composed of 95% hydroxyapatite, 4% water and 1% collagen fibers, while dentin is composed of 70% hydroxyapatite, 20% collagen fibers and 10% water.[39] The hard tissue of the tooth affected by caries contains more water than healthy tissue, which means that the speed of laser treatment in dentin is higher than that in enamel. This allows the caries tissue to be removed more easily and quickly than healthy tissue. It is essential to control the parameters during laser cavity treatment in accordance with the principles of minimally invasive dentistry. When working in enamel, the frequency parameters may exceed those employed in dentin, particularly within the region of the intraosseous wall.[37,40]

Another parameter that was assessed was the diameter of the applicator used. The diameter of 600 μm (C600F) was identified as the most frequently used applicator. The diameter of 400 μm (C400F) was primarily employed for cavity opening and enamel working, whereas the diameter of 800 μm (C800F) was utilized for dentin treatment in cavities with a larger surface area. The present study delineated the area and type of tooth on which the applicator was utilized in detail. In each case, it was stated whether the contact or non-contact treatment was used, the movements executed during the treatment, the angle at which the applicator was positioned in relation to the tissue being treated, and the type of cavity and tooth treated. Observations revealed that the pulse energy was higher when working in enamel and lower when working in dentin.

A review of the available research papers[21,29,30,35,36,38,40–43] revealed that it is not possible to ascertain which parameters were employed to treat a specific tooth surface or to remove caries in enamel and dentin. A single paper provided this information, stating the fluence, the applicator type, the diameter, the non-contact mode 1 mm from the target area, the type of teeth (canines), and the surface to be treated (class V cavity).[36] The study did not specify the angle of the applicator in relation to the working surface and the type of movement performed by the operator. The study by Rossi et al. appears to be one of the most accurate among the works evaluated, containing the data necessary for determining the optimal physical settings of the Er,Cr:YSGG laser for working in the enamel and dentin of deciduous teeth in a class V cavity.[36]

The study by Valério et al. employed a 900-µm diameter applicator in a non-contact mode with a focal distance of 7 mm, utilizing a non-contact head.[38] In contrast, Johar et al. used a 600-µm diameter applicator in a non-contact mode, yet the distance from the surface to be treated and the type of applicator were not specified.[30] None of the papers provided information on the specific tissues that the given parameters were used for, nor did they indicate whether the parameters were applied to enamel or dentin of deciduous teeth.

Following the analysis and evaluation of the laser settings in the presented studies, it is evident that despite specifying the diameter of the applicator used,[21,29,30,35,36,38] only 3 publications[31,42,43] described the type of applicator. Furthermore, none of the reviewed studies specified the angle of the applicator in relation to the treated tissue or the type of movement performed during the procedure. All of the evaluated studies used the non-contact mode, with the distance from the target area varying between 1 mm,[21,29,36] 7 mm[37] and 17 mm,[35] with no specification of the length from the working surface.[30] However, the values provided do not elucidate the reason behind the selected distances, such as the rationale behind the choice of 1 mm in one instance and 7 mm or 17 mm in another.

In many studies, the data concerning the applicator type and diameter were described in general terms, without linking them to the specific tissue or tooth type. Several publications, including those by Rossi et al.,[36] Valério et al.,[38] and Johar et al.[30] have explicitly examined primary dentition. However, most of the reviewed studies failed to specify the movements performed while operating in the hard tissue, nor did they indicate the angle of incidence of the laser beam, which are crucial parameters for standardizing laser-based protocols in pediatric restorative dentistry.

The studies on the treatment of permanent teeth (with regard to the type of tissue to be treated) revealed a more detailed treatment algorithm, which was not included in the studies on deciduous teeth. In their description of laser treatment of cavities in permanent and deciduous teeth in children with the Er:YAG laser, Kato et al. provided detailed information on the pulse duration, type, and diameter of the applicator used, as well as the physical parameters of the laser applied to enamel and dentin.[28] However, the authors did not specify the movement of the applicator when working in hard tissue and did not distinguish between working in deciduous and permanent teeth. Consequently, the paper is not suitable for determining the baseline parameters of the physical settings of the laser for any type of tooth.[28]

The analysis by Eren et al. describes the specific head and applicator used and is the only study to specify the movements of the applicator.[26]

Based on the published studies, it can be claimed that the use of erbium lasers is beneficial due to the lack of contact with tooth tissues and the absence of vibration.[26,29,31,34,35,44–46] A significant proportion of children (90%) accept laser tooth treatment, whereas patients aged 10 years and older show no preference between laser and conventional treatment methods.[1,46] The reduction of turbine noise is of particular significance for children, given the importance of comfort during operation.[26,29,31,34,35,44,45,47] In the majority of cases, laser treatment indicates the absence of local anesthesia,[20,24,29–31,34–36,38,41,42] as observed in the present study. In our study, the difficulty of removing carious lesions from the lateral walls of the cavity was not evaluated, as previously described by Valério et al.[38] The removal of carious lesions from the lateral cavity walls is more challenging than conventional methods, as the laser applicator does not function in the same manner as a bur. Future studies should aim to investigate the preparation of infected dentin from the lateral cavity wall.

Upon analysis of data from our study and other research papers, it can be concluded that there is a pressing need for further laboratory and clinical studies. It is imperative that the data included in a research publication adheres to the following protocol: firstly, the laser wavelength employed in the study should be specified; then, the type of the head used, the type of the applicator, the diameter of the applicator, the mode of operation (contact or non-contact), the distance of the applicator from the tissue being treated, and the angle of the applicator's movement should be outlined. Additionally, the position of the applicator in relation to the tissue being treated, the type of movement performed, the type of tissue being treated (enamel or dentin), the type of tooth to be treated (deciduous, immature or mature permanent tooth), the surface of the tooth to be treated, and finally, the physical parameters of the laser (pulse duration, frequency, pulse energy, power, as well as water spray and air cooling) must be specified.

Conclusions

Er:YAG lasers offer a promising alternative to traditional rotary instruments for treating caries in primary teeth. The best results were achieved for enamel preparation using the 600-µm diameter applicator with a pulse energy of 230 mJ, mainly on occlusal and proximal surfaces. For dentin preparation, the optimal outcomes were obtained when using the applicator with the 600-µm diameter, and a pulse energy of 120 mJ, or the 800-µm diameter with a pulse energy of 150 mJ. In the context of the enamel modification procedure, positive results were obtained using the 400-µm applicator at a pulse energy of 50 mJ and the 600-µm applicator at a pulse energy of 70 mJ. The optimal operating frequencies of 10 Hz (enamel and dentin caries) and 20 Hz (enamel modification) were identified, with a water spray volume of 2.5–3.0 mL/min (enamel) and 2.0–2.5 mL/min

(dentin). Despite the extended treatment duration, the reduction in pain and the preservation of pulp vitality make laser treatment a favorable option for pediatric patients. Further research is necessary to refine laser settings and improve the efficiency of this technique.

Trial registration

The trial was registered with the ISRCTN (International Standard Randomised Controlled Trial Number) Registry under the reference No. 45696-OH7CP3.

Ethics approval and consent to participate

The Ethics Committee of Wroclaw Medical University, Poland, approved the study (approval No. KB-547/2021), and informed consent was obtained from all participants, as stipulated in the Declaration of Helsinki.

Data availability

The datasets generated and/or analyzed during the current study are available from the corresponding author on reasonable request.

Consent for publication

Not applicable.

Use of AI and AI-assisted technologies

Not applicable.

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References

1. Evans DJ, Matthews S, Pitts NB, Longbottom C, Nugent ZJ. A clinical evaluation of an Erbium:YAG laser for dental cavity preparation. *Br Dent J*. 2000;188(12):677–679. doi:10.1038/sj.bdj.4800575
2. Ferreira JMS, Pinheiro SL, Sampaio FC, de Menezes VA. Caries removal in primary teeth – a systematic review. *Quintessence Int*. 2012;43(1):e9–e15. PMID:22259813.
3. Walsh LJ, Brostek AM. Minimum intervention dentistry principles and objectives. *Aust Dent J*. 2013;58 Suppl 1:3–16. doi:10.1111/adj.12045
4. Zhegova G, Rashkova M, Rocca JP. Minimally invasive treatment of dental caries in primary teeth using an Er:YAG laser. *Laser Ther*. 2014;23(4):249–254. doi:10.5978/islsm.14-OR-18
5. Golob Deeb J, Reddy N, Kitten T, Carrico CK, Grzech-Leśniak K. Viability of bacteria associated with root caries after Nd:YAG laser application in combination with various antimicrobial agents: An in vitro study. *Dent Med Probl*. 2023;60(4):649–655. doi:10.17219/dmp/171690
6. Golob Deeb J, Smith J, Belvin BR, Lewis J, Grzech-Leśniak K. Er:YAG laser irradiation reduces microbial viability when used in combination with irrigation with sodium hypochlorite, chlorhexidine, and hydrogen peroxide. *Microorganisms*. 2019;7(12):612. doi:10.3390/microorganisms7120612
7. El Mobadder M, Nammour S, Namour M, Namour A, Grzech-Leśniak K. Disinfection potential of 980 nm diode laser and hydrogen peroxide (3%) in "critical probing depths" periodontal pockets: Retrospective study. *Life (Basel)*. 2022;12(3):370. doi:10.3390/life12030370
8. Grzech-Leśniak K, Nowicka J, Pajączkowska M, et al. Effects of Nd:YAG laser irradiation on the growth of *Candida albicans* and *Streptococcus mutans*: In vitro study. *Lasers Med Sci*. 2019;34(1):129–137. doi:10.1007/s10103-018-2622-6
9. Nammour S, El Mobadder M, Maalouf E, et al. Clinical evaluation of diode (980 nm) laser-assisted nonsurgical periodontal pocket therapy: A randomized comparative clinical trial and bacteriological study. *Photobiomodul Photomed Laser Surg*. 2021;39(1):10–22. doi:10.1089/photob.2020.4818
10. Grzech-Leśniak K. Making use of lasers in periodontal treatment: A new gold standard? *Photomed Laser Surg*. 2017;35(10):513–514. doi:10.1089/pho.2017.4323
11. Grzech-Leśniak K, Belvin BR, Lewis JP, Golob Deeb J. Treatment with Nd:YAG laser irradiation combined with sodium hypochlorite or hydrogen peroxide irrigation on periodontal pathogens: An in vitro study. *Photobiomodul Photomed Laser Surg*. 2021;39(1):46–52. doi:10.1089/photob.2019.4775
12. Matys J, Grzech-Leśniak K, Flieger R, Dominiak M. Assessment of an impact of a diode laser mode with wavelength of 980 nm on a temperature rise measured by means of k-02 thermocouple: Preliminary results [in Polish]. *Dent Med Probl*. 2016;53(3):345–351. doi:10.17219/dmp/62575
13. Arnabat-Dominguez J, Del Vecchio A, Todea C, et al. Laser dentistry in daily practice during the COVID-19 pandemic: Benefits, risks and recommendations for safe treatments. *Adv Clin Exp Med*. 2021;30(2):119–125. doi:10.17219/acem/130598
14. El Mobadder M, Nammour S, Grzech-Leśniak Z, Grzech-Leśniak K. Efficacy of the adjunct use of povidone-iodine or sodium hypochlorite with non-surgical management of periodontitis: A systematic review and meta-analysis. *J Clin Med*. 2022;11(21):6593. doi:10.3390/jcm11216593
15. Diaci J, Gaspirc B. Comparison of Er:YAG and Er,Cr:YSGG lasers used in dentistry. *LA&HA*. 2012;1:1–13. https://www.laserandhealthacademy.com/media/objave/academy/priponke/1_13_laha_journal_2012_1a.pdf. Accessed August 15, 2024.
16. Grzech-Leśniak Z, Pyrkosz J, Szwach J, et al. In vitro evaluation of the effect of Er:YAG laser with a fractional PS04 handpiece on microbial biofilm survival. *Dent Med Probl*. 2025. doi:10.17219/dmp/201941
17. Fekrazad R, Gholami GA, Sadr Eshkevari P, Nokhbatolfoghahaei H, Mohaghegh S. Management of recurrent aphthous ulcers with therapeutic Nd:YAG laser, using two different methods. *Dent Med Probl*. 2023;60(3):467–472. doi:10.17219/dmp/147048
18. Pion LA, De Matos LLM, Gimenez T, Palma-Dibb RG, Faraoni JJ. Treatment outcome for dentin hypersensitivity with laser therapy: Systematic review and meta-analysis. *Dent Med Probl*. 2023;60(1):153–166. doi:10.17219/dmp/151482
19. Sabel N. Enamel of primary teeth – morphological and chemical aspects. *Swed Dent J Suppl*. 2012;222:1–77. PMID:22515039.
20. Chowdhary N, Subba Reddy VV. Dentin comparison in primary and permanent molars under transmitted and polarised light microscopy: An in vitro study. *J Indian Soc Pedod Prev Dent*. 2010;28(3):167–172. doi:10.4103/0970-4388.73793
21. Hibst R, Stock K, Gall R, Keller U. Controlled tooth surface heating and sterilization by Er:YAG laser radiation. *Proc SPIE*. 1996;2922:119–161. doi:10.1117/12.260668
22. Yaneva BK, Zagorchev PI, Firkova EI, Glavinkov IT. In vitro study of temperature changes in pulp chamber during root planing procedure using Er:YAG laser. *Folia Med (Plovdiv)*. 2016;58(3):206–210. doi:10.1515/folmed-2016-0022

23. Krmek SJ, Miletic I, Simeon P, Mehicić GP, Anić I, Radisić B. The temperature changes in the pulp chamber during cavity preparation with the Er:YAG laser using a very short pulse. *Photomed Laser Surg*. 2009;27(2):351–355. doi:10.1089/pho.2008.2247
24. Kolberg-Babrzyńska I, Grzech-Leśniak K, Kiryk J, Dominiak M, Matys J. Effects of endodontic retreatment by conventional therapy compared to combined therapy with an Er:YAG laser and photobiomodulation: A randomized clinical trial. *Dent Med Probl*. 2025. doi:10.17219/dmp/188864
25. Korkut E, Gezgin O, Özer H, Şener Y. Evaluation of Er:YAG lasers on pain perception in pediatric patients during caries removal: A split-mouth study. *Acta Odontol Turc*. 2018;35(3):81–86. doi:10.17214/gaziaot.296473
26. Eren F, Altinok B, Ertugral F, Tanboga I. The effect of erbium, chromium:yttrium-scandium-gallium-garnet (Er,Cr:YSGG) laser therapy on pain during cavity preparation in paediatric dental patients: A pilot study. *Oral Health Dent Manag*. 2013;12(2):80–84. PMID:23756423.
27. Genovese MD, Olivi G. Laser in paediatric dentistry: Patient acceptance of hard and soft tissue therapy. *Eur J Paediatr Dent*. 2008;9(1):13–17. PMID:18380525.
28. Kato J, Moriya K, Jayawardena JA, Wijeyeweera RL. Clinical application of Er:YAG laser for cavity preparation in children. *J Clin Laser Med Surg*. 2003;21(3):151–155. doi:10.1089/104454703321895608
29. Liu JF, Lai YL, Shu WY, Lee SY. Acceptance and efficiency of Er:YAG laser for cavity preparation in children. *Photomed Laser Surg*. 2006;24(4):489–493. doi:10.1089/pho.2006.24.489
30. Johar S, Goswami M, Kumar G, Dhillon JK. Caries removal by Er,Cr:YSGG laser and Air-rotor handpiece comparison in primary teeth treatment: An in vivo study. *Laser Ther*. 2019;28(2):116–122. doi:10.5978/islsm.19-OR-08
31. Afkhami F, Ebrahimi H, Aghazadeh A, Sooratgar A, Chiniforush N. Evaluation of the effect of the photobiomodulation therapy on the pain related to dental injections: A preliminary clinical trial. *Dent Med Probl*. 2022;59(3):421–425. doi:10.17219/dmp/133645
32. Wigdor H, Abt E, Ashrafi S, Walsh JT. The effect of lasers on dental hard tissues. *J Am Dent Assoc*. 1993;124(2):65–70. doi:10.14219/jada.archive.1993.0041
33. Paghdiwala AF, Vaidyanathan TK, Paghdiwala MF. Evaluation of erbium:YAG laser radiation of hard dental tissues: Analysis of temperature changes, depth of cuts and structural effects. *Scanning Microsc*. 1993;7(3):989–997. PMID:8146625.
34. Kumazaki M, Toyoda K. Removal of hard dental tissue (cavity preparation) with the Er:YAG laser. *J Jpn Soc Laser Dent*. 1995;6(1):16–24. doi:10.5984/jjpnsoclaserdent.6.16
35. Wanderley RL, Monghini EM, Pecora JD, Palma-Dibb RG, Borsatto MC. Shear bond strength to enamel of primary teeth irradiated with varying Er:YAG laser energies and SEM examination of the surface morphology: An in vitro study. *Photomed Laser Surg*. 2005;23(3):260–267. doi:10.1089/pho.2005.23.260
36. Rossi RR, Aranha AC, Eduardo CP, Ferreira LS, Navarro RS, Zezell DM. Microleakage of glass ionomer restoration in cavities prepared by Er,Cr:YSGG laser irradiation in primary teeth. *J Dent Child (Chic)*. 2008;75(2):151–157. PMID:18647510.
37. Zhang S, Chen T, Ge LH. Evaluation of clinical outcomes for Er:YAG laser application in caries therapy of children [in Chinese]. *Beijing Da Xue Xue Bao Yi Xue Ban*. 2013;45(1):87–91. PMID:23411526.
38. Valério RA, Borsatto MC, Serra MC, et al. Caries removal in deciduous teeth using an Er:YAG laser: A randomized split-mouth clinical trial. *Clin Oral Investig*. 2016;20(1):65–73. doi:10.1007/s00784-015-1470-z
39. Zijp JR, Bosch JJ. Theoretical model for the scattering of light by dentin and comparison with measurements. *Appl Opt*. 1993;32(4):411–415. doi:10.1364/AO.32.000411
40. Kornblit R, Trapani D, Bossù M, Muller-Bolla M, Rocca JP, Polimeni A. The use of Erbium:YAG laser for caries removal in paediatric patients following minimally invasive dentistry concepts. *Eur J Paediatr Dent*. 2008;9(2):81–87. PMID:18605890.
41. Keller U, Raab WH, Hibst R. Pulp reactions during Erbium YAG laser irradiation of hard tooth structure [in German]. *Dtsch Zahnarztl Z*. 1991;46(2):158–160. PMID:1814713.
42. Jacobson B, Asgari A. Restorative dentistry for children using a hard tissue laser. *Alpha Omegan*. 2008;101(3):133–139. doi:10.1016/j.aodf.2008.07.025
43. Sungurtekin E, Oztaş N. The effect of erbium, chromium:yttrium-scandium-gallium-garnet laser etching on marginal integrity of a resin-based fissure sealant in primary teeth. *Lasers Med Sci*. 2010;25(6):841–847. doi:10.1007/s10103-009-0720-1
44. Keller U, Hibst R, Geurtsen W, et al. Erbium:YAG laser application in caries therapy. Evaluation of patient perception and acceptance. *J Dent*. 1998;26(8):649–656. doi:10.1016/s0300-5712(97)00036-5
45. Alia S, Khan SA, Navit S, et al. Comparison of pain and anxiety level induced by laser vs rotary cavity preparation: An in vivo study. *Int J Clin Pediatr Dent*. 2020;13(6):590–594. doi: 10.5005/jp-journals-10005-1820
46. Deeb JG, McCall C, Carrico CK, Dahlke WO, Grzech-Leśniak K. Retrieval of prefabricated zirconia crowns with Er,Cr:YSGG laser from primary and permanent molars. *Materials (Basel)*. 2020;13(23):5569. doi:10.3390/ma13235569
47. Matsumoto K, Wang X, Zhang C, Kinoshita JI. Effect of a novel Er:YAG laser in caries removal and cavity preparation: A clinical observation. *Photomed Laser Surg*. 2007;25(1):8–13. doi:10.1089/pho.2006.2020

Original paper

Cognitive status and periodontitis: Is there a correlation? A cross-sectional study

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Abstract

Background. The Montreal Cognitive Assessment (MoCA) is the most accurate cognitive screening tool for early diagnoses of mild cognitive impairment (MCI). However, the majority of research on the correlation between MCI and periodontitis has been conducted using the Mini–Mental State Examination (MMSE).

Objectives. The study aimed to determine a possible correlation between the gingival index (GI), periodontitis staging and MCI as evaluated by the MoCA in adults aged ≥55 years.

Material and methods. A total of 120 Mexican patients with or without periodontal disease who met the selection criteria were included in this cross-sectional study. A comprehensive clinical and radiographic evaluation was conducted to determine the periodontal status of all patients. The MoCA was applied to assess cognitive impairment. Descriptive statistics and logistic model-based multivariate statistical analyses were performed to identify variables associated with MCI.

Results. The cognitive status of subjects aged ≥55 years with and without periodontitis was mildly impaired in more than half of the studied sample. Thirty-four patients were diagnosed as periodontally healthy (group 1), 48 were diagnosed with mild to moderate periodontitis (group 2; stages I and II), and 38 had severe periodontitis (group 3; stages III and IV); 54% of the subjects exhibited MCI, mostly in groups 2 and 3. A negative correlation was observed between GI and MCI ($p = 0.025$). Other prognostic variables associated with MCI were loss of clinical attachment level (CAL) (odds ratio (*OR*) = 1.68 (95% confidence interval (*CI*): 1.06–2.68)), sex and education level.

Conclusions. These findings highlight the potential clinical relevance of gingival and periodontal parameters, such as GI and CAL, and their association with MCI in patients of advanced age.

Keywords: mental status, neurocognitive tests, Montreal Cognitive Assessment, chronic periodontitis

Highlights

- Gingival inflammation (GI) and clinical attachment loss (CAL) should be assessed in patients aged 55 and older, as these parameters are linked to mild cognitive impairment (MCI) and serve as objective indicators within periodontal care.
- Given the association between periodontitis and cognitive decline, clinicians should consider using cognitive screening tools such as the Montreal Cognitive Assessment (MoCA) for older patients with periodontal disease.
- Females have nearly 3 times greater odds of developing MCI, while higher education levels are associated with a reduced MCI risk.
- Effective management of periodontal disease may reduce the risk of dementia, as severe periodontal conditions or lack of therapy correlate with an increased risk of cognitive decline, underscoring the need for comprehensive periodontal treatment.

Introduction

The process of aging in humans is frequently associated with an increased prevalence of oral diseases, including periodontitis. There is a bidirectional link between periodontitis and ischemic heart disease,[1] pregnancy,[2] myocardial strain,[3] and cognitive changes, including neurodegenerative diseases.[4] The early detection of cognitive dysfunction can be accomplished by the utilization of several screening and evaluation instruments to identify affected individuals.[5–7]

The Montreal Cognitive Assessment (MoCA) is a brief, standardized, and scoring cognitive screening tool that is one of the most widely used screening tests in healthcare settings for detecting cognitive impairments that may not be as readily apparent on other cognitive tests.[5,8,9] It is a freely available tool that includes extensive neuropsychological test batteries covering several cognitive conditions.[7] The global score ranges from 0 to 30 points, and a higher score reflects better cognitive function. The test's sensitivity and specificity for the detection of mild cognitive impairment (MCI) are 90% and 87%, respectively.[7,8,10] Therefore, it is critical to early identify the associated risk factors for this neurological condition to implement opportune therapeutic interventions and prevent complications.[6,11]

Previous reports have linked temporomandibular disorders (TMD), impaired masticatory function[12] and periodontitis to an increased risk of MCI. Additionally, reduced masticatory function,[12] TMD and periodontitis have been reported as risk factors associated with MCI. Temporomandibular disorders are characterized by pain in the temporomandibular joint and masticatory muscles, which decreases chewing efficiency. Chewing is a constant sensory stimulus for the brain. Impaired memory functions in the hippocampus, masticatory muscle loss and weakness are associated with systemic and central inflammation and dementia.[12] On the other hand, mastication activates various cortical regions of the brain, and increased cerebral blood oxygenation levels in the hippocampus and prefrontal cortex facilitate learning and memory processes.[13] Periodontitis causes structural damage to the tissues surrounding the alveolar bone and periodontal ligament collagen fiber-affected teeth, leading to tooth loss.[14] The host response is a complex interplay between numerous cells and inflammatory mediators.[14,15] The host response to periodontal bacteria may induce chronic systemic inflammation, which, in turn, stimulates the production of high levels of inflammatory mediators (P-selectin, receptor activator of nuclear factor kappa-B ligand (RANKL), intercellular adhesion molecule 1 (ICAM-1)), cytokines (tumor necrosis factor alpha (TNF-α), interferon gamma (INF-γ), interleukin (IL)-1, IL-6, prostaglandin E_2 (PGE_2)), prostanoids, and matrix metalloproteinases (MMPs).[15] The evidence suggests that these inflammatory mediators reach all systems through blood circulation.[15] Studies have proposed that peripheral inflammation and dysbiotic conditions contribute to the pathogenesis of MCI.[14,15] Epidemiological data has demonstrated an association between the systemic inflammatory influence of periodontal disease and an increase in the neuroinflammatory response, which can result in cognitive impairment in elderly individuals.[15] These studies have reported odds ratios (*OR*s) and hazard risk ratios within the mild to moderate range.[11,16] Other studies,[14,16–23] including 3 systematic reviews,[14,16,17] have also suggested a relationship between periodontitis and cognitive impairment.

The evidence presented thus far suggests that periodontal disease modulates neuroinflammation and neurodegeneration, which may result in cognitive impairment. Therefore, clinicians must objectively assess the medical comorbidities of their patients, including possible cognitive dysfunctions.[24] The present analytical cross-sectional study aimed to find a correlation between the gingival index (GI) staging, grading of periodontitis and MCI, as evaluated through the MoCA in Mexican adults aged ≥55 years.

Material and methods

Study population

This cross-sectional study was approved by the Institutional Ethics Committee of the Faculty of Dentistry at the Autonomous University of San Luis Potosí in Mexico (approval No. CEI-FE-036-021). Individuals attending the outpatient Periodontics Clinic (Periodontics Postgraduate Program, University of San Luis Potosí) from October 2022 to June 2023, who met the inclusion criteria, were consecutively selected for the study (non-probability sampling). Prior to enrollment, written informed consent forms were obtained from all participants.

The inclusion criteria were as follows: participants aged 55 years or older with at least 10 natural teeth present; individuals with or without periodontal disease, at different stages of severity, diagnosed through a full-mouth clinical and radiographic examination at baseline. Patients were excluded from the study if they had a history of severe cranial trauma, cerebrovascular disease, chronic headache or migraine, neurodegenerative conditions, brain tumors, chronic subdural hematoma, cryptococcosis, pellagra, hypothyroidism, uncontrolled hypertension, current medication likely to affect the cognition level, or oral acute infectious process.

The demographic information collected encompassed various parameters, such as age, sex, education level, diet, smoking or alcohol consumption, and medical history.

Clinical and radiographic intraoral examination

The number of present teeth and the degree of tooth mobility were recorded. Additionally, the GI was determined according to the Löe–Silness criteria.[25] Then, an experienced and precalibrated periodontist documented the probing pocket depth (PPD) and clinical attachment level (CAL) with the use of a North Carolina periodontal probe. These procedures were performed at 6 sites around each tooth (mesiobuccal, buccal, distobuccal, mesiolingual/palatine, lingual/palatine, and distolingual/palatine). Probing pocket depth was measured as the distance from the gingival margin to the base of the pocket. The CAL was determined by measuring the distance from the cementoenamel junction to the base of the pocket. The pocket's base was established at the level where the periodontal probe encountered maximum resistance.

The presence of bleeding was evaluated through careful probing around each present tooth, and the total percentage of bleeding sites was recorded.[26] Additionally, periapical radiographs were taken in all subjects to assess bone height loss and the pattern of loss, as well as other periodontally relevant features, such as loss of cortical and crest bone continuity, widening of the periodontal ligament space, calculus deposits, and periapical/furcation lesions.[26]

Each subject was diagnosed according to the periodontal parameters described in the current classification of periodontal diseases[26]: CAL; PPD; the percentage of bone loss; the presence and extent of angular bony defects and furcation involvement; tooth mobility; and tooth loss due to periodontitis. In addition, periodontal disease was categorized into 3 grades according to the risk of disease progression, the anticipated treatment response, and the impact on systemic health,[26] as follows: grade A – low risk; grade B – moderate risk; and grade C – high risk. This evaluation includes the assessment of the patient's general health status and the presence of additional risk factors, such as smoking or diabetes mellitus. The staging of periodontitis was determined as follows: stage I – initial periodontitis (no tooth loss), CAL: 1–2 mm; stage II – moderate periodontitis (no tooth loss), CAL: 3–4 mm; stage III – severe periodontitis with potential for additional tooth loss (≤4 lost teeth), CAL ≥ 5 mm; and stage IV – advanced periodontitis with extensive tooth loss and potential for loss of dentition (≥5 lost teeth), CAL ≥ 5 mm. The CAL was measured interdentally at the site of the greatest loss. The control group consisted of patients with a confirmed absence of periodontal disease, as determined by clinical and radiographic examination.

Cognitive assessment

A complete cognitive evaluation was conducted for each participant. The validated Spanish version of the MoCA tool[7,8,10] was used to assess the presence of cognitive impairment among the study participants. The test took approx. 10 min to complete and evaluated multiple cognitive skills. Following the intraoral examination, the MoCA test was administered to each patient. The participant was provided with detailed instructions as they proceeded with the test. Ultimately, a single trained evaluator (AMG) interpreted the individual results. The MoCA score ranges from 0 to 30 points. The severity of cognitive impairment was classified according to the final score, as follows: 10–17 points – moderate; 18–26 points – mild; and >26 points – absent.

Evaluator calibration and statistical analysis

To ensure the quality of the measurement of clinical and radiographic periodontal variables, the evaluator (AMG) was calibrated with an expert (interobserver agreement) through kappa statistic (0.82). With regard to the performance of the MoCA tool, the same evaluator was previously trained and duly certified (MoCA Certificate of Completion, 2021/10/07).

A preliminary descriptive analysis was conducted on the collected data. It incorporated the calculation of various statistical measures, including frequencies, means and standard deviations, medians, interquartile ranges, ranges, and percentages. The distribution of the data was evaluated

using the Shapiro–Wilk test. Based on the results, diverse statistical hypothesis tests, both parametric and non-parametric, were carried out for quantitative and categorical data. Initially, exploratory correlation analyses were conducted between quantitative variables, reporting Pearson's correlation coefficients (r and corresponding p-values), but only of those with significant correlation. Then, multiple binary logistic regression analyses were performed, incorporating diverse prognostic variables in the models. The approach yielded respective *OR*s (95% confidence intervals (*CI*s)), which were subsequently used to predict MCI. The statistical analysis was conducted using the GraphPad Prism v. 8 (GraphPad Software, Boston, USA) and STATA v. 14 (StataCorp LLC, College Station, USA) software. More than 80% of the power was achieved, indicating an adequate sample size to contrast the hypothesis of the association between the periodontal status and MCI.[24] A statistically significant value of 0.05 was set.

Results

Demographic characteristics of the participants

A total of 120 participants were included in the study; 50% were female, and 50% were male, with a mean age of 63.34 ±4.86 years. Their median age was 59.5 years (interquartile range: 55–95 years). Regarding education level, 46% of the participants had finished undergraduate studies, while only 6% obtained a postgraduate degree. Patients were divided into 3 groups according to their periodontal characteristics. The control group (group 1, n = 34) comprised periodontally healthy individuals (<10% bleeding on probing and PPD < 4 mm), with or without minimal insertion loss. The second group (group 2, n = 48) consisted of patients diagnosed with stage I and II (moderate) periodontitis, grades A–C. The third group (group 3, n = 38) consisted of patients diagnosed with stage III and IV (severe) periodontitis, grades A–C (Table 1).

There were 15% smokers in the sample, with the majority (20%) belonging to group 2. Regarding the body mass index (BMI), it was observed that all 3 groups were overweight. Additionally, the severity of periodontal disease increased along with the BMI. A comparison of the BMI among the 3 study groups revealed no statistically significant differences (p = 0.268). Regarding comorbidities, 68% of the participants exhibited one or more systemic diseases, with the highest prevalence observed among individuals with stage III and IV periodontitis (84%). Thirty different types of diseases were identified. The most prevalent conditions were arterial hypertension (33%; predominantly observed in group 3) and type 2 diabetes mellitus (20%). The occurrence of benign prostatic hyperplasia, hypothyroidism and depression was also documented. Furthermore, 74 different types of medications were reported, with metformin, losartan, enalapril, and omeprazole being the most prevalent.

Periodontal characteristics

The clinical parameters (number of teeth, bleeding sites, bleeding on probing, PPD, GI, and CAL) were described and compared between the study groups (Table 2). Significant differences were identified between the groups. The severity of these parameters increased concomitantly with the severity of periodontitis, thereby validating the periodontal diagnosis and classification.

Cognitive assessment

Fifty-four percent of the participants exhibited MCI, with a cut-off point of 23, as established by Thomann et al.[7] When the sample was divided according to the periodontal status, the percentage of MCI was obsrved to increase with the severity of periodontitis. Patients in groups 2 and 3 showed a mild degree of cognition, while the overall score of healthy patients corresponded to a normal cognitive level. However, no statistically significant differences were identified between the groups (p > 0.05) (Table 3).

Table 1. Demographic characteristics of the study sample according to the periodontal status

Variable		Group 1 (n = 34)	Group 2 (n = 48)	Group 3 (n = 38)	Total (N = 120)	p-value
Age [years] *Me* (*IQR*)		56.5 (55–64)	60 (56–65)	60 (56–67)	59.5 (55–65)	0.308[#]
Sex, n (%)	female	20 (59)	27 (56)	13 (34)	60 (50)	0.062[##]
	male	14 (41)	21 (44)	25 (66)	60 (50)	
Education level, n (%)	elementary	12 (35)	12 (25)	12 (32)	36 (30)	0.491[##]
	high school	7 (21)	11 (23)	8 (21)	26 (22)	
	bachelor	15 (44)	22 (46)	18 (47)	55 (46)	
	postgraduate	0 (0)	3 (6)	0 (0)	3 (2)	

group 1 – periodontally healthy individuals (<10% bleeding on probing and probing pocket depth (PPD) <4 mm); group 2 – patients diagnosed with stage I and II (moderate) periodontitis; group 3 – patients diagnosed with stage III and IV (severe) periodontitis; *Me* – median; *IQR* – interquartile range; [#] Kruskall–Wallis test; [##] χ^2 test.

Table 2. Periodontal parameters of the study sample according to the periodontal status

Variable	Group 1 (*n* = 34)	Group 2 (*n* = 48)	Group 3 (*n* = 38)	*p*-value
GI *Me* (*IQR*)	0.995 (0.69–1.30)	1.13 (0.76–1.82)	1.48 (1.20–2.46)	0.001*
CAL *Me* (*IQR*)	2.58 (2.28–2.83)	2.88 (2.68–3.30)	3.92 (3.51–5.20)	0.001*
Bleeding sites *Me* (*IQR*)	28.50 (17.00–48.00)	33.00 (23.00–58.00)	46.00 (33.00–70.00)	0.014*
Bleeding on probing *M* ±*SD*	0.26 ±0.17	0.28 ±0.18	0.44 ±0.22	0.000*
PPD *Me* (*IQR*)	2.12 (2.08–2.29)	2.53 (2.41–2.77)	3.18 (2.95–3.70)	0.000*
Number of teeth *Me* (*IQR*)	23.50 (18.00–26.00)	25.00 (22.00–26.00)	21.50 (15.00–24.00)	0.001*

group 1 – periodontally healthy individuals (<10% bleeding on probing and PPD < 4 mm); group 2 – patients diagnosed with stage I and II (moderate) periodontitis; group 3 – patients diagnosed with stage III and IV (severe) periodontitis; GI – gingival index; CAL – clinical attachment level; *M* – mean; *SD* – standard deviation; * statistically significant ($p < 0.05$, Kruskall–Wallis test).

Table 3. Cognitive assessment of the study sample according to the periodontal status

Variable	Group 1 (*n* = 34)	Group 2 (*n* = 48)	Group 3 (*n* = 38)	Total (*N* = 120)	*p*-value (one-way ANOVA)
MoCA score *M* ±*SD*	23.15 ±3.23	22.98 ±3.52	22.76 ±3.30	22.95 ±3.33	0.889
MCI, *n* (%)	18 (53)	26 (54)	21 (55)	65 (54)	0.981

group 1 – periodontally healthy individuals (<10% bleeding on probing and PPD < 4 mm); group 2 – patients diagnosed with stage I and II (moderate) periodontitis; group 3 – patients diagnosed with stage III and IV (severe) periodontitis; MoCA – Montreal Cognitive Assessment; MCI – mild cognitive impairment.

Cognitive status and gingival index

Possible relationships between different gingival and periodontal variables and MCI, as evaluated by the MoCA, were determined. Only the GI showed a significant negative correlation with MCI ($p = 0.025$) (Fig. 1).

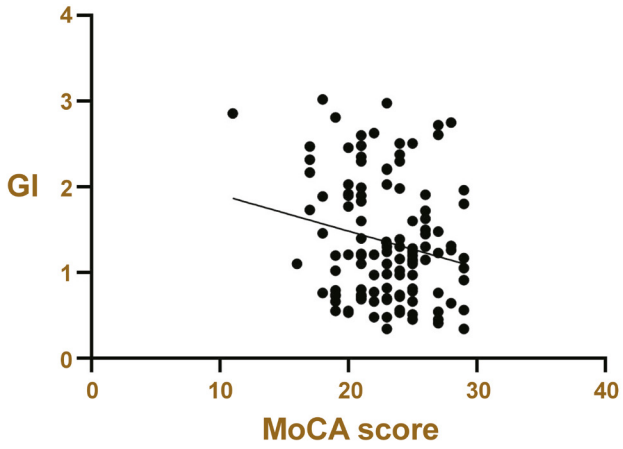

Fig. 1. Correlation between the gingival index (GI) and the cognitive state of study participants

MoCA – Montreal Cognitive Assessment.

In light of these findings, MoCA scores were obtained for all participants, who were then classified into 3 groups according to the extent of inflammation: mild (23.28 ±3.03); moderate (23.29 ±3.22); and severe (21.50 ±2.86).

A correlation was identified between the GI and the MoCA score (coefficient = –1.01, $p = 0.032$) with adjustments made for age, sex, BMI, education level, and comorbidities (arterial hypertension, diabetes mellitus and depression) (Table 4). Furthermore, the likelihood of presenting MCI in cases of severe gingival inflammation was 3.03 ($p = 0.062$), and the individuals with a high level of education showed a 63% decrease in the likelihood of developing MCI compared to those with a low level of education (*OR* = 0.37, 95% *CI*: 0.15–0.86, $p = 0.021$) (Table 5).

Cognitive status according to sex and age

Based on the MoCA results, female patients with periodontal disease exhibited a greater tendency for developing MCI than males. However, this difference was not statistically significant. The participants were divided

Table 4. Multivariate linear regression analysis of the association between the Montreal Cognitive Assessment (MoCA) score and the gingival index (GI)

Variable		Coefficient	95% *CI*	Robust standard error	*t*	*p*-value
GI		–1.01	–1.93––0.09	0.47	–2.17	0.032*
Female sex		–0.77	–1.92–0.38	0.58	–1.31	0.186
Age		–0.07	–0.18–0.04	0.06	–1.33	0.193
Education level		1.00	–0.23–2.24	0.62	1.61	0.110
BMI	normal weight	–	–	–	–	–
	overweight and obesity grade I	–0.79	–2.23–0.65	0.73	–1.09	0.279
	obesity grades II and III	–1.00	–2.63–0.61	0.82	–1.23	0.221
Comorbidities	diabetes mellitus	1.15	–0.46–2.76	0.81	1.41	0.161
	arterial hypertension	0.17	–1.03–1.37	0.61	0.28	0.782
	depression	2.04	–0.97–4.29	1.13	1.81	0.073

BMI – body mass index; *CI* – confidence interval; * statistically significant ($p < 0.05$).

into 3 age groups (55–59, 60–74, and ≥75 years). Lower MoCA scores were observed among the oldest participants. The presence of MCI was detected only in patients over the age of 60.

Association of mild cognitive impairment with periodontal parameters and other variables

Another logistic regression model for the occurrence of MCI was conducted, with adjustments made for the number of teeth, age, BMI, sex, comorbidities (arterial hypertension, diabetes mellitus, depression), and education level. Participants with loss of CAL exhibited a 68% higher possibility of developing MCI than participants without loss of CAL (*OR* = 1.68, 95% *CI*: 1.06–2.68, $p = 0.027$). Female participants were 2.97 times more likely to have MCI than male subjects (*OR* = 2.97, 95% *CI*: 1.24–7.14, $p = 0.015$) (Table 6).

Discussion

This cross-sectional study aimed to determine the cognitive state evaluated by the MoCA in Mexican adults aged ≥55 years, with or without periodontal disease. A contemporary classification system for periodontal diseases was implemented, encompassing the general status of each patient and the presence of comorbidities or risk factors.[25–29] Consequently, it was determined that more than 50% of the subjects exhibited MCI. This finding can be explained by the presence of chronic inflammation caused by periodontal disease and the production of inflammatory mediators and bacterial products. These factors may exacerbate neurodegeneration processes systemically or trigeminally, favoring the presence of MCI.[20,21] However, in periodontally healthy subjects, MCI was present in more than half of the participants, which may be the result of other risk factors associated with systemic inflammation comorbidities, such as obesity, smoking and genetic alterations.[15,30,31]

Table 5. Multivariate logistic regression analysis of the association between mild cognitive impairment (MCI) and the severity of gingival inflammation

Variable		Adjusted *OR*	95% *CI*	Standard error	*z*	*p*-value
GI (inflammation level)	mild	1.00	–	–	–	–
	moderate	0.78	0.31–1.93	0.36	–0.54	0.611
	severe	3.03	0.92–10.0	1.85	1.82	0.062
Sex	male	1.00	–	–	–	–
	female	2.16	0.95–4.93	0.91	1.84	0.066
Age		1.02	0.96–1.09	0.03	0.72	0.469
Education level		0.37	0.15–0.86	0.16	–2.31	0.021*
BMI	normal weight	1.00	–	–	–	–
	overweight and obesity grade I	1.46	0.54–3.94	0.74	0.74	0.456
	obesity grades II and III	1.29	0.43–3.85	0.72	0.45	0.652
Comorbidities	diabetes mellitus	0.42	0.13–1.33	0.25	–1.46	0.114
	arterial hypertension	0.96	0.37–2.49	0.47	–0.08	0.938
	depression	0.21	0.04–1.20	0.19	–1.75	0.080

OR – odds ratio; * statistically significant ($p < 0.05$). A cut-off point of 23 was determined, as established by Thomann et al.[7]

Table 6. Multivariate logistic regression analysis of the association between mild cognitive impairment (MCI) and periodontal parameters

Variable		Adjusted *OR*	95% *CI*	Standard error	*z*	*p*-value
CAL		1.68	1.06–2.68	0.34	2.22	0.027*
Number of teeth		1.09	0.97–1.22	0.63	1.49	0.137
Sex	male	1.00	–	–	–	–
	female	2.97	1.24–7.14	1.33	2.44	0.015*
Age		1.02	0.96–1.10	0.03	0.82	0.410
Education level		0.39	0.17–0.92	0.17	–2.16	0.031*
BMI	normal weight	1.00	–	–	–	–
	overweight and obesity grade I	1.56	0.57–4.25	0.80	0.86	0.388
	obesity grades II and III	1.27	0.43–3.77	0.70	0.43	0.664
Comorbidities	diabetes mellitus	0.41	0.14–1.20	0.22	–1.63	0.103
	arterial hypertension	0.91	0.37–2.25	0.42	–0.19	0.846
	depression	0.15	0.02–1.12	0.16	–1.84	0.065

* statistically significant ($p < 0.05$). A cut-off point of 23 was determined, as established by Thomann et al.[7]

The evidence demonstrates that severe gingival inflammation and/or chronic periodontitis are peripheral sources of inflammatory cytokines. The cytokines increase the levels of C-reactive protein, which favors the activation of microglial cells and the production of misfolded proteins. These proteins are biomarkers that contribute to the occurrence of dementia and accelerate neurodegeneration.[11,31–33] Consequently, subjects with severe gingival inflammation show a greater tendency to MCI compared to those with a mild or moderate GI. Likewise, a significant negative correlation was observed between the MoCA score and the level of gingival inflammation, suggesting that reduced gingival inflammation is associated with enhanced cognitive function. This finding indicates that gingival inflammation may contribute to the development of MCI, which is consistent with the results of studies by Hategan et al.[11] and Stewart et al.,[34] who found a positive correlation between inflammation, IL-1β levels and MCI. Moreover, a logistic regression model implemented in our study demonstrated a tendency toward MCI in cases of severe gingival inflammation. In this regard, Shin et al. stated in their cohort study that the presence of chronic periodontal inflammation (with severe gingivitis and CAL) is not sufficient to observe its positive association with cognitive decline.[16] A period of persistent inflammation is also necessary for the cognitive status to be affected.

In the present study, a negative correlation was observed between MCI and the GI. A case–control study conducted on patients with active periodontitis reported significant cognitive impairment over a period of 6 months compared to patients with inactive periodontitis.[35] This evidence suggests that the level of disease activity, the amount of bone destruction, inflammation, and exposure time are potential factors that activate neurodegeneration and trigger MCI. Yoneyama et al. found that there was an increase in the Mini–Mental State Examination (MMSE) test scores in a 24-month study of oral care in older adults.[36] The MMSE is a brief screening tool commonly used to assess dementia, orientation, memory, attention, language, and visuospatial skills. These findings demonstrated that reducing periodontal inflammation can reverse the progression of MCI.[36] In addition, clinical and animal studies have reported that the administration of paracetamol, ibuprofen, and nonsteroidal anti-inflammatory drugs (NSAIDs) is associated with a significant reduction in IL-1β, IL-6, TNF-α, and PGE_2, improved cognitive function, and reduced systemic inflammation.[37–40] It can thus be inferred that neuroinflammation is related to inflammatory systemic states, and that the modification of the related processes could have a positive effect on cognitive function.

Concerning clinical attachment loss, which is equally essential to the diagnosis of periodontitis, the present study identified a positive association between clinical attachment loss and MCI, with an *OR* of 1.68 (95% *CI*: 1.06–2.68). Subjects with evident loss of CAL were 68% more likely to develop MCI than those without CAL loss. The progression of clinical attachment loss is contingent upon the stage of periodontal disease. Therefore, the longer the periodontal disease, the greater the loss and the subsequent development of chronic neuroinflammation and MCI. The phenomenon of cognitive deterioration is preceded by a loss of insertion. Furthermore, Gil Montoya et al., in their case–control study, used cognitive tests ("Phototest") in conjunction with 29 inflammatory biomarkers to determine the level of peripheral inflammation and to establish a potential association between periodontal disease and MCI or dementia.[18] The authors demonstrated that clinical attachment loss was significantly associated with MCI, reporting an *OR* of 2.97 (95% *CI*: 1.61–5.48).[18]

Other periodontal variables have been linked to MCI. In the current study, as the severity of periodontitis increased, a greater tendency toward the condition was observed. Periodontally healthy subjects showed no impairment (with an average MoCA score of 23.15 points) compared to groups with periodontitis. However, this difference was not statistically significant, possibly due to the cross-sectional design of the study. The study design is subject to limitations, particularly due to constraints in sample size and the measurement of all variables at a single time point. Nevertheless, robust evidence supports a link between periodontal indicators, specifically the GI and CAL, and cognitive deterioration.[4,11,14–16]

This outcome is consistent with the results of the study by Hategan et al., which revealed that subjects diagnosed with aggressive periodontitis had lower MoCA scores compared to those with mild periodontitis or periodontally healthy subjects.[11] A lower test score was also observed in subjects with moderate periodontitis compared to those without periodontal disease. Two other studies have reached similar conclusions. These studies were conducted in 2021 by Hu et al.[14] (using a cohort design) and Guo et al.[41] through a systematic review/meta-analysis. On the other hand, a cross-sectional study conducted by Naorungroj et al. revealed no significant association between severe periodontitis and MCI.[42]

In their meta-analysis, Guo et al. included 20 observational studies.[41] When periodontitis and cognitive impairment were associated, the *OR* was 1.77 (95% *CI*: 1.31–2.38), indicating a strong relationship between the 2 conditions. However, the statistical analysis revealed no significant effect of periodontitis on dementia, with an *OR* of 1.59 (95% *CI*: 0.92–2.76). Additionally, Guo et al. stated that the MMSE is less sensitive and specific than the MoCA, and that it could generate false positive results. In addition, they considered the MMSE as a test designed specifically for dementia and not for MCI, as the MoCA is.[41] In this regard, and according to Nasreddine et al.,[8] the MoCA is specifically designed to detect subtler cognitive deficits, which characterize MCI, earlier and more accurately.

Multiple chronic comorbidities, such as high blood pressure, hyperlipidemia, coronary heart disease, or heart attacks, are considered high-risk prognostic factors for the development of MCI. Depression, diabetes mellitus and chronic obstructive pulmonary disease have been identified as moderate risk factors.[15,18,32,43] Several chronic conditions were present in the studied sample, which were subsequently incorporated into multivariate regression models. However, a significant association could not be demonstrated in all 3 study groups between hypertension and type 2 diabetes mellitus with MCI. Both diseases are prevalent among the Mexican population and are considered major contributors to mortality after the age of 50.[44] No association was identified between depression and MCI.

Regarding the variables of age, sex and education level, a positive association was identified between MCI and female sex (*OR* = 2.97 (95% *CI*: 1.24–7.14)). In postmenopausal women, the amount of estrogen, a vital component of cognitive function, decreases. Estrogens increase synaptic properties in neurons, act as antioxidants to protect neurons from free radicals, improve mitochondrial function in the brain, and induce DNA repair. Our results are consistent with several previous reports.[40] The logistic regression model did not demonstrate a correlation between the age of the participants and MCI. However, as expected, a decline in performance was evident with increasing age, particularly in the 75–90 age group. This decline was not statistically significant. Cecato et al. reported analogous results in their cross-sectional study of 136 elderly participants with more than 4 years of education.[45] The current study found a positive association between the education level and MCI (*OR* = 0.39 (95% *CI*: 0.17–0.91)), in concordance with the study of Langa and Levine.[46]

It is important to note that a rapid growth of dementia usually occurs in subjects of advanced age. Currently, there is no known cure for this disease. Therefore, it is necessary to apply diagnostic tests during the early stages of the condition, encompassing electrophysiological studies, magnetoencephalography, and magnetic resonance spectroscopy.[47–49] However, these technologies are not available in a traditional dentist's office. The present study claims that the application of the MoCA in periodontitis-affected patients may facilitate the identification of an association with their cognitive status.

According to recent research, the management of periodontal disease may play a pivotal role in the prevention of cognitive impairment. In a recent retrospective cohort study, Lee et al. examined the extent and temporal aspect of the relationship between periodontal disease, dental health and dementia.[50] The participants were assigned to one of the 4 study groups, namely dental prophylaxis, intensive periodontal therapy, tooth extraction, and no treatment at all. Next, a comparison was made between the groups' dementia incidence rates. The study found that the incidence of dementia was notably higher in the groups that did not receive treatment (0.76% annually) and those that underwent tooth extraction (0.57% annually) compared to the groups that received dental prophylaxis (0.39% annually) and underwent intensive periodontitis treatment (0.35% annually). The risk of dementia was higher in subjects with more severe periodontal disease or in those who did not receive any periodontal therapy.

Strengths and limitations

This study, conducted in Mexico, is among the first to demonstrate a connection between periodontitis and MCI using the MoCA. The Montreal Cognitive Assessment is more precise and accurate in detecting MCI compared to the widely used the MMSE test. In addition, the test was administered by a researcher who had received certification for its application and interpretation of subsequent results. Another strength of the study is the implementation of a new and improved classification of periodontal diseases, as proposed by Tonetti et al.[26] and Caton et al.[30]

The study's main limitations were the limited sample size and the narrow range of selection criteria chosen. The following risk factors associated with MCI were not considered: lifestyle; intake of NSAIDs; family history of cognitive dysfunction; socioeconomic status; and sedentary lifestyle. There was no strict control of potential confounding variables. The authors propose the execution of future longitudinal clinical studies of higher methodological quality, with the objective of establishing whether periodontal treatment modifies the MoCA score.

Conclusions

The findings reported in the present cross-sectional study highlight the potential clinical relevance of gingival and periodontal parameters, such as GI and clinical attachment loss, and their association with MCI. A comprehensive periodontal evaluation is imperative for patients aged ≥55 years old, as it can serve as an objective indicator of their cognitive status within the context of periodontal care.

Ethics approval and consent to participate

The study was approved by the Institutional Ethics Committee of the Faculty of Dentistry at the Autonomous University of San Luis Potosí in Mexico (approval No. CEI-FE-036-021). Prior to enrollment, written informed consent forms were obtained from all participants.

Data availability

The datasets generated and/or analyzed during the current study are available from the corresponding author on reasonable request.

Consent for publication

Not applicable.

Use of AI and AI-assisted technologies

Not applicable.

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References

1. Ghorbani P, Rezaei Esfahrood Z, Foroughi M. Paraoxonase-1, a novel link between periodontitis and ischemic heart disease: A case–control study. *Dent Med Probl*. 2023;60(1):55–59. doi:10.17219/dmp/152181
2. Waligóra J, Kuśnierz-Cabala B, Pawlica-Gosiewska D, Gawlik K, Chomyszyn-Gajewska M, Pytko-Polończyk J. Salivary matrix metalloproteinase-9 (MMP-9) as a biomarker of periodontitis in pregnant patients with diabetes. *Dent Med Probl*. 2023;60(1):35–45. doi:10.17219/dmp/147104
3. Narendran N, Shenoy S, Kodangala S, Raghavendra Vamsi A, Kamath V. Assessment of myocardial strain in hypertensive patients with periodontitis. *Dent Med Probl*. 2023;60(1):61–69. doi:10.17219/dmp/152707
4. Dziedzic A. Is periodontitis associated with age-related cognitive impairment? The systematic review, confounders assessment and meta-analysis of clinical studies. *Int J Mol Sci*. 2022;23(23):15320. doi:10.3390/ijms232315320
5. Ciesielska N, Sokołowski R, Mazur E, Podhorecka M, Polak-Szabela A, Kędziora-Kornatowska K. Is the Montreal Cognitive Assessment (MoCA) test better suited than the Mini-Mental State Examination (MMSE) in mild cognitive impairment (MCI) detection among people aged over 60? Meta-analysis. *Psychiatr Pol*. 2016;50(5):1039–1052. doi:10.12740/PP/45368
6. Tsoi KKF, Chan JYC, Hirai HW, Wong SYS, Kwok TCY. Cognitive tests to detect dementia: A systematic review and meta-analysis. *JAMA Intern Med*. 2015;175(9):1450–1458. doi:10.1001/jamainternmed.2015.2152
7. Thomann AE, Berres M, Goettel N, Steiner LA, Monsch AU. Enhanced diagnostic accuracy for neurocognitive disorders: A revised cut-off approach for the Montreal Cognitive Assessment. *Alzheimers Res Ther*. 2020;12(1):39. doi:10.1186/s13195-020-00603-8
8. Nasreddine ZS, Phillips NA, Bédirian V, et al. The Montreal Cognitive Assessment, MoCA: A brief screening tool for mild cognitive impairment [published correction appears in *J Am Geriatr Soc*. 2019;67(9):1991]. *J Am Geriatr Soc*. 2005;53(4):695–699. doi:10.1111/j.1532-5415.2005.53221.x
9. Pinto TCC, Machado L, Bulgacov TM, et al. Is the Montreal Cognitive Assessment (MoCA) screening superior to the Mini-Mental State Examination (MMSE) in the detection of mild cognitive impairment (MCI) and Alzheimer's disease (AD) in the elderly? *Int Psychogeriatr*. 2019;31(4):491–504. doi:10.1017/S1041610218001370
10. Aguilar-Navarro SG, Mimenza-Alvarado AJ, Palacios-García AA, Samudio-Cruz A, Gutiérrez-Gutiérrez LA, Ávila-Funes JA. Validity and reliability of the Spanish version of the Montreal Cognitive Assessment (MoCA) for the detection of cognitive impairment in Mexico. *Rev Colomb Psiquiatr (Engl Ed)*. 2018;47(4):237–243. doi:10.1016/j.rcp.2017.05.003
11. Hategan SI, Kamer SA, Craig RG, et al. Cognitive dysfunction in young subjects with periodontal disease. *Neurol Sci*. 2021;42(11):4511–4519. doi:10.1007/s10072-021-05115-3
12. Lee YH. Association of temporomandibular disorders and reduced masticatory function with dementia in elderly. *Spec Care Dentist*. 2024;44(2):613–614. doi:10.1111/scd.12908
13. Sigurdsson T, Duvarci S. Hippocampal-prefrontal interactions in cognition, behavior, and psychiatric disease. *Front Syst Neurosci*. 2015;9:190. doi:10.3389/fnsys.2015.00190
14. Hu X, Zhang J, Qiu Y, Liu Z. Periodontal disease and the risk of Alzheimer's disease and mild cognitive impairment: A systematic review and meta-analysis. *Psychogeriatrics*. 2021;21(5):813–825. doi:10.1111/psyg.12743
15. Wang RPH, Ho YS, Leung WK, Goto T, Chang RCC. Systemic inflammation linking chronic periodontitis to cognitive decline. *Brain Behav Immun*. 2019;81:63–73. doi:10.1016/j.bbi.2019.07.002
16. Shin HS, Shin MS, Ahn YB, Choi BY, Nam JH, Kim HD. Periodontitis is associated with cognitive impairment in elderly Koreans: Results from the Yangpyeong cohort study. *J Am Geriatr Soc*. 2016;64(1):162–167. doi:10.1111/jgs.13781
17. Nascimento PC, Castro MML, Magno MB, et al. Association between periodontitis and cognitive impairment in adults: A systematic review. *Front Neurol*. 2019;10:323. doi:10.3389/fneur.2019.00323
18. Gil Montoya JA, Barrios R, Sanchez-Lara I, et al. Systemic inflammatory impact of periodontitis on cognitive impairment. *Gerodontology*. 2020;37(1):11–18. doi:10.1111/ger.12431
19. Kamer AR, Craig RG, Pirraglia E, et al. TNF-alpha and antibodies to periodontal bacteria discriminate between Alzheimer's disease patients and normal subjects. *J Neuroimmunol*. 2009;216(1–2):92–97. doi:10.1016/j.jneuroim.2009.08.013
20. Gaur S, Agnihotri R. Alzheimer's disease and chronic periodontitis: Is there an association? *Geriatr Gerontol Int*. 2015;15(4):391–404. doi:10.1111/ggi.12425
21. Teixeira FB, Saito MT, Matheus FC, et al. Periodontitis and Alzheimer's disease: A possible comorbidity between oral chronic inflammatory condition and neuroinflammation. *Front Aging Neurosci*. 2017;9:327. doi:10.3389/fnagi.2017.00327
22. Nilsson H, Sanmartin Berglund J, Renvert S. Longitudinal evaluation of periodontitis and development of cognitive decline among older adults. *J Clin Periodontol*. 2018;45(10):1142–1149. doi:10.1111/jcpe.12992
23. Iwasaki M, Kimura Y, Ogawa H, et al. Periodontitis, periodontal inflammation, and mild cognitive impairment: A 5-year cohort study. *J Periodontal Res*. 2019;54(3):233–240. doi:10.1111/jre.12623
24. Sung CE, Huang RY, Cheng WC, Kao TW, Chen WL. Association between periodontitis and cognitive impairment: Analysis of national health and nutrition examination survey (NHANES) III. *J Clin Periodontol*. 2019;46(8):790–798. doi:10.1111/jcpe.13155
25. McClanahan SF, Bartizek RD, Biesbrock AR. Identification and consequences of distinct Löe–Silness gingival index examiner styles for the clinical assessment of gingivitis. *J Periodontol*. 2001;72(3):383–392. doi:10.1902/jop.2001.72.3.383
26. Tonetti MS, Greenwell H, Kornman KS. Staging and grading of periodontitis: Framework and proposal of a new classification and case definition [published correction appears in *J Periodontol*. 2018;89(12):1475]. *J Periodontol*. 2018;89 Suppl 1:S159–S172. doi:10.1002/JPER.18-0006
27. Liccardo D, Marzano F, Carraturo F, et al. Potential bidirectional relationship between periodontitis and Alzheimer's disease. *Front Physiol*. 2020;11:683. doi:10.3389/fphys.2020.00683
28. Sharma S, Nayak SU, Uppoor A, Rao S, Pai K, Natarajan S. Evaluation of cognitive impairment in type 2 diabetic patients with chronic periodontitis: A cross-sectional study. *J Int Soc Prev Community Dent*. 2021;11(1):50–57. doi:10.4103/jispcd.JISPCD_349_20
29. Tugnait A, Carmichael F. Use of radiographs in the diagnosis of periodontal disease. *Dent Update*. 2005;32(9):536–542. doi:10.12968/denu.2005.32.9.536
30. Caton JG, Armitage G, Berglundh T, et al. A new classification scheme for periodontal and peri-implant diseases and conditions – Introduction and key changes from the 1999 classification. *J Clin Periodontol*. 2018;45 Suppl 20:S1–S8. doi:10.1111/jcpe.12935
31. Holmes C, Cunningham C, Zotova E, et al. Systemic inflammation and disease progression in Alzheimer disease. *Neurology*. 2009;73(10):768–774. doi:10.1212/WNL.0b013e3181b6bb95

32. Kapila YL. Oral health's inextricable connection to systemic health: Special populations bring to bear multimodal relationships and factors connecting periodontal disease to systemic diseases and conditions. *Periodontol 2000*. 2021;87(1):11–16. doi:10.1111/prd.12398
33. Li Y, Liu L, Barger SW, Griffin WST. Interleukin-1 mediates pathological effects of microglia on tau phosphorylation and on synaptophysin synthesis in cortical neurons through a p38-MAPK pathway. *J Neurosci*. 2003;23(5):1605–1611. doi:10.1523/JNEUROSCI.23-05-01605.2003
34. Stewart R, Weyant RJ, Garcia ME, et al. Adverse oral health and cognitive decline: The health, aging and body composition study. *J Am Geriatr Soc*. 2013;61(2):177–184. doi:10.1111/jgs.12094
35. Holmer J, Eriksdotter M, Schultzberg M, Pussinen PJ, Buhlin K. Association between periodontitis and risk of Alzheimer's disease, mild cognitive impairment and subjective cognitive decline: A case–control study. *J Clin Periodontol*. 2018;45(11):1287–1298. doi:10.1111/jcpe.13016
36. Yoneyama T, Yoshida M, Ohrui T, et al.; Oral Care Working Group. Oral care reduces pneumonia in older patients in nursing homes. *J Am Geriatr Soc*. 2002;50(3):430–433. doi:10.1046/j.1532-5415.2002.50106.x
37. Kamer AR, Dasanayake AP, Craig RG, Glodzik-Sobanska L, Bry M, de Leon MJ. Alzheimer's disease and peripheral infections: The possible contribution from periodontal infections, model and hypothesis. *J Alzheimers Dis*. 2008;13(4):437–449. doi:10.3233/jad-2008-13408
38. Le V, Kurnutala L, SchianodiCola J, et al. Premedication with intravenous Ibuprofen improves recovery characteristics and stress response in adults undergoing laparoscopic cholecystectomy: A randomized controlled trial. *Pain Med*. 2016;17(6):1163–1173. doi:10.1093/pm/pnv113
39. Peng M, Wang YL, Wang FF, Chen C, Wang CY. The cyclooxygenase-2 inhibitor parecoxib inhibits surgery-induced proinflammatory cytokine expression in the hippocampus in aged rats. *J Surg Res*. 2012;178(1):e1–e8. doi:10.1016/j.jss.2012.08.030
40. Zhao WX, Zhang JH, Cao JB, et al. Acetaminophen attenuates lipopolysaccharide-induced cognitive impairment through anti-oxidant activity. *J Neuroinflammation*. 2017;14(1):17. doi:10.1186/s12974-016-0781-6
41. Guo H, Chang S, Pi X, et al. The effect of periodontitis on dementia and cognitive impairment: A meta-analysis. *Int J Environ Res Public Health*. 2021;18(13):6823. doi:10.3390/ijerph18136823
42. Naorungroj S, Schoenbach VJ, Wruck L, et al. Tooth loss, periodontal disease, and cognitive decline in the Atherosclerosis Risk in Communities (ARIC) study. *Community Dent Oral Epidemiol*. 2015;43(1):47–57. doi:10.1111/cdoe.12128
43. Vassilaki M, Aakre JA, Cha RH, et al. Multimorbidity and risk of mild cognitive impairment. *J Am Geriatr Soc*. 2015;63(9):1783–1790. doi:10.1111/jgs.13612
44. Soto-Estrada G, Moreno-Altamirano L, Pahua Díaz D. Epidemiological overview of Mexico's leading causes of morbidity and mortality. *Rev Fac Med (Méx)*. 2016;59(6):8–22. http://www.scielo.org.mx/scielo.php?script=sci_arttext&pid=S0026-17422016000600008&lng=es. Accessed November 1, 2023.
45. Cecato JF, Martinelli JE, Izbicki R, Yassuda MS, Aprahamian I. A subtest analysis of the Montreal Cognitive Assessment (MoCA): Which subtests can best discriminate between healthy controls, mild cognitive impairment and Alzheimer's disease? *Int Psychogeriatr*. 2017;29(4):701. doi:10.1017/S104161021600212X
46. Langa KM, Levine DA. The diagnosis and management of mild cognitive impairment: A clinical review. *JAMA*. 2014;312(23):2551–2561. doi:10.1001/jama.2014.13806
47. Petersen RC, Caracciolo B, Brayne C, Gauthier S, Jelic V, Fratiglioni L. Mild cognitive impairment: A concept in evolution. *J Intern Med*. 2014;275(3):214–228. doi:10.1111/joim.12190
48. López-Sanz D, Bruña R, Delgado-Losada ML, et al. Electrophysiologial brain signatures for the classification of subjective cognitive decline: Towards an individual detection in the preclinical stage of dementia. *Alzheimers Res Ther*. 2019;11(1):49. doi:10.1186/s13195-019-0502-3
49. Waliszewska-Prosół M, Bladowska J, Budrewicz S, Sąsiadek M, Dziadkowiak E, Ejma M. The evaluation of Hashimoto's thyroiditis with event-related potentials and magnetic resonance spectroscopy and its relation to cognitive function. 2021;11(1):2480. doi:10.1038/s41598-021-82281-6
50. Lee YL, Hu HY, Huang LY, Chou P, Chu D. Periodontal disease associated with higher risk of dementia: Population-based cohort study in Taiwan. *J Am Geriatr Soc*. 2017;65(9):1975–1980. doi:10.1111/jgs.14944

Original paper

Early healing after regenerative periodontal surgery with or without antibiotics

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Abstract

Background. The effectiveness of guided tissue regeneration (GTR) has been thoroughly documented. Since most post-GTR complications are related to the exposure of the membrane and its subsequent bacterial contamination, clinicians treat the incorporation of systemic antibiotics as an integral component of the standard surgical procedure. However, this approach remains controversial.

Objectives. The aim of the study was to assess the effect of postsurgical administration of antibiotics on early healing and patient morbidity after treatment of periodontal intrabony defects with deproteinized bovine bone mineral (DBBM) and a collagen membrane (GTR).

Material and methods. The study encompassed 41 patients with 41 intrabony defects. The subjects were randomly assigned to either the test group (DBBM/GTR+AB (postsurgical antibiotic)) or the control group (DBBM/GTR). In the test group, 1 g of amoxicillin was administered twice daily for 7 days. The early healing index (EHI) was assessed 1 and 2 weeks after the procedure. Patient morbidity was recorded. The clinical attachment level (CAL), probing depth (PD) and gingival recession (GR) were measured at baseline and 6 months after surgery.

Results. Early healing was uneventful in both groups. No adverse events were recorded in either group. In the second week, EHI-defined primary flap closure was evident in 95% of the test group sites and 80% of the control group sites. The CAL changed significantly in both groups: from 8.7 ±1.4 mm to 4.6 ±1.7 mm in the DBBM/GTR+AB group ($p < 0.0001$); and from 8.6 ±1.9 mm to 5.7 ±1.2 mm in the DBBM/GTR group ($p < 0.0001$). The significant outcome was in favor of the test group ($p = 0.010$). Probing depth significantly decreased in both groups, without any observed intergroup differences.

Conclusions. The administration of systemic amoxicillin after regenerative periodontal surgery did not have any effect on early wound healing; however, it yielded a statistically significant CAL gain after 6 months compared to the group treated without antibiotics.

Keywords: antibiotics, intrabony defects, guided tissue regeneration, regenerative dentistry

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Highlights

- Systemic antibiotics after regenerative periodontal surgery did not result in a substantial improvement in early wound healing.
- The use of systemic antibiotics was associated with a significantly greater clinical attachment level (CAL) gain after 6 months.
- Strict adherence to aseptic protocols during surgery is essential to minimize postsurgical complications.

Introduction

The primary aim of periodontal therapy is to facilitate the regeneration of tissues that have been lost due to inflammation. In essence, periodontal regeneration comprises the reconstitution of the functionally arranged collagen fibers inserting into the new cementum and bone. The periodontium itself has exceptional natural, self-restoring properties that can result in regeneration after surgery, but only under optimal conditions. Wound stability, healing per primam and enhanced space provision for regeneration are the key factors that stimulate the healing process.[1,2]

The establishment of knowledge on the regenerative potential of the periodontal ligament, as well as a thorough understanding of regeneration, fostered a revolutionary change in surgery.[3] Papilla-preserving incisions and minimally invasive surgical techniques have largely limited the risk of developing postoperative complications.[4–7] Regenerative surgery has so far used barrier membranes, bone grafts, bone substitutes, bioactive agents, and their combinations. Certain surgical techniques and materials result in a reduced probing depth (PD) and greater clinical attachment level (CAL) gain. In contrast, the application of flap elevation as a standalone procedure has a lesser potential for this outcome.[8,9]

The effectiveness of guided tissue regeneration (GTR) has been documented both clinically and histologically.[8,10,11] This modality uses barrier membranes to separate the epithelium and gingival tissues from the root, thus enabling periodontal ligament cells to repopulate the isolated space.[12] Histological studies have demonstrated that the primary function of the membrane is to provide space for regeneration and to stabilize the clot.[13,14]

Although the routine administration of systemic antibiotics in conjunction with periodontal surgical procedures prevents postsurgical complications and optimizes expected outcomes, this approach is guided by practical experience rather than by evidence.[15] Some researchers claim that perioperative antibiotics promote CAL gain,[16,17] while others argue that they offer no greater benefits than the surgery itself.[18] It is also worth noting that the number of postsurgical infections is statistically low,[19] and the uncontrolled use of antibiotics may favor bacterial resistance.[20] In addition, hypersensitivity, allergic reactions and interactions with other drugs necessitate caution.[21]

Since most post-GTR complications that affect the outcome are related to the exposure of the membrane and its consequent bacterial contamination,[22] some clinicians treat the inclusion of systemic antibiotics as inseparable from the standard surgical procedure.[23,24] Currently, however, the routine administration of systemic antibiotics after regenerative periodontal surgery using barrier membranes remains controversial, with no clear protocols available. It is of great importance to reduce the bacterial and viral load before surgery, a result that can be achieved through initial periodontal therapy.[25,26] Studies are also being conducted on topical agents that will be able to modulate healing in the periodontium. Plant preparations are an important focus of this research.[27,28]

The process of healing is initiated by hemostasis, early clot formation and inflammatory cell infiltration. Subsequently, epithelial cells begin to proliferate, and fibroblasts migrate into the wound. Wound closure is essential for periodontal wound stability in the early phase of healing,[1,2] and the first postoperative week is deemed to be critical.[29] Uneventful wound healing after GTR is a major factor for clinical success.[30] Trombelli et al. have emphasized the importance of primary intention healing and have observed substantial clinical improvements in cases without membrane exposure.[31] A number of indices have been developed to characterize the early wound healing process, namely the healing index (HI),[32] the early healing index (EHI),[7] the wound healing index (WHI),[33] and the early wound healing score (EHS).[34] At the same time, several surrogate parameters have been used to describe the healing process, encompassing the tissue color, bleeding, characteristics of incision margins, the presence of suppuration, the assessment of wound closure, abscess formation, fibrin and necrosis, edema, erythema, suppuration, patient discomfort, and flap dehiscence.[35]

Taking into consideration the complexity of periodontal healing, the present study aimed to evaluate early postoperative healing, as measured by the EHI,[7] and to assess the possible complications that may arise after regenerative surgery. The assessment utilized a deproteinized bovine bone mineral (DBBM) and a collagen membrane, with or without a postoperative antibiotic regimen. The secondary objective of the study was to identify a possible relationship between early healing and the clinical outcomes measured at 6 months following regenerative surgery.

Material and methods

Study design and participants

The project was designed as a randomized, prospective, controlled clinical study. The study was approved by the Bioethics Committee of Medical University of Bialystok, Poland (approval No. R-I-002-302-2013), and was compliant with the Declaration of Helsinki. Patients diagnosed with stage III periodontitis[36] and having at least 1 intrabony defect were enrolled in the study, which encompassed 41 generally healthy adults.

The patients were required to meet the following inclusion criteria: age ≥18 years; at least 1 intrabony defect with a PD ≥ 6 mm, radiographically tested using the long cone paralleling technique (radiographic depth (RxD) ≥3 mm, radiographic width (RxW) ≥2 mm); no allergic reaction to the penicillin in the family history; smoking status: non-smoker; effective plaque control (full mouth plaque score (FMPS) <20%); and signed informed consent.

Subjects who had received antibiotics within 3 months prior to the study, those diagnosed with systemic diseases that affect healing, as well as pregnant or breastfeeding women were excluded.

During the inclusion period, 99 patients with intrabony defects were screened. Twenty three subjects were excluded due to the presence of a radiological defect with a depth smaller than 3 mm. Fourteen individuals resigned without any reason. Six patients were in need of complete oral rehabilitation. Fourteen patients were excluded for the following reasons: diabetes (n = 4); insufficient oral hygiene (n = 3); age <18 years (n = 2); acute endo-perio lesions (n = 2); pregnancy (n = 1); breastfeeding (n = 1); radiotherapy (n = 1).

The random allocation software was used to allocate 41 patients into 2 groups: the study group (DBBM/GTR+AB (postsurgical antibiotic), n = 21); and the control group (DBBM/GTR, n = 20).

Clinical measurements

Prior to and 6 months after surgery, an experienced periodontist performed clinical measurements, including PD, gingival recession (GR) and mathematically calculated CAL. The probe was calibrated in 1-mm increments (PCPUNC 15; Hu-Friedy, Chicago, USA) and the readings were rounded up to the nearest millimeter. Six locations around each tooth with an intrabony defect were probed: the mesio-, mid- and distobuccal regions, as well as the mesio-, mid- and distolingual regions. The cementoenamel junction (CEJ) was used as a fixed reference point, unless the CEJ was not detectable, in which case a filling margin was used instead. The deepest value in the pre-surgery test was relevant for the statistical assessment.

The FMPS and the full mouth bleeding on probing (FMBOP) were dichotomously calculated as percentages for the 4 surfaces of each tooth.[37]

At 1st, 2nd and 4th week after surgery, the sites were examined for signs of suppuration (absence or presence) and dehiscence (in mm).

Photographic documentation of each site encompassed both an inner occlusal view and occlusal and non-occlusal side views. The photographs were taken with a professional camera designed for intraoral photography (Olympus OM-D E-M10; Olympus Corporation, Tokyo, Japan; lens: M. Zuiko Digital ED 60mm F:2.8 Macro; Olympus Corporation; macro flash: Metz 15 MS-1; Metz Consumer Electronics GmbH, Zirndorf, Germany).

The EHI was assessed based on the photographs taken at the 1st (EHI1) and 2nd weeks (EHI2) post-surgery, as follows:

- EHI = 1: a complete flap closure with no visible fibrin line in the interproximal area (Fig. 1A);
- EHI = 2: a complete flap closure with a visible fine fibrin line in the interproximal area (Fig. 1B);
- EHI = 3: a complete flap closure with a visible fibrin clot in the interproximal area (Fig. 1C);
- EHI = 4: an incomplete flap closure with partial necrosis of the interproximal tissue (Fig. 1D,E);
- EHI = 5: an incomplete flap closure with complete necrosis of the interproximal tissue.

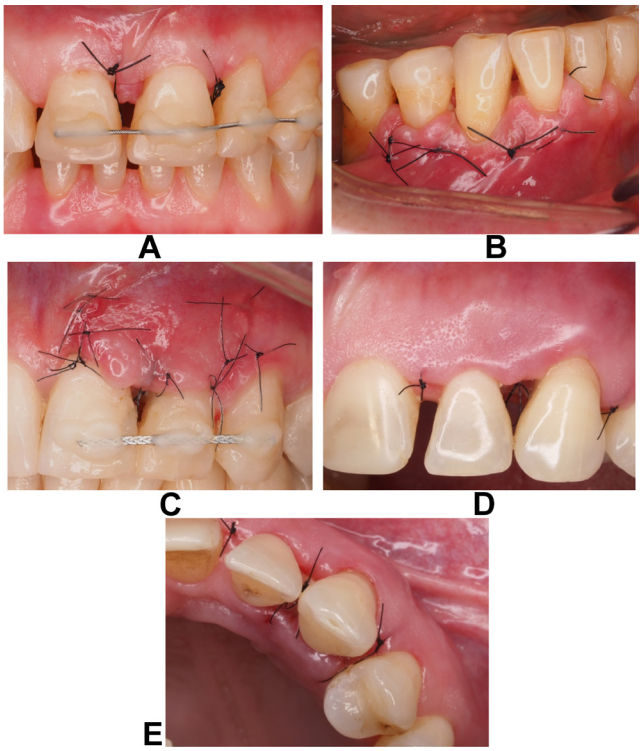

Fig. 1. Photographs taken at the 1st week (EHI1) after regenerative surgery
A. Tooth 36, EHI1 = 1 (test group); B. Tooth 43, EHI1 = 2 (test group); C. Tooth 22, EHI1 = 3 (control group); D. Tooth 23, EHI1 = 4 (test group); E. Occlusal photo of tooth 23, EHI1 = 4.

Surgical procedures

An experienced surgeon carried out all surgical procedures. Mucoperiosteal flaps were elevated buccally and lingually via an intrasulcular incision under local anesthesia (Ubistesin Forte; 3M ESPE, Seefeld, Germany). Vertical incisions were made exclusively in cases where easier access to the defect was necessary. The modified papilla preservation technique (MPPT) or simplified papilla preservation flap (SPPF)[4,5] was employed within interdental spaces. Subsequently, the granulation tissue was removed, and the root surfaces were thoroughly scaled and root planed using conventional Gracey currettes (Hu-Friedy) and an ultrasonic scaler (Tip PS, Piezon EMS; EMS, Nyon, Switzerland). The defect was then filled with the gently pressed DBBM (cerabone®; botiss biomaterials GmbH, Zossen, Germany) and covered with a trimmed collagen membrane (collprotect®; botiss biomaterials GmbH). Once the membrane was in place, the mucoperiosteal flap was coronally displaced and stabilized over the defect with vertical modified mattress sutures. The remaining papillae and vertical incisions were closed with loop sutures (monofilament non-resorbable Ethilon™ 5-0; Johnson & Johnson Company, New Brunswick, USA).

Intrasurgery measurements

The elevation of the flap and debridement allowed for the assessment of the extent of each intrabony defect with a periodontal probe (PCPUNC 15) and its subsequent classification as one-, two- or three-walled according to the morphological criteria. In instances of evident tooth mobility after surgery, the tooth was immobilized.

Postoperative care

On the day of the surgery, patients randomized to the test group were instructed to take 1 g of amoxicillin every 12 h for 7 days (Ospamox; Sandoz GmbH, Kundl, Austria). Additionally, they were advised to use mouthrinses containing 0.2% chlorhexidine digluconate solution for 2 weeks (Eludril; Pierre Fabre Laboratories, Paris, France). After that time, the sutures were removed, and the patients were permitted to resume brushing in the healing area with an ultra-soft brush.

The 6-month observation period was a time of scrupulous periodontal care for the patients. The 1-, 2- and 4-week postoperative appointments covered an evaluation of the patient's general oral hygiene, polishing of the site, and detection of potential suppuration and/or wound dehiscence. In addition to supragingival scaling, the 2-, 3- and 6-month appointments involved the assessment of FMPS and FMBOP. Intraoral photographs were taken at each appointment. No subgingival instrumentation was performed in the treated areas during the first 6 months after surgery.

Statistical analysis

The data analysis was conducted using the Statistica™ 13.1 software (StatSoft, Tulsa, USA), with each patient considered a statistical unit. The primary outcome variables encompassed EHI characteristics and the parameters corresponding to suppuration, wound dehiscence and severe postsurgical pain. The alterations in CAL and PD, as determined by probing at the 6th month, and their association with EHI, were considered secondary variables. The analysis used the maximum value of CAL recorded at baseline. The variables were expressed as the mean (*M*) and standard deviation (*SD*), as well as the median (*Me*) and quartiles. Within each group, a statistical analysis was performed using the Wilcoxon matched-pairs signed-rank test, while between-group comparisons were conducted employing the Mann–Whitney rank sum test. The value of $p < 0.05$ was considered statistically significant. In addition, the Spearman's rank correlation coefficient was calculated to assess the relationship between early healing, as described by EHI1 and EHI2, and 6-month clinical parameters.

The sample size calculation was performed a priori, under the assumption that the *SD* of the change in CAL was 1 mm, and that a mean difference of 1 mm could be detected with a test power of 80% in 32 subjects. However, we considered the possibility of dropouts and thus recruited and randomized 41 patients.

Results

The earliest outcome of the study, in which 41 adult patients underwent a surgical procedure (27 women, 14 men, mean age: 41.78 years), revealed predominantly undisturbed healing in both the test and control groups. The only exceptions were slight, temporary dehiscences. Otherwise, we did not observe any signs of suppuration or severe swelling, and the patients did not complain of intense pain, fever or feeling unwell. Only 2 patients in the test group experienced tooth hypersensitivity, and 1 subject in the control group reported a cold sore in the corner of the mouth. The characteristics of the defects at baseline are presented in Table 1.

One week after surgery, the mean EHI1 scores were 2.14 ±1.2 for the DBBM/GTR+AB group, and 2.4 ±1.3 for the DBBM/GTR group. Two-week postoperative EHI2 scores were 1.48 ±0.87 and 2.05 ±1.28, respectively. None of the sites exhibited an EHI score of 5, indicating complete necrosis of the interproximal tissue. An incomplete flap closure resulting in a dehiscence was observed in 4 test patients and 6 control patients at week 1, while at week 2, it was observed in 1 test patient and 4 control patients. After 4 weeks, dehiscence was observed in 3 test patients and 1 control group patient. However, the observed dehiscences did not exceed 2 mm. After 2 weeks, 95% of the DBBM/GTR+AB sites and 80% of the DBBM/GTR sites

Table 1. Characteristics of the defects at baseline

Group	1-wall/2-wall/3-wall, *n*	PD [mm]	GR [mm]	CAL [mm]	Intra [mm]
Test (DBBM/GTR+AB)	5/12/4	7.8 ±1.5	0.9 ±1.2	8.7 ±1.4	5.5 ±2.0
Control (DBBM/GTR)	6/9/5	7.4 ±1.3	1.2 ±1.3	8.6 ±1.9	5.0 ±1.6
p-value (Mann–Whitney rank sum test)		NS	NS	NS	NS

NS – non-significant; PD – pocket depth; GR – gingival recession; CAL – clinical attachment level; Intra – depth of intrabony component; DBBM – deproteinized bovine bone material; GTR – guided tissue regeneration; AB – postsurgical antibiotic.

demonstrated primary flap closure. The distribution of the EHI scores is reported in Table 2.

At baseline, no statistically significant differences were observed between the groups in terms of PD, GR and CAL. At 6 months, a statistically significant decrease in PD and CAL gain was observed in both groups. Additionally, GR increased significantly in the control but not in the test group. The between-group comparison revealed statistically significant differences in terms of CAL, while PD and GR were not significantly different (Table 3).

The investigation revealed a modest correlation between EHI1 and EHI2 paired with GR and CAL in the control group after a 6-month period. There were no statistically significant correlations in the test group (Table 4).

The patients in both groups maintained high standards of oral hygiene throughout the study, as evidenced by FMPS < 20% and FMBOP < 20%.

Table 2. Distribution of the early healing index (EHI) scores at the 1st week and 2nd week after regenerative surgery

EHI score	Test group (DBBM/GTR+AB)		Control group (DBBM/GTR)	
	1st week	2nd week	1st week	2nd week
1	9 (43)	15 (71)	7 (35)	11 (55)
2	4 (19)	3 (14)	4 (20)	1 (5)
3	4 (19)	2 (10)	3 (15)	4 (20)
4	4 (19)	1 (5)	6 (30)	4 (20)
5	0 (0)	0 (0)	0 (0)	0 (0)

Data presented as frequency (percentage) (*n* (%)).

Table 4. Results of the Spearman's rank correlation analysis

Pair of variables	Control group (DBBM/GTR) (*n* = 20)		Test group (DBBM/GTR+AB) (*n* = 21)	
	Spearman's *r*	*p*-value	Spearman's *r*	*p*-value
EHI1 + GR6	0.50	0.020*	0.16	NS
EHI1 + CAL6	0.55	0.010*	0.22	NS
EHI2 + GR6	0.45	0.049*	0.24	NS
EHI2 + CAL6	0.51	0.020*	0.21	NS

* statistically significant ($p < 0.05$); GR6 – gingival recession at the 6th month after surgery; CAL6 – clinical attachment level at the 6th month after surgery.

Discussion

The primary aim of the present study was to determine the impact of a routine course of antibiotic therapy on the early healing of periodontal wounds after regenerative surgery employing collagen membranes and DBBM. Early postoperative complications, such as suppuration, wound dehiscence and severe postoperative pain were meticulously monitored to ascertain the necessity of an antibiotic regimen following regenerative surgery for intrabony defects. Irrespective of the presence or absence of an antibiotic regimen, no cases of suppuration or severe inflammatory complications were observed in any of the study groups.

The rationale for selecting the EHI was based on the fact that this index was specifically developed to monitor

Table 3. Between-group comparison of clinical parameters at baseline and 6 months after regenerative surgery

Parameter		Baseline *M* ±*SD*	6th month *M* ±*SD*	*p*-value	Baseline *Me*	6th month *Me*	Difference *M* ±*SD*
PD [mm]	DBBM/GTR+AB	7.8 ±1.5	3.6 ±0.9	<0.0001	8	3	4.2 ±1.4
	DBBM/GTR	7.4 ±1.3	4.0 ±0.9	<0.0001	7	4	3.4 ±1.7
	p-value	–	NS	–	–	–	0.053
GR [mm]	DBBM/GTR+AB	0.9 ±1.2	1.0 ±1.2	NS	0	1	−0.2 ±0.8
	DBBM/GTR	1.2 ±1.3	1.8 ±1.4	0.020*	1	1.5	−0.6 ±0.8
	p-value	–	NS	–	–	–	NS
CAL [mm]	DBBM/GTR+AB	8.7 ±1.4	4.6 ±1.7	<0.0001	9	4	4.0 ±1.7
	DBBM/GTR	8.6 ±1.9	5.7 ±1.2	<0.0001	8	6	2.9 ±1.4
	p-value	–	0.010*	–	–	–	0.008*

M – mean; *SD* – standard deviation; *Me* – median; * statistically significant ($p < 0.05$). The Mann–Whitney rank sum test was used for the between-group comparisons. Changes over time were assessed using the Wilcoxon matched-pairs signed-rank test.

the healing of surgically treated intrabony defects. The EHI scores correspond to 5 characteristics indicative of complete or incomplete flap closure, taking into account the presence of fibrin and necrosis. None of the sites in our study displayed an EHI of 5, which would indicate a complete necrosis of the interproximal tissues. However, the present study revealed that the EHI scores were slightly higher in comparison to those documented by Wachtel et al.[7] In that study, EHI decreased from 1.85 to 1.39 within 2 weeks in the enamel matrix derivative group and from 1.65 to 1.19 in the control group.[7] The observed discrepancy in the results may be ascribed to the different surgical technique and biomaterials used for periodontal tissue regeneration. The surgical interventions performed in this study incorporated grafting materials and collagen membranes, necessitating more extensive flap preparation. Notably, unlike enamel matrix derivative, the biomaterial itself was required to preserve a space for regeneration under the flap.

The importance of primary intention healing has been demonstrated in the study on GTR procedures by Trombelli et al.[31] The paper demonstrated significantly lower values of bone gain when the membrane was previously exposed.[31] Wound closure for periodontal regeneration enables primary intention healing and is considered a prerequisite to stabilize the blood clot and promote a regeneration process.[35] The EHI is a clinically available method to determine the early healing phase of periodontal wounds. The relevance of EHI as a predictor of the outcomes of regenerative procedures has been investigated by Farina et al.[38] However, the study was not able to demonstrate the association between early healing and 6-month clinical results, such as a CAL gain and pocket depth reduction. The authors hypothesized that the observed findings could be related to a short observation period, suggesting that the EHI may not be sensitive enough to detect significant differences in early postoperative healing, which could have a substantial impact on the outcomes over a period of half a year.[38] The influence of the EHI on 1-year CAL gain was examined by Liu et al.[39] Lower EHI scores positively influenced the CAL gain. Sites demonstrating optimal healing were more likely to achieve a greater CAL gain during the 1-year observation period.[39] Apart from the abovementioned success, it is important to acknowledge that regenerative therapy is multifactorial and contingent on numerous patient-related and site-specific factors, other than the EHI.[40]

The results of the current study demonstrated that GTR with DBBM led to a notable decrease in PD and an increase in CAL after 6 months, irrespective of whether the surgery was followed by a course of postoperative antibiotics. The only exception was an unchanged GR in the DBBM/GTR+AB group, which proved advantageous when the expected outcome was a CAL gain rather than a recession. Thus, the efficacy of both postoperative protocols has been demonstrated.

Six months after surgery, the attained gain in CAL was 4.6 ±1.7 mm in the test group and 2.9 ±1.4 mm in the control group. The difference between the groups was statistically significant, favoring the antibiotic regimen. However, the statistical significance in the CAL gain was not confirmed after the 1-year observation period. As previously documented, a 1-year CAL gain in the test group was 3.6 ±1.6 mm, while in the control group it was 2.7 ±1.6 mm.[41] There were no statistically significant intergroup differences.[41] Other authors who used collagen membranes and xenogeneic biomaterials with antibiotics reported comparable gain in CAL. Camargo et al. observed a mean gain of 3.2 mm after 6 months.[42] Sculean et al. reported a mean gain of 4.0 mm after 1 year.[43] Esposito et al. noted a probing attachment level (PAL) of 3.6 mm after 1 year.[44] Similar clinical studies without postsurgical antibiotics revealed a mean CAL gain of 4.1 mm[45] and 3.7 mm after 1 year.[46]

Histological studies in humans have confirmed the effectiveness of GTR. For instance, in the study by Sculean et al., concurrently with a mean attachment gain of 3.6 mm, there was 2.4 mm of new cementum and 2.1 mm of new bone.[47] Thus, it may be anticipated that the clinical improvements obtained in the present study may also reflect, at least to a certain extent, periodontal regeneration. However, only a histological analysis can provide definite evidence regarding the quality of the newly formed tissues, which, due to evident reasons, could not be performed in the present study.[48]

Postsurgical antibiotic regimens vary in type and dosage. They range from 250 mg tetracycline administered 4 times daily for a week[4] to 1.5 g of amoxicillin once a day for a week,[49] 200 mg of doxycycline administered daily for a week,[50] or 1 g of amoxicillin plus clavulanic acid once a day for 8 days.[51] What is more, clinicians are not unanimous in their views on the potential benefits of antibiotic treatment. The main postoperative hazards to prevent are membrane exposure and wound infection. A probable bacterial contamination of the exposed membrane impedes clinical effects of the procedure. The novel papilla preservation techniques (MPPT, SPPF)[23] have led to a notable decrease in the incidence of such complications.[23] These methods ensure the secure placement of the flap above the barrier membrane. It is, however, practically impossible to keep the membrane aseptic, as it becomes contaminated within the first 3 minutes in the oral cavity.[22]

Antibiotics are also frequently recommended in another modality of regenerative therapy that uses enamel matrix derivative (EMD), even though the risk of infection is lower.[47,52] However, the results of randomized clinical studies have failed to reveal any advantages of a postoperative antibiotic regimen on the clinical outcomes obtained after regenerative surgery with EMD, thus corroborating our findings.[26,53]

A review of the relevant literature from 2005 to 2014 reveals that systemic antibiotics were routinely administered

in 75.4% of cases involving flap elevation despite the low overall proportion of postoperative infections. These amounted to 0.073% when antibiotics were included and to 0.693% when they were not. This difference was neither statistically nor clinically significant.[54] Previous studies have noted a limited occurrence of postoperative complications following periodontal surgery,[19,55] with reported rates of 2.09% and 4.2%, respectively. Some of these complications may be related to inadequate preoperative preparation of the patient. A thorough execution of the hygienic phase, comprising subgingival mechanical debridement (i.e., scaling and root planing), is imperative to reduce the bacterial load and minimize postoperative complication rates.[26] Finally, a strictly executed, asepsis-oriented surgical protocol, as evidenced in the present study, is crucial to minimize the need for postsurgical antibiotic therapy.[56]

Conclusions

The results of the present study do not support the routine administration of antibiotics in systemically healthy patients treated with a collagen membrane and DBBM xenograft. This type of regenerative therapy, carried out under aseptic conditions, ensured undisturbed healing. Although a supplemental 1-g dose of amoxicillin did not significantly influence EHI and PD reduction, it led to a CAL gain after 6 months.

Ethics approval and consent to participate

The study was approved by the Bioethics Committee of Medical University of Bialystok, Poland (approval No. R-I-002-302-2013), and was compliant with the Declaration of Helsinki. All patients provided signed informed consent.

Data availability

The datasets generated and/or analyzed during the current study are available from the corresponding author on reasonable request.

Consent for publication

Not applicable.

Use of AI and AI-assisted technologies

Not applicable.

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References

1. Wikesjö UM, Selvig KA. Periodontal wound healing and regeneration. *Periodontol 2000*. 1999;19:21–39. doi:10.1111/j.1600-0757.1999.tb00145.x
2. Susin C, Fiorini T, Lee J, De Stefano JA, Dickinson DP, Wikesjö UME. Wound healing following surgical and regenerative periodontal therapy. *Periodontol 2000*. 2015;68(1):83–98. doi:10.1111/prd.12057
3. Susin C, Wikesjö UME. Regenerative periodontal therapy: 30 years of lessons learned and unlearned. *Periodontol 2000*. 2013;62(1):232–242. doi:10.1111/prd.12003
4. Cortellini P, Prato GP, Tonetti MS. The modified papilla preservation technique. A new surgical approach for interproximal regenerative procedures. *J Periodontol*. 1995;66(4):261–266. doi:10.1902/jop.1995.66.4.261
5. Cortellini P, Prato GP, Tonetti MS. The simplified papilla preservation flap. A novel surgical approach for the management of soft tissues in regenerative procedures. *Int J Periodontics Restorative Dent*. 1999;19(6):589–599. PMID:10815597.
6. Cortellini P, Tonetti MS. Microsurgical approach to periodontal regeneration. Initial evaluation in a case cohort. *J Periodontol*. 2001;72(4):559–569. doi:10.1902/jop.2001.72.4.559
7. Wachtel H, Schenk G, Böhm S, Weng D, Zuhr O, Hürzeler MB. Microsurgical access flap and enamel matrix derivative for the treatment of periodontal intrabony defects: A controlled clinical study. *J Clin Periodontol*. 2003;30(6):496–504. doi:10.1034/j.1600-051x.2003.00013.x
8. Sculean A, Nikolidakis D, Nikou G, Ivanovic A, Chapple ILC, Stavropoulos A. Biomaterials for promoting periodontal regeneration in human intrabony defects: A systematic review. *Periodontol 2000*. 2015;68(1):182–216. doi:10.1111/prd.12086
9. Larsson L, Decker AM, Nibali L, Pilipchuk SP, Berglundh T, Giannobile WV. Regenerative medicine for periodontal and peri-implant diseases. *J Dent Res*. 2016;95(3):255–266. doi:10.1177/0022034515618887
10. Kao RT, Nares S, Reynolds MA. Periodontal regeneration – intrabony defects: A systematic review from the AAP Regeneration Workshop. *J Periodontol*. 2015;86(2 Suppl):S77–S104. doi:10.1902/jop.2015.130685
11. Reynolds MA, Kao RT, Camargo PM, et al. Periodontal regeneration – intrabony defects: A consensus report from the AAP Regeneration Workshop. *J Periodontol*. 2015;86(2 Suppl):S105–S107. doi:10.1902/jop.2015.140378
12. Gottlow J, Nyman S, Karring T, Lindhe J. New attachment formation as the result of controlled tissue regeneration. *J Clin Periodontol*. 1984;11(8):494–503. doi:10.1111/j.1600-051x.1984.tb00901.x
13. Haney JM, Nilvéus RE, McMillan PJ, Wikesjö UM. Periodontal repair in dogs: Expanded polytetrafluoroethylene barrier membranes support wound stabilization and enhance bone regeneration. *J Periodontol*. 1993;64(9):883–890. doi:10.1902/jop.1993.64.9.883
14. Sigurdsson TJ, Hardwick R, Bogle GC, Wikesjö UM. Periodontal repair in dogs: Space provision by reinforced ePTFE membranes enhances bone and cementum regeneration in large supra alveolar defects. *J Periodontol*. 1994;65(4):350–356. doi:10.1902/jop.1994.65.4.350
15. Herrera D, Alonso B, León R, Roldán S, Sanz M. Antimicrobial therapy in periodontitis: The use of systemic antimicrobials against the subgingival biofilm. *J Clin Periodontol*. 2008;35(8 Suppl):45–66. doi:10.1111/j.1600-051X.2008.01260.x
16. Nowzari H, Matian F, Slots J. Periodontal pathogens on polytetrafluoroethylene membrane for guided tissue regeneration inhibit healing. *J Clin Periodontol*. 1995;22(6):469–474. doi:10.1111/j.1600-051x.1995.tb00179.x
17. Mombelli A, Zappa U, Brägger U, Lang NP. Systemic antimicrobial treatment and guided tissue regeneration. Clinical and microbiological effects in furcation defects. *J Clin Periodontol*. 1996;23(4):386–396. doi:10.1111/j.1600-051x.1996.tb00562.x
18. Vest TM, Greenwell H, Drisko C, et al. The effect of postsurgical antibiotics and a membrane on regenerative healing in Class II furcation defects. *J Periodontol*. 1999;70(8):878–887. doi:10.1902/jop.1999.70.8.878
19. Powell CA, Mealey BL, Deas DE, McDonnell HT, Moritz AJ. Postsurgical infections: Prevalence associated with various periodontal surgical procedures. *J Periodontol*. 2005;76(3):329–333. doi:10.1902/jop.2005.76.3.329

20. van Winkelhoff AJ, Winkel EG. Antibiotics in periodontics: Right or wrong? *J Periodontol*. 2009;80(10):1555–1558. doi:10.1902/jop.2009.090276
21. Addy M, Martin MV. Systemic antimicrobials in the treatment of chronic periodontal diseases: A dilemma. *Oral Dis*. 2003;9 Suppl 1:38–44. doi:10.1034/j.1601-0825.9.s1.7.x
22. Nowzari H, MacDonald ES, Flynn J, London RM, Morrison JL, Slots J. The dynamics of microbial colonization of barrier membranes for guided tissue regeneration. *J Periodontol*. 1996;67(7):694–702. doi:10.1902/jop.1996.67.7.694
23. Cortellini P, Tonetti MS. Focus on intrabony defects: Guided tissue regeneration. *Periodontol 2000*. 2000;22:104–132. doi:10.1034/j.1600-0757.2000.2220108.x
24. Sanz M, Giovanolli JL. Focus on furcation defects: Guided tissue regeneration. *Periodontol 2000*. 2000;22:169–189. doi:10.1034/j.1600-0757.2000.2220111.x
25. Mahendra J, Mahendra L, Mugri MH, et al. Role of periodontal bacteria, viruses, and placental mir155 in chronic periodontitis and preeclampsia – a genetic microbiological study. *Curr Issues Mol Biol*. 2021;43(2):831–844. doi:10.3390/cimb43020060
26. Sculean A, Blaes A, Arweiler N, Reich E, Donos N, Brecx M. The effect of postsurgical antibiotics on the healing of intrabony defects following treatment with enamel matrix proteins. *J Periodontol*. 2001;72(2):190–195. doi:10.1902/jop.2001.72.2.190
27. Rahman S, Nagur Karibasappa S, Mehta DS. Evaluation of the wound-healing potential of the kiwifruit extract by assessing its effects on human gingival fibroblasts and angiogenesis. *Dent Med Probl*. 2023;60(1):71–77. doi:10.17219/dmp/146635
28. Ananda N, Ariawan D, Juniantito V. Effects of the *Hydnophytum formicarum* plant extract on collagen density, angiogenesis, wound length, and re-epithelialization in wound healing: Experimental study on rats. *Dent Med Probl*. 2022;59(1):67–73. doi:10.17219/dmp/140208
29. Werfully S, Areibi G, Toner M, et al. Tensile strength, histological and immunohistochemical observations of periodontal wound healing in the dog. *J Periodontal Res*. 2002;37(5):366–374. doi:10.1034/j.1600-0765.2002.01375.x
30. Polimeni G, Xiropaidis AV, Wikesjö UME. Biology and principles of periodontal wound healing/regeneration. *Periodontol 2000*. 2006;41:30–47. doi:10.1111/j.1600-0757.2006.00157.x
31. Trombelli L, Kim CK, Zimmerman GJ, Wikesjö UM. Retrospective analysis of factors related to clinical outcome of guided tissue regeneration procedures in intrabony defects. *J Clin Periodontol*. 1997;24(6):366–371. doi:10.1111/j.1600-051x.1997.tb00199.x
32. Landry RG. *Effectiveness of Benzydamine HC1 in the Treatment of Periodontal Post-Surgical Patients* [dissertation]. Toronto: University of Toronto; 1985.
33. Hagenaars S, Louwerse PHG, Timmerman MF, Van der Velden U, Van der Weijden GA. Soft-tissue wound healing following periodontal surgery and Emdogain application. *J Clin Periodontol*. 2004;31(10):850–856. doi:10.1111/j.1600-051X.2004.00571.x
34. Marini L, Rojas MA, Sahrmann P, Aghazada R, Pilloni A. Early wound healing score: A system to evaluate the early healing of periodontal soft tissue wounds. *J Periodontal Implant Sci*. 2008;48(5):274–283. doi:10.5051/jpis.2018.48.5.274
35. Rojas MA, Marini L, Pilloni A, Sahrmann P. Early wound healing outcomes after regenerative periodontal surgery with enamel matrix derivatives or guided tissue regeneration: A systematic review. *BMC Oral Health*. 2019;19(1):76. doi:10.1186/s12903-019-0766-9
36. Tonetti MS, Greenwell H, Kornman KS. Staging and grading of periodontitis: Framework and proposal of a new classification and case definition. *J Clin Periodontol*. 2018;45 Suppl 20:S149–S161. doi:10.1111/jcpe.12945
37. O'Leary TJ, Drake RB, Naylor JE. The plaque control record. *J Periodontol*. 1972;43(1):38. doi:10.1902/jop.1972.43.1.38
38. Farina R, Simonelli A, Rizzi A, Pramstraller M, Cucchi A, Trombelli L. Early postoperative healing following buccal single flap approach to access intraosseous periodontal defects. *Clin Oral Investig*. 2013;17(6):1573–1583. doi:10.1007/s00784-012-0838-6
39. Liu B, Ouyang X, Kang J, et al. Efficacy of periodontal minimally invasive surgery with and without regenerative materials for treatment of intrabony defect: A randomized clinical trial. *Clin Oral Investig*. 2022;26(2):1613–1623. doi:10.1007/s00784-021-04134-w
40. Tonetti MS, Prato GP, Cortellini P. Factors affecting the healing response of intrabony defects following guided tissue regeneration and access flap surgery. *J Clin Periodontol*. 1996;23(6):548–556. doi:10.1111/j.1600-051x.1996.tb01823.x
41. Pietruska M, Dolińska E, Milewski R, Sculean A. Effect of systemic antibiotics on the outcomes of regenerative periodontal surgery in intrabony defects: A randomized, controlled, clinical study. *Clin Oral Investig*. 2021;25(5):2959–2968. doi:10.1007/s00784-020-03616-7
42. Camargo PM, Lekovic V, Weinlaender M, et al. A controlled re-entry study on the effectiveness of bovine porous bone mineral used in combination with a collagen membrane of porcine origin in the treatment of intrabony defects in humans. *J Clin Periodontol*. 2000;27(12):889–896. doi:10.1034/j.1600-051x.2000.027012889.x
43. Sculean A, Berakdar M, Chiantella GC, Donos N, Arweiler NB, Brecx M. Healing of intrabony defects following treatment with a bovine-derived xenograft and collagen membrane. A controlled clinical study. *J Clin Periodontol*. 2003;30(1):73–80. doi:10.1034/j.1600-051x.2003.10192.x
44. Esposito M, Grusovin MG, Lambert F, et al. The effectiveness of a resorbable bone substitute with resorbable membrane in the treatment of periodontal infrabony defect – A multicenter randomized controlled trial. *Eur J Oral Implantol*. 2015;8(3):233–244. PMID:26355168.
45. Sculean A, Chiantella GC, Windisch P, Arweiler NB, Brecx M, Gera I. Healing of intra-bony defects following treatment with a composite bovine-derived xenograft (Bio-Oss Collagen) in combination with a collagen membrane (Bio-Gide PERIO). *J Clin Periodontol*. 2005;32(7):720–724. doi:10.1111/j.1600-051X.2005.00758.x
46. Iorio-Siciliano V, Andreuccetti G, Blasi A, Matarasso M, Sculean A, Salvi GE. Clinical outcomes following regenerative therapy of non-contained intrabony defects using a deproteinized bovine bone mineral combined with either enamel matrix derivative or collagen membrane. *J Periodontol*. 2014;85(10):1342–1350. doi:10.1902/jop.2014.130420
47. Sculean A, Donos N, Blaes A, Lauermann M, Reich E, Brecx M. Comparison of enamel matrix proteins and bioabsorbable membranes in the treatment of intrabony periodontal defects. A split-mouth study. *J Periodontol*. 1999;70(3):255–262. doi:10.1902/jop.1999.70.3.255
48. Reddy MS, Jeffcoat MK. Methods of assessing periodontal regeneration. *Periodontol 2000*. 1999;19:87–103. doi:10.1111/j.1600-0757.1999.tb00149.x
49. Cortellini P, Carnevale G, Sanz M, Tonetti MS. Treatment of deep and shallow intrabony defects. A multicenter randomized controlled clinical trial. *J Clin Periodontol*. 1998;25(12):981–987. doi:10.1111/j.1600-051x.1998.tb02402.x
50. Tonetti MS, Cortellini P, Suvan JE, et al. Generalizability of the added benefits of guided tissue regeneration in the treatment of deep infrabony defects. Evaluation in a multicenter randomized controlled clinical trial. *J Periodontol*. 1998;69(11):1183–1192. doi:10.1902/jop.1998.69.11.1183
51. De Sanctis M, Zucchelli G. Interleukin-1 gene polymorphisms and long-term stability following guided tissue regeneration therapy. *J Periodontol*. 2000;71(4):606–613. doi:10.1902/jop.2000.71.4.606
52. Heijl L, Heden G, Svärdström G, Östgren A. Enamel matrix derivative (EMDOGAIN) in the treatment of intrabony periodontal defects. *J Clin Periodontol*. 1997;24(9 Pt 2):705–714. doi:10.1111/j.1600-051x.1997.tb00253.x
53. Röllke L, Schacher B, Wohlfeil M, et al. Regenerative therapy of infrabony defects with and without postsurgical doxycycline. A randomized placebo-controlled trial. *J Clin Periodontol*. 2012;39(5):448–456. doi:10.1111/j.1600-051X.2012.01861.x
54. Liu Y, Duan D, Xin Y, et al. A review of the literature: Antibiotic usage and its relevance to the infection in periodontal flaps. *Acta Odontol Scand*. 2017;75(4):288–293. doi:10.1080/00016357.2017.1295165
55. Checchi L, Trombelli L, Nonato M. Postoperative infections and tetracycline prophylaxis in periodontal surgery: A retrospective study. *Quintessence Int*. 1992;23(3):191–195. PMID:1641461.
56. Abu-Ta'a M, Quirynen M, Teughels W, van Steenberghe D. Asepsis during periodontal surgery involving oral implants and the usefulness of peri-operative antibiotics: A prospective, randomized, controlled clinical trial. *J Clin Periodontol*. 2008;35(1):58–63. doi:10.1111/j.1600-051X.2007.01162.x

Original paper

Leveraging independence and mental fitness – keys to reducing in-hospital mortality among geriatric COVID-19 patients in the intensive care unit: A cross-sectional study in Poland

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Abstract

Background. Coronavirus disease 2019 (COVID-19) quickly reached the pandemic status, with 765.22 million confirmed cases of COVID-19 and 6.92 million COVID-19 deaths reported worldwide by May 2023. Due to its sudden and global nature, the COVID-19 pandemic has had a significant impact on the emotional and mental health of many people. A group of COVID-19 patients who frequently require intensive care are geriatric patients. The cognitive performance of these patients and their independence in instrumental activities of daily living (IADL) may be crucial to their prognosis and risk of in-hospital death.

Objectives. The present study aimed to assess the level of independence in activities of daily living (ADL), mental fitness, the level of fear of COVID-19, and cognitive functions to determine their impact on in-hospital mortality in geriatric COVID-19 patients.

Material and methods. A total of 300 intensive care unit (ICU) patients with COVID-19 were included in the cross-sectional study, using the following questionnaires: the Lawton IADL scale, the Katz ADL index of independence, the fear of COVID-19 scale (FCV-19S), the abbreviated mental test score (AMTS), and the 15-item geriatric depression scale (GDS15).

Results. Patients aged 64 or below reported significantly greater independence on the IADL scale and the basic ADL scale, and showed a significantly higher level of mental fitness (Mann–Whitney *U* test; $p = 0.001$). Patient survival and in-hospital mortality were influenced by independence in basic and complex ADL.

Conclusions. The level of independence is an important prognostic indicator for in-hospital mortality in geriatric COVID-19 patients. The higher the level of mental fitness, the higher the level of independence in basic and instrumental activities of daily living. Patients aged ≥65 years are less independent in basic and instrumental activities of daily living. Moreover, they show a significantly lower level of cognitive functions.

Keywords: quality of life, depression, COVID-19, intensive care unit

Introduction

The novel coronavirus disease 2019 (COVID-19) quickly reached the pandemic status, and as of May 3, 2023, 765.22 million confirmed COVID-19 cases and 6.92 million COVID-19 deaths had been reported worldwide.[1]

Due to its sudden and global nature, the COVID-19 pandemic has had a significant impact on the emotional and mental health of many people.[2] Its influence on healthcare professionals and patients requiring long-term hospital treatment, including intensive care, has been particularly profound.

According to the Centers for Disease Control and Prevention (CDC) report, 41% of respondents in the surveys conducted across the United States reported at least one adverse mental health condition directly attributable to the pandemic.[3] Over 50% of the respondents reported the symptoms of an anxiety disorder or a depressive disorder, or the symptoms of a trauma- and stressor-related disorder with regard to the pandemic, and as many as 11% reported having seriously considered suicide as a result of the pandemic.[3]

During the COVID-19 pandemic, the number of patients requiring intensive care increased significantly. COVID-19 patients hospitalized in intensive care units (ICUs) are under extreme psychological strain and they exhibit high levels of stress, as they are aware of how deadly the disease can be. In addition, an ICU stay is itself a risk factor for psychological difficulties.[4] Another factor that has an impact on the severity of anxiety and depression symptoms and confusion in COVID-19 patients under intensive care are ICU visitor restrictions, which make conscious patients feel they do not receive sufficient psychological support.[5] Isolation and the lack of contact due to COVID-19 negatively affect the mental health of patients. Research shows that isolation has a significant influence on the incidence of anxiety and depression, making patients more likely to show anger, feel lonely or be dissatisfied with the healthcare in the ward.[5,6] In addition, staff in overburdened wards are not always able to ensure that patients are provided with appropriate psychological care, which increases patients' stress and fear.[7] The literature notes that women are more likely to show a fear of the disease.[8,9] Studies also suggest that women's greater sensitivity and emotionality, which are dependent on the level of sex hormones, may be an influencing factor.[8,9] On the other hand, men find it more challenging to express fear. Therefore, women might be more vulnerable to the fear of COVID-19.[8,9]

COVID-19 patients requiring intensive care are often those who have developed acute respiratory distress syndrome (ARDS) and require prolonged mechanical ventilation (PMV). Prolonged mechanical ventilation is associated with an extended stay in ICU. It contributes to emotional stress and the deterioration of patients' well-being, as well as enhances the possibility of developing depression or even post-traumatic stress disorder (PTSD). The severity of the disease and the prolonged hospital stay negatively affect the mental well-being of patients.[10–13] The apparatus used makes it difficult for these patients to communicate with others. Moreover, they suffer from dyspnea, chest pain and chest tightness, which increases their anxiety about their health and long-term prognosis.[14–16] All these factors can lead to anxiety and depressive disorders, reducing patients' quality of life.

A common group of COVID-19 patients requiring intensive care are geriatric patients. Since geriatric patients, in addition to the underlying disease, often suffer from other comorbidities, and are more likely to have their daily independence deteriorated or their cognitive functions impaired, or to feel loneliness, there is a risk that their hospitalization and further prognosis may differ as compared to patients below 65 years of age. Studies show that in-hospital mortality in geriatric patients in ICUs, apart from somatic disorders, is associated with the impairment of their independence (in terms of activities of daily living (ADL) and instrumental activities of daily living (IADL)), and the loss or deterioration of cognitive functions.[17] The presence of cognitive dysfunction worsens the prognosis of patients, independently of other medical comorbidities.[18,19] The cognitive performance of patients aged 65 and above and their independence in IADL may be crucial to their prognosis and risk of in-hospital death.

Since the above factors may influence the occurrence of either depression or in-hospital mortality, the authors decided to use multiple scales. It was crucial to assess independence in basic and complex activities of daily living (the IADL and ADL scales), as well as the mental state, and this was done using the 15-item geriatric depression scale (GDS15). In the study, we used scales for evaluating the fear of COVID-19 (FCV-19S) and the abbreviated mental test score (AMTS), which assesses the cognitive functions of patients. Based on the existing research, our study hypothesizes that the loss of independence and the deterioration of cognitive functions worsen the prognosis of geriatric patients and increase the risk of in-hospital mortality.

The present study aimed to assess the level of independence in activities of daily living, mental fitness, the level of fear of COVID-19, and cognitive functions to determine their impact on in-hospital mortality in geriatric COVID-19 patients in comparison with patients aged ≤64 years.

Material and methods

This cross-sectional study was conducted among 300 ICU patients with COVID-19 from 2 hospitals in Wroclaw, Poland: the 4th Military Teaching Hospital; and the Independent Public Healthcare Center of the Ministry of the Interior and Administration. Among the patients examined, 234 were from the former hospital, and 66 from the latter one. The group was a convenient sample.

The data was collected from November 2020 to March 2022. The inclusion criteria were as follows: adult patients (aged more than 18 years); admission to ICU; a diagnosis of COVID-19; and informed consent. The exclusion criteria were: age under 18; admission to ICU without being diagnosed with COVID-19; being unable to complete the questionnaire; and the lack of consent to participate in the study. The following questionnaires were used: the Lawton IADL scale; the Katz ADL index of independence; FCV-19S; AMTS; and GDS15. Each of the questionnaires was administered to each patient only once. The patients were informed about the purpose of the study, and were made aware that they could withdraw from it at any time. Patient confidentiality was maintained by interviewing the patients in individual rooms. The questionnaires were not signed with the patient's first and last name; only gender and age were indicated in the questionnaire. The conversation with the patient was always conducted individually, devoting a lot of attention to each patient and an individually selected amount of time. The interviewer was the lead author – a qualified nurse with 25 years of experience with older people and considerable knowledge of the scales used in the study. The interviewer read the questions to each patient. When a given question was incomprehensible, the interviewer explained it thoroughly. At the end of the survey, the patient personally confirmed with their signature that all the data was correct, and signed informed consent to participate in the study.

If a given questionnaire was incomplete, and it was possible to talk to a particular patient, the conversation was repeated and the missing data was completed. If this was not possible, an incomplete questionnaire was excluded from the study. There were 15 incomplete questionnaires. For the 300 patients presented in the study, all data was completed in full. The study was approved by the relevant bioethics committee (Military Medical Chamber, Warsaw, Poland; approval No. KB-191/22).

The Lawton IADL scale is used to assess the patient's ability to carry out instrumental activities of daily living. There are 8 questions about the ability to use the telephone, do the shopping, prepare food, do housekeeping, do the laundry, travel independently, take medications, and handle finances. In each question, the patient may score from 1 point to 3 points, where 3 points refers to full independence in a given activity, 2 points means that the patient needs help with an activity, and 1 point that the patient is almost completely dependent on another person's help. The scale has a score range of 8–24. The higher the score, the higher the level of IADL independence. A minimum score of 8 points indicates full dependence, a score of 9–23 points means that the patient is moderately dependent, and a maximum score of 24 points stands for full independence.[20,21] The validity of the IADL scale was assessed using the Guttman and Rasch scoring system. The validity coefficients were consistent across the scoring methods.[22]

The Katz ADL index of independence is used to assess performance in basic activities of daily living. The scale consists of 6 questions (activities) measuring the ability to bathe, dress/undress, eat, and use the toilet independently, basic mobility, and the ability to control urination and defecation. The patient answers positively (1 point) or negatively (0 points) to each question. The total score on the scale (0–6 points) is the number of activities the patient can carry out independently. A score of 2 and less means that patient is significantly disabled.[23] The reliability of the Katz ADL index of independence was assessed with Cronbach's alpha of 0.87. Validity was assessed as a coefficient of scalability of 0.6 and a correlation with the activity index of 0.95.[24]

The FCV-19S is used to assess the fear of COVID-19. It has a score range of 7–35. The higher the score, the higher the level of fear. There are no standards as to what score on the scale indicates a high level of fear and what score denotes a low level of fear. However, the average number of points per question can be calculated and interpreted using the scoring scale for a single question, where 1 denotes a definite lack of fear, 2 denotes the lack of fear, 3 denotes a neutral response, 4 denotes the presence of fear, and 5 denotes the presence of a significant fear.[25] The interpretation of the results was adopted from the existing literature in Polish: 27–35 points stands for a high level of COVID-19 anxiety; 20–26 points indicates that anxiety is at a moderate level; 9–19 points means low anxiety; and <9 points indicates no COVID-19 fear.[26–28] In European publications, the cut-off point is a score of 16.5 or higher.[29] It is recommended not to use that cut-off point as a diagnostic value, but only as a value for screening for a group of patients with an increased risk of COVID-19 fear.[27–29] The FCV-19S was validated in Poland by testing 708 people.[26] The results showed high internal consistency of the scale (Cronbach's alpha of 0.89), and that the criteria of scale invariance and correlation with other variables were met.[26]

The AMTS is used to assess cognitive functions. It comprises 10 questions. The total score on AMTS is the number of correct answers to those questions. When answering, the patient has to supply information about their age, year of birth, address, the current time, the current year, the date of the First World War, and the name and surname of the current president. The patient is asked to repeat and remember an address given by the doctor, and count backward from 20 to 1. A score of 0–3 suggests a severe impairment of cognitive functions, a score of 4–6 suggests a moderate impairment, a score of 7–8 indicates a mild impairment, and a score of 9–10 indicates normal cognitive function.[30] The AMTS was validated by comparison to 7 AMTS versions and the mini-mental state examination (MMSE). Based on the linear regression and C statistics, AMTS showed a significant correlation and no significant differences from the C statistic (0.87), which proves the usefulness of this tool for assessing cognitive impairment.[31]

The GDS15 is used to assess the severity of depressive symptoms in the elderly. The scale consists of 15 questions that are answered affirmatively or negatively. The questions focus on evaluating satisfaction with life, being happy, feeling bored or inferior to others, feeling helpless or anxious, having memory problems, and not wanting to leave the house. The scale has a score range of 0–15, where the higher the score, the higher the severity of depressive symptoms. A score of 0–5 indicates the absence of depressive symptoms, a score of 6–10 indicates a moderate severity of depressive symptoms and a score of 11–15 indicates severe depression.[32] The validity of GDS15 was assessed by meta-analysis of the 69 studies identified.[33]

The following hypothesis was tested: the IADL, ADL, FCV-19S, AMTS, and GDS15 scores depend on age (24–64 years vs. 65–97 years) and gender (female vs. male). Other hypotheses were: the IADL and ADL scores correlate with the AMTS scores; the AMTS scores correlate with the FCV-19S results; the IADL and ADL scores correlate with the FCV-19S results; the FCV-19S scores correlate with survival; the GDS15 scores correlate with survival; the IADL and ADL scores correlate with survival; and the FCV-19S results correlate with the length of the hospital stay.

All the scales used in the study had been proved to be valid and reliable in the population corresponding to the study group of patients.[22,24,26,31,33,34]

Statistical analysis

For the assessment of the sample size, we used convenience sampling. The study was conducted within a specific timeframe from November 2020 to March 2022 in the 4th Military Teaching Hospital and the Independent Public Healthcare Center of the Ministry of the Interior and Administration, Wroclaw, Poland. We included all patients available at that time and place who met the study inclusion criteria; therefore, our sample was the largest possible. The analysis of quantitative variables was carried out by calculating means and standard deviations (*M* ±*SD*), and medians and interquartile ranges (*Me* (*IQR*)). Qualitative variables were analyzed by calculating frequencies and percentages (*n* (%)). The values of quantitative variables were compared between the 2 groups (patients aged 24–64 years vs. 65–97 years) using the Mann–Whitney *U* test. Correlations between quantitative variables were analyzed using Spearman's correlation coefficient (*r*). A univariate analysis of the impact of a number of variables on a dichotomous variable was carried out using logistic regression. The results are reported as odds ratio (*OR*) values with a 95% confidence interval (*CI*). Statistical significance was set at 0.05. Thus, all *p*-values of less than 0.05 were interpreted as indicating significant relationships.

The comparison of the values of quantitative variables in the 2 groups was made using the Mann–Whitney *U* test, since the data being analyzed did not have a normal distribution (as checked with the Shapiro–Wilk test). Correlations between quantitative variables were analyzed using Spearman's correlation coefficient, since the data did not have a normal distribution (as checked with the Shapiro–Wilk test). A univariate analysis of the influence of many variables on a binary variable was performed using the logistic regression method. The results are presented as *OR* parameter values with a 95% *CI*, as the modelled variable was a two-state one.

The analysis was carried out using the R software, v. 4.1.3 (https://www.r-project.org). Logistic regression was performed by entering the appropriate command (glm) in the R program, which then performed calculations according to the formulas.[35]

Results

Of the 300 patients included in the study, 161 were female and 139 were male. The mean age of the patients was 70.41 years. Eighty-one patients were aged 64 or below, and 219 patients were aged 65 or above. Of the 300 patients examined, 34 died during hospitalization (the mortality rate was 11.33%). Among the variables examined, there were dependent variables, including death, and independent variables – the IADL, ADL, FCV-19S, AMTS, and GDS15 scores, which were treated as continuous variables, so in the analysis of their impact on mortality they were not divided into categories. Hence, there is no calculation of the number of deaths in each category.

The Lawton IADL results showed that of the 300 patients included in the study, 153 (51.00%) were partially independent, 145 (48.33%) were fully independent, and 2 (0.67%) were fully dependent in IADL. The Katz ADL results showed that of the 300 patients studied, 288 (96.00%) were fully functional, 6 (2.00%) had a significant degree of ADL disability, and 6 (2.00%) showed a moderate degree of ADL disability. The characteristics of the study group, including the results of the IADL and ADL independence assessment, are presented in Table 1.

The mean score of the patients on FCV-19S was 18.93 ±5.61, i.e., 2.7 points per question (rounded to 3). The minimum score was 7, while the maximum score was 35 points. The mean score of 18.93 is interpreted in the Polish adaptation of the scale as indicating a low level of fear,[27,28] while in the European adaptation, the score is above the cut-off point; therefore, these patients should be evaluated further to assess their mental well-being.[29]

The AMTS results showed that of the 300 patients included in the study, 196 (65.33%) had normal cognitive function, 62 (20.67%) had mild cognitive impairment,

Table 1. Characteristics of the study group

Scale	*N*	Data gaps	*M* ±*SD*	*Me* (*IQR*)	min	max	Score	Interpretation	*n* (%)
Lawton IADL scale	300	0	21.01 ±4.32	23 (20–24)	8	25	8	fully dependent	2 (0.67)
							9–23	partially dependent	153 (51.00)
							24	fully independent	145 (48.33)
Katz ADL index of independence	300	0	5.80 ±0.87	6 (6–6)	0	6	0–2	significant degree of disability	6 (2.00)
							3–4	moderate degree of disability	6 (2.00)
							5–6	fully functional	288 (96.00)
AMTS	300	0	8.37 ±2.13	9 (8–10)	0	10	0–3	severe cognitive impairment	17 (5.67)
							4–6	moderate cognitive impairment	25 (8.33)
							7–8	mild cognitive impairment	62 (20.67)
							9–10	normal cognitive function	196 (65.33)
GDS15	219	0	4.07 ±3.09	4 (2–6)	0	14	0–5	absence of depressive symptoms	156 (71.23)
							6–10	moderate severity of depressive symptoms	56 (25.57)
							11–15	severe depression	7 (3.20)

M – mean; *SD* – standard deviation. *Me* – median; *IQR* – interquartile range; min – minimum; max – maximum; IADL – instrumental activities of daily living; ADL – activities of daily living; AMTS – abbreviated mental test score; GDS15 – 15-item geriatric depression scale.

25 (8.33%) had moderate cognitive impairment, and 17 (5.67%) had severe cognitive impairment (Table 1). The analysis showed that the level of cognitive functions in geriatric patients differed from that in younger patients. Mental performance was significantly higher in the under-65 age group ($p < 0.001$) (Table 2).

Using GDS15, the authors assessed only the group of geriatric patients (≥65 years old) without making a comparison to the younger group, and this was due to the reliability of this scale only in a geriatric group of patients. The GDS15 results showed that of the 219 patients studied, 156 (71.23%) had no depressive symptoms, 56 (25.57%) displayed a moderate severity of depressive symptoms, and 7 (3.20%) had severe depression (Table 1).

Our analysis showed certain statistically significant correlations ($p < 0.05$). Based on the results, we may notice that age is an important factor affecting the level of independence among patients. Patients aged 64 or below reported significantly greater independence on the IADL scale and the basic ADL scale, and showed a significantly higher level of mental fitness. The correlations between age and independence in IADL, independence in basic ADL and mental fitness are collected and presented in Table 2. Our analysis using the Mann–Whitney *U* test showed that gender was a factor influencing the level of fear of COVID-19 ($p = 0.001$). Female patients reported a significantly higher fear of COVID-19 as compared to male patients. The mean score on FCV-19S in the female group ($N = 161$) was 19.84 ±5.60, while in the male group ($N = 139$) it was 17.88 ±5.45.

The analysis showed statistically significant relationships ($p < 0.05$), indicating that the higher the level of mental fitness, the higher the level of IADL and ADL independence. The correlation between the AMTS and Lawton IADL scale scores was $r = 0.438$, whereas between the AMTS and Katz ADL scale scores it was $r = 0.270$.

Our study presents a novel, previously unpublished result regarding the impact of IADL and ADL independence on in-hospital mortality. It shows that patient survival and

Table 2. Statistically significant relationships between age and independence in instrumental activities of daily living (IADL) and basic activities of daily living (ADL) and mental fitness

Variable	Age [years]	*M* ±*SD*	*Me* (*IQR*)	*p*-value
Lawton IADL scale score	≤64 (*N* = 81)	23.25 ±2.28	24 (24–24)	<0.001*
	≥65 (*N* = 219)	20.18 ±4.60	22 (17–24)	
Katz ADL scale score	≤64 (*N* = 81)	5.99 ±0.11	6 (6–6)	0.007*
	≥65 (*N* = 219)	5.73 ±1.00	6 (6–6)	
AMTS score	≤64 (*N* = 81)	9.33 ±1.13	10 (9–10)	<0.001*
	≥65 (*N* = 219)	8.01 ±2.29	9 (7–10)	

* statistically significant (Mann –Whitney *U* test).

in-hospital mortality are influenced by independence in basic and complex activities of daily living. The dichotomous variable was the variable determining whether the patient died or not. The FCV-19S and GDS15 scores did not affect the dichotomous outcome, each point on the Lawton IADL scale reduced the chance of death by 11.1%, and each point on the Katz ADL scale reduced the chance of death by 31% ($p < 0.05$). Table 3 demonstrates the correlations between the IADL and ADL scores and the likelihood of death. Therefore, the higher the IADL and ADL scores, the higher chance of patient survival.

No statistically significant relationships ($p > 0.05$) were found between age and the level of fear of COVID-19, between gender and the level of IADL and ADL independence, between gender and the level of mental fitness, between gender and the severity of depressive symptoms, between mental fitness and the level of fear of COVID-19, between the level of fear COVID-19 as well as the severity of depressive symptoms and survival, and between the level of fear of COVID-19 and the length of the hospital stay. In conclusion, gender had an impact on the level of fear of COVID-19, but it had no significance on the level of independence and mental fitness, and the severity of depression. There was no significant correlation between age and the level of fear of COVID-19. The intensity of fear of COVID-19 did not significantly affect the level of cognitive functions, the level of independence (IADL, ADL), survivability, and the length of the hospital stay. The age of the patients showed a significant correlation with the level of independence and the level of cognitive functions. However, age did not determine the level of fear of the disease. The statistically significant and non-significant relationships and correlations are presented in Tables 4 and 5.

Table 3. Correlations between the independence in instrumental activities of daily living (IADL) and basic activities of daily living (ADL) scores and the likelihood of death

Variable	*OR*	95% *CI*	*p*-value
Lawton IADL scale score	0.889	0.829–0.953	0.001*
Katz ADL scale score	0.690	0.522–0.914	0.010*

OR – odds ratio; *CI* – confidence interval; * statistically significant (univariate logistic regression).

Table 5. Statistically non-significant correlations with regard to the fear of COVID-19

Correlation	*r*	*p*-value
IADL vs. FCV-19S	–0.043	0.460
ADL vs. FCV-19S	–0.045	0.436
AMTS vs. FCV-19S	0.014	0.807
FCV-19S vs. the length of hospitalization	0.051	0.379

r – Spearman's correlation coefficient.

Discussion

Illnesses requiring hospital admission, and in particular those requiring intensive care management, have a negative impact on the well-being of patients.[36] According to the available literature, up to 67% of hospitalized patients suffer from symptoms of anxiety and depression, and 45% are diagnosed with PTSD,[37–48] which has a negative impact on their health-related quality of life (HRQoL).[40,44,45,47]

COVID-19 patients hospitalized in ICUs suffer from stress and anxiety. A study by Kupeli et al. found that 37.1% of the COVID-19 patients studied showed symptoms of anxiety and 43.6% showed symptoms of depression in the first 24 h of admission to ICU.[49] Such a response to a stressor is considered acute stress disorder (ASD). This disorder is associated with feelings of great uncertainty, vulnerability and even fear resulting from exposure to a potentially fatal event.[50] COVID-19 patients hospitalized in ICUs need to be kept isolated from other patients, which may increase their feelings of uncertainty and vulnerability. They do not know what is going to happen to them next. Moreover, the personal protective equipment (PPE) that must be worn by health workers while treating patients with COVID-19 can evoke negative connotations, making patients aware of the seriousness of their situation.

Studies have shown that certain demographic features and factors relating to the hospital stay itself can have a negative impact on the mental well-being of patients. These include a low level of education, being unemployed, female gender, the duration of the stay in ICU, and disease severity.[41,51–53]

Table 4. Comparison of scores on various scales with regard to age and gender (statistically significant and non-significant relationships)

Variable		Lawton IADL scale score	Katz ADL scale score	AMTS score	FCV-19S score	GDS15 score
Age [years]	≤64 (*N* = 81)	23.25 ±2.28	5.99 ±0.11	9.33 ±1.13	18.48 ±4.60	–
	≥65 (*N* = 219)	20.18 ±4.60	5.73 ±1.00	8.01 ±2.29	19.10 ±5.94	–
p-value		<0.001*	0.007*	<0.001*	0.675	–
Gender	F (*N* = 161)	20.96 ±4.09	5.75 ±0.95	8.17 ±2.30	19.84 ±5.60	4.18 ±3.11
	M (*N* = 139)	21.07 ±4.58	5.86 ±0.76	8.60 ±1.89	17.88 ±5.45	3.94 ±3.08
p-value		0.167	0.060	0.112	0.001*	0.583

Data presented as *M* ±*SD*.
FCV-19S – fear of COVID-19 scale; F – female; M – male; * statistically significant (Mann –Whitney *U* test).

In their study, Daltaban and Aytekin showed that there is a strong association between female gender and an increased level of fear during the pandemic.[54] Similarly, the present study found a correlation between female gender and a greater fear of COVID-19. Women are more likely to respond more emotionally to such difficulties. Moreover, given the different life roles of men and women, the fear of infection experienced by women may be due to their concerns that no one would replace them in taking care of their children, grandchildren or living partners, or in carrying out their duties if they became ill. Studies suggest that women's greater sensitivity and emotionality, which are dependent on the level of sex hormones, may be an influencing factor. However, we cannot name the exact causes of women's higher levels of fear. Therefore, the suggested factors are the subject of research; we propose to consider them a hypothesis.[8,9,55–57]

In our study, we found no relationship between age and the level of fear of COVID-19. Kaya and Bayındır observed a higher level of fear during the COVID-19 pandemic among geriatric patients, and suggested that healthcare professionals should help patients normalize the level of fear with physical activity and social support.[58] In a study by Nino et al., the researchers noticed that the level of anxiety increased with age, which might be related to more frequent comorbidities in older people.[59] However, these results have not been confirmed in every ethnic group studied. In a study by Lin et al.[25] and also one by Łazarz-Półkoszek et al.[60], children and older people showed a lower level of anxiety than young or middle-aged adults, which was attributed to their different perceptions of the disease and its impact on health and social functioning, including financial functioning.

In addition, the mental fitness of patients and their level of IADL independence prior to hospitalization have an impact on their perception of the disease and their awareness of the risk it poses. The present study showed that patients aged over 64 exhibited a lower level of cognitive function and were less independent in activities of daily living as compared to younger patients. Moreover, the study found that the level of mental fitness and the level of independence in daily life influence each another, and that there is a statistically significant correlation between them. This also has implications for the patient's hospital stay, and especially their prognosis. In the present study, a higher level of independence was found to be associated with a lower likelihood of death. Bruno et al. concluded in their study that the Katz ADL index of independence provides additional information that can help assess the risk of in-hospital death in COVID-19 patients.[61] Patients with low Katz ADL scores, i.e., those with limitations in ADL, were found to be at a particularly high risk of deah.[61] Similarly, Ting-Jie et al.[62] and Ocagli et al.[63] noted in their studies that the Barthel index could be used as a prognostic indicator for mortality. A study by Bo et al. confirms the assumed hypothesis regarding the impact of independence and the quality of cognitive function on the prognosis of geriatric patients hospitalized in ICUs; the lower the level of self-reliance and high or moderate cognitive impairment, the higher the in-hospital mortality.[17]

With age, the loss of interneuronal networks and brain atrophy can be observed. It is often associated with a cognitive decline. However, cognitive impairment may also be associated with dementia, due to damage to the vessels, brain tumors, post-stroke changes, or the impairment of the dopaminergic system, or with potentially reversible causes, such as depression, endocrine causes, such as hypothyroidism, or vitamin B12 deficiency.[64–67] Older age also affects the level of independence, but it is not the only important factor. Depression, a low level of social support, not living and participating in social relationships, as well as the impairment of cognitive functions, also reduce independence. There is an age–cognition–independence connection, where all factors influence each other.[68,69]

In addition to a low level of independence, other risk factors for in-hospital death reported in the available literature include dementia, cognitive impairment and older age (>85 years).[70] It has been observed that older age is not a sufficient indicator of the risk of death. The independence of the patient and their functioning in everyday life are more important indicators. Some geriatric patients live alone, some rely on support from their families or professionals, and some live in care homes. However, this does not mean that the impairment of functioning is the main cause of death. Independence is also influenced by factors such as comorbidities and the severity of their symptoms, as well as cognitive impairment.[71] It is these factors that are crucial in determining risk factors for mortality in older COVID-19 patients.[72] Elderly COVID-19 patients under intensive care, especially those with impaired physical or mental function, require special care and support.[53,62]

The mental state of patients, and their levels of independence and cognitive function have a significant impact on the course of hospitalization and treatment. Clinicians should pay attention to the presence of risk factors for increased mortality in these patients and provide them with appropriate care. Based on our research and the literature cited, we can conclude that acute physiological impairment is not the most important prognostic factor. Despite treating the patient according to the established guidelines, the effect on each patient may vary. We hope that clinicians, especially in ICUs, will pay attention to the factors mentioned in the present article.

Determining these factors on admission to hospital is of great prognostic importance and should lead to modifications in patient care so that the therapeutic effect is as good as possible. Our work proposes future research directions in searching for factors that would improve patients' cognitive function and level of independence. We recommend focusing on the role of social and family

relationships and psychological care during the ICU stay, as well as on minimizing the state of isolation to improve patients' mental well-being. Our research suggests a wider use of scales (the Lawton IADL scale, the Katz ADL scale and AMTS) to assess the levels of independence and cognitive function on hospital admission. Whilst assessing cognitive function, we recommend keeping in mind the potentially reversible causes of dementia.

It is essential to approach patients holistically so that, in addition to the therapeutic effect, patients' subsequent quality of life would be as good as possible. It is worth considering what appropriate measures could be taken to improve the condition of patients at an early stage. The mental state, the level of cognitive function and the level of independence of patients seem difficult to modify. However, an attempt to improve them or to implement appropriate treatment would contribute to a better functioning of patients, their better survival, and from a far perspective, a better functioning of the wards.

Limitations

The results of the study were based on observations. A potential limitation to this work is a small cohort group and a smaller group of patients under 65 years. Due to the low expected values resulting from the insufficient number of patients, the conclusions presented in this study should be treated cautiously. Another potential limitation is not including dependent variables, such as comorbidities, disease severity, race or ethnicity, the socioeconomic status, medications, and the length of the hospital stay, in the assessment. Based on the current literature, the influence of the level of education, unemployment and disease severity on the well-being of patients has been noted. Our study primarily addressed factors such as independence, the level of fear of COVID-19 and the level of cognitive function. A limitation to our work is not mentioning other possible factors that may affect the well-being of patients. However, as we could not obtain such data from all respondents, it was impossible to analyze those factors with due reliability. We suggest that future research on this topic should investigate the factors mentioned above. We recommend conducting a larger and more age-differentiated population study over a longer period of time.

Conclusions

The level of independence in basic and instrumental activities of daily living is associated with in-hospital mortality in geriatric COVID-19 patients, which is a novel conclusion, not published in the previous literature.

A higher level of cognitive function is positively correlated with higher levels of independence in basic and instrumental activities of daily living.

Patients aged 65 and older are more likely to be less independent in basic and instrumental activities of daily living as compared to younger patients. Moreover, this cohort group exhibits a statistically significantly lower level of cognitive function.

These findings highlight the importance of considering both cognitive function and independence in daily living activities when planning care and treatment strategies for geriatric COVID-19 patients.

Ethics approval and consent to participate

The study was approved by the relevant bioethics committee (Military Medical Chamber, Warsaw, Poland; approval No. KB-191/22). All patients provided written informed consent to participate in the study.

Data availability

The datasets supporting the findings of the current study are available from the corresponding author on reasonable request.

Consent for publication

Not applicable.

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References

1. World Health Organization (WHO). WHO COVID-19 dashboard. Geneva, Switzerland. 2020. https://covid19.who.int. Accessed July 21, 2023.
2. Rajkumar RP. COVID-19 and mental health: A review of the existing literature. *Asian J Psychiatr.* 2020;52:102066. doi:10.1016/j.ajp.2020.102066
3. Czeisler MÉ, Lane RI, Petrosky E, et al. Mental health, substance use, and suicidal ideation during the COVID-19 pandemic – United States, June 24–30, 2020. *MMWR Morb Mortal Wkly Rep*. 2020;69(32):1049–1057. doi:10.15585/mmwr.mm6932a1
4. Tingey JL, Bentley JA, Hosey MM. COVID-19: Understanding and mitigating trauma in ICU survivors. *Psychol Trauma*. 2020;12(S1):S100–S104. doi:10.1037/tra0000884
5. Hosey MM, Needham DM. Survivorship after COVID-19 ICU stay. *Nat Rev Dis Primers*. 2020;6(1):60. doi:10.1038/s41572-020-0201-1
6. Abad C, Fearday A, Safdar N. Adverse effects of isolation in hospitalised patients: A systematic review. *J Hosp Infect*. 2010;76(2):97–102. doi:10.1016/j.jhin.2010.04.027
7. Schittek GA, Bornemann-Cimenti H, Sandner-Kiesling A. Well-being of ICU patients with COVID-19. *Intensive Crit Care Nurs*. 2021;65:103050. doi:10.1016/j.iccn.2021.103050
8. Metin A, Erbiçer ES, Şen S, Çetinkaya A. Gender and COVID-19 related fear and anxiety: A meta-analysis. *J Affect Disord*. 2022;310:384–395. doi:10.1016/j.jad.2022.05.036
9. Sürme Y, Özmen N, Arik BE. Fear of COVID-19 and related factors in emergency department patients. *Int J Ment Health Addict*. 2023;21(1):28–36. doi:10.1007/s11469-021-00575-2

10. Sivanathan L, Wunsch H, Vigod S, Hill A, Pinto R, Scales DC. Mental illness after admission to an intensive care unit. *Intensive Care Med*. 2019;45(11):1550–1558. doi:10.1007/s00134-019-05752-5
11. Vlake JH, Van Bommel J, Wils EJ, et al. Effect of intensive care unit-specific virtual reality (ICU-VR) to improve psychological well-being and quality of life in COVID-19 ICU survivors: A study protocol for a multicentre, randomized controlled trial. *Trials*. 2021;22(1):328. doi:10.1186/s13063-021-05271-z
12. Jubran A, Lawm G, Kelly J, et al. Depressive disorders during weaning from prolonged mechanical ventilation. *Intensive Care Med*. 2010;36(5):828–835. doi:10.1007/s00134-010-1842-4
13. Wintermann GB, Weidner K, Strauss B, Rosendahl J. Rates and predictors of mental health care utilisation in patients following a prolonged stay on intensive care unit: A prospective cohort study. *BMJ Open*. 2023;13(1):e063468. doi:10.1136/bmjopen-2022-063468
14. Rose L, Nonoyama M, Rezaie S, Fraser I. Psychological wellbeing, health related quality of life and memories of intensive care and a specialised weaning centre reported by survivors of prolonged mechanical ventilation. *Intensive Crit Care Nurs*. 2014;30(3):145–151. doi:10.1016/j.iccn.2013.11.002
15. Engström Å, Nyström N, Sundelin G, Rattray J. People's experiences of being mechanically ventilated in an ICU: A qualitative study. *Intensive Crit Care Nurs*. 2013;29(2):88–95. doi:10.1016/j.iccn.2012.07.003
16. Arslanian-Engoren C, Scott LD. The lived experience of survivors of prolonged mechanical ventilation: A phenomenological study. *Heart Lung*. 2003;32(5):328–334. doi:10.1016/s0147-9563(03)00043-8
17. Bo M, Massaia M, Raspo S, et al. Predictive factors of in-hospital mortality in older patients admitted to a medical intensive care unit. *J Am Geriatr Soc*. 2003;51(4):529–533. doi:10.1046/j.1532-5415.2003.51163.x
18. Pisani MA, McNicoll L, Inouye SK. Cognitive impairment in the intensive care unit. *Clin Chest Med*. 2003;24(4):727–737. doi:10.1016/s0272-5231(03)00092-3
19. Smits CH, Deeg DJ, Kriegsman DM, Schmand B. Cognitive functioning and health as determinants of mortality in an older population. *Am J Epidemiol*. 1999;150(9):978–986. doi:10.1093/oxfordjournals.aje.a010107
20. Graf C. The Lawton instrumental activities of daily living scale. *Am J Nurs*. 2008;108(4):52–62. doi:10.1097/01.NAJ.0000314810.46029.74
21. Millán-Calenti JC, Tubío J, Pita-Fernández S, et al. Prevalence of functional disability in activities of daily living (ADL), instrumental activities of daily living (IADL) and associated factors, as predictors of morbidity and mortality. *Arch Gerontol Geriatr*. 2010;50(3):306–310. doi:10.1016/j.archger.2009.04.017
22. Vittengl JR, White CN, McGovern RJ, Morton BJ. Comparative validity of seven scoring systems for the instrumental activities of daily living scale in rural elders. *Aging Ment Health*. 2006;10(1):40–47. doi:10.1080/13607860500307944
23. Pashmdarfard M, Azad A. Assessment tools to evaluate Activities of Daily Living (ADL) and Instrumental Activities of Daily Living (IADL) in older adults: A systematic review. *Med J Islam Repub Iran*. 2020;34:33. doi:10.34171/mjiri.34.33
24. Wallace M, Shelkey M. Reliability and validity of Katz ADL index. *Am J Nurs*. 2008;108(4). doi:10.1097/01.NAJ.0000315266.66521.e7
25. Lin CY, Hou WL, Mamun MA, et al. Fear of COVID-19 Scale (FCV-19S) across countries: Measurement invariance issues. *Nurs Open*. 2021;8(4):1892–1908. doi:10.1002/nop2.855
26. Pilch I, Kurasz Z, Turska-Kawa A. Experiencing fear during the pandemic: Validation of the fear of COVID-19 scale in Polish. *PeerJ*. 2021;9:e11263. doi:10.7717/peerj.11263
27. Grajek M, Krupa-Kotara K, Rozmiarek M, et al. The level of COVID-19 anxiety among oncology patients in Poland. *Int J Environ Res Public Health*. 2022;19(18):11418. doi:10.3390/ijerph191811418
28. Grajek M, Działach E, Buczkowska M, Górski M, Nowara E. Feelings related to the COVID-19 pandemic among patients treated in the oncology clinics (Poland). *Front Psychol*. 2021;12:647196. doi:10.3389/fpsyg.2021.647196
29. Nikopoulou VA, Holeva V, Parlapani E, et al. Mental health screening for COVID-19: A proposed cutoff score for the Greek version of the Fear of COVID-19 Scale (FCV-19S). *Int J Ment Health Addict*. 2020;20(2):907–922. doi:10.1007/s11469-020-00414-w
30. Pendlebury ST, Klaus SP, Mather M, De Brito M, Wharton RM. Routine cognitive screening in older patients admitted to acute medicine: Abbreviated mental test score (AMTS) and subjective memory complaint versus Montreal Cognitive Assessment and IQCODE. *Age Ageing*. 2015;44(6):1000–1005. doi:10.1093/ageing/afv134
31. Piotrowicz K, Romanik W, Skalska A, et al. The comparison of the 1972 Hodkinson's Abbreviated Mental Test Score (AMTS) and its variants in screening for cognitive impairment. *Aging Clin Exp Res*. 2019;31(4):561–566. doi:10.1007/s40520-018-1009-7
32. Mowla A, Ghaedsharaf M, Pani A. Psychopathology in elderly COVID-19 survivors and controls. *J Geriatr Psychiatry Neurol*. 2022;35(3):467–471. doi:10.1177/08919887211002664
33. Mitchell AJ, Bird V, Rizzo M, Meader N. Diagnostic validity and added value of the Geriatric Depression Scale for depression in primary care: A meta-analysis of GDS30 and GDS15. *J Affect Disord*. 2010;125(1–3):10-17. doi:10.1016/j.jad.2009.08.019
34. Brorsson B, Asberg KH. Katz index of independence in ADL. Reliability and validity in short-term care. *Scand J Rehabil Med*. 1984;16(3):125–132. PMID:6494836.
35. R Core Team. A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria. 2022. https://www.r-project.org. Accessed November 12, 2023.
36. Vlake JH, Wesselius S, Van Genderen ME, Van Bommel J, Boxma-de Klerk B, Wils EJ. Psychological distress and health-related quality of ife in patients after hospitalization during the COVID-19 pandemic: A single-center, observational study. *PloS One*. 2021;16(8):e0255774. doi:10.1371/journal.pone.0255774
37. Kendrick D, Dhiman P, Kellezi B, et al. Psychological morbidity and return to work after injury: Multicentre cohort study. *Br J Gen Pract*. 2017;67(661):e555–e564. doi:10.3399/bjgp17X691673
38. Paredes Molina CS, Berry S, Nielsen A, Winfield R. PTSD in civilian populations after hospitalization following traumatic injury: A comprehensive review. *Am J Surg*. 2018;216(4):745–753. doi:10.1016/j.amjsurg.2018.07.035
39. Sheldrick R, Tarrier N, Berry E, Kincey J. Post-traumatic stress disorder and illness perceptions over time following myocardial infarction and subarachnoid haemorrhage. *Br J Health Psychol*. 2006;11(Pt 3):387–400. doi:10.1348/135910705X71434
40. Van Seben R, Covinsky KE, Reichardt LA, et al. Insight into the posthospital syndrome: A 3-month longitudinal follow up on geriatric syndromes and their association with functional decline, readmission, and mortality. *J Gerontol A Biol Sci Med Sci*. 2020;75(7):1403–1410. doi:10.1093/gerona/glaa039
41. Visser E, Gosens T, Den Oudsten BL, De Vries J. The course, prediction, and treatment of acute and posttraumatic stress in trauma patients: A systematic review. *J Trauma Acute Care Surg*. 2017;82(6):1158–1183. doi:10.1097/TA.0000000000001447
42. Walker J, Burke K, Wanat M, et al. The prevalence of depression in general hospital inpatients: A systematic review and meta-analysis of interview-based studies. *Psychol Med*. 2018;48(14):2285–2298. doi:10.1017/S0033291718000624
43. Davydow DS, Desai SV, Needham DM, Bienvenu OJ. Psychiatric morbidity in survivors of the acute respiratory distress syndrome: A systematic review. *Psychosom Med*. 2008;70(4):512–519. doi:10.1097/PSY.0b013e31816aa0dd
44. Gerth AM, Hatch RA, Young JD, Watkinson PJ. Changes in health-related quality of life after discharge from an intensive care unit: A systematic review. *Anaesthesia*. 2019;74(1):100–108. doi:10.1111/anae.14444
45. Nikayin S, Rabiee A, Hashem MD, et al. Anxiety symptoms in survivors of critical illness: A systematic review and meta-analysis. *Gen Hosp Psychiatry*. 2016;43:23–29. doi:10.1016/j.genhosppsych.2016.08.005
46. Rabiee A, Nikayin S, Hashem MD, et al. Depressive symptoms after critical illness: A systematic review and meta-analysis. *Crit Care Med*. 2016;44(9):1744–1753. doi:10.1097/CCM.0000000000001811
47. Oeyen SG, Vandijck DM, Benoit DD, Annemans L, Decruyenaere JM. Quality of life after intensive care: A systematic review of the literature. *Crit Care Med*. 2010;38(12):2386–2400. doi:10.1097/CCM.0b013e3181f3dec5

48. Righy C, Rosa RG, Amancio da Silva RT, et al. Prevalence of post-traumatic stress disorder symptoms in adult critical care survivors: A systematic review and meta-analysis. *Crit Care*. 2019;23(1):213. doi:10.1186/s13054-019-2489-3
49. Kupeli I, Kara MY, Yakın I, Caglayan AC. Anxiety and depression in the first 24 h in COVID-19 patients who underwent non-invasive mechanical ventilation in the intensive care unit. *Ir J Med Sci*. 2022;191(5):2291–2295. doi:10.1007/s11845-021-02808-8
50. Bryant RA. Acute stress disorder. *Curr Opin Psychol*. 2017;14:127–131. doi:10.1016/j.copsyc.2017.01.005
51. Davydow DS, Katon WJ, Zatzick DF. Psychiatric morbidity and functional impairments in survivors of burns, traumatic injuries, and ICU stays for other critical illnesses: A review of the literature. *Int Rev Psychiatry*. 2009;21(6):531–538. doi:10.3109/09540260903343877
52. De Vries GJ, Olff M. The lifetime prevalence of traumatic events and posttraumatic stress disorder in the Netherlands. *J Trauma Stress*. 2009;22(4):259–267. doi:10.1002/jts.20429
53. De Graaf MA, Antoni ML, Ter Kuile MM, et al. Short-term outpatient follow-up of COVID-19 patients: A multidisciplinary approach. *EClinicalMedicine*. 2021;32:100731. doi:10.1016/j.eclinm.2021.100731
54. Daltaban Ö, Aytekin Z. Fear and anxiety of COVID-19 in dental patients during the COVID-19 pandemic: A cross-sectional survey in Turkey. *Dent Med Probl*. 2022;59(3):343–350. doi:10.17219/dmp/150075
55. Albert KM, Newhouse PA. Estrogen, stress, and depression: Cognitive and biological interactions. *Annu Rev Clin Psychol*. 2019;15:399–423. doi:10.1146/annurev-clinpsy-050718-095557
56. Li SH, Graham BM. Why are women so vulnerable to anxiety, trauma-related and stress-related disorders? The potential role of sex hormones. *Lancet Psychiatry*. 2017;4(1):73–82. doi:10.1016/S2215-0366(16)30358-3
57. Rubinow DR, Schmidt PJ. Sex differences and the neurobiology of affective disorders. *Neuropsychopharmacology*. 2019;44(1):111–128. doi:10.1038/s41386-018-0148-z
58. Kaya N, Bayındır F. Evaluation of the relationship between the geriatric anxiety and COVID-19 anxiety and fear levels in geriatric dental patients during the COVID-19 pandemic. *Dent Med Probl*. 2023;60(1):5–11. doi:10.17219/dmp/157345
59. Niño M, Harris C, Drawve G, Fitzpatrick KM. Race and ethnicity, gender, and age on perceived threats and fear of COVID-19: Evidence from two national data sources. *SSM Popul Health*. 2021;13:100717. doi:10.1016/j.ssmph.2020.100717
60. Łazarz-Półkoszek MJ, Orczykowska M, Gala A, Pihut M. Impact of the COVID-19 pandemic on patients' anxiety levels related to dental appointments in Poland. *Dent Med Probl*. 2023;60(3):367–373. doi:10.17219/dmp/163476
61. Bruno RR, Wernly B, Flaatten H, et al. The association of the Activities of Daily Living and the outcome of old intensive care patients suffering from COVID-19. *Ann Intensive Care*. 2022;12(1):26. doi:10.1186/s13613-022-00996-9
62. Ting-Jie I, Tsai YL, Cheng YY. Predictors of basic activity in daily living and length of hospitalization in patients with COVID-19. *Healthcare*. 2022;10(8):1589. doi:10.3390/healthcare10081589
63. Ocagli H, Cella N, Stivanello L, Degan M, Canova C. The Barthel index as an indicator of hospital outcomes: A retrospective cross-sectional study with healthcare data from older people. *J Adv Nurs*. 2021;77(4):1751–1761. doi:10.1111/jan.14708
64. Murman DL. The impact of age on cognition. *Semin Hear*. 2015;36(3):111–121. doi:10.1055/s-0035-1555115
65. Flicker L, Ames D. Metabolic and endocrinological causes of dementia. *Int Psychogeriatr*. 2005;17(Suppl 1):S79–S92. doi:10.1017/s1041610205001961
66. Paterniti S, Verdier-Taillefer MH, Dufouil C, Alpérovitch A. Depressive symptoms and cognitive decline in elderly people: Longitudinal study. *Br J Psychiatry*. 2002;181:406–410. doi:10.1192/bjp.181.5.406
67. Iadecola C. The pathobiology of vascular dementia. *Neuron*. 2013;80(4):844–866. doi:10.1016/j.neuron.2013.10.008
68. Chen SW, Chippendale T. Factors associated with IADL independence: Implications for OT practice. *Scand J Occup Ther*. 2017;24(2):109–115. doi:10.1080/11038128.2016.1194464
69. Beltz S, Gloystein S, Litschko T, Laag S, Van den Berg N. Multivariate analysis of independent determinants of ADL/IADL and quality of life in the elderly. *BMC Geriatr*. 2022;22(1):894. doi:10.1186/s12877-022-03621-3
70. Covino M, De Matteis G, Della Polla DA, et al. Predictors of in-hospital mortality and death risk stratification among COVID-19 patients aged ≥80 years old. *Arch Gerontol Geriatr*. 2021;95:104383. doi:10.1016/j.archger.2021.104383
71. Zekry D, Loures Valle BH, Lardi C, et al. Geriatrics index of comorbidity was the most accurate predictor of death in geriatric hospital among six comorbidity scores. *J Clin Epidemiol*. 2010;63(9):1036–1044. doi:10.1016/j.jclinepi.2009.11.013
72. Mendes A, Serratrice C, Herrmann FR, et al. Predictors of in-hospital mortality in older patients with COVID-19: The COVIDAge study. *J Am Med Dir Assoc*. 2020;21(11):1546–1554.e3. doi:10.1016/j.jamda.2020.09.014

Original paper

Lesions located on the tongue after SARS-CoV-2 infection: A retrospective study

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Abstract

Background. The post-coronavirus disease (post-COVID) syndrome (PCS), which occurs after severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) infection, can manifest a variety of symptoms in the oral cavity. Changes to the tongue tend to persist longer than other symptoms in this area.

Objectives. The aim of the study was to present the changes and lesions that occur on the tongue after SARS-CoV-2 infection, as well as their healing as a consequence of the therapy used or lack thereof.

Material and methods. The study sample included 426 individuals who had contracted SARS-CoV-2 and presented with changes on the tongue. Periodic checkups enabled to determine their variability and duration in response to treatment or lack thereof.

Results. The presence of various oral manifestations was reported, including strawberry tongue (women (F): 143; men (M): 65), depapillation (F: 86, M: 156), geographic tongue (F: 65, M: 124), vascular changes (F: 102, M: 46), aphthous changes (F: 106, M: 58), candidiasis (F: 89, M: 57), edema (F: 42, M: 52), herpetic lesions (F: 38, M: 49), ulcers (F: 38, M: 46), erosions (F: 32, M: 28), nodular lesions (F: 6, M: 19), and necrotic changes (F: 9, M: 7). Fungiform papillae were found to be enlarged in 189 women and 213 men. On average, from 3 to 5 changes were identified concurrently. In the majority of cases, the changes disappeared on their own and persisted from 4 weeks to 36 months. In 20% of cases, they recurred. Local therapy resulted in a 50% reduction in the duration of PCS.

Conclusions. Changes that manifest on the tongue require observation and basic or specialized treatment. In the absence of pain, monitoring is recommended for a period of 4 weeks, after which a spontaneous disappearance should be expected. In the event that various changes occur in the oral cavity, the patient should be referred for specialized treatment.

Keywords: treatment, duration, SARS-CoV-2, long COVID, tongue changes

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Highlights

- Severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) is associated with various tongue pathologies, with emerging variants contributing to their diversity.
- Tongue lesions may present at the onset of infection or as a symptom of long coronavirus disease (COVID).
- A retrospective analysis identified enlarged fungiform papillae, depapillation and strawberry tongue as the most frequent symptoms, with many patients exhibiting from 3 to 5 concurrent pathologies.
- Tongue lesions may resolve spontaneously under observation or require basic to specialized therapeutic intervention.

Introduction

Coronavirus disease 2019 (COVID-19) manifests a variety of symptoms in the oral cavity[1–4] and promotes the appearance of the post-COVID syndrome (PCS). The syndrome occurs in individuals who have been infected with the severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2), persists for a minimum of 2 months, and cannot be explained by an alternative diagnosis. A variety of symptoms may occur in the oral cavity during the acute phase of COVID-19 or after infection, and they are recurrent.[5] Single cases of autoimmune diseases have also been observed in the aftermath of SARS-CoV-2 infection.[6] Oral symptoms may also emerge following COVID-19 vaccination.[7] The mechanism of action of the SARS-CoV-2 is not known. Despite the passage of time, it is not possible to confirm whether the changes located in the oral cavity are the result of the systemic reaction to the viral infection, the congenital immune response to those infections, the action of cytokines,[8,9] or are secondary.[10,11] However, it has been validated that the oral cavity is the gateway and reservoir of the virus because the high concentration of angiotensin-converting enzyme 2 (ACE2) receptors in this area predisposes to the appearance of pathological changes.[12,13] The virus causes the activation of cytokines, leading to cell apoptosis and subsequent loss of taste buds, resulting in dysfunction of taste sensation. It also affects the trajectory of the gustatory tract by damaging cells in peripheral taste neurosensory chemoreceptors and/or by directly damaging cranial nerves (VII, IX and X) that are responsible for taste.[14]

The progression and severity of the infection are also influenced by the patient's oral hygiene, which is inseparably linked to their mental health.[15] A retrospective analysis of patients after the SARS-CoV-2 infection has revealed that tongue lesions tend to persist longer than other changes in this area.

Initially, tongue lesions were observed in 38% of patients with confirmed SARS-CoV-2 infection.[3] The first cases of tongue swelling were reported at the beginning of the pandemic. It affected hospitalized patients with severe COVID-19, intubated patients and Black individuals.[16,17] An image of a strawberry tongue was featured on social media in January 2021.[18] Fungal lesions were also prevalent, resulting from poor hygiene practices. Subsequent examinations revealed depapillation of the tongue (red papilla-free areas surrounded by an irregular white border), swelling, inflammation, ulceration, nodules, and geographic tongue. The manifestation of these symptoms in conjunction with the SARS-CoV-2 infection is referred to as "the COVID tongue."[19,20] However, it remains unclear whether the COVID tongue is an early symptom of the disease or develops with its progression. Geographic tongue resulting from the SARS-CoV-2 infection persists for years without causing pain, only discomfort when consuming spicy foods. Vascular,[21] drug-induced[22] and reinfection-related[23] lesions have also been documented.

A review of the available literature revealed no publications describing the occurrence, treatment and disappearance of all 13 tongue pathologies that appeared throughout the pandemic and were the result of infection with all known variants of the SARS-CoV-2 virus.

The objective of this study is to present the changes that occur on the tongue after SARS-CoV-2 infection, as well as their healing as a consequence of the therapy used or lack thereof.

Material and methods

The study was conducted according to the STROBE (Strengthening the Reporting of Observational Studies in Epidemiology) guidelines. The Bioethical Commission of Medical University of Warsaw, Poland, has approved the scope of this retrospective study (commission statement No. AKBE/318/2023). The medical records of 1,090 patients aged 20–40 who had contracted the virus between 2020 and 2023 were transferred from the IT system of the dentistry clinic to an external hard drive.

The data was password-protected using the AES 256 encryption in the VeraCrypt software (https://veracrypt.io/en/Home.html). A random ID number was assigned to each patient, distinguishing between male and female subjects, and was not associated with their existing medical data.

The documentation was searched using the following terms: "SARS-CoV-2"; "RT-PCR (reverse-transcription polymerase chain reaction)"; "post-COVID-19 syndrome"; different tongue changes and treatment methods ("enlarged fungiform papillae", "depapillation", "strawberry tongue", "geographic tongue", "vascular changes", "aphthous changes", "candidiasis", "edema", "herpetic lesions", "ulcers", "erosions", "nodular lesions", and "necrotic changes"). After analyzing the data, the documentation of 426 individuals (254 women and 172 men) who had mild forms of SARS-CoV-2 infection that persisted for up to 7 days and manifested with lesions on the tongue was included. The patients were previously healthy and did not exhibit any oral lesions. The exclusion criteria were comorbidities, pregnancy and addictions.

During the initial visit, all patients underwent a comprehensive examination, in accordance with the checklist presented in Table 1. Particular attention was paid to the presence of lesions on the tongue. As outlined in the documentation, periodic checkups allow to determine variability and duration of lesions, depending on the therapy or lack thereof.

In the case of ulcers, erosions and aphthous changes, laser therapy was recommended (5 treatments every 3 days) using a semiconductor laser (SMART; Lasotronix, Piaseczno, Poland). The treatment area was rinsed 3 times a day for 14 days with a 0.2% chlorhexidine solution or Alfa Implant fluid (ATOS, Warsaw, Poland), which contains sage, chamomile, arnica, oak bark extracts, linseed, xylitol, and chlorhexidine. For the treatment of fungal lesions, Nystatin (Teva Pharmaceuticals Polska Sp. z o.o., Warsaw, Poland) at a concentration of 100,000 IU/mL was applied topically to the tongue (2–3 times a day) until the symptoms disappeared and for 48 h after their disappearance.

All patients with lesions located on the tongue were advised, in addition to standard hygiene practices, to brush their tongue twice a day and to use undiluted antiseptic rinses 3 times a day for at least 30 s.

Statistical analysis

The following variables were used for the statistical analysis of quantitative data (MS Excel 365 (Microsoft Corporation, Redmond, USA)): the total number of patients (*N*); the number of valid observations in the occurrence of different tongue pathologies divided by gender (*n*); minimum value; maximum value; arithmetic mean (μ); median (*Me*); and standard deviation (*SD*). Additionally, the percentage of patients affected by these changes and the duration of symptoms relative to the therapy or lack thereof were calculated. The study adopted a significance threshold of $\alpha = 0.05$.

Results

The distribution of different types of changes observed on the tongue is shown in Fig. 1. The most prevalent symptom was the enlargement of fungiform papillae, which was found in 44.4% of women and 50% of men. According to the findings, 36.6% of the male population

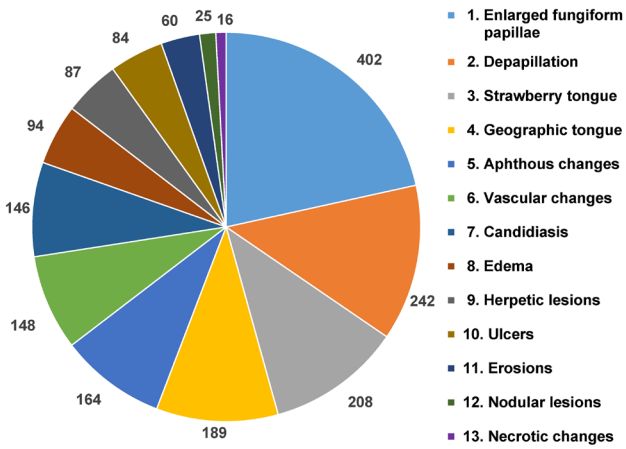

Fig. 1. Distribution of different types of lesions observed on the tongue in the study sample (total number of patients (*N*) = 426; median (*Me*) = 127; standard deviation (*SD*) = 107.1973)

Table 1. Checklist of the medical examinations conducted on patients participating in the research program

Medical examination	Periodicity	Description
Subjective and objective examination	during the first visit, as well as 1 and 2 weeks after the implementation of the treatment; during the first year of observation – every month, and in subsequent years – every 2 months or whenever a problem occurs	RT-PCR results, oral condition, date of the appearance of the first and subsequent symptoms, complications, current ailments, systemic diseases, medications taken, addictions
Laboratory tests	during the first visit, and subsequently – every 6 months	level of D-dimer and SARS-CoV-2 IgG, fungal test; in case of deviations from the norm – a referral to a specialist
Chest X-ray or CT scan	during the first visit, and subsequently – every 12 months	in smoking and COPD patients
OPG/OPT	during the first visit, and subsequently – every 6 months	assessment of the condition of the teeth and bone tissue
USG of the tongue	during the first visit, and subsequently – every 6 months	nodular lesions, unilateral swelling of the tongue; in case of deviations from the norm – a referral to a specialist

RT-PCR – reverse transcription polymerase chain reaction; COPD – chronic obstructive pulmonary disease; OPG/OPT – orthopantomography; USG – ultrasonography; CT – computed tomography; SARS-CoV-2 – severe acute respiratory syndrome coronavirus 2; IgG – immunoglobulin G.

presented with depapillation, 33.6% had strawberry tongue and 29.1% demonstrated geographic tongue. However, in the case of women, 23.9% had vascular changes, 20.9% – candidiasis, 20.2% – depapillation, and 19.7% – aphthous changes. The least common symptoms in men were necrotic changes (1.2%), erosions (3.8%) and nodular lesions (4.5%), while in women, nodular lesions (0.7%), necrotic changes (2.1%) and herpetic lesions (4.7%) were the least prevalent.

The first variants of the virus gave a characteristic picture of strawberry tongue, which occurred in 208 analyzed patients. Subsequently, depapillation (Fig. 2) was identified in 86 women and 156 men. The Omicron variant caused the intensification of clinical changes on the tongue, which included geographic tongue in 65 women and 124 men with strongly marked furrows on the entire dorsal and lateral surfaces. Vascular changes on the ventral surface (Fig. 3) were identified in 102 women and 46 men. Large aphthous changes were found in 106 women and 58 men, candidiasis – in 89 women and 57 men, edema – in 42 women and 52 men, herpetic lesions – in 38 women and 49 men, ulcers – in 38 women and 46 men, erosions – in 32 women and 28 men, nodular lesions (Fig. 4) – in 6 women and 19 men, and necrotic changes were found in 9 women and 7 men. Enlarged fungiform papillae were reported in 189 women and 213 men.

The number of tongue changes and their duration after and without treatment are presented in Table 2. On average, from 3 to 5 changes were identified concurrently. In most cases, the changes disappeared spontaneously and persisted from 4 weeks to 36 months (4.69% of cases). In 20% of cases, they recurred.

Fig. 2. Example of tongue depapillation selected from the patient records

Fig. 3. Example of vascular changes on the tongue, selected from the patient records

Fig. 4. Example of nodular lesions selected from the patient records

The local therapy used resulted in a 50% reduction in the duration of PCS.

In 2 male patients, unilateral swelling of the tongue accompanied by depapillatory changes and a fungal coating was found to be cancerous. As indicated in the interview, the patients exhibited a history of good health, refrained from smoking, and did not consume alcohol. The first patient (KP), a 38-year-old male, experienced a short-term loss of smell and taste, mild fever, and muscle and bone soreness for a duration of 4 days. The second patient (WA), a 36-year-old male, exhibited a loss of taste for 7 days during the infection. The reverse transcription polymerase chain reaction (RT-PCR) test was positive in both patients. At present, patients are being treated in the oncology department.

Table 2. Frequency distribution of tongue lesions and their duration after and without treatment ($N = 426$)

No.	Tongue pathology	Patients, *n*		Duration [days]					
				treated patients			not treated patients		
		treated	not treated	Min	Max	*Me*	Min	Max	*Me*
1.	Enlarged fungiform papillae	349	53	14	60	37	300	1,080	690
2.	Depapillation	130	112	90	120	105	360	1,080	720
3.	Strawberry tongue	144	64	60	90	75	180	360	270
4.	Geographic tongue	120	69	120	180	150	300	1,080	690
5.	Aphthous changes	142	22	5	7	8.5	10	14	12
6.	Vascular changes	120	28	60	90	75	180	720	450
7.	Candidiasis	127	19	5	7	6	14	21	17
8.	Edema	82	12	10	14	12	10	30	20
9.	Herpetic lesions	69	18	5	7	6	10	14	12
10.	Ulcers	68	16	7	10	8.5	10	21	10
11.	Erosions	48	12	5	7	6	7	14	10
12.	Nodular lesions	22	3	7	21	14	300	360	330
13	Necrotic changes	14	2	10	14	12	60	90	75

Min – minimum value; Max – maximum value; *Me* – median.

Discussion

At the onset of the COVID-19 pandemic, the predominant focus was on respiratory and systemic manifestations. In the case of pathologies located within the oral cavity,[1–4] only cases of single lesions on the tongue have been documented.[24–26]

A survey of 665 Egyptian patients after the SARS-CoV-2 infection revealed that xerostomia occurred in 47.6% of cases, oral pain was experienced by 23% of patients, ulcerations were present in 20.4% of cases, 12% of patients reported pain in bones or joints, and 10.5% of individuals experienced halitosis. In 28.3% of cases, 2–3 symptoms manifested concurrently.[27]

Based on our retrospective study, it was found that lesions located on the tongue are characterized by clinical diversity and that they undergo transformation with the emergence of new coronavirus variants. All patients with tongue lesions reported no pain symptoms, only discomfort when swallowing. Patients requiring consultation with a specialist were referred to reference centers.

The documentation of the group under study revealed that 94.36% of patients demonstrated enlarged fungiform papillae, 56.81% had depapillation, 48.82% had strawberry tongue, and 44.36% presented with geographic tongue.

The tongue pathologies persisted from 5 to 1,080 days. The shortest time after treatment concerned aphthous changes, herpetic lesions, candidiasis, and erosions. The average duration of ulceration, necrotic changes, and swelling of the tongue did not exceed 12 days, and the average duration of swelling of the fungiform papillae, strawberry tongue, vascular changes, depapillation, and geographic tongue was between 5 and 22 weeks.

Based on other research, aphthous-like lesions persist from 5 to 10 days, ulcerations and erosions from 5 to 21 days, and erosions from 14 to 28 days after treatment.[3] Other authors have observed that lesions in the oral cavity may disappear from 10 to 42 days after the disappearance of systemic symptoms and disappear spontaneously or after basic treatment.[14,24] The longest persisting lesion is geographic tongue (120–180 days).[28]

A review of patient records indicated that untreated lesions, erosions, ulcers, aphthous changes, and herpetic changes exhibited the shortest duration, ranging from 10 to 12 days. Candidiasis and edema demonstrated a duration of approx. 20 days, necrotic changes – about 11 weeks, and the remaining conditions – from 9 to 36 months.

Many authors have observed tongue pathologies related to acute SARS-CoV-2 infection, as well as the impact of commonly prescribed pharmaceutical agents.[1,3,22] Some patients present with a white infiltrative plaque on the dorsum of the tongue, located centrally, that resembles the late stage of recurrent herpetic lesions and candidiasis, manifesting as multiple, small, yellowish, circular ulcers.[3,29] In other cases, irregular tongue ulceration has been observed,[2,3,29,30] accompanied by the presence of many small extravasations[31] or depapillation of the tongue.[26,32]

The medical records of the patients included in the study indicated that candidiasis occurred in 29.96% of patients, herpetic lesions – in 16.19%, ulcers – in 15.96%, erosions – in 11.26%, and tongue depapillation affected 56.8% of individuals.

In a study conducted in Spain in 2020, 304 out of 666 patients presented with mucocutaneous manifestations, including transient lingual papillitis (11.5%), aphthous inflammation (6.9%), glossitis with pitting on its lateral

surface (6.6%), and glossitis with uneven depapillation (3.9%).[31] In the current study, aphthous changes were found in 38.5% of the subjects.

Analyzing the data from the available literature, it should be stated that no clear relationship has been established between COVID-19 and changes on the tongue.[33] Some researchers believe that these changes are a consequence of stress,[1,3,32,33] hygiene neglect,[1,3] opportunistic infections,[1,3,8,22,23] immunosuppression,[1,3,28] vascular changes,[1,20,21] or excessive inflammatory response.[34–38] It is also suggested that systemic health deterioration, an acute onset of infection, and multidrug therapy may induce pathological changes in the oral cavity.[33] The occurrence of secondary ulcers and the immune response are associated with the presence of viral infections.[29,36,37] Coronavirus disease may, therefore, cause the overactivation of the humoral response to inflammatory factors, resulting in a cytokine storm and immune exhaustion, which may lead to early changes in the oral cavity.[35]

A lack of oral hygiene among hospitalized patients connected to a respirator is a probable cause of opportunistic fungal infections.[35]An impaired immune system can cause recurrent herpes simplex virus (HSV-1) infections, non-specific ulcers and drug eruptions.[37,38]

In the present study, the hypothesis of hygiene neglect was rejected, as evidenced by good oral hygiene and overall health of the patients. The adequate selection of oral hygiene products, especially those based on natural ingredients, ensures adequate protection of the oral cavity.[39] Initially, it was thought that antibacterial mouthwashes could reduce viral load. The study on hypochlorous acid (HClO) and povidone-iodine (PVP-I) revealed no evidence to support the hypothesis that these preparations reduce the viral load.[40] At the beginning of the pandemic, 73% of respondents reported feelings of fear and anxiety when considering a dental visit due to the possibility of being infected with the SARS-CoV-2.[41] Isolation and anxiety caused deterioration of oral hygiene in some patients, increased caries and inflammation of the oral cavity, and temporomandibular joint (47.8%) and muscle disorders, leading to parafunctions, bruxism (31%), headaches, and mental disorders.[42,43] The resumption of medical activities was permitted after the implementation of increased hygiene and antiseptic procedures in dental offices, based on the guidelines from the World Health Organization (WHO) and the Polish Dental Association (PDA). The use of the ultraviolet C (UVC) radiation, ozone, disinfectants, and protective equipment ensured the safety of staff and patients.[44,45]

Some studies suggest that changes in the oral cavity may be the direct effect of the virus.[4,21,38]

Published papers on tongue lesions during the SARS-CoV-2 infection confirm the relationship with organic damage or complications of thrombocytopenia, anticoagulant treatment, disseminated intravascular coagulation, and systemic inflammation. It is suggested that the presence of long COVID lesions results from primary or secondary vascular/hematological changes and lymphocytic thrombophilic arteritis.[33,38]

Some authors hypothesize that prolonged manifestations of the disease may result from co-infections and/or secondary changes.[1,8,32,35]

The results of the conducted cross-sectional study indicated the occurrence of erythematous spots, single ulcers (3%), atrophic glossitis (4.6%), and candidiasis (1%).[38]

The differential diagnosis of the COVID tongue includes herpetic glossitis, Melkersson–Rosenthal syndrome, lichen planus, and fungal infections.[1] When diagnosing a patient with PCS symptoms localized on the tongue, reinfection should be considered, which may occur many months after the initial infection with the SARS-CoV-2.[1,3,35]

In addition, there have been reports of post-vaccination lesions on the tongue.[46] A survey conducted among vaccinated individuals in Poland, Italy and other EU countries revealed post-vaccination symptoms in the oral cavity after the administration of the first dose of the vaccine (3.1%) and after the second dose (5.4%). The undesirable post-vaccination reactions include changes in sensitivity and facial paresis, a burning sensation, aphthous changes, taste changes, xerostomia, depapillation of the tongue, pain, stomatitis, cheilitis, and candidiasis.[47] Many published studies demonstrate that changes located on the tongue are the result of acute infection, reduced immunity, polypragmasia, hygiene neglect, or stress.[1,3] This hypothesis was not confirmed in the current study.

However, the medical documentation of young patients, those without comorbidities and previous changes in the tongue, as well as the emergence of subsequent variants of the virus, indicate that they cannot be underestimated. The presence of 2 cases of cancer lesions suggest the necessity for thorough diagnostics.

Research on lesions located on the tongue should also be continued based on other age groups, as well as those affected by systemic diseases or addictions.

Conclusions

The infection caused by the SARS-CoV-2 affects not only the general condition of the body but also the health of the oral cavity. Changes located on the tongue require observation and basic or specialized treatment. In the absence of pain symptoms, the patient should be monitored for a period of 4 weeks, with the expectation that the symptoms will spontaneously resolve. In case of pain, a good solution is the application of laser bio-stimulation. If various changes co-occur in the oral cavity, it is advised that the patient be referred for specialized treatment. The findings of this study suggest that the issue of the COVID tongue is not acknowledged by a considerable portion of the medical community.

Ethics approval and consent to participate

The Bioethical Commission of Medical University of Warsaw, Poland, has approved the scope of this retrospective study (commission statement No. AKBE/318/2023). In addition to the standard consent to treatment, as required by national regulations, all patients provided written consent to participate in this study.

Data availability

The datasets generated and/or analyzed during the current study are available from the corresponding author on reasonable request.

Consent for publication

Not applicable.

Use of AI and AI-assisted technologies

Not applicable.

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References

1. Sharma P, Malik S, Wadhwan V, Palakshappa SG, Singh R. Prevalence of oral manifestations in COVID-19: A systematic review. *Rev Med Virol*. 2022;32(6):e2345. doi:10.1002/rmv.2345
2. Chaux-Bodard AG, Deneuve S, Desoutter A. Oral manifestation of COVID-19 as an inaugural symptom? *J Oral Med Oral Surg*. 2020;26:18. doi:10.1051/mbcb/2020011
3. Iranmanesh B, Khalili M, Amiri R, Zartab H, Aflatoonian M. Oral manifestations of COVID-19 disease: A review article. *Dermatol Ther*. 2021;34(1):e14578. doi:10.1111/dth.14578
4. Paradowska-Stolarz AM. Oral manifestations of COVID-19: Brief review. *Dent Med Probl*. 2021;58(1):123–126. doi:10.17219/dmp/131989
5. World Health Organization. Coronavirus disease (COVID-19): Post COVID-19 condition. 2023. https://www.who.int/news-room/questions-and-answers/item/coronavirus-disease-(covid-19)-post-covid-19-condition. Accessed March 28, 2023.
6. Galeotti C, Bayry J. Autoimmune and inflammatory diseases following COVID-19. *Nat Rev Rheumatol*. 2020;16(8):413–414. doi:10.1038/s41584-020-0448-7
7. Duś-Ilnicka I, Mazur M, Rybińska A, Radwan-Oczko M, Jurczyszyn K, Paradowska-Stolarz A. SARS CoV-2 IgG seropositivity post-vaccination among dental professionals: A prospective study. *BMC Infect Dis*. 2023;23(1):539. doi:10.1186/s12879-023-08534-z
8. Straburzyński M, Nowaczewska M, Budrewicz S, Waliszewska-Prosół M. COVID-19-related headache and sinonasal inflammation: A longitudinal study analysing the role of acute rhinosinusitis and ICHD-3 classification difficulties in SARS-CoV-2 infection. *Cephalalgia*. 2022;42(3):218–228. doi:10.1177/03331024211040753
9. Straburzyński M, Kuca-Warnawin E, Waliszewska-Prosół M. COVID-19-related headache and innate immune response – a narrative review. *Neurol Neurochir Pol*. 2023;57(1):43–52. doi:10.5603/PJNNS.a2022.0049
10. Tomo S, Miyahara GI, Simonato LE. Oral mucositis in a SARS-CoV-2 infected patient: Secondary or truly associated condition? *Oral Dis*. 2022;Suppl 1:963–967. doi:10.1111/odi.13570
11. Hathway RW. COVID tongue. *Br Dent J*. 2021;230(3):114. doi:10.1038/s41415-021-2666-z
12. Tsuchiya H. The oral cavity potentially serving as a reservoir for SARS-CoV-2 but not necessarily facilitating the spread of COVID-19 in dental practice. *Eur J Dent*. 2023;17(2):310–318. doi:10.1055/s-0042-1757909
13. Bilińska K, Jakubowska P, Von Bartheld CS, Butowt R. Expression of the SARS-CoV-2 entry proteins, ACE2 and TMPRSS2, in cells of the olfactory epithelium: Identification of cell types and trends with age. *ACS Chem Neurosci*. 2020;11(11):1555–1562. doi:10.1021/acschemneuro.0c00210
14. Lozada-Nur F, Chainani-Wu N, Fortuna G, Sroussi H. Dysgeusia in COVID-19: Possible mechanisms and implications. *Oral Surg Oral Med Oral Pathol Oral Radiol*. 2020;130(3):344–346. doi:10.1016/j.oooo.2020.06.016
15. Więckiewicz M, Danel D, Pondel M, et al. Identification of risk groups for mental disorders, headache and oral behaviors in adults during the COVID-19 pandemic. *Sci Rep*. 2021;11(1):10964. doi:10.1038/s41598-021-90566-z
16. Hassainin EH, Islam N, Weyer A, Haney D, Devine MS. Macroglossia in the setting of COVID-19. Poster presented at: *Nashville Chest 2022*; October 16–19, 2022; Nashville, TN. doi:10.1016/jchest.2022.08.244
17. Colombo D, Del Nonno F, Nardacci R, Falasca L. May macroglossia in COVID-19 be related not only to angioedema? *J Infect Public Health*. 2022;15(1):112–115. doi:10.1016/j.jiph.2021.10.026
18. @timspector. One in five people with COVID still present with less common symptoms that dont get on the official PHE list – such as skin rashes. January 31, 2021. https://twitter.com/timspector/status/1349421278034325506/photo/1. Accessed January 31, 2021.
19. Picciani BLS, Santos LR, Amin TN, et al. Applicability of the geographic tongue area and severity index among healthcare professionals: A cross-sectional clinical validation of a newly developed geographic tongue scoring system. *J Clin Med*. 2021;24:10(23):5493. doi:10.3390/jcm10235493
20. Díaz Rodríguez M, Jimenez Romera A, Villarroel M. Oral manifestations associated with COVID-19. *Oral Dis*. 2022;28 Suppl 1:960–962. doi:10.1111/odi.13555
21. Pérez-Sayáns M, Ortega KL, Braz-Silva PH, Carreras-Presas CM, Carrión AB. Can "COVID-19 tongue" be considered a pathognomonic finding in SARS-CoV-2 infection? *Oral Dis*. 2022;28 Suppl 2:2579–2580. doi:10.1111/odi.13807
22. Katz J. Prevalence of candidiasis and oral candidiasis in COVID-19 patients: A cross-sectional pilot study from the patients' registry in a large health center. *Quintessence Int*. 2021;52(8):714–718. doi:10.3290/j.qi.b1491959
23. Pang W, Zhang D, Zhang J, et al. Tongue features of patients with coronavirus disease 2019: A retrospective cross-sectional study. *Integr Med Res*. 2020;9(3):100493. doi:10.1016/j.imr.2020.100493
24. Scotto G, Fazio V, Spirito F, Lo Muzio E, Lo Muzio L. COVID tongue: Suggestive hypothesis or clinical reality? *Oral Dis*. 2022;28 Suppl 2:2618–2619. doi:10.1111/odi.14134
25. Sofi-Mahmudi A. Patients with COVID-19 may present some oral manifestations. *Evid Based Dent*. 2021;22(2):80–81. doi:10.1038/s41432-021-0173-3
26. Afshar ZM, Barary M, Ebrahimpour S, et al. Pathophysiology and management of tongue involvement in COVID-19 patients. *Indian J Otolaryngol Head Neck Surg*. 2022;74(Suppl 2):3235–3238. doi:10.1007/s12070-021-03052-3
27. Abubakr N, Salem ZA, Kamel AHM. Oral manifestations in mild-to-moderate cases of COVID-19 viral infection in the adult population. *Dent Med Probl*. 2021;58(1):7–15. doi:10.17219/dmp/130814
28. de Moraes Melo Neto CL, Bannwart LC, de Melo Moreno AL, Coelho Goiato M. SARS-CoV-2 and dentistry – review. *Eur J Dent*. 2020;14(S01):S130–S139. doi:10.1055/s-0040-1716438
29. Dos Santos JA, Normando AGC, Carvalho da Silva RL, et al. Oral mucosal lesions in a COVID-19 patient: New signs or secondary manifestations? *Int J Infect Dis*. 2020;97:326–328. doi:10.1016/j.ijid.2020.06.012
30. Corchuelo J, Ulloa FC. Oral manifestations in a patient with a history of asymptomatic COVID-19: Case report. *Int J Infect Dis*. 2020;100:154–157. doi:10.1016/j.ijid.2020.08.071

31. Nuno-Gonzalez A, Martin-Carrillo P, Magaletsky K, et al. Prevalence of mucocutaneous manifestations in 666 patients with COVID-19 in a field hospital in Spain: Oral and palmoplantar findings. *Br J Dermatol*. 2021;184(1):184–185. doi:10.1111/bjd.19564
32. Sarruf MBJM, Quinelato V, Sarruf GJM, et al. Stress as worsening of the signs and symptoms of the geographic tongue during the COVID-19 pandemic: A pilot study. *BMC Oral Health*. 2022;22(1):565. doi:10.1186/s12903-022-02609-0
33. Carreras-Presas CM, Sánchez JA, López-Sánchez AF, Jané-Salas E, Somacarrera Pérez ML. Oral vesiculobullous lesions associated with SARS-CoV-2 infection. *Oral Dis*. 2021;27 Suppl 3:710–712. doi:10.1111/odi.13382
34. Moreno-Pérez O, Merino E, Leon-Ramirez JM, et al.; COVID19-ALC research group. Post-acute COVID-19 syndrome. Incidence and risk factors: A Mediterranean cohort study. *J Infect*. 2021;82(3):378–383. doi:10.1016/j.jinf.2021.01.004
35. Dziedzic A, Wojtyczka R. The impact of coronavirus infectious disease 19 (COVID-19) on oral health. *Oral Dis*. 2021;27 Suppl 3:703–706. doi:10.1111/odi.13359
36. Riad A, Kassem I, Hockova B, Badrah M, Klugar M. Tongue ulcers associated with SARS-CoV-2 infection: A case series. *Oral Dis*. 2022;28 Suppl 1:988–990. doi:10.1111/odi.13635
37. Cruz Tapia RO, Peraza Labrador AJ, Guimaraes DM, Valdez LHM. Oral mucosal lesions in patients with SARS-CoV-2 infection. Report of four cases. Are they a true sign of COVID-19 disease? *Spec Care Dentist*. 2020;40(6):555–560. doi:10.1111/scd.12520
38. Ganesan A, Kumar S, Kaur A, et al. Oral manifestations of COVID-19 infection: An analytical cross-sectional study. *J Maxillofac Oral Surg*. 2022;21(4):1326–1335. doi:10.1007/s12663-021-01679-x
39. Mazur M, Ndokaj A, Bietolini S, Nisii V, Duś-Ilnicka I, Ottolenghi L. Green dentistry: Organic toothpaste formulations. A literature review. *Dent Med Probl*. 2022;59(3):461–474. doi:10.17219/dmp/146133
40. Sevinç Gül SN, Dilsiz A, Sağlık İ, Aydın NN. Effect of oral antiseptics on the viral load of SARS-CoV-2: A randomized controlled trial. *Dent Med Probl*. 2022;59(3):357–363. doi:10.17219/dmp/150831
41. Daltaban Ö, Aytekin Z. Fear and anxiety of COVID-19 in dental patients during the COVID-19 pandemic: A cross-sectional survey in Turkey. *Dent Med Probl*. 2022;59(3):343–350. doi:10.17219/dmp/150075
42. Holm-Hadulla RM, Wendler H, Baracsi G, Storck T, Möltner A, Herpertz SC. Depression and social isolation during the COVID-19 pandemic in a student population: The effects of establishing and relaxing social restrictions. *Front Psychiatry*. 2023;14:1200643. doi:10.3389/fpsyt.2023.1200643
43. Emodi-Perlman A, Eli I, Smardz J, et al. Temporomandibular disorders and bruxism outbreak as a possible factor of orofacial pain worsening during the COVID-19 pandemic—concomitant research in two countries. *J Clin Med*. 2020;9(10):3250. doi:10.3390/jcm9103250
44. Wezgowiec J, Wieczynska A, Wieckiewicz M, et al. Evaluation of antimicrobial efficacy of UVC radiation, gaseous ozone, and liquid chemicals used for disinfection of silicone dental impression materials. *Materials (Basel)*. 2022;15(7):2553. doi:10.3390/ma15072553
45. Dominiak M, Różyło-Kalinowska I, Gedrange T, et al. COVID-19 and professional dental practice. The Polish Dental Association Working Group recommendations for procedures in dental office during an increased epidemiological risk. *J Stoma*. 2020;73(1):1–10. doi:10.5114/jos.2020.94168
46. Khazeei Tabari MA, Najary S, Khadivi G, Yousefi MJ, Samieefar N, Abdollahimajd F. Oral lesions after COVID-19 vaccination: Immune mechanisms and clinical approach. *Infect Med (Beijing)*. 2022;1(3):171–179. doi:10.1016/j.imj.2022.06.004
47. Mazur M, Duś-Ilnicka I, Jedliński M, et al. Facial and oral manifestations following COVID-19 vaccination: A survey-based study and a first perspective. *Int J Environ Res Public Health*. 2021;18(9):4965. doi:10.3390/ijerph18094965

Original paper

Odontomas as a cause of tooth eruption disorders

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Abstract

Background. Odontomas are odontogenic tumors located in the jawbone, referred to by the World Health Organization (WHO) as hamartomatous (Greek *hamartion* – a bodily defect) developmental malformations. The term was first used by Brock in 1866 to refer to every odontogenic tumor. Today an odontoma is believed to be a congenital malformation of dental hard tissues. Odontomas can contribute to the development of dental abnormalities and bite disorders.

Objectives. The study aimed to present tooth eruption disorders caused by the occurrence of odontomas in 20 patients treated at the Dental Surgery Clinic for Children.

Material and methods. The study group comprised 20 patients (13 in good general health, 6 with systemic diseases and 1 with a congenital malformation), aged 2–17 years (mean age: 8.96 years), with jawbone odontomas diagnosed through clinical and radiological examination. These odontomas were the cause of disorders in the eruption of deciduous and permanent teeth. The treatment method applied in all patients involved the enucleation of the tumor with its capsule and sending the material for histopathological examination.

Results. Odontomas were discovered accidentally or were accompanied by such clinical symptoms as retained deciduous teeth, impacted deciduous and permanent teeth, missing teeth in the oral cavity, gaps, displaced teeth, and the distension of the alveolar process of the maxilla/alveolar part of the mandible. The odontomas occurred in both the maxilla and the mandible, more often in boys. The compound odontoma (OCp) was the most common type.

Conclusions. Odontomas are benign odontogenic tumors that do not undergo neoplastic transformation. The treatment of choice is a surgical procedure involving the complete removal of the tumor with its capsule, which results in full recovery.

Keywords: children, oral surgery, orthodontics, dental anomalies, tumors

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Highlights

- Odontomas are benign tumors that do not cause neoplastic transformation.
- The compound odontoma is the most common type of odontoma.
- Odontomas often cause malocclusion.
- The treatment of choice is a surgical procedure involving the complete enucleation of the tumor with its capsule, which results in full recovery.
- Orthodontic treatment is often required after surgical treatment.

Introduction

Odontomas are among the most common odontogenic neoplasm-like tumors of the jawbone.[1,2] They are not typical neoplasms, as they are only hamartomatous developmental malformations.[3] According to the literature, they most frequently occur in the 2nd and 3rd decades of life, though they are also not infrequent in the 1st decade.[4] Their etiopathogenesis has not been fully explained and there are a number of theories concerning the origin of odontomas, such as traumas, infections, genetic disorders, or anomalies in the development of the tooth bud.[3–5] Odontomas are often discovered incidentally, as they rarely cause pain. The most common reasons for their discovery are retained deciduous teeth, impacted deciduous and permanent teeth, missing teeth in the oral cavity, gaps, displaced teeth, and the distension of the alveolar process of the maxilla/alveolar part of the mandible. Odontomas usually have a diameter not exceeding 3 cm.[1] Since they are odontogenic tumors, they contain an epithelial and mesenchymal component. The World Health Organization (WHO) divides them into 2 groups: complex odontomas (OC); and compound odontomas (OCp).[6–8] The complex odontoma is a hamartomatous developmental lesion in which structures such as enamel, dentin, and sometimes cementum are arranged chaotically.[4] The compound odontoma is an encapsulated developmental hamartomatous malformation containing numerous denticles – odontoids. In the case of OCp, structures such as enamel, dentin, cementum, and pulp are arranged in an organized fashion, so they resemble a properly formed tooth.[4,9] Some authors distinguish another variant – erupted odontoma – in the past referred to as peripheral odontoma. It is a variant of OCp that erupts in the oral cavity.[4,5]

In histological examination, individual tooth tissues like enamel, dentin, cementum, and pulp can be distinguished.[9] Diagnostic methods include radiological imaging – tooth X-rays, panoramic radiography, cone-beam computed tomography (CBCT) and/or computed tomography (CT). The treatment of choice is a surgical procedure involving the enucleation of the tumor with the capsule around it.[2,4,9]

The study aimed to present a type of tooth eruption disorders caused by the occurrence of odontomas, as well as treatment methods for odontomas in children and adolescents.

Material and methods

A total of 20 patients (13 (65%) boys and 7 (35%) girls), aged between 2.8 and 17.6 years (mean age: 8.96 ±4.20 years), were included in the study. The study involved general and local history-taking concerning chronic comorbidities and the medications received, dental history (the reason for the visit, presenting complaints), and extraoral (facial symmetry and lymph nodes) and intraoral (the number and type of erupted teeth, the assessment of the shape and consistency of the alveolar process) clinical examination. All patients underwent radiological and histopathological examination. Depending on the indications, tooth X-rays, panoramic radiography, CBCT and/or CT were performed. Radiological imaging was used to assess the location and type of odontoma, the presence of impacted or retained teeth, gaps, and tooth displacement. While OCp were found in X-ray images as high-density radiopacities arranged as numerous small deformed denticles (odontoids) surrounded by a band of radiolucency with an osteosclerotic border, OC were characterized by irregular radiopacities with a radiolucent edge and an osteosclerotic rim. The treatment of choice for all patients was the enucleation of the tumor with the capsule surrounding it.

Results

Six participants (30%) had a diagnosis of a systemic disease: nephritic syndrome; bicuspid aortic valve (BAV) dysfunction; epilepsy; atrial septal defect (ASD) II; autoimmune polyglandular syndrome (APS); and a history of chemotherapy for anaplastic ependymoma. One patient (5%) had a congenital malformation – cleft lip and cleft palate. The remaining patients were in good general health. Eight children (40%) had deciduous teeth, and 12 (60%) had permanent or mixed dentition (Table 1).

Table 1. Characteristics of patients and clinical signs of odontomas

Parameter	Deciduous dentition (n = 8)	Mixed/permanent dentition (n = 12)	Total N = 20
Male gender	7	6	13 (65)
Age [years]	4.18 ±0.98	11.62 ±2.75	8.96 ±4.20
Distention/protrusion of the alveolar process	5	4	9 (45)
Gaps between the teeth	1 (coexisting with alveolar protrusion)	1	2 (10)
Patients with missing teeth (impacted) teeth	4	7	11 (55)
How many teeth are missing?	5	8	13 (65)
Numbers of the missing teeth	63, 64, 65, 71, and 81	21, 21, 43, 44, and 46 12, 33 and 43	–
Retained deciduous teeth	0	3	3 (15)
Pain	1	0	1 (5)
Lesions located in the maxilla	6	5	11 (55)
Lesions located in the mandible	2	7	9 (45)

Data presented as number, number (percentage) (n (%)) or as mean ± standard deviation (M ±SD).

All patients were referred for diagnostic examination by dentists: 11 (55%) of them due to missing teeth; 3 (15%) due to retained deciduous teeth; 9 (45%) had limited distension of the alveolar process of the maxilla or the alveolar part of the mandible; 2 (10%) had gaps between their teeth; 1 (5%) experienced pain; 2 (10%) had exophytic lesions; in 3 patients (15%), the lesion was discovered incidentally in plain radiography. Some patients had 2 or more symptoms simultaneously, for example, gaps between the teeth and the distension of the alveolar process of the maxilla or the alveolar part of the mandible (1 (5%)) or an impacted tooth and the distension of the alveolar process of the maxilla or the alveolar part of the mandible (2 (10%)). Most patients reported no pain. Only one boy at the age of 3.6 years reported moderate pain of a distending and continuous nature. In 2 boys (aged 2.8 and 3.6 years), changes in facial features in the form of cheek protrusion were observed. The buccal skin in the area of the lesions was unaffected, without signs of inflammation. None of the participants had palpable or painful submandibular lymph nodes. The most common intraoral symptoms included the absence of a deciduous or permanent tooth typical for the patient's age despite the presence of a corresponding tooth on the other side, and alveolar process enlargement (protrusion, a gap between the teeth). Five of the 9 patients with alveolar enlargement also lacked a tooth typical for their age, of which 1 child was from the group with deciduous teeth (1/5) and 4 children were from the group with mixed/permanent dentition (4/5). Nine patients were lacking a single tooth, the remaining 2 (each of them) were lacking 2 teeth (teeth 71 and 81, 43 and 44). The most commonly missing teeth were deciduous or permanent canines. Radiological examination showed that all the missing teeth were impacted. In 3 patients from the group with mixed/permanent dentition, the absence of a tooth was accompanied by the presence of retained deciduous tooth/teeth (teeth 61, 73, 84, and 85). Lesions were more frequently observed in the maxilla (55%), predominantly in the area of canines (50%) and incisors (25%), and they were least frequent in the lateral segment (25%) (Table 1, Fig. 1).

The patients had the following examination performed: tooth X-ray (11 (55%)); panoramic radiography (11 (55%)); CBCT (12 (60%)); or CT (1 (5%)). X-ray images revealed 18 OCp (of which 2 were described as erupted) and 2 OC.

In 11 patients (55%), odontomas occurred in the maxilla – 4 (20%) in the midline, 5 (25%) on the left side (Fig. 2) and 2 (10%) on the right side, and they were all OCp, of which 1 (5%) on the right side was an erupted odontoma. Nine patients (45%) were diagnosed with lesions in the mandible, with 5 (25%) of them having lesions on the right side (including 4 (20%) OCp and 1 (5%) OC) (Fig. 3), 3 (15%) of them with lesions on the left side (1 (5%) OCp, 1 (5%) OC and 1 (5%) erupted odontoma) and 1 patient (5%) with OCp in the area of incisors.

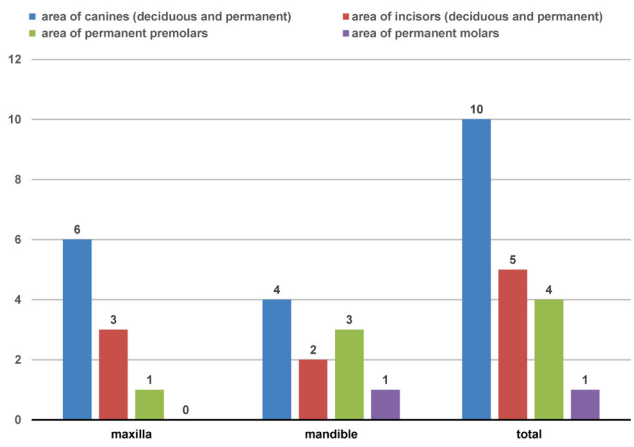

Fig. 1. Location of the lesions in relation to the types of teeth in the maxilla and the mandible

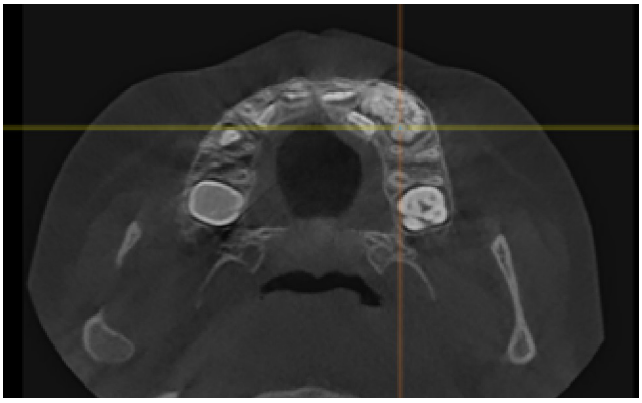

Fig. 2. Compound odontoma (OCp) in a 5-year-old patient

There are visible radiopacities with a radiodensity characteristic for dental tissues (enamel and dentin) sized 3 cm, arranged as characteristic small denticles (odontoids) with a band of radiolucency and a sclerotic rim on the left side of the maxilla.

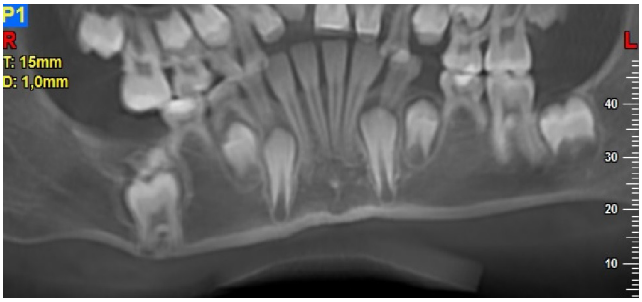

Fig. 3. Complex odontoma (OC) in a 9-year-old patient

There is a visible opaque 1-centimeter mass of irregular shape, resembling dental tissues, above the crown of a molar on the right side of the mandible

In the case of all patients, it was decided that their tumors and the capsules surrounding the tumors would be enucleated surgically in local or general anesthesia (depending on the patient's age and cooperation). After the procedure, the patients received antibiotics – amoxicillin, or alternatively clindamycin in the case of an allergy to penicillins – at doses adjusted to the body weight (b.w.) and age of the child in mg/kg b.w./day, or at doses as for adults (more than 40 kg). The material was sent for histopathological examination. On microscopic examination, in 16 cases (80%), the lesions were described as OCp, 2 (10%) as OC, and 2 (10%) as erupted odontomas (visible in the oral cavity). Compound odontomas were described as fragments of the dental pulp, dentin, enamel matrix, epithelium with fragments of connective tissue, or fragments of a tooth with preserved histological structure, with fibrovascular tissue without epithelial covering and with foci of chronic inflammatory granulation tissue (Fig. 4). Complex odontomas, on the other hand, were described as fragments of dentin and connective bone tissue (Fig. 5).

Follow-up appointments were arranged for patients 1 day after the procedure, 1 week after the procedure, 1 month after the procedure, and then at 3 and 6 months after the first follow-up. On follow-up examinations, the wounds were observed to heal properly. After approx. 6 months, in

Fig. 4. Microscopic image of a compound odontoma (OCp)

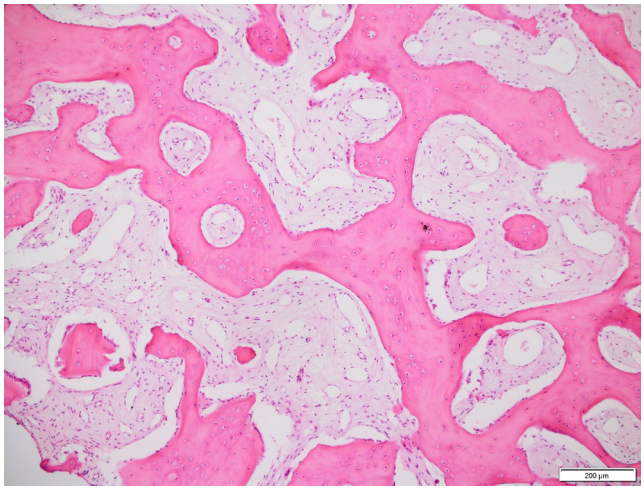

Fig. 5. Microscopic image of a complex odontoma (OC)

3 patients (15%), the impacted teeth were erupting or were situated higher as compared to their position during previous X-ray examinations. A 12-year-old girl who had 2 impacted teeth (the lower right first premolar 44 and the lower right canine 43), after 2 years from the procedure, had tooth 44 erupted and tooth 43 situated higher, but impacted by the crown of tooth 44. In another case of a 14-year-old boy who had the odontoma removed when impacted tooth 33 had completed root development, after about half a year, the impacted tooth did not change its position. Both patients were referred to an orthodontist and in both cases, the impacted teeth were orthodontically inserted into the dental arch. In no case was a recurrence observed after the removal of an odontoma.

Discussion

Odontomas account for approx. 22% of odontogenic tumors.[1,10] They are found mainly in deciduous and mixed dentition.[3] Many authors claim that OCp are more

common than OC.[4,9–11] On the other hand, according to Uma, OC are more common in older people, while OCp in the younger population.[3] In our radiological and histopathological examinations, in the age group between 2 and 17 years, the OCp was the most common lesion – it was found in 16 patients (80%). Two patients (10%) were diagnosed with OC, and in 2 boys (10%), the lesions were visible in the oral cavity, and thus classified as erupted odontomas.

Khanum et al. mention one more variant of mixed odontoma (compound-complex odontoma), which they described in a 13-year-old boy.[8] They reported it as an extremely rare type of odontoma, and their diagnosis was based on the histopathological examination showing that in some places, dental tissues were haphazardly arranged, and in other places, they presented regular, small tooth-like structures – odontoids.[8]

According to some researchers, the incidence of odontomas can be gender-specific.[12–18] Uma in her article notes a predilection toward females in the case of OC, and OCp being more common in males,[3] whereas Gedik and Müftüoğlu claim that odontomas of the maxilla are more often found in girls.[11] In our study, OC were found in 1 boy (5%) and 1 girl (5%), and OCp in 10 boys (50%) and 6 girls (30%), which is inconsistent with the findings of Uma and Gredik and Müftüoğlu.[3,11]

With regard to the typical locations of odontomas described in the literature, for OC, it is mainly the lateral section of the mandible and the anterior section of the maxilla, and for OCp, it is predominantly the anterior section of the maxilla.[3,4,10,19] In our study, similarly, OC were found in the posterior region of mandible (10%), while OCp was diagnosed in the mandible in 6 patients (30%), and in the maxilla – in 10 patients (50%), which points to its predominance in the maxilla.

According to Kale et al., odontomas occur more frequently on the right than on the left side (62% of OCp and 68% of OC), regardless of whether they are located in the maxilla or the mandible.[10] In our patients, odontomas occurred on the left side – 8 (40%), on the right side – 7 (35%), and 5 (25%) of them in the midline, which does not confirm the findings of Kale et al.[10]

Amailuk and Grubor claim that traumas are among the causes of odontomas.[5] The authors described a case of a 15-year-old boy from Sudan with OCp that was erupted, together with malformed crown and root of the upper left central incisor 21. It was decided that the odontoma and tooth 21 would be removed, and the crown of the upper right central incisor 11 reconstructed (tooth 11 had erupted with a malformed crown). The authors report that the odontoma and abnormalities in teeth 11 and 21 were caused by a childhood trauma. In some African countries, in accordance with local beliefs, children aged from 3 months to 3 years have the buds of their deciduous teeth removed by their healer for protection against viruses and vomiting. The authors mention 2 more cases of children who had their deciduous tooth buds removed in a ritual in their childhood, and then were diagnosed with an odontoma in their adolescence.[5] In one of our patients (17 years old, with nephritic syndrome), the odontoma was visible in the oral cavity in the region of lower left canine 33 and lower left first premolar 34. During the first appointment, the visible nodule was diagnosed as an erupting supernumerary tooth, and it was not until an X-ray was performed that the correct diagnosis of OCp was reached, which, due to its occurrence in the oral cavity, was described as an erupted odontoma (Fig. 6). In this case, similarly to our other patients, the patient's parents could not remember past traumas or inflammations in the region of the odontoma.

Custódio et al. report that odontomas can occur not only in the bone of the jaws, but also peripherally.[20] In their article, they describe a case of an 11-year-old girl who had on her palatal mucosa a 1.5-centimeter non-painful nodular lesion with a smooth surface, covered with an unaffected mucous membrane, which had been growing slowly for more than 2 years. Clinically, the picture was consistent with a peripheral ossifying fibroma or a peripheral giant-cell granuloma. Radiological and intraoperative examinations revealed no erosion of (damage to) the bone tissue. In was not until histopathological examination that a complex odontoma was revealed. In their study, the authors refer to 17 publications from the years 1989–2014 discussing peripheral odontomas (gingival odontomas/soft tissue odontomas), 15 of which were found in children.[20]

Clinically, in 18 (90%) of our patients, the lesions occurred in bones, and in 2 patients (10%), there were exophytic lesions in the oral cavity (in a 3-year-old patient on the lingual side, near teeth 83 and 84, and in a 14-year-old patient after cleft lip and cleft palate on the palatal side near tooth 13). An erupted odontoma was discovered accidentally in a 3-year-old patient during a dental appointment; his parents did not know how long it had been present in the oral cavity. The lesion was not painful, covered with an unchanged mucous membrane. Due to

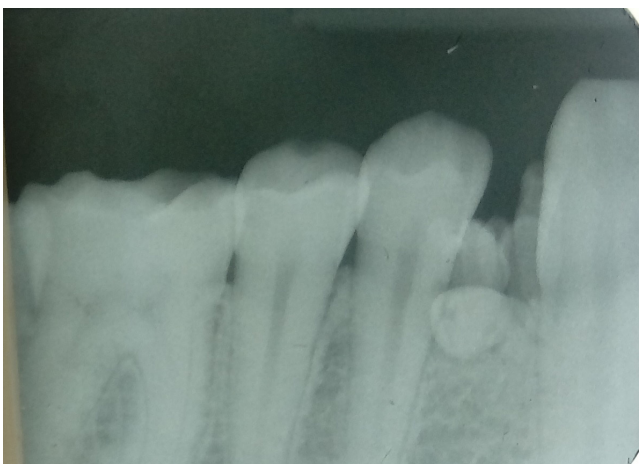

Fig. 6. Erupted odontoma

the patient's lack of cooperation, X-ray images were inaccurate and did not indicate any pathologies. In general anesthesia, CT was performed, revealing a small radiopacity located lingually at the level of the neck of tooth no. 83, characteristic of OCp. A 14-year-old patient was admitted to the Dental Surgery Clinic for the removal of a palatal lesion. On clinical examination, tooth 12 was missing, tooth 13 was displaced, the alveolar process of the maxilla was slightly distended, and an odontoma was erupting from the palatal side – it was removed in local anesthesia.

Clinical signs of odontomas described in the literature include retained deciduous teeth, impacted permanent teeth, the distension of the alveolar process of the maxilla or the alveolar part of the mandible, gaps, mild pain (in only one case), a changed shape of teeth (one patient after a childhood trauma), a nodular non-painful lesion covered with an unaffected mucous membrane (diagnosed as a gingival complex odontoma), discovered accidentally in an X-ray image.[2–10,19,20] In most of our cases, the symptoms were similar: tooth eruption disorders; the protrusion of the alveolar process of the maxilla or the alveolar part of the mandible; retained deciduous teeth; impacted permanent teeth without abnormalities; gaps; no pain. Only one boy was referred to a dentist due to pain and the swelling of the cheek, as his tumor was large and caused a significant distension of the alveolar process of the maxilla. Normal tooth eruption was observed at follow-up visits, with no indications for orthodontic treatment.

Radiographically, OC is a highly dense radiopacity of irregular shape, surrounded by a band of radiolucency with a sclerotic rim,[6,19] and OCp is a dense radiopacity arranged as numerous small deformed denticles (odontoids) surrounded by a band of radiolucency and a sclerotic rim.[6] Similar radiological pictures were obtained for our patients. Histopathological examination should be conducted to confirm the clinical and radiological diagnosis. On microscopic examination, OCp is described as fragments of the dental pulp, dentin, enamel matrix, epithelium, and connective tissue, or as fragments of a tooth with preserved histological structure, with fragments of fibrovascular tissue. On the other hand, OC is described in histopathological findings as fragments of dentin and connective bone tissue. The histopathological picture of OC is very similar to that of an ameloblastic fibro-odontoma (AFO).[20,21] And this was the microscopic picture obtained for the lesions removed in our patients.

Recently, there has been improvement in X-ray diagnostic imaging methods, especially with regard to CBCT, which certainly facilitates arriving at diagnosis as early as at the stage of radiological examination. Jayam et al. describe a case of an 11-year-old girl with a radiological picture (panoramic radiography, a tooth X-ray and a maxillary occlusal radiograph) description of impacted tooth 11 with an additional cusp and an extensive dentigerous cyst.[2] In the course of marsupialization, the dentigerous cyst was found to have no contact with tooth 11, while it contacted the adjacent additional cusp. On histopathological examination, OCp was diagnosed with a cyst. According to some authors, X-ray images – panoramic radiographs and targeted dental X-rays – may not accurately reveal the pattern of the lesion, which is why cross-sectional CBCT is recommended in the case of doubts or for more accurately determining the location.[2] In the case of 2 of our patients, it was not until a CBCT was performed that the adequate diagnosis was reached, and it was later confirmed during the procedure and histopathological examination. The first patient was a 9-year-old girl referred to an orthodontist in our clinic due to impacted tooth 21 for the exposure of the tooth and the application of a bracket to the crown of tooth 21. The patient had with her a current panoramic radiograph with visible impacted tooth 21, showing a small non-distinctive radiopacity in the tooth crown, which could be consistent with an additional cusp. An X-ray of tooth 21 was performed at our clinic, revealing a non-distinctive area of radiopacity next to the crown of tooth 21 (Fig. 7). The patient was referred for cross-sectional CBCT. After verification, OCp was diagnosed and removed with its capsule, and an orthodontic bracket was attached to tooth 21 in local anesthesia (Fig. 8). Histopathological examination confirmed the preliminary diagnosis of OCp. The other patient was a 5-year-old girl who was admitted to our clinic with a missing deciduous tooth 65. The patient had a current panoramic radiograph with her, revealing an impacted tooth (most probably tooth 65) with a non-distinctive area of radiopacity (Fig. 9). Cross-sectional CBCT was recommended. In the X-ray image, the preliminary diagnosis suggested OCp with the impacted deciduous tooth 65 above it and the tooth bud of permanent tooth 25 on the palatal side (Fig. 10). The procedure involved the resection of the

Fig. 7. Fragment of a panoramic radiograph of impacted tooth 21 and a compound odontoma (OCp)

impacted deciduous tooth 65 and the enucleation of the odontoma with its capsule; the tooth bud of tooth 25 was left. Due to the child's age and her poor cooperation, the procedure was carried out in general anesthesia. Histopathological findings revealed OCp.

Preoteasa and Preoteasa mention the possibility of asymmetric resorption of the roots of deciduous teeth, caused not by a permanent tooth, but by a tumor, which may lead to tooth eruption disorders, i.e., to the occurrence of an impacted deciduous tooth and an impacted permanent tooth.[9] The authors describe a case of a 9-year-old patient who was admitted with impacted upper left central incisor 21, ectopically located upper lateral incisor, and retained deciduous incisors – left central 61 and left lateral 62. The X-ray image revealed OCp located between teeth 61 and 21, which caused asymmetric resorption of the root of deciduous tooth 61, which could have caused its delayed exfoliation.[9] Other authors, Amailuk and Grubor, report the possibility of changes in the shape of the roots of permanent teeth adjacent to a tumor.[5] In the group of our patients, there were 2 cases (10%) with a retained deciduous tooth above which an odontoma was found with a permanent tooth over it, but no changes were observed in the shape of the root of permanent teeth.

A 14-year-old patient, after the treatment of complete cleft lip and palate, came to us for the treatment of an odontoma. Paradowska-Stolarz and Kawala compared the incidence of dental anomalies in 154 patients with complete cleft lip and palate and in 151 healthy patients.[22] According to the authors, dental anomalies were much more common in patients with cleft and on the cleft side.[22] In our case, the odontoma was on the right side and the cleft was on the left side.

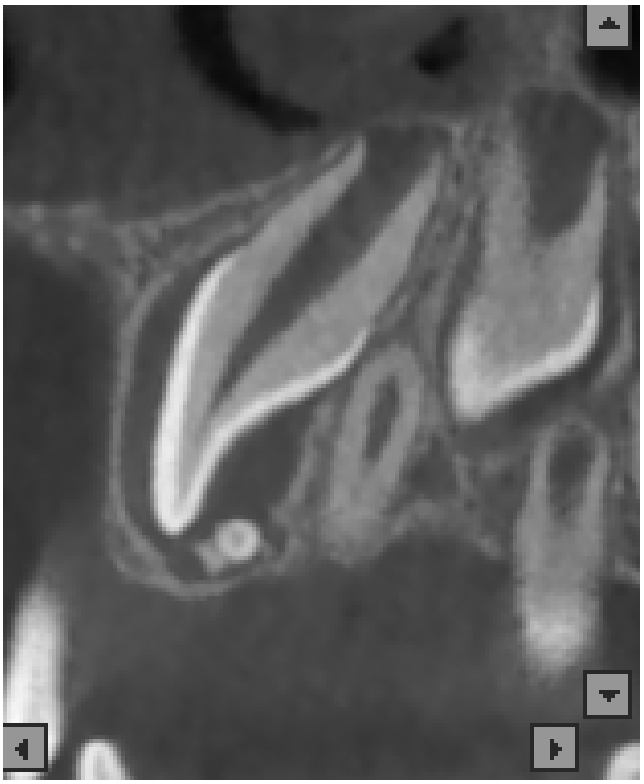

Fig. 8. Cone-beam computed tomography (CBCT) section showing unerupted tooth 21 and a compound odontoma (OCp)

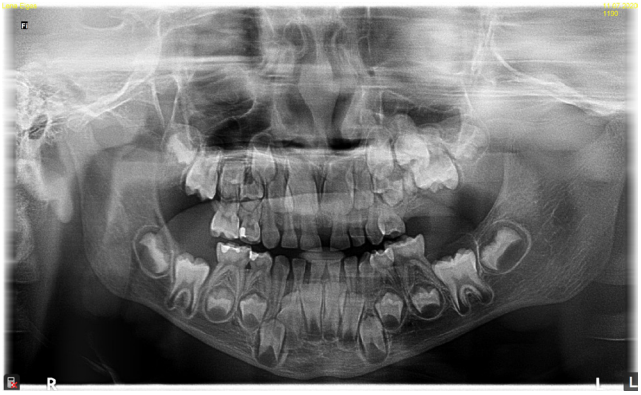

Fig. 9. Panoramic radiograph with impacted deciduous tooth 65 and a compound odontoma (OCp) in a 5-year-old patien

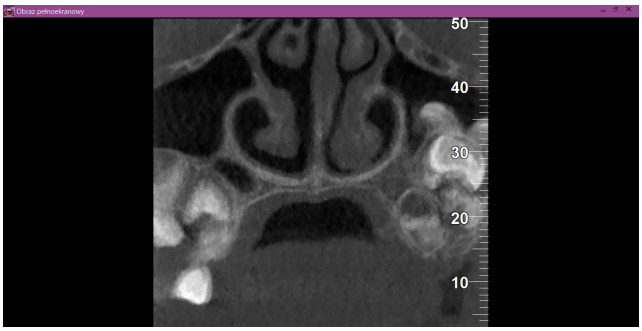

Fig. 10. CBCT of a 5-year-old patient with a compound odontoma (OCp) and impacted tooth 65 above it, and a tooth bud of permanent tooth 25 on the palatal side

The treatment of choice for odontomas is a surgery involving the complete enucleation of the tumor with its capsule. According to de Oliveira et al.[4] and Qazi et al.,[23] if an odontoma causes the impaction of the tooth bud of a permanent tooth, the tumor should be removed as soon as possible, before the root of the permanent tooth is fully developed (e.g., before it reaches half of its length) to prevent its permanent impaction. According to Uma, in turn, if after the removal of an odontoma, the impacted tooth does not erupt within 3 months of the procedure, the patient should be referred to an orthodontist.[3] In conclusion, odontomas should be removed as soon as possible to prevent the occurrence of complications in the form of dental abnormalities. In our clinic, the tumors were removed as soon as possible (within a month from the first appointment). In one case, the procedure was postponed in a 4-year-old boy (with antiphospholipid syndrome) who was receiving chemotherapy. On clinical examination, the protrusion of the alveolar process of the maxilla and a small gap between deciduous teeth 62 and 63 were observed, and there were no impacted teeth. The procedure was performed 4 months after the first appointment, during which OCp was diagnosed.

Many authors recommend the use of platelet-rich fibrin (PRF) and platelet-rich plasma (PRP) after surgical

procedures, presenting their benefits.[24–26] Healing after procedures in children very often proceeds without complications. In our cases, we did not observe dry sockets or bone inflammation. We did not perform odontoma removal procedures in patients taking bisphosphonates. In our research, we did not use PRP and PRF, but based on many positive studies and opinions, we are considering the use of PRP and PRF after odontoma removal procedures.

The literature does not report recurrences after the complete enucleation of the tumor with its capsule.[8,9] So far, we have not observed recurrences in any of our patients, either.

Conclusions

Odontomas are benign tumors that do not undergo neoplastic transformation. Such clinical signs as dental abnormalities, missing teeth, a protrusion on the alveolar process of the maxilla or the alveolar part of the mandible should prompt a dentist to perform diagnostic imaging. It is very often that, due to the absence of clinical signs, odontomas are discovered by accident during an X-ray performed before orthodontic treatment. The compound odontoma is the most common type of odontoma. Its radiological picture is characteristic – highly dense tooth-like radiopacities (odontoids) surrounded by a band of radiolucency with a sclerotic rim. The treatment of choice is a surgical procedure involving the complete enucleation of the tumor with its capsule, which results in full recovery.

Ethics approval and consent to participate

This retrospective study was approved by the institutional review board at The Children's Memorial Health Institute, Warsaw, Poland (No. of approval: 28/KBE/2021).

Data availability

The datasets supporting the findings of the current study are available from the corresponding author on reasonable request.

Consent for publication

Not applicable.

Use of AI and AI-assisted technologies

Not applicable.

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References

1. Park JC, Yang JH, Jo SY, Kim BC, Lee J, Lee W. Giant complex odontoma in the posteriori mandible: A case report and literature review. *Imaging Sci Dent*. 2018;48(4):289–293. doi:10.5624/isd.2018.48.4.289
2. Jayam C, Bandlapalli A, Patel N, Kashinath Choudhary RS. A case of impacted central incisor due to dentigerous cust associated with impacted compound odontome. *BMJ Case Rep*. 2014;2014:bcr2013202447. doi:10.1136/bcr-2013-202447
3. Uma E. Compound odontoma in anterior mandible – a case report. *Malays J Med Sci*. 2017;24(3):92–95. doi:10.21315/mjms2017.24.3.11
4. de Oliveira BH, Campos V, Marçal S. Compound odontoma – diagnosis and treatment: Three case reports. *Pediatr Dent*. 2001;23(2):151–157. PMID:11340730.
5. Amailuk P, Grubor D. Erupted compound odontoma: Case report of a 15-year-old Sudanese boy with a history of traditional dental mutilation. *Br Dent J*. 2008;204(1):11–14. doi:10.1038/bdj.2007.1184
6. Deliverska E, Gagov L, Stefanov L, Rubiev M. Odontoma – report of two cases and literature review. *MedInform*. 2016;1:414–421. doi:10.18044/Medinform.201631.414
7. Katiyar A, Gupta K, Solanki S, Gupta S. A case of dental hamartoma associated with unerupted primary canine in maxillary region. *Natl J Maxillofac Surg*. 2018;9(2):232–234. doi:10.4103/njms.NJMS_22_18
8. Khanum N, Shivalingu MM, Lingaraju N, Basappa S. Compound-complex odontoma: A case report of a rare variant. *J Indian Acad Oral Med Radiol*. 2014;26(4):463–466. doi:10.4103/0972-1363.155668
9. Preoteasa CT, Preoteasa E. Compound odontoma – morphology, clinical findings and treatment. Case report. *Rom J Morphol Embryol*. 2018;59(3):997–1000. PMID:30534846.
10. Kale YJ, Dahake PT, Patil PS, Dadpe MV. Management of a massive compound odontoma in a 9-year child. *Contemp Clin Dent*. 2016;7(3):409–412. doi:10.4103/0976-237X.188582
11. Gedik R, Müftüoğlu S. Compound odontoma: Differential diagnosis and review of the literature. *West Indian Med J*. 2014;63(7):793–795. doi:10.7727/wimj.2013.272
12. González-Alva P, Inoue H, Miyazaki Y, et al. Podoplanin expression in odontomas: Clinicopathological study and immunohistochemical analysis of 86 cases. *J Oral Sci*. 2011;53(1):67–75. doi:10.2334/josnusd.53.67
13. Boffano P, Zavattero E, Roccia F, Gallesio C. Complex and compound odontomas. *J Craniofac Surg*. 2012;23(3):685–688. doi:10.1097/SCS.0b013e31824dba1f
14. Baden E. Odontogenic tumors. *Pathol Annu*. 1971;6:475–568. PMID:4342310.
15. Nelson BL, Thompson LD. Compound odontoma. *Head Neck Pathol*. 2010;4(4):290–291. doi:10.1007/s12105-010-0186-2
16. Srivastava A, Annaji AG, Nyamati SB, Singh G, Shivakumar GC, Sahana S. Complex odontoma in both the jaws: A rare case report. *J Orofac Res*. 2012;2(1):56–60.
17. Bordini J Jr., Contar CM, Sarot JR, Fernandes A, Naval Machado MA. Multiple compound odontomas in the jaw: Case report and analysis of the literature. *J Oral Maxillofac Surg*. 2008;66(12):2617–2620. doi:10.1016/j.joms.2007.08.027
18. Yildrim-Oz G, Tosun G, Kiziloglu D, Durmuş E, Sener Y. An unusual association of odontomas with primary teeth. *Eur J Dent*. 2007;1(1):45–49. PMID:19212497. PMCID:PMC2612947.
19. Chhavi J, Aakriti G. A Case of complex odontoma associated with supplemental supernumerary teeth. *Ann Dent Res*. 2011;1(1):62–72.
20. Custódio M, Araujo JP, de Barros Gallo C, Trierveiler M. Gingival complex odontoma: A rare case report with a review of the literature. *Autops Case Rep*. 2018;8(1):e2018009. doi:10.4322/acr.2018.009
21. Aly H, Amer H, El Khatib O. Ameloblastic fibro-odontoma with chondroid tissue formation. *Contemp Oncol (Pozn)*. 2018;22(1):50–53. doi:10.5114/wo.2018.74395
22. Paradowska-Stolarz A, Kawala B. Dental anomalies in maxillary incisors and canines among patients with total cleft lip and palate. *Appl Sci*. 2023;13(11):6635. doi:10.3390/app13116635
23. Qazi HS, Ibrahim MW, Siddiqi KM, Zia AU. Compound odontoma causing impaction of mandibular canine: A case report. *POJ*. 2011;3(1):25–31.

24. Pietruszka P, Chruścicka I, Duś-Ilnicka I, Paradowska-Stolarz A. PRP and PRF – subgroups and divisions when used in dentistry. *J Pers Med*. 2021;11(10):944. doi:10.3390/jpm11100944
25. Egierska D, Perszke M, Mazur M, Duś-Ilnicka I. Platelet-rich plasma and platelet-rich fibrin in oral surgery: A narrative review. *Dent Med Probl*. 2023;60(1):177–186. doi:10.17219/dmp/147298
26. Gunasekaran S, Sakthivel S, Shanthala BM, Babu G, Vijayan V. Clinical application of platelet-rich fibrin in pediatric dentistry. *J Health Allied Sci*. 2022;12:186–190. doi:10.1055/s-0041-1736269

Original paper

Validation of the oral screen test as a tool for screening pediatric nasal obstruction

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D – writing the article; E – critical revision of the article; F – final approval of the article

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Abstract

Background. Nasal obstruction in pediatric patients can lead to serious issues, such as facial growth alterations, otitis media with effusion, and sleep disorders. Diagnosing nasal obstruction is challenging because subjective evaluations are often inaccurate, and objective measures like rhinomanometry are difficult to perform in children. This study proposes using an oral screen test as a rapid and cost-effective diagnostic method.

Objectives. The aim of the study is to validate the oral screen test as a method for diagnosing nasal obstruction in children. This objective is based on the observation that children with nasal obstruction do not tolerate the oral screen test well.

Material and methods. The validation of the diagnostic test was assessed based on the results of 104 children aged 4–15 years undergoing rhinomanometry. A silicone oral screen (Forwardontics®) and the Spanish version of the Sinus and Nasal Quality of Life Survey (SN-5) were used.

Results. The oral screen test yielded a positive result in 78 participants (75%). The children with a positive test result exhibited higher nasal resistance (561.3 ±140.5%) than those with a negative test result (102.0 ±3.4%), with an odds ratio (*OR*) of 18.5 (95% confidence interval (*CI*): 5.5–64.1).

Conclusions. The oral screen test is a highly sensitive and moderately specific method for diagnosing nasal obstruction in children, making it a useful screening tool in clinical practice.

Keywords: rhinomanometry, nasal obstruction, rhinitis, oral screen, SN-5

Highlights

- This study is the first to validate the oral screen as a tool for pediatric nasal obstruction screening.
- The oral screen test demonstrated high sensitivity and acceptable specificity in identifying children with nasal obstruction.
- A positive oral screen test was strongly correlated with increased nasal resistance.
- The oral screen test offers a simple, rapid and cost-effective method suitable for routine use in pediatric, otorhinolaryngology, and dental settings.

Introduction

Nasal obstruction is a common complaint in general pediatric consultations, otolaryngology, as well as pediatric odontology, either directly or indirectly due to its consequences, such as facial growth alterations,[1] otitis media with effusion,[2] and sleep disturbances.[3]

However, nasal obstruction is challenging to diagnose.[4] Subjective assessment is not enough, as children and their parents often lack the capacity to accurately self-diagnose nasal patency,[5] and symptoms are not always related to objective nasal obstruction.[6] Objective measures such as rhinomanometry, which is considered the gold standard, are not universally feasible due to the need for child collaboration and the time-consuming nature of the procedure.[4]

As children with nasal obstruction are forced to mouth breathe, it is supposed that they will not tolerate mouth taping. A simple method to force nasal breathing is the use of oral screens. Oral screens are common instruments in orthodontic consultations. These silicone pieces are designed to be placed between the cheeks, the lips and the teeth.[7] Oral screens are mainly used to prevent the perioral muscles from exerting forces on the teeth. The instruments block the oral air passage and force nasal breathing. Based on the observations from daily consultations, children with nasal obstruction do not tolerate oral screens and dismantle them. In consequence, it is hypothesized that the oral screen test may serve as a fast screening test to diagnose nasal obstruction.

The present research is designed with the aim of assessing the oral screen test as a diagnostic method of nasal obstruction.

Material and methods

Study sample

The diagnostic test validation method was followed. Before performing the physical examination, the parents were requested to provide informed consent for their children's participation in the study. The data collection process was planned before the index test and reference standard were performed.

Inclusion criteria

The inclusion criteria were as follows: 4–15 years of age; children attending the pediatric otolaryngology unit at the University Hospital Complex of Santiago de Compostela, Spain, and undergoing rhinomanometry.

The participants were selected consecutively from January 2022 to January 2023. All subjects undergoing rhinomanometry were included, irrespective of the indication for the procedure.

Exclusion criteria

The exclusion criteria encompassed children whose parents declined to participate in the study, subjects not collaborating during rhinomanometry, and those for whom rhinomanometry could not be performed (e.g., due to complete nasal obstruction, septal perforation).

The age limit was set at 4 years, as children under this age usually do not collaborate during rhinomanometry. The superior age limit was established at 15 years, as in Spain, this is the maximum age at which a patient may be considered pediatric.

The sample included healthy children undergoing a health-related consultation prior to the initiation of pediatric orthodontics, postoperative controls, as well as children with symptoms of nasal obstruction due to nasal septum deviation, adenoid hypertrophy, or rhinitis. The study population comprised healthy children in order to encompass the entire spectrum of nasal patency for the external validation of the diagnostic test.

Validation

The oral screen test was the primary evaluation, with rhinomanometry established as the gold standard. True positives were defined as cases where nasal resistance exceeded 100% of the normative data for the child's age. True negatives (controls) were defined as children with an unobstructed nose, showing nasal resistance below 100% of the normative data for their age. For the external validation process, a contingency table was used to assess sensitivity, specificity, as well as positive and negative predictive values.

Oral screen test

A silicone oral screen (Up-Locker Vacuum Activator; Forwardontics®, San Mateo, USA) was used (Fig. 1). It was recommended that children keep the oral screen in their mouth for 2 min, ensuring that their lips were firmly closed. The examiner observed the children, as some subjects may leave their lips apart and breathe through the mouth even with the oral screen. The participants were informed that they can stop the test if they experienced difficulty breathing through their nose. If a child tolerated the test, it was marked as negative. If the test was not tolerated, it was recorded as positive.

The test was performed before rhinomanometry to prevent observation bias.

Physical examination

The direct examination by nasofibroscope is currently considered the gold standard for the evaluation of adenoid and turbinate hypertrophy.[8] Turbinates were classified based on the study by Camacho et al.,[6] while adenoids were classified according to the study by Cassano et al.[7] The assessment of septal deviation was determined based on the study by Mariño-Sánchez et al.[8]

Adenoid hypertrophy was defined as a Cassano score greater than 2,[9] turbinate hypertrophy as a full Camacho score greater than 4,[10] and obstructive septal deviation as a Mariño-Sánchez score of 2.[11]

Sinus and Nasal Quality of Life Survey (SN-5)

In this study, the Spanish version of the Sinus and Nasal Quality of Life Survey (SN-5) was used.[12] The survey evaluates 5 clusters of symptoms, namely sinus infection, nasal obstruction, allergy, emotional distress, and activity limitations.[13] Each cluster contains symptoms that have been selected to assist parents in comprehending the nature of the assessment. Each cluster is rated on a 7-point Likert scale, ranging from 0 (never) to 6 (all the time). In addition to the symptomatic evaluation, caregivers were instructed to evaluate the child's overall quality of life on a visual analog scale (VAS) ranging from 0 (worst possible) to 10 (best possible).

Fig. 1. Silicone oral screen (Up-Locker Vacuum Activator; Forwardontics®, San Mateo, USA)

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Rhinomanometry

The recommendations of the International Committee on Standardization of Rhinomanometry were followed throughout the study.[11] Rhinomanometry was performed after 30 min of acclimatization, in a room where humidity was constant and the temperature regulated with a thermostat. The results were assessed using a reference pressure gradient across the nose of 150 Pa.[14]

Following the recommendations outlined in previous reports,[15] the results of nasal resistance were standardized according to pediatric reference values for each age subgroup.[16] The nasal resistance value that corresponded to the normative value of an age subgroup was designated as 100%. The results falling below 100% corresponded to children with unobstructed nasal passages, while the results exceeding 100% corresponded to subjects with increased levels of resistance. For the contingency table, 110% was selected as the resistance value of a patient with nasal obstruction.

In cases where rhinomanometry could not be performed due to severe obstruction, the data was designated as missing.

Statistical analysis

The normality of the quantitative variables was assessed through the implementation of the Shapiro–Wilk test. A comparison between quantitative and dichotomic variables was performed using the *t*-test for a normal distribution or the non-parametric variation rank sum test for a non-normal distribution. The statistical significance was set at $p < 0.05$. The statistical analysis was conducted using Stata 17 (StataCorp LLC, College Station, USA).

Results

Description of the study sample

After the selection process, a total of 104 participants were included in the study: 75 individuals with known obstructive nasal disorders; and 29 healthy participants. Diagnosed obstructive nasal disorders included hypertrophic rhinitis ($n = 68$), adenoid hypertrophy ($n = 46$) and septal deviation ($n = 5$).

The results pertaining to age, sex, nasal resistance, and SN-5 are summarized in Table 1.

Table 1. Characteristics of the study sample

Variable	Healthy individuals ($n = 29$)	Patients with obstructive nasal disorder ($n = 75$)	Total ($N = 104$)	Statistics
Age [years] *M* ±*SD*	9.4 ±0.5	10.4 ±0.3	10.1 ±0.2	$t = -1.80^{\#}$ $p = 0.075$
Sex, *n* (%)	F: 10 (34.48) M: 19 (65.52)	F: 35 (46.67) M: 40 (53.33)	F: 45 (43.27) M: 59 (56.73)	$\chi^2 = 1.26^{\#\#\#}$ $p = 0.261$
Nasal resistance [% of the normative age value] *M* ±*SD*	93.8 ±1.7	582.8 ±145.6	446.5 ±107.0	$t = -2.08^{\#}$ $p = 0.040$*
SN-5 obstruction score *M* ±*SD*	3.1 ±0.3	3.8 ±0.2	3.6 ±0.2	$z = -1.99^{\#\#}$ $p = 0.046$*
SN-5 VAS score *M* ±*SD*	6.1 ±0.4	6.0 ±0.2	6.0 ±0.2	$z = 0.20^{\#\#}$ $p = 0.841$
SN-5 full score *M* ±*SD*	10.3 ±1.1	11.2 ±0.6	10.9 ±0.5	$z = -0.20^{\#\#}$ $p = 0.475$

M – mean; *SD* – standard deviation; F – females; M – males; SN-5 – Sinus and Nasal Quality of Life Survey; VAS – visual analog scale; * statistically significant ($p < 0.05$); # Student's *t*-test; ## Mann–Whitney rank sum test; ### χ^2 test.

Oral screen test results

All the initially included participants underwent the oral screen test. The test yielded a positive result in 78 participants (75%).

Nasal resistance was higher in patients with a positive test result (561.3 ±140.5%) than those with a negative result (102.0 ±3.4%), with an odds ratio (*OR*) of 18.5 (95% confidence interval (*CI*): 5.5–64.1) (Table 2) (Fig. 2).

The SN-5 subdomain of nasal obstruction was higher in subjects with a positive test result ($z = -4.38$; $p < 0.001$) (Table 3) (Fig. 3).

Table 2. Contingency table used during the external validation process

Variable	Healthy individuals ($n = 29$)	Patients with obstructive nasal disorder ($n = 75$)	Statistics
Negative oral screen test ($n = 26$), *n* (%)	19 (65.52)	7 (9.33)	NPV = 73.08
Positive oral screen test ($n = 78$), *n* (%)	10 (34.48)	68 (90.67)	PPV = 87.18
Statistics	Sp = 65.52	Se = 90.67	*OR* =18.5 95% *CI*: 5.5–64.1

Sp – specificity; Se – sensitivity; NPV – negative predictive value; PPV – positive predictive value; *OR* – odds ratio; *CI* – confidence interval.

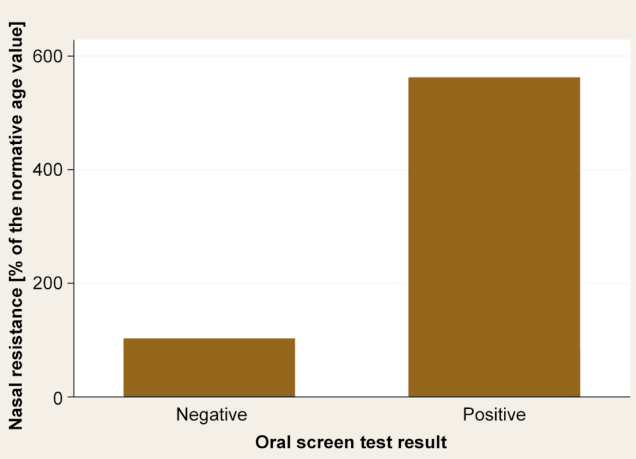

Fig. 2. Nasal resistance in oral screen test subgroups

Table 3. Sinus and Nasal Quality of Life Survey (SN-5) scores in oral screen test subgroups

Variable	Negative oral screen test ($n = 26$)	Positive oral screen test ($n = 78$)	Statistics
Nasal resistance [% of the normative age value]	102.0 ±3.4	561.3 ±140.5	$t = -1.8810^{\#}$ $p = 0.063$
SN-5 VAS score	6.7 ±0.4	5.8 ±0.2	$z = 1.64^{\#\#}$ $p = 0.101$
SN-5 full score	8.7 ±1.0	11.7 ±0.6	$z = -2.36^{\#\#}$ $p = 0.018$*
SN-5 obstruction score	2.5 ±0.2	4.0 ±0.2	$z = -4.38^{\#\#}$ $p < 0.001$*

* statistically significant ($p < 0.05$); # Student's t-test; ## Mann–Whitney rank sum test. Data presented as *M* ±*SD*.

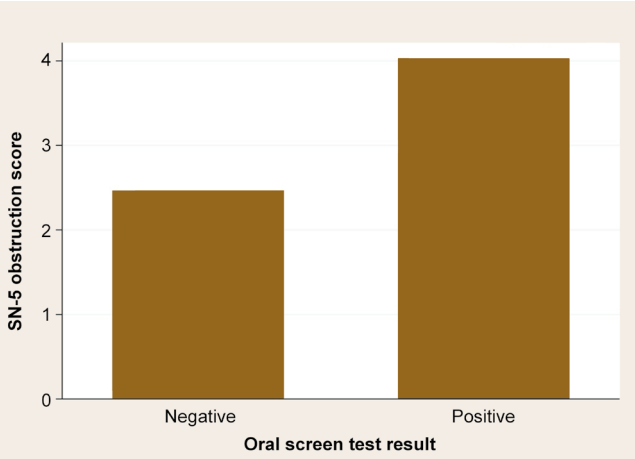

Fig. 3. Sinus and Nasal Quality of Life Survey (SN-5) subdomain of nasal obstruction in oral screen test subgroups

Discussion

The present study validated the oral screen test as an effective tool for diagnosing nasal obstruction in children, obtaining high sensitivity and a moderate to high specificity. The children with a positive result were found to be 18.5 times more likely to have nasal obstruction.

The oral screen test is not intended to substitute any other objective test. It has been developed as a fast, inexpensive and innocuous method to identify children at risk of nasal obstruction. If nasal obstruction is suspected, a specialist must be consulted to confirm the diagnosis and determine its exact cause.

For a diagnostic test to be considered suitable for screening purposes, it must meet specific criteria. Firstly, the evaluation must be affordable, easy to perform, innocuous, and have high sensitivity.[17] Secondly, the illness or condition under investigation must be highly prevalent, with early treatment altering the course of the disease. The oral screen test fulfills all these characteristics.

Pediatric nasal obstruction is a prevalent symptom that merits attention and prompt diagnosis. The exact prevalence of pediatric nasal obstruction remains unknown. However, some of its most common causes have been well studied. In Spain, for instance, rhinitis has been documented in 39% of the population,[18] while adenoid hypertrophy has been diagnosed in 42%.[19]

Nasal obstruction has been associated with several conditions, including caries,[20] middle ear disease, sinusitis, and alterations in facial growth. The early treatment of nasal obstruction has been related to the improvement in facial growth,[21] middle ear ventilation,[22] and the reduction of otitis media[23] and sinusitis.[24]

Despite its moderate specificity, the current study demonstrated a satisfactory level of sensitivity. This finding indicates that the test accurately identified the majority of children with nasal obstruction; however, several healthy children were misdiagnosed as obstructed. This phenomenon is presumably attributed to the fact that some children were restless during the test, stating that they could not appropriately breathe only to be authorized to remove the oral screen.

Regarding the cost of the procedure, it must be noted that, in our practice, we sterilize the oral screens. However, the utilization of single-use screens has the potential to result in an unacceptable level of plastic waste.

It was challenging to choose the gold standard test to compare against the oral screen test. Physical examination is widely regarded as the preferred method for diagnosing nasal obstruction. However, this technique can diagnose causes, but not the obstruction itself. The diagnosis of nasal obstruction can be made on the basis of either a subjective complaint, or an objective method. The subjective assessment by parents or children has been proven to be inadequate, as children and their parents often misdiagnose their own symptoms.[22] Several objective tests are available, each with its own advantages and limitations, such as rhinomanometry, acoustic rhinometry, rhinohigrometry, and nasal peak flow. Despite its limitations, the most widely accepted gold standard procedure is rhinomanometry.[4]

Rhinomanometry is criticized due to the low rate of collaboration exhibited by pediatric patients. Despite being true in some cases, most patients over 4 years of age can cooperate during rhinomanometry. Rhinomanometry is a time-consuming test. Both of these reasons justify using the oral screen test.

Another critique of rhinomanometry is that its reference values vary with age. Therefore, we have used normative data for each age subgroup in order to use relative values. This approach enabled the comparison of measurements of all the children despite their different ages.

The third limitation asserts that there is a low correlation between subjective measurements and rhinomanometric values. This claim is generally valid. However, in this study, we have assessed the quality of life via the SN-5. The study found that children with a positive oral screen test exhibited worse results in the SN-5 and the nasal obstruction subdomain of SN-5 (Table 3). Despite these outcomes, the overall quality of life assessed using the VAS score did not reach statistical significance. This is in contrast with previous studies, as most available evidence reported a low correlation between both measures. This phenomenon can be attributed, at least in part, to the fact that a number of children under study were regular patients and were more aware of their own bodies and illnesses. However, the absence of an association between the 2 measurements should not be a limitation for diagnosing nasal obstruction alone, independent of symptoms, as children are treated not only based on their self-reported symptoms. Nasal breathing is in itself a favorable outcome. As previously discussed, adequate nasal breathing promotes optimal facial growth, dental health, and reduced incidence of middle ear illnesses.[24] Therefore, the objective of diagnosing and treating nasal obstruction could be complementary to diagnosing and treating its symptoms.

Another highly debated question in pediatric rhinomanometry is whether the procedure should be anterior or posterior. In the anterior rhinomanometry, the differential pressure is measured anterior to the adenoids. However, in the posterior rhinomanometry, the pressure is measured in the oropharynx, inferior to the adenoid pad. Conceptually, posterior rhinomanometry is a superior method for diagnosing nasal obstruction caused by adenoid hypertrophy. However, we decided to use anterior rhinomanometry, given that children tend to be less collaborative with the posterior method.[25]

When assessing nasal obstruction, oral screen tests do not measure the severity of the obstruction but whether a certain cut-off value is surpassed or not. Once the nasal cavity is obstructed, oral breathing is forced, thereby

precipitating the majority of the observed complications. This phenomenon can be attributed to the non-linear relationship between nasal airflow and nasal resistance, which exhibits an exponential relationship to the fourth power (Fig. 4). Therefore, at around 0.3 Pa/mL, small changes in nasal resistance are related to substantial variations in nasal airflow. The oral screen test is a diagnostic tool that can be used to identify cases of oral breathing, irrespective of the extent of nasal obstruction.

The present study did not use nasal decongestants, which have been demonstrated to be effective in addressing turbinate hypertrophy[26] and adenoid hypertrophy[27] in children. Future studies will assess this topic.

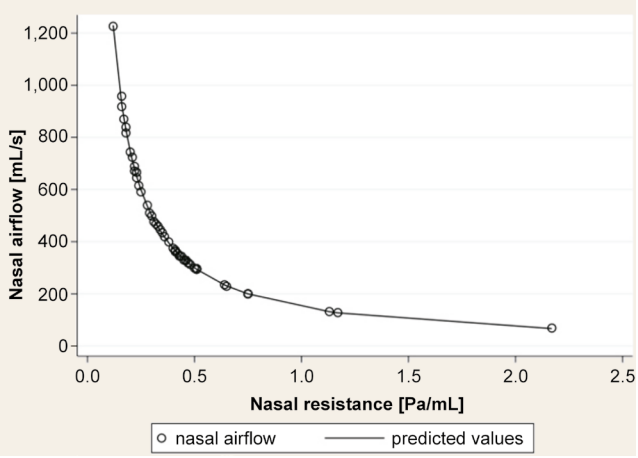

Fig. 4. Relationship between nasal airflow and nasal resistance in the study sample

Limitations

Despite the aforementioned strengths of the study, it is important to acknowledge its limitations. The study was a validation test, and a small sample size was used. However, given the characteristics of the test, it is our intention to progressively increase this sample size in order to obtain more solid results. The second limitation is that nasal obstruction can be caused by several entities, such as septal deviation, allergic rhinitis or adenoid hypertrophy, among others.[28] In this study, we have included different causes of nasal obstruction in order to diminish the potential selection bias. Even though not all causes of nasal obstruction have been included, the findings of this study should be interpreted with caution.

Conclusions

Nasal obstruction in pediatric patients has garnered increasing attention from otolaryngologists, pediatricians and odontologists. This trend is evidenced by a notable increase in the number of papers addressing this subject. This study is the first to assess an odontologic tool, the oral screen, as a screening method for diagnosing pediatric nasal obstruction. It demonstrated a high level of sensitivity and a positive predictive value. Given the characteristics of the test and the high prevalence of nasal obstruction, the oral screen test can be incorporated into the daily practice of professionals who treat pediatric patients.

Ethics approval and consent to participate

The parents of the children who participated in the study provided informed consent.

Data availability

The datasets generated and/or analyzed during the current study are available from the corresponding author on reasonable request.

Consent for publication

Not applicable.

Use of AI and AI-assisted technologies

Not applicable.

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References

1. Fernandes P, Pinto J, Ustrell-Torrent J. Relationship between oro and nasopharynx permeability and the direction of facial growth. *Eur J Paediatr Dent*. 2017;18(1):37–40. doi:10.23804/ejpd.2017.18.01.08
2. Calvo-Henriquez C, Martínez-Seijas P, Boronat-Catalá B, et al. Assessing the ability of children and parents to rate their nasal patency. A cross sectional study. *Int J Pediatr Otorhinolaryngol*. 2022;156:111094. doi:10.1016/j.ijporl.2022.111094
3. Ta NH, Gao J, Philpott C. A systematic review to examine the relationship between objective and patient-reported outcome measures in sinonasal disorders: Recommendations for use in research and clinical practice. *Int Forum Allergy Rhinol*. 2021;11(5):910–923. doi:10.1002/alr.22744
4. Rai A, Jain A, Datarkar A, Bhawalkar A. Use of oral screen for preventing soft tissue injuries associated with use of arch bars: A prospective randomized clinical study. *Oral Maxillofac Surg*. 2019;23(3):291–295. doi:10.1007/s10006-019-00780-3
5. Kindermann CA, Roithmann R, Lubianca Neto JF. Sensitivity and specificity of nasal flexible fiberoptic endoscopy in the diagnosis of adenoid hypertrophy in children. *Int J Pediatr Otorhinolaryngol*. 2008;72(1):63–67. doi:10.1016/j.ijporl.2007.09.013
6. Camacho M, Zaghi S, Certal V, et al. Inferior turbinate classification system, grades 1 to 4: Development and validation study. *Laryngoscope*. 2015;125(2):296–302. doi:10.1002/lary.24923
7. Cassano P, Gelardi M, Cassano M, Fiorella ML, Fiorella R. Adenoid tissue rhinopharyngeal obstruction grading based on fiberendoscopic findings: A novel approach to therapeutic management. *Int J Pediatr Otorhinolaryngol*. 2003;67(12):1303–1309. doi:10.1016/j.ijporl.2003.07.018

8. Mariño-Sánchez FS, Valls-Mateus M, Ruiz-Echevarría K, et al. Nasal obstructive disorders induce medical treatment failure in paediatric persistent allergic rhinitis (the NODPAR Study). *Pediatr Allergy Immunol*. 2017;28(2):176–184. doi:10.1111/pai.12679
9. Calvo-Henríquez C, Valencia-Blanco B, Boronat-Catalá B, et al. Cross-cultural adaptation of the sinus and nasal quality of life survey (SN-5) to Spanish. *Int J Pediatr Otorhinolaryngol*. 2020;139:110425. doi:10.1016/j.ijporl.2020.110425
10. Kay DJ, Rosenfeld RM. Quality of life for children with persistent sinonasal symptoms. *Otolaryngol Head Neck Surg*. 2003;128(1):17–26. doi:10.1067/mhn.2003.41
11. Clement PAR, Gordts F; Standardisation Committee on Objective Assessment of the Nasal Airway, IRS, and ERS. Consensus report on acoustic rhinometry and rhinomanometry. *Rhinology*. 2005;43(3):169–179. PMID:16218509.
12. Zapletal A, Chalupová J. Nasal airflow and resistance measured by active anterior rhinomanometry in healthy children and adolescents. *Pediatr Pulmonol*. 2002;33(3):174–180. doi:10.1002/ppul.10066
13. Principato JJ, Wolf P. Pediatric nasal resistance. *Laryngoscope*. 1985;95(9 Pt 1):1067–1069. PMID:4033328.
14. Wilson JM, Jungner YG. Principles and practice of mass screening for disease [in Spanish]. *Bol Oficina Sanit Panam*. 1968;65(4):281–393. PMID:4234760.
15. Batlles-Garrido J, Torres-Borrego J, Rubí-Ruiz T, et al. Prevalence and factors linked to allergic rhinitis in 10 and 11-year-old children in Almería. Isaac Phase II, Spain. *Allergol Immunopathol (Madr)*. 2010;38(3):135–141. doi:10.1016/j.aller.2009.09.005
16. Pereira L, Monyror J, Almeida FT, et al. Prevalence of adenoid hypertrophy: A systematic review and meta-analysis. *Sleep Med Rev*. 2018;38:101–112. doi:10.1016/j.smrv.2017.06.001
17. Calvo Henriquez C, Rodríguez-Rivas P, Mayo-Yáñez M, et al. Allergic rhinitis and dental caries: A systematic review. *Allergol Immunopathol (Madr)*. 2023;51(2):168–176. doi:10.15586/aei.v51i2.752
18. do Nascimento RR, Masterson D, Trindade Mattos C, de Vasconcellos Vilella O. Facial growth direction after surgical intervention to relieve mouth breathing: A systematic review and meta-analysis. *J Orofac Orthop*. 2018;79(6):412–426. doi:10.1007/s00056-018-0155-z
19. Mikals SJ, Brigger MT. Adenoidectomy as an adjuvant to primary tympanostomy tube placement: A systematic review and meta-analysis. *JAMA Otolaryngol Head Neck Surg*. 2014;140(2):95–101. doi:10.1001/jamaoto.2013.5842
20. van den Aardweg MT, Schilder AG, Herkert E, Boonacker CW, Rovers MM. Adenoidectomy for otitis media in children. *Cochrane Database Syst Rev*. 2010;2010(1):CD007810. doi:10.1002/14651858.CD007810.pub2
21. Vasco CTC, Morais HC, Avelino MAG. Systematic review of the literature on surgical treatment of chronic rhinosinusitis in children: What is the best approach? *Rev Paul Pediatr*. 2020;38:e2018068. doi:10.1590/1984-0462/2020/38/2018068
22. Occasi F, Duse M, Vittori T, et al. Primary school children often underestimate their nasal obstruction. *Rhinology*. 2016;54(2):164–169. doi:10.4193/Rhino15.120
23. Valero A, Navarro AM, Del Cuvillo A, et al. Position paper on nasal obstruction: Evaluation and treatment. *J Investig Allergol Clin Immunol*. 2018;28(2):67–90. doi:10.18176/jiaci.0232
24. Saito K, Ono T, Mochida M, Ohyama K. Changes in nasorespiratory function in association with maxillary distraction osteogenesis in subjects with cleft lip and palate. *Cleft Palate Craniofac J*. 2006;43(1):75–83. doi:10.1597/04-094r1.1
25. Parker AJ, Maw AR, Powell JE. Rhinomanometry in the selection for adenoidectomy and its relation to preoperative radiology. *Int J Pediatr Otorhinolaryngol*. 1989;17(2):155–161. doi:10.1016/0165-5876(89)90090-6
26. Calvo-Henriquez C, Mayo-Yáñez M, Lechien JR, et al. Looking for a cutoff value for the decongestant test in children suffering with turbinate hypertrophy. *Eur Arch Otorhinolaryngol*. 2021;278(10):3821–3826. doi:10.1007/s00405-021-06657-0
27. Calvo-Henriquez C, Rivero I, Maldonado-Alvarado B, et al. Rhinomanometry with and without decongestant used to select children for adenoidectomy: A cohort study. *Eur Arch Otorhinolaryngol*. 2022;280(2):723–729. doi:10.1007/s00405-021-06657-0
28. Isaac A, Major M, Witmans M, et al. Correlations between acoustic rhinometry, subjective symptoms, and endoscopic findings in symptomatic children with nasal obstruction. *JAMA Otolaryngol Head Neck Surg*. 2015;141(6):550–555. doi:10.1001/jamaoto.2015.0468

Original paper

Knowledge, perception and attitude of dentists regarding the role of artificial intelligence in the field of pediatric dentistry: An online questionnaire study

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D – writing the article; E – critical revision of the article; F – final approval of the article

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Abstract

Background. Knowledge on the potential applications of artificial intelligence (AI) as a diagnostic instrument in the domain of pediatric dentistry is imperative, as AI may significantly influence present and future dental practice.

Objectives. The present study aimed to evaluate the knowledge, perception and attitude of pediatric dentists and postgraduate students in the pediatric specialty with regard to the employment of AI in pediatric dental practice.

Material and methods. An online questionnaire survey was conducted among 375 participants (92 postgraduates, 203 faculty members and 80 private practitioners), who were provided with 19 close-ended questions through the Google Forms link sent via email. The questions referred to the knowledge, perception and attitude of the participants, with 17 questions answered using a three-point Likert scale and 2 of them being multiple-choice questions. The responses were analyzed using the χ^2, Kruskal–Wallis and Mann–Whitney *U* tests.

Results. A total of 62% of the participants were familiar with the term 'artificial intelligence', and the majority confirmed that AI could be used for the identification of plaque (57%) and supernumerary teeth (52%), the detection of early childhood caries (ECC) (68%) and the ectopic eruption of first permanent molars (67%), the assessment of child psychology (82%), and the estimation of chronological age (67%). Most participants felt that AI training should be incorporated into the postgraduate curriculum (82%) and were willing to introduce AI to clinical practice (87%). The barriers related to the use of AI were high costs (83%), the lack of training after graduation (78%), the lack of technical knowledge (77%), the fear of misdiagnosis (73%), and the lack of awareness (71%).

Conclusions. The present study concluded that although most pedodontists and postgraduate students had knowledge on AI, there were many obstacles connected with the use of AI in the field of pediatric dentistry. Therefore, the basic training of AI should be included in the curriculum of postgraduate studies.

Keywords: knowledge, survey, questionnaire, pediatric dentistry, artificial intelligence

Highlights

- Faculty members and postgraduates support the integration of AI, while private practitioners remain skeptical about the technology.
- Challenges in AI implementation in dentistry are high costs, the lack of proper training and insufficient technical knowledge.
- Younger pediatric dentists are more knowledgeable and open to incorporating AI into their professional practice.

Introduction

In 1956, John McCarthy introduced the concept of artificial intelligence (AI).[1] This field of study can be defined as the practical application of computer science, which aims to replicate the cognitive abilities of humans. Artificial intelligence is focused on the development of intelligent systems that possess the capacity to think and learn, similar to their human counterparts.[1] Artificial intelligence and its subcategories – machine learning (ML) and deep learning (DL) – have been integrated into numerous facets of dentistry, encompassing the field of pediatric dentistry.[2]

Machine learning, a subset of AI, relies on algorithms to make predictions based on datasets. Its objective is to enable machines to learn from the available data and solve problems without human intervention. In contrast, neural networks consist of algorithms that process signals using artificial neurons. Deep learning, a subset of ML, utilizes a deep neural network with multiple computational layers to analyze the input data. The purpose is to create a neural network that would be able to automatically identify patterns and enhance feature detection. Deep learning is also referred to as a convolutional neural network (CNN). In the field of pediatric dentistry, CNNs are primarily employed to analyze large and intricate images, thereby improving the accuracy and efficiency of diagnostic processes.[3]

The implementation of AI can potentially eliminate some laborious and time-consuming procedures so far performed by dental professionals. Furthermore, it is plausible to enhance the health of the general population at decreased expenditure, provide customized, preventative and predictive dental treatment, and consolidate the accessibility of healthcare for all. Artificial intelligence possesses the capability to elevate the benchmarks of dental care, refine the precision and efficacy of diagnostic procedures, devise enhanced visual aids for treatment, simulate outcomes, and forecast oral ailments and well-being.[2]

Artificial intelligence has been commonly employed in the field of pediatric dentistry to provide support for pedodontists in improving the accuracy of their diagnoses. These models prove to be immensely beneficial at both the individual and community levels, as they effectively classify children into various risk groups, at the same time enabling the identification and enumeration of teeth, the identification of supernumerary teeth, the diagnosis of the early ectopic eruption of first permanent molars, and the assessment of age, among other functionalities. Furthermore, they can serve as valuable tools in the planning and evaluation of oral health programs in schools, thereby increasing the awareness of dental well-being among children.[2,4]

Despite the numerous advantages of AI, its use in pediatric dentistry remains significantly restricted. This can be ascribed to a multitude of factors, including the limited knowledge and understanding of AI principles among pedodontists, fear and apprehension with regard to using AI software, and the lack of proper knowledge, training and skills for using AI in clinical practice. Moreover, the public is reluctant to place confidence in the outcomes of AI. As a result, various challenges persist, necessitating a proactive approach to finding a resolution.[5]

Owing to the lack of previous studies, the present study was conducted to assess the knowledge, perception, attitude, and barriers regarding the use of AI in the field of pediatric dentistry among postgraduates, faculty members and private practitioners.

Material and methods

Study design and setting

The present cross-sectional, descriptive, questionnaire-based study was conducted from July to September 2023 at the Department of Pedodontics at Yogita Dental College and Hospital, Khed, India, on postgraduate students, faculty members and private practitioners in the field of pedodontics, who were registered with the Indian Society of Pedodontics and Preventive Dentistry (ISPPD). This survey was approved by the institutional ethics committee (approval No. EC/NEW/INST/2022/2959/Y23/212), and was conducted in accordance with the ethical principles outlined in the Declaration of Helsinki.

Study participants and data collection procedure

A formal letter of request was submitted to the administrative officers of ISPPD, seeking authorization to disclose the list of pedodontists, along with their

corresponding email addresses, who have been duly registered as postgraduate students, faculty members and private practitioners in the field of pediatric dentistry. From the list of 2,974 individuals provided by ISPPD, it was observed that there were 680 postgraduate students, 694 private practitioners and 1,600 individuals who were faculty members. This distribution followed an approximate ratio of 1:1:2.3. Consequently, for the purpose of the present study, the participants were selected through the non-probabilistic convenience sampling technique. Specifically, a Google Forms link, accompanied by a set of instructions and consent forms, was randomly dispatched via email to 650 ISPPD members (150 postgraduate students, 150 private practitioners and 350 faculty members). This distribution was carried out in accordance with the initial ratio of 1:1:2.3. The participants were afforded the opportunity to complete the questionnaire on a single occasion, with no time restrictions. Subsequently, after clarifying the objective and ensuring the preservation of confidentiality, and after the participants had provided informed consent and expressed willingness to participate in the survey, their responses were gathered. The survey guaranteed the maintenance of confidentiality, as it did not require the disclosure of personal information regarding the participants' identities. Additionally, it was explicitly stated in the participant recruitment statement that participation in the survey was completely voluntary. The responses were provided anonymously, without any identifying information. The principal investigator was the sole individual granted access to the data. Duplicated entries were rectified, and only responses that were fully completed were considered. Reminder emails to complete the forms were sent after 1 month.

Pedodontists employed as faculty members and in private practice, along with postgraduate students pursuing their course in the specialty of pedodontics, who agreed to participate in the study, were included. Undergraduates, participants who did not provide consent, as well as those who provided incomplete forms, were excluded from the study.

Sample size estimation

The sample size estimator provided by Calculator.net (https://www.calculator.net/sample-size-calculator.html) was used to establish the appropriate sample size. According to the statistics from ISPPD, the total number of pedodontists in 2023, including postgraduate students, was 2,974. Based on this, statistical power analysis was performed to calculate the sample size. The suggested sample size was 341, with a design effect of 50%, a margin of error of 5% and a confidence level of 95%. Considering a 60% response rate, the estimated sample size was 569. Previous studies yielded a fluctuating rate of response ranging from 50% to 60%.[6–8] Consequently, the current study was undertaken encompassing a sample size of 650 individuals.

Study instrument

The questionnaire was divided into 4 domains. The first one, known as part A, focused on 6 open-ended questions on sociodemographic characteristics; the participants entered their age, gender, designation, workplace, region, and years of experience. Part B consisted of 8 close-ended questions identifying the basic knowledge of the participants on the use of AI in the field of pediatric dentistry (7 questions used a three-point Likert scale (agree, neutral, disagree) and 1 was a multiple-choice question). Part C consisted of 6 questions assessing the participants' perception with regard to the use of AI (5 questions used a three-point Likert scale and 1 was a multiple-choice question). Part D comprised 5 questions on a Likert scale, regarding the attitude of the participants toward AI.

Pre-testing and content validity of the questionnaire

The questionnaire was created through the collaborative efforts of 5 professionals, including 3 pedodontists, one AI expert and one researcher with a decade of experience, who were not part of the study. Subsequent to the evaluation conducted by these 5 specialists, Aiken's V statistic was calculated, revealing a value of 0.92, indicating a favorable level of content validity. To assess the dependability of the inquiries, a preliminary examination or pre-testing of the questionnaire was conducted on 35 individuals who were not involved in the study. The reliability of the questionnaire was assessed by employing Cronbach's alpha; the test yielded a value of 0.87 for knowledge-based questions, and 0.92 for perception- and attitude-based questions. To determine the level of agreement among the questions, the questionnaire was administered to the same cohort after a 2-week interval. Inter-observer agreement was evaluated using the kappa coefficient, which amounted to 0.94.

Scoring criteria

Each question (except for questions 2 and 14) was scored as agree = 5, neutral = 2 or disagree = 0. The maximum score for knowledge was 35 (7 × 5), 25 (5 × 5) for perception and 25 (5 × 5) for attitude. The mean and standard deviation ($M \pm SD$) values were calculated for knowledge, perception and attitude.

Statistical analysis

The collected data was subjected to statistical analysis with the use of the IBM SPSS Statistics for Windows software, v. 22.0 (IBM Corp., Armonk, USA). The normal distribution of the data was confirmed using the Shapiro–Wilk test. The compilation and presentation of sociodemographic variables and the participants' responses

were achieved using frequency distributions. To determine significant differences between the variables, non-parametric tests, including the χ^2 test, were employed. The knowledge, perception and attitude scores were calculated and analyzed using the Mann–Whitney *U* test (for gender) and the Kruskal–Wallis test (for other variables), followed by post-hoc analysis with Dunn's test. The correlation for ordinal data, such as the knowledge, perception and attitude scores, was calculated with Spearman's ρ test. The level of significance was set at $p \leq 0.05$.

Results

Demographic details of the respondents

The baseline demographic characteristics of the participants are presented in Table 1. A total of 375 participants completed the online survey, with a response rate of 58%. A total of 118 participants represented the age group of 26–30 years, and the smallest number of participants were in the age group of 56–60 years. With regard to gender, 57% of the participants were male, and 43% were female. Of the 375 participants, 54% were faculty members, 25% were postgraduates and 21% were private practitioners. Of the sample, 41% had less than 5 years of experience, and 34% had more than 10 years of experience (Table 1).

Table 1. Baseline characteristics of the study participants (*N* = 375)

Variable		*n* (%)
Age [years]	26–30	118 (31.47)
	31–35	78 (20.80)
	36–40	42 (11.20)
	41–45	52 (13.87)
	46–50	36 (9.60)
	51–55	28 (7.47)
	56–60	21 (5.60)
Gender	M	214 (57.07)
	F	161 (42.93)
Designation	postgraduate	92 (24.53)
	faculty member	203 (54.13)
	private practitioner	80 (21.33)
Experience [years]	<5	154 (41.07)
	5–10	93 (24.80)
	>10	128 (34.13)

M – male; F – female.

Assessing the knowledge of the participants on AI and its applications

A total of 62% of the participants were familiar with the term 'artificial intelligence', with a statistically significant difference between the groups ($p = 0.001$); among them, there were 74% of faculty members, followed by 61% of postgraduates. As many as 44% of private practitioners were unaware of the term 'AI'. The most common source of AI information was industry news (78%), and the least common professional organizations (45%). Most of the participants agreed that AI could be used for identifying plaque in children, with no significant differences between the participants ($p > 0.05$), whereas statistically significant differences were observed when the participants were asked about the use of AI for the identification of supernumerary teeth, the detection of early childhood caries (ECC) and the ectopic eruption of first permanent molars, and the assessment of child psychology ($p < 0.05$). Most faculty members agreed with the use of AI for various purposes in the field of pediatric dentistry. A total of 67% of postgraduates expressed their agreement with the implementation of AI in the identification of supernumerary teeth, while 38% of private practitioners held a different opinion. The majority of faculty members (81%) and private practitioners (56%) were in favor of utilizing AI for the detection of EEC, although 41% of postgraduates disagreed with this notion. Similarly, the majority of faculty members (77%) and postgraduates (71%) accepted the use of AI for the estimation of chronological age in children, whereas 33% of private practitioners expressed their disagreement. With regard to the detection of the ectopic eruption of first permanent molars in early mixed dentition, most faculty members and private practitioners were supportive of employing AI, with only 59% of postgraduates agreeing to the same. Furthermore, the majority of the respondents agreed with the use of AI for assessing child psychology (Table 2).

Assessing the perception of AI by the participants

A total of 81% of the participants wanted to use AI software in the future, and the highest agreement was noticed among the faculty members. Most of the respondents (82%) felt that it should be part of postgraduate training, with a statistically significant difference between the groups ($p = 0.026$). As many as 85% of the participants, mostly faculty members (88%) and private practitioners (85%), wanted a professional training course on the use of AI ($p > 0.05$). Also, 87% of the participants agreed that AI could be used in clinical practice, and 85% of the participants would recommend the use of AI to their fellow colleagues, with statistically significant differences noted ($p < 0.05$), where the highest agreement was noticed among the private practitioners and the lowest among the postgraduates. The barriers related to the use of AI in the field of pediatric dentistry were high costs (83%), the lack of postgraduate training (78%), the lack of technical knowledge (77%), the fear of incorrect diagnosis (73%), and the lack of awareness (71%), as shown in Table 2.

Table 2. Comparison of the response frequencies of the participants according to their designation

Questions and answer options			Postgraduates *N* = 92	Faculty members *N* = 203	Private practitioners *N* = 80	Total *N* = 375	*p*-value
Knowledge	1. The term 'artificial intelligence' is known by most of us	agree	56 (60.87)	151 (74.38)	24 (30.00)	231 (62)	0.001*
		neutral	15 (16.30)	18 (8.87)	21 (26.25)	54 (14)	
		disagree	21 (22.83)	34 (16.75)	35 (43.75)	90 (24)	
	2. What is the source of information about AI (multiple answers can be ticked)?	research papers	45 (48.91)	167 (82.27)	25 (31.25)	237 (63)	0.015*
		online data	56 (60.87)	154 (75.86)	34 (42.50)	244 (65)	
		professional organizations	23 (25.00)	121 (59.61)	26 (32.50)	170 (45)	
		industry news	54 (58.70)	176 (86.70)	62 (77.50)	292 (78)	
		conferences	45 (48.91)	178 (87.68)	45 (56.25)	268 (71)	
	3. AI can be used for the identification of plaque-affected teeth in children	agree	56 (60.87)	123 (60.59)	34 (42.50)	213 (57)	0.056
		neutral	12 (13.04)	25 (12.32)	12 (15.00)	49 (13)	
		disagree	24 (26.09)	55 (27.09)	34 (42.50)	113 (30)	
	4. AI can be used for the identification of supernumerary teeth in children	agree	62 (67.39)	106 (52.22)	26 (32.50)	194 (52)	0.001*
		neutral	8 (8.70)	25 (12.32)	24 (30.00)	57 (15)	
		disagree	22 (23.91)	72 (35.47)	30 (37.50)	124 (33)	
	5. AI can detect EEC	agree	45 (48.91)	165 (81.28)	45 (56.25)	255 (68)	0.001*
		neutral	9 (9.78)	12 (5.91)	22 (27.50)	43 (11)	
		disagree	38 (41.30)	26 (12.81)	13 (16.25)	77 (21)	
	6. AI can estimate chorological age in children	agree	65 (70.65)	156 (76.85)	32 (40.00)	253 (67)	0.001*
		neutral	12 (13.04)	28 (13.79)	22 (27.50)	62 (17)	
		disagree	15 (16.30)	19 (9.36)	26 (32.50)	60 (16)	
	7. AI can detect the ectopic eruption of first permanent molars in early mixed dentition	agree	54 (58.70)	145 (71.43)	52 (65.00)	251 (67)	0.012*
		neutral	12 (13.04)	25 (12.32)	18 (22.50)	55 (15)	
		disagree	26 (28.26)	33 (16.26)	10 (12.50)	69 (18)	
	8. AI can be effectively used for assessing child psychology	agree	62 (67.39)	188 (92.61)	56 (70.00)	306 (82)	0.001*
		neutral	8 (8.70)	8 (3.94)	14 (17.50)	30 (8)	
		disagree	22 (23.91)	7 (3.45)	10 (12.50)	39 (10)	
Perception	1. Will you use AI software on patients in the future?	agree	76 (82.61)	176 (86.70)	52 (65.00)	304 (81)	0.001*
		neutral	8 (8.70)	19 (9.36)	22 (27.50)	49 (13)	
		disagree	8 (8.70)	8 (3.94)	6 (7.50)	22 (6)	
	2. Will you go for any professional training on the use of AI in pediatric dentistry?	agree	72 (78.26)	178 (87.68)	68 (85.00)	318 (85)	0.247
		neutral	8 (8.70)	8 (3.94)	6 (7.50)	22 (6)	
		disagree	12 (13.04)	17 (8.37)	6 (7.50)	35 (9)	
	3. AI should be an integral part of postgraduate training	agree	75 (81.52)	165 (81.28)	66 (82.50)	306 (82)	0.026*
		neutral	5 (5.43)	25 (12.32)	12 (15.00)	42 (11)	
		disagree	12 (13.04)	13 (6.40)	2 (2.50)	27 (7)	
	4. AI can be employed in clinical practice after proper training only	agree	65 (70.65)	186 (91.63)	74 (92.50)	325 (87)	0.001*
		neutral	15 (16.30)	6 (2.96)	4 (5.00)	25 (7)	
		disagree	12 (13.04)	11 (5.42)	2 (2.50)	25 (7)	
	5. Do you recommend the use of AI in pediatric dentistry to fellow colleagues?	agree	67 (72.83)	178 (87.68)	72 (90.00)	317 (85)	0.007*
		neutral	6 (6.52)	8 (3.94)	6 (7.50)	20 (5)	
		disagree	19 (20.65)	17 (8.37)	2 (2.50)	38 (10)	
	6. What are the barriers in the use of AI in the field of pediatric dentistry? (multiple answers can be ticked)?	lack of awareness	56 (60.87)	154 (75.86)	56 (70.00)	266 (71)	0.010*
		lack of training at the postgraduate level	52 (56.52)	178 (87.68)	63 (78.75)	293 (78)	
		lack of technical knowledge	65 (70.65)	167 (82.27)	56 (70.00)	288 (77)	
		fear of over- or underestimating the problem	32 (34.78)	176 (86.70)	64 (80.00)	272 (73)	
		high costs	78 (84.78)	180 (88.67)	54 (67.50)	312 (83)	

Questions and answer options			Postgraduates $N = 92$	Faculty members $N = 203$	Private practitioners $N = 80$	Total $N = 375$	p-value
Attitude	1. I am impressed by what AI can do	agree	62 (67.39)	168 (82.76)	67 (83.75)	297 (79)	0.033*
		neutral	10 (10.87)	12 (5.91)	5 (6.25)	27 (7)	
		disagree	20 (21.74)	23 (11.33)	8 (10.00)	51 (14)	
	2. AI has great future in the field of pediatric dentistry	agree	68 (73.91)	163 (80.30)	75 (93.75)	306 (82)	0.010*
		neutral	6 (6.52)	15 (7.39)	2 (2.50)	23 (6)	
		disagree	18 (19.57)	25 (12.32)	3 (3.75)	46 (12)	
	3. AI has better diagnostic ability than an experienced pedodontist	agree	56 (60.87)	123 (60.59)	56 (70.00)	235 (63)	0.269
		neutral	16 (17.39)	25 (12.32)	6 (7.50)	47 (13)	
		disagree	20 (21.74)	55 (27.09)	18 (22.50)	93 (25)	
	4. I think AI systems make many errors in the treatment protocol	agree	31 (33.70)	134 (66.01)	42 (52.50)	207 (55)	0.001*
		neutral	16 (17.39)	25 (12.32)	21 (26.25)	62 (17)	
		disagree	45 (48.91)	44 (21.67)	17 (21.25)	106 (28)	
	5. Organizations use AI unethically	agree	54 (58.70)	121 (59.61)	44 (55.00)	219 (58)	0.053
		neutral	12 (13.04)	34 (16.75)	23 (28.75)	69 (18)	
		disagree	26 (28.26)	48 (23.65)	13 (16.25)	87 (23)	

Data presented as number (percentage) (n (%)).
EEC – early childhood caries; * statistically significant (χ^2 test).

Assessing the attitude of the participants toward AI

Aggregating the attitude scores for the 5 questions could result in biased interpretation. Therefore, each question was individually scrutinized to enhance comprehension. A considerable number of male faculty members and private practitioners aged between 26 and 55 years demonstrated elevated mean scores and exhibited a favorable disposition toward incorporating AI into the field of pediatric dentistry. They firmly believed that AI had immense prospects, with the difference being statistically significant at $p < 0.05$. In contrast, female postgraduate students and older individuals aged 56–60 years displayed comparatively lower mean scores and held a less optimistic attitude toward the integration of AI. The postgraduates and those with less than 5 years of experience, ranging in age from 46 to 50 years, exhibited low mean scores and expressed uncertainty regarding the superior diagnostic capability of AI as compared to pedodontists. A substantial majority, constituting 66% of the academic faculty, conveyed a belief that AI may generate errors in the treatment protocols. By contrast, 49% of postgraduate students maintained a divergent perspective; the difference was statistically significant ($p = 0.001$). A considerable proportion of female respondents, postgraduates and older individuals between the age of 56 and 60 years had low scores and held the view that AI is prone to committing diagnostic errors, as well as expressed concern over the unethical utilization of AI by certain institutions (Tables 2 and 3).

Mean scores for the knowledge, perception and attitude of the participants

There were statistically significant gender differences in the mean knowledge scores of the participants, with males having higher scores (28.12 ±3.78) than females (26.67 ±3.12) ($p < 0.05$). The faculty members had the greatest mean perception scores (22.40 ±2.98), followed by the postgraduates (21.86 ±3.67) and the private practitioners

Table 3. Mean attitude scores with regard to age, gender, designation, and experience

Variable		A1	A2	A3	A4	A5
Age [years]	26–30	3.828	4.103	3.724	2.586	3.552
	31–35	4.053	4.053	3.105	3.053	2.895
	36–40	4.727	4.727	3.818	4.091	3.545
	41–45	4.769	4.769	4.308	4.308	4.538
	46–50	3.556	4.111	1.556	2.444	2.111
	51–55	4.000	4.000	3.125	3.375	3.250
	56–60	3.000	3.000	3.000	1.400	1.400
Gender	M	4.143	4.500	3.857	3.405	3.548
	F	3.981	3.942	3.019	2.827	3.058
Designation	postgraduate	3.522	3.870	3.391	2.087	3.304
	faculty member	4.176	4.176	3.275	3.569	3.294
	private practitioner	4.350	4.600	3.700	3.000	3.200
Experience [years]	<5	4.783	4.783	3.565	3.522	3.261
	5–10	3.718	3.923	3.641	2.872	3.513
	>10	3.938	4.094	2.969	3.031	3.000

A1–A5 – mean attitude scores for particular questions.

(18.56 ±2.03). The participants with less than 5 years of experience had better mean knowledge scores (27.39 ±3.12), while the participants with 5–10 years of experience had better mean perception scores (23.40 ±2.32). The lowest mean knowledge (22.03 ±2.34) and perception scores (19.56 ±2.45) were noticed in the participants with more than 10 years of experience. Statistically significant age differences were observed in the participants' mean knowledge and perception scores ($p < 0.05$). The participants aged less than 45 years had better scores than the participants aged more than 45 years, as shown in Table 4.

Post-hoc analysis was conducted to perform pairwise comparisons between different age groups. The analysis revealed that there were significant differences in the knowledge scores between age groups 26–30 years and 31–35 years, 36–40 years, 46–50 years, and 56–60 years. Furthermore, significant differences were observed between age groups 41–45 years and 46–50 years as well as 56–60 years. In terms of perception scores, significant differences were found between age groups 31–35 years and 46–50 years, 51–55 years and 56–60 years. Additionally, differences were evident between age groups 36–40 years and 46–50 years, 51–55 years as well as 56–60 years. Lastly, significant differences were noted in the attitude scores between age groups 26–30 years, 31–35 years, 36–40 years, and 41–45 when compared to age group 56–60 years. Pairwise comparisons for designation demonstrated that significant differences were apparent between the postgraduates and the faculty members in relation to the scores for knowledge and attitude. The perception scores of the private practitioners were significantly different from those of the faculty members and postgraduates. Pairwise comparisons in terms of experience indicate the presence of notable disparities among individuals with varying levels of experience. Specifically, when examining the knowledge and perception scores, it was observed that individuals with less than 5 years of experience differed significantly from those with 5–10 years of experience or more than 10 years of experience. Conversely, in relation to the attitude scores, discernible differences were identified between individuals with less than 5 years of experience and those with 5–10 years of experience (Table 5).

Correlation between the knowledge, perception and attitude scores and the demographic variables of the participants

The knowledge, perception and attitude exhibited by the participants showed a negative correlation with the age of the participants, suggesting that older individuals possessed less profound mindset, knowledge and interpretation pertaining to the utilization of AI in the realm of pediatric dentistry as compared to younger individuals. However, there was a weak correlation between age and attitude, which suggests that there might be a tendency for attitude to decrease with an increasing age, but this relationship is not strong enough to be considered significant. There was a positive correlation between knowledge and attitude, and between knowledge and perception. These findings indicate that as pedodontists' comprehension of AI increased, their mindset and interpretation also improved. This is evident from the data presented in Table 6.

Table 4. Mean scores for knowledge, attitude and perception with regard to age, gender, designation, and experience

Variable		*N*	Knowledge score (max = 35)	*p*-value	Perception score (max = 25)	*p*-value	Attitude score (max = 25)	*p*-value
			M ±*SD*		*M* ±*SD*		*M* ±*SD*	
Age [years][b]	26–30	118	23.34 ±2.12	0.029*	19.98 ±3.46	0.048*	17.34 ±1.45	0.081
	31–35	78	27.23 ±3.46		21.28 ±2.45		16.98 ±2.23	
	36–40	42	26.12 ±3.12		22.21 ±2.78		17.78 ±2.12	
	41–45	52	23.34 ±3.02		21.78 ±3.12		14.23 ±1.87	
	46–50	36	25.23 ±2.67		19.12 ±2.65		15.29 ±1.93	
	51–55	28	22.31 ±2.78		17.34 ±3.23		16.23 ±2.34	
	56–60	21	16.23 ±1.98		17.27 ±3.12		15.34 ±1.78	
Gender[a]	M	214	28.12 ±3.78	0.032*	20.34 ±3.89	0.068	18.67 ±3.41	0.068
	F	161	26.67 ±3.12		21.21 ±2.76		19.23 ±2.78	
Designation[b]	postgraduate	92	23.39 ±3.45	0.921	21.86 ±3.67	0.0367*	20.13 ±3.03	0.093
	faculty member	203	27.21 ±2.65		22.40 ±2.98		22.00 ±2.45	
	private practitioner	80	20.03 ±2.43		18.56 ±2.03		19.17 ±3.21	
Experience [years][b]	<5	154	27.39 ±3.12	0.029*	21.86 ±3.15	0.044*	18.13 ±3.03	0.082
	5–10	93	23.21 ±2.78		23.40 ±2.32		22.03 ±2.34	
	>10	128	22.03 ±2.34		19.56 ±2.45		17.15 ±2.11	

M – mean; *SD* – standard deviation; max – maximum; * statistically significant ([a] Mann–Whitney *U* test, [b] Kruskal–Wallis test).

Table 5. Post-hoc analysis with Dunn's test for pairwise comparisons (*p*-values)

Variable	Pairwise comparisons		Knowledge	Perception	Attitude
Age [years]	26–30	31–35	0.000*	0.055	0.854
		36–40	0.000*	0.001*	0.858
		41–45	1.000	0.008*	0.056
		46–50	0.007*	0.753	0.072
		51–55	0.575	0.001*	0.084
		56–60	0.000*	0.004*	0.000*
	31–35	36–40	0.364	0.684	0.300
		41–45	0.000*	0.969	0.072
		46–50	0.007*	0.009*	0.080
		51–55	0.000*	0.000*	0.557
		56–60	0.000*	0.000*	0.009*
	36–40	41–45	0.000*	0.994	0.052
		46–50	0.797	0.000*	0.231
		51–55	0.000*	0.000*	0.066
		56–60	0.000*	0.000*	0.000*
	41–45	46–50	0.030*	0.001*	0.139
		51–55	0.696	0.000*	0.000*
		56–60	0.000*	0.000*	0.269
	46–50	51–55	0.001*	0.236	0.442
		56–60	0.000*	0.289	1.000
	51–55	56–60	0.000*	1.000	0.670
Designation	postgraduate	faculty member	0.042*	0.321	0.036*
		private practitioner	0.063	0.000*	0.081
	faculty member	private practitioner	0.167	0.000*	0.062
Experience [years]	<5	5–10	0.000*	0.000*	0.000*
		>10	0.000*	0.000*	0.062
	5–10	>10	0.005*	0.000*	0.081

* statistically significant.

Table 6. Correlation of the scores for knowledge, perception and attitude with the participants' age

Variable	Correlation parameters	Knowledge score	Perception score	Attitude score	Age
Knowledge score	ρ	–	0.608	0.456	−0.233
	p-value	–	<0.001*	<0.001*	<0.001*
Perception score	ρ	0.608	–	0.585	−0.249
	p-value	<0.001*	–	<0.001*	<0.001*
Attitude score	ρ	0.456	0.585	–	−0.048
	p-value	<0.001*	<0.001*	–	0.355
Age	ρ	−0.233	−0.249	−0.048	–
	p-value	<0.001*	<0.001*	0.355	–

* statistically significant.

Discussion

Artificial intelligence has a vast array of medical applications and has recently experienced a surge of popularity, necessitating the meticulous exploration of its potential implementation in the field of dentistry. However, a considerable number of scientists and medical professionals are not familiar with AI and its potential impact on both their personal and professional lives. To the best of our knowledge, this survey stands out among the existing research because of its distinctive focus on the application of AI in the field of pediatric dentistry.

The results of the present study indicated that 62% of the participants, mostly faculty members, were aware of the existence of AI. Our findings are similar to those of previous studies.[6–8] The reason behind this phenomenon may lie in the fact that AI encompasses a wide range of emerging technologies that have a lasting impact on everyday life. Artificial intelligence enables the analysis of large sets of data, thus providing accurate and dependable information, and ultimately enhancing the process of making informed decisions.[9] The main sources of knowledge on AI were industry news, conferences and online data from both social media and academics. This is in contrast with the findings of a previous study, in which social media were the main source, and not academics.[10]

Regarding the field of pediatric dentistry, AI can be used to identify plaque in children. An AI model demonstrated satisfactory clinical efficacy in the identification of dental plaque on primary teeth in comparison with an experienced pediatric dentist.[11] The utilization of an AI model can potentially offer support to parents in their everyday existence, as it can significantly diminish the challenges associated with identifying dental plaque on their offspring's teeth, thereby aiding in the prevention of dental caries.[4] In our study, 52% of the participants felt that AI could also be used for the identification of supernumerary teeth. The CNN-based deep learning represents a highly promising strategy for identifying supernumerary teeth in the initial stage of mixed dentition.[12]

As many as 68% of the participants felt that AI could detect ECC, as reported in previous studies as well.[13,14] Parents can utilize their customary smartphones to capture the images of their offspring's dentition, which can then be analyzed with the aid of AICaries. This enables parents to proactively pursue the treatment of their children during the initial and remediable phases of ECC. The incorporation of AI has the potential to enhance the diagnostic acumen of dentists, primarily by augmenting their ability to detect enamel lesions with greater sensitivity.[14] In the present study, 67% of the participants agreed that AI could be used for the estimation of chronological age in children. AI-supported standards can offer considerably more precise forecasts of chronological age, with mean errors of less than 0.05 years. Conversely, conventional approaches have yielded inflated outcomes for both genders.[15]

Moreover, 67% of the participants agreed that AI could effectively detect the ectopic eruption of first permanent molars, which was also observed in previous studies,[4,16] and 82% of the participants agreed upon the use of AI in assessing child psychology. There are numerous mechanisms by which AI can assist parents, ranging from the provision of tailored guidance that aligns with the requirements of each family to prompt intervention upon the detection of the preliminary indications of developmental challenges or parental strain.[17]

Most of the participants wanted to use AI software in the future and to attend professional courses to learn about the use of AI in pediatric dentistry. They felt a strong need to include AI training during their postgraduation years and would recommend their colleagues to use AI in their practice. This is in accordance with previous studies.[10,18] These results show the interest of pedodontists in learning new technologies, such as AI, and their willingness to incorporate it into clinical practice. When asked about the barriers related to the use of AI in the field of pediatric dentistry, the majority enumerated the high costs of AI software, the lack of proper training and technical knowledge, the fear of misdiagnosis, and the lack of awareness, as stated in previous studies as well.[5,10] While 82% of the participants felt that AI had a promising future in the field of pediatric dentistry, 55% felt that AI could make errors in diagnosis and treatment planning. Some of them also felt that AI was used unethically by some organizations. Mistakes in the procedure or protocol within the realm of healthcare can potentially yield dire ramifications for the individual undergoing treatment, consequently serving as the unfortunate recipient of said errors. Currently, the absence of clearly established regulations hinders the resolution of legal and ethical dilemmas that could emerge because of the integration of AI within healthcare environments. There is an undeniable requirement for the disclosure of algorithms, safeguarding data privacy and the protection of all parties who benefit from these systems while also ensuring the security of the vulnerabilities linked to cybersecurity.[19]

The present study revealed no significant difference in the attitude scores between individuals with less than 5 years of experience and those with more than 10 years of experience, which suggests that the level of professional experience may not be a strong predictor of attitude toward AI. This might be due to fact that professionals with different levels of experience may possess a relatively homogeneous knowledge base when it comes to AI in pediatric dentistry. If decision-making processes within the field involve input from professionals of varying experience levels, it could contribute to a consensus in attitude toward AI. However, a better understanding of the specific reasons behind the lack of difference in the attitude scores would likely involve a more in-depth examination, including surveys, interviews or focus group discussions with professionals from both experience groups. Although AI has shown promising results in specific diagnostic tasks within dentistry, it is not a substitute for skilled pedodontists' comprehensive care, judgment and experience. The treatment protocols in medicine and dentistry can be highly complex and involve multiple factors, such as patient history, comorbidities and individual responses to treatment. Artificial intelligence systems may not yet be sufficiently sophisticated to fully understand and integrate all these variables as effectively as human practitioners.

Our study also revealed that females demonstrated less knowledge about AI than males. However, they were eager to learn about AI. The faculty members and postgraduates had better knowledge and perception scores than private practitioners. Better scores, particularly the knowledge and perception scores, were reported for individuals aged 26–30 years and with less than 5 years of experience, which represents a younger population, mainly postgraduates, as compared to an older age group of 56–60 years and with more than 5 years of experience. This phenomenon could potentially be attributed to a rise in consciousness, availability and enthusiasm among the younger generation to acquire knowledge about the emerging technologies, such as AI, whereas the older

generations are more afraid of using AI for diagnosis, as they feel that a machine cannot replace or work better than an experienced pedodontist; most contemporary AI applications offer limited information, which falls short of fully explaining the intricate decision-making process in clinical care.[20] Younger individuals, particularly those from generations that have been immersed in technology, are prone to possess a greater level of knowledge and ease with regard to AI concepts. They might have obtained more up-to-date education or training, and may have been exposed to the most recent advancements in AI and its potential applications in pediatric dentistry. Generational perspectives toward technology, innovation and change may influence the way individuals perceive the integration of AI in the field of pediatric dentistry. Differences in the sources of information and exposure to AI-related content through media and educational platforms can also contribute to variations in the knowledge and perception scores across different age groups. Despite this, older individuals demonstrate a strong interest in acquiring knowledge about AI and express a desire to integrate it into clinical practice.

Limitations

As we explored the limitations of the study, it is important to acknowledge the presence of an age bias, as the concepts employed in the survey might be unfamiliar to older individuals. Consequently, this selection bias resulted in a greater representation of younger age groups within the sample. Considering the ease of distributing and accessing the survey via the Google Forms link, coupled with the subject matter's appeal to a younger population, these circumstances may have contributed to the overrepresentation of young individuals within the study sample. The questionnaire was formulated in English, which is the dominant language in the area under investigation. This might have hindered the involvement of individuals who did not possess proficiency in English. The use of the Likert scale in close-ended questions may have hindered the generation of suggestions or concepts for inquiries that require a multitude of perspectives, thereby leading to misinterpretation. The novel scoring system used in the study needs to be tested in a diverse population for generalizability, external validation and reliability assessment.

Recommendations

According to the findings of the current survey, most pedodontists generally exhibit a predominantly positive attitude toward AI technology. A significant proportion of them expressed their willingness to incorporate AI into their professional practice, with numerous individuals indicating their readiness to acquire new technological skills and embrace novel advancements. Nevertheless, the findings also revealed that these professionals harbor concerns regarding the financial and time investments required for the implementation of AI in their practice, as well as the lack of awareness and technical expertise. To address these concerns effectively, it is recommended that AI be seamlessly integrated into academic curricula for undergraduate and postgraduate students. This integration will cater to the specific needs of pedodontists across disciplines. Furthermore, targeted efforts should be made to increase the comfort level of pedodontists in the age range of 45–64 years. To achieve this, it is important to provide training programs, workshops and other resources that are specifically designed to assist users in feeling more comfortable and confident when utilizing AI tools.

Conclusions

The majority of participants exhibited knowledge regarding the advantages of employing AI in the field of pediatric dentistry and expressed confidence in its potential as an advantageous tool. The investigation revealed that the presence of enhanced technical facilities within clinics and the provision of education to professionals at both undergraduate and postgraduate levels could potentially overcome obstacles in utilizing AI within the domain of pediatric dentistry.

Ethics approval and consent to participate

The study was approved by the institutional ethics committee at Yogita Dental College and Hospital, Khed, India (approval No. EC/NEW/INST/2022/2959/Y23/212). All participants provided written informed consent.

Data availability

The datasets supporting the findings of the current study are available from the corresponding author on reasonable request.

Consent for publication

Not applicable.

Use of AI and AI-assisted technologies

Not applicable.

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References

1. McCarthy J, Hayes PJ. Some philosophical problems from the standpoint of artificial intelligence. In: Meltzer B, Michie D, eds. *Machine Intelligence, 4*. Edinburgh, UK: Edinburgh University Press; 1964:463–502.
2. Mahajan K, Kunte SS, Patil KV, Shah PP, Shah RV, Jajoo SS. Artificial intelligence in pediatric dentistry – a systematic review. *J Dent Res Rev*. 2023;10(1):7–12. doi:10.4103/jdrr.jdrr_199_22
3. Agrawal P, Nikhade P. Artificial intelligence in dentistry: Past, present, and future. *Cureus*. 2022;14(7):e27405. doi:10.7759/cureus.27405
4. Vishwanathaiah S, Fageeh HN, Khanagar SB, Maganur PC. Artificial intelligence its uses and application in pediatric dentistry: A review. *Biomedicines*. 2023;11(3):788. doi:10.3390/biomedicines11030788
5. Müller A, Mertens SM, Göstemeyer G, Krois J, Schwendicke F. Barriers and enablers for artificial intelligence in dental diagnostics: A qualitative study. *J Clin Med*. 2021;10(8):1612. doi:10.3390/jcm10081612
6. Murali S, Bagewadi A, Kumar L, et al. Knowledge, attitude, and perception of dentists regarding the role of artificial intelligence and its applications in oral medicine and radiology: A cross sectional study. *J Oral Med Oral Surg*. 2023;29:1–12. doi:10.1051/mbcb/2023018
7. Ranjana V, Gayathri R, Priya VV, Kavitha S. Awareness on application of artificial intelligence in medicine among dental students – a survey. *Ann Rom Soc Cell Biol*. 2021:1137–1153. http://annalsofrscb.ro/index.php/journal/article/view/1568. Accessed June 12, 2023.
8. Asmatahasin M, Pratap KV, Padma TM, Kalyan VS, Kumar VS. Attitude and perception of dental students towards artificial intelligence. *Indian J Basic Appl Med Res*. 2021;10(3):305–314. doi:10.36848/IJBAMR/2020/29215.557818
9. Lee JH, Kim DH, Jeong SN, Choi SH. Diagnosis and prediction of periodontally compromised teeth using a deep learning-based convolutional neural network algorithm. *J Periodontal Implant Sci*. 2018;48(2):114–123. doi:10.5051/jpis.2018.48.2.114
10. Singh N, Pandey A, Tikku AP, Verma P, Singh BP. Attitude, perception and barriers of dental professionals towards artificial intelligence. *J Oral Biol Craniofac Res*. 2023;13(5):584–588. doi:10.1016/j.jobcr.2023.06.006
11. You W, Hao A, Li S, Wang Y, Xia B. Deep learning-based dental plaque detection on primary teeth: A comparison with clinical assessments. *BMC Oral Health*. 2020;20(1):141. doi:10.1186/s12903-020-01114-6
12. Mine Y, Iwamoto Y, Okazaki S, et al. Detecting the presence of supernumerary teeth during the early mixed dentition stage using deep learning algorithms: A pilot study. *Int J Paediatr Dent*. 2022;32(5):678–685. doi:10.1111/ipd.12946
13. Xiao J, Luo J, Ly-Mapes O, et al. Assessing a smartphone app (AICaries) that uses artificial intelligence to detect dental caries in children and provides interactive oral health education: Protocol for a design and usability testing study. *JMIR Res Protoc*. 2021;10(10):e32921. doi:10.2196/32921
14. Hegde S, Gao J. Deep learning algorithms show some potential as an adjunctive tool in caries diagnosis. *J Evid Based Dent Pract*. 2022;22(4):101772. doi:10.1016/j.jebdp.2022.101772
15. Wu TJ, Tsai CL, Gao QZ, Chen YP, Kuo CF, Huang YH. The application of artificial-intelligence-assisted dental age assessment in children with growth delay. *J Pers Med*. 2022;12(7):1158. doi:10.3390/jpm12071158
16. Liu J, Liu Y, Li S, Ying S, Zheng L, Zhao Z. Artificial intelligence-aided detection of ectopic eruption of maxillary first molars based on panoramic radiographs. *J Dent*. 2022;125:104239. doi:10.1016/j.jdent.2022.104239
17. Aylward BS, Abbas H, Taraman S, et al. An introduction to artificial intelligence in developmental and behavioral pediatrics. *J Dev Behav Pediatr*. 2023;44(2):e126–e134. doi:10.1097/DBP.0000000000001149
18. Chen YW, Stanley K, Att W. Artificial intelligence in dentistry: Current applications and future perspectives. *Quintessence Int*. 2020;51(3):248–257. doi:10.3290/j.qi.a43952
19. Naik N, Zeeshan Hameed BM, Shetty DK, et al. Legal and ethical consideration in artificial intelligence in healthcare: Who takes responsibility? *Front Surg*. 2022;9:862322. doi:10.3389/fsurg.2022.862322
20. Schwendicke F, Samek W, Krois J. Artificial intelligence in dentistry: Chances and challenges. *J Dent Res*. 2020;99(7):769–774. doi:10.1177/0022034520915714

Original paper

Evaluation of the use of artificial intelligence in evidence-based endodontology: Bibliometric and scientometric analysis

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Abstract

Background. Artificial intelligence (AI) systems have the potential to revolutionize the fields of medicine and dentistry by identifying solutions for managing multiple clinical problems. This greatly facilitates the tasks of physicians. Bibliometric studies not only provide insight into the history of a particular topic, but also help to determine how the work evolves over time, and to identify interesting new research.

Objectives. The aim of the present study was to identify and analyze bibliographically recent research articles on the use of AI in endodontics.

Material and methods. The search was conducted in March 2024 in the Web of Science Core Collection (WoS-CC), using the Clarivate™ search engine. The search strategy in all fields included in the database was as follows: "endodontics" was the main keyword, and the other keywords were "artificial intelligence", "deep learning", "machine learning", "artificial neural network", and "convolutional neural network". The title, authors, institution, country, impact factor, total number of citations, year of publication, journal name, number of authors, keywords, abstracts, and other topics of interest were recorded. Bibliometric networks were generated and analyzed using the Visualization of Similarities Viewer (VOSviewer).

Results. Of the 54 articles published by the journals indexed in the WoS-CC between 2012 and 2024 that contained the search terms, 40 were included in this study. The article citations ranged from 0 to168, with an average of 18.97. The number of countries contributing to the research was 29. The country with the highest contribution rate in the field was the USA ranked first (27.5 %), followed by Germany (17.5 %), China (15.0%), and India (15.0%).

Conclusions. Based on this review, it can be concluded that a more significant research interest in AI and endodontics was observed in the USA. The most cited research articles dealt with dental image diagnosis with the use of convolutional neural networks (CNN), the radiologic diagnosis of apical lesions using AI, and the computer-aided diagnosis of periapical lesions using AI in computed tomography (CT) analyses.

Keywords: artificial intelligence, endodontics, citation analysis, bibliometrics

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Highlights

- Artificial intelligence (AI) applications in endodontics have grown rapidly since 2019, addressing key challenges, such as diagnosis, treatment outcome prediction and case difficulty assessment.
- Convolutional neural networks showed particular promise for the radiographic detection of apical and periapical lesions, though further improvement in sensitivity is needed for routine clinical use.
- Bibliometric evidence underscores AI as an emerging transformative approach in endodontics, with the capacity to reshape both the trajectory of research and future clinical applications.

Introduction

Planning a research idea, obtaining study data by successfully applying an appropriate method, and analyzing and comparing the data with other studies in the literature are the stages that must be carefully managed in preparing a scientific publication.[1] Research is deemed worthy of publication in a scientific journal if it will attract the attention of researchers, be frequently cited and make a scientifically meaningful contribution to the field under investigation.[2] Bibliometrics assesses academic productivity using quantitative measures, with the aim of analyzing and monitoring the development and structure of science.[3] Bibliometric studies enable the evaluation of the contribution of particular countries to the scientific literature in certain disciplines and topics, as well as the assessment of journals through comparative studies.[4] The number of citations an article receives may not reflect its scientific quality or impact on clinical practice, but it is considered an objective indicator for researchers to track new trends and make evidence-based decisions with regard to technological development in the field.[5,6]

The rationale underlying bibliometric research is that analyzing articles on a specific topic in peer-reviewed journals can provide clinicians and researchers with a historical perspective on the progress of the topic. It can also help to identify where research is most concentrated and how areas of interest have changed over time.[3] The first bibliometric analysis in endodontics addressed trends in endodontic research by examining the top 100 most cited articles.[7]

Artificial intelligence (AI) is a field of applied computer science, first defined by John McCarthy. The development of AI is often referred to as the "fourth industrial revolution" due to its ability to simulate critical thinking, intelligent behavior and human-like decision making based on computer technology.[8,9] Recent interest in AI is due to the development of a new generation of specialized algorithms capable of analyzing and predicting, using large datasets.[10]

Artificial intelligence has found its application in areas of medicine and dentistry, as well as in all kinds of industries.[11,12] Artificial intelligence systems have the potential to revolutionize the fields of medicine and dentistry by identifying solutions for managing multiple clinical problems. This greatly facilitates the tasks of physicians.[8] Today, modern medicine is witnessing a revolution with the application of AI in clinical decision making. Artificial intelligence has been shown to improve efficiency and accuracy in a timely manner at a lower cost, with results comparable to those reached by medical professionals.[13] However, the implementation of AI systems is becoming increasingly complex due to emerging potential risks and ethical challenges that must be considered from a legal standpoint.[14]

The application of AI has not become routine in dental clinical practice.[15] However, pathology detection,[16] caries diagnosis,[17] robotic assistance,[18] and electronic record retrieval[19] with the use of AI have been gaining acceptance in dentistry.

In recent years, there has been a significant increase in the number of papers reporting the application of AI models in endodontics. The determination of root canal working length,[20] the detection of vertical root fractures,[21,22] the success of root canal treatment,[23] the detection of pulp diseases,[24] the detection and diagnosis of periapical lesions,[25] the detection of unfilled canals,[26] the prediction of postoperative pain[27] and case difficulty[28] are the main focus in some of the studies conducted in this field.

Artificial intelligence research in endodontics has grown alongside other dental specialities.[8] In recent years, bibliometric research has been conducted in many fields of dentistry.[7,29,30] As far as we are aware, no bibliometric analysis of AI studies in the field of endodontics has been published. From this perspective, the aim of the present study was to bibliometrically evaluate studies using AI models in the field of endodontics.

Material and methods

A systematic search of the literature was conducted using the Web of Science Core Collection (WoS-CC) online database and the Clarivate™ search engine to identify

relevant research in the field. An electronic search was conducted on March 2024. The search strategy for all fields included in the database applied the main keyword "endodontics" and other keywords: "artificial intelligence"; "deep learning"; "machine learning"; "artificial neural network"; and "convolutional neural network" [((artificial intelligence) OR (deep learning) OR (machine learning) OR (artificial neural network) OR (convolutional neural network)) AND (endodontics)]. A total of 54 studies published between 2012 and March 2024 were identified in the scanning. Articles that were not directly related to the subject, or were related only to disciplines other than endodontics, were excluded. After a thorough review of all articles, 2 investigators excluded 6 proceeding papers, 1 editorial material and 7 irrelevant articles. Thus, a total of 40 articles were included in the study. They were then ranked according to the frequency of citations and were further analyzed.

Each article was thoroughly reviewed, and basic information, including the study design, was recorded. The data was imported into the application using a tab-delimited file tool; this data included the full record and the cited references. The following information was recorded: title; authors; institution; country; impact factor; total number of citations; year of publication; journal name; number of authors; keywords; abstracts; and other topics. The Visualization of Similarities Viewer (VOS Viewer) software, v. 1.6.19 (Centre for Science and Technology Studies, Leiden University, the Netherlands), was used. An algorithm for automatic term data identification was used to map the bibliometric network of the exported data.[31]

With regard to the aim of the present study, the questions to be answered are listed below:

1. What is the distribution of articles written on the topic by years?
2. Which countries are the highest contributors?
3. Which journals publish the highest number of articles in the field?
4. Who are the most cited authors and what are the most cited publications?
5. What is the most common use of AI in endodontics?

Results

A total of 40 articles were included in the study; they were all published in indexed journals (WoS-CC, the Science Citation Index Expanded (SCIE)) between 2012 and 2024. The distribution of the number of publications and citations over the years is shown in Fig. 1.

The countries of all authors (not only first authors) who contributed to the articles are shown in Fig. 2. The highest contribution came from the USA, with 11 articles. The countries of origin of the authors who published the most articles on the topic were the USA (11) ranked first,

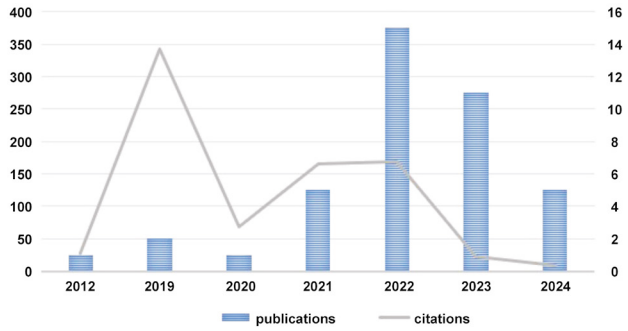

Fig. 1. Distribution of the number of publications and citations over the years 2021–2024

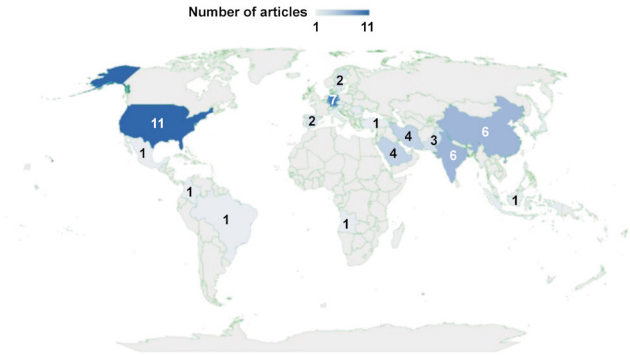

Fig. 2. Contribution to the literature by country of all authors

followed by Germany (7), China (6), and India (6). Other countries with the number of articles published are as follows: Saudi Arabia (4); Iran (4); South Korea (3); Denmark (2); Pakistan (2); and Spain (2); and the UAE, Angola, Austria, Belgium, Brazil, Czech Republic, Colombia, England, Indonesia, Italy, Japan, Mexico, the Netherlands, Romania, Slovakia, Sweden, Thailand, Turkey, and Wales (1). Although a total of 40 articles were included in the study, the number of contributing countries exceeded the number of articles, since some publications had international co-authorship from multiple countries.

A total number of 103 contributing institutions was determined based on the authors' addresses. The analysis of the country of origin using VOSviewer showed that 29 countries contributed to research and publications in the field, both with and without collaboration.

The citation analysis of articles was conducted by selecting countries that had published at least one article and received at least one citation, and 24 countries were observed to have the most connections (Fig. 3). While the countries of origin of the most cited articles were Germany and the USA, the countries of origin of the authors who published the most articles on the topic were the USA, Germany, China, and India. In terms of the total link strength of the countries of origin of the articles on the topic, the USA ranked first, Germany ranked second, followed by China and India. The article citations ranged from 0 to 168, with an average of 18.97. Two articles received over 100 citations.

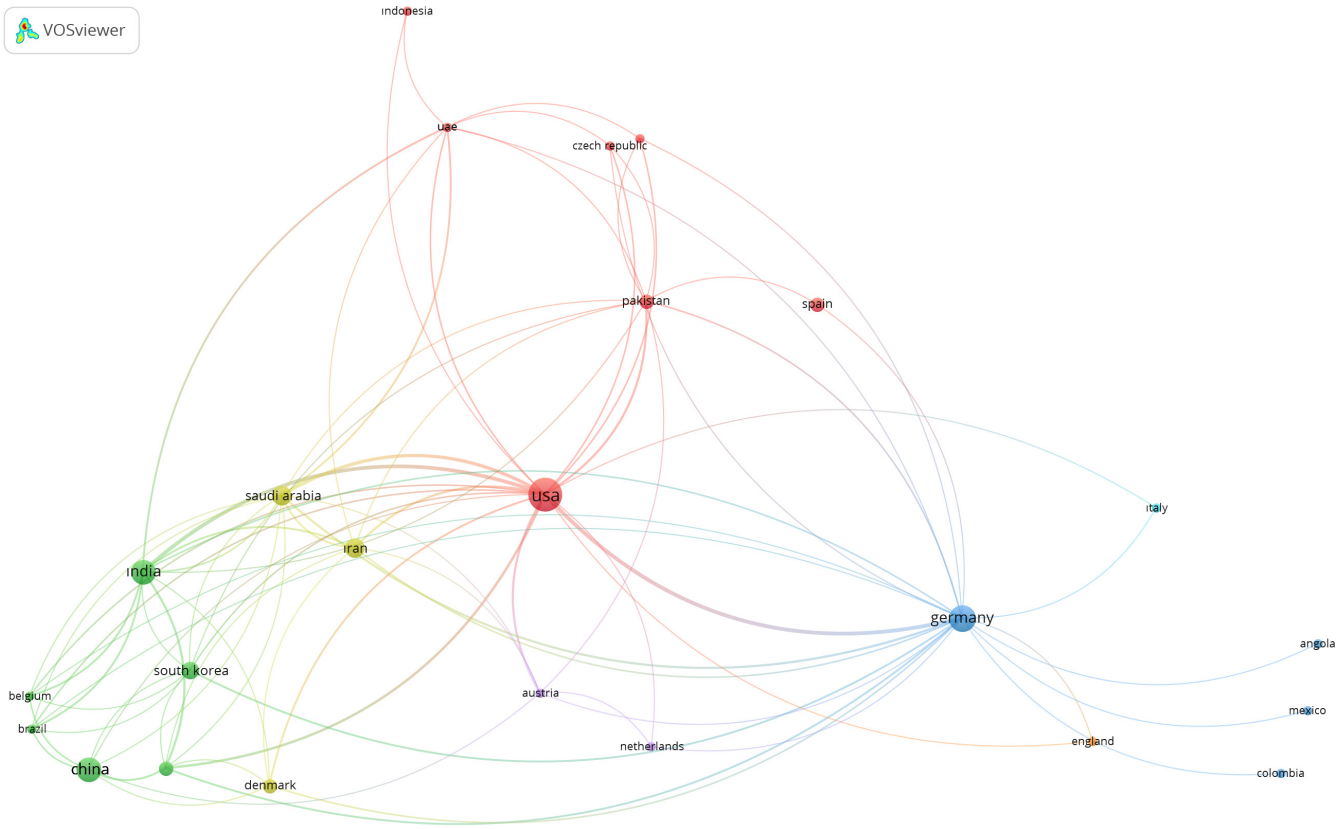

Fig. 3. Citation analysis based on country

The comparison of the total number of citations of the journals related to the subject and the number of publications is shown in Fig. 4. The *Journal of Endodontics* has published the highest number of articles – 14 articles (35%). The *Journal of Dentistry* with 4 articles (10%) followed the *Journal of Endodontics*. The *International Endodontic Journal* took the 3rd place with 3 articles (7.5%).

When examining the first authors of the 3 most cited publications, Schwendicke is ranked first, followed by Ekert in the 2nd place and Setzer in the 3rd place. It was found that 217 authors were involved in articles on the topic of AI in endodontics. Schwendicke was the most cited author with a total of 335 citations in 3 articles. Krois and Golla followed, with a total number of 317 citations, each with 2 articles (Table 1).

Based on the scientometric evaluation, the AI application that received the most focus in endodontics was the detection and segmentation of anatomical structures using AI on dental radiographs, and the diagnosis of periapical lesions in radiographic and tomographic images, followed by the analysis of canal morphology (C-shaped canals and second mesial buccal (MB2) canals).

The 10 most commonly used keywords and their frequency of occurrence in the included articles, as well as their total link strength, are shown in Table 2. The most cited article was published in the *Journal of Dentistry* in 2019, the 2nd most cited article was published in the *Journal of Endodontics* in 2019 and the 3rd most cited article was published in the *Journal of Endodontics* in 2020. The list of publications is presented in Table 1 in order from the most cited to the least cited.

When a bibliometric analysis was performed according to keyword, it was observed that the most frequently used words in studies on AI in endodontics were "artificial intelligence", "endodontics" and "deep learning", followed by "machine learning". In total, 124 unique keywords were identified in the included studies. In Fig. 5, the size of a node represents the frequency of use of the word in the published articles.

Discussion

Bibliometric studies not only provide insight into the history of a particular topic, but also help to determine how the work evolves over time, and to identify interesting new research.[32] Bibliometric studies can evaluate the most cited publications of a journal.[33] Moreover, they can focus on the scientific production of a particular country or field of research.[34] Given this background, the aim of our study was to provide a bibliometric evaluation of research on AI in endodontics. This is a current concept in endodontics and dentistry.

The WoS-CC database was searched following the methodology used in another recent endodontic bibliometric review.[35] The WoS-CC is a popular and suitable database for bibliometric analysis, as it has a large da-

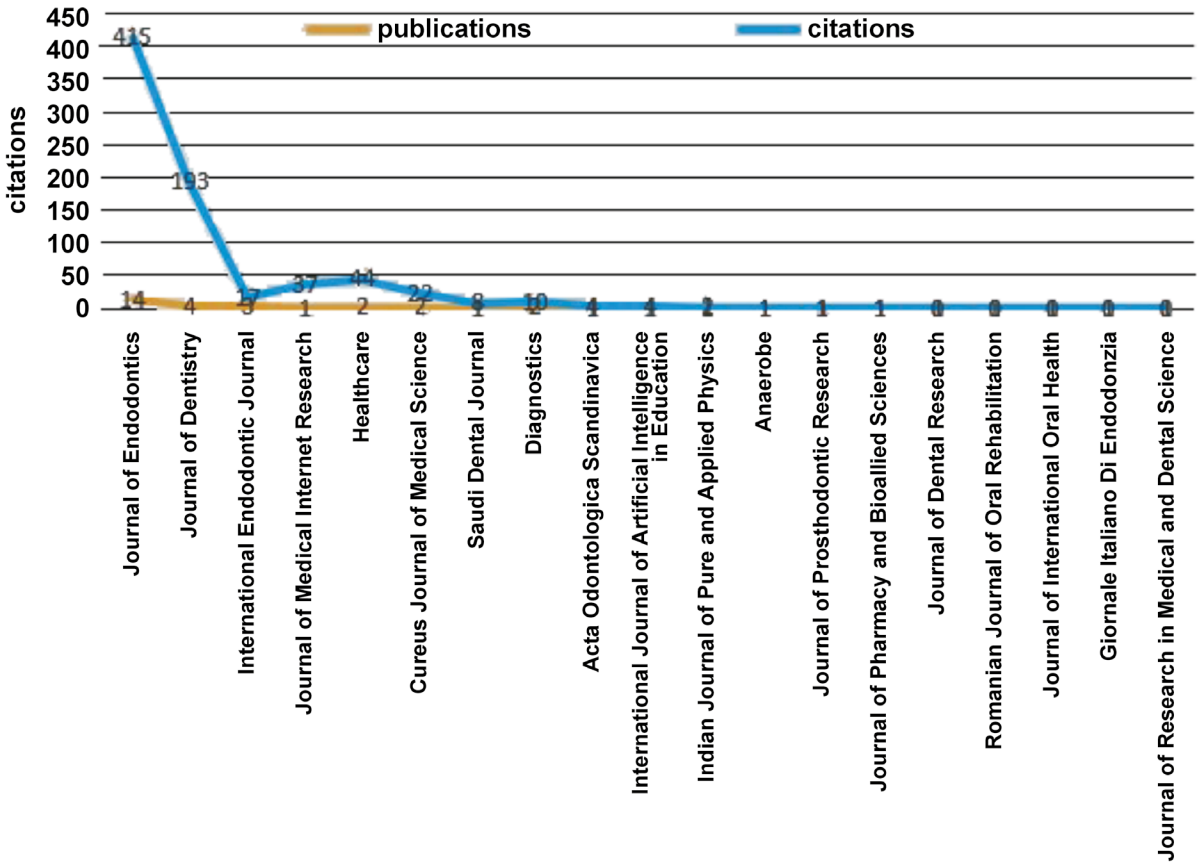

Fig. 4. Comparison of journals in terms of the number of published articles and the total citation value

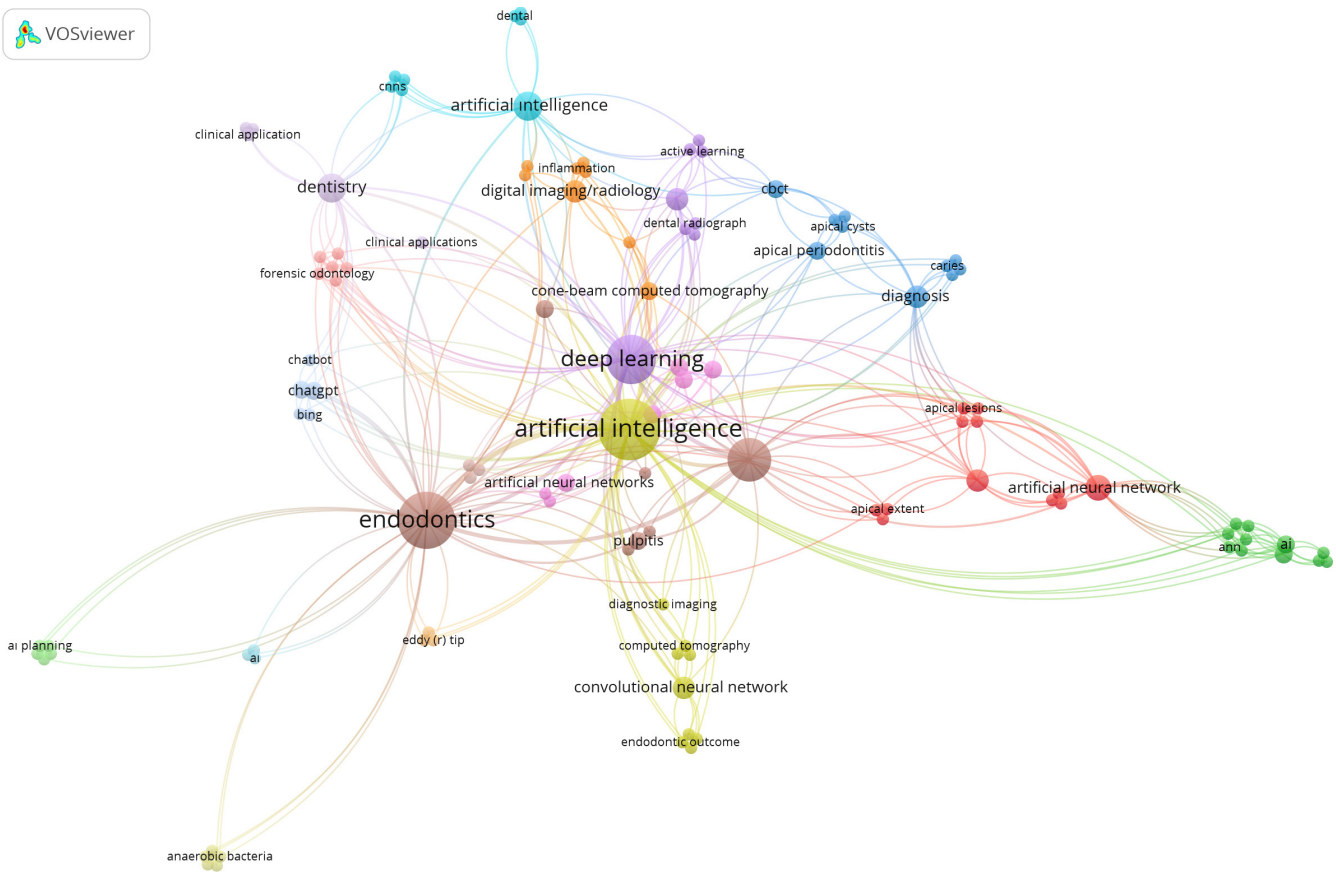

Fig. 5. Research focus according to keyword. Frequency and interaction of the main keywords associated with the study

Table 1. List of publications on artificial intelligence (AI) in endodontics

Authors	Research article	Journal	Institution (corresponding author)	Times cited (WoS-CC)	Times cited (all databases)	Year	Main topic	Conclusion
Schwendicke Golla Dreher Krois	Convolutional neural networks for dental image diagnostics: A scoping review	Journal of Dentistry	Charité – Universitätsmedizin Berlin, Freie Universität Berlin, Humboldt-Universität zu Berlin, and Berlin Institute of Health	168	182	2019	review dental diagnosis with AI	Convolutional neural networks are increasingly used for dental image diagnosis in research settings.
Ekert Krois Meinhold Elhennawy Emara Golla Schwendicke	Deep learning for the radiographic detection of apical lesions	Journal of Endodontics	CODE University of Applied Sciences	149	161	2019	the diagnosis of apical lesions	The convolutional neural network has been satisfactory in differentiating periapical lesions on panoramic radiographs.
Setzer Shi Zhang Yan Yoon Mupparapu Li	Artificial intelligence for the computer-aided detection of periapical lesions in cone-beam computed tomographic images	Journal of Endodontics	University of Pennsylvania	64	68	2020	the diagnosis of periapical lesions	The deep learning algorithm trained in a limited CBCT environment showed excellent lesion detection accuracy and voxel matching accuracy.
Lahoud EzEldeen Beznik Willems Leite Van Gerven Jacobs	Artificial intelligence for fast and accurate 3-dimensional tooth segmentation on cone-beam computed tomography	Journal of Endodontics	University of Leuven	55	55	2021	the diagnosis of tooth segmentation	Successfully performed AI-guided automatic tooth segmentation in CBCT imaging.
Li Ye H Ye F Liu Lv Zhang P Zhang X Zhou	The current situation and future prospects of simulators in dental education	Journal of Medical Internet Research	Peking University School and Hospital of Stomatology	37	42	2021	review the use of AI in dental education	The review was to provide an overview of current dental simulators on related technologies, advantages and disadvantages, the methods of evaluating effectiveness, and future directions for development.
Aminoshariae Kulild Nagendrababu	Artificial intelligence in endodontics: Current applications and future directions	Journal of Endodontics	University of Missouri	33	35	2021	review current applications and future directions	AI has demonstrated accuracy and precision in terms of detection, identification and disease prediction in endodontics.
Thurzo Urbanová Novák Czako Siebert Stano Mareková Fountoulaki Kosnáčová Varga	Where is the artificial intelligence applied in dentistry? Systematic review and literature analysis	Healthcare (Basel)	Comenius University in Bratislava	33	33	2022	systematic review AI applied in dentistry	The research confirms that the current use of AI in dentistry is concentrated mainly around the evaluation of digital diagnostic methods, especially radiology, while it is gradually penetrating all parts of the dental profession.
Saghiri Garcia-Godoy Gutmann Lotfi Asgar	The reliability of artificial neural network in locating minor apical foramen: A cadaver study	Journal of Endodontics	Azad University	28	29	2012	the evaluation of working length determination	The artificial neural network is an accurate method for determining the working length.

Authors	Research article	Journal	Institution (corresponding author)	Times cited (WoS-CC)	Times cited (all databases)	Year	Main topic	Conclusion
Li S Liu Zhou Zirui Zhou Zilin Wu Li Y Wang Liao Ying Zhao	Artificial intelligence for caries and periapical periodontitis detection	Journal of Dentistry	Sichuan University	25	25	2022	the diagnosis of periapical periodontitis and caries	Deep learning can improve accuracy and consistency in the assessment of dental caries and periapical periodontitis on periapical radiographs.
Umer Habib	Critical analysis of artificial intelligence in endodontics: A scoping review	Journal of Endodontics	Aga Khan University Hospital	24	25	2022	scoping review the analysis of AI	AI models had acceptable performance, i.e., more than 90% accuracy, in various diagnostic tasks.
Agrawal Nikhade	Artificial intelligence in dentistry: Past, present, and future	Cureus Journal of Medical Science	Datta Meghe Institute of Medical Sciences University	21	23	2022	review the past, present and future of AI	AI can help advance endodontic diagnosis and treatment, which in turn can improve endodontic treatment outcomes.
Sherwood AA Sherwood AI Setzer Sheela Devi K Shamili John Schwendicke	A deep learning approach to segment and classify C-shaped canal morphologies in mandibular second molars using cone-beam computed tomography	Journal of Endodontics	University of Pennsylvania	18	19	2021	the diagnosis and classification of C-shaped canal morphology in mandibular second molars	Deep learning can help detect and classify C-shaped canal anatomy.
Lin Fu Ren Yang Duan Chen Zhang	Micro-computed tomography-guided artificial intelligence for pulp cavity and tooth segmentation on cone-beam computed tomography	Journal of Endodontics	Tongji University	12	14	2021	guided AI for pulp cavity and tooth segmentation	AI has been shown to provide an accurate and automated approach for tooth and pulp cavity segmentation on CBCT images, applicable in research and clinical tasks.
Fatima Shafi Afzal Díez Lourdes Breñosa Martínez Espinosa Ashraf	Advancements in dentistry with artificial intelligence: Current clinical applications and future perspectives	Healthcare (Basel)	Yeungnam University	11	11	2022	systematic review the current clinical applications of AI	AI has the potential to improve dental care, disease diagnosis and prognosis, treatment planning, and risk assessment.
Sadr Mohammad-Rahimi Motamedian Zahedrozegar Motie Vinayahalingam Dianat Nosrat	Deep learning for detection of periapical radiolucent lesions: A systematic review and meta-analysis of diagnostic test accuracy	Journal of Endodontics	University of Maryland	11	11	2023	systematic review deep learning models in detecting periapical lesions	Deep learning showed highly accurate results in detecting periapical radiolucent lesions on dental radiographs.
Cotti Schirru	Present status and future directions: Imaging techniques for the detection of periapical lesions	International Endodontic Journal	University of Cagliari	10	11	2022	review the detection of periapical lesions	All imaging techniques should be associated with a thorough clinical examination and good calibration of the operator.

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Kirnbauer Hadzic Jakse Bischof Stern	Automatic detection of periapical osteolytic lesions on cone-beam computed tomography using deep convolutional neuronal networks	Journal of Endodontics	Medical University of Graz	10	10	2022	the diagnosis of periapical osteolytic lesions	Although periapical lesions vary in appearance, size and shape in the CBCT dataset, and there is a high imbalance between teeth with and without periapical lesions, the proposed fully automated method provided excellent results as compared to the relevant literature.
Yang Lee Jang Kim K Kim J Kim H Park	Development and validation of a visually explainable deep learning model for classification of C-shaped canals of the mandibular second molars in periapical and panoramic dental radiographs	Journal of Endodontics	Yonsei University	10	10	2022	the diagnosis of C-shaped canals	A deep learning system may be expected to effectively diagnose C-shaped canals and aid clinicians in practice and education.
Asiri Altuwalah	The role of neural artificial intelligence for diagnosis and treatment planning in endodontics: A qualitative review	Saudi Dental Journal	Majmaah University	8	8	2022	qualitative review endodontic treatment planning and diagnosis with AI	AI with different models or frameworks and algorithms can help dentists diagnose and manage endodontic problems more accurately.
Suárez Díaz-Flores García Algar Sanchez de Pedro Freire	Unveiling the ChatGPT phenomenon: Evaluating the consistency and accuracy of endodontic question answers	International Endodontic Journal	Universidad of Europea de Madrid	7	7	2024	the evaluationof the accuracy and consistency of ChatGPT in endodontic clinical questions	ChatGPT is not capable of replacing dentists in clinical decision making.
Albitar Zhao Huang Mahdian	Artificial intelligence (AI) for detection and localization of unobturated second mesial buccal (MB2) canals in cone-beam computed tomography (CBCT)	Diagnostics (Basel)	Stony Brook University	6	6	2022	the detection and localization of unobturated second mesial buccal (MB2) canals	The current AI algorithm has the potential to identify obturated and unobturated canals in endodontically treated teeth.
Vannaprathip Haddawy Schultheis Suebnukarn	Intelligent tutoring for surgical decision making: A planning-based approach	International Journal of Artificial Intelligence in Education	Mahidol University	4	4	2022	intelligent tutoring for surgical decision making	The experts evaluating the results of AI in surgical intervention decision making (SDMentor) show that it can only predict which interventions come from SDMentor with 15% accuracy, as compared to a randomized initial accuracy of 9%.
Khanagar Alfadley Alfouzan Awawdeh Alaqla Jamleh	Developments and performance of artificial intelligence models designed for application in endodontics: A systematic review	Diagnostics (Basel)	King Saud Bin Abdulaziz University for Helath Sciences	4	4	2023	the design of AI models for endodontics	The models can be used as supplementary tools in clinical practice to expedite the clinical decision-making process.
Ramezanzade Laurentiu Bakhshandah Ibragimov Kvist Bjørndal	The efficiency of artificial intelligence methods for finding radiographic features in different endodontic treatments – a systematic review	Acta Odontologica Scandivanica	University of Copenhagen	4	4	2023	systematic review finding radiographic features in different kinds of endodontic treatment	AI-based models have shown effectiveness in finding radiographic features in different kinds of endodontic treatment.

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Thakur Kankar Parey Jain A Jain P	Prediction of apical extent using ensemble machine learning technique in the root canal through biomechanical preparation: In-vitro study	Indian Journal of Pure and Applied Physics	Indian Institute of Technology Indore	2	2	2022	the prediction of the apical extent, using a machine learning model	Machine learning approaches can improve the treatment practice and root canal treatment results, and provide a suitable decision support system.
Kumar Ravindranath Srilatha Alobaoid Kulkarni Mathew Tiwari	Analysis of advances in research trends in robotic and digital dentistry: An original research	Journal of Pharmacy and Bioallied Sciences	Gitam Dental Collage and Hospital	1	1	2022	questionnaire study the evaluation of the awareness and application of these technologies by clinicians	The awareness of these advanced technologies and routine practices is low. These technologies show higher sensitivity and accuracy.
Ahlat Aydin Kaya Baysallar	Identification of root canal microbiota profiles of periapical tissue diseases using matrix-assisted laser desorption/ionization time-of-flight mass spectrometer	Anaerobe	University of Health Sciences	1	1	2023	the classification of periapical tissue microorganisms by machine learning models	MALDI-TOF MS can be considered a fast and high-throughput screening technique for microbial species identification in endodontics.
Alzaid Ghulam Albani Alharbi Othman Taher Albaradie Ahmed	Revolutionizing dental care: A comprehensive review of artificial intelligence applications among various dental specialties	Cureus Journal of Medical Science	Riyadh Elm University	1	1	2023	the use of AI in various specialties of modern dentistry	Dental practice also involves administering treatment to patients. While AI cannot replace dentists, a comprehensive understanding of AI concepts and techniques will be advantageous in the future.
Li Inamochi Wang Fueki	Clinical application of robots in dentistry: A scoping review	Journal of Prosthodontic Research	Tokyo Medical and Dental University (TMDU)	1	1	2024	scoping review robots in dentistry	It was revealed that there are still limitations and gaps between research and the application of dental robots.
Huang Farpour Yang Mupparapu Lure Li Yan Setzer	Uncertainty-based active learning by Bayesian U-Net for multi-label cone-beam CT segmentation	Journal of Endodontics	University of Pennsylvania	1	1	2024	the efficacy of active learning strategies while training AI models, using a limited dataset	Active learning may contribute to reducing extensive labeling needs for training AI algorithms for biomedical image analysis in dentistry.
Peeters Silitonga Zuhal	Application of artificial intelligence in a visual-based fluid motion estimator surrounding a vibrating EDDY® tip	Giornale Italiano Di Endodonzia	Laser Research Center in Dentistry	0	0	2022	the AI visualization of sonic activation (Eddy)	The proposed motion estimation program supported by LiteFlowNet can perform detailed flow estimation of a non-PIV experiment.
Kawale Choudhari Sedani	Artificial intelligence: A boon to conservative dentistry	Journal of Research in Medical and Dental Science	Datta Meghe Institute of Medical Sciences (Deemed to be University)	0	0	2022	review AI in endodontics and conservative dentistry	AI is being studied for a range of applications, including the detection of normal and different structures, disease diagnosis and treatment prediction.
Lee J Seo Choi Lee C Kim S Lee Y Lee S Kim E	An endodontic forecasting model based on the analysis of preoperative dental radiographs: A pilot study on an endodontic predictive deep neural network	Journal of Endodontics	Yonsei University	0	0	2023	the prediction of the endodontic outcome with AI	Deep convolutional neural networks can accurately detect various clinical features on periapical radiographs.

Authors	Research article	Journal	Institution (corresponding author)	Times cited (WoS-CC)	Times cited (all databases)	Year	Main topic	Conclusion
Qu Wen Chen Guo Huang Gu	Predicting case difficulty in endodontic microsurgery using machine learning algorithms	Journal of Dentistry	Sun Yat-sen University	0	0	2023	the prediction of case difficulty in endodontic microsurgery	The relative feature importance provides a reference for developing a scoring system for case difficulty assessment in endodontic microsurgery.
Ramezanzade Dascalu Ibragimov Bakhshandeh Bjørndal	Prediction of pulp exposure before caries excavation using artificial intelligence: Deep learning-based image data versus standard dental radiographs	Journal of Dentistry	University of Copenhagen	0	0	2023	the investigation of the impact of providing dental students with AI-based radiographic information on their ability to predict pulp exposure	Although the AI model outperformed all groups, the participants only benefited 'slightly' from the AI predictions. While AI technology shows promise, it is important to provide more explainable AI predictions and a learning curve.
Pop Choudhary Porumb Avram Badea I Picos Badea A Jiman Muntean	The future of endodontics. A systematic review	Romanian Journal of Oral Rehabilitation	Iuliu Haţieganu University of Medicine and Pharmacy	0	0	2023	review current trends in endodontics based on the latest scientific literature	Overall, endodontic treatment is becoming more precise, leading to a reduction in complications. In conclusion, the field of endodontics is expected to experience numerous exciting advances in the coming years, some of which will be incremental, while others will be paradigm-shifting.
Sudeep Gehlot Murali Mariswamy	Artificial intelligence in endodontics: A narrative review	Journal of International Oral Health	JSS Dental College and Hospital JSS Academy of Higher Education and Research (JSSAHER)	0	0	2023	review AI in endodontics	Being a potential game changer and the beginning of something dubbed the "fourth industrial revolution", AI has what it takes to revolutionize endodontics over time.
Mohammad-Rahimi Ourang Pourhoseingholi Dianat Howell Dummer Nosrat	Validity and reliability of artificial intelligence chatbots as public sources of information on endodontics	International Endodontic Journal	University of Maryland	0	0	2024	the comparison of the validity of responses between chatbots	In comparison with Google Bard and Bing, GPT-3.5 provided more credible information on topics related to endodontics.
Mohammad-Rahimi Dianat Abbasi Zahedrozegar Ashkan Motamedian Rohban Nosrat	Artificial intelligence for detection of external cervical resorption using label-efficient self-supervised learning method	Journal of Endodontics	University of Maryland	0	0	2024	the evaluation of radiographic images by endodontists or 7 basic deep learning models as well as 9 contrast-enhanced self-supervised learning models	AI can aid clinicians in detecting early caries lesions and distinguishing them from other lesions. Furthermore, self-supervised learning has been introduced as a means of detecting early caries lesions.
Fu Zhu Li Wang Deng Chen Shen Meng Bian	Clinically oriented CBCT periapical lesion evaluation via 3D CNN algorithm	Journal of Dental Research	Wuhan University	0	0	2024	the detection and segmentation of periapical lesions associated with apical periodontitis on CBCT images, called PAL-Net	PAL-Net can enhance dentists' diagnostic performance and speed when working with CBCT images, provide clinically relevant volume information, and potentially be used in dental clinics without the need for expert-level dentists or radiologists.

WoS-CC – Web of Science Core Collection; AI – artificial intelligence; CBCT – cone-beam computed tomography; MALDI-TOF MS – matrix-assisted laser desorption/ionization time-of-flight mass spectrometer; PIV – particle image velocimetry.

Table 2. Article keywords with the frequency of occurrence and their total link strength

Serial No.	Keyword	Frequency	Total link strength
1	artificial intelligence	22	103
2	endodontics	19	84
3	deep learning	14	74
4	machine learning	11	50
5	dentistry	5	25
6	artificial neural network	4	23
7	diagnosis	3	18
8	root canal treatment	3	17
9	periapical lesion	3	16
10	convolutional neural network	3	15

tabase of publications dating back to 1945.[36] However, other similar scientometric or bibliometric studies in the field have used other databases, such as Scopus or MEDLINE/PubMed®, to obtain data.[34,37] After comparing the available evidence in this area, it was decided to conduct an individual search in a single database (WoS-CC) for the current study. Although the use of more than one database for search may have made more studies available, at this stage, there could have been duplicate publications needing to be removed manually. The presence of duplicate records would alter all the quantitative bibliometric parameters, potentially leading to the misinterpretation of the relationships between different research components. Thus, only one database was included.

Although it has been claimed that bibliometric analysis should be carried out on topics that have been studied for many years, it is confirmed that the articles published and cited in journals tend to be current topics.[33,38] Artificial intelligence in endodontics is a hot topic. Therefore, in this study, the authors do not consider the fact that the research was conducted on a current issue as a limitation.

The combined data analysis with the use of the bibliometric and network tools of WoS-CC provided many results referring to endodontics and AI. Publications on AI in endodontics continued to increase from 2021 to 2022, peaking in 2022, and decreasing slightly in 2023. This could be an indication that AI is becoming more popular as technology advances, while the topic is becoming more interesting and worthy of discussion through research publication.

The analysis of country of origin and the research institution was conducted using VOSviewer; it showed that 29 countries contributed to research and publications in this field, either collaboratively or non-collaboratively. The country that contributed the most in the field of AI and endodontics was the USA, which can be attributed to the fact that this country has experienced an increase in the number of researchers and working groups in the field of endodontics, and it has followed the technological development.

It is also important to identify the most influential journals for the publication and dissemination of AI and endodontics research. This can help researchers to follow specific journals and aim to publish their research in sources that show specificity in a scientific field.[37] Comparing the number of articles n AI published in scientific journals, the *Journal of Endodontics* had the highest number of publications in the categories of "Dentistry" and "Endodontics", with 14 articles. As a result of the analysis, the *Journal of Endodontics* was also found to be the journal with the highest total number of citations of articles related to AI in the field of endodontics. One of the reasons for this may be that this journal has published the highest number of articles on the topic and that the journal closely follows current topics in the field of endodontics, and finds them worthy of publication. Additionally, as the journal is in the Q1 quartile of the "Dentistry, Medicine and Oral Surgery" category, its citation rate is high and very popular. The fact that the journal follows all advances in endodontics and that it is indexed in SCIE may be attractive factors for researchers when choosing a journal. The *Journal of Dentistry* followed the *Journal of Endodontics*, with 4 articles. The journal *International Endodontic Journal* took the 3rd place, with 3 articles. An important observation about journals is that, although AI is mostly about diagnostics and radiology, diagnostic and radiological reviews in endodontics are more likely to be published in endodontic journals rather than in general dentistry or radiology journals.

The most cited articles on AI in endodontics play a crucial role in the field. Their knowledge draws public attention to researchers who have influenced the growth and development of the work.[7] According to the analysis conducted in our study, Schwendicke was the most cited author on the topic. In a 2019 review, Schwendicke et al. reported studies on the detection and segmentation of anatomical structures in different areas of dentistry using AI on dental radiographs.[39] The 2nd most cited article was that of Ekert et al., who detected apical lesions on radiographs using AI, and found that AI gave satisfactory results in diagnosing apical lesions on panoramic radiographs.[40] In the 3rd most cited article, Setzer et al. used AI to detect apical lesions on cone-beam computed tomography (CBCT) images and reported that, in the long term, the addition of automated image analysis based on three-dimensional (3D) imaging could assist clinicians in lesion detection and the differential diagnosis of periapical lesions (pathological and/or non-odontogenic lesions) in combination with previous examinations.[41] In this context, it can be concluded that the most cited articles have paved the way for dental diagnosis, the detection of dental pathology, and even differential diagnosis using AI in endodontics. Dental diagnosis using AI may also help to reduce the time spent by clinicians on making a diagnosis.

Although the most cited article on AI is a review article analyzing general dentistry fields, including endodontics,

the 2nd and 3rd most cited articles are about the use of AI in the diagnosis of apical lesions in panoramic radiography and on CBCT images, published in a journal in the field of endodontics. We can say that the number of citations of publications on AI in the field of endodontics competes with publications on the use of AI in dentistry.

The keyword co-occurrence analysis can be an insight into the development patterns of a particular area of scientific research.[35] As shown in Table 2, the first 10 keywords were often repeated, and the most often repeated keyword among them was "artificial intelligence". This was followed by the keywords "endodontics" and "deep learning". However, it was also observed that keywords such as "machine learning", "artificial neural network" and "convolutional neural network" were used as well. Artificial intelligence is a general term that comprises more specific topics, including deep learning, machine learning and artificial neural networks. The results of our keyword analysis show that studies using the concept of AI have begun to specialize in the branches of AI, leaving the general concept behind over time.

One limitation could be a small number of articles included in our study. This was due to the fact that the concept of AI is a current topic, and studies using AI have recently been undertaken in the field of endodontics, as in almost all fields.

Conclusions

The use of AI in dental research has steadily increased since the first applications were reported in 2012, but the "explosion" of research occurred in 2019.[39] Indeed, the number of papers reporting on AI models applied in endodontics has raised significantly in recent years. The determination of root canal working length,[20] the detection of vertical root fractures,[21,22] the success of root canal treatment,[23] the detection of pulp diseases,[24] the detection and diagnosis of periapical lesions,[25] the detection of unfilled canals,[26] the prediction of postoperative pain[27] and case difficulty[28] are some of the issues covered by the studies conducted in this field. In the years that followed, the use of AI in endodontics expanded. Studies were conducted on its use in preclinical education.[42] To date, AI in endodontics has had many applications, which we have analyzed bibliometrically. For example, the use of a conventional neural network to detect apical lesions based on panoramic radiographs can help dentists in their diagnostic efforts. However, for such approaches to be widely used in clinical settings, their sensitivity needs to be improved via further studies.

The present bibliometric analysis reviewed current trends, as well as leading countries and journals in terms of research focusing on the use of AI applications in endodontics. Our study considered publications related to AI in endodontics to highlight the bibliometric characteristics of a specific topic. Further research could focus on AI in endodontics in a broader context.

Ethics approval and consent to participate

Not applicable.

Data availability

The datasets supporting the findings of the current study are available from the corresponding author on reasonable request.

Consent for publication

Not applicable.

Use of AI and AI-assisted technologies

Not applicable.

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References

1. Ahlstrom D. How to publish in academic journals: Writing a strong and organized introduction section. *J East Eur Cent Asian Res*. 2017;4(2):9. doi:10.15549/jeecar.v4i2.180
2. Chatterjee A, Ghosh A, Chakrabarti BK. Universality of citation distributions for academic institutions and journals. *PLoS One*. 2016;11(1):e0146762. doi:10.1371/journal.pone.0146762
3. Diane Cooper I. Bibliometrics basics. *J Med Libr Assoc*. 2015;103(4):217–218. doi:10.3163/1536-5050.103.4.013
4. Garcovich D, Marques Martinez L, Adobes Martin M. Citation classics in paediatric dentistry: A bibliometric study on the 100 most-cited articles. *Eur Arch Paediatr Dent*. 2020;21(2):249–261. doi:10.1007/s40368-019-00483-z
5. Aksoy U, Küçük M, Versiani MA, Orhan K. Publication trends in micro-CT endodontic research: A bibliometric analysis over a 25-year period. *Int Endod J*. 2021;54(3):343–353. doi:10.1111/iej.13433
6. Mishra L, Kim HC, Singh NR, Rath PP. The top 10 most-cited articles on the management of fractured instruments: A bibliometric analysis. *Restor Dent Endod*. 2019;44(1):e2. doi:10.5395/rde.2019.44.e2
7. Fardi A, Kodonas K, Gogos C, Economides N. Top-cited articles in endodontic journals. *J Endod*. 2011;37(9):1183–1190. doi:10.1016/j.joen.2011.05.037
8. Aminoshariae A, Kulild J, Nagendrababu V. Artificial intelligence in endodontics: Current applications and future directions. *J Endod*. 2021;47(9):1352–1357. doi:10.1016/j.joen.2021.06.003
9. Peeters HH, Silitonga F, Zuhal L. Application of artificial intelligence in a visual-based fluid motion estimator surrounding a vibrating EDDY® tip. *G Ital Endod*. 2022;36(1):151–159. doi:10.32067/GIE.2021.35.02.50
10. Umer F, Habib S. Critical analysis of artificial intelligence in endodontics: A scoping review. *J Endod*. 2022;48(2):152–160. doi:10.1016/j.joen.2021.11.007
11. Yüce F, Taşsöker M. The applications of artificial intelligence in dentistry [in Turkish]. *Yeditepe Dent J*. 2023;19(2):141–149. doi:10.5505/yeditepe.2023.05668
12. Orhan H, Yavşan E. Artificial intelligence-assisted detection model for melanoma diagnosis using deep learning techniques. *Math Model Numer Simul Appl*. 2023;3(2):159–169. doi:10.53391/mmnsa.1311943

13. Murphy M, Killen C, Burnham R, Sarvari F, Wu K, Brown N. Artificial intelligence accurately identifies total hip arthroplasty implants: A tool for revision surgery. *Hip Int*. 2022;32(6):766–770. doi:10.1177/1120700020987526
14. Nebeker C, Torous J, Bartlett Ellis RJ. Building the case for actionable ethics in digital health research supported by artificial intelligence. *BMC Med*. 2019;17(1):137. doi:10.1186/s12916-019-1377-7
15. Schwendicke F, Samek W, Krois J. Artificial intelligence in dentistry: Chances and challenges. *J Dent Res*. 2020;99(7):769–774. doi:10.1177/0022034520915714
16. Ilhan B, Lin K, Guneri P, Wilder-Smith P. Improving oral cancer outcomes with imaging and artificial intelligence. *J Dent Res*. 2020;99(3):241–248. doi:10.1177/0022034520902128
17. Lian L, Zhu T, Zhu F, Zhu H. Deep learning for caries detection and classification. *Diagnostics (Basel)*. 2021;11(9):1672. doi:10.3390/diagnostics11091672
18. Grischke J, Johannsmeier L, Eich L, Griga L, Haddadin S. Dentronics: Towards robotics and artificial intelligence in dentistry. *Dent Mater*. 2020;36(6):765–778. doi:10.1016/j.dental.2020.03.021
19. Mahmood H, Shaban M, Indave BI, Santos-Silva AR, Rajpoot N, Khurram SA. Use of artificial intelligence in diagnosis of head and neck precancerous and cancerous lesions: A systematic review. *Oral Oncol*. 2020;110:104885. doi:10.1016/j.oraloncology.2020.104885
20. Saghiri MA, Asgar K, Boukani KK, et al. A new approach for locating the minor apical foramen using an artificial neural network. *Int Endod J*. 2012;45(3):257–265. doi:10.1111/j.1365-2591.2011.01970.x
21. Fukuda M, Inamoto K, Shibata N, et al. Evaluation of an artificial intelligence system for detecting vertical root fracture on panoramic radiography. *Oral Radiol*. 2020;36(4):337–343. doi:10.1007/s11282-019-00409-x
22. Kapralos V, Koutroulis A, Irinakis E, et al. Digital subtraction radiography in detection of vertical root fractures: Accuracy evaluation for root canal filling, fracture orientation and width variables. An ex-vivo study. *Clin Oral Investig*. 2020;24(10):3671–3681. doi:10.1007/s00784-020-03245-0
23. Lee J, Seo H, Choi YJ, et al. An endodontic forecasting model based on the analysis of preoperative dental radiographs: A pilot study on an endodontic predictive deep neural network. *J Endod*. 2023;49(6):710–719. doi:10.1016/j.joen.2023.03.015
24. Tumbelaka BY, Oscandar F, Baihaki FN, Sitam S, Rukmo M. Identification of pulpitis at dental X-ray periapical radiography based on edge detection, texture description and artificial neural networks. *Saudi Endod J*. 2014;4(3):115–121. doi:10.4103/1658-5984.138139
25. Cotti E, Schirru E. Present status and future directions: Imaging techniques for the detection of periapical lesions. *Int Endod J*. 2022;55(Suppl 4):1085–1099. doi:10.1111/iej.13828
26. Albitar L, Zhao T, Huang C, Mahdian M. Artificial intelligence (AI) for detection and localization of unobturated second mesial buccal (MB2) canals in cone-beam computed tomography (CBCT). *Diagnostics (Basel)*. 2022;12(12):3214. doi:10.3390/diagnostics12123214
27. Gao X, Xin X, Li Z, Zhang W. Predicting postoperative pain following root canal treatment by using artificial neural network evaluation. *Sci Rep*. 2021;11(1):17243. doi:10.1038/s41598-021-96777-8
28. Qu Y, Wen Y, Chen M, Guo K, Huang X, Gu L. Predicting case difficulty in endodontic microsurgery using machine learning algorithms. *J Dent*. 2023;133:104522. doi:10.1016/j.jdent.2023.104522
29. Feijoo JF, Limeres J, Fernández-Varela M, Ramos I, Diz P. The 100 most cited articles in dentistry. *Clin Oral Investig*. 2014;18(3):699–706. doi:10.1007/s00784-013-1017-0
30. Hui J, Han Z, Geng G, Yan W, Shao P. The 100 top-cited articles in orthodontics from 1975 to 2011. *Angle Orthod*. 2013;83(3):491–499. doi:10.2319/040512-284.1
31. Visualization of Similarities Viewer (VOSviewer). https://www.vosviewer.com.
32. Barboza-Palomino M, Salas G, Vega-Arce M, et al. Thirty years of Psicothema: A bibliometric analysis (1989–2018). *Psicothema*. 2020;32(4):459–468. doi:10.7334/psicothema2020.145
33. Ahmad P, Dummer PM, Noorani TY, Asif JA. The top 50 most-cited articles published in the *International Endodontic Journal*. *Int Endod J*. 2019;52(6):803–818. doi:10.1111/iej.13083
34. Shamszadeh S, Asgary S, Nosrat A. Regenerative endodontics: A scientometric and bibliometric analysis. *J Endod*. 2019;45(3):272–280. doi:10.1016/j.joen.2018.11.010
35. Guerrero-Gironés J, Forner L, Sanz JL, et al. Scientific production on silicate-based endodontic materials: Evolution and current state: A bibliometric analysis. *Clin Oral Investig*. 2022;26(9):5611–5624. doi:10.1007/s00784-022-04605-8
36. Jafarzadeh H, Shirazi AS, Andersson L. The most-cited articles in dental, oral, and maxillofacial traumatology during 64 years. *Dent Traumatol*. 2015;31(5):350–360. doi:10.1111/edt.12195
37. Kodonas K, Fardi A, Gogos C, Economides N. Scientometric analysis of vital pulp therapy studies. *Int Endod J*. 2021;54(2):220–230. doi:10.1111/iej.13422
38. Ahmad P, Mohamed Elgamal HA. Citation classics in the *Journal of Endodontics* and a comparative bibliometric analysis with the most downloaded articles in 2017 and 2018. *J Endod*. 2020;46(8):1042–1051. doi:10.1016/j.joen.2020.04.014
39. Schwendicke F, Golla T, Dreher M, Krois J. Convolutional neural networks for dental image diagnostics: A scoping review. *J Dent*. 2019;91:103226. doi:10.1016/j.jdent.2019.103226
40. Ekert T, Krois J, Meinhold L, et al. Deep learning for the radiographic detection of apical lesions. *J Endod*. 2019;45(7):917–922.e5. doi:10.1016/j.joen.2019.03.016
41. Setzer FC, Shi KJ, Zhang Z, et al. Artificial intelligence for the computer-aided detection of periapical lesions in cone-beam computed tomographic images. *J Endod*. 2020;46(7):987–993. doi:10.1016/j.joen.2020.03.025
42. Choi S, Choi J, Peters OA, Peters CI. Design of an interactive system for access cavity assessment: A novel feedback tool for preclinical endodontics. *Eur J Dent Educ*. 2023;27(4):1031–1039. doi:10.1111/eje.12895

Original paper

Quality of education and the evaluation of its outcomes in tooth preparation among dental students in Poland

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Abstract

Background. The preparation of model teeth for prosthetic crowns plays a crucial role in preclinical dental education. Its primary objective is to ensure optimal conditions for training students to function in clinical settings.

Objectives. The aim of the study was to assess the quality of dental students' education regarding the preparation of model teeth for cast metal-ceramic crowns. An additional objective was to compare the agreement between measurements made by examiners on the model teeth prepared by the students.

Material and methods. A total of 180 model premolar teeth (KaVo) were used for the tests. The teeth were prepared by the third-year dental students, who created a shoulder finish line on the buccal surface and a chamfer finish line on the palatal surface. Two examiners assessed the convergence angles of walls, the margin width of surfaces, the reduction in the height of functional and non-functional cusps, the extent of the preparation, the smoothness of the preparation, and the continuity of the finish line using simple measuring tools.

Results. The students' most proficient parameters were margin width, the extent of the preparation, and the finish line continuity. The least accurate results were observed in the wall convergence angles and the reduction in the height of functional and non-functional cusps. The interexaminer reliability was considerable for convergence angles, the margin width and the reduction in the height of cusps (intraclass correlation coefficient (*ICC*) >0.75). The examiners demonstrated moderate agreement (kappa: 0.60–0.79) for parameters such as the extent of the preparation, wall smoothness and finish line continuity.

Conclusions. The exercises provided a solid foundation for the students. However, practice and more targeted feedback are necessary to improve performance in the more challenging aspects of tooth preparation. The traditional analytical approach that uses simple measuring tools to assess the tooth preparation procedure is a reproducible method that examiners can use to effectively evaluate students' work.

Keywords: dental students, evaluation, dental education, prosthodontic tooth preparation

Highlights

- Dental students need practical exercises and well-targeted feedback from instructors to enhance their clinical skills.
- In supervising crown preparations, dentists should closely evaluate wall convergence and the reduction height of functional and non-functional cusps.
- The traditional analytical approach using basic measuring tools remains a reliable and reproducible method for assessing the quality of students' tooth preparations.

Introduction

The quality of education in dentistry is contingent upon numerous factors, including a well-organized curriculum, effective teaching methods, modern educational tools, and appropriate academic support. A well-designed curriculum should cover a wide range of topics and be regularly updated. The efficacy of teaching methods and tools is paramount in ensuring that students can thoroughly master both theoretical and practical issues. Teachers play a crucial role in education, as their approach to delivering knowledge, assessing students and providing feedback significantly influences students' skills. These elements contribute to the training of skilled dentists who can provide patients with a high standard of dental care. Prosthodontics is a field of dentistry that requires theoretical knowledge and high practical skills. Students initially develop their precision and manual skills through preclinical exercises, followed by clinical exercises.[1,2]

Preparing simulated model teeth for prosthetic crowns plays a crucial role in preclinical dental education in the field of prosthodontics. Its primary objective is to ensure optimal conditions for training students to function in clinical settings. The preclinical training assumes the form of practical sessions that offer dental students the opportunity to engage with a novel learning environment. Besides providing theoretical knowledge, these classes are designed to develop the students' manual skills and foster an understanding of "spatial vision".[2,3] Initially, students are required to complete a series of theoretical sessions, encompassing seminars and lectures, that prepare them for practical exercises. Subsequently, they perform activities demonstrated to them beforehand on specially prepared simulated models, and only after successfully completing these sessions they can transition to working with actual patients. Preclinical sessions introduce students to deviations from the ideal (standard) preparation and encourage them to identify their shortcomings and to work on improvements. The evaluation of students' performance at the preclinical stage is a pivotal aspect of education, as it serves to reinforce the learning process. It enables early error detection and guides students in their work. Furthermore, it provides teachers with insights into the specific work stages or parameters that pose the greatest challenges for students and require special attention or repetition. All these factors translate into easier, less stressful, and improved patient care.[4,5] There is a positive correlation between students' grades in preclinical and clinical exams, indicating that appropriate preparation in the earlier stages of education can positively influence clinical competencies of students.[6]

The identification and implementation of effective methods for assessing students' performance remains a significant challenge for academic teachers and educators.[7,8] There are noticeable differences in tooth preparation assessments, with discrepancies noted both between assessors and between the assessments made by the same examiner evaluating the same procedure at different times.[9] The development of objective approaches to evaluating tooth preparation is a critical aspect of dental education. Current methods for the assessment of tooth preparation can be divided into traditional and digital techniques.[4]

The traditional approach is the most often adopted by examiners experienced in a specific field, who compare the work of students with a standard model. In traditional practice, researchers distinguish between a global assessment (inspection and evaluation) method and an analytical (using rubrics) method. The former is a straightforward visual technique in which the examiner provides a comprehensive grade for the student's work. This subjective method rarely produces consistent results.[10] According to some researchers, examiner fatigue is a particularly important factor contributing to inconsistencies between ratings.[9] Conversely, the analytical method involves the creation of rubrics, based on textbooks and publications, to assess individual components of tooth preparation. Specific parameters are evaluated by measuring certain distances and angles using basic measuring tools such as a ruler, a compass and a protractor. The parameters that are frequently measured in this manner include the convergence angles between the walls, the width of the prepared margin, the reduction in the height of the occlusal surface, the extent of the prepared finish line, and the smoothness of the preparation.[4] In this method, proper calibration of the assessors is vital. This process involves the formulation of the objective assessment criteria for the preparations, so that the latter are easy to measure,

interpret and reproduce. This approach enhances accuracy and precision, thereby mitigating the risk of subjectivity and inconsistency in evaluations. For students engaged in the learning process, consistent, definite and repeatable feedback from educators regarding the quality of their work is of the utmost importance. Such feedback enables students to absorb knowledge in a specific field more rapidly and to focus on the learning process rather than on wondering whether the grade they have received is fair or accurate.[8] Research suggests that the reliability of student assessments can be increased by employing 2 assessors, as opposed to the traditional method in which a single examiner is involved.[8,11] Without assessor calibration, there is a risk of diverging assessments made by different teachers.

In recent years, more advanced evaluation methods have been introduced, such as digital assessments of tooth preparations that employ various scanners and computer-aided design software. The efficacy of these methods is primarily attributable to their accuracy. They can also serve as an additional educational approach in preclinical sessions.[12,13] While digital technologies facilitate the acquisition of manual skills, there is no evidence regarding the long-term impact of digital training methods on clinical outcomes and student competencies.[14] Despite the immediate feedback provided by these methods, studies have demonstrated that students prefer receiving feedback from a teacher during sessions and also benefit from digital methods. Similarly, during exams, they seek evaluation from both human examiners and computers. Students perceive digital methods as an objective source of feedback and a valuable addition to teaching practical skills. Yet, they do not want to entirely abandon traditional assessment methods in favor of newer technologies.[15,16] Sometimes, students achieve low or inconsistent results when first interacting with a digital assessment system.[3] The implementation of these methods requires appropriate training, and due to their relatively high cost and the need for special maintenance, they remain unavailable in most academic centers for use in preclinical sessions. As a result, the primary educational approach continues to rely on the traditional method for assessing students' readiness.[15,16]

The aim of the study was to assess the quality of dental students' education regarding the preparation of model teeth for cast metal-ceramic crowns. An additional objective was to compare the agreement between measurements made by examiners on the model teeth prepared by the students.

Material and methods

The present study was based on 180 models of maxillary right first premolars provided by KaVo (Biberach, Germany) and prepared by third-year dental students at the Jagiellonian University Medical College (Krakow, Poland). A total of 180 students, with an average age of 22 years, participated in the study. The study protocol followed the ethical guidelines established by the 1975 Declaration of Helsinki and was approved by the Institutional Ethics Committee at Jagiellonian University Medical College (approval No. 118.6120.109.2023). Prior to their involvement in the study, each student provided informed consent.

The preparation stage was preceded by a series of theoretical lessons, including lectures and seminars. The students attended a total of 3 h of lectures, which were delivered by a professor and included a range of subjects, such as the definition of prosthetic crowns, the various types of prosthetics crowns and the indications for their use. Additionally, the lectures covered the general principles of tooth preparation. The seminars were led by academic teachers and consisted of 2 sessions, each with a duration of 45 min. During these sessions, students were acquainted with the principles for properly preparing the maxillary right first premolar for a cast metal-ceramic crown (Table 1). The students received instructions verbally and in the form of an instructional video. The preparation process involved creating a shoulder margin on the buccal surface, a chamfer margin on the palatal surface, and a smooth transition between these finish lines on the proximal surfaces. Directly after the second seminar, the students began the practical exercises. The participants had 2 h and 30 min to perform the preparation. This task constituted the students' first experience with a tooth preparation procedure. A specific bur set, designed by Prof. Stanisław W. Majewski and Prof. Bartłomiej W. Loster (Olident, Cologne, Germany) and intended for a crown with a step-type finish line preparation was used. To mimic the natural conditions of the oral cavity, the model teeth were mounted on partial dentition study models (OK T12; KaVo) and then on dental patient simulators (KaVo). The practical part was supervised by academic teachers. During the preparation phase, the students did not communicate with each other. Once the exercises were completed, the teeth were extracted from the study models, collected and numbered by the examiners. Figure 1 presents the premolar tooth 14 mounted on the partial dentition study model before and after preparation.

The authors adopted an analytical method for evaluating the prepared teeth, wherein each parameter was assessed individually. The measurements were made by 2 independent examiners (examiner A and examiner B) who were dental practitioners and academic teachers with over a decade of professional experience. The examiners familiarized themselves with the preparation assessment criteria (Table 1), and they were calibrated before proceeding with their evaluations. The calibration was performed on a model of an ideal

Table 1. Assessment criteria for a maxillary right first premolar prepared for a cast metal-ceramic crown

Type of parameter	Standard parameter	Assessment method
Reduction in the height of the functional cusp	1.5 mm	measurement from the cementoenamel junction to the highest point of the palatal cusp
Reduction in the height of the non-functional cusp	1 mm	measurement from the cementoenamel junction to the highest point of the buccal cusp
Wall convergence angle	4–6 degrees (ideal) 7–16 degrees (acceptable)	measurement of the mesiodistal and buccopalatal angles
Width of the chamfer margin	0.5–0.8 mm	measurement of the palatal margin
Width of the shoulder margin	1.0–1.2 mm	measurement of the buccal margin
Extent of the preparation	supragingival	visual assessment from the mesial, distal, buccal, and palatal sides (supragingival/above the gingiva, epigingival/to the gingiva, subgingival/under the gingiva)
Smoothness of the walls and the margin	high	finish line and margin are continuous, clearly defined, and smooth; walls are smooth
	medium	finish line and margin are continuous but not smooth; walls are moderately smooth
	low	finish line and/or margin are not continuous, considerable roughness of the finish line and/or walls and/or margin is noted
Finish line continuity	yes	margin is continuous throughout
	no	margin is interrupted

preparation, indicating how deviations from the standard model should be assessed. After the calibration and before the evaluation, the examiners assessed a random sample of 15 prepared teeth to ensure the consistency of the evaluations and the comprehension of the assessment criteria. Each examiner independently evaluated 15 preparations, after which the results were analyzed and discussed collectively.

The authors assessed the convergence angles mesiodistally and buccopalatally, the width of the margin at the midpoint of the mesial (m), distal (d), buccal (b), and palatal (p) surfaces, the reduction in the height of functional and non-functional cusps, the extent of the preparation from the mesial, distal, buccal, and palatal sides, the smoothness of the preparation, and the continuity of the finish line. The convergence angles were measured using a white sheet of paper, a pencil (Faber-Castell Grip 2001 "H"; Faber Castell, Stein, Germany), a plastic ruler (Herlitz, Hanover, Germany), and a plastic protractor (Herlitz). The analysis involved placing and stabilizing the tooth on a piece of paper, marking 2 lines using a pencil and a ruler along the prepared tooth surfaces, and measuring the angle at which the lines intersected using a protractor. This procedure was repeated twice for each tooth – first for the mesiodistal angle and then for the buccopalatal angle after the tooth was flipped. The margin widths were measured at the midpoint of each surface with an analog calliper (Högert Technik GmbH, Pruszków, Poland) after which they were remeasured with a ruler.

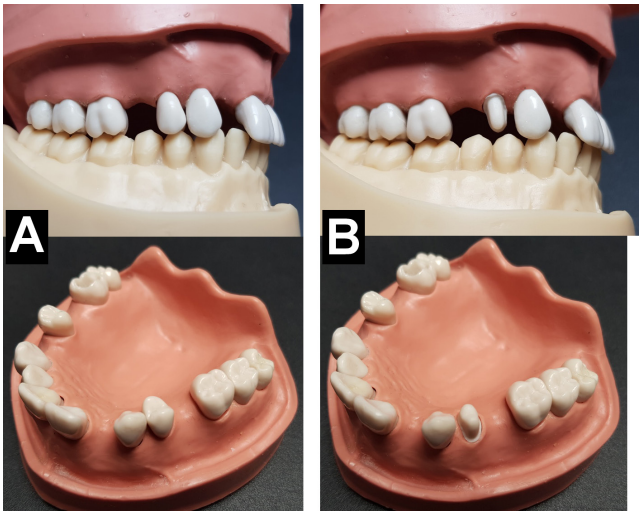

Fig. 1. Premolar tooth 14 mounted on the partial dentition study model before (A) and after (B) preparation

The height of the cusp reduction was measured using a calliper, with the measurement extending from the cementoenamel junction to the highest point of each cusp. The actual value was then determined with a ruler [mm]. The remaining parameters were visually assessed by the examiners, according to the established criteria (Table 1). Each examiner had 3 min to complete a single tooth assessment. The examiners were instructed to perform this task independently, that is, without consulting one another.

Statistical analysis

The continuous variables were presented as medians and interquartile ranges, while the categorical variables were expressed as frequencies and percentages. The interexaminer reliability in the case of convergence angles, margin width and reduction height was assessed by means of the intraclass correlation coefficient (*ICC*). In the case of finish line continuity, Cohen's kappa was used, while for the extent and smoothness of the preparation, the weighted kappa was the method of choice. The significance level was set at $p < 0.05$. The calculations were performed using the IBM SPSS Statistics for Windows software, v. 26.0 (IBM Corp., Armonk, USA).

Results

For examiners A and B, the mesiodistal (m-d) walls exhibited median convergence angles of 22.5 (*IQR*: 17–28) and 20.5 (*IQR*: 15–28) degrees (*ICC* = 0.821), and for the buccopalatal (b-p) walls, the angles were 23 (17–30) and 22 (16–29) degrees (*ICC* = 0.938), respectively. The differences in the margin width, as measured by examiners A and B, yielded the following medians: for the mesial margin – 0.50 (0.25–0.75) mm and 0.50 (0.25–0.50) mm, respectively (*ICC* = 0.774); for the distal margin – 0.75 (0.50–1.00) mm and 0.50 (0.25–1.00) mm, respectively (*ICC* = 0.866); for the buccal margin – 1.00 (0.75–1.00) mm and 1.00 (0.50–1.00) mm, respectively (*ICC* = 0.818); for the palatal margin – 1.00 (0.75–1.25) mm in the case of both examiners (*ICC* = 0.858). The median values for the functional cusp reduction height were 1 (*IQR*: 1–1.5) mm for examiner A and 1 (*IQR*: 1–2) mm for examiner B (*ICC* = 0.830). For the non-functional cusps, the medians were 2.0 (1.5–2.3) mm and 2.0 (1.5–2.5) mm, respectively (*ICC* = 0.893). With regard to the agreement between the doctors on the students' measurements of the extent of the preparation, the values were as follows: mesial kappa = 0.694; distal kappa = 0.600; buccal kappa = 0.728; and palatal kappa = 0.616. The smoothness of the wall and finish line continuity yielded kappa values of 0.726 and 0.617, respectively.

There was considerable agreement between the examiners (*ICC* > 0.75) regarding aspects such as the m-d and b-p wall convergence angles, margin widths, and the reduction heights of both the functional and non-functional cusps. Moderate interexaminer reliability was observed for the extent of the preparation, wall smoothness, and finish line continuity, with the kappa values ranging from 0.600 to 0.728. The most significant discrepancy between the examiners' measurements concerned the extent of the preparation on the distal side, while the b-p convergence angle exhibited the least variance (Table 2).

Table 2 presents the number of students who met the optimal benchmarks for individual preparation parameters, segmented by each evaluator's assessment. According to examiner A, only 6 (3.3%) students achieved the optimal m-d wall convergence angle, whereas examiner B identified 4 (2.2%) such students. For the b-p angle, examiners A and B identified 6 (3.3%) and 8 (4.4%) students, respectively. In the case of the acceptable m-d wall convergence angle, they found 37 (20.7%) and 50 (27.8%) students meeting the criteria, respectively. The corresponding numbers for the b-p angle were 37 (20.7%) and 46 (25.6%). With regard to the ideal shoulder margin width, 89 (49.4%) students satisfied the criteria according to examiner A and 113 (62.8%) according to examiner B. The optimal chamfer margin width was achieved by 152 (84.4%) students for examiner A and 136 (75.6%) for examiner B. Only 25 (13.9%) students achieved the correct reduction height for functional cusps in examiner A's assessment, while 39 (21.7%) did so according to examiner B. Examiner A reported that 26 (14.4%) students attained the correct reduction height for non-functional cusps, while examiner B noted this achievement in only 15 (8.3%) individuals. One area in which the students excelled was the extent of the preparation on specific tooth surfaces. According to examiners A and B, the supragingival preparation was accomplished by 85.6–96.1% and 75.0–93.3% of the students, respectively, and depended on the tooth surface. A majority of the students, namely 80 (44.4%) students in the case of examiner A and 89 (49.4%) according to examiner B, produced a preparation with average wall smoothness. According to examiners A and B, a high degree of surface smoothness was achieved by only 61 (33.9%) and 46 (25.6%) students, respectively. The majority of the students managed to maintain the finish line continuity of the maxillary right first premolar, e.g., 112 (62.2%) of the students evaluated by examiner A and 94 (52.2%) of those assessed by examiner B. The students' most proficient parameters were the shoulder and chamfer margin widths, the extent of the preparation, and the finish line continuity. Conversely, the most challenging areas were the wall convergence angles and the reduction heights of both the functional and non-functional cusps.

Discussion

Preparing a tooth for prosthetic crowns is one of the most challenging manual procedures that students encounter during their dental training. In a study conducted by Hattar et al., the confidence levels of fifth-year students were evaluated by a questionnaire when performing various dental procedures.[17] The students felt most competent and assured when performing direct restorations and endodontics, and least confident when dealing with fixed prosthodontics. In the context of fabricating indirect restorations, they exhibited a notable decrease in confidence during the preparation of tooth crowns when compared to taking impressions or trying in the prosthetic restoration.[17] Appropriate training during preclinical sessions is of the utmost importance in enhancing student skills and bolstering their confidence in the field of fixed prosthodontics.

The present study identifies specific areas in dental education that require improvement to enhance students' practical skills in tooth preparation for prosthetic crowns. It also highlights the importance of detailed and consistent assessments in education.

The students demonstrated the highest level of skills in achieving optimal margin widths on various surfaces, the extent of the preparation, and maintaining finish line continuity.

A high percentage of students met the criteria for optimal shoulder and chamfer margin widths. Specifically, from

Table 2. Comparison between the ratings of the 2 examiners for each variable with the corresponding agreement measure

Variable		Examiner A		Examiner B		Agreement measure
		n	%	*n*	%	
m-d convergence angle	0–6°	6	3.3	4	2.2	0.821 (0.760–0.867)
	7–16°	37	20.6	50	27.8	
	>16	137	76.1	126	70.0	
b-p convergence angle	0–6°	6	3.3	8	4.4	0.983 (0.915–0.954)
	7–16°	37	20.6	46	25.6	
	>16	137	76.1	126	70.0	
m width	<0.5 mm	80	44.4	76	42.2	0.774 (0.697–0.832)
	0.5–1 mm	97	53.9	101	56.1	
	>1 mm	3	1.7	3	1.7	
d width	<1 mm	105	58.3	115	63.9	0.866 (0.810–0.904)
	1–1.5 mm	71	39.4	61	33.9	
	>1.5 mm	4	2.2	4	2.2	
b width (shoulder)	<1 mm	73	40.6	46	25.6	0.858 (0.806–0.896)
	1–1.5 mm	89	49.4	113	62.8	
	>1.5 mm	18	10.0	21	11.7	
p width (chamfer)	<0.5 mm	7	3.9	20	11.1	0.818 (0.741–0.871)
	0.5–1 mm	152	84.4	136	75.6	
	>1 mm	21	11.7	24	13.3	
Reduction in the height of the functional cusp	<1 mm	30	16.7	16	8.9	0.830 (0.630–0.906)
	1 mm	103	57.2	78	43.3	
	1.5 mm	25	13.9	39	21.7	
	2 mm	16	8.9	39	21.7	
	>2 mm	6	3.3	8	4.4	
Reduction in the height of the non-functional cusp	<1 mm	5	2.8	2	1.1	0.893 (0.756–0.943)
	1 mm	26	14.4	15	8.3	
	1.5 mm	43	23.9	37	20.6	
	2 mm	61	33.9	58	32.2	
	>2 mm	45	25.0	68	37.8	
m preparation extent	above	154	85.6	140	77.8	0.694 (0.548–0.841)
	to	17	9.4	27	15.0	
	under	9	5.0	13	7.2	
d preparation extent	above	160	88.9	135	75.0	0.600 (0.437–0.763)
	to	12	6.7	32	17.8	
	under	8	4.4	13	7.2	
p preparation extent	above	164	91.1	150	83.3	0.616 (0.430–0.803)
	to	14	7.8	25	13.9	
	under	2	1.1	5	2.8	
b preparation extent	above	173	96.1	168	93.3	0.728 (0.531–0.925)
	to	5	2.8	5	2.8	
	under	2	1.1	7	3.9	
Preparation smoothness	low	39	21.7	45	25.0	0.726 (0.640–0.812)
	medium	80	44.4	89	49.4	
	high	61	33.9	46	25.6	
Finish line continuity	yes	112	62.2	94	52.2	0.617 (0.503–0.730)
	no	68	37.8	86	47.8	

m-d – mesiodistal; p-b – buccopalatal; m – mesial; d – distal; p – palatal; b – buccal; above – above the gingiva; to – to the gingiva; under – under the gingiva. The agreement measure values are presented as kappa (95% confidence interval (*CI*)) for the preparation extent and smoothness, weighted kappa (95% *CI*) for the continuity of the finish line, and intraclass correlation coefficient (*ICC*) (95% *CI*) for the remaining variables.

49.4% (examiner A) to 62.8% (examiner B) of students achieved the ideal shoulder margin width, and from 75.6% (examiner B) to 84.4% (examiner A) achieved the optimal chamfer margin width. Maintaining finish line continuity was achieved by more than half of the students. This finding aligns with the conclusions of other authors. In a study by Sadid-Zadeh et al., 75% of the cases attained the desired width and continuity for the given crown type.[18] Al-Omari and Al-Wahadni reported an average margin width of 0.86 mm buccally, 0.74 mm palatally, 0.7 mm mesially, and 0.66 mm distally.[19] These values were lower than the recommended 1.0–1.2 mm for a shoulder-type margin. An excessively wide margin violates the biological principles of tooth preparation. Conversely, a margin that is too narrow may result in an aesthetically pleasing tooth restoration, especially when it comes to less translucent materials. The presence of a discontinuous finish line may compromise the marginal adaptation of the restorations.

One parameter in which students achieved good results was the location of the finish line relative to the gum line. In over 75% of the prepared specimens, the required supragingival margin was achieved. This adequate tooth preparation, which approaches the standard, is instrumental in maintaining proper hygiene and sealing prosthetic restorations. This finding is consistent with the research, wherein students also obtained favorable results for this parameter.[4]

Most students achieved an average level of smoothness of the prepared tooth stump surface. A lack of wall smoothness can reduce the fit of the prosthetic restoration. Smoothness is particularly important at the gingival level of the finish line, as its absence in this area can cause the prosthetic restorations to leak. In addition, the findings of other authors indicate that this is not an easy parameter to achieve.[18] The required smoothness of the prepared stumps was achieved by less than half of the students.[18]

In the present study, the students were the least precise in their preparation of the convergence angles of the mesiodistal and buccopalatal walls, greatly exceeding the presumed margins of error (optimal values). The standard guidelines for wall convergence angles during the crown preparation procedure consider an angle within the range of 4–6 degrees as ideal, and 7–16 degrees as acceptable.[20,21] The more the opposing prepared walls lie parallel to one another, the greater the retention of the final prosthetic restoration. However, achieving these recommendations in clinical practice can present challenges as it requires a high level of precision and clear visibility in the operative field. Factors such as the position of the tooth within the dental arch, anatomical structure and the practitioner's experience all play significant roles. Achieving almost parallel walls without the risk of undercuts in the tooth preparation is challenging. Excessive reduction of the tooth stump, resulting in an increased convergence angle, may compromise the retentive properties of the abutment.[20,22] Numerous researchers have noted overly high convergence angles among dental students practicing on model teeth. In a study by Ayad et al., no more than 47% of the teeth prepared by dental students had acceptable convergence angles.[21] Al-Omari and Al-Wahadni examined 157 tooth samples prepared for metal-ceramic crowns, finding average convergence angles of 22.4 degrees and 25.3 degrees for buccopalatal and mesiodistal walls, respectively.[19] Strain et al. analyzed articles from 11 countries, published between 1978 and 2014, encompassing a total of 2,306 preparations.[23] In these studies, students failed to achieve wall convergence angles between 4 and 14 degrees. Most papers reported angles ranging from 10 to 20 degrees.[23]

Another challenging parameter in the study was the reduction in the height of both functional and non-functional cusps. Less than one-fourth of the students achieved correct results in these parameters. Excessive reduction of the occlusal surface may lead to poorer retention of the prosthetic restoration. Insufficient occlusal reduction necessitates thinner layers of restorative materials, which can potentially affect the crown's resilience, longevity and aesthetics. Sadid-Zadeh et al. found optimal cusp reduction values difficult to achieve.[18] Out of 223 teeth prepared by students in preclinical sessions, only 53.7% obtained optimal cusp reduction values.[18] Al-Omari and Al-Wahadni reported occlusal surface reduction ranging from 1.8 mm to 2.2 mm, with an average reduction of 2 mm. These values are slightly above the ideal but nevertheless still ensure crown durability and good aesthetics.[19]

Achieving the correct convergence angle and cusp height reduction values is challenging for the majority of dental students and requires vigilant oversight from supervising clinicians. Students should prioritize refining their skills in areas in which they received lower scores from the examiners.

In the present study, the work of the students was assessed using the traditional analytical method. To further increase the credibility of the assessments, 2 independent examiners participated in the research. Each examiner evaluated the work of the students separately. The traditional analytical method of evaluation has been used in the field of dentistry for many years, and it is considered reliable and repeatable.[24] A validated and properly executed assessment process can reduce students' stress levels and have an impact on achieving better results in exercises. Studies indicate that the most significant sources of stress for students include assessments, exams, and the atmosphere created by clinical professors.[25,26] According to the literature, the level of specialist knowledge did not influence preclinical assessments based on the analytical method. Both younger and older lecturers reported similar results. Differences in assessments were only observed in groups with no prior experience in fixed prosthodontics.[9,27] Thus, it can be regarded as a universal method of assessment that can be successfully applied by less experienced individuals in the event of a shortage of senior training staff.

However, the criteria for individual parameters should be precisely defined and the evaluators properly calibrated. Esser et al. observed significant discrepancies between the correlations of maxima and medians when examiners visually assessed students' individual preparation parameters, thus indicating insufficient calibration of the evaluators.[5] They emphasized that both targeted calibration and more precisely formulated criteria could improve the quality of results.[5] The study noted a very high level of interexaminer reliability in the case of the wall convergence angle, cusp reduction height and margin width, as assessed using simple measuring tools. These results demonstrate that when an examination is correctly prepared, even simple methods and instruments such as a compass and a ruler may suffice to properly assess students' work in preclinical classes. There was a moderate agreement between the evaluators with regard to parameters such as the extent of the preparation, surface smoothness, and finish line continuity. These parameters were assessed visually based on the established criteria. The evaluation of parameters that were not quantifiable was more difficult despite the existence of precise criteria and the calibration of examiners.

The conducted study emphasizes the importance of standardizing dental curricula across educational institutions as a pivotal component of dental education. This would ensure the uniformity of educational standards and enhance its quality. Additionally, the quality of dental education can be improved by the adoption of innovative teaching methods such as simulation-based training, formative assessments and other interactive techniques, as well as by the establishment of robust feedback systems to provide students with regular and constructive critiques of their work. There is a clear need for longitudinal studies assessing the long-term impact of educational interventions on clinical competency.

The strengths of the present study include comprehensive assessment, combining both quantitative and qualitative data. Moreover, a diverse sample provided a broad perspective on educational quality. The study also offers practical recommendations based on empirical evidence for conducting preclinical dental preparation courses.

Limitations

The present research was subject to certain limitations. It was conducted in a single educational center and only by 2 examiners. In the future, it would be beneficial to expand the research to include other university dental schools and increase the number of examiners. The criteria adopted for the study were not assessed by any independent external institution. Another limitation is the possibility of bias in the self-reported qualitative data, as well as the cross-sectional nature of the study, which lacks long-term outcome assessment. The present research was limited to the domain of tooth preparation and may not be generalized to other aspects of dental education.

Conclusions

A combination of theoretical and practical exercises allowed students to acquire basic knowledge and skills in the preparation of teeth for prosthetic crowns. In order to improve their performance during the more difficult stages of preparations, students need practical exercises and well-targeted feedback from instructors. Dentists who supervise students should pay particular attention to the parameters in which students achieved their lowest scores in terms of precision, namely the degree of wall convergence and the reduction in the height of both functional and non-functional cusps.

The traditional analytical approach to assessing the tooth preparation procedure is a reproducible method that examiners can use to effectively evaluate students' work. Targeted calibration of the evaluators is essential to ensure more accurate results. Parameters that are not easily quantifiable are more challenging to evaluate.

Ethics approval and consent to participate

The study protocol followed the ethical guidelines established by the 1975 Declaration of Helsinki and was approved by the Institutional Ethics Committee at Jagiellonian University Medical College, Krakow, Poland (approval No. 118.6120.109.2023). Prior to their involvement in the study, each student provided informed consent.

Data availability

The datasets generated and/or analyzed during the current study are available from the corresponding author on reasonable request.

Consent for publication

Not applicable.

Use of AI and AI-assisted technologies

Not applicable.

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References

1. Field J, Spencer R, Johnson I, Cure R. Professional standards in dental education. *Br Dent J*. 2020;228(11):875–881. doi:10.1038/s41415-020-1640-5
2. Capehart KL, Harris RL, Britton EE, et al. Dental school quality assessment: Effect of student calibration on the outcomes of fixed prosthodontic cases. *J Dent Educ*. 2023;87(9):1294–1301. doi:10.1002/jdd.13253

3. Kateeb ET, Kamal MS, Kadamani AM, Abu Hantash RO, Abu Arqoub MM. Utilising an innovative digital software to grade pre-clinical crown preparation exercise. *Eur J Dent Educ*. 2017;21(4):220–227. doi:10.1111/eje.12204
4. Habib SR. Rubric system for evaluation of crown preparation performed by dental students. *Eur J Dent Educ*. 2018;22(3):e506–e513. doi:10.1111/eje.12333
5. Esser C, Kerschbaum T, Winkelmann V, Krage T, Faber FJ. A comparison of the visual and technical assessment of preparations made by dental students. *Eur J Dent Educ*. 2006;10(3):157–161. doi:10.1111/j.1600-0579.2006.00408.x
6. Al-Angari NS, Aldaham AS, Masuadi E, Nadeem M, Alkadi L. The effectiveness of students' performance in preclinical fixed prosthodontics course in predicting subsequent clinical performance. *J Prosthodont*. 2022;31(1):45–49. doi:10.1111/jopr.13363
7. Tang L, Cao Y, Liu Z, et al. Improving the quality of preclinical simulation training for dental students using a new digital real-time evaluation system. *Eur J Dent Educ*. 2021;25(1):100–107. doi:10.1111/eje.12580
8. Habib SR, Sherfudhin H. Students' self-assessment: A learning tool and its comparison with the faculty assessments. *J Contemp Dent Pract*. 2015;16(1):48–53. doi:10.5005/jp-journals-10024-1634
9. Al Amri MD, Sherfudhin HR, Habib SR. Effects of evaluator's fatigue and level of expertise on the global and analytical evaluation of preclinical tooth preparation. *J Prosthodont*. 2018;27(7):636–643. doi:10.1111/jopr.12558
10. O'Donnell JA, Oakley M, Haney S, O'Neill PN, Taylor D. Rubrics 101: A primer for rubric development in dental education. *J Dent Educ*. 2011;75(9):1163–1175. PMID:21890846.
11. Feil PH. An analysis of the reliability of a laboratory evaluation system. *J Dent Educ*. 1982;46(8):489–494. PMID:6954179.
12. Park CF, Sheinbaum JM, Tamada Y, et al. Dental students' perceptions of digital assessment software for preclinical tooth preparation exercises. *J Dent Educ*. 2017;81(5):597–603. doi:10.21815/JDE.016.015
13. Liu L, Li J, Yuan S, et al. Evaluating the effectiveness of a preclinical practice of tooth preparation using digital training system: A randomised controlled trial. *Eur J Dent Educ*. 2018;22(4):e679–e686. doi:10.1111/eje.12378
14. Erdilek D, Gümüştaş B, Güray Efes B. Digitalization era of dental education: A systematic review. *Dent Med Probl*. 2023;60(3):513–525. doi:10.17219/dmp/156804
15. Schepke U, van Wulfften Palthe ME, Meisberger EW, Kerdijk W, Cune MS, Blok B. Digital assessment of a retentive full crown preparation–An evaluation of prepCheck in an undergraduate preclinical teaching environment. *Eur J Dent Educ*. 2020;24(3):407–424. doi:10.1111/eje.12516
16. Seet RH, Soo PR, Leong KJM, Pang JJH, Lee FKF, Tan MY. Crown preparations by undergraduate dental students: A comparison of conventional versus digital assessment via an intraoral scanner. *J Dent Educ*. 2020;84(11):1303–1313. doi:10.1002/jdd.12285
17. Hattar S, AlHadidi A, Altarawneh S, Hamdan AAS, Shaini FJ, Wahab FK. Dental students' experience and perceived confidence level in different restorative procedures. *Eur J Dent Educ*. 2021;25(1):207–214. doi:10.1111/eje.12592
18. Sadid-Zadeh R, DeLuca JT, Arany H, Li R. Assessment of teeth prepared by senior dental students for CAD/CAM restorations. *J Dent Educ*. 2020;84(3):358–366. doi:10.21815/JDE.019.178
19. Al-Omari WM, Al-Wahadni AM. Convergence angle, occlusal reduction, and finish line depth of full-crown preparations made by dental students. *Quintessence Int*. 2004;35(4):287–293. PMID:15119714.
20. Amine M, Wahid HO, Fahi S, Lehmouddi S, Hamza M, Elarabi S. Assessment of convergence angle of tooth preparations for complete crowns among dental students: Typodont vs simulator. *Int J Dent*. 2022;2022:7615892. doi:10.1155/2022/7615892
21. Ayad MF, Maghrabi AA, Rosenstiel SF. Assessment of convergence angles of tooth preparations for complete crowns among dental students. *J Dent*. 2005;33(8):633–638. doi:10.1016/j.jdent.2004.12.008
22. Marghalani TY. Convergence angles of metal ceramic crowns prepared by dental students. *J Prosthet Dent*. 2014;112(5):1250–1256. doi:10.1016/j.prosdent.2014.03.024
23. Strain KJ, Mackie J, Bonsor SJ, Macfarlane TV. Crown taper angles achieved by dental students: A systematic review. *J Dent Educ*. 2018;82(11):1203–1212. doi:10.21815/JDE.018.125
24. Dhuru VB, Rypel TS, Johnston WM. Criterion-oriented grading system for preclinical operative dentistry laboratory course. *J Dent Educ*. 1978;42(9):528–531. PMID:280591.
25. Smolana A, Loster Z, Loster J. Assessment of stress burden among dental students: A systematic literature review and meta-analysis of data. *Dent Med Probl*. 2022;59(2):301–307. doi:10.17219/dmp/133955
26. Ertürk Avunduk AT, Delikan E. Satisfaction and stress levels of dentistry students relating to distance education. *Dent Med Probl*. 2021;58(3):291–298. doi:10.17219/dmp/135318
27. Strain K, Horsfall L, Bonsor S, Ibbetson R. The accuracy of dental students and dentists assessing the total occlusal convergence of crown preparations. *Eur J Prosthodont Restor Dent*. 2020;28(2):86–93. doi:10.1922/EJPRD_1984Strain07

Original paper

Effects of the probiotics on the proliferation phase in oral wound healing: In vivo study

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Abstract

Background. Probiotics, known for their anti-inflammatory properties and ability to balance the oral microbiome, show promise in enhancing wound healing, particularly through topical application, in oral healthcare.

Objectives. The main objective of the present study was to investigate the topical application of probiotics to accelerate oral wound healing, focusing on key indicators, like collagen density, angiogenesis, the re-epithelization rate, the wound area, and the wound length.

Material and methods. Palatal wounds were induced in 60 male Sprague–Dawley® rats, which were divided into 4 groups. Probiotics, including *Streptococcus salivarius* K12 (BLIS K12®) and *Lactobacillus reuteri* (Interlac® Pro-D and Interlac®), were topically administered. The rates were sacrificed on days 3, 7 and 14 to evaluate the early, middle and late proliferation phases. Histopathological examinations assessed collagen density, angiogenesis, the re-epithelialization rate, the wound area, and the wound length.

Results. Probiotics showed beneficial effects on the oral wound healing indicators examined in this study. This study demonstrates the significant benefits of applying probiotics in enhancing wound healing throughout various proliferation stages. Our findings consistently highlight their positive impact across key indicators. With 3 different probiotic types, we observed improvement in all aspects of wound healing, from early to late stages.

Conclusions. The study underscores the potential of probiotics as effective agents in promoting wound repair and regeneration, offering promising avenues for enhanced clinical outcomes.

Keywords: *Streptococcus salivarius*, angiogenesis, probiotics, wound healing, *Limosilactobacillus reuteri*

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Highlights

- The study demonstrates that probiotics significantly improve wound healing throughout various stages of the proliferation phase.
- Improvement was observed in 5 critical aspects of healing – collagen density, angiogenesis, the re-epithelialization rate, the wound area, and the wound length.
- All 3 tested probiotic strains contributed to enhanced wound repair, indicating broad therapeutic potential.
- The findings suggest probiotics could be effective agents in wound management, highlighting the need for further research to understand mechanisms and refine clinical application.

Introduction

Oral and maxillofacial surgeons deal with surgeries in the mouth, jaw and face, which involves the intricate process of oral wound healing.[1,2] Oral wounds pose unique challenges due to the warm oral environment and the presence of abundant microorganisms. Wound healing, a complex physiological process involving various cell types, can be influenced by multiple factors. Understanding these intricacies is crucial for addressing the complexities of healing oral wounds within this surgical domain.[3]

The wound healing process in the oral cavity, especially in the proliferation phase, relies on several critical factors. In this phase, which is part of the 4 accurately programmed stages of wound healing – hemostasis, inflammation, proliferation, and remodeling or maturation, specific biophysiological mechanisms are activated.[4] These include collagen production, the development of new blood vessels through angiogenesis, and the restoration of the protective epithelial layer via re-epithelialization. The interactions between these elements are intricately tied to the size and morphology of the oral wound, amplifying the complexity of the overall healing process.[3]

Advancement in wound care has led to the development of materials specifically designed for wound dressing. These materials not only aim to expedite the healing process, but also address the unique challenges posed by mucosal wounds within the oral cavity. Such advances offer promising avenues for enhancing the management and treatment strategies for oral wounds, marking a significant progression in oral cavity care and wound management.[5]

In recent years, research delving into the potential of probiotic agents in oral healthcare has seen a significant surge. Probiotics, renowned for their role in fostering a balanced and healthy oral microbiome, have demonstrated considerable promise due to their notable anti-inflammatory properties. This particular attribute presents an opportunity to potentially alleviate inflammatory responses, thereby potentially enhancing the subsequent stages of wound healing. Studies conducted in the field have validated that both the oral administration and injection of probiotics can yield benefits in wound healing, particularly observed in rat models, where these interventions have stimulated the inflammatory processes, aiding in the healing of wounds.[6,7] This revelation has sparked interest and optimism in leveraging probiotics as a therapeutic approach for wound management in oral healthcare. Furthermore, investigations have highlighted the superiority of topical treatment over systemic approaches in addressing the physical traumas and chemical injuries of the oral mucosa.[8] This finding emphasizes the potential efficacy of localized application in the wound healing process within the oral cavity.

A deliberate push for further research in oral healthcare revolves around the topical application of probiotics to enhance the healing of oral wounds. This investigation aims to unravel the specific mechanisms through which topically applied probiotics can positively impact and expedite oral wound healing. By focusing on indicators like collagen density, angiogenesis and re-epithelialization, this research endeavors to deeply explore the interplay between probiotics and the intricate mechanisms governing oral wound healing. The ultimate goal is to develop innovative strategies that could revolutionize the management and treatment of oral wounds, offering promising new avenues in oral healthcare.

Material and methods

This research was conducted at the Veterinary and Biomedical Sciences Hospital and the Faculty of Veterinary Medicine and Biomedical Sciences of IPB University (Institut Pertanian Bogor), Bogor, Indonesia, from May to July 2023.

Probiotics

In this research study, 3 commercially available probiotics were employed: the *Streptococcus salivarius* K12 lozenge containing 1 billion colony-forming units (CFU) (BLIS K12®); *Lactobacillus reuteri* ATCC PTA 5289 at 1×10^8 CFU (Interlac® Pro-D) in lozenge form; and *Lactobacillus reuteri* DSM 17938 at 1×10^8 CFU provided

in powder sachet form. It is worth noting that the preparation of BLIS K12 and Interlac Pro-D formulations involved grinding down in a mortar, leading to the transformation of all tested probiotics into a powdered state, which facilitated the experimental procedures.

Rats

Based on the Federer formula, it was determined that the minimum sample size per intervention is 3. In this study, 5 rats were used per intervention; therefore sixty 8-week-old Sprague–Dawley® male rats weighing between 200–300 g were utilized. The rats were randomly assigned to 4 distinct groups, with each group consisting of 15 rats: group 1, receiving treatment with BLIS K12; group 2, treated with Interlac Pro-D; group 3, administered with Interlac; and finally, group 4 as the control group. During the adaptation period lasting 7 days, the rats were given the prophylactic deworming medication albendazole at a dosage of 30 mg/kg body weight (b.w.) orally. This was aimed at ensuring the health of the rats and eliminating any parasitic infestation before commencing the research. The 60 rats were housed in 30 cages, with each cage accommodating 2 rats. The cages were plastic boxes measuring 30 × 40 cm, equipped with wood shavings as bedding. The rat housing was situated within a specially regulated room maintained at a temperature of 25 ±2°C, with humidity levels set at 55 ±10% and a lighting cycle of 12 h of bright light, followed by 12 h of darkness.

Wound model

Under sterile conditions, surgical wounds were created on the rats' palates. Anesthesia for the experimental animals was induced by an intraperitoneal (i.p.) injection of Ketamine-Hameln® as the primary anesthetic, dosed at 95 mg/kg b.w. Additionally, xylazine was administered at 5 mg/kg b.w. to promote muscle relaxation and prolong the anesthesia. Both medications were injected into the rat's abdominal area, using a 1-cc syringe equipped with a 30G needle, allowing approx. 5 min for the drugs to take effect. Following successful anesthesia, the rat's palate was sterilized using gauze, forceps and 70% alcohol. A 5-mm diameter punch biopsy was then performed at the midline of the rat's hard palate to create a wound, followed by swabbing the wound site until bleeding ceased. Notably, no analgesics or antibiotics were administered to the rats after the creation of the wound.

Application of treatment

After creating the wounds on the rats' palates, the test groups were administered probiotics in powdered form, applied evenly to the wounds at a dosage of 4 mg, using a cotton pellet. Subsequently, a mucosal patch, Curatick™, was placed over the wounds. In the control group, only the mucosal patch was applied directly onto the wounds. This treatment was administered daily throughout the experimental period, and the mucosal patch was replaced daily. Then, 5 rats from each group were anesthetized and sacrificed on days 3, 7 and 14, using the exsanguination technique.

Observation of wound healing

Tissue samples were excised and fixed in Bouin's fixative solution for 24 h. Subsequently, they were sectioned into pieces measuring 2 × 1 mm with a thickness of 2 mm, using a No. 12 surgical blade knife. Dehydration followed using 100% alcohol before embedding the tissue blocks in paraffin of a temperature of 70°C within a base mold. After setting, the tissues underwent cryotome sectioning and were placed on glass slides for subsequent staining. The excised wound areas were then fixed and subjected to histopathological examination involving hematoxylin and eosin (H&E) staining, as well as Masson's trichrome staining. Quantitative analysis of collagen density, angiogenesis, re-epithelialization, the wound area, and the wound length was performed utilizing the ImageJ software (https://imagej.net/ij/index.html).

The method employed for assessing collagen density areas involved Masson's trichrome staining, with the areas being identified by distinctive bluish coloration, and characterized by thick, wavy cytoplasm with transverse fibers and the absence of a nucleus. Quantification of the collagen density area [mm^2] was accomplished by determining the total collagen area relative to the overall wound area in 6 distinct regions. Angiogenesis, visualized through H&E staining, showcased purple endothelial cells and red-colored erythrocytes. The measurement of angiogenesis was based on the total count of blood vessels identified in the 6 specified regions. The microscopic wound length was measured using the H&E-stained slides by calculating the distance between both wound edges, relying on the examination of the epithelial tissue under a microscope with ×4 magnification. The wound area was measured macroscopically by calculating the clinical wound area based on the clinical photographs of the rat wounds. A ruler was placed next to the wound in the photographs as a size reference, which was then used to measure the wound area accurately. Finally, the re-epithelialization rate was computed using a specific formula (Equation 1):

$$\text{re-epithelialization rate} = \frac{S_t}{S_0} \times 100\% \quad (1)$$

where:
S_t – residual wound area at the indicated time; and
S_0 – initial wound area.

Blinding

Blinding was implemented during the allocation, outcome assessment and data analysis phases of the study. This approach was used to minimize bias, and ensure that the allocation of participants, the evaluation of outcomes and the interpretation of data remained objective and uninfluenced by the knowledge of the treatment groups.

Statistical analysis

The data was statistically analyzed using the IBM SPSS Statistics for Windows, v. 26.0 (IBM Crop., Armonk, USA). The normality of data distribution was tested using the Shapiro–Wilk test ($p > 0.05$), and the multivariate analysis of variance (MANOVA) with Tukey's post hoc honestly significant difference (HSD) was performed to compare differences between the groups and across the necropsy days.

Results

The data pertaining to collagen density, angiogenesis, the re-epithelialization rate, the wound area, and the wound length is displayed in Table 1. In a descriptive analysis of the collagen density variable, it was observed that there was an increase in collagen density across all groups, corresponding directly with the necropsy/observation days. The group of rats serving as controls and necropsied on the 3rd day exhibited the lowest collagen density value, whereas the group treated with the probiotic Interlac and necropsied on the 14th day showed the highest collagen density value, suggesting enhanced wound healing over time (Fig. 1). This increase in collagen density corresponds with the body's natural healing process, where collagen deposition plays a critical role in strengthening the wound.

Regarding the dependent variable angiogenesis, there is a pattern of an initial increase followed by a slight decline; the lowest angiogenesis values appeared on day 3, gradually increasing by day 7, and slightly decreasing by day 14 (Fig. 2). This trend likely reflects the peak of new blood

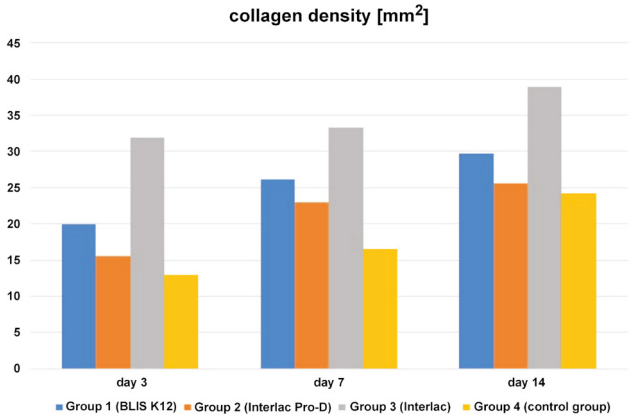

Fig. 1. Comparison of collagen density [mm^2] between the study groups over 3 time points (days 3, 7, and 14)

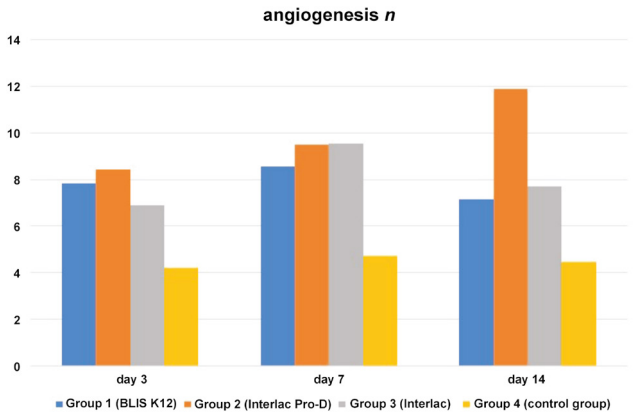

Fig. 2. Comparison of angiogenesis between the study groups over 3 time points (days 3, 7, and 14)

Table 1. Collagen density, angiogenesis, re-epithelialization rate, wound area, and wound length for all groups across various necropsy days

Group	Necropsy day	Collagen density [mm^2]	Angiogenesis *n*	Re-epithelialization rate [%]	Wound area [mm^2]	Wound length [mm]
Group 1	day 3	19.95 ±1.40	7.85 ±1.04	71.88 ±4.65	4.07 ±0.25	3.35 ±0.60
	day 7	26.07 ±2.51	8.55 ±1.43	78.31 ±16.85	2.47 ±0.98	2.74 ±0.36
	day 14	29.65 ±6.88	7.15 ±0.53	90.68 ±18.64	0.23 ±0.45	1.17 ±0.45
Group 2	day 3	15.49 ±3.20	8.45 ±2.11	57.77 ±8.16	3.92 ±2.39	4.16 ±0.36
	day 7	22.92 ±1.27	9.50 ±1.37	67.15 ±16.05	1.63 ±1.40	3.63 ±1.08
	day 14	25.54 ±1.27	11.90 ±2.31	94.26 ±11.48	0.22 ±0.43	3.18 ±0.59
Group 3	day 3	31.92 ±9.49	6.90 ±1.62	56.30 ±11.25	3.55 ±0.26	3.64 ±1.19
	day 7	33.30 ±9.14	9.55 ±4.75	87.46 ±15.63	0.89 ±1.35	2.46 ±0.49
	day 14	38.86 ±4.47	7.70 ±2.87	82.06 ±21.53	0.55 ±0.68	2.14 ±0.72
Group 4	day 3	12.98 ±4.58	4.20 ±0.59	42.95 ±7.33	7.76 ±2.49	4.37 ±1.06
	day 7	16.53 ±2.80	4.70 ±0.58	45.17 ±6.27	5.24 ±6.49	2.90 ±0.59
	day 14	24.14 ±3.41	4.45 ±0.68	43.61 ±10.58	2.00 ±0.91	6.77 ±1.21

Data presented as mean ± standard deviation (*M* ±*SD*).
Groups: group 1 – BLIS K12; group 2 – Interlac Pro-D; group 3 – Interlac; group 4 – control.

vessel formation necessary for tissue repair, and subsequent stabilization as healing progresses. The Interlac Pro-D group exhibited the highest angiogenesis by day 14, indicating a robust vascular response that supports tissue regeneration.

The dependent variable re-epithelialization rate logically shows an inverse relationship with the dependent variables wound area and wound length. Initially, a low re-epithelialization rate is expected, increasing gradually in subsequent days (Fig. 3). Conversely, in the early days, the wound area and the wound length are expected to be large, gradually decreasing in the following days (Fig. 4 and 5). For the re-epithelialization rate, the lowest value was in the control group necropsied on the 3rd day, while the highest was in the group of rats treated with the probiotic Interlac Pro-D and necropsied on the 14th day. Regarding the wound area, the lowest value was in the group of rats treated with the probiotic Interlac Pro-D and necropsied on the 14th day, while the highest was in the control group necropsied on the 3rd day. As for the wound length, the lowest value was in the group of rats treated with the probiotic BLIS K12 and necropsied on the 14th day, while the highest was in the control group necropsied on the 14th day.

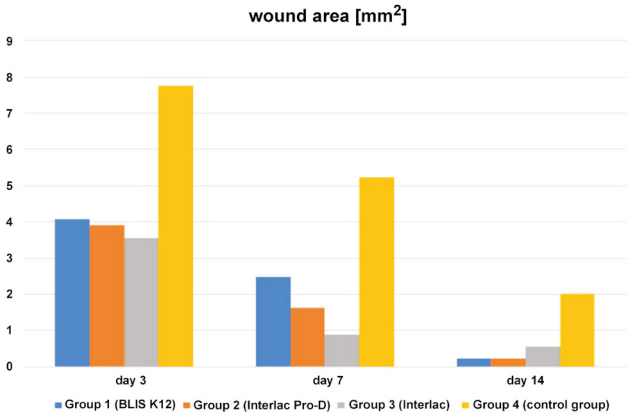

Fig. 4. Comparison of the wound area [mm²] between the study groups over 3 time points (days 3, 7, and 14)

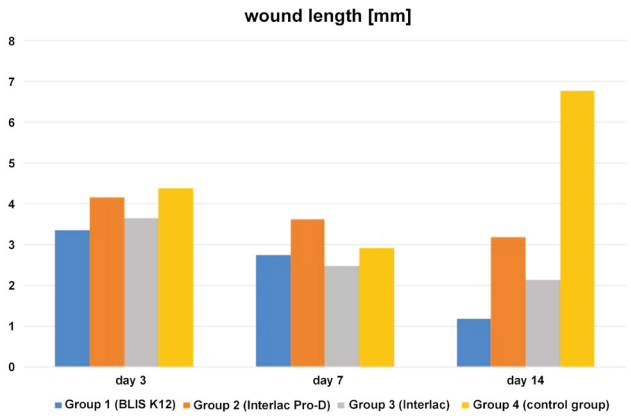

Fig. 5. Comparison of the wound length [mm] between the study groups over 3 time points (days 3, 7, and 14)

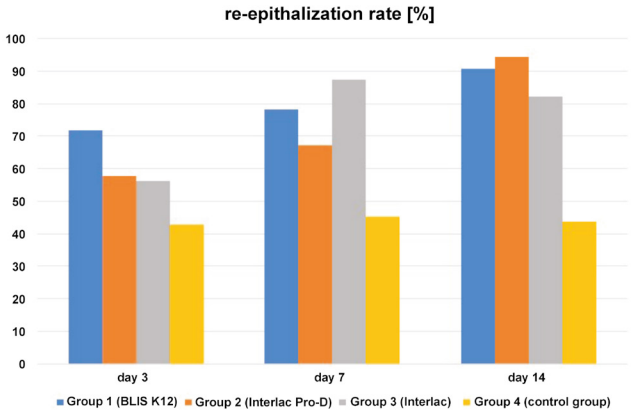

Fig. 3. Comparison of the re-epithelization rate [%] between the study groups over 3 time points (days 3, 7, and 14)

The Shapiro–Wilk test indicated that the data was normally distributed ($p > 0.05$). To compare the wound healing variables (collagen density, angiogenesis, the rate of re-epithelialization, the wound area, and the wound length) between the groups and across the necropsy days, MANOVA with Tukey's post hoc HSD test was employed. The results of these analyses are presented in Tables 2–4.

Table 2 presents the results of the MANOVA test aimed at evaluating the effects of 2 independent variables, namely the groups and the necropsy days, on 5 dependent variables: collagen density; angiogenesis; the re-epithelialization rate; the wound area; and the wound length. The analysis also assessed the interaction between the independent variables and their impact on the dependent variables. The MANOVA test revealed that all the dependent variables were significantly affected by different groups. Additionally, with the exception of angiogenesis,

Table 2. Results of the MANOVA test with regard to differences in collagen density, angiogenesis, the re-epithelialization rate, the wound area, and the wound length between the groups and the necropsy days

Comparison		F	*p*-value
Between the groups	collagen density	25.026	0.000*
	angiogenesis	15.211	0.000*
	re-epithelialization rate	18.095	0.000*
	wound area	5.660	0.003*
	wound length	19.870	0.000*
Between the necropsy days	collagen density	14.175	0.000*
	angiogenesis	1.600	0.216
	re-epithelialization rate	9.451	0.001*
	wound area	13.189	0.000*
	wound length	5.822	0.006*
Interaction between the groups and the necropsy days	collagen density	0.405	0.063
	angiogenesis	1.251	0.173
	re-epithelialization rate	2.279	0.275
	wound area	0.415	0.065
	wound length	10.812	0.643

* statistically significant.

all the dependent variables show significant differences across different necropsy days. However, the interaction between the groups and the necropsy day did not have a significant effect on any of the dependent variables. The lack of a significant interaction between the groups and the necropsy days suggests that while each factor individually affected the healing outcomes, their combined influence did not further alter the patterns observed.

Table 3 shows the results of Tukey's post hoc HSD test for intergroup comparisons. It reveals significant differences in all the dependent variables when comparing group 4 (the control group) with the other treated groups (groups 1–3), except for collagen density when comparing group 4 (control) with group 2 (Interlac Pro-D). Interestingly, when comparing between the treated groups, most of the dependent variables showed no significant differences. However, there are exceptions: collagen density showed significant differences when comparing group 1 (BLIS K12) with group 3 (Interlac), as well as group 2 (Interlac Pro-D) with group 3 (Interlac). Additionally, the wound length exhibited significant differences when comparing group 1 (BLIS K12) with group 2 (Interlac Pro-D), and when comparing group 2 (Interlac Pro-D) with group 3 (Interlac).

Table 3. Results of Tukey's post hoc HSD test for intergroup comparisons

Variable	Comparison		*p*-value
Collagen density	group 4 (control group)	group 1 (BLIS K12)	0.005*
		group 2 (Interlac Pro-D)	0.352
		group 3 (Interlac)	0.000*
	group 1 (BLIS K12)	group 2 (Interlac Pro-D)	0.245
		group 3 (Interlac)	0.000*
	group 2 (Interlac Pro-D)	group 3 (Interlac)	0.000*
Angiogenesis	group 4 (control group)	group 1 (BLIS K12)	0.001*
		group 2 (Interlac Pro-D)	0.000*
		group 3 (Interlac)	0.001*
	group 1 (BLIS K12)	group 2 (Interlac Pro-D)	0.072
		group 3 (Interlac)	0.995
	group 2 (Interlac Pro-D)	group 3 (Interlac)	0.119
Re-epithelialization rate	group 4 (control group)	group 1 (BLIS K12)	0.000*
		group 2 (Interlac Pro-D)	0.000*
		group 3 (Interlac)	0.000*
	group 1 (BLIS K12)	group 2 (Interlac Pro-D)	0.554
		group 3 (Interlac)	0.796
	group 2 (Interlac Pro-D)	group 3 (Interlac)	0.977
Wound area	group 4 (control group)	group 1 (BLIS K12)	0.025*
		group 2 (Interlac Pro-D)	0.010*
		group 3 (Interlac)	0.005*
	group 1 (BLIS K12)	group 2 (Interlac Pro-D)	0.983
		group 3 (Interlac)	0.916
	group 2 (Interlac Pro-D)	group 3 (Interlac)	0.992
Wound length	group 4 (control group)	group 1 (BLIS K12)	0.000*
		group 2 (Interlac Pro-D)	0.015*
		group 3 (Interlac)	0.000*
	group 1 (BLIS K12)	group 2 (Interlac Pro-D)	0.003*
		group 3 (Interlac)	0.747
	group 2 (Interlac Pro-D)	group 3 (Interlac)	0.036*

* statistically significant.

Table 4. Results of Tukey's post hoc HSD test for comparisons among the necropsy days

Variable	Comparison		*p*-value
Collagen density	necropsy day 3	necropsy day 7	0.035*
		necropsy day 14	0.000*
	necropsy day 7	necropsy day 14	0.026*
Angiogenesis	necropsy day 3	necropsy day 7	0.217
		necropsy day 14	0.392
	necropsy day 7	necropsy day 14	0.923
Re-epithelialization rate	necropsy day 3	necropsy day 7	0.035*
		necropsy day 14	0.000*
	necropsy day 7	necropsy day 14	0.212
Wound area	necropsy day 3	necropsy day 7	0.019*
		necropsy day 14	0.000*
	necropsy day 7	necropsy day 14	0.072
Wound length	necropsy day 3	necropsy day 7	0.005*
		necropsy day 14	0.120
	necropsy day 7	necropsy day 14	0.370

* statistically significant.

Table 4 displays the outcomes of Tukey's post hoc HSD test for comparisons among the necropsy days. The analysis indicated significant differences in collagen density between each pair of necropsy days (day 3 and day 7, day 3 and day 14, and day 7 and day 14). However, angiogenesis did not exhibit any significant differences between the various necropsy day pairs. Notably, the re-epithelialization rate and the wound area demonstrated significant differences between necropsy day 3 and day 7, as well as between necropsy day 3 and day 14, but not between necropsy day 7 and day 14. Similarly, the wound length displayed a significant difference between necropsy day 3 and day 7, but no significant differences were observed between necropsy day 3 and day 14, or between necropsy day 7 and day 14.

Discussion

Intraoral wounds, prone to bacterial contamination, often result in postoperative complications, such as wound dehiscence, infection and pain.[1] However, effectively managing these problems remains a challenge. While conventional treatment, like silver dressings, have been historically used, recent research questions their effec-

tiveness in promoting wound healing. Topical antibiotics, though frequently prescribed, can lead to resistance and skin irritation. Similarly, iodine has been associated with cellular toxicity. Conversely, topical probiotics offer broad antimicrobial activity with minimal systemic side effects and effectively inhibit biofilm formation. Both human and animal studies have shown promising results in using probiotics to enhance wound healing.[9]

Probiotics and their byproducts are gaining attention for their ability to help the body heal by regulating vital biological processes. Probiotics can be applied directly to the wound or taken orally.[6] However, using active ingredients systemically, either through the mouth or by injection, has drawbacks, like needing higher doses for effectiveness.[10] Recent studies show that applying probiotics directly to wounds can reduce bacterial growth and speed up tissue repair.[6] Based on these reasons, this study was undertaken utilizing probiotics applied topically.

The hypothesis that probiotics *L. reuteri* and *S. salivarius* K12 can enhance the wound healing process is based on their ability to inhibit the formation of pathogenic bacterial biofilms.[11,12] Biofilms often act as a physical barrier that hinders the migration of epithelial cells, which is essential for wound closure. This delayed epithelialization is a hallmark of chronic wounds, contributing to slower healing.[13] Additionally, the presence of biofilms can trigger an excessive inflammatory response, leading to prolonged inflammation and subsequent tissue damage. Chronic inflammation further impedes the wound healing process.[14] By inhibiting biofilm formation, *L.reuteri* and *S. salivarius* K12 may help reduce inflammation and promote faster epithelialization, ultimately enhancing the overall wound healing process.[13,14]

The use of antibiotics and analgesics can significantly influence the wound healing process. Antibiotics have been shown to suppress key inflammatory mediators, such as interleukin 1 beta (IL-1β), C-C motif chemokine ligand 2 (CCL2) and interferon alpha/beta (IFN-α/β), potentially leading to delayed wound healing. Moreover, although studies on non-steroidal anti-inflammatory drugs (NSAIDs) have yielded mixed results, it is well-established that NSAIDs inhibit the COX pathway, which plays a critical role in the proliferation phase of wound healing. To avoid potential bias, no antibiotics or analgesics were used in this study. The absence of these medications did not affect the wound healing process, as normal wound healing morphology was observed in both groups, and none of the rats exhibited signs of secondary infection. Ethical considerations were strictly adhered to, ensuring that the well-being of the rats was monitored closely throughout the study.[10]

One notable limitation of this study is its reliance on Sprague–Dawley rats as the experimental subjects, which may not fully reflect the physiological responses of humans. Despite the differences between humans and Sprague–Dawley rats in oral wound healing, this rat strain remains a valuable model for pilot studies. Their biological processes, particularly the fibroblast growth factor (FGF) pathways, closely resemble those of humans and provide useful insights into oral wound repair.[15,16] However, the inherent differences in metabolism, immune responses and the speed of wound healing between rats and humans can influence the outcomes, and these variations may affect the generalizability of findings to human subjects. While Sprague–Dawley rats are practical for understanding the basic mechanisms of tissue regeneration and wound contraction, further research on human subjects is crucial to confirm the applicability and clinical relevance of these results.[17]

Additionally, the presence of outliers in the data introduces the potential for bias in the analysis of certain variables, which could influence the interpretation of results. However, it is essential to emphasize our commitment to transparently and honestly reporting the data within this study, ensuring that the findings presented reflect the genuine observations made during the research process. Despite these limitations, this study lays valuable groundwork for future investigations aimed at understanding the potential effects of probiotics in wound healing.

Given the potential of probiotics and notwithstanding the aforementioned limitations, further research into probiotics as alternative, safe antimicrobial agents for wound care is still imperative. This study aims to explore the effects of commercially available probiotics on wound healing, particularly focusing on the proliferation phase. Assessment at the 3rd, 7th and 14th days post-application provides insight into their impact during the early, mid and late stages of proliferation, respectively.

The statistical analysis demonstrated that the interaction between the treatment groups and the necropsy days did not exert a significant effect on any of the dependent variables, encompassing collagen density, angiogenesis, the re-epithelialization rate, the wound area, and the wound length. This suggests that the influence of different treatment groups remained consistent throughout the evaluation days, indicating stable effects regardless of the assessment time. This underscores the reliability and consistency of the observed outcomes across the study timeline. However, the application of probiotics still significantly impacted these variables, as evidenced by the findings. Therefore, each variable will be discussed in detail in the following subsections.

Collagen density

Notably, group 4 (control group) exhibited the lowest collagen density across all evaluation days, suggesting a discernible effect of probiotics on this metric. Statistical analysis revealed significant differences between the control group and both group 1 (BLIS K12) and group 3 (Interlac), indicating their effectiveness in enhancing collagen density. Surprisingly, while group 2 (Interlac Pro-D)

demonstrated a tendency toward higher collagen density as compared to the control group, the difference was not statistically significant, suggesting limited efficacy in this context. Of particular interest is the observation that group 3 (Interlac) displayed the highest collagen density among all groups, with a statistically significant difference as compared to other groups. This underscores the superior effectiveness of Interlac in promoting collagen density during wound healing within the scope of this study.

A previous study conducted by Moraes et al. investigated the effects of both live probiotic and paraprobiotic forms of Interlac Pro-D on the collagen levels in periodontitis-induced rats.[18] Their findings indicated that the live probiotic Interlac Pro-D did not significantly alter collagen amounts, while the paraprobiotic forms containing non-viable *L reuteri* DSM 17938 and ATCC PTA 5289 showed a significant increase in collagen production.[18] This aligns with the results of our study, which also found that Interlac Pro-D did not significantly affect collagen density. Furthermore, previous research by Garcia et al. demonstrated improvement in the collagen levels in the probiotic-treated group using Interlac as compared to both the control group and those treated with systemic saline solution.[19] These findings parallel our own, highlighting the potential of Interlac in enhancing collagen production. Our research revealed that the topical application of *L. reuteri* DSM 17938 can enhance collagen production, although combining this strain with others, such as ATCC PTA 5289, may yield different results. Additionally, the potential influence of *S. salivarius* K12 on collagen deposition remains largely unexplored. However, the results of this study serve as a foundational or pilot project, providing insight into the notion that the probiotic *S. salivarius* K12 may indeed enhance collagen production in oral wound healing.

The findings of this study indicate a progressive increase in collagen density over time, particularly in the early, middle and late proliferation phases. Statistical analysis further corroborates these results, demonstrating significant differences in collagen density among all evaluation days. This observed temporal variation suggests a dynamic process of collagen deposition during the course of wound healing. The underlying mechanism driving this phenomenon can be attributed to the heightened fibroblast activity essential for tissue repair, particularly during the granulation phase.[20]

Angiogenesis

The benefits of probiotic bacteria for wound healing, including their potential role in promoting angiogenesis, have been suggested in previous research.[20] Angiogenesis, a crucial aspect of the wound healing process, involves a meticulously orchestrated series of biological events. These events facilitate the recruitment of inflammatory cells and the production of cytokines, matrix-degrading enzymes, and chemokines, ultimately leading to the formation of new capillaries from the existing ones.[21] This research provided a valuable perspective on the impact of various probiotics on angiogenesis during the proliferation phase of wound healing. The findings indicate that the angiogenesis levels in the control group were consistently lower as compared to all other groups across all evaluation days.

Statistical analysis further confirms substantial differences in angiogenesis between the control group and all other groups, suggesting that the topical application of the 3 types of probiotics investigated in this study does indeed influence angiogenesis. However, intriguingly, there were no statistically significant differences observed among the probiotic groups themselves. This implies that each of the 3 probiotics utilized in this research yields similar outcomes in enhancing angiogenesis during the proliferation phase of wound healing.

Consistent with the findings of this study, Zhou et al. reported in their research that the utilization of the probiotic *L. reuteri* led to an increase in the CD31 and vascular endothelial growth factor (VEGF) levels, both of which are proteins associated with angiogenesis.[22] Moreover, their study revealed that the application of metal-phenolic self-assembly shielded *L. reuteri* in a reinforced hydrogel further enhanced the expression of these proteins as compared to using *L. reuteri* alone. These results suggest that the incorporation of probiotics, particularly in novel delivery systems such as hydrogels, holds promise for augmenting angiogenesis and potentially improving wound healing outcomes.[22]

Limited in vivo and in vitro studies have suggested that certain probiotics, whether in live form or in a bacterial culture supernatant, can locally stimulate angiogenesis. They have been shown to induce the production of VEGF, thereby promoting endothelial cell growth and migration.[23] However, no study on the influence of *S. salivarius* K12 on angiogenesis has been found. This research can provide foundational knowledge that the effect of enhancing angiogenesis by *S. salivarius* K12 is comparable to that of *L. reuteri*, which has been previously researched.

Moreover, the lack of significant differences in angiogenesis across time points may be due to the nature of angiogenesis itself, which typically peaks early in the wound healing process, around days 3 to 7, when new blood vessels are formed to support tissue regeneration. By day 14, angiogenesis may have already stabilized, meaning that further changes in blood vessel formation are minimal, which could explain why no significant differences were observed at the later necropsy days.[10]

Re-epithelialization rate, wound area and wound length

The wound area and length exhibit an inverse relationship with the level of re-epithelialization and fibroblast

activity. Therefore, it is unsurprising to observe in this research that there is an inverse correlation between the re-epithelialization rate and both the wound area and the wound length. This is because a higher rate of re-epithelialization leads to smaller wound areas and lengths.[10] In this study, statistically significant differences were found in the re-epithelialization rate, the wound area and the wound length between the control group and the 3 treatment groups receiving different types of probiotics. This indicates that the topical application of the 3 types of probiotics on oral wounds can enhance re-epithelialization and expedite wound closure, as evidenced by the reduced wound area and shorter wound length over time.

This study revealed that all 3 types of probiotics showed no significant differences in enhancing re-epithelialization and reducing the wound area. However, notable variations emerged regarding the wound length, with significant differences observed between group 2 (Interlac Pro-D) and both group 1 (BLIS K12) and group 3 (Interlac). This suggests that Interlac Pro-D may be less effective in reducing wound length as compared to BLIS K12 and Interlac. Conversely, no statistical differences were observed in the effect of reducing the wound length between group 1 (BLIS K12) and group 3 (Interlac), indicating comparable effects for both these probiotics.

The difference in outcomes between the wound area and length measurements could be due to the methodology employed for wound assessment. The wound length is determined by measuring the longest linear dimension within the wound, which may not offer such a precise measurement as in the case of the wound area in fully capturing the overall dimensions of the wound. This discrepancy arises because, even if the wound area seems small, variations in the wound width could result in a greater length measurement.[24]

Moysidis et al. in their study concluded that their experimental findings support the notion that the manipulation of the wound environment using beneficial bacteria, such as probiotics, positively impacts the healing process.[25] In their research, they utilized *Lactobacillus plantarum* UBLP-40, as well as a combination of *Lactobacillus rhamnosus* UBLR-58 and *Bifidobacterium longum* UBBL-64. The authors further stated that probiotics operated through various, potentially unique mechanisms that are specific to each strain, yet collectively lead to faster wound healing.[25] These findings align with our own research, which demonstrates that the topical application of 3 probiotic strains on oral wounds enhances re-epithelialization and expedites wound closure.

On the other hand, Öhnstedt et al. demonstrated the efficacy and tolerability of the novel drug candidate resulting from the significant development of *Limosilactobacillus reuteri* R2LC.[26] This strain has been genetically modified to encode human CXCL12 1 alpha, leading to the creation of the drug candidate ILP100. This innovative approach has facilitated the development of ILP100 in a freeze-dried formulation. Their research revealed that topical treatment with ILP100 significantly accelerated the healing process of full-thickness wounds in minipigs.[26]

Conclusions

In conclusion, the present study demonstrates the significant benefits of probiotics in enhancing wound healing across various timing of the proliferation phase. Through the analysis of 5 key aspects, including collagen density, angiogenesis, the re-epithelialization rate, the wound area, and wound length, our findings reveal a consistent positive impact of probiotic use. Utilizing 3 distinct types of probiotics, we observed improvement in all aspects of wound healing, from the early to late stages of proliferation. This underscores the potential of probiotics as effective agents in promoting wound repair and regeneration, offering promising avenues for enhancing clinical outcomes in wound management. Further research and clinical trials are warranted to elucidate the specific mechanisms underlying the therapeutic effects of probiotics, and to optimize their utilization in clinical practice.

Ethics approval and consent to participate

The study protocol was approved by the Animal Ethics Committee at the Faculty of Veterinary Medicine and Biomedical Sciences of IPB University (Institut Pertanian Bogor), Bogor, Indonesia (approval No. 108/KEH/SKE/IX/2023).

Data availability

The datasets supporting the findings of the current study are available from the corresponding author on reasonable request.

Consent for publication

Not applicable.

Use of AI and AI-assisted technologies

Not applicable.

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References

1. Raghavan SL, Panneerselvam E, Mudigonda SK, Raja KK. Protection of an intraoral surgical wound with a new dressing: A randomised controlled clinical trial. *Br J Oral Maxillofac Surg*. 2020;58(7):766–770. doi:10.1016/j.bjoms.2020.03.017
2. Rahman S, Nagur Karibasappa S, Mehta DS. Evaluation of the wound-healing potential of the kiwifruit extract by assessing its effects on human gingival fibroblasts and angiogenesis. *Dent Med Probl*. 2023;60(1):71–77. doi:10.17219/dmp/146635
3. Shah R, Domah F, Shah N, Domah J. Surgical wound healing in the oral cavity: A review. *Dent Update*. 2020;47(2):135–143. doi:10.12968/denu.2020.47.2.135
4. Toma AI, Fuller JM, Willett NJ, Goudy SL. Oral wound healing models and emerging regenerative therapies. *Transl Res*. 2021;236:17–34. doi:10.1016/j.trsl.2021.06.003
5. Kibe T, Koga T, Nishihara K, et al. Examination of the early wound healing process under different wound dressing conditions. *Oral Surg Oral Med Oral Pathol Oral Radiol*. 2017;123(3):310–319. doi:10.1016/j.oooo.2016.10.023
6. Tagliari E, Campos LF, Campos AC, Costa-Casagrande TA, de Noronha L. Effect of probiotic oral administration on skin. *Arq Bras Cir Dig*. 2019;32(3):e1457. doi:10.1590/0102-672020190001e1457
7. Han N, Jia L, Su Y, et al. *Lactobacillus reuteri* extracts promoted wound healing via PI3K/AKT/β-catenin/TGF β1 pathway. *Stem Cell Res Ther*. 2019;10(1):243. doi:10.1186/s13287-019-1324-8
8. Kang S, Jang EJ, Jo HM, et al. Effects of a topically applied oral wound dressing film on intraoral wound healing in rabbits. *In Vivo*. 2022;36(4):1745–1752. doi:10.21873/invivo.12887
9. Knackstedt R, Knackstedt T, Gatherwright J. The role of topical probiotics on wound healing: A review of animal and human studies. *Int Wound J*. 2020;17(6):1687–1694. doi:10.1111/iwj.13451
10. Ananda N, Ariawan D, Juniantito V. Effects of the *Hydnophytum formicarum* plant extract on collagen density, angiogenesis, wound length, and re-epithelialization in wound healing: Experimental study on rats. *Dent Med Probl*. 2022;59(1):67–73. doi:10.17219/dmp/140208
11. Widyarman AS, Ramajayanti T, Theodorea CF. Indonesian strain of *Lactobacillus reuteri* probiotic reduces the initial biofilm colonization. *Open Dent J*. 2020;14:544–553. doi:10.2174/1874210602014010544
12. Stašková A, Sondorová M, Nemcová R, Kačírová J, Maďar M. Antimicrobial and antibiofilm activity of the probiotic strain *Streptococcus salivarius* K12 against oral potential pathogens. *Antibiotics (Basel)*. 2021;10(7):793. doi:10.3390/antibiotics10070793
13. Metcalf DG, Bowler PG. Biofilm delays wound healing: A review of the evidence. *Burns Trauma*. 2013;1(1):5–12. doi:10.4103/2321-3868.113329
14. Davis SC, Li J, Gil J, et al. The wound-healing effects of a next-generation anti-biofilm silver Hydrofiber wound dressing on deep partial-thickness wounds using a porcine model. *Int Wound J*. 2018;15(5):834–839. doi:10.1111/iwj.12935.
15. Huang NC, Huang NC, Kang LY, et al. Enhanced diabetic rat wound healing by platelet-rich plasma adhesion zwitterionic hydrogel. *Ann Plast Surg*. 2024;92(Suppl 1):S2–S11. doi:10.1097/SAP.0000000000003796
16. Maddaluno L, Urwyler C, Werner S. Fibroblast growth factors: Key players in regeneration and tissue repair. *Development*. 2017;144(22):4047–4060. doi:10.1242/dev.152587
17. Weber B, Lackner I, Haffner-Luntzer M, et al. Modeling trauma in rats: Similarities to humans and potential pitfalls to consider. *J Transl Med*. 2019;17(1):305. doi:10.1186/s12967-019-2052-7
18. Moraes RM, Lescura CM, Milhan NV, Ribeiro JL, Silva FA, Anbinder AL. Live and heat-killed *Lactobacillus reuteri* reduce alveolar bone loss on induced periodontitis in rats. *Arch Oral Biol* 2020;119:104894. doi:10.1016/j.archoralbio.2020.104894
19. Garcia VG, Janjácomo Miessi DM, da Rocha TE, et al. The effects of *Lactobacillus reuteri* on the inflammation and periodontal tissue repair in rats: A pilot study. *Saudi Dent J*. 2022;34(6):516–526. doi:10.1016/j.sdentj.2022.05.004
20. Golkar N, Ashoori Y, Heidari R, et al. A novel effective formulation of bioactive compounds for wound healing: Preparation, in vivo characterization, and comparison of various postbiotics cold creams in a rat model. *Evid Based Complement Alternat Med*. 2021;2021:8577116. doi:10.1155/2021/8577116
21. Manzoor S, Wani SM, Mir SA, Rizwan D. Role of probiotics and prebiotics in mitigation of different diseases. *Nutrition*. 2022;96:111602. doi:10.1016/j.nut.2022.111602
22. Zhou C, Zou Y, Xu R, et al. Metal-phenolic self-assembly shielded probiotics in hydrogel reinforced wound healing with antibiotic treatment. *Mater Horiz*. 2023;10(8):3114–3123. doi:10.1039/d3mh00033h
23. Xu J, Chen C, Gan S, et al. The potential value of probiotics after dental implant placement. *Microorganisms*. 2023;11(7):1845. doi:10.3390/microorganisms11071845
24. Saiko G. The impact of the wound shape on wound healing dynamics: Is it time to revisit wound healing measures? In: Proceedings of the 14th International Joint Conference on Biomedical Engineering Systems and Technologies (BIOSTEC 2021) – Volume 2: BIOIMAGING:182–187. doi:10.5220/0010337601820187
25. Moysidis M, Stavrou G, Cheva A, et al. The 3-D configuration of excisional skin wound healing after topical probiotic application. *Injury*. 2022;53(4):1385–1393. doi:10.1016/j.injury.2022.02.006
26. Öhnstedt E, Tomenius HL, Frank P, Roos S, Vågesjö E, Phillipson M. Accelerated wound healing in minipigs by on-site production and delivery of CXCL12 by transformed lactic acid bacteria. *Pharmaceutics*. 2022;14(2):229. doi:10.3390/pharmaceutics14020229

Original paper

Optical properties of advanced lithium disilicate

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Abstract

Background. A variety of firing protocols are available for the IPS e.max lithium disilicate (LD) and can be used for new, 'advanced' LD (ALD). However, the impact of firing protocols on the optical properties of ALD is still unknown.

Objectives. The aim of the present study was to evaluate the color difference (ΔE_{00}), the translucency parameter (TP_{00}) and the whiteness index for dentistry (WID) for both LD glass ceramics after the processes of firing/glazing.

Material and methods. Fifty disk-shaped specimens, with a diameter of 10 mm and a thickness of 1.2 mm, were fabricated from IPS e.max CAD (LD; Ivoclar) and another 50 from CEREC Tessera™ (ALD; Dentsply Sirona). The specimens from each group were further divided into 5 subgroups ($n = 10$) according to the firing/glazing protocol applied: crystallization (c); one-step crystallization and glazing (cg); crystallization and refiring (c-r); two-step crystallization and glazing (c-g); or long-firing crystallization (lfc). The ΔE_{00}, TP_{00} and WID were assessed. The statistical analysis of ΔE_{00} was performed using the one-way analysis of variance (ANOVA) and Tukey's post hoc test, while TP_{00} and WID were analyzed with the two-way ANOVA and Tukey's post hoc test at a statistical significance level of 0.05. The cg groups were designated as the reference.

Results. The ANOVA showed that the firing procedures had no effect on ΔE_{00}, TP_{00} and WID in the case of LD. In addition, LD exhibited greater translucency and brightness as compared to ALD. For ALD, all color changes observed in relation to the reference firing protocol were clinically unacceptable. The ALD specimens which underwent 1 standard firing cycle showed higher TP_{00} and WID values than other ALD groups.

Conclusions. The choice of the firing protocol has no impact on the color, TP_{00} or WID of LD. Additionally, LD presents higher WID values than ALD, irrespective of the firing protocol used. Alternative firing protocols result in clinically unacceptable color variations when compared to the manufacturer-recommended protocol for ALD. Advanced LD is more sensitive to different firing protocols with regard to its optical properties, which makes the workflow less predictable in comparison with LD.

Keywords: CAD/CAM, optical properties, dental materials, glass ceramics

Introduction

In recent years, glass ceramics have become widely used due to their adequate bond strength, mechanical behavior and excellent optical properties.[1,2] For instance, the translucency of lithium disilicate (LD) is higher as compared to the majority of zirconia ceramics.[1] Therefore, LD is one of the most commonly used ceramics for anterior veneers, posterior inlays, onlays and overlays, crowns, and bridges.[3]

When considering the specific needs of each clinical case, the satisfaction of the patient depends on providing a functional restoration whilst mimicking natural tooth appearance. Translucency, along with color, texture, size, and shape, determine the appearance and optical properties of restorations.[4–6] These esthetic parameters are affected by various factors, including the thickness of the restoration, surface treatment, the firing temperature, the number of firing cycles, the type of substructure, and differences in the manufacturing process.[5] Despite the favorable characteristics of the well-known LD, its esthetic properties can be altered, depending on the firing/glazing protocols applied,[5,7–9] which reduces the predictability of the treatment outcome.

Consequently, alternative materials have been developed to address the limitations of LD.[10] The manufacturer of CEREC Tessera™ (Dentsply Sirona, Charlotte, USA) refers to this new material as 'advanced' LD (ALD), and claims ALD has increased its mechanical strength as compared to conventional LD while maintaining high esthetic parameters. Advanced LD consists of a zirconia-enriched glass matrix and lithium aluminum silicate (LAS) crystals called virgilite.[11] The material is indicated for single-unit crowns, inlays, onlays, and veneers. Despite sharing several indications with LD, the firing protocol differs. According to the manufacturer, the use of a glaze layer as a finishing protocol is a mandatory step to achieve the desired mechanical properties. Yet, the effects of using or not using this protocol on the esthetic parameters of color, translucency and whiteness remain unknown.

The color evaluation of a natural-looking tooth is not easy, since the internal build-up is layered and complex. In addition, visual color assessment can be subjective due to both psychological and physiological aspects.[12] Therefore, instruments such as colorimeters and spectrophotometers are commonly used to evaluate color changes in dental materials.[13] To calculate the color difference (ΔE_{00}), the International Commission on Illumination (Commission internationale de l'éclairage – CIE) recommends the use of the CIEDE2000 color difference formula, which is currently the standard and most commonly used equation in the dental field to quantify color.[12] To define translucency, specimens should be evaluated over a black-and-white background. The difference between the reflected colors (the translucency parameter – TP_{00}) provides a value corresponding to the human visual perception of translucency.[14] Another parameter determined to assess the esthetic performance of a dental material is the whiteness index for dentistry (WID), which is crucial in terms of the patient's demands.[15] Since ALD requires the application of a glaze layer, dentists and dental technicians need to know if and how this layer can affect the optical properties of the material in comparison with LD. Additionally, it is important to investigate the impact of different firing protocols and processing methods on the esthetic outcome.[16]

Therefore, the present study investigated the impact of different firing/glazing protocols on the optical properties of LD and ALD. The null hypotheses were as follows: different firing protocols would not affect the (1) color, (2) translucency and (3) WID of both LD and ALD, and (4) no difference would be observed between the tested materials.

Material and methods

In the present study, 2 different reinforced glass-ceramic systems were tested. Their brand names, composition and manufacturer information are summarized in Table 1.

Fifty disk-shaped (10 mm in diameter and 1.2 mm in thickness) LD and ALD specimens were fabricated from each ceramic in the A2 shade in accordance with the VITA® classical shade guide (VITA Zahnfabrik, Bad Säckingen, Germany). Then, the ceramic disks from each group were further divided into 5 subgroups (n = 10) according to the evaluated firing/glazing protocols: crystallization (c); one-step crystallization and glazing (cg); crystallization and refiring (c-r); two-step crystallization and glazing (c-g); or long-firing crystallization (lfc) (Fig. 1, Table 2). As indicated by the ALD manufacturer, the recommended firing protocol is crystallization with glazing in one step. This firing protocol was also adopted for LD as the reference.

The sample size was calculated using statistical software (OpenEpi, v. 3.01; https://www.openepi.com/Menu/OE_Menu.htm) to achieve 80% statistical power performed with a 95% confidence interval (*CI*). The mean (*M*) and standard deviation (*SD*) values from a previous report that evaluated the relative translucency of LD and ALD[17] were employed.

Table 1. Characteristics of the ceramic dental materials evaluated in the study

Material	Brand name	Chemical composition	Manufacturer
LD	IPS e.max CAD	SiO_2: 57–80% Li_2O: 11–19% K_2O: 0–13% P_2O_5: 0–11% ZrO: 0–8% ZnO_2: 0–8% coloring oxides: 0–8%	Ivoclar, Schaan, Liechtenstein
ALD	CEREC Tessera	$Li_2Si_2O_5$: 90% Li_3PO_4: 5% $Li_{0.5}Al_{0.5}Si_{2.5}O_6$ (virgilite): 5%	Dentsply Sirona, Charlotte, USA

LD – lithium disilicate; ALD – advanced lithium disilicate.

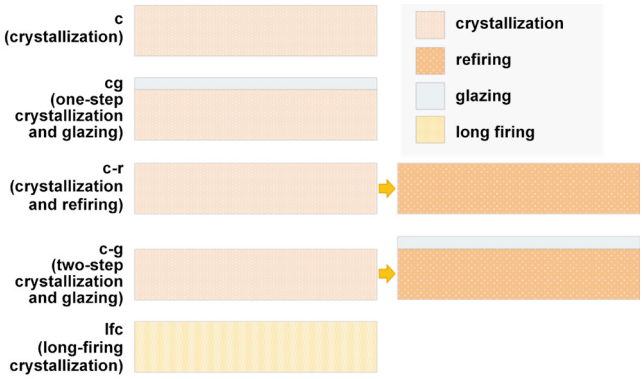

Fig. 1. Schematic illustration of the evaluated firing protocols, applied to both lithium disilicate (LD) and advanced lithium disilicate (ALD) specimens

Ceramic blocks of both materials were shaped into cylinders with a diamond drill of an internal diameter of 10 mm (Diamant Boart, Brussels, Belgium), connected to a bench drill (SBE 1010 Plus; Metabo, Nürtingen, Germany) under constant water cooling. All specimens were cut and polished with #800, #1,000 and #1,200 grit sandpaper (CarbiMet® SiC abrasive paper; Buehler, Lake Bluff, USA) in a polishing machine (EcoMet® 30; Buehler) until the final thickness of 1.20 ±0.09 mm was achieved. The disks were divided into 5 subgroups (n = 10) according to the firing protocol. The IPS e.max CAD Crystall Glaze Spray (Ivoclar) was used for LD, and the Universal Spray Glaze (Dentsply Sirona) was used for ALD. All firing cycles for the 5 subgroups were preprogrammed in a ceramic oven (Programat® P300; Ivoclar) according to the manufacturers' instructions.

Table 2. Distribution of the study groups according to the ceramic material and the firing protocol (as per the manufacturers' instructions)

Glass ceramic	Subgroup	Step 1	Step 2
IPS e.max CAD (LD)	LDc	LD firing crystallization: – closing time: 6 min – stand-by temperature: 403°C – heating rate: 60°C/min – firing temperature: 770°C – holding time: 10 s – heating rate: 30°C/min – firing temperature: 850°C – holding time: 10 min – vacuum 1: 550–770°C – vacuum 2: 770–850°C – long-term cooling: 700°C/min	–
	LDcg		–
	LDc-r		another LD firing crystallization
	LDc-g		IPS e.max CAD Crystall Glaze Spray firing: – closing time: 6 min – pre-heating temperature: 403°C – heating rate: 90°C/min – firing temperature: 820°C – holding time: 10 s – heating rate: 30°C/min – firing temperature: 840°C – holding time: 3 min – vacuum 1: 550–820°C
	LDlfc	LD long-firing crystallization: – closing time: 6 min – stand-by temperature: 403°C – heating rate: 60°C/min – firing temperature: 770°C – holding time: 10 s – heating rate: 30°C/min – firing temperature: 850°C – holding time: 13 min – vacuum 1: 550–770°C – vacuum 2: 770–850°C – long-term cooling: 700°C/min	–
CEREC Tessera (ALD)	ALDc	ALD firing crystallization: – closing time: 2 min – pre-heating temperature: 400°C – heating rate: 55°C/min – firing temperature: 760°C – holding time: 2 min – vacuum 1 and vacuum 2: off – long-term cooling: 0°C/min	–
	ALDcg		–
	ALDc-r		another ALD firing crystallization
	ALDc-g		Universal Spray Glaze firing
	ALDlfc	ALD long-firing crystallization: – closing time: 2 min – pre-heating temperature: 400°C – heating rate: 55°C/min – firing temperature: 760°C – holding time: 4 min – vacuum 1 and vacuum 2: off – long-term cooling: 0°C/min	–

Firing protocols: c – crystallization; cg – one-step crystallization and glazing; c-r – crystallization and refiring; c-g – two-step crystallization and glazing; lfc – long-firing crystallization.

The color was measured with a spectrophotometer (VITA Easyshade®; VITA Zahnfabrik), which provided the lightness (L*), red–green axis (a*) and yellow–blue axis (b*) values from the CIELab color space for the specimens against white, black and grey backgrounds, using box-blocking harsh lighting (Table 3). Before each measurement session, the spectrophotometer was calibrated according to the manufacturer's instructions. Each specimen was measured 3 times consecutively on each background, and the average was calculated to give the final value. The values obtained against the grey background were used to calculate the difference in color perception (ΔE_{00}), using the CIEDE2000 formula (Equation 1)[12]:

$$\Delta E_{00} = \left[\left(\frac{DL}{k_L * S_L}\right)^2 + \left(\frac{DC}{k_C * S_C}\right)^2 + \left(\frac{DH}{k_H * S_H}\right)^2 + R_T\left(\frac{DC}{k_C * S_C}\right) * \left(\frac{DH}{k_H * S_H}\right)\right]^{0.5} \quad (1),$$

where DL, DC and DH refer to the differences in lightness (L'), chroma (C') and hue (H') among the specimens. The weighting functions S_L, S_C, and S_H adjust the overall color difference to account for variations in the position of the color difference in the L*a*b* coordinates. Meanwhile, the parametric factors k_L, k_C and k_H serve as correction terms for the experimental conditions. RT is the rotation function that compensates for the interaction between the chroma and hue differences, specifically in the blue region.[13] For ΔE_{00}, the acceptability threshold (AT) is 1.77, while the perceptibility threshold (PT) is 0.81.[12] Thus, values under 0.81 can be considered irrelevant and negligible. Values between 0.81 and 1.77 are visible to the untrained eye, yet clinically acceptable, whereas values above 1.77 are clinically unacceptable.

Additionally, TP_{00} was calculated using the L*a*b* values from the white and black backgrounds (Equation 2):

$$TP_{00} = \left[\left(\frac{L'_B - L'_W}{k_L * S_L}\right)^2 + \left(\frac{C'_B - C'_W}{k_C * S_C}\right)^2 + \left(\frac{H'_B - H'_W}{k_H * S_H}\right)^2 + R_T\left(\frac{C'_B - C'_W}{k_C * S_C}\right) * \left(\frac{H'_B - H'_W}{k_H * S_H}\right)\right]^{0.5} \quad (2)$$

where:
TP_{00} – transparency parameter;
other parameters – as defined above;
W refers to the white background, and
B refers to the black background.

Table 3. L*a*b* parameters for the backgrounds used

Background	L* value	a* value	b* value
White	16.90 ±0.62	0.40 ±0.00	4.87 ±0.21
Black	1.23 ±0.19	24.60 ±0.94	45.63 ±6.30
Grey	13.00 ±1.72	0.50 ±0.08	3.97 ±0.49

Data presented as mean ± standard deviation (*M* ±*SD*).
L* – lightness; a* – value on the red–green axis; b* – value on the yellow–blue axis.

The TP_{00} values closer to 100 indicate more transparent specimens, and the TP_{00} values closer to 0 indicate more opaque specimens.[18]

The L*a*b* coordinates were obtained over the black background and WID was calculated according to the following equation (Equation 3)[15]:

$$WID = 0.511L^* - 2.324a^* - 1.100b^* \quad (3)$$

where:
WID – whiteness index for dentistry;
L* – lightness;
a* – value on the red–green axis; and
b* – value on the yellow–blue axis.

Higher WID values indicate whiter specimens, while lower WID values indicate less white specimens.

Statistical analysis

To evaluate the color differences for the ceramic groups, the visible and clinical acceptability thresholds were determined, and no further statistical analysis was performed. The cg groups were used as a point of reference in comparison to other methods, in accordance with the indication set forth by the ALD manufacturer. The ΔE_{00} was analyzed using the one-way analysis of variance (ANOVA) with $\alpha = 0.05$ and Tukey's post hoc test within each ceramic. The TP_{00} and WID were evaluated using the two-way ANOVA ($\alpha = 0.05$) and Tukey's post hoc test, based on the firing protocol and the ceramic used.

Results

Table 4 presents the mean L*, a* and b* values against the white, black and grey backgrounds. The mean values of L*, a* and b* on the grey background were used to calculate ΔE_{00} for different protocols within the same material group (Table 5). For LD, when considering LDcg as the reference for the firing protocol, there were no discernible color differences as compared to other protocols ($\Delta E_{00} < 0.81$; $p > 0.05$). However, when comparing the firing protocols with each other, there were perceptible and acceptable color differences between LDc-r and LDc-g, between LDc-g and LDc, and between LDc-g and LDlfc ($0.81 < \Delta E_{00} < 1.77$). For ALD, unacceptable color variations were encountered in all comparisons to ALDcg (the recommended crystallization protocol), with $\Delta E_{00} > 1.77$. Additionally, ALDc and ALDc-r were significantly different from each other ($p = 0.041$).

For TP_{00}, the two-way ANOVA showed significant differences with regard to the type of ceramic ($p < 0.001$), the firing protocol ($p < 0.001$), and their interaction ($p < 0.001$). Lithium disilicate was more translucent

Table 4. L*a* b* parameters for the backgrounds used according to the study groups

Background	Group	L* value	a* value	b* value
White	LDc	93.08 ±0.32	−0.83 ±0.07	14.20 ±0.35
	LDcg	92.91 ±0.42	−0.83 ±0.10	13.82 ±0.48
	LDc-r	92.99 ±0.20	−0.74 ±0.13	14.30 ±0.63
	LDc-g	92.80 ±0.33	−0.89 ±0.10	13.05 ±0.48
	LDlfc	93.06 ±0.43	−0.79 ±0.07	14.28 ±0.30
	ALDc	84.65 ±1.52	−0.67 ±1.45	37.85 ±5.43
	ALDcg	85.64 ±1.72	−0.88 ±1.87	37.30 ±7.09
	ALDc-r	85.91 ±1.20	4.93 ±1.90	52.63 ±4.82
	ALDc-g	87.10 ±0.55	4.09 ±2.19	50.14 ±5.73
	ALDlfc	85.63 ±0.56	2.76 ±2.12	48.54 ±5.95
Black	LDc	75.56 ±0.24	−1.67 ±0.05	8.50 ±0.23
	LDcg	75.24 ±0.38	−1.66 ±0.07	8.21 ±0.40
	LDc-r	75.24 ±0.46	−1.56 ±0.10	8.63 ±0.39
	LDc-g	74.99 ±0.52	−1.68 ±0.07	7.67 ±0.35
	LDlfc	75.36 ±0.26	−1.62 ±0.06	8.52 ±0.26
	ALDc	70.72 ±2.14	−4.72 ±1.23	24.13 ±5.67
	ALDcg	71.35 ±2.81	−4.81 ±1.47	23.54 ±6.88
	ALDc-r	74.61 ±0.85	0.16 ±1.78	40.16 ±5.16
	ALDc-g	75.32 ±1.09	−0.64 ±2.07	37.25 ±6.67
	ALDlfc	73.52 ±1.03	−1.80 ±1.72	34.93 ±5.88
Grey	LDc	86.31 ±0.29	−1.19 ±0.05	12.07 ±0.32
	LDcg	86.07 ±0.21	−1.18 ±0.09	11.69 ±0.53
	LDc-r	85.99 ±0.28	−1.15 ±0.10	12.10 ±0.48
	LDc-g	85.89 ±0.34	−1.25 ±0.09	10.97 ±0.46
	LDlfc	86.26 ±0.38	−1.16 ±0.07	12.15 ±0.24
	ALDc	79.04 ±1.67	−2.13 ±1.33	32.57 ±5.40
	ALDcg	79.97 ±1.95	−2.27 ±1.66	32.14 ±6.75
	ALDc-r	80.99 ±1.16	2.93 ±1.85	46.85 ±5.43
	ALDc-g	82.25 ±0.60	2.20 ±2.06	44.35 ±5.86
	ALDlfc	80.49 ±0.44	1.09 ±1.94	43.27 ±5.71

Data presented as *M* ±*SD*.

(12.40 ±0.07) than ALD (10.76 ±1.41). Regarding the firing protocol, the highest mean TP_{00} values were found in the cg (12.38 ±0.03) and c (12.23 ±0.08) groups, followed by the lfc (11.32 ±1.55), c-g (11.11 ±1.94) and c-r (10.88 ±2.20) groups. Considering the interaction of factors, no statistical differences in translucency were observed between the LD firing protocols. In the case of ALD, the cg and c groups showed the highest translucency, showing similar values. Protocols lfc, c-g and c-r showed the lowest values, with no significant differences between them. The statistical outcomes are summarized in Table 6 and graphically depicted in Fig. 2.

The two-way ANOVA revealed differences in terms of WID, based on the type of ceramic, the firing protocol, and the interaction of factors ($p < 0.001$ in all cases). The LD specimens exhibited greater whiteness

Table 5. Color difference from shade A2 according to the CIELab color space (ΔE) and CIEDE2000 (ΔE_{00}) with regard to the ceramic material and the firing protocol

Group	ΔE					ΔE_{00}					Group	ΔE					ΔE_{00}				
	LDcg	LDc	LDc-r	LDc-g	LDlfc	LDcg	LDc	LDc-r	LDc-g	LDlfc		ALDcg	ALDc	ALDc-r	ALDc-g	ALDlfc	ALDcg	ALDc	ALDc-r	ALDc-g	ALDlfc
LDcg	–	0.67 ±0.53^{A}	0.73 ±0.38^{A}	1.06 ±0.49^{A}	0.71 ±0.50^{A}	–	0.47 ±0.34^{A}	0.49 ±0.27^{A}	0.73 ±0.34^{A}	0.48 ±0.33^{A}	ALDcg	–	5.46 ±5.33^{A}	15.85 ±7.78^{B}	13.40 ±8.95^{AB}	13.06 ±7.86^{AB}	–	2.74 ±2.47^{A}	6.90 ±3.37^{B}	6.02 ±3.92^{AB}	5.68 ±3.47^{AB}
LDc	0.67 ±0.53^{A}	–	0.56 ±0.33	1.24 ±0.45	0.48 ±0.28	0.47 ±0.34^{A}	–	0.39 ±0.21	0.84 ±0.32	0.34 ±0.17	ALDc	5.46 ±5.33^{A}	–	15.51 ±7.60	14.59 ±7.10	11.36 ±6.50	2.74 ±2.47^{A}	–	6.69 ±3.03	6.51 ±3.12	4.90 ±2.72
LDc-r	0.73 ±0.38^{A}	0.56 ±0.33	–	1.20 ±0.53	0.62 ±0.29	0.49 ±0.27^{A}	0.39 ±0.21	–	0.82 ±0.36	0.43 ±0.19	ALDc-r	15.85 ±7.78^{B}	15.51 ±7.60	–	6.48 ±3.89	6.37 ±4.44	6.90 ±3.37^{B}	6.69 ±3.03	–	2.70 ±1.49	2.70 ±1.54
LDc-g	1.06 ±0.49^{A}	1.24 ±0.45	1.20 ±0.53	–	1.29 ±0.52	0.73 ±0.34^{A}	0.84 ±0.32	0.82 ±0.36	–	0.88 ±0.37	ALDc-g	13.40 ±8.95^{AB}	14.59 ±7.10	6.48 ±3.89	–	8.87 ±5.10	6.02 ±3.92^{AB}	6.51 ±3.12	2.70 ±1.49	–	3.81 ±1.80
LDlfc	0.71 ±0.50^{A}	0.48 ±0.28	0.62 ±0.29	1.29 ±0.52	–	0.48 ±0.33^{A}	0.34 ±0.17	0.43 ±0.19	0.88 ±0.37	–	ALDlfc	13.06 ±7.86^{AB}	11.36 ±6.50	6.37 ±4.44	8.87 ±5.10	–	5.68 ±3.47^{AB}	4.90 ±2.72	2.70 ±1.54	3.81 ±1.80	–

Data presented as *M* ±*SD*. Different capital letters show statistically significant differences between the groups (ANOVA, Tukey's post hoc test; $p < 0.05$). Considered thresholds: 50:50% perceptibility threshold (PT) – ΔE = 1.22, ΔE_{00} = 0.81; 50:50% acceptability threshold (AT) – ΔE = 2.66, ΔE_{00} = 1.77.

Table 6. Translucency parameter according to the CIELab color space (TP) and CIEDE2000 (TP_{00}), and the whiteness index for dentistry (WID) with regard to the ceramic material and the firing protocol

Group	TP	TP_{00}	WID
LDcg	18.56 ±0.57^{BC}	12.40 ±0.39^{A}	34.21 ±0.86^{A}
LDc	18.44 ±0.39^{BC}	12.29 ±0.26^{A}	33.87 ±0.57^{A}
LDc-r	18.65 ±0.43^{BC}	12.44 ±0.31^{A}	33.50 ±0.96^{A}
LDc-g	18.62 ±0.34^{BC}	12.48 ±0.26^{A}	35.13 ±0.75^{A}
LDlfc	18.63 ±0.35^{BC}	12.42 ±0.22^{A}	33.69 ±0.64^{A}
ALDcg	20.25 ±1.01^{A}	12.36 ±1.54^{A}	4.78 ±11.31^{B}
ALDc	19.98 ±0.83^{A}	12.17 ±1.18^{A}	3.18 ±8.69^{B}
ALDc-r	17.56 ±1.12^{C}	9.33 ±0.68^{B}	−25.45 ±9.97^{C}
ALDc-g	18.10 ±1.42^{BC}	9.74 ±1.13^{B}	−20.15 ±11.41^{C}
ALDlfc	18.81 ±0.75^{B}	10.23 ±1.03^{B}	−16.05 ±11.63^{C}

Data presented as *M* ±*SD*. Different capital letters show statistically significant differences between the groups (ANOVA, Tukey's post hoc test; $p < 0.05$).

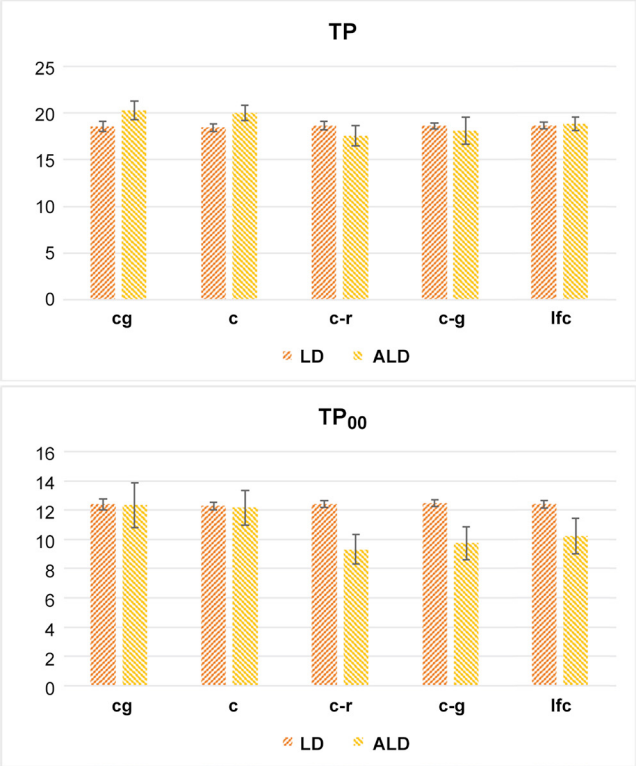

Fig. 2. Column charts of the translucency parameter according to the CIELab color space (TP) and CIEDE2000 (TP_{00}) with regard to the ceramic material and the firing protocol

(34.08 ±0.64) than the ALD specimens (−10.74 ±13.85). The highest WID values could be noticed in groups cg (19.49 ±20.81) and c (18.52 ±21.70), while the lowest values were encountered in the lfc (8.82 ±35.16), c-g (7.49 ±39.09) and c-r (4.02 ±41.69) groups. Different protocols did not significantly affect the WID of LD. However, for ALD, the lfc, c-g and c-r exhibited darker values. The statistical analysis of the WID values is summarized in Table 6 and graphically depicted in Fig. 3.

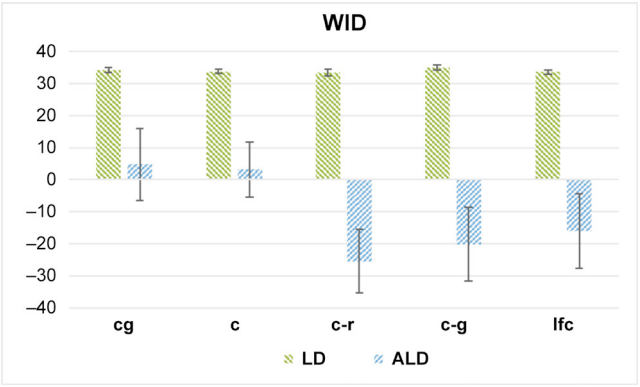

Fig. 3. Column chart of the whiteness index for dentistry (WID) with regard to the ceramic material and the firing protocol

Discussion

The present study investigated ΔE_{00}, TP_{00} and WID for 2 types of LD after subjecting the materials to different firing/glazing protocols. For LD, all color differences were either not visible or negligible and acceptable. All ALD groups presented visible and clinically unacceptable color differences in relation to the reference firing protocol (cg), which leads to the partial acceptance of the 1st hypothesis. It is well established that the color remains stable under different conditions for conventional LD.[17,19–21] In general, the mean ΔE_{00} values were below the perceivable threshold, indicating that the firing times and temperatures, combined or not with glazing, do not affect the color of LD.[1] For ALD, all ΔE_{00} values were clinically unacceptable when compared to the reference group, indicating that ALD is highly susceptible to the evaluated firing protocols. When observing the alternative firing protocols, the highest ΔE_{00} values relative to the reference protocol were observed for the c-r group, which underwent 2 standard firing cycles without glazing.

Considering translucency, LD exhibited comparable behavior across different firing protocols, indicating that the processing protocol also does not affect translucency, which disagrees with previous literature.[19] Conversely, the translucency of the ALD samples was affected, which partially supports the 2nd hypothesis. In a study by Miranda et al., translucency changes occurred in most LD groups after more than 2 firing sessions,[20] which exceeds the number of sessions conducted in the present study. On the contrary, ALD has only recently become available, and variations among different firing protocols are still being verified.[22] Lithium disilicate showed higher TP_{00} values as compared to ALD, which differs from a previous study that reported similar and higher relative TP_{00} for LD and ALD.[17] The authors used similar specimen dimensions to those utilized in the present study; however, the measurement was performed using a different device. The authors claimed that different chemical composition and crystalline structures had a greater influence on translucency.[17]

The present results corroborate the existing literature, indicating that the grain size also has a considerable effect on translucency[23]; LD with larger crystals than ALD,[24,25] exhibits greater translucency.[23]

Similar WID values were found for the LD groups, while the ALD groups presented differences among the firing protocols, which leads to the partial acceptance of the 3rd hypothesis.

When comparing both materials, variations in translucency and WID were noted, rejecting the 4th hypothesis. The WID for ALD presented statistically significant differences as compared to LD, regardless of the firing protocol.

A similar effect in terms of color changes can be observed for TP_{00} and WID, wherein the ALDc-r group exhibited lower mean values as compared to the ALDcg group. It was previously reported that the crystallization temperature can alter the optical properties of the material[8] and ALD appeared to be more susceptible to the firing protocols than LD. There is a wider variety of firing protocols for LD offered by the manufacturer: 1 firing cycle without glazing (c); 1 firing cycle with glazing (cg); and 1 crystallization cycle followed by another firing cycle with glazing (c-g). It seems that this variety of firing protocols cannot be implemented in ALD if its optical properties are taken into consideration. When compared to the protocol indicated by the manufacturer (cg), the color difference of the c-g specimens was greater. However, no statistical difference was observed in comparison with the c-r protocol. This suggests that the glaze serves as a protection layer against optical degradation in case of firing cycles longer than the one indicated by the manufacturer of ALD. Similar behavior could be noticed regarding the translucency and WID parameters. The refiring of ceramics can result in alterations to their mechanical and optical properties.[22,26] Moreover, esthetic degradation is notable relative to groups with a single standard firing protocol for the ALD samples.

Nevertheless, for the ALD samples, crystallization alone (c) promoted the lowest ΔE_{00}, and did not result in significant differences in TP_{00} and WID when compared to the cg protocol. One similarity between these groups is that they underwent just one firing cycle at the same parameters, which seems to have had a less harmful effect on the esthetic outcome. It appears that ALD behaves better with a single firing cycle applied in accordance with the parameters set forth by the manufacturer, differing from those for the lfc group. The lfc protocol was used to observe whether the effects found for groups c-g and c-r would be withdrawn due to the elimination of the temperature drop between the firing cycles. This would eliminate material cooling, which is a relevant factor influencing the optical properties of the material.[26] Unacceptable color changes, and differences in the TP_{00} and WID values were observed upon comparison with the reference protocol, indicating that longer firing cycles than the one indicated by the manufacturer impair the optical properties of ALD, reinforcing the idea that ALD is highly susceptible to firing protocols. The WID is an important parameter, especially when considering restorations in esthetic areas and the necessity for matching the bleached teeth.[15] Translucency represents the degree of light transmittance through an object and is determined by its characteristics, such as the reflection and/or scattering of light and the absorption of radiation. Interestingly, a recent study has demonstrated a significant increase in roughness for LD and ALD when applying a glaze layer, which would be expected to affect the esthetic parameters.[22] In the present study, however, the glaze did not seem to be a determinant of color changes and translucency, contrary to the one-step glaze application protocol (cg), since another firing decreased translucency and WID.

When working in collaboration with a dental laboratory, it is common for the clinician to receive the final restoration without being aware of the firing protocol that has been employed. Moreover, the advent of new restorative materials has brought processing variations. For instance, some studies have advised additional firing for ALD to increase its strength and decrease or eliminate microfissures.[22,24] Ultimately, clinical decisions should be made based on the best scientific evidence available, the clinician's expertise and the patient's perceptions. Notably, the colorimeter device used to assess the color in the present study has an average accuracy of $\Delta E < 0.5$, and performs equally well in laboratory and clinical settings, making it suitable for clinically relevant interpretation.[27] The human eye starts to detect color differences at different values.[12] The perceptibility and acceptability thresholds for color differences are, therefore, a point of discussion in the literature. The conclusions derived from these values are subjective, given that color perception can be influenced by external stimuli, or even memory.[15] Furthermore, it is important to ensure that the documentation is well conducted, and that the patient's needs and expectations are taken into account.

One of the main physical aspects that influence the optical properties of the material is its surface roughness.[28] As reported by Lu et al., the surface roughness of LD and ALD was not affected by an additional firing cycle, but by glazing.[22] Although the 2nd firing cycle with glaze application impaired the fracture resistance of LD, the same strategy improved the mechanical behavior of ALD. In addition, scanning electron microscopy (SEM) indicated that refiring could not alter the surface morphology of the material, while glaze application led to smoother surfaces. Thus, it was suggested that the glassy phase on the ceramic could be partially molten during the 2nd firing, leading to better mechanical behavior, which implies the formation of different internal and superficial structures.[22] Nonetheless, what is beneficial for ALD in terms of its mechanical properties has proven to be a critical problem when optical properties are considered.

When examining its mechanical behavior, ALD benefited from the 2nd firing cycle with glaze application (the c-g protocol), whereas the manufacturer's protocol (cg – 1 firing cycle with glaze application) resulted in lower flexural strength, which was still superior to that obtained with a single firing cycle without glazing (c).[22,26] This study revealed that only the c protocol promoted similar translucency and whiteness, although this was accompanied by an unacceptable color change. Therefore, to maintain the optical properties of ALD indicated by the manufacturer, the cg protocol is advocated. Dental restorations require a balance between mechanical and optical properties. Thus, if a modification to the ALD firing protocol were to be implemented with the objective of improving mechanical properties, this would result in alterations to the optical properties of the restoration.

Limitations

Although the study provides valuable insights into the optical properties of ALD under various firing protocols, several limitations should be acknowledged. Firstly, the study did not investigate the potential influence of different kinds of surface treatment on the optical outcomes, which could have impacted the results.[29,30] Additionally, variations in the restoration thickness and design were not accounted for, which could have affected light transmission and color perception.[31] Moreover, the absence of fatigue testing limits the understanding of the long-term performance and durability of the materials under different firing conditions.[32] Furthermore, the study did not explore the effects of bonding to different substrates, which could have influenced the overall optical behavior of the restorations.[33] Addressing these limitations in future research would provide a more comprehensive understanding of the optical behavior of LD and ALD restorations. The use of only one measuring device (VITA Easyshade) and only one specimen thickness can also be considered as the limitations of this study. To imitate clinical use, varying disk thicknesses should be investigated. Future research should employ different measuring devices, and consider the use of esthetic characterization and pigment solutions.[34] Additionally, there are dipping glazes which can influence the studied outcomes, and researchers are encouraged to explore them in future studies.

Conclusions

Advanced LD is highly susceptible to changes in color, translucency and WID, whereas LD demonstrates stability in the evaluated optical properties. All alternative firing protocols for ALD result in clinically unacceptable color variations when compared to the manufacturer-recommended protocol (cg).

Ethics approval and consent to participate

Not applicable.

Data availability

The datasets supporting the findings of the current study are available from the corresponding author on reasonable request.

Consent for publication

Not applicable.

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References

1. Park JH, Bang HJ, Choi NH, Park EJ. Translucency and masking ability of translucent zirconia; comparison with conventional zirconia and lithium disilicate. *J Adv Prosthodont*. 2022;14(5):324–333. doi:10.4047/jap.2022.14.5.324
2. Aziz A, El-Mowafy O, Paredes S. Clinical outcomes of lithium disilicate glass-ceramic crowns fabricated with CAD/CAM technology: A systematic review. *Dent Med Probl*. 2020;57(2):197–206. doi:10.17219/dmp/115522
3. Li RWK, Chow TW, Matinlinna JP. Ceramic dental biomaterials and CAD/CAM technology: State of the art. *J Prosthodont Res*. 2014;58(4):208–216. doi:10.1016/j.jpor.2014.07.003
4. Eldwakhly E, Ahmed DRM, Soliman M, Abbas MM, Badrawy W. Color and translucency stability of novel restorative CAD/CAM materials. *Dent Med Probl*. 2019;56(4):349–356. doi:10.17219/dmp/111400
5. Revilla-León M, Sorensen JA, Nelson LY, Gamborena I, Yeh YM, Özcan M. Effect of fluorescent and nonfluorescent glaze pastes on lithium disilicate pressed ceramic color at different thicknesses. *J Prosthet Dent*. 2021;125(6):932–939. doi:10.1016/j.prosdent.2020.05.003
6. Tango RN, Todorović A, Stamenković D, Karasan DN, Sailer I, Paravina RD. Effect of staining and aging on translucency parameter of CAD-CAM materials. *Acta Stomatol Croat*. 2021;55(1):2–9. doi:10.15644/asc55/1/1
7. Nejatidanesh F, Azadbakht K, Savabi O, Sharifi M, Shirani M. Effect of repeated firing on the translucency of CAD-CAM monolithic glass-ceramics. *J Prosthet Dent*. 2020;123(3):530.e1–530.e6. doi:10.1016/j.prosdent.2019.10.028
8. Simba BG, Ribeiro MV, Alves MF, Vasconcelos Amarante JE, Strecker K, dos Santos C. Effect of the temperature on the mechanical properties and translucency of lithium silicate dental glass-ceramic. *Ceram Int*. 2021;47(7(Pt A)):9933–9940. doi:10.1016/j.ceramint.2020.12.137
9. Yılmaz K, Gonuldas F, Ozturk C. The effect of repeated firings on the color change of dental ceramics using different glazing methods. *J Adv Prosthodont*. 2014;6(6):427–433. doi:10.4047/jap.2014.6.6.427
10. Zhang Y, Vardhaman S, Rodrigues CS, Lawn BR. A Critical review of dental lithia-based glass-ceramics. *J Dent Res*. 2023;102(3):245–253. doi:10.1177/00220345221142755
11. Lu Y, Dal Piva AMdO, Tribst JPM, Feilzer AJ, Kleverlaan CJ. Does glaze firing affect the strength of advanced lithium disilicate after simulated defects? *Clin Oral Investig*. 2023;27(11):6429–6438. doi:10.1007/s00784-023-05246-1

12. Paravina RD, Ghinea R, Herrera LJ, et al. Color difference thresholds in dentistry. *J Esthet Restor Dent*. 2015;27(Suppl 1):S1–S9. doi:10.1111/jerd.12149
13. Ataol AS, Ergun G, Yayman M. Effects of the substructure thickness, the resin cement color and the finishing procedure on the color and translucency of zirconia-based ceramic restorations. *Dent Med Probl*. 2023;60(1):137–144. doi:10.17219/dmp/149356
14. Salas M, Lucena C, Herrera LJ, Yebra A, Della Bona A, Pérez MM. Translucency thresholds for dental materials. *Dent Mater*. 2018;34(8):1168–1174. doi:10.1016/j.dental.2018.05.001
15. del Mar Pérez M, Ghinea R, Rivas MJ, et al. Development of a customized whiteness index for dentistry based on CIELAB color space. *Dent Mater*. 2016;32(3):461–467. doi:10.1016/j.dental.2015.12.008
16. Leite Madruga CF, Dal Piva AMdO, Pereira GKR, Ferraz Caneppele TM, Valandro LF, Bottino MA. Sintering mode of a translucent Y-TZP: Effects on its biaxial flexure fatigue strength, surface morphology and translucency. *J Esthet Restor Dent*. 2022;34(8):1197–1205. doi:10.1111/jerd.12923
17. Demirel M, Diken Türksayar AA, Donmez MB. Translucency, color stability, and biaxial flexural strength of advanced lithium disilicate ceramic after coffee thermocycling. *J Esthet Restor Dent*. 2023;35(2):390–396. doi:10.1111/jerd.12960
18. Acar O, Yilmaz B, Altintas SH, Chandrasekaran I, Johnston WM. Color stainability of CAD/CAM and nanocomposite resin materials. *J Prosthet Dent*. 2016;115(1):71–75. doi:10.1016/j.prosdent.2015.06.014
19. Seyidaliyeva A, Rues S, Evagorou Z, Hassel AJ, Rammelsberg P, Zenthöfer A. Color stability of polymer-infiltrated-ceramics compared with lithium disilicate ceramics and composite. *J Esthet Restor Dent*. 2020;32(1):43–50. doi:10.1111/jerd.12525
20. Miranda JS, Barcellos AS, MartinelliLobo CM, Caneppele TM, Amaral M, Kimpara ET. Effect of staining and repeated firing on the surface and optical properties of lithium disilicate. *J Esthet Restor Dent*. 2020;32(1):113–118. doi:10.1111/jerd.12558
21. Arif R, Yilmaz B, Johnston WM. In vitro color stainability and relative translucency of CAD-CAM restorative materials used for laminate veneers and complete crowns. *J Prosthet Dent*. 2019;122(2):160–166. doi:10.1016/j.prosdent.2018.09.011
22. Lu Y, Dal Piva AMdO, Nedeljkovic I, Tribst JPM, Feilzer AJ, Kleverlaan CJ. Effect of glazing technique and firing on surface roughness and flexural strength of an advanced lithium disilicate. *Clin Oral Investig*. 2023;27(7):3917–3926. doi:10.1007/s00784-023-05014-1
23. Subaşı MG, Alp G, Johnston WM, Yilmaz B. Effect of thickness on optical properties of monolithic CAD-CAM ceramics. *J Dent*. 2018;71:38–42. doi:10.1016/j.jdent.2018.01.010
24. Phark JH, Duarte S Jr. Microstructural considerations for novel lithium disilicate glass ceramics: A review. *J Esthet Restor Dent*. 2022;34(1):92–103. doi:10.1111/jerd.12864
25. Lubauer J, Belli R, Peterlik H, Hurle K, Lohbauer U. Grasping the lithium hype: Insights into modern dental lithium silicate glass-ceramics. *Dent Mater*. 2022;38(2):318–332. doi:10.1016/j.dental.2021.12.013
26. Juntavee N, Uasuwan P. Influence of thermal tempering processes on color characteristics of different monolithic computer-assisted design and computer-assisted manufacturing ceramic materials. *J Clin Exp Dent*. 2019;11(7):e614–e624. doi:10.4317/jced.55869
27. Dozić A, Kleverlaan CJ, El-Zohairy A, Feilzer AJ, Khashayar G. Performance of five commercially available tooth color-measuring devices. *J Prosthodont*. 2007;16(2):93–100. doi:10.1111/j.1532-849X.2007.00163.x
28. Al-Thobity AM, AlOtaibi AM, Alhumaidan AE, et al. Impact of thermocycling on surface roughness, microhardness and optical properties of three different lithium disilicate ceramics. *Saudi Dent J*. 2022;34(7):589–595. doi:10.1016/j.sdentj.2022.08.004
29. Duarte Moura DM, Medeiros de Araújo AM, de Souza KB, Veríssimo AH, Tribst JPM, de Assunção E Souza RO. Hydrofluoric acid concentration, time and use of phosphoric acid on the bond strength of feldspathic ceramics. *Braz Oral Res*. 2020;34:e018. doi:10.1590/1807-3107bor-2020.vol34.0018
30. Dal Piva AMdO, Carvalho RL, Lima AL, Bottino MA, Melo RM, Valandro LF. Silica coating followed by heat-treatment of MDP-primer for resin bond stability to yttria-stabilized zirconia polycrystals. *J Biomed Mater Res B Appl Biomater*. 2019;107(1):104–111. doi:10.1002/jbm.b.34100
31. da Fonseca GF, de Andrade GS, Dal Piva AMdO, Tribst JPM, Souto Borges AL. Computer-aided design finite element modeling of different approaches to rehabilitate endodontically treated teeth. *J Indian Prosthodont Soc*. 2018;18(4):329–335. doi:10.4103/jips.jips_168_18
32. Leite Madruga CF, Bueno MG, Dal Piva AMdO, et al. Sequential usage of diamond bur for CAD/CAM milling: Effect on the roughness, topography and fatigue strength of lithium disilicate glass ceramic. *J Mech Behav Biomed Mater*. 2019;91:326–334. doi:10.1016/j.jmbbm.2018.12.037
33. Ausiello PP, Ciaramella S, Lanzotti A, et al. Mechanical behavior of Class I cavities restored by different material combinations under loading and polymerization shrinkage stress. A 3D-FEA study. *Am J Dent*. 2019;32(2):55–60. PMID:31094138.
34. Freitas JS, Brum Souza LF, Dellazzana FZ, et al. Advanced lithium disilicate: A comparative evaluation of translucency and fatigue failure load to other ceramics for monolithic restorations. *J Mech Behav Biomed Mater*. 2023;148:106192. doi:10.1016/j.jmbbm.2023.106192

Original paper

Influence of alcoholic and non-alcoholic beverages on the compressive strength of core build-up bulk-fill resin materials

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Abstract

Background. In the oral cavity, restorations and teeth are subject to occlusal stresses, as well as physiological and chemical wear processes. The consumption of alcoholic and non-alcoholic beverages is popular among adolescents, and this habit often persists into adulthood, contributing to tooth erosion. The erosive potential of these beverages is influenced by various factors, including the pH, acid type, buffer capacity, adhesion, chelating effect, and phosphate or calcium or fluoride content of the drink. Several studies have confirmed that the excessive consumption of alcoholic and non-alcoholic beverages reduces the microhardness and flexural strength of composites.

Objectives. The purpose of this study is to compare the compressive strength (CS) of 3 core build-up resin composite materials in the presence of different alcoholic and non-alcoholic beverages.

Material and methods. A total of 135 specimens (blocks measuring 5 mm × 4 mm) made from different composites (NexCore™, spee-dee™ build-up, and Ecosite Bulk Fill) were divided into 3 experimental groups ($n = 45$). The experimental liquids used in each group were artificial saliva, Kingfisher® beer (an alcoholic beverage) and Thums Up® (a non-alcoholic beverage), which were maintained for 24 h at 37°C. The compressive strength of the samples was measured with the use of a universal testing machine.

Results. Nexcore™ and Ecosite Bulk Fill exhibited the highest fracture resistance among the tested liquid mediums. The mean CS for NexCore™ was significantly higher after immersion in alcohol compared to saliva and non-alcoholic medium ($p < 0.0001$).

Conclusions. The immersion medium affects the CS of bulk-fill composites. Moreover, NexCore™ and Ecosite Bulk Fill composites exhibited substantial CS, while spee-dee™ build-up demonstrated the lowest CS among the tested media.

Keywords: compressive strength, beverages, composite

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Highlights

- The immersion medium affects the compressive strength (CS) of bulk-fill composites.
- NexCore™ demonstrated the highest CS values in alcoholic media.
- Ecosite Bulk Fill showed superior CS in artificial saliva.
- Spee-dee™ build-up exhibited the lowest CS values across all tested media.

Introduction

A root canal-treated tooth with extensive loss of the coronal tooth structure is susceptible to fracture.[1] A significant reduction in the remaining tooth tissue represents a major restorative challenge. It has been established that the quality of coronal restoration affects the outcome of endodontic therapy.[2] According to the guidelines and treatment standards established by the American Association of Endodontists (AAE), endodontically treated teeth (ETT) should be restored with a core build-up (optionally, a root canal post) and a full crown to ensure the longevity of the tooth–restoration complex.[3] Moreover, the quality of the coronal restoration is equally or more important for the outcome of endodontic treatment than the quality of the actual root canal treatment.[4] Therefore, root canal treatment cannot be successful without timely and adequate definitive restoration.[3,4] However, most of the ETT undergo full coverage crown restorations, which lead to the removal of tooth structure.[5,6] The restorations should always be designed conservatively with the aim of retaining as much sound residual tooth structure as possible.

In recent years, advancements in the field of adhesive restoration and a shift toward minimally invasive access cavity preparation have led to a preference for direct or indirect bonded restorations of ETT with intact proximal walls.[7] In clinical practice, direct restorations offer several advantages, including time efficiency, conservation of tooth structure, and cost efficiency compared to laboratory restoration.[8]

Light-cure or dual-cure composite resins are commonly used with or without posts as a restoration material for ETT. Dual-cure materials can be further categorized into those with higher viscosity (paste-like) or those more flowable.[9] The composites intended to be used as a core build-up demonstrate higher levels of inorganic filler content, increased molecular weight per reactive group and reduced shrinkage.[9,10] The resin core build-up can be executed through either the conventional multi-increment fill or the bulk-fill technique. Recent advancements in bulk-fill composites have enabled incremental filling of up to 4–5 mm, exhibiting adequate polymerization.[11,12] This enhancement can be attributed to the high color translucency of the composites, which increases the depth of cure, and a more innovative initiator system, which shortens the light-curing time.[13]

Within the oral cavity, both restorations and teeth are subject to occlusal stresses, as well as physiological and chemical wear processes. The consumption of alcoholic and non-alcoholic beverages is popular among adolescents, and the habit often persists into adulthood, contributing to tooth erosion.[14,15] The erosive potential of these beverages is influenced by various factors, including the pH, acid type (phosphoric acid or citric acid), buffer capacity, adhesion, chelating effect, and phosphate, calcium or fluoride content of the drink.[14,15] Several studies have confirmed that the excessive consumption of alcoholic and non-alcoholic beverages reduces the microhardness and flexural strength of composites.[14–18] A potential explanation for this phenomenon is that polymer materials absorb water, while coupling agents cause hydrolysis, resulting in the loss of chemical bonds between filler particles and the resin matrix.[14,17,19,20] Filler particles dislodge from the outer surface of the material, causing surface roughness and decreasing hardness. The effect on the resin matrix, the degradation of the resin–filler interface, and the inorganic fillers may also play a role in the reduction of surface hardness.[17–19]

Given the paucity of data on the influence of beverages on the compressive strength (CS) of core build-up materials, the authors decided to investigate this area. The aim of the study was to evaluate the CS of 3 core build-up materials after exposure to alcoholic and non-alcoholic liquids. The null hypothesis (H0) states that there is no difference in the CS of core build-up bulk-fill resin materials after immersion in alcoholic beverages compared to non-alcoholic beverages.

Material and methods

Sample preparation

The study evaluated 3 core build-up materials: NexCore™ (Meta Biomed, Cheongju-si, South Korea) (NC group), spee-dee™ build-up (PULPDENT Cooperation, Watertown, USA) (S-D group), and Ecosite Bulk Fill (DMG, Hamburg, Germany) (E group). The characteristics of the materials are presented in Table 1. In each study group (NC, S-D and E), 45 samples were prepared using cylindrical Teflon molds measuring 5 mm in diameter and 4 mm in thickness. The molds were filled

with core build-up material using a composite instrument (PF21; GDC Fine Crafted Dental Pvt. Ltd., Punjab, India).

Photoactivation was performed for 20 s, in accordance with the manufacturer's instructions, using a light-emitting diode (LED) curing unit (Bluephase; Ivoclar Vivadent, Schaan, Liechtenstein) operating in a continuous high-power mode. The light tip was positioned in contact with a glass slide placed on the top surface of the specimen. A calibrated LED radiometer (DB686 NANO; COXO Medical Instrument Co., Ltd., Foshan, China) was used to verify the light intensity output of the curing unit before each sample preparation. Next, the mold and any excess material were removed.

Storage solutions

The study groups were divided into 3 subgroups of 5 specimens each, based on the storage fluid. Within each subgroup, the samples were immersed in an experimental fluid: a saliva substitute (Wet Mouth; ICPA Health Products Ltd, Mumbai, India); Kingfisher® Strong Premium beer (United Breweries, Mangaluru, India) (an alcoholic beverage); or Thums Up® (Coca-Cola Company, Bengaluru, India) (a non-alcoholic beverage) (Table 2). Containers with specimens and solutions were secured to prevent evaporation and maintained at 37°C for 24 h.

Fig. 1. Sample placed in a universal testing machine (Model No. 8801; Instron, Norwood, USA)

Compressive strength calculation

Next, all samples were individually mounted on a computer control universal testing machine (Model No. 8801; Instron, Norwood, USA) with a loadcell of 5 kN (Fig. 1). The data was recorded using the WaveMatrix™3 Dynamic Testing Software and Bluehill Universal® software (Instron, Norwood, USA). All samples were subjected to a compressive load at the center of the machine, at a crosshead speed of 1 mm/min, until fracture of each specimen.

Table 1. Characteristics of the composite materials used in the study

Study group	Product name (manufacturer)	Type of material	Adhesive procedures	Composition	Filler load [wt%]	Filler size [µm]
NC	NexCore™ (Meta Biomed, Cheongju-si, South Korea)	dual-cure core build-up composite	etching/conditioning of enamel, followed by the universal adhesion procedure	barium glass, ytterbium (III) fluoride, pyrogenic silicic acid, Bis-GMA, TEGDMA	70–75	0.1–1.5
S-D	spee-dee™ build-up (PULPDENT Cooperation, Watertown, USA)	fluoride-releasing, dual-cure core build-up composite	self-adhesion/self-etching to dentin (etching and bonding agents are optional)	two-part uncured acrylate ester monomers, amorphous silica and glass (BPA/Bis-GMA-free)	54.5	0.012 (silica), 4 (glass)
E	Ecosite Bulk Fill (DMG, Hamburg, Germany)	light-cure core build-up composite	etching/conditioning of enamel, followed by the universal adhesion procedure	Bis-GMA, barium glass	82	0.02–0.70

NC – NexCore™; S-D – spee-dee™ build-up; E – Ecosite Bulk Fill; Bis-GMA – bisphenol A-glycidyl methacrylate; TEGDMA – triethylene glycol dimethacrylate; BPA – bisphenol A.

Table 2. Characteristics of the storage media used in the study

Storage medium	Type	Composition	pH
Wet Mouth (ICPA Health Products Ltd, Mumbai, India)	saliva substitute	water, glycerin, sorbitol, propylene glycol, PEG-40-HCO, poloxamer, sodium benzoate, sodium CMC, flavor, cetylpyridinium chloride, parabens, xylitol, xanthan gum, disodium hydrogen phosphate, sodium dihydrogen phosphate	7
Kingfisher® Strong Premium (United Breweries, Mangaluru, India)	alcoholic beverage (beer)	alcohol content: 7.2 vol% prenylated flavonoids, phenolic acids, simple phenols, flavanols, hydroxycoumarins, flavones, proanthocyanidins, tannins, amino-phenolic compounds	3.94
Thums Up® (Coca-Cola Company, Bengaluru, India)	non-alcoholic beverage	carbonated water, sugar, acidity regulator (E338), caffeine, natural color (150d), flavors	2.68

PEG-40-HCO – polyethylene glycol-40-hydrogenated castor oil; CMC – carboxymethyl cellulose.

The load required to fracture the specimen was recorded in kN, and the CS was calculated according to the following formula (Equation 1):

$$CS\,[\mathrm{MPa}] = \frac{F_{\max}}{A} \quad (1)$$

where:
F – force [N], calculated as load [kN] × 10^3;
A – cross-sectional area [mm^2], calculated as $\pi \times r^2$ (r – radius of the specimen) or as $\pi \times (d/2)^2$ (d – diameter).

Power analysis

The post hoc power analysis for the comparison of 2 distributions with the Mann–Whitney *U* test and analysis of variance (ANOVA) in 3 groups was conducted using the G*power v. 3.1.9.4 software.[21] The power exceeded 0.90 for the various tests conducted in the study.

Statistical analysis

The data analysis was performed using the IBM SPSS Statistics for Windows software, v. 25.0 (IBM Corp., Armonk, USA). The variables obtained from the 3 fluid groups were subjected to the Shapiro–Wilk test of normality. The saliva data in the NC and S-D groups, alcohol data in the S-D and E groups and data on all liquids in the E group did not conform to a normal distribution. Hence, the comparison of distribution was conducted using the non-parametric Kruskal–Wallis and Mann–Whitney *U* tests. The non-alcoholic liquid data for the 3 material groups conformed to normality, enabling a comparison of mean fracture resistance. This comparison was performed using parametric ANOVA and the Bonferroni post hoc test.

Results

Compressive strength in artificial saliva

For the saliva, the CS values were the highest in the E group (median (*Me*) (interquartile range (*IQR*)): 275.1 (259.8–295.5) MPa) and the lowest in the S-D group (203.8 (163.0–219.1) MPa). The CS values for Ecosite Bulk Fill were significantly higher than those for NexCore™ and spee-dee™ build-up. The significant differences were observed in the distributions across the 3 material groups, as evidenced by a pairwise comparison ($p = 0.000$) (Table 3).

Compressive strength in an alcoholic liquid

For the alcoholic liquid, the highest CS values were observed in the NC group (285.3 (270.0–295.5) MPa) and the lowest values were noted in the S-D group (224.2 (198.7–239.4) MPa). In the NC group, the values were significantly higher when compared to the other groups. The significant differences were observed in the distributions across the 3 material groups, as demonstrated by the results of the pairwise comparison ($p < 0.05$) (Table 4).

Table 3. Comparison of the compressive strength (CS) between the groups after immersion in saliva

Study group	CS [MPa]			*p*-value
	M ±*SD*	*Me* (IQR)	range	
NC	244.2 ±14.9	249.6 (235.4–255.7)	217.5–258.8	0.000*
S-D	199.7 ±25.6	203.8 (163.0–219.1)	163.0–234.3	
E	279.4 ±22.6	275.1 (259.8–295.5)	251.2–322.0	

* statistically significant ($p < 0.05$, Kruskal–Wallis test); *M* – mean; *SD* – standard deviation; *Me* – median; IQR – interquartile range. The results of the pairwise comparison (Mann–Whitney *U* test) were as follows: NC vs. S-D: $p = 0.000$; NC vs. E: $p = 0.000$; S-D vs. E: $p = 0.000$.

Compressive strength in a non-alcoholic liquid

For the non-alcoholic liquid, the highest CS values were noted in the E group (254.7 (214.0–295.5) MPa) and the lowest values were observed in the S-D group (234.3 (193.6–275.1) MPa). However, the distribution of the CS across 3 materials did not show any significant differences, as evidenced by a pairwise comparison ($p > 0.05$) (Table 5).

Table 4. Comparison of the compressive strength (CS) between the groups after immersion in an alcoholic liquid

Study group	CS [MPa]			*p*-value
	M ±*SD*	*Me* (IQR)	range	
NC	282.2 ±17.3	285.3 (270.0–295.5)	254.7–305.7	0.0001*
S-D	218.4 ±33.3	224.2 (198.7–239.4)	122.3–249.6	
E	236.7 ±44.0	254.7 (203.8–264.9)	117.2–285.3	

* statistically significant ($p < 0.05$, Kruskal–Wallis test). The results of the pairwise comparison (Mann–Whitney *U* test) were as follows: NC vs. S-D: $p = 0.0001$; NC vs. E: $p = 0.0001$; S-D vs. E: $p = 0.036$.

Table 5. Comparison of the compressive strength (CS) between the groups after immersion in a non-alcoholic liquid

Study group	CS [MPa]			*p*-value (ANOVA)
	M ±*SD*	*Me* (IQR)	range	
NC	246.2 ±21.1	244.5 (229.2–264.9)	214.0–285.3	0.396
S-D	233.3 ±41.2	234.3 (193.6–275.1)	173.2–295.5	
E	252.0 ±47.0	254.7 (214.0–295.5)	168.1–315.8	

The results of the pairwise comparison (Bonferroni post hoc test) were as follows: NC vs. S-D: $p = 1.000$; NC vs. E: $p = 1.000$; S-D vs. E: $p = 0.559$.

Comparison of the compressive strength of composites across different subgroups

The mean CS values were compared between the liquids within each of the 3 materials (Table 6,7) (Fig. 2). The mean CS for the NC group was the highest for alcohol (282.2 ±17.3 MPa) and the lowest for saliva (244.2 ±14.9 MPa). These values were significantly higher after storage in alcohol than in saliva and non-alcoholic liquid (p = 0.0001).

In the S-D group, the CS was the highest for the non-alcoholic liquid (233.3 ±41.2 MPa) and the lowest for saliva (199.7 ±25.6 MPa). These values were significantly higher after exposure to the non-alcoholic liquid compared to saliva (p = 0.029).

In the E group, the CS was the highest for saliva (279.4 ±22.6 MPa) and the lowest for the alcoholic liquid (236.7 ±44.0 MPa). Compressive strength was significantly higher after storage in saliva in comparison to alcohol (p = 0.015).

Table 6. Comparison of the impact of liquids on the compressive strength (CS) of composites within study groups

Study group	Medium	*M* ±*SD* [MPa]	*p*-value
NC	saliva	244.2 ±14.9	0.0001*
	alcohol	282.2 ±17.3	
	non-alcoholic beverage	246.2 ±21.1	
S-D	saliva	199.7 ±25.6	0.033*
	alcohol	218.4 ±33.3	
	non-alcoholic beverage	233.3 ±41.2	
E	saliva	279.4 ±22.6	0.017*
	alcohol	236.7 ±44.0	
	non-alcoholic beverage	252.0 ±47.0	

* statistically significant (p < 0.05, ANOVA).

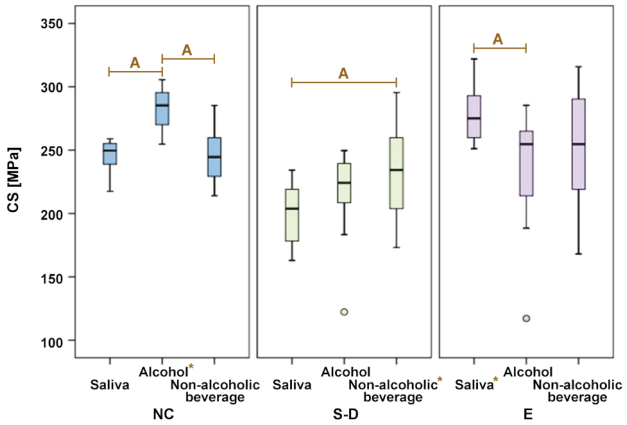

Fig. 2. Compressive strength (CS) of different materials after 24-h immersion in artificial saliva, alcohol and non-alcoholic liquid

NC – NexCore™; S-D – spee-dee™ build-up; E – Ecosite Bulk Fill. Statistically significant values within the groups are denoted by an "A" (p < 0.05), and the highest values within the groups are marked with an asterisk (*).

Table 7. Results of the Bonferroni post hoc test

Group	Medium		*p*-value
NC	saliva	alcohol	0.0001*
		non-alcoholic beverage	1.000
	alcohol	non-alcoholic beverage	0.0001*
S-D	saliva	alcohol	0.419
		non-alcoholic beverage	0.029*
	alcohol	non-alcoholic beverage	0.705
E	saliva	alcohol	0.015*
		non-alcoholic beverage	0.191
	alcohol	non-alcoholic beverage	0.883

* statistically significant (p < 0.05).

Discussion

Composite resin materials are being increasingly applied in modern reconstructive dentistry. However, despite the improvement in their physical and chemical properties, the long-term clinical performance in the oral cavity remains a challenge.

The present investigation demonstrates that the type of immersing liquid affects the CS of the bulk-fill composites applied as core build-up restorations. Therefore, the null hypothesis should be rejected. These findings are consistent with those of previous studies that evaluated the physical properties of different core build-up materials when immersed in food-simulating liquids.[22,23]

The duration of sample fabrication and storage were chosen based on the literature.[23–29] The storage time of 12 h simulated a 1-year exposure of 2 min/day. Consequently, the calculated time equaled 2 years of clinical service.[24,30] Compressive strength is a vital parameter for the performance and longevity of core build-up materials. It influences the ability to withstand functional loads and prevent fractures, ultimately contributing to better clinical outcomes.[31] Core build-ups are subject to occlusal forces due to load transfer from the crown to the supporting material during function.

A substantial body of research has indicated that the mechanical properties of resin materials deteriorated after immersion in solutions such as coffee, tea and juices.[14–17,22–25,28,30,32–45] However, only few studies investigated the influence of alcoholic and non-alcoholic beverages on core build-up bulk-fill materials.[14,22] None of the studies have evaluated the CS of NexCore™ and spee-dee™ build-up after storage in beverages. Additionally, in the present study, a salivary substitute was used as a control to imitate the oral cavity environment, as recommended by literature.[46] Beer was chosen as one of the liquids, due to its status as the most ancient and frequently consumed alcoholic beverage in the world.[41] Likewise, one of the most popular brands of soft drink was selected for the non-alcoholic beverage.[43] Moreover, both Kingfisher® beer and Thums Up® are popular products.[41]

The current study revealed a significantly higher CS of Ecosite Bulk Fill when immersed in the salivary substitute and of NexCore™ after storage in the alcoholic fluid when compared to the other liquids tested. Spee-dee™ build-up demonstrated the lowest CS after storage in both salivary substitute and alcohol. However, in the non-alcoholic fluid, no statistically significant differences in CS were observed between the investigated materials. It is worth mentioning that the materials under investigation exhibited CS within the average range, demonstrating sufficient performance for clinical purposes.[47,48]

Several studies have demonstrated that liquids have an impact on the physical and mechanical properties of composites.[14,22,23,25,28,37,49–54] It has been shown that composites aged in distilled water exhibit reduced microhardness and flexural strength values when compared to dry samples.[17,44,55] The increased content of triethylene glycol dimethacrylate (TEGDMA) in the resin matrix may result in an increased water uptake due to the higher hydrophilicity of this monomer compared to bisphenol A-glycidyl methacrylate (Bis-GMA) and urethane dimethacrylate (UDMA).[56] This increased hydrophilicity could be a contributing factor to the lower CS of the TEGDMA-containing composite, NexCore™. Water immersion causes hydrolytic degradation of the filler surface and erosion of resin composite by filler matrix debonding.[54] Sorption, as a co-existing process, results in the swelling of the matrix and a subsequent formation of pores inside the material.[54] Additionally, unbound monomers and additives may be eluted by solvents after setting. The second mechanism causing mass loss is surface erosion, which leads to the long-term release of components.[43,49] The type of inorganic filler may influence the material's properties after immersion in water. Specifically, zinc, barium glass and zirconia/silica fillers have been found to modify the material's properties in comparison to those containing quartz fillers.[36,52]

The most frequently investigated and consumed alcoholic beverage is wine. A high risk of dental erosion has been identified in individuals with a predilection for wine.[42] However, beer is the second most widely consumed alcoholic beverage worldwide.[42] In the present study, it was noted that beer (7.2 vol%) reduced the CS of Ecosite Bulk Fill and spee-dee™ build-up composites. These findings are consistent with the conclusions of previous studies, which demonstrated that the presence of alcohol leads to a reduction in the physical properties of composites.[14,57,58] Due to their low pH and ethanol content, alcoholic beverages can cause erosion and modify the properties of composites.[30] The ethanol component of alcoholic beverages and the citric acid present in non-alcoholic beverages induce plasticization, increase surface roughness and solubility, and decrease flexural strength and flexural modulus.[35,54,59–61] However, some studies have reported no changes in the surface roughness of composite materials following exposure to alcoholic and non-alcoholic liquids.[35,52] The investigators of these studies have posited that the absence of marked changes in pH over time in the stored solutions, in comparison to freshly prepared solutions, may be a contributing factor. Secondly, the filler components (zirconia or silica) of the composites used were more inert in different solutions.

Interestingly, beer was found to increase surface roughness when compared to strong alcohols.[49] This phenomenon can be attributed to the higher acidity levels of beer compared to vodka. In other words, an increase in the acidic concentration of beverages corresponds to an increase in the surface roughness of composite materials.[51,62] Moreover, low pH leads to the degradation of calcium, aluminum and silicon ions present in restorative resins, consequently resulting in an increased roughness of the material. Acids penetrate the resin matrix, triggering the release of unreacted monomers into the environment.[63] The increased surface roughness may result in staining of resin restoration and bacterial adherence, leading to the formation of secondary caries.

It is worth mentioning that the majority of in vitro studies focus on the erosive potential of beverages and disregard the protective properties of saliva, which are attributable to buffer substances that facilitate pH neutralization. However, it should be noted that clinical performance of composite materials, apart from erosive factors, is influenced by occlusal forces, endogenic acids, eating disorders, diet and hygienic habits, smoking, and the use of drugs.[64]

Additionally, NexCore™, after immersion in beer, exhibited statistically higher CS than other study groups. NexCore™ core build-up contains a filler matrix composed of barium glass, ytterbium (III) fluoride and pyrogenic silicic acid, and a resin matrix of Bis-GMA with TEGDM.[65] The alcoholic liquid increases the ductility of composites by acting as a plasticizer of the polymeric matrix.[59] Moreover, ethanol can contribute to the reduction in the bond between the resin matrix and inorganic fillers, causing a decrease in mechanical properties.[38]

All investigated bulk-fill composites exhibited comparable CS after immersion in non-alcoholic beverage (Thums Up®). A potential explanation for this phenomenon is that the major component of this beverage is citric acid (pH = 2.5). However, this component did not cause significant changes in the flexural strength of both silorane- and methacrylate-based composites.[19,66] In other studies, citric acid immersion resulted in a significant reduction in flexural strength and hardness, and an increased roughness of methacrylate-based materials.[26,53] The reduced values of these parameters could be attributed to the high acidic and low fluoride and calcium levels of Thums Up® cola drink, which exerts a softening effect on Bis-GMA.[45,66] Bis-GMA constitutes the main resin matrix component of NexCore™ and Ecosite Bulk Fill. These findings align with those reported in previous studies.[14,37]

In the present study, a significantly higher CS of the dual-cure core build-up material (spee-dee™ build-up) was observed after immersion in the non-alcoholic beverage compared to artificial saliva. According to the literature, the consumption of non-alcoholic beverages (i.e., Coca Cola) may increase surface roughness and decrease microhardness of microhybrid and hybrid resin composites.[18,37,55]

Spee-dee™ build-up exhibited the lowest CS in all liquids. It is a hydrophilic material based on the Embrace monomer that is believed to be more hydrophilic than UDMA- or Bis-GMA-based composites.[19] Moreover, the low CS value may be due to high water sorption of spee-dee™ build-up when compared to other composites.[66]

The tested composite materials differ in terms of their filler content and size. In the present study, NexCore™ and Ecosite Bulk Fill had higher filler content (75 wt% and 82 wt%) than spee-dee™ build-up (54.5 wt%). Filler loading affects the flexural strength, modulus of elasticity, fracture toughness, microhardness, and surface roughness of composites.[56] Increasing the filler load beyond 55% results in the crack browning effect, when crack branching occurs or the matrix around the filler plastically deforms. This could explain why both NexCore™ and Ecosite Bulk Fill have higher CS than spee-dee™ build-up.

Limitations

There are some limitations of the study that should be acknowledged. First, the study design did not fully replicate the multifactorial oral environment (saliva flow, different types of saliva composition released by glands, plaque, temperature changes, and occlusal load). Artificial saliva lacks the full range of proteins and enzymes present in natural saliva. Therefore, it does not provide antibacterial properties and buffering capacity. Moreover, the utilization of composites and beverages was restricted, and the study did not evaluate the tooth–restoration complex samples. Another limitation is the non-normal distribution, which is attributable to the relatively small sample size. It is therefore recommended that the sample size be increased in future studies. Another aspect that affects the normality of distribution are outliers, which mainly influence the average.[67] The results of the present in vitro research cannot be extrapolated to clinical conditions. Therefore, further studies should be carried out to evaluate other types of beverages and their influence on various resin composites. Additionally, pH cycling simulation, microhardness, three-point bending, and diametral tensile strength testing of bulk-fill composites should be carried out. Furthermore, a comprehensive evaluation of the tooth–restoration complex and interface is imperative to ascertain their quality and longevity. Subsequent studies should also incorporate the complexity of oral cavity conditions to fully investigate the influence of beverages on the tooth–restoration complex in clinical settings.

Conclusions

The immersion medium affects the CS of bulk-fill composites. Moreover, NexCore™ and Ecosite Bulk Fill exhibited significant CS, while the CS for spee-dee™ build-up was the lowest after storage in the tested liquids.

Ethics approval and consent to participate

Not applicable.

Data availability

The datasets generated and/or analyzed during the current study are available from the corresponding author on reasonable request.

Consent for publication

Not applicable.

Use of AI and AI-assisted technologies

Not applicable.

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References

1. Reeh ES, Messer HH, Douglas WH. Reduction in tooth stiffness as a result of endodontic and restorative procedures. *J Endod*. 1989;15(11):512–516. doi:10.1016/S0099-2399(89)80191-8
2. Gillen BM, Looney SW, Gu LS, et al. Impact of the quality of coronal restoration versus the quality of root canal fillings on success of root canal treatment: A systematic review and meta-analysis. *J Endod*. 2011;37(7):895–902. doi:10.1016/j.joen.2011.04.002
3. American Association of Endodontists. Treatment Standards. https://www.aae.org/specialty/wp-content/uploads/sites/2/2018/04/TreatmentStandards_Whitepaper.pdf. Accessed January 16, 2024.
4. AAE guidelines. https://www.aae.org/specialty/publications-research/endodontics-colleagues-excellence. Accessed January 16, 2024.
5. Spinhayer L, Bui ATB, Leprince JG, Hardy CMF. Core build-up resin composites: An in-vitro comparative study. *Biomater Investig Dent*. 2020;7(1):159–166. doi:10.1080/26415275.2020.1838283
6. Spielman H, Schaffer SB, Cohen MG, et al.; Practitioners Engaged in Applied Research and Learning (PEARL) Network. Restorative outcomes for endodontically treated teeth in the Practitioners Engaged in Applied Research and Learning Network. *J Am Dent Assoc*. 2012;143(7):746–755. doi:10.14219/jada.archive.2012.0262
7. Suksaphar W, Banomyong D, Jirathanyanatt T, Ngoenwiwatkul Y. Survival rates from fracture of endodontically treated premolars restored with full-coverage crowns or direct resin composite restorations: A retrospective study. *J Endod*. 2018;44(2):233–238. doi:10.1016/j.joen.2017.09.013
8. Dietschi D, Duc O, Krejci I, Sadan A. Biomechanical considerations for the restoration of endodontically treated teeth: A systematic review of the literature--Part 1. Composition and micro- and macrostructure alterations. *Quintessence Int*. 2007;38(9):733–743. PMID:17873980.

9. Magne P, Goldberg J, Edelhoff D, Güth JF. Composite resin core buildups with and without post for the restoration of endodontically treated molars without ferrule. *Oper Dent*. 2016;41(1):64–75. doi:10.2341/14-258-L
10. Warangkulkasemkit S, Pumpaluk P. Comparison of physical properties of three commercial composite core build-up materials. *Dent Mater J*. 2019;38(2):177–181. doi:10.4012/dmj.2018-038
11. Hatipoğlu Ö, Par M, Pertek Hatipoğlu F. Comparison of degree of conversion performance of bulk-fill resin composites: A systematic review and network meta-analysis of in vitro studies. *J Dent*. 2024;149:105289. doi:10.1016/j.jdent.2024.105289
12. Nijakowski K, Jankowski J, Lehmann A. Bibliometric analysis of literature on bulk-fill composite resins in dentistry. *J Endod Restor Dent*. 2023;1(1):2–7. doi:10.5281/zenodo.8306021
13. Belli S, Eraslan O, Eskitascioglu G. Direct restoration of endodontically treated teeth: A brief summary of materials and techniques. *Curr Oral Health Rep*. 2015;2(4):182–189. doi:10.1007/s40496-015-0068-5
14. Yusuf F, Srirekha A, Hegde J, Karale R, Bashetty K, Adiga S. Effect of alcoholic and non-alcoholic beverages on the wear and fracture toughness of teeth and resin composite materials: In vitro study. *J Restor Dent*. 2013;1(1):11–17. doi:10.4103/2321-4619.111227
15. Zimmer S, Kirchner G, Bizhang M, Benedix M. Influence of various acidic beverages on tooth erosion. Evaluation by a new method. *PLoS One*. 2015;10(6):e0129462. doi:10.1371/journal.pone.0129462
16. Wongkhantee S, Patanapiradej V, Maneenut C, Tantbirojn D. Effect of acidic food and drinks on surface hardness of enamel, dentine, and tooth-coloured filling materials. *J Dent*. 2006;34(3):214–220. doi:10.1016/j.jdent.2005.06.003
17. Badra VV, Faraoni JJ, Pereira Ramos R, Palma-Dibb RG. Influence of different beverages on the microhardness and surface roughness of resin composites. *Oper Dent*. 2005;30(2):213–219. PMID:15853107.
18. Szalewski L, Wójcik D, Bogucki M, Szkutnik J, Różyło-Kalinowska I. The influence of popular beverages on mechanical properties of composite resins. *Materials (Basel)*. 2021;14(11):3097. doi:10.3390/ma14113097
19. Santos C, Clarke RL, Braden M, Guitian F, Davy KW. Water absorption characteristics of dental composites incorporating hydroxyapatite filler. *Biomaterials*. 2002;23(8):1897–1904. doi:10.1016/S0142-9612(01)00331-3
20. Hatipoğlu Ö, Turumtay EA, Saygın AG. Evaluation of monomer elution, microhardness, and roughness of experimental dental composite resins prepared from Bis-EFMA, a novel monomer system. *Polym Compos*. 2022;43(1):584–592. doi:10.1002/pc.26401
21. Faul F, Erdfelder E, Lang AG, Buchner A. G*Power 3: A flexible statistical power analysis program for the social, behavioral, and biomedical sciences. *Behav Res Methods*. 2007;39(2):175–191. doi:10.3758/BF03193146
22. Borges MG, Soares CJ, Maia TS, et al. Effect of acidic drinks on shade matching, surface topography, and mechanical properties of conventional and bulk-fill composite resins. *J Prosthet Dent*. 2019;121(5):868.e1–868.e8. doi:10.1016/j.prosdent.2019.02.006
23. Bansal K, Acharya SR, Saraswathi V. Effect of alcoholic and non-alcoholic beverages on color stability and surface roughness of resin composites: An in vitro study. *J Conserv Dent*. 2012;15(3):283–288. doi:10.4103/0972-0707.97961
24. Geha O, Inagaki LT, Favaro JC, et al. Effect of chemical challenges on the properties of composite resins. *Int J Dent*. 2021;2021:4895846. doi:10.1155/2021/4895846
25. Eweis AH, Yap AUJ, Yahya NA. Impact of dietary solvents on flexural properties of bulk-fill composites. *Saudi Dent J*. 2018;30(3):232–239. doi:10.1016/j.sdentj.2018.04.002
26. Maganur P, Satish V, Prabhakar AR, Namineni S. Effect of soft drinks and fresh fruit juice on surface roughness of commonly used restorative materials. *Int J Clin Pediatr Dent*. 2015;8(1):1–5. doi:10.5005/jp-journals-10005-1274
27. Jain G, Narad A, Boruah LC, Rajkumar B. Comparative evaluation of shear bond strength of three resin based dual-cure core build-up materials: An in-vitro study. *J Conserv Dent*. 2015;18(4):337–341. doi:10.4103/0972-0707.159754
28. Abouelleil H, Pradelle N, Villat C, Attik N, Colon P, Grosgogeat B. Comparison of mechanical properties of a new fiber reinforced composite and bulk filling composites. *Restor Dent Endod*. 2015;40(4):262–270. doi:10.5395/rde.2015.40.4.262
29. Jayanthi N, Vinod V. Comparative evaluation of compressive strength and flexural strength of conventional core materials with nanohybrid composite resin core material an in vitro study. *J Indian Prosthodont Soc*. 2013;13(3):281–289. doi:10.1007/s13191-012-0236-4
30. Gürgan S, Onen A, Köprülü H. In vitro effects of alcohol-containing and alcohol-free mouthrinses on microhardness of some restorative materials. *J Oral Rehabil*. 1997;24(3):244–246. PMID:9131482.
31. Wang X, Huyang G, Palagummi SV, et al. High performance dental resin composites with hydrolytically stable monomers. *Dent Mater*. 2018;34(2):228–237. doi:10.1016/j.dental.2017.10.007
32. Ozkanoglu S, Akin EGG. Evaluation of the effect of various beverages on the color stability and microhardness of restorative materials. *Niger J Clin Pract*. 2020;23(3):322–328. doi:10.4103/njcp.njcp_306_19
33. Mohammadi E, Pishevar L, Mirzakouchaki Boroujeni P. Effect of food simulating liquids on the flexural strength of a methacrylate and silorane-based composite. *PLoS One*. 2017;12(12):e0188829. doi:10.1371/journal.pone.0188829
34. Hengtrakool C, Kukiattrakoon B, Kedjarune-Leggat U. Effect of naturally acidic agents on microhardness and surface micromorphology of restorative materials. *Eur J Dent*. 2011;5(1):89–100. PMID:21311608.
35. Tanthanuch S, Kukiattrakoon B, Siriporananon C, et al. The effect of different beverages on surface hardness of nanohybrid resin composite and giomer. *J Conserv Dent*. 2014;17(3):261–265. doi:10.4103/0972-0707.131791
36. Poggio C, Dagna A, Chiesa M, Colombo M, Scribante A. Surface roughness of flowable resin composites eroded by acidic and alcoholic drinks. *J Conserv Dent*. 2012;15(2):137–140. doi:10.4103/0972-0707.94581
37. Correr GM, Bruschi Alonso RC, Baratto-Filho F, Correr-Sobrinho L, Sinhoreti MAC, Puppin-Rontani RM. In vitro long-term degradation of aesthetic restorative materials in food-simulating media. *Acta Odontol Scand*. 2012;70(2):101–108. doi:10.3109/00016357.2011.600701
38. Omata Y, Uno S, Nakaoki Y, et al. Staining of hybrid composites with coffee, oolong tea, or red wine. *Dent Mater J*. 2006;25(1):125–131. doi:10.4012/dmj.25.125
39. Palani Swamy UK, Amravai AR, Mandadi SR, Habeeb A. Effect of alcoholic beverages on shear bond strength of composites to enamel. *J Conserv Dent*. 2018;21(5):542–545. doi:10.4103/JCD.JCD_180_18
40. Eapen AM, Amirtharaj LV, Sanjeev K, Mahalaxmi S. Fracture resistance of endodontically treated teeth restored with 2 different fiber-reinforced composite and 2 conventional composite resin core buildup materials: An in vitro study. *J Endod*. 2017;43(9):1499–1504. doi:10.1016/j.joen.2017.03.031
41. Siegel M, DeJong W, Naimi TS, et al. Alcohol brand preferences of underage youth: Results from a pilot survey among a national sample. *Subst Abus*. 2011;32(4):191–201. doi:10.1080/08897077.2011.601250
42. Brand HS, Tjoe Fat GM, Veerman ECI. The effects of saliva on the erosive potential of three different wines. *Aust Dent J*. 2009;54(3):228–232. doi:10.1111/j.1834-7819.2009.01124.x
43. Somavarapu S, Mubeena B. A consumer survey on preferences of soft drinks in different stages of adolescence. *Res Rev: J Dairy Sci Technol*. 2017;6(3):54–73. doi:10.37591/rrjodst.v6i3.142
44. World Health Organization. Global status report on alcohol and health. 2011. https://apps.who.int/iris/handle/10665/44499. Accessed December 17, 2021.
45. Yanikoğlu N, Duymuş ZY, Yilmaz B. Effects of different solutions on the surface hardness of composite resin materials. *Dent Mater J*. 2009;28(3):344–351. PMID:19662734.
46. Veloso SRM, Lemos CAA, de Moraes SLD, do Egito Vasconcelos BC, Pellizzer EP, de Melo Monteiro GQ. Clinical performance of bulk-fill and conventional resin composite restorations in posterior teeth: A systematic review and meta-analysis. *Clin Oral Invest*. 2019;23(1):221–233. doi:10.1007/s00784-018-2429-7
47. Zankuli MA, Silikas N, Devlin H. The effect of cyclic loading on the compressive strength of core build-up materials. *J Prosthodont*. 2015;24(7):549–552. doi:10.1111/jopr.12264
48. Cho GC, Kaneko LM, Donovan TE, White SN. Diametral and compressive strength of dental core materials. *J Prosthet Dent*. 1999;82(3):272–276. doi:10.1016/S0022-3913(99)70079-X

49. Vaidya N, Kumar P, Pathak K, Punia SK, Choudhary A, Patnana AK. Comparative evaluation of the influence of different sports/energy drinks and alcoholic beverages on the surface roughness of three different flowable esthetic restorative materials: An in vitro analysis. *J Int Soc Prev Community Dent*. 2020;10(5):585–590. doi:10.4103/jispcd.JISPCD_246_20
50. Borah D, Shetty S, KB J, Nadig P, I S, DS S. Effect of dietary drinks on the microhardness of two nanocomposites: An in-vitro study. *Saudi J Oral Dent Res*. 2017;2(6):134–139. https://saudijournals.com/media/articles/SJODR-26134-139.pdf. Accessed February 1, 2024.
51. Mahajan RP, Shenoy VU, Sumanthini MV, Mahajan HP, Walzade PS, Mangrolia R. Comparative evaluation of the discoloration of microhybrid and nanohybrid composite resins by different beverages: A spectrophotometric analysis. *J Contemp Dent Pract*. 2019;20(2):226–230. PMID:31058640.
52. Yap AU, Low JS, Ong LF. Effect of food-simulating liquids on surface characteristics of composite and polyacid-modified composite restoratives. *Oper Dent*. 2000;25(3):170–176. PMID:11203812.
53. Akova T, Ozkomur A, Uysal H. Effect of food-simulating liquids on the mechanical properties of provisional restorative materials. *Dent Mater*. 2006;22(12):1130–1134. doi:10.1016/j.dental.2005.09.009
54. Rajavardhan K, Sankar A, Kumar M, Kumar K, Pranitha K, Kishore K. Erosive potential of cola and orange fruit juice on tooth colored restorative materials. *Ann Med Health Sci Res*. 2014;4(Suppl 3):S208–S212. doi:10.4103/2141-9248.141960
55. Da Silva MAB, Vitti RP, Sinhoreti MAC, Consani RLX, da Silva-Júnior JG, Tonholo J. Effect of alcoholic beverages on surface roughness and microhardness of dental composites. *Dent Mater J*. 2016;35(4):621–626. doi:10.4012/dmj.2015-383
56. Calais JG, Söderholm KJM. Influence of filler type and water exposure on flexural strength of experimental composite resins. *J Dent Res*. 1988;67(5):836–840. doi:10.1177/00220345880670050801
57. Scotti N, Eruli C, Comba A, et al. Longevity of class 2 direct restorations in root-filled teeth: A retrospective clinical study. *J Dent*. 2015;43(5):499–505. doi:10.1016/j.jdent.2015.02.006
58. Reis AF, Vestphal M, do Amaral RC, Rodrigues JA, Roulet JF, Roscoe MG. Efficiency of polymerization of bulk-fill composite resins: A systematic review. *Braz Oral Res*. 2017;31(Suppl 1):e59. doi:10.1590/1807-3107bor-2017.vol31.0059
59. Vouvoudi EC, Sideridou ID. Dynamic mechanical properties of dental nanofilled light-cured resin composites: Effect of food-simulating liquids. *J Mech Behav Biomed Mater*. 2012;10:87–96. doi:10.1016/j.jmbbm.2012.02.007
60. Rahim TNAT, Mohamad D, Md Akil H, Ab Rahman I. Water sorption characteristics of restorative dental composites immersed in acidic drinks. *Dent Mater*. 2012;28(6):e63–e70. doi:10.1016/j.dental.2012.03.011
61. Schmidt C, Ilie N. The mechanical stability of nano-hybrid composites with new methacrylate monomers for matrix compositions. *Dent Mater*. 2012;28(2):152–159. doi:10.1016/j.dental.2011.11.007
62. Reddy PS, Tejaswi KLS, Shetty S, Annapoorna BM, Pujari SC, Thippeswamy HM. Effects of commonly consumed beverages on surface roughness and color stability of the nano, microhybrid and hybrid composite resins: An in vitro study. *J Contemp Dent Pract*. 2013;14(4):718–723. doi:10.5005/jp-journals-10024-1390
63. Erdemir U, Yildiz E, Eren MM, Ozel S. Surface hardness evaluation of different composite resin materials: Influence of sports and energy drinks immersion after a short-term period. *J Appl Oral Sci*. 2013;21(2):124–131. doi:10.1590/1678-7757201302185
64. Meira IA, Fernandes NLS, de Lavôr JR, Sampaio FC, de Oliveira AFB. Investigation of the erosive potential of different types of alcoholic beverages. *Pesqui Bras Odontopediatria Clín Integr*. 2020;20:e4920. doi:10.1590/pboci.2020.050
65. Meta® Biomed Europe. Beyond Expectations. Product Catalogue 2019. https://www.meta-europe.com/wp-content/uploads/2019/08/Meta_Biomed_Katalog_EN_Download.pdf. Accessed August 5, 2025.
66. Yesilyurt C, Yoldas O, Altintas SH, Kusgoz A. Effects of food-simulating liquids on the mechanical properties of a silorane-based dental composite. *Dent Mater J*. 2009;28(3):362–367. doi:10.4012/dmj.28.362
67. Krithikadatta J. Normal distribution. *J Conserv Dent*. 2014;17(1):96–97. doi:10.4103/0972-0707.124171

Original paper

Challenges in defining an efficient cleansing protocol against mono- and multi-species biofilms on a cobalt-chromium alloy surface

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Conflict of interest
None declared

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Abstract

Background. Effective cleaning protocols are crucial for controlling biofilm formation on oral prostheses and preserving the oral health of patients relying on removable partial dentures (RPDs).

Objectives. The present study aimed to investigate the antibiofilm efficacy of 4 cleansing protocols on a cobalt-chromium (Co-Cr) alloy surface, which is commonly used as the base-metal framework material in dental prosthodontics.

Material and methods. Cobalt-chromium specimens were contaminated with isolated strains of *Candida albicans*, *Candida glabrata*, *Staphylococcus aureus*, and *Streptococcus mutans* to form mono-species biofilms. For a multi-species biofilm, all strains were grown simultaneously on the surfaces of the specimens. After biofilm maturation, the specimens were immersed in different solutions: *Ricinus communis* 2%; *R. communis* 10%; Cepacol™; NitrAdine™; and distilled water (control). After applying the hygiene protocols, the viability of the microorganisms and the amount of residual biofilm were assessed.

Results. Immersion in *R. communis*-based solutions did not significantly alter the viability of the microorganisms. Cepacol reduced the viability of *C. albicans*, *C. glabrata* and *S. aureus* in the mono-species biofilms, as well as *C. glabrata* in the multi-species biofilm. NitrAdine demonstrated effectiveness in reducing the viability of *C. glabrata* and *S. mutans* in both the mono- and multi-species biofilms. However, its efficacy against *S. aureus* was only observed in the mono-species pattern. NitrAdine also reduced the area covered by the living biofilm.

Conclusions. The studied cleansing protocols exhibited reduced antimicrobial efficacy on the multi-species biofilm as compared to the mono-species model. NitrAdine showed potential as a complementary agent for controlling biofilm formation on removable partial dentures.

Keywords: removable partial denture, cleansers, antimicrobial action, cobalt-chromium alloy

Highlights

- The cleansing protocols were less effective against the multi-species biofilm as compared to the mono-species biofilms.
- NitrAdine™ showed potential as a complementary agent for controlling biofilm on removable partial dentures.
- None of the tested solutions could significantly reduce both microbial viability and the biofilm-covered areas.
- NitrAdine™ reduced the viability of the most species, but had no effect on the *Candida albicans* microbial load.
- Cepacol™ was effective in the mono-species biofilms, but performed poorly in the multi-species biofilm.

Introduction

Oral biofilms are composed of bacteria and yeast-like fungi, which adhere and grow on biotic and abiotic surfaces.[1] Biofilms, with an inadequate hygiene of prostheses, constitute a source of microorganisms and act as a gate to systemic diseases.[2] Although it is known that the control of biofilm formation on oral prostheses is crucial for maintaining general health, there is no consensus regarding a suitable solution for removable partial dentures (RPDs).[3–5]

Compatibility with constituent materials is a requisite for an ideal RPD cleanser.[4,5] Additionally, other aspects, such as a low cost, easy manipulation and antibiofilm activity, are desirable.[6,7] There is evidence that diluted sodium hypochlorite is efficient in controlling biofilm formation; however, it is not recommended for to cleaning RPDs, taking into consideration their metal components.[3,8] Mouthwashes are popular in oral care and are frequently used as prosthesis cleansing solutions,[9] even though there are no specific guidelines regarding their use. Generally, these formulations include chlorhexidine, chlorine dioxide, cetylpyridinium chloride, and essential oils (e.g., eucalyptol, menthol, thymol, and methyl salicylate).[10] Besides mouthwashes, effervescent tablets are also largely used, partly due to their pleasant taste and odor characteristics. They are composed of different active ingredients, such as titanium dioxide, sodium lauryl sulfate and ethylenediaminetetraacetic acid (EDTA).[11] Both mouthwashes and effervescent tablets are complex chemical combinations that can damage the dental alloy as a result of ion release in the presence of oxidizing compounds.[12] According to previous studies, the NitrAdine™ effervescent tablet acts against oral biofilms[13,14] and may be indicated as an RPD cleanser, as its 5-year use did not induce deleterious effects to the dental alloy.[4,5] Nonetheless, given different characteristics of dental materials, it is fundamental to verify the antibiofilm activity of the cleanser on a metallic surface.

Broadening the knowledge about the antimicrobial properties of natural substances can have an impact on the selection of appropriate products to deal with the resistance of microorganisms.[15] Furthermore, it has been suggested that natural products do not have adverse effects inherent to synthetic compounds, and contribute to environmental and economic sustainability.[16] The *Ricinus communis* or castor oil plant belongs to the *Euphorbiaceae* family and is easily found in tropical zones. The *R. communis* oil has been used since antiquity, and has been demonstrated in medical and dental research to bring significant benefits.[17] Regarding its biological effects, the literature reports its healing, antioxidant, anti-inflammatory,[18] and antimicrobial properties.[19–21] In dentistry, previous studies indicated its potential use for prothesis hygiene[22,23] and the improvement of clinical conditions of denture-related stomatitis.[24] Even though the scientific literature has pointed out the compatibility of *R. communis* with the cobalt-chromium (Co-Cr) alloy,[5] its antibiofilm effect on a metallic surface has not been investigated.

Considering that inconsistent RPD hygiene can favor the manifestation of opportunistic pathologies,[25] new hygiene solutions should be investigated. Given this point, it is important to advertise that the presence of *Candida* spp. on a denture surface is an etiological factor for denture-related stomatitis.[26] In addition, the presence of other species, such as *Staphylococcus aureus* and *Streptococcus mutans*, may contribute to the pathogenicity of the biofilm.[2,27] The physical interactions of *Candida albicans* with various species go beyond simple synergistic and antagonistic associations. These interactions significantly influence the expression of virulence factors, directly impacting colonization and tissue invasion.[26] *Staphylococcus aureus*, *S. mutans*, *C. albicans*, and *Candida glabrata* are common species colonizing the abutment and non-abutment teeth in RPD wearers.[28] The presence of respiratory pathogens in the denture biofilm has already been investigated, and prostheses seem to act as a reservoir for *S. aureus*.[29] It is evident that the oral environment in RDP wearers is the habitat a polymicrobial community that interacts and forms a structured biofilm within a short period after clinical rehabilitation. However, the majority of studies refer to hygiene protocols only with regard to mono-species biofilms. Therefore, it is crucial that the antimicrobial analysis of RPD cleansers should explore different biofilm models.

For the aforementioned reasons, the present study analyzed the antimicrobial activity of cleansing solutions (mouthwash Cepacol™, effervescent tablet NitrAdine, and experimental solutions of *R. communis* (2% and 10%))

against mono- and multi-species biofilms (*C. albicans*, *C. glabrata*, *S. aureus*, and *S. mutans*) grown on a Co-Cr surface. The null hypothesis of this study was that the viability of the microorganisms and the biofilm-covered areas would be influenced by the cleansing protocols.

Material and methods

Experimental solutions

Castor oil was extracted from seeds, using the cold pressing method (Chemical Institute of São Carlos, University of São Paulo, São Carlos, Brazil). Initially, to formulate the *R. communis* solution, an esterification reaction with alcohols was performed. Afterward, the ester-containing solution was diluted in distilled water at final concentrations of 2% (RC02) and 10% (RC10) (v/v). The commercial mouthwash Cepacol (Reckitt Benckiser, São Paulo, Brazil) (CPC) was directly applied without dilution. The peroxide-based solution (Ni) was prepared by diluting one NitrAdine effervescent tablet (Bonyf, Vaduz, Liechtenstein) in 150 mL of water at 37°C, as directed by the manufacturer (Table 1).

Specimen manufacturing

A total of 244 Co-Cr disks were manufactured using the lost-wax casting method. Circular wax patterns (Ø 12 × 3 mm) were made using a metal matrix. The wax patterns were covered with the Micro-fine 1700 phosphate coating (Talladium Brazil, Curitiba, Brazil) and casting was performed using the Neutrodyn Easyti electronic machine (F.Lli Manfredi, Turin, Italy) by vacuum electroinduction. The disks were deflated and blasted with 100-micrometer aluminum oxide particles (Aluminum Oxide 100; Asfer Indústria Química, Sao Caetano do Sul, Brazil) at a pressure of 3 bar, using the Microjet III device (EDG, São Carlos, Brazil) for cleaning. After being separated from the feed channel, the opposing surfaces were progressively polished with 220-, 400-, 600-, and 1,200-grit sandpaper (Norton Abrasivos Brasil, Guarulhos, Brazil). The surface roughness of the specimens was standardized in the range of 0.04–0.10 µm.[5] The specimens were packaged in envelopes and sterilized with ethylene oxide.

Culture conditions

Four strains from the American Type Culture Collection (ATCC) were used for biofilm development: *C. albicans* (ATCC 10231); *C. glabrata* (ATCC 2001); *S. aureus* (ATCC 25923); and *S. mutans* (ATCC 25175). The experiment was carried out in 3 biological replications with 3 technical repetitions each, totaling in 9 specimens per group.

Biofilm growth was conducted under aseptic conditions, following the protocol described previously.[11] Briefly, the strains kept at –80°C in a glycerol stock were thawed and streaked out on a selected agar culture medium: for *C. albicans* and *C. glabrata* – Sabouraud Dextrose Agar (SDA) (HiMedia Laboratories, Mumbai, India); and for *S. aureus* and *S. mutans* – Brain Heart Infusion (BHI) broth (HiMedia). The plates were incubated at 37°C for 24 h. Subsequently, a microbial colony was transferred to its respective broth medium and re-incubated at 37°C for 24 h to obtain cells in the exponential growth phase. The cultures were then centrifuged at 4,200 g for 5 min. The resulting pellet was washed twice with phosphate-buffered saline (PBS). *Candida* spp. counting was performed in the Neubauer chamber (Kasvi, Curitiba, Brazil) due to the variable morphology of the genus. To adjust the cell concentration (10^8 colony-forming units per milliliter (CFU/mL)), the bacterial suspension was read on a spectrophotometer (Multiskan GO; Thermo Scientific, Waltham, USA) at 625 nm.

For mono-species biofilms, the inoculum was separately prepared in Sabouraud Dextrose Broth (SDB) (HiMedia) (*C. albicans* and *C. glabrata*) and BHI Broth (HiMedia) (*S. aureus* and *S. mutans*) at a cell concentration of 10^6 CFU/mL. The specimens were randomly assigned into 12-well cell culture plates (TPP Techno Plastic Products, Trasadingen, Switzerland) and filled with 2 mL of the inoculated culture media. In this model, each specimen was contaminated with only one species.

For a multi-species biofilm, the inoculum was prepared with the mixture of the 4 evaluated microorganisms for the specimens to be simultaneously contaminated with

Table 1. Characteristics of the hygiene solutions

Hygiene solution	Active ingredients*	Directions for use
Ricinus. communis 2%	castor oil rich in fatty acids, of which C18:1OH-ricinoleic is predominant (~85.0%)	20 min[19,20,22]
Ricinus communis 10%		
Cepacol™	water, alcohol denat. 14%, glycerin, cetylpyridinium chloride (Ceepryn) 0.05%, flavors, sodium phosphate, disodium phosphate, Polysorbate 80, saccharin, disodium EDTA, FD&C Yellow No. 5 (tartrazine)	10 min[3,12]
NitrAdine™	citric acid, sodium lauryl sulfate, lactose monohydrate, sodium bicarbonate, sodium chloride, potassium hydrogen monopersulfate	15 min**

EDTA – ethylenediaminetetraacetic acid; * according to the manufacturer's information; ** according to the manufacturer's instructions.

the *Candida* spp. and bacteria. The inoculum was prepared in BHI Broth at a cell concentration of 10^7 CFU/mL for bacteria and 10^6 CFU/mL for *Candida* spp. As in the case of the mono-species biofilms, the specimens were randomly assigned into 12-well cell culture plates and filled with 2 mL of the inoculated culture medium.

To attest the sterility of the experiment, one additional specimen received a sterile culture medium. The specimens were kept in an incubator (Shaker Incubator CE-320; Cienlab, Campinas, Brazil) at 37°C for 90 min under agitation (75 rpm) for the adhesion period. After this period, the specimens were washed twice with PBS and the same volume of a sterile culture medium was added to the wells. The plates were re-incubated for 48 h. After 24 h, 1 mL of the culture medium was removed, and the same volume of a fresh culture medium was added to the wells. All cultivation steps were performed in a microaerophilic environment.

Hygiene protocols

The specimens were transferred to sterile perforated stainless-steel baskets[30] and placed inside containers with 150 mL of a cleanser solution, remaining fully immersed. An adapted stainless-steel wire allowed the baskets to remain suspended and not touch the bottom of the container. Immersion in the *R. communis*-based solutions and CPC was performed for 20 min and 10 min, respectively. The immersion times were chosen based on the results of previous studies, which demonstrated both antibiofilm effects and the absence of adverse effects for the hygiene solutions.[3,12,19,20,22] Immersion in Ni was performed for 15 min, according to the manufacturer's instructions. Immersion in distilled water for 20 min was used as a control, and the rationale for the immersion time was based on the longest evaluated period. The specimens of negative control (without contamination) were also immersed in distilled water for 20 min. At the end of the immersion periods, the specimens were rinsed 3 times with sterile PBS to eliminate cleanser residues.

Viability assay

After immersion, the specimens were transferred to a tube containing 10 mL of the Letheen Broth medium (BD Difco™, Sparks, USA). The tubes were sonicated (200W, 40 KHz) (Clean 9CA; Altsonic, Ribeirão Preto, Brazil) for 20 min to detach the remaining microorganisms. The resulting suspension was vortexed for 30 s, and serial dilutions (10^{-1} to 10^{-4}) were seeded in a selected culture medium: for *C. albicans* and *C. glabrata* – CHROMagar Candida Medium (CAC) (BD Difco); for *S. aureus* – Mannitol Salt Agar (MSA) (HiMedia), supplemented with nystatin (200 U/mL); and for *S. mutans* – Mitis Salivarius Agar (HiMedia), supplemented with nystatin (200 U/mL) and bacitracin (0.2 U/mL). The plates were incubated at 37°C for 48 h. The incubation of *S. mutans* was performed in microaerophilic conditions. The number of colonies was registered and expressed in $\log_{10}$CFU/mL.

Biofilm removal capacity

Since elevated resistance to the hygiene protocols was observed in the multi-species biofilm, an evaluation of the biofilm removal capacity was performed in this case. Thus, the specimens with the multi-species biofilm were analyzed by visualizing the amount of live and dead cells on the surfaces of the specimens. After conducting the hygiene protocols, 2 specimens from each group were transferred to a new 12-well plate and stained with 1.5 mL of LIVE/DEAD BacLight™ Kit (Invitrogen Molecular Probes, Eugene, USA), prepared according to the manufacturer's instructions. Briefly, the working solution was prepared by adding 3 µL of the SYTO® 9 stain and 3 µL of the propidium iodide stain to 1 mL of distilled-sterilized water.

The plates were incubated for 15 min at room temperature, protected from light. The surfaces of the specimens were subsequently washed with PBS and analyzed under an inverted fluorescence microscope (Carl Zeiss, Oberkochen, Germany) with the appropriate filters. Twenty random fields were captured at ×630 magnification to quantify the total area occupied by green and red cells. Images were captured with the ZEN Lite software, v. 2.3 (Carl Zeiss), and the biofilm-covered areas [μm^2] were quantified with the AxioVision software, v. 4.8.2 (Carl Zeiss). Since all cells are dyed green, the area was considered as the total biofilm (live and dead cells). Red staining indicated dead cells. The area of the living biofilm was calculated as the difference between the green-stained cell area and the red-stained cell area.[14]

Statistical analysis

At first, the data was tested to check for normal and homogeneous distribution by the Shapiro–Wilk and Levene tests, respectively. According to distribution, the Kruskal–Wallis test, followed by Dunn's post-test, or the analysis of variance (ANOVA), followed by Tukey's post-test were used to compare the results. Statistical analysis was performed using the IBM SPSS for Windows software, v. 21.0 (IBM Corp., Armonk, USA), at a significance level of 0.05.

Results

Viability of microorganisms

Based on the analysis of the biofilm viability, Ni showed the strongest antimicrobial action. In comparison with the control group, immersion in Ni reduced the microbial load of *C. glabrata* (2.18 log; $p < 0.001$), *S. aureus*

(1.37 log; p = 0.012) and *S. mutans* (4.38 log; p = 0.002) when grown singly. The solution was also effective against *C. glabrata* (1.20 log; p = 0.006) and *S. mutans* (4.67 log; p = 0.010) when grown in association with other species. Cepacol promoted reduction in the viability of *C. albicans* (1.08 log; p = 0.018), *C. glabrata* (1.82 log; p = 0.001) and *S. aureus* (4.14 log; p < 0.001) grown in the mono-species biofilms. Regarding the multi-species biofilm, antimicrobial action was observed only against *C. glabrata* (1.34 log; p = 0.035). The experimental hygiene solutions RC02 and RC10 were not effective in reducing the viability of microorganisms grown in different biofilm patterns (Fig. 1 and Table 2).

Growth in association with different species seems to have increased the resistance of *C. albicans* and *S. aureus*, since CPC and Ni did not reduce the viability of the microorganisms in the multi-species biofilm, as happened for single biofilms. In contrast, this behavior was not observed for *C. glabrata* and *S. mutans* (Table 2).

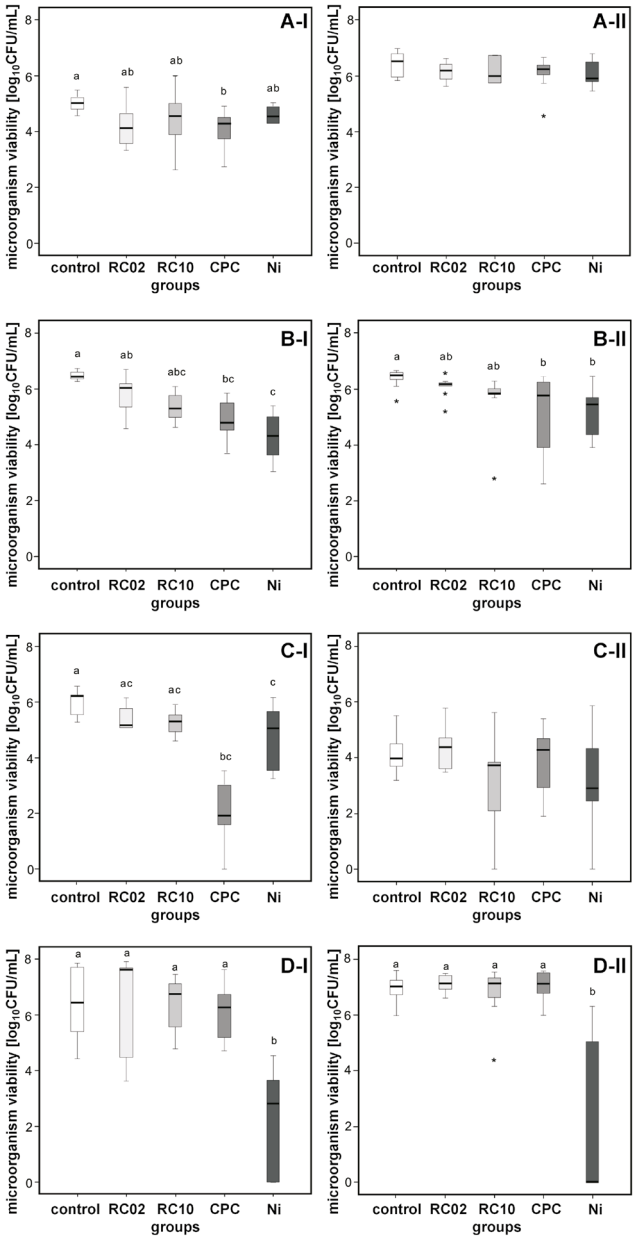

Fig. 1. Comparative analysis of microorganism viability [log_{10}CFU/mL] in the mono-species (I) and multi-species biofilms (II) after immersion in different hygiene solutions

A – *Candida albicans*; B – *Candida glabrata*; C – *Staphylococcus aureus*; D – *Streptococcus mutans*.
Groups: RC02 – *Ricinus communis* 2%; RC10 – *Ricinus communis* 10%; CPC – Cepacol™ ; Ni – NitrAdine™. Different lowercase letters indicate a statistically significant difference.

Biofilm removal capacity

With regard to the biofilm removal capacity, lower rates of live biofilm (green-stained cells) could be observed in comparison with the rates of total biofilm (green- and red-stained cells) (p < 0.001). In agreement with the viability results, Ni promoted a considerable reduction of the living biofilm (p < 0.001). Cepacol, RC2 and RC10 presented moderate efficacy in reducing the amount of the living biofilm. When the total biofilm areas were compared, it was found that Ni resulted in a greater removal of the biofilm than other solutions (Table 3). After immersion in all different hygiene solutions, a large amount of the aggregated dead biofilm (red-stained cells) remained, covering an extensive portion of the surfaces of the specimens (Fig. 2). This finding indicates that, although Ni and CPC reduced cell viability (green-stained cells), they could not widely eliminate the biofilm from the surfaces of the specimens.

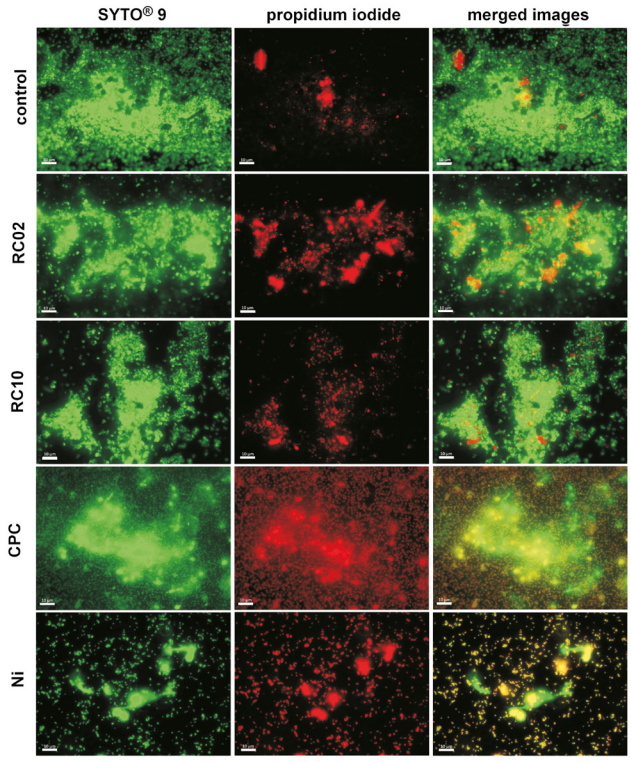

Fig. 2. Representative fluorescent microscopy micrographs of the multi-species biofilm grown on the cobalt-chromium (Co-Cr) surfaces immersed in different hygiene solutions (the cells were stained with the LIVE/DEAD BacLight™ Kit) ×630 magnification.

Table 2. Biofilm viability [log_{10}CFU/mL] after immersion in different hygiene solutions

Biofilm model	Microorganisms	Hygiene solutions	*M* ±*SD* (*Me*)	95% *CI* (range)	*p*-value	Pairwise comparisons
Mono-species biofilm	*C. albicans*	control	5.15 ±0.51 (5.03)	4.82; 5.49 (4.56–6.30)	0.007**	control vs. CPC: 0.018
		R. communis 2%	4.23 ±0.83 (4.12)	3.59; 4.87 (3.32–5.59)		
		R. communis 10%	4.49 ±1.06 (4.55)	3.67; 5.30 (2.64–6.00)		
		Cepacol	4.07 ±0.71 (4.29)	3.60; 4.53 (2.75–4.89)		
		NitrAdine	4.48 ±0.60 (4.54)	4.02; 4.93 (3.08–5.03)		
	C. glabrata	control	6.41 ±0.91 (5.59)	4.64; 6.03 (3.00–6.14)	0.004**	control vs. CPC: 0.001 control vs. Ni: <0.001 RC02 vs. Ni: 0.010
		R. communis 2%	5.33 ±0.49 (5.29)	4.96; 5.72 (4.62 – 6.08)		
		R. communis 10%	4.76 ±0.68 (5.06)	4.23; 5.28 (3.64–5.70)		
		Cepacol	4.59 ±0.37 (4.64)	4.31; 4.87 (4.00–5.19)		
		NitrAdine	4.23 ±0.82 (4.31)	3.60; 4.85 (3.03–5.38)		
	S. aureus	control	5.99 ±0.43 (6.21)	5.65; 6.32 (5.28–6.54)	0.014*	control vs. CPC: <0.001 control vs. Ni: 0.012 RC02 vs. CPC: 0.014 RC10 vs. CPS: 0.015
		R. communis 2%	5.28 ±0.62 (5.16)	4.80; 5.75 (4.00–6.12)		
		R. communis 10%	5.24 ±0.41 (5.27)	4.92; 5.56 (4.61–5.89)		
		Cepacol	1.85 ±1.57 (2.30)	0.60; 3.11 (0.00–3.51)		
		NitrAdine	4.62 ±1.16 (5.05)	3.73; 5.51 (3.26–6.15)		
	S. mutans	control	6.41 ±1.27 (6.42)	5.43; 7.38 (4.43–7.84)	<0.001*	control vs. Ni: 0.002 RC02 vs. Ni: 0.005 RC10 vs. Ni: 0.001 CPC vs. Ni: 0.033
		R. communis 2%	6.41 ±1.82 (7.61)	5.01; 7.80 (3.64–7.90)		
		R. communis 10%	6.40 ±1.01 (6.73)	5.62; 7.18 (4.80 – 7.43)		
		Cepacol	6.02 ±0.98 (6.26)	5.27; 6.78 (4.70–7.60)		
		NitrAdine	2.03 ±2.00 (2.81)	0.49; 3.57 (0.00–4.53)		
Multi-species biofilm	*C. albicans*	control	6.42 ±0.42 (6.52)	6.10; 6.75 (5.85–6.97)	0.463*	–
		R. communis 2%	6.05 ±0.54 (6.19)	5.64; 6.47 (4.86–6.61)		
		R. communis 10%	5.70 ±1.28 (5.99)	4.72; 6.68 (3.36–6.74)		
		Cepacol	6.05 ±0.62 (6.24)	5.58; 6.53 (4.55–6.65)		
		NitrAdine	5.96 ±0.68 (5.92)	5.44; 6.48 (4.56–6.78)		
	C. glabrata	control	6.40 ±0.39 (6.48)	6.10; 6.70 (5.54–6.92)	0.004*	control vs. CPC: 0.035 control vs. Ni: 0.006
		R. communis 2%	6.08 ±0.38 (6.17)	5.78; 6.37 (5.18–6.56)		
		R. communis 10%	5.58 ±1.07 (5.83)	4.76; 6.40 (2.78–6.27)		
		Cepacol	5.06 ±1.39 (5.76)	3.99; 6.14 (2.60–6.45)		
		NitrAdine	5.20 ±0.88 (5.45)	4.52; 5.87 (3.90–6.45)		
	S. aureus	control	4.13 ±0.70 (3.97)	3.59; 4.67 (3.19–5.49)	0.349*	control vs. Ni: 0.010 RC02 vs. Ni: 0.003 RC10 vs. Ni: 0.010 CPC vs. Ni: 0.001
		R. communis 2%	4.35 ±0.78 (4.37)	3.75; 4.95 (3.49–5.76)		
		R. communis 10%	3.13 ±1.74 (3.72)	1.79; 4.47 (0.00–5.61)		
		Cepacol	3.89 ±1.15 (4.27)	3.01; 4.77 (1.91–5.39)		
		NitrAdine	3.20 ±1.72 (2.88)	1.88; 4.52 (0.00–5.84)		
	S. mutans	control	6.94 ±0.48 (7.01)	6.58; 7.31 (5.98–7.57)	<0.001*	–
		R. communis 2%	7.09 ±0.33 (7.13)	6.83; 7.35 (6.60–7.47)		
		R. communis 10%	6.75 ±0.97 (7.12)	6.00; 7.50 (4.37–7.51)		
		Cepacol	7.02 ±0.57 (7.11)	6.59; 7.46 (6.00–7.55)		
		NitrAdine	2.27 ±2.75 (0.00)	0.16; 4.39 (0.00–6.30)		

M – mean; *SD* – standard deviation; *Me* – median; *CI* – confidence interval; * Kruskal–Wallis test and Dunn's post-test; ** ANOVA and Tukey's post-test.

Table 3. Biofilm-covered areas [%] after immersion in different hygiene solutions

Biofilm	Hygiene solutions	*M* ±*SD* (*Me*)	95% *CI* (range)
Living	control	41.81 ± 10.34 (40.56)^{a*}	37.10; 46.51 (22.38–61.66)
	R. communis 2%	20.62 ±7.41 (21.16)^{b*}	17.25; 24.00 (8.43–32.44)
	R. communis 10%	16.94 ±6.28 (17.08)^{b*}	14.09; 19.80 (5.61–32.66)
	Cepacol	17.55 ±11.56 (17.45)^{b*}	12.29; 22.81 (4.63–43.82)
	NitrAdine	7.42 ±5.56 (6.40)^{c*}	4.89; 9.96 (0.00–21.31)
Total	control	64.96 ±12.08 (67.36)^{A}	59.46; 70.46 (32.07–81.49)
	R. communis 2%	46.16 ±9.43 (45.27)^{B}	41.87; 50.46 (27.55–65.09)
	R. communis 10%	36.70 ±12.77 (32.74)^{B}	30.89; 42.51 (19.88 – 65.77)
	Cepacol	45.24 ±11.05 (44.35)^{B}	40.21; 50.27 (27.08–66.44)
	NitrAdine	20.33 ±9.15 (18.56)^{C}	16.17; 24.50 (10.08–45.47)

Comparisons of the biofilm-covered areas were conducted using the two-way ANOVA with independent levels (the living and total biofilm and the hygiene solutions) and the Bonferroni post-hoc test. Different lowercase letters indicate statistically significant differences among the hygiene solutions in the living biofilm area. Different uppercase letters indicate statistically significant differences among the hygiene solutions in the total biofilm area. * statistically significant difference between the living biofilm area and the total biofilm area for the same hygiene solution ($p < 0.05$).

Discussion

The scientific literature has demonstrated that biofilm development is a remarkable issue in medical device-associated infections.[31] This study was carried out using mono- and multi-species biofilms in order to clarify if biofilms developed by single strains have greater susceptibility to hygiene solutions than those developed by multiple strains. The species association seems to have increased the resistance of *C. albicans* and *S. aureus*, since CPC and Ni had no effect when the microorganisms grew in a multi-species biofilm model.

Promising biological findings involving *R. communis* suggest that the ethanolic, methanolic or hexane fractions obtained from its leaves and seeds can be an alternative source of therapeutic substances.[17,18,21] Previous studies showed that the solutions obtained by the esterification of ricinoleic acid were beneficial for the control of biofilm formation on acrylic resin and silicone surfaces.[19,20,22,23] Nonetheless, the scientific literature has brought to light a cascade of controversial results regarding the concentration of the *R. communis* solution capable of exerting biological effects. The investigated concentrations ranged from 2% to 10%; however, until now, no ideal concentration has been established, leaving researchers struggling with conflicting evidence.[8,19,20,22] Therefore, in this study 2 extreme concentrations were evaluated. What should also be taken into account is the fact that RPDs are composed of artificial teeth, acrylic resin and the dental alloy. Since cell adhesion and biofilm formation depend on the composition of the surface,[32] one cannot assume that hygiene solutions will have the same effect on all surfaces.

The antibiofilm activity of the *R. communis* solutions was slightly disappointing. The solutions only promoted a modest reduction of the viability of *C. albicans*, *C. glabrata* and *S. aureus* in both the mono- and multi-species biofilms, yet the reduction was not statistically significant. Andrade et al., investigating the 2% concentration, indicated that the solution had an intermediate antibiofilm action, comparable to the that of an effervescent tablet (Polident).[19] The authors concluded that single immersion was insufficient for broadly promoting biofilm removal and suggested that association with mechanical brushing would be suitable for better results.[19] Some clinical studies showed biofilm removal capacity, the reduction of the microbial load and the remission of denture-related stomatitis after using *R. communis* solutions.[8,22,24]

The antimicrobial effect of *R. communis* is probably linked to its toxicity, which is attributed to the protein ricin. The seeds have ricin at a percentage of up to 5%; the biological function of the protein is inhibiting protein synthesis.[33] Worbs et al. indicates that ricin removes adenine from the so-called sarcin-ricin loop of 28S rRNA, thereby preventing the binding of elongation factors and further protein synthesis.[33] As reviewed by Yeboah et al., the composition and properties of castor oil vary with respect to the method of extraction, geographical location and the type of cultivar.[34] Thus, in view of these statements, we suggest 2 different reasons to explain the insufficient antimicrobial effect of *R. communis* in this study. First, the discrepancy of results presented by the literature, as well as the absence of antibiofilm activity presented here, might be associated with extraction methods and oil purity. Second, as the antimicrobial effect seems to be attributed to the inhibition of protein synthesis, one single 20-minute application would not alter protein synthesis to the point of presenting reduction in the microbial load.

The mouthwash Cepacol was more effective against the mono-species biofilms. In the multi species biofilm pattern, it only reduced the viability of *C. glabrata*. These findings are in line with microscopy evaluations. The images obtained from the CPC group showed a modest reduction of the multi-species biofilm-covered areas, suggesting a limited disaggregating capacity. Cepacol has 0.05% of cetylpyridinium chloride as an active ingredient. This is a quaternary ammonium compound that affects cell integrity by interfering with osmoregulation and homeostasis. Diverse in vitro studies report the antibacterial activity of Cepacol against planktonic bacteria.[35] The apparent discrepancy between our findings and those of other researchers can be related to the antimicrobial susceptibility of microorganisms in biofilm- and non-biofilm-associated states. Biofilm tolerance to antimicrobial agents is about 100–1,000 times greater as compared to that of the planktonic form.[36]

NitrAdine presented the best antibiofilm action against the largest number of species evaluated. In both the mono- and multi-species biofilms, Ni reduced the viability of *S. mutans* and *C. glabrata* in about 4 log and 1 log, respectively. The *S. aureus* mono-species biofilm was also reduced in about 1 log after immersion in Ni. The antimicrobial effect of Ni is attributed to sodium lauryl sulfate and sodium bicarbonate that act through injuring the microbial cell membrane.[37,38] In addition, another active ingredient of Ni, citric acid, is associated with the capability of disturbing the microbial metabolism.[39] Controversies about the antibiofilm effectiveness of effervescent tablets emerge in the scientific literature. Supporting our findings, Coimbra et al. reported that Ni exhibited satisfactory antibiofilm activity, reducing the microbial load and metabolic activity, and the area covered by the multi-species biofilm composed of *C. albicans*, *S. aureus* and *Pseudomonas aeruginosa*.[14] Effective antibiofilm activity of Ni against the *S. mutans* biofilm in a multi-species biofilm model was demonstrated by Lopes Vasconcelos et al.[11]

In agreement with the viability reduction observed in CFU counts, the microscopy images indicated a significant reduction of the living biofilm after immersion in Ni. The reaction between sodium bicarbonate and citric acid, the active ingredients of Ni, in the presence of water leads to the liberation of carbon dioxide, promoting the effervescent aspect. It has been postulated that the release of effervescence can induce a mechanical effect that disrupts biofilms, which could explain the superior ability of effervescent tablets in removing microbial deposits.[40] Nonetheless, despite significant antimicrobial activity, about 31% of the surfaces of the specimens remained covered by the residual aggregated biofilm after immersion in Ni. This can be interpreted as evident antimicrobial action of Ni, but also as its incapability to completely remove all aggregates. It was suggested by the Council on Dental Materials, Instruments, and Equipment that the release of bubbles from effervescent tablets might promote a mechanical action favoring the detaching of the biofilm from the surface of the prosthesis.[40]

The current study was limited by the fact that biofilms were grown considering only ATCC samples. It is recognized that hygiene solutions should be tested on clinical samples employing multidrug-resistant strains. In addition, single short-time immersion was applied. In light of future studies, we believe that investigating distinct multi-species biofilms is essential, considering the high diversity of the buccal microbiome. Combinations involving members of both Gram-positive and Gram-negative groups, as well as other *Streptococcus* spp. or anaerobic rods, could better represent the microbiome of RPDs. Evaluating the clinical effect of RPD immersion in the tested solutions in association with mechanical biofilm removal is another important aspect to be considered in further studies.

Conclusions

Considering the limitations of the study, the findings clearly illustrate that none of the evaluated solutions was able to widely reduce the viability of the microorganisms and the biofilm-covered areas. Although NitrAdine reduced the viability of the largest number of species, it did not alter the microbial load of *C. albicans*. Cepacol reduced the viability of microorganisms in the mono-species biofilms; however, its action was unsatisfactory in the multi-species biofilms.

Ethics approval and consent to participate

Not applicable.

Data availability

The datasets supporting the findings of the current study are available from the corresponding author on reasonable request.

Consent for publication

Not applicable.

Use of AI and AI-assisted technologies

Not applicable.

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References

1. O'Donnell LE, Millhouse E, Sherry L, et al. Polymicrobial *Candida* biofilms: Friends and foe in the oral cavity. *FEMS Yeast Res*. 2015;15(7):fov077. doi:10.1093/femsyr/fov077
2. Yumoto H, Hirota K, Hirao K, et al. The pathogenic factors from oral streptococci for systemic diseases. *Int J Mol Sci*. 2019;20(18):4571. doi:10.3390/ijms20184571
3. Borges Felipucci DN, Davi LR, Oliveira Paranhos HF, Bezzon OL, Silva RF, Pagnano VO. Effect of different cleansers on the surface of removable partial denture. *Braz Dent J*. 2011;22(5):392–397. doi:10.1590/s0103-64402011000500008
4. Lopes Vasconcelos GL, Curylofo PA, Raile PN, Macedo AP, Oliveira Paranhos HF, Pagnano VO. Effect of alkaline peroxides on the surface of cobalt chrome alloy: An in vitro study. *J Prosthodont*. 2019;28(1):e337–e341. doi:10.1111/jopr.12789
5. Curylofo PA, Raile PN, Lopes Vasconcelos GL, Macedo AP, Pagnano VO. Effect of denture cleansers on cobalt-chromium alloy surface: A simulated period of 5 years' use. *J Prosthodont*. 2020;29(2):142–150. doi:10.1111/jopr.12996
6. Jose A, Coco BJ, Milligan S, et al. Reducing the incidence of denture stomatitis: Are denture cleansers sufficient? *J Prosthodont*. 2010;19(4):252–257. doi:10.1111/j.1532-849X.2009.00561.x

7. de Lucena-Ferreira SC, Ricomini-Filho AP, da Silva WJ, Cury JA, Del Bel Cury AA. Influence of daily immersion in denture cleanser on multispecies biofilm. *Clin Oral Invest*. 2014;18(9):2179–2185. doi:10.1007/s00784-014-1210-9
8. Ferraz de Arruda CN, Salles MM, Badaró MM, et al. Effect of sodium hypochlorite and *Ricinus communis* solutions on control of denture biofilm: A randomized crossover clinical trial. *J Prosthet Dent*. 2017;117(6):729–734. doi:10.1016/j.prosdent.2016.08.035
9. Axe AS, Varghese R, Bosma M, Kitson N, Bradshaw DJ. Dental health professional recommendation and consumer habits in denture cleansing. *J Prosthet Dent*. 2016;115(2):183–188. doi:10.1016/j.prosdent.2015.08.007
10. Linde N. Mouthwash. In: Wexler P, ed. *Encyclopedia of Toxicology*. 2nd ed. Amsterdam, the Netherlands: Elsevier/Academic Press; 2005:162–163.
11. Lopes Vasconcelos GL, Curylofo PA, Targa Coimbra FC, et al. In vitro antimicrobial activity of effervescent denture tablets on the components of removable partial dentures. *Int J Prosthodont*. 2020;33(3):315–320. doi:10.11607/ijp.6436
12. de Freitas Oliveira Paranhos H, Bezzon OL, Davi LR, Borges Felipucci DN, Lovato da Silva CH, Pagnano VO. Effect of cleanser solutions on the color of acrylic resins associated with titanium and nickel-chromium alloys. *Braz Oral Res*. 2014;28:S1806-83242014000100234. doi:10.1590/1807-3107bor-2014.vol28.0017
13. Silva-Lovato CH, de Wever B, Adriaens E, et al. Clinical and antimicrobial efficacy of NitrAdine™-based disinfecting cleaning tablets in complete denture wearers. *J Appl Oral Sci*. 2010;18(6):560–565. doi:10.1590/s1678-77572010000600005
14. Coimbra FC, Rocha MM, Oliveira VC, et al. Antimicrobial activity of effervescent denture tablets on multispecies biofilms. *Gerodontology*. 2021;38(1):87–94. doi:10.1111/ger.12500
15. Wu SC, Liu F, Zhu K, Shen JZ. Natural products that target virulence factors in antibiotic-resistant *Staphylococcus aureus*. *J Agric Food Chem*. 2019;67(48):13195–13211. doi:10.1021/acs.jafc.9b05595
16. Atanasov AG, Waltenberger B, Pferschy-Wenzig EM, et al. Discovery and resupply of pharmacologically active plant-derived natural products: A review. *Biotechnol Adv*. 2015;33(8):1582–1614. doi:10.1016/j.biotechadv.2015.08.001
17. Hussein HM, Hameed RH, Hameed IH. Screening of bioactive compounds of *Ricinus communis* using GC-MS and FTIR and evaluation of its antibacterial and antifungal activity. Indian J Public Health Res Dev. 2018;9(5):463–469. doi:10.5958/0976-5506.2018.00488.6
18. Abdul WM, Hajrah NH, Sabir JS, et al. Therapeutic role of *Ricinus communis* L. and its bioactive compounds in disease prevention and treatment. *Asian Pac J Trop Med*. 2018;11(3):177–185. doi:10.4103/1995-7645.228431
19. de Andrade IM, de Andrade KM, Pisani MX, Silva-Lovato CH, de Souza RF, de Freitas Oliveira Paranhos H. Trial of an experimental castor oil solution for cleaning dentures. *Braz Dent J*. 2014;25(1):43–47. doi:10.1590/0103-6440201302327
20. Salles MM, de Cássia Oliveira V, Souza RF, Silva-Lovato CH, de Freitas Oliveira Paranhos H. Antimicrobial action of sodium hypochlorite and castor oil solutions for denture cleaning – in vitro evaluation. *Braz Oral Res*. 2015;29:1–6. doi:10.1590/1807-3107BOR-2015.vol29.0104
21. Umaarasu T, Padmavathy K, Thirunavukkarasu D, Rajesh SV, Govindaraj J, Shanmugam G. Evaluation of the antimicrobial activity and phytochemical investigation of the leaf extracts of *Ricinus communis* Linn. against drug-resistant bacterial pathogens. *Drug Invent Today*. 2019;11(6):1299–1303.
22. Badaró MM, Salles MM, Fagundes Leite VM, et al. Clinical trial for evaluation of *Ricinus communis* and sodium hypochlorite as denture cleanser. *J Appl Oral Sci*. 2017;25(3):324–334. doi:10.1590/1678-7757-2016-0222
23. Pinheiro JB, Vomero MP, do Nascimento C, et al. Genomic identification of microbial species adhering to maxillofacial prostheses and susceptibility to different hygiene protocols. *Biofouling*. 2018;34(1):15–25. doi:10.1080/08927014.2017.1403591
24. Pinelli LA, Montandon AA, Corbi SC, Moraes TA, Fais LM. *Ricinus communis* treatment of denture stomatitis in institutionalized elderly. *J Oral Rehabil*. 2013;40(5):375–380. doi:10.1111/joor.12039
25. Neppelenbroek KH. The importance of daily removal of the denture biofilm for oral and systemic diseases prevention. *J Appl Oral Sci*. 2015;23(6):547–548. doi:10.1590/1678-77572015ed006
26. Lohse MB, Gulati M, Johnson AD, Nobile CJ. Development and regulation of single- and multi-species *Candida albicans* biofilms. *Nat Rev Microbiol*. 2018;16(1):19–31. doi:10.1038/nrmicro.2017.107
27. Schlecht LM, Peters BM, Krom BP, et al. Systemic *Staphylococcus aureus* infection mediated by *Candida albicans* hyphal invasion of mucosal tissue. *Microbioliology (Reading)*. 2015;161(Pt 1):168–181. doi:10.1099/mic.0.083485-0
28. Costa L, do Nascimento C, Pagnano de Souza VO, Pedrazzi V. Microbiological and clinical assessment of the abutment and non-abutment teeth of partial removable denture wearers. *Arch Oral Biol*. 2017;75:74–80. doi:10.1016/j.archoralbio.2016.11.002
29. O'Donnell LE, Smith K, Williams C, et al. Dentures are a reservoir for respiratory pathogens. *J Prosthodont*. 2016;25(2):99–104. doi:10.1111/jopr.12342
30. Oliveira Paranhos HF, Silva-Lovato CH, de Souza RF, et al. Effect of three methods for cleaning dentures on biofilms formed in vitro on acrylic resin. *J Prosthodont*. 2009;18(5):427–431. doi:10.1111/j.1532-849X.2009.00450.x
31. Stewart PS, Bjarnsholt T. Risk factors for chronic biofilm-related infection associated with implanted medical devices. *Clin Microbiol Infect*. 2020;26(8):1034–1038. doi:10.1016/j.cmi.2020.02.027
32. Zhou Y, Xiao Y, Qiu Y, et al. Adhesion and proliferation of cells and bacteria on microchip with different surfaces microstructures. *Biomed Tech (Berl)*. 2016;61(5):475–482. doi:10.1515/bmt-2015-0075
33. Worbs S, Köhler K, Pauly D, et al. *Ricinus communis* intoxications in human and veterinary medicine – a summary of real cases. *Toxins (Basel)*. 2011;3(10):1332–1372. doi:10.3390/toxins3101332
34. Yeboah A, Ying S, Lu J, et al. Castor oil (*Ricinus communis*): A review on the chemical composition and physicochemical properties. *Food Sci Technol*. 2020;41(2):399–413. doi:10.1590/fst.19620
35. Silva VC, de Cássia Oliveira V, Lopes de Sousa AF, et al. Prevalence and susceptibility profile of *Candida* spp. isolated from patients in cancer therapy. *Arch Oral Biol*. 2020;119:104906. doi:10.1016/j.archoralbio.2020.104906
36. Macià MD, Rojo-Molinero E, Oliver A. Antimicrobial susceptibility testing in biofilm-growing bacteria. *Clin Microbiol Infect*. 2014;20(10):981–990. doi:10.1111/1469-0691.12651
37. Pellizzaro D, Polyzois G, Machado AL, Giampaolo ET, Sanitá PV, Vergani CE. Effectiveness of mechanical brushing with different denture cleansing agents in reducing in vitro *Candida albicans* biofilm viability. *Braz Dent J*. 2012;23(5):547–554. doi:10.1590/s0103-64402012000500013
38. Dhamande MM, Pakhan AJ, Thombare RU, Ghodpage SL. Evaluation of efficacy of commercial denture cleansing agents to reduce the fungal biofilm activity from heat polymerized denture acrylic resin: An in vitro study. *Contemp Clin Dent*. 2012;3(2):168–172. doi:10.4103/0976-237X.96820
39. Izumi S, Ryu M, Ueda T, Ishihara K, Sakurai K. Evaluation of application possibility of water containing organic acids for chemical denture cleaning for older adults. *Geriatr Gerontol Int*. 2016;16(3):300–306. doi:10.1111/ggi.12467
40. Council on Dental Materials, Instruments, and Equipment. Denture cleansers. *J Am Dent Assoc*. 1983;106(1):77–79. PMID:6574171.

Original paper

Effects of mechanical and chemical hygiene methods on the O-ring system-retained overdenture: A simulated two-year evaluation

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Abstract

Background. Cleaning overdentures is challenging due to their complex metallic structures, which often create small and irregular areas that are difficult to clean. Thus, it is necessary to find an effective and safe method for their maintenance.

Objectives. The aim of the study was to evaluate the effects of hygiene methods on the dimensional changes and retention force of the O-ring system over a simulated two-year period.

Material and methods. A total of 54 specimens with 2 O-rings each were distributed into 9 groups: control – no cleaning; brushing – blue®m toothpaste (BM); brushing – Trihydral toothpaste (TR); immersion – NitrAdine® (Ni); immersion – 5 mg/mL chitosan solution (Ch); and combined methods: BM+Ni, BM+Ch, TR+Ni, TR+Ch. The deformation and retention force were evaluated every 6 months for 2 years. The statistical analysis was performed using two-way analysis of variance (ANOVA) and Pearson's correlation test ($\alpha = 0.05$).

Results. The BM+Ch group exhibited smaller changes in diameter after 6 months (Δ(T1–T0)) than the control and BM+Ni groups, both internally and externally ($p < 0.001$). In addition, the BM+Ch group demonstrated greater retention forces at 6 months ($p = 0.024$) and 18 months ($p = 0.028$) in comparison to the TR+Ch group. The TR+Ch group exhibited a negative correlation with changes in the external diameter ($r = -0.334$, $p = 0.021$), while BM+Ch showed a negative correlation with changes in the internal diameter ($r = -0.334$, $p = 0.021$). The Ni group demonstrated a negative relationship with changes in both the internal ($r = -0.296$, $p = 0.041$) and external diameters ($r = -0.405$, $p = 0.004$).

Conclusions. A combination of brushing with blue®m toothpaste and immersion in the chitosan solution demonstrated satisfactory results in maintaining the retention force of the O-ring system.

Keywords: denture cleanser, prosthodontics, toothpastes, dental prosthesis

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Highlights

- Cleaning methods influence dimensional changes and retention force of O-rings in implant-supported overdentures.
- Brushing with blue®m toothpaste combined with immersion in chitosan solution provided satisfactory hygiene and retention outcomes.
- Further studies are required to confirm clinical applicability and assess long-term effects of these cleaning methods on overdentures.

Introduction

Edentulism remains a prevalent condition affecting a significant portion of the population. Among the available options for oral rehabilitation, the use of overdentures has emerged as an excellent alternative to conventional complete dentures. Overdentures offer various advantages, including the prevention of soft tissue injuries, enhanced retention, stability, masticatory function, phonetics, and esthetics. Success rates for overdentures range between 94% and 100%.[1]

The O-ring system comprises an attachment connected to the implant and a metallic capsule with an internal silicone ring that is retained in the prosthesis.[2] This system effectively distributes masticatory forces among the supporting structures and is favored by patients due to its ease of insertion and removal.[2]

Proper hygiene of overdentures is crucial for the success and longevity of the treatment.[3–5] Regular recalls and maintenance of gingival health are also essential.[6] The literature describes various hygiene protocols, with mechanical hygiene (brushing) being the most commonly used method, and the combined approach involving both mechanical and chemical methods as the most effective in eliminating biofilm and preventing periodontal disease.[7–16] Therefore, the significance of denture cleansers and toothpastes in maintaining oral health cannot be neglected.[17,18]

Given the absence of conventional toothpastes designed for the maintenance of overdentures, the present study focuses on products such as Trihydral and blue®m, which are suitable for both prosthesis and oral cavity hygiene without causing metal degradation. Effervescent tablets for dentures, particularly NitrAdine®, have been frequently recommended as a chemical hygiene method. NitrAdine® has demonstrated effectiveness in cleaning removable partial dentures containing metal, as well as overdentures.[19–21] However, since overdentures are composed of various materials, and few studies have examined the effects of cleansers on these prostheses,[8,22–26] it is necessary to evaluate the efficacy of denture cleansers to recommend an effective and safe method for patients with overdentures. A recent systematic review indicated that hygiene solutions have the most significant impact on retention for the bar–clip attachments in different materials.[27]

To explore alternative immersion solutions that are natural, easy to use, accessible, and cost-effective, researchers have investigated the potential of chitosan. Chitosan, a natural biopolymer with proven antimicrobial properties, is found in abundance in nature and has been applied in various fields, including pharmaceuticals, foods, cosmetics, and chemicals.[28–32]

Despite the significance of overdenture hygiene, a consensus on the most effective hygiene method remains elusive.[33] Studies have reported conflicting outcomes with the use of effervescent tablets,[5,21] sodium bicarbonate/sodium perborate,[34] mouthwash containing cetylpyridinium chloride and fluoride, and sodium hypochlorite, with consequent changes in the surface of the O-ring capsules.[24,25,35] However, while the combined mechanical and chemical hygiene method is regarded the most suitable for the hygiene of dentures, there is a paucity of literature addressing its application in overdentures.[36–38]

The present study aims to evaluate the effects of isolated and combined hygiene methods on the dimensional changes and retention force of the O-ring system over a simulated two-year period. The null hypothesis posits that the utilization of these methods will not have a significant influence on the O-ring diameters or retention force.

Material and methods

Experimental design

Fifty-four specimens of acrylic resin simulating prosthetic bases, with 2 O-rings each, were obtained[39] and articulated. Each base contained 2 implants (grand morse™; Neodent Serviços Odontológicos LTDA, Curitiba, Brazil). In a previous study, the data indicated that the difference in the response of matched pairs is normally distributed with a standard deviation (*SD*) of 0.88.[35] If the true difference in the mean response of matched pairs is 1.38, 5 pairs of subjects would be needed to reject the null hypothesis

that this response difference is 0 with a probability (power) of 0.8. The type I error probability associated with this test of the null hypothesis is 0.05. Thus, 6 specimens per group were selected based on the sample size of studies employing the same methodology.[35–37]

The specimens were divided into 9 groups (Table 1). Each group, encompassing 6 samples, underwent a hygiene process and had outcomes measured at 4 different times. The selected O-ring contains the pink O-ring rubber (code 102.107), which the manufacturer specifies as having low retention. The resin bases with the implants (Fig. 1) were obtained from condensation silicone impressions (Perfil; Vigodent SA Indústria e Comércio, Rio de Janeiro, Brazil) of metallic bases. The O-ring retainers were installed over the implants with a torque of 20 Ncm.[35]

Table 1. Distribution of the evaluated groups

Intervention	Group
No cleaning	control
Mechanical brushing	blue®m
	Trihydral
Chemical immersion	NitrAdine®
	chitosan
Combined (brushing + immersion)	blue®m + NitrAdine®
	blue®m + chitosan
	Trihydral + NitrAdine®
	Trihydral + chitosan

Initial measurements

Before simulation, measurements were obtained through stereomicroscopic analysis (SMZ800; Nikon, Melville, USA). The O-rings were subjected to a visual examination, and images were captured using the NIS-Elements for Basic Research software (Nikon) at a magnification of ×20 to evaluate any dimensional changes that occurred due to the cleaning procedures. The horizontal and vertical distances of the internal and external diameters of the O-rings were measured as demonstrated in Fig. 2.

The specimens were also subjected to the retention force analysis using a mechanical testing machine equipped with a 196-N load cell. The machine performed 20 insertion/removal cycles per minute,[38] resulting in 216 readings (9 groups × 4 times × 6 repetitions). The base that simulated the mandible was fitted and fixed in an acrylic container filled with water at 37°C. Subsequently, the testing machine performed the insertion and removal of the specimen in the base.

The experimental design of the study is presented in Fig. 3.

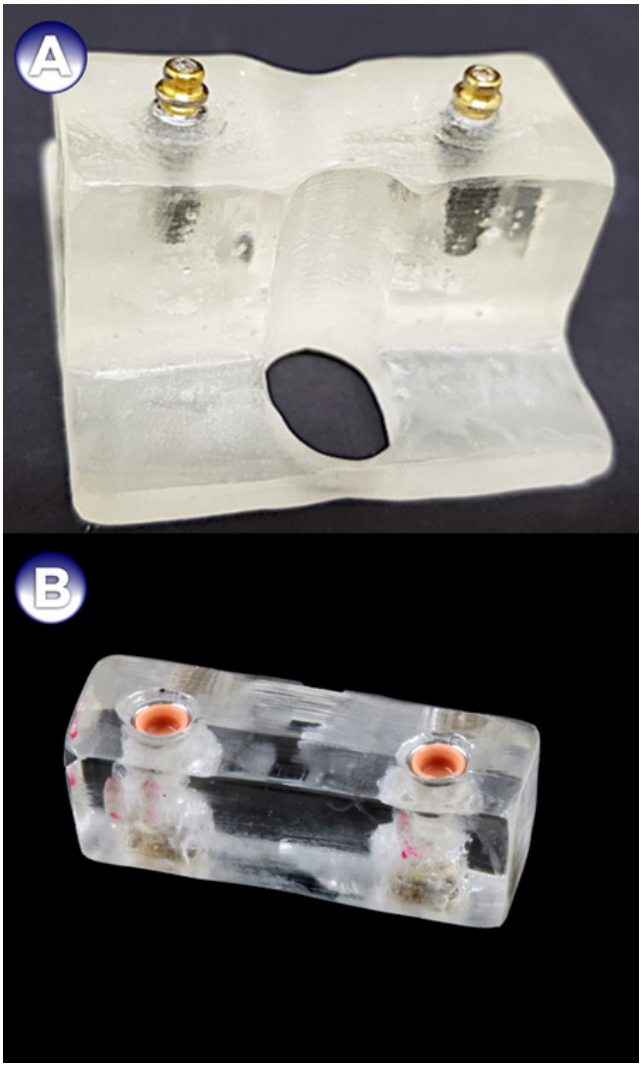

Fig. 1. Resin bases used in the study
A. Base with implants and fittings; B. Capsule base with specimens.

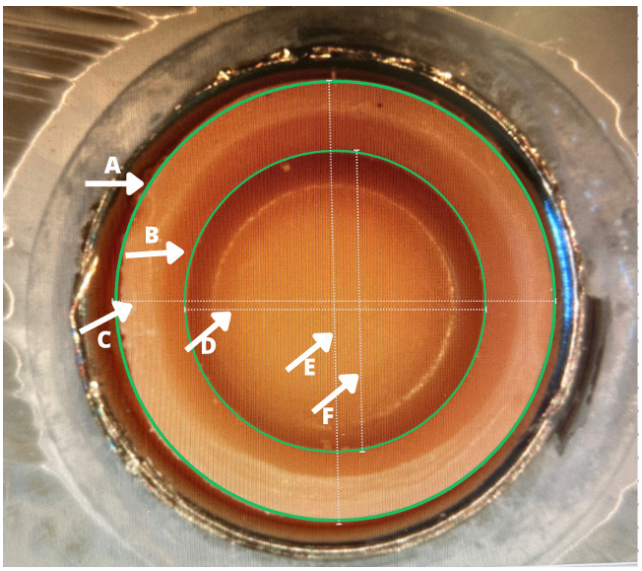

Fig. 2. Measurement of the internal and external diameters of the O-rings
A. Outer circle; B. Inner circle; C. External horizontal distance; D. Internal horizontal distance; E. External vertical distance; F. Internal vertical distance.

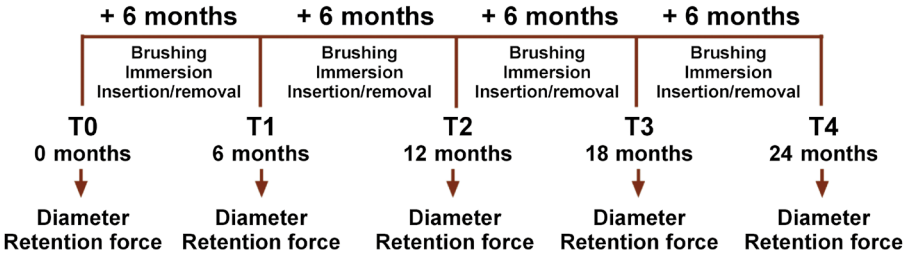

Fig. 3. Experimental design of the study

Cleaning

Mechanical hygiene

The mechanical cleaning method involved the use of an artificial brushing machine to standardize the strength and frequency of the brushing (MAVTEC Brush Simulator; Mavtec Comércio de Peças Acessórios Serviços Ltda ME, Ribeirão Preto, Brazil). Additionally, soft-bristle toothbrushes (Colgate Slim Soft; Colgate-Palmolive Company, Rio de Janeiro, Brazil) were utilized. Two types of toothpaste were used: blue®m (Bluem Brazil, Curitiba, Brazil); and Trihydral (Perland Pharmacos Ltda, Cornélio Procópio, Brazil). The toothpastes were suspended in distilled water at a ratio of 1:1 (Table 1).[18,40] To calculate the brushing time at the frequency of 356 rotations per minute, 8,600 cycles were performed, which corresponded to 6 months of brushing exposure. This cycle was repeated until 2 years of use. The toothbrushes and toothpaste solutions were replaced at the conclusion of each 6-month cycle.[12,39]

Chemical hygiene

Two methods for the chemical hygiene of overdentures were employed. The first technique involved the immersion of the specimens for 12 h in a solution containing effervescent tablets (NitrAdine®; MST Laboratories AG, Vaduz, Liechtenstein) diluted in 150 mL of water at an average temperature of 37°C.[41] This exposure is equivalent to a routine use of the product twice a week for 15 min, according to the manufacturer's instructions, thereby simulating a period of 6 months. During the immersion, the solution was not changed because studies on the degradation curve of the tablet indicated that the product remains highly effective within the specified time frame.[35]

The second immersion method entailed the use of an experimental solution of chitosan at a concentration of 5 mg/mL.[28] For this purpose, low molecular weight chitosan (Sigma-Aldrich, St. Louis, USA) was utilized. A daily 15-min immersion routine was simulated for a period equivalent to 6 months (42 h) (Table 2).

Combined hygiene methods

In the combined hygiene protocols, mechanical methods were first used, followed by chemical methods, as described in the methodology.[38]

Outcomes of interest

After each 6-month simulation period, measurements were conducted once more using stereomicroscopy (SMZ800; Nikon) as previously described.

The specimens were also subjected to the retention force analysis using a mechanical testing machine. A total of 720 cycles were performed to simulate 6 months of denture use, based on the assumption of 4 insertions and removals of the prosthesis by the patient per day.[40] The force required for each insertion/removal cycle was recorded using the LabVIEW 8.0 software (National Instruments, Austin, USA) connected to the load cell of the testing machine.

Table 2. Products used the study

Commercial name	Manufacturer	Composition
NitrAdine®	MST Laboratories AG, Vaduz, Liechtenstein	citric acid, sodium lauryl sulfate, lactose monohydrate, sodium bicarbonate, sodium chloride, potassium monopersulfate, sodium carbonate, mint flavor, PVP
Chitosan	Sigma-Aldrich, St. Louis, USA	water, low molecular weight chitosan (5 mg/mL), and negligible amounts of peracetic acid and calcium hydroxide
blue®m	Bluem Brazil, Curitiba, Brazil	glycerin, water, hydrated silica, honey, silica, PEG-32, aroma, cocamidopropyl betaine, cellulosic gum, magnesium sulfate, sodium sulfate, sodium citrate, lactoferrin, sodium saccharin, xylitol, methyl salicylate, sodium chloride, sodium perborate, sodium methylparaben, citric acid, limonene, CI 42090
Trihydral	Perland Pharmacos Ltda, Cornélio Procópio, Brazil	1500 ppm of fluoride, monofluorophosphate, chloramine-T (0.2%), calcium carbonate, propylene glycol, sorbitol, hydroxyethylcellulose, tetrasodium, pyrophosphate, sodium lauryl sulfate, decyl polyglucoside, methylparaben, sodium saccharin, flavor, water, sodium monofluorophosphate

PVP – polyvinylpyrrolidone; PEG – polyethylene glycol.

Statistical analysis

The data analysis was performed using the IBM SPSS Statistics for Windows software, v. 21.0 (IBM Corp., Armonk, USA). The mean values, which exhibited a normal distribution as indicated by the Shapiro–Wilk test, were compared using two-way analysis of variance (ANOVA) and Tukey's tests with Bonferroni correction. The Pearson's correlation (r) test was applied to verify the correlation between the dimensional changes of the diameters and the retention force ($\alpha = 0.05$ for all tests).

Results

The results of this study demonstrated that the dimensional changes of the internal (ID) and external diameters (ED) of the O-ring system were affected by immersion ($p < 0.05$), time ($p < 0.001$) and brushing techniques ($p < 0.001$). Likewise, the retention force was found to be time-dependent, exhibiting interactions with both immersion and brushing techniques ($p = 0.005$).

Immersion treatments demonstrated a better preservation of the ID ($p = 0.012$) in comparison to the absence of immersion, with the use of NitrAdine® yielding a smaller change in ID (26.2 µm (SD = 54.2)) after 6 months (Δ(T1–T0)) compared to the no immersion group, which exhibited a mean change of 40.4 µm ($SD = 7.8$) ($p = 0.009$). Similarly, changes in the ED were significantly less pronounced in the presence of NitrAdine®, with a mean change of –19.3 µm ($SD = 12.3$), in contrast to the no immersion group's mean change of 12.0 µm ($SD = 22.5$) at Δ(T1–T0) ($p = 0.004$).

The brushing method analyses revealed distinct dimensional alterations. The application of Trihydral brushing without immersion led to an increase in the ID ($p < 0.001$), contrasting with the stabilizing effects of NitrAdine® and chitosan immersion. Brushing with blue®m without immersion resulted in a decrease in diameter measurements at Δ(T1–T0). Moreover, when used in conjunction with chitosan immersion, it exhibited a reduction trend in diameters in the ED ($p \leq 0.001$) (Table 3).

The retention force analysis revealed a significant variation over time ($p < 0.0001$). The initial force (T0) of the O-rings, without immersion or brushing, was 14.41 ±2.69 N. In the subsequent temporal analyses, both NitrAdine® and chitosan immersions demonstrated enhanced retention forces compared to the non-immersion condition ($p = 0.006$ and $p = 0.005$, respectively). By the final time point (T4), the retention force in the chitosan immersion group exhibited less variance than that of the NitrAdine® group ($p = 0.001$). The application of Trihydral brushing demonstrated no significant differences between immersion types across all temporal frames (Table 4).

Table 3. Differences in the dimensions of the internal and external diameters of the O-rings over time

Diameter	Immersion	Brushing	Δ(T1–T0) [µm]	Δ(T2–T0) [µm]	Δ(T3–T0) [µm]	Δ(T4–T0) [µm]
Internal	no immersion	no brushing	40.4 ±7.8$^{\alpha aA}$	–49.6 ±21.5$^{\alpha aB}$	–45.4 ±24.4$^{\alpha aB}$	–44.8 ±23.6$^{\alpha aB}$
		Trihydral	29.6 ±9.1$^{\alpha aA}$	34.5 ±9.3$^{\alpha bA}$	42.9 ±8.2$^{\alpha bAB}$	44.5 ±10.3$^{\alpha bB}$
		blue®m	44.8 ±53.3$^{\alpha aA}$	–16.5 ±20.6$^{\alpha aBC}$	–18.5 ±25.5$^{\alpha aB}$	–9.7 ±30.8$^{\alpha aC}$
	NitrAdine®	no brushing	26.2 ±54.2$^{\alpha aA}$	22.2 ±52.2$^{\beta abA}$	29.5 ±48.8$^{\beta aA}$	26.4 ±53.0$^{\beta abA}$
		Trihydral	–19.3 ±12.3$^{\beta bA}$	–12.7 ±17.3$^{\beta bA}$	–11.8 ±17.6$^{\beta bA}$	–7.8 ±18.0$^{\beta bA}$
		blue®m	54.9 ±17.0$^{\alpha aA}$	52.6 ±18.1$^{\beta aA}$	52.8 ±16.4$^{\beta aA}$	59.8 ±15.8$^{\beta aA}$
	chitosan	no brushing	44.4 ±20.8$^{\alpha aAB}$	38.4 ±20.5$^{\beta aA}$	48.2 ±19.4$^{\beta aB}$	46.4 ±19.1$^{\beta aAB}$
		Trihydral	–18.0 ±34.3$^{\beta bA}$	–12.6 ±31.0$^{\beta bA}$	–1.3 ±29.5$^{\beta bB}$	2.9 ±32.2$^{\beta bB}$
		blue®m	–29.2 ±45.2$^{\beta bA}$	–22.1 ±50.8$^{\alpha bAB}$	–15.9 ±52.0$^{\alpha bAB}$	–14.1 ±51.7$^{\alpha bB}$
External	no immersion	no brushing	12.0 ±22.5$^{\alpha\beta aA}$	–130.9 ±25.7$^{\alpha aB}$	–141.9 ±28.5$^{\alpha aB}$	–145.5 ±27.4$^{\alpha aB}$
		Trihydral	–4.4 ±13.5$^{\alpha aA}$	13.5 ±10.7$^{\alpha bA}$	6.8 ±18.9$^{\alpha bA}$	16.6 ±18.8$^{\alpha bA}$
		blue®m	29.9 ±95.6$^{\alpha aA}$	–112.2 ±63.5$^{\alpha aBC}$	–125.2 ±50.7$^{\alpha aB}$	–92.1 ±59.1$^{\alpha aC}$
	NitrAdine®	no brushing	10.9 ±64.1$^{\alpha aA}$	–27.2 ±54.7$^{\beta abA}$	–18.1 ±42.4$^{\beta abA}$	–24.7 ±44.8$^{\beta abA}$
		Trihydral	–76.2 ±27.7$^{\beta bA}$	–76.2 ±27.2$^{\beta aA}$	–70.4 ±17.9$^{\beta aA}$	–63.2 ±27.6$^{\beta aA}$
		blue®m	38.1 ±27.3$^{\alpha aA}$	13.9 ±22.0$^{\beta bA}$	17.8 ±24.5$^{\beta bA}$	14.9 ±16.5$^{\beta bA}$
	chitosan	no brushing	54.3 ±29.7$^{\alpha aAB}$	–0.4 ±60.8$^{\beta aA}$	–10.1 ±63.5$^{\beta aB}$	2.1 ±53.0$^{\delta aB}$
		Trihydral	–72.2 ±50.4$^{\beta bA}$	–76.9 ±46.9$^{\beta bA}$	–72.9 ±48.4$^{\beta bA}$	–60.8 ±48.6$^{\beta bA}$
		blue®m	–66.6 ±50.4$^{\beta bA}$	–92.8 ±67.3$^{\alpha bA}$	–82.9 ±63.6$^{\alpha bA}$	–55.4 ±66.6$^{\alpha bA}$

T0 – 0 months; T1 – 6 months; T2 – 12 months; T3 – 18 months; T4 – 24 months. Data presented as mean ± standard deviation ($M \pm SD$). The same Greek letters (α, β, δ) indicate statistical similarity between immersion techniques for the same brushing method and time. The use of the same lowercase letters indicates statistical similarity between brushing methods for the same immersion technique and time. The same capital letters denote statistical similarity between time points for the same immersion technique and brushing method. The mean values, which exhibited a normal distribution as indicated by the Shapiro–Wilk test, were compared using two-way ANOVA and Tukey's test with Bonferroni correction.

Table 4. Correlation between retention force values in different groups over time

Immersion	Brushing	Retention force [N]			
		T1	T2	T3	T4
No immersion	no brushing	12.14 ±4.53$^{\alpha aA}$	7.72 ±1.92$^{\alpha aA}$	7.81 ±1.89$^{\alpha aA}$	9.16 ±2.42$^{\alpha\beta aA}$
	blue®m	10.93 ±3.13$^{\alpha aAB}$	14.70 ±3.15$^{\alpha bA}$	15.37 ±3.71$^{\alpha\beta bA}$	9.62 ±3.58$^{\alpha aB}$
	Trihydral	9.65 ±3.84$^{\alpha aA}$	13.70 ±3.50$^{\alpha bA}$	12.37 ±1.42$^{\alpha bA}$	10.77 ±2.38$^{\alpha aA}$
NitrAdine®	no brushing	12.73 ±3.68$^{\alpha aA}$	14.48 ±4.34$^{\beta aA}$	10.63 ±3.82$^{\alpha\beta aA}$	12.32 ±1.76$^{\alpha aA}$
	blue®m	11.37 ±3.38$^{\alpha aA}$	11.83 ±3.67$^{\alpha aA}$	11.97 ±1.60$^{\alpha aA}$	9.78 ±1.32$^{\alpha aA}$
	Trihydral	10.60 ±4.09$^{\alpha aA}$	13.29 ±2.37$^{\alpha aA}$	9.70 ±4.91$^{\alpha aA}$	11.27 ±2.96$^{\alpha aA}$
Chitosan	no brushing	10.20 ±2.93$^{\alpha abAB}$	14.58 ±3.58$^{\beta aA}$	14.68 ±2.18$^{\beta abAB}$	5.99 ±2.03$^{\beta aB}$
	blue®m	13.46 ±4.57$^{\alpha bB}$	13.18 ±4.15$^{\alpha aA}$	16.38 ±1.86$^{\beta bB}$	7.54 ±3.30$^{\alpha abAB}$
	Trihydral	7.58 ±2.43$^{\alpha aA}$	11.93 ±4.64$^{\alpha aA}$	11.89 ±2.22$^{\alpha aA}$	11.32 ±3.99$^{\alpha bB}$

Data presented as *M* ±*SD*. The same Greek letters (α, β) indicate statistical similarity between immersion techniques for the same brushing method and time. The use of the same lowercase letters indicates statistical similarity between brushing methods for the same immersion technique and time. The same capital letters denote statistical similarity between time points for the same immersion technique and brushing method. The mean values, which exhibited a normal distribution as indicated by the Shapiro–Wilk test, were compared using two-way ANOVA and Tukey's test with Bonferroni correction.

The correlation analysis between dimensional changes and retention force indicated a weak negative correlation among specimens immersed in NitrAdine®, where less dimensional change was associated with higher retention force (Table 5). These complex interactions were supported by the statistical analysis, according to the data provided in Tables 3–5.

Table 5. Correlation between retention force and dimensional change in diameters

Factor	Group	Variable	Diameter	
			internal	external
Retention force	control	Pearson's correlation	0.162	0.039
		p-value	0.272	0.790
	Trihydral	Pearson's correlation	0.074	0.214
		p-value	0.618	0.144
	blue®m	Pearson's correlation	−0.003	0.136
		p-value	0.987	0.356
	NitrAdine®	Pearson's correlation	−0.296	−0.405
		p-value	0.041*	0.004*
	NitrAdine® + Trihydral	Pearson's correlation	0.186	−0.055
		p-value	0.205	0.709
	NitrAdine® + blue®m	Pearson's correlation	−0.013	0.033
		p-value	0.928	0.826
	chitosan	Pearson's correlation	−0.238	−0.22
		p-value	0.104	0.134
	chitosan + Trihydral	Pearson's correlation	−0.148	−0.334
		p-value	0.315	0.021*
	chitosan + blue®m	Pearson's correlation	−0.334	−0.275
		p-value	0.021*	0.058

* statistically significant ($p < 0.05$).

Discussion

The present study aimed to evaluate the dimensional changes in the internal and external diameters of the O-rings and the retention force before and after the application of different hygiene methods for overdentures. The results indicated that the null hypothesis was not accepted, as the evaluated hygiene methods promoted deformations in the capsules and changed the retention force at the different times analyzed. This highlights the influence of the hygiene method on the properties of O-rings. The present study focused on the evaluation of O-ring systems, which are widely accepted by patients because they are easy to insert and remove, simple to clean, improve quality of life, and allow for effective distribution of forces.[3–5] Previous studies have demonstrated that the retention of systems can be affected by different hygiene solutions.[4,21,24,25,27,34,42] Additionally, color changes in the retentive elements have also been reported.[25,42–44]

Regarding the retention force, the combination of blue®m toothpaste and chitosan solution demonstrated optimal outcomes. This combination proved effective in preserving the dimensions and the retention force of the O-ring system. The use of a 5 mg/mL chitosan solution as a complementary chemical method, in addition to brushing, appears to be a promising approach. Chitosan, an abundant and affordable substance of animal origin, has been considered a favorable option for denture hygiene due to its non-toxic nature, biopolymer properties, and proven antimicrobial action.[30,33] The necessity for easily accessible products, coupled with the prevalence of chitosan in nature, has led to the development of blue®m, a product that is free from chemical components such as triclosan and fluoride. This toothpaste contains active oxygen and biological honey derivatives, and is characterized by a neutral pH, as reported by the manufacturer. It is recommended for oral cavity hygiene and for use with overdentures, as it is purported to not cause adverse effects, such as oxidation, on prosthetic components.

The denture retention is directly related to patient satisfaction and chewing difficulties, which can necessitate the replacement of the retaining component of the prosthesis and frequent returns to the dental office for maintenance.

In relation to the dimensional changes, brushing with Trihydral in conjunction with immersion in NitrAdine® as well as brushing with blue®m followed by the immersion in a chitosan solution resulted in the smallest dimensional changes in the O-ring diameter. Trihydral toothpaste does not contain highly abrasive particles that could cause prosthesis wear. Its active ingredient, chloramine-T, has antiseptic and biocidal properties, and has been used as a periodontal surgical adjuvant.[43] It exhibits excellent biofilm-penetrating properties and effectively neutralizes bacteria, while also inhibiting gum inflammation and promoting tissue regeneration and repair.[43,44] Martinez et al. conducted a systematic review and meta-analysis to evaluate the effectiveness of effervescent tablets in cleaning and sanitizing removable dentures compared to other chemical and physical methods.[45] The authors assessed biofilm reduction, microbial levels and material stability. The study concluded that the combination of brushing with the use of an effervescent tablet yielded significantly better results.[45] According to many studies, the NitrAdine® effervescent tablet has antimicrobial properties and is compatible with cobalt–chromium metal alloy.[19,20,35]

A study by de Campos et al. stated that overdentures are subject to loss of retention when undergoing continuous insertion and removal of the prosthesis.[46] The authors highlighted several factors that can influence the efficacy of overdentures, including implant angulation, implant type, connector size, retainer depth, and retainer material. However, they did not evaluate the impact of cleaning agents.[46] Overdentures are considered to be relatively easy to clean; nevertheless, there is currently no ideal product that could be recommended for their hygiene.[47–49]

The results of this study demonstrate that the retention values of the control group were less satisfactory, suggesting that the hygiene method played a crucial role in maintaining adequate retention force. Even with the wear resulting from the insertion and removal of the prosthesis, the efficacy of the cleansers in protecting the retainers likely contributed to the enhanced retention observed in these cases. All groups exhibited higher retention force in comparison to the control group. A comprehensive analysis of the temporal dimension, coupled with the examination of the internal and external diameters of the O-rings, revealed that all groups, including the control group, exhibited dimensional changes over time, characterized by an increase or decrease in diameter. In a study by Ayyıldız et al., the effect of effervescent tablets was evaluated.[23] The results indicated that their use resulted in lower changes compared to the control group.[23] Effervescent tablets have been proposed for patients with implant-retained overdentures.[19,20] These findings are consistent with the results of other studies that have demonstrated the preservation of retainer system characteristics by cleansers.[22] Varghese et al. showed that the use of Polident for 5 min did not significantly alter the properties of the O-ring, while Fittydent and chlorhexidine gluconate resulted in an increase in its retention force when compared to the control group.[22]

In the present study, a weak negative correlation was observed between the dimensional changes of the internal and external diameters and the retention force after immersion in NitrAdine®. However, the mechanisms of retention loss cannot be solely explained by dimensional and surface changes, as not all groups that experienced retention loss exhibited significant dimensional changes. These results corroborate the findings of Rutkunas et al., who used scanning electron microscopy to evaluate the effects of wear on overdenture retainers and concluded that not all groups exhibiting retention loss demonstrated substantial dimensional changes.[47] The changes in the physical and mechanical properties of the O-ring, such as decreased flexibility, may contribute to maintaining the retention force during the use of prosthesis.

Limitations

This study was subject to certain limitations, as it was conducted in vitro and did not evaluate the effect of chewing cycles on the retainers. In clinical settings, chewing forces, as well as patient forces and the insertion axis involved in prosthesis removal and insertion, can undergo changes. Additionally, thermal cycling was not performed, and it is acknowledged that the conditions of the oral cavity can influence the results, as thermal and pH changes can alter the retention force.[23] The present study is a pioneering analysis of the combination of mechanical and chemical methods for the hygiene of overdentures, aiming to provide guidelines for improving denture hygiene and informing professionals about the safety of overdenture hygiene. The evaluated methods had satisfactory effects, as they ensured the stability of the O-ring properties within acceptable limits. However, further studies, particularly clinical trials, are necessary to evaluate not only the antimicrobial activity, but also the potential deleterious effects on different surfaces of overdentures.

Conclusions

This study contributes to the understanding of the effects of different cleaning methods on the dimensional changes and retention force of O-rings in implant-supported overdentures. The findings suggest that the combination of brushing with blue®m toothpaste and immersion in a chitosan solution can yield satisfactory results in terms of overdenture hygiene and retention force. Further research is warranted to explore the clinical applicability and long-term effects of these hygiene methods on the overdentures.

Ethics approval and consent to participate

Not applicable.

Data availability

The datasets generated and/or analyzed during the current study are available from the corresponding author on reasonable request.

Consent for publication

Not applicable.

Use of AI and AI-assisted technologies

Not applicable.

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References

1. Cune M, Burgers M, van Kampen F, de Putter C, van der Bilt A. Mandibular overdentures retained by two implants: 10-year results from a crossover clinical trial comparing ball-socket and bar-clip attachments. *Int J Prosthodont*. 2010;23(4):310–317. PMID:20617218.
2. Swelem AA, Abdelnabi MH. Attachment-retained removable prostheses: Patient satisfaction and quality of life assessment. *J Prosthet Dent*. 2021;125(4):636–644. doi:10.1016/j.prosdent.2020.07.006
3. Ellis JS, Burawi G, Walls A, Thomason JM. Patient satisfaction with two designs of implant supported removable overdentures; ball attachment and magnets. *Clin Oral Implants Res*. 2009;20(11):1293–1298. doi:10.1111/j.1600-0501.2009.01810.x
4. Chaware SH, Thakkar ST. A systematic review and meta-analysis of the attachments used in implant-supported overdentures. *J Indian Prosthodont Soc*. 2020;20(3):255–268. doi:10.4103/jips.jips_368_19
5. Derafshi R, Mohaghegh M, Saki M, Safari A, Rabee Haghighi M. The effects of denture cleansing solutions on the retention of attachments of implant supported overdentures. *J Dent (Shiraz)*. 2015;16(Suppl 1):68–72. PMID:26106638.
6. Ting M, Faulkner RJ, Donatelli DP, Suzuki JB. Tooth-to-implant-supported fixed partial denture: A comprehensive overview of systematic reviews. *Implant Dent*. 2019;28(5):490–499. doi:10.1097/ID.0000000000000901
7. Ettinger RL, Taylor TD, Scandrett FR. Treatment needs of overdenture patients in a longitudinal study: Five-year results. *J Prosthet Dent*. 1984;52(4):532–537. doi:10.1016/0022-3913(84)90340-8
8. Nguyen CT, Masri R, Driscoll CF, Romberg E. The effect of denture cleansing solutions on the retention of pink Locator attachments: An in vitro study. *J Prosthodont*. 2010;19(3):226–230. doi:10.1111/j.1532-849X.2009.00550.x
9. You W, Masri R, Romberg E, Driscoll CF, You T. The effect of denture cleansing solutions on the retention of pink locator attachments after multiple pulls: An in vitro study. *J Prosthodont*. 2011;20(6):464–469. doi:10.1111/j.1532-849X.2011.00722.x
10. Fermandes RAG, Lovato-Silva CH, Paranhos HFO, Ito IY. Efficacy of three denture brushes on biofilm removal from complete dentures. *J Appl Oral Sci*. 2007;15(1):39–43. doi:10.1590/s1678-77572007000100009
11. Salles MM, de Cássia Oliveira V, Macedo AP, Silva-Lovato CH, de Freitas de Oliveira Paranhos H. Effectiveness of brushing associated with oral irrigation in maintenance of peri-implant tissues and overdentures: Clinical parameters and patient satisfaction. *J Oral Implantol*. 2021;47(2):117–123. doi:10.1563/aaid-joi-D-19-00092
12. Paranhos HFO, Silva-Lovato CH, de Souza RF, et al. Effect of three methods for cleaning dentures on biofilms formed in vitro on acrylic resin. *J Prosthodont*. 2009;18(5):427–431. doi:10.1111/j.1532-849X.2009.00450.x
13. Baba Y, Sato Y, Owada G, Minakuchi S. Effectiveness of a combination denture-cleaning method versus a mechanical method: Comparison of denture cleanliness, patient satisfaction, and oral health-related quality of life. *J Prosthodont Res*. 2018;62(3):353–358. doi:10.1016/j.jpor.2018.01.005
14. Schmutzler A, Rauch A, Nitschke I, Lethaus B, Hahnel S. Cleaning of removable dental prostheses – a systematic review. *J Evid Based Dent Pract*. 2021;21(4):101644. doi:10.1016/j.jebdp.2021.101644
15. Papadiochou S, Polyzois G. Hygiene practices in removable prosthodontics: A systematic review. *Int J Dent Hyg*. 2018;16(2):179–201. doi:10.1111/idh.12323
16. Kamonkhantikul K, Homsiang W, Arksornnukit M. Brushing effect on the retentive force of retentive inserts in three denture attachments: An in vitro study. *J Prosthet Dent*. 2022;128(3):487.e1–487.e12. doi:10.1016/j.prosdent.2022.06.014
17. Brown JL, Young T, McKloud E, et al. An in vitro evaluation of denture cleansing regimens against a polymicrobial denture biofilm model. *Antibiotics (Basel)*. 2022;11(1):113. doi:10.3390/antibiotics11010113
18. Kiesow A, Sarembe S, Pizzey RL, Axe AS, Bradshaw DJ. Material compatibility and antimicrobial activity of consumer products commonly used to clean dentures. *J Prosthet Dent*. 2016;115(2):189–198.e8. doi:10.1016/j.prosdent.2015.08.010
19. Coenye T, De Prijck K, De Wever B, Nelis HJ. Use of the modified Robbins device to study the in vitro biofilm removal efficacy of NitrAdine, a novel disinfecting formula for the maintenance of oral medical devices. *J Appl Microbiol*. 2008;105(3):733–740. doi:10.1111/j.1365-2672.2008.03784.x
20. Silva-Lovato CH, de Wever B, Adriaens E, et al. Clinical and antimicrobial efficacy of NitrAdine™-based disinfecting cleaning tablets in complete denture wearers. *J Appl Oral Sci*. 2010;18(6):560–565. doi:10.1590/s1678-77572010000600005
21. Coimbra FCT, Rocha MM, Oliveira VC, et al. Antimicrobial activity of effervescent denture tablets on multispecies biofilms. *Gerodontology*. 2021;38(1):87–94. doi:10.1111/ger.12500
22. Varghese RM, Masri R, Driscoll CF, Romberg E. The effect of denture cleansing solutions on the retention of yellow Hader clips: An in vitro study. *J Prosthodont*. 2007;16(3):165–171. doi:10.1111/j.1532-849X.2006.00167.x
23. Ayyıldız S, Şahin C, Emir F, Ersu B. Effect of denture cleansing solutions on the retention of locator attachments over time. *J Prosthodont*. 2020;29(3):237–242. doi:10.1111/jopr.13144
24. AlRumaih HS, Albarrak AA, AlMedaires M, et al. Influence of denture cleansers on the retention loss of attachment systems retained implant overdenture. *Int J Biomater*. 2023;2023:5077785. doi:10.1155/2023/5077785
25. Kürkcüoğlu I, Özkir SE, Köroğlu A, Sahin O, Yilmaz B. Effect of denture cleansing solutions on different retentive attachments. *J Prosthet Dent*. 2016;115(5):606–610. doi:10.1016/j.prosdent.2015.11.012
26. Bal B, Erkul S, Erzincanlı A, Ozkurt-Kayahan Z, Kazazoglu E. The effect of different denture cleansing solutions on the retention of polyetheretherketone locator attachments. *J Prosthodont*. 2023;32(5):417–422. doi:10.1111/jopr.13661
27. de Campos MR, Marcondes Agnelli JA, Cândido Dos Reis A. Factors influencing retention and durability of attachments for overdentures – adverse effects of cleansings, pH, and temperature: A systematic review. *Heliyon*. 2022;8(12):e12411. doi:10.1016/j.heliyon.2022.e12411
28. Curylofo PC, Raile PN, Oliveira VC, et al. Antimicrobial activity of experimental chitosan solutions on acrylic resin and cobalt-chromium surfaces. *Int J Prosthodont*. 2023;36(5):650. doi:10.11607/ijp.7264

29. Costa EM, Silva S, Madureira AR, Cardelle-Cobas A, Tavaria FK, Pintado MM. A comprehensive study into the impact of a chitosan mouthwash upon oral microorganism's biofilm formation in vitro. *Carbohydr Polym*. 2014;101:1081–1086. doi:10.1016/j.carbpol.2013.09.041
30. Hosseinnejad M, Jafari SM. Evaluation of different factors affecting antimicrobial properties of chitosan. *Int J Biol Macromol*. 2016;85:467–475. doi:10.1016/j.ijbiomac.2016.01.022
31. Srimaneepong V, Thanamee T, Wattanasirmkit K, Muangsawat S, Matangkasombut O. Efficacy of low-molecular weight chitosan against *Candida albicans* biofilm on polymethyl methacrylate resin. *Aust Dent J*. 2021;66(3):262–269. doi:10.1111/adj.12826
32. Raile PN, Oliveira VC, Macedo AP, et al. Action of chitosan-based solutions against a model four-species biofilm formed on cobalt-chromium and acrylic resin surfaces. *Gerodontology*. 2023;40(4):472–483. doi:10.1111/ger.12672
33. Verlee A, Mincke S, Stevens CV. Recent developments in antibacterial and antifungal chitosan and its derivatives. *Carbohydr Polym*. 2017;164:268–283. doi:10.1016/j.carbpol.2017.02.001
34. Axe AS, Varghese R, Bosma ML, Kitson N, Bradshaw DJ. Dental health professional recommendation and consumer habits in denture cleansing. *J Prosthet Dent*. 2016;115(2):183–188. doi:10.1016/j.prosdent.2015.08.007
35. Felipucci DNB, Curylofo PA, Crizóstomo LC, et al. Effect of cleanser solutions on the retention force of O'ring attachment: An in vitro study. *Braz Dent J*. 2020;31(6):657–663. doi:10.1590/0103-6440202003268
36. Rodrigues RCS, Faria ACL, Macedo AP, de Mattos MGC, Ribeiro RF. Retention and stress distribution in distal extension removable partial dentures with and without implant association. *J Prosthodont Res*. 2013;57(1):24–29. doi:10.1016/j.jpor.2012.07.001
37. Rodrigues RCS, Macedo AP, de Torres EM, de Mattos MGC, Ribeiro RF. Retention force of T-bar clasps for titanium and cobalt-chromium removable partial dentures. *Braz Dent J*. 2008;19(3):209–213. doi:10.1590/S0103-64402008000300006
38. Mariotto LG, da Costa Valente ML, de Castro DT, Dos Reis AC. Effects of denture cleansing solutions on different materials used for fabrication of polymer attachment components. *Int J Prosthodont*. 2020;33(1):74–80. doi:10.11607/ijp.6445
39. Paranhos HFO, Salles AES, de Macedo LD, Silva-Lovato CH, Pagnano VO, Watanabe E. Complete denture biofilm after brushing with specific denture paste, neutral soap and artificial saliva. *Braz Dent J*. 2013;24(1):47–52. doi:10.1590/0103-6440201301946
40. da Silva GG, Shimano MVW, Macedo AP, Valente MLC, Dos Reis AC. In vitro assessment of polyetheretherketone for an attachment component for an implant-retained overdenture. *J Prosthet Dent*. 2022;127(2):319.e1–319.e8. doi:10.1016/j.prosdent.2021.07.031
41. Lopes Vasconcelos GL, Curylofo PA, Targa Coimbra FC, et al. In vitro antimicrobial activity of effervescent denture tablets on the components of removable partial dentures. *Int J Prosthodont*. 2020;33(3):315–320. doi:10.11607/ijp.6436
42. Khodadadkashi F, Alihemmati M, Yahyazadehfar N, Jamalighomi A, Hakimaneh SMR, Shayegh SS. Effect of denture cleansing solutions on retention of two types of overdenture attachments. *J Islam Dent Assoc Iran*. 2019;31(4):203–213. doi:10.30699/jidai.31.4.203
43. Tirapelli C, Landi F, Ribas JP, Panzeri H, Lara EH. Evaluating an experimental dentifrice containing chloramine-T: A preliminary study. *Oral Health Prev Dent*. 2010;8(4):375–381. PMID:21180675.
44. Martori E, Ayuso-Montero R, Martinez-Gomis J, Viñas M, Peraire M. Risk factors for denture-related oral mucosal lesions in a geriatric population. *J Prosthet Dent*. 2014;111(4):273–279. doi:10.1016/j.prosdent.2013.07.015
45. Martinez Y, Ausina V, Llena C, Montiel JM. Scientific evidence on the efficacy of effervescent tablets for cleaning removable prostheses. A systematic review and meta-analysis. *J Prosthet Dent*. 2024;131(6):1071–1083. doi:10.1016/j.prosdent.2023.01.031
46. de Campos MR, Botelho AL, Dos Reis AC. Reasons for the fatigue of ball attachments and their O-rings: A systematic review. *Dent Med Probl*. 2023;60(1):167–176. doi:10.17219/dmp/146719
47. Rutkunas V, Mizutani H, Takahashi H, Iwasaki N. Wear simulation effects on overdenture stud attachments. *Dent Mater J*. 2011;30(6):845–853. doi:10.4012/dmj.2011-057
48. Ramadan R, Elsherbeeny Y, Thabet Y, Kandil B, Ghali R. Retention of a telescopic overdenture on customized abutments after the simulation of 1 year in function. *Dent Med Probl*. 2021;58(2):201–206. doi:10.17219/dmp/127963
49. Silveira MPM, Campaner LM, Bottino MA, Nishioka RS, Borges ALS, Tribst JPM. Influence of the dental implant number and load direction on stress distribution in a 3-unit implant-supported fixed dental prosthesis. *Dent Med Probl*. 2021;58(1):69–74. doi:10.17219/dmp/130847

Original paper

Effects of EDTA, fumaric acid, and the chitosan solutions prepared with distilled water and acetic acid on dentin microhardness and tubular penetration: An in vitro study

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Abstract

Background. Chitosan prepared with acetic acid is commonly used as an endodontic irrigant. However, the chitosan solution prepared with distilled water has not been evaluated for endodontic usage.

Objectives. The present study aimed to compare the effects of ethylenediaminetetraacetic acid (EDTA), fumaric acid, and the chitosan solutions prepared with distilled water (C-DW) and acetic acid (C-AA) on dentin microhardness and dentinal tubule penetration.

Material and methods. Eighty maxillary central incisors were endodontically instrumented and randomly divided into 2 main groups ($n = 40$) for the evaluation of dentin microhardness and tubular penetration, with 4 subgroups in each main group ($n = 10$) according to the final irrigation solutions used (EDTA, fumaric acid, C-DW, and C-AA). The C-AA solution was prepared by diluting medium-molecular-weight chitosan in acetic acid. The C-DW solution was prepared with distilled water and chitosan ammonium salts, which were synthesized using trichloroacetic acid and low-molecular-weight chitosan. After irrigation, the roots were sectioned horizontally 2 mm (the apical third) and 5 mm (the middle third) from the apex. The microhardness measurements were taken at depths of 500 µm and 1,000 µm from the canal lumen. The sections were examined for tubular penetration using confocal laser scanning microscopy. The data was analyzed using the analysis of variance (ANOVA), with a significance level set at $p < 0.05$.

Results. The microhardness values were statistically similar at either depth for each third ($p > 0.05$), except for the 1,000-µm depth in the apical third, where the use of the C-AA solution resulted in lower microhardness as compared to fumaric acid ($p < 0.05$). No significant differences were observed in tubular penetration with regard to each third ($p > 0.05$).

Conclusions. All solutions showed a similar penetration ability in each third. At the 1,000-µm depth in the apical third, the fumaric acid solution provided a higher microhardness value than the C-AA solution.

Keywords: chitosan, chelating agents, confocal laser scanning microscopy, fumaric acid

Highlights

- All the tested irrigation solutions (EDTA, fumaric acid, chitosan prepared with distilled water (C-DW), and chitosan prepared with acetic acid (C-AA)) demonstrated comparable ability to penetrate dentinal tubules in both the apical and middle thirds of the root canals.
- While the microhardness values were similar across most conditions, fumaric acid showed superior preservation of dentin microhardness at the 1,000-µm depth in the apical third as compared to C-AA.
- C-DW and C-AA performed similarly in terms of tubular penetration and microhardness, indicating that C-DW is a viable alternative to C-AA for clinical use in root canal treatment.

Introduction

The success of endodontic treatment depends on the effective chemo-mechanical preparation and three-dimensional (3D) filling of the root canal system.[1] Chemo-mechanical preparation aims to shape and clean the root canals. Mechanical preparation is insufficient to reach root canal irregularities and dentinal tubules, and it cannot eliminate microorganisms from those untouched surfaces.[2] Mechanical preparation promotes the formation of the smear layer, which consists of organic and inorganic tissue remnants, microorganisms, and their by-products.[3] The smear layer negatively affects the penetration of irrigation solutions and the adhesion of the filling material.[4] The irrigation solutions used for chemical preparation mainly aim to reach and eliminate microorganisms in the uninstrumented areas, and dissolve the components of the smear layer.[5] The efficiency of irrigants is affected by their ability to penetrate into dentinal tubules, the irrigation method and root canal structures.[6]

Irrigation solutions may act similarly on the smear layer and root canal dentin.[7] Irrigants can cause changes in the chemical and mechanical properties of dentin by affecting its mineral content.[8] The determination of microhardness shows mineral changes in root canal dentin.[9] A decreased microhardness value may indicate the dissolution or deterioration of dentin composition,[10] which adversely affects the adhesion of the sealer.[11]

Several irrigation solutions are preferred for smear layer removal, e.g., ethylenediaminetetraacetic acid (EDTA), maleic acid, QMix®, MTAD, fumaric acid, and chitosan.[7,12–14] EDTA is the most widely used irrigation solution for this purpose. Irrigation with a 0.7% concentration of fumaric acid has been considered efficient in smear layer removal.[13] Fumaric acid is a trans isomer of maleic acid and is produced in a citric acid cycle.[15] Its esters have been successfully used for the treatment of psoriasis and multiple sclerosis.[16]

Recently, 0.2% chitosan has been applied in irrigation procedures.[17] Chitosan is a natural polysaccharide obtained from crab and shrimp shells.[18] Its use in the biomedical and dentistry fields is highly popular due to its good biocompatibility, biodegradability, antimicrobial properties, and chelating activity toward metal ions.[19] The 0.2% chitosan solution prepared using medium-molecular-weight chitosan and acetic acid (C-AA) has been found effective in removing the smear layer from root canal walls.[20] However, chitosan dissolves in acidic conditions and has poor water solubility, which limits its further industrial applications.[21] To overcome this drawback, chitosan derivatives have been synthesized from halogens and low-molecular-weight chitosan to provide good water solubility. These chitosan ammonium salts are considered to have better antifungal and antibacterial properties than chitosan itself.[21,22] The chitosan solution with distilled water (C-DW) is prepared by dissolving chitosan ammonium salts in distilled water.

Previously, EDTA and 0.2% chitosan solutions were compared with regard to dentin microhardness changes and the ability of the irrigant to penetrate into dentinal tubules.[4] A literature review showed that fumaric acid and the C-DW solution had not been evaluated in terms of their effects on dentin microhardness and tubular penetration ability. Thus, the present study aimed to compare the effects of different final irrigation solutions, including EDTA, fumaric acid, and the C-DW and C-AA solutions, on dentinal tubule penetration capacity and dentin microhardness changes. The null hypothesis stated that no differences would be observed in the effects of various irrigants on dentinal tubule penetration and dentin microhardness.

Material and methods

This study was approved by the ethics committee of Ankara Yıldırım Beyazıt University, Turkey (research code: 2020-149; decision date and No. of approval: 09.07.2020/39). Eighty extracted human maxillary central incisor teeth with full apex formation were selected. The teeth were visually and radiographically examined to have one single root and root canal, and to have no caries, resorptions or calcifications. The teeth were decoronated at the cemento-enamel junction (CEJ) under water coolant to obtain a 17-mm root length. The working length was determined as 16 mm. The root canals were prepared using the ProTaper Next X4 instruments (Dentsply Maillefer, Ballaigues, Switzerland) with an endodontic

electronic torque-controlled motor. The root canals were irrigated with 2 mL of 2.5% NaOCl irrigation solution at each instrument change. The roots were equally divided into 2 main groups to evaluate the effects of the tested irrigants on dentin microhardness and dentinal tubule penetration. The tested irrigants were as follows: EDTA; fumaric acid; C-DW; and C-AA.

Preparation of irrigation solutions

The EDTA solution: 17% EDTA solution was used.

The fumaric acid solution: The solution was prepared at a 0.7% concentration by mixing fumaric acid with distilled water.

The C-DW solution: Chitosan ammonium salts with halogens were prepared according to the method of a previous study,[22] using trichloroacetic acid and low-molecular-weight chitosan. The solution was prepared by dissolving the chitosan-trichloroacetate compound in distilled water at a concentration of 5 mg/mL.

The C-AA solution: Chitosan with a deacetylation degree of 70–85% (Sigma-Aldrich, St. Louis, USA) was dissolved in 1% acetic acid solution to obtain 0.2% chitosan solution.[7] The acetic acid solution was added to the weighed amount of chitosan and the mixture was stirred overnight with the help of a magnetic stirrer. Then, the solution was filtrated through Whatman 42 filter paper to remove the undissolved material.

Evaluation of dentin microhardness

The 40 roots were randomly divided into 4 subgroups (n = 10) for microhardness evaluation: EDTA; fumaric acid; C-AA; and C-DW. The roots were irrigated using 5 mL of the solution for 1 min. Afterward, irrigation with 5 mL of distilled water was performed to prevent the residual action of the solutions on dentin. The roots were embedded into acrylic resin blocks. The roots were sectioned vertically along the long axis under distilled water coolant, using a low-speed saw (Micracut 151; Metkon Instruments Inc., Bursa, Turkey), and 2.0-millimeter-thick slices were obtained at 2 mm (the apical third) and 5 mm (the middle third) from the root apex.

A Vickers microhardness testing machine (HMV-700; Shimadzu Corporation, Tokyo, Japan) was used at a load of 100 g for 10 s. Three indentations were made at 500 µm and 1,000 µm from the canal lumen, with 100 µm between the indentations, under a stereomicroscope integrated into the microhardness testing machine at ×40 magnification. The arithmetic mean of the Vickers microhardness values was calculated for each distance.

Evaluation of dentinal tubule penetration

All final irrigants were mixed with 0.1% fluorescent rhodamine B isothiocyanate (Merck, Darmstadt, Germany) to visualize dentinal tubules under confocal laser scanning microscopy. Root surfaces were sealed with nail polish. The apical foramina of the roots were covered with wax. The group was randomly divided into 4 subgroups (n = 10) according to the final irrigation solution: EDTA; fumaric acid; C-AA; and C-DW. All groups were irrigated with 5 mL of the solution for 1 min. The roots were embedded in acrylic resin and sectioned vertically along the long axis under distilled water coolant, using a low-speed saw (Micracut 151). At 2 mm (the apical third) and 5 mm (the middle third) from the root apex, 1.0-millimeter-thick slices were obtained. The coronal sides of the slices were polished with silicon carbide abrasive paper. The samples were fixed on glass coverslips, with the apical sides of the slices contacting the coverslips.

The specimens were scanned and photographed using a confocal laser scanning microscope (Zeiss LSM 510; Carl Zeiss, Jena, Germany) under the Ar/HeNe laser excitation, with a wavelength of 543 nm (Fig. 1). The images were evaluated using the ImageJ software, v. 1.48 (https://imagej.net/ij; National Institutes of Health (NIH), Bethesda, USA), to analyze the irrigant penetration areas. In each image, the outline of the penetration area and the circumference of the root canal were marked. The percentage of irrigant penetration was calculated by dividing the outlined area by the root canal area.

Statistical analysis

Statistical analysis was performed using IBM SPSS Statistics for Windows, v. 22.0 (IBM Corp., Armonk, USA). The normal distribution of the data in each group was evaluated using the Shapiro–Wilk test. The data showing a normal distribution was evaluated using the one-way analysis of variance (ANOVA), post-hoc Tukey's test and t test. The significance level was set at $p < 0.05$.

Results

Evaluation of dentin microhardness

The results regarding Vickers microhardness are shown in Table 1. No significant differences were found between the tested irrigants in the 500-µm and 1,000-µm depth microhardness values for the middle third ($p > 0.05$), and in the 500-µm depth microhardness values for the apical third ($p > 0.05$). C-AA provided a statistically significantly lower microhardness value than fumaric acid at the 1,000-µm depth for the apical third ($p < 0.05$). Regardless of the irrigant used, the irrigation procedure resulted in a statistically significantly lower microhardness values in the middle third than in the apical third at either depth ($p < 0.05$) (Table 2).

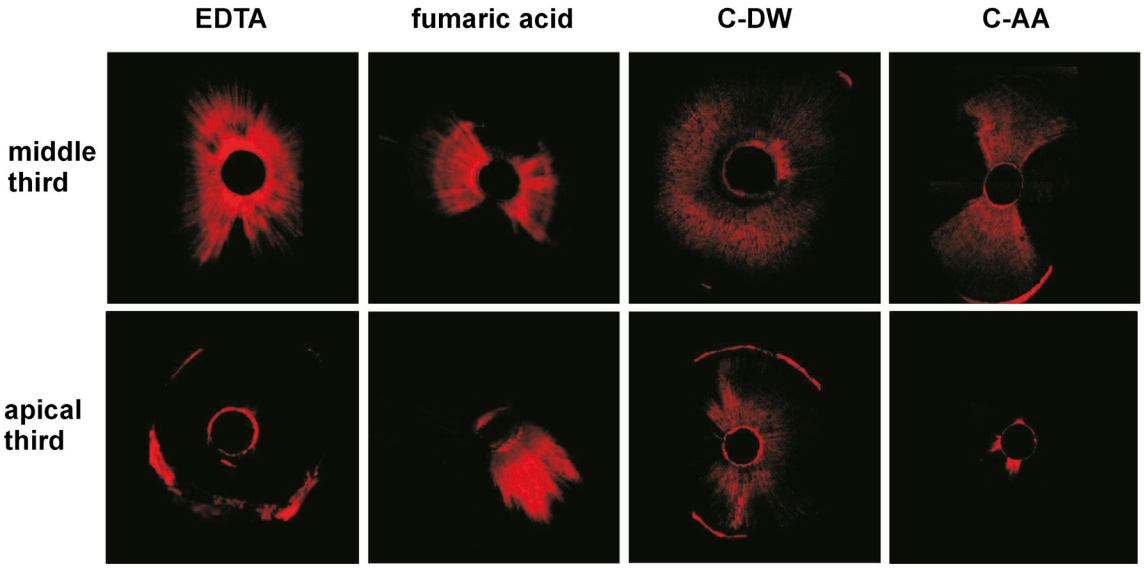

Fig. 1. Representative confocal laser scanning microscopic images for each irrigation solution at the middle and apical thirds
EDTA – ethylenediaminetetraacetic acid; C-DW – chitosan prepared with distilled water; C-AA – chitosan prepared with acetic acid.

Table 1. Comparison of the irrigants with regard to the Vickers microhardness values obtained at depths of 500 µm and 1,000 µm from the canal lumen for the apical and middle thirds of the root canals

Irrigant	Apical third						Middle third					
	500 µm			1,000 µm			500 µm			1,000 µm		
	Vickers microhardness [HV]	F	*p*-value	Vickers microhardness [HV]	F	*p*-value	Vickers microhardness [HV]	F	p-value	Vickers microhardness [HV]	F	*p*-value
EDTA (n = 10)	76.99 ±9.38	1.601	0.206	83.33 ±6.42^{a,b}	5.405	0.004*	75.56 ±7.62	1.436	0.248	82.22 ±3.75	1.620	0.202
Fumaric acid (n = 10)	76.57 ±5.12			86.01 ±3.66^{a}			75.47 ±4.61			83.98 ±4.52		
C-DW (n = 10)	76.04 ±4.85			82.08 ±3.78^{a,b}			77.10 ±6.25			81.65 ±3.34		
C-AA (n = 10)	71.71 ±3.43			78.27 ±2.81^{b}			70.87 ±9.14			78.87 ±8.08		

Data presented as mean ± standard deviation (*M* ±*SD*).
* statistically significant (ANOVA and post hoc Tukey's test). Different superscript letters indicate significant differences within each column.

Table 2. Comparison of the apical and middle thirds with regard to the dentin microhardness values and the irrigant penetration ratios, regardless of the type of irrigant

Part of the root canal	500 µ (n = 40)			1,000 µ (n = 40)			Penetration ratio (n = 40)	*t*	*p*-value
	Vickers microhardness [HV]	*t*	*p*-value	Vickers microhardness [HV]	*t*	*p*-value			
Apical third	78.52 ±6.94	2.382	0.020*	82.42 ±5.07	0.054	0.003*	7.94 ±4.97	2.690	0.009*
Middle third	74.75 ±7.22			78.49 ±6.38			11.61 ±7.03		

Data presented as *M* ±*SD*.
* statistically significant (*t* test).

Evaluation of dentinal tubule penetration

No significant differences were observed between the tested irrigants in terms of dentinal tubule penetration in the apical and middle thirds ($p > 0.05$) (Table 3). The penetration ratio obtained in the middle third was statistically higher than that in the apical third, regardless of the irrigant used ($p < 0.05$) (Table 2).

Table 3. Comparison of the irrigants with regard to the percentage of dentinal tubule penetration

Irrigant	Apical third			Middle third		
	dentinal tubule penetration [%]	F	*p*-value	dentinal tubule penetration [%]	F	*p*-value
EDTA ($n = 10$)	7.625 ±5.864	0.629	0.601	11.497 ±5.556	2.435	0.081
Fumaric acid ($n = 10$)	6.687 ±3.083			8.321 ±4.890		
C-DW ($n = 10$)	9.699 ±5.491			10.477 ±6.351		
C-AA ($n = 10$)	7.764 ±5.251			16.128 ±9.107		

Data presented as *M* ±*SD* (ANOVA).

Discussion

An irrigation solution should have the abilities of smear layer removal and dentinal tubule penetration without promoting a decrease in dentin microhardness. In this study, the effects of the EDTA, fumaric acid, C-AA, and C-DW solutions on dentin microhardness and dentinal tubule penetration were evaluated. The effects of the solutions in terms of dentinal tubule penetration were similar in the apical and middle thirds of the root canals. Dentin microhardness values for the solutions were similar at the 500-µm and 1,000-µm depths in the middle third, and at the 500-µm depth in the apical third. However, in the apical third, C-AA showed a lower microhardness value than fumaric acid at the 1,000-µm depth. Thus, the null hypothesis was partially accepted.

The dentin microhardness measurement is a non-destructive method that indirectly proves mineral loss or gain in dental hard tissues.[9] A positive correlation has been proven between dentin mineralization and microhardness.[23] The Vickers indenter method has been found to be more sensitive for evaluating deep dentin microhardness than the Knoop method.[24] In this study, Vickers indentation was preferred.

Chitosan irrigation solutions prepared with different concentrations were previously evaluated. Chitosan solutions diluted with acetic acid at 0.2% and 0.5% concentrations were similar in terms of the Ca/P ratio changes and smear layer removal in root canal dentin.[25] Chitosan solutions at 0.37% and 0.2% concentrations were similar with regard to removing the smear layer, but 0.37% chitosan caused a much greater erosive effect.[26] Therefore, in this study, the 0.2% concentration was preferred for the preparation of the C-AA solution. A 0.7% concentration of fumaric acid was used, as this concentration represents the maximum dissolution rate of fumaric acid at room temperature.[13]

The chelating feature of EDTA reduces dentin microhardness. EDTA causes demineralization by binding the Ca ions of dentin, thus softening the intraradicular dentin.[27] The effect of the 0.2% chitosan solution prepared with medium-molecular-weight chitosan and acetic acid on dentin microhardness was found to be similar to that of 17% EDTA solution,[7,28] which is in accordance with the results of this study. Supporting these results, the C-AA solution prepared with acetic acid and the C-DW solution prepared with distilled water similarly affected dentin microhardness in each third and at either depth in this study. The chelating mechanism of chitosan is not yet clear, but the chelating process is considered to occur between the amino group of chitosan and metallic ions.[7] Using acetic acid or distilled water for a chitosan solution similarly affected dentin microhardness. Further studies should be conducted to evaluate the effect of C-AA and C-DW on the physical and chemical properties of dentin.

Regardless of the irrigant used, higher microhardness values were found in the apical third at either depth. The intraradicular dentin in the apical third was more sclerosed and the concentration of non-collagenous proteins decreased. Thus, the level of demineralization in the apical third area of the root canal also decreased.[29] In the apical third at the 1,000-µm depth, fumaric acid showed the highest microhardness value, although the difference was statistically significant only in comparison with C-AA. Fumaric acid has also been found to be more effective in smear layer removal than EDTA due to 2 carboxylic groups opposite each other in its molecular form, which provides a greater binding ability to Ca ions.[13] In this study, fumaric acid also showed a similar penetration ability to that of other irrigation solutions.

Scanning electron microscopy (SEM) is a widely used method to evaluate dentinal tubule penetration. However, this technique has some drawbacks, such as the requirement for the vacuum and gold sputtering procedures, promoting artifacts, a limited number of analyzed surfaces, and the subjective examination of images.[30] Confocal laser scanning microscopy does not require any specimen preparation and provides fewer artifacts.[31] In the present study, confocal laser scanning microscopy was used for the evaluation of irrigant penetration. A fluorescent dye, rhodamine B, in a low concentration (0.1%) was used to detect the irrigant. The sections were obtained 2 mm and 5 mm from the apical foramen, as observed in previous studies.[32–34]

Regardless of the irrigant used, tubular penetration was lower in the apical third, as dentinal tubules are more sclerosed and fewer there than in the middle third.[35]

Better penetration of the irrigant can be achieved by better smear layer removal. The similar penetration ability of the solutions shows that they have a similar smear layer removal ability. The dentinal tubule penetration of irrigation solutions not only improves the flow of the sealer, but also provides better antimicrobial activity. The dentin wettability depends on the viscosity and surface tension of the irrigant, which both affect the tubular penetration of the irrigants.[36] Similar mean values of tubular penetration were observed for all irrigation solutions, regardless of the root canal third assessed. The tested solutions could also have similar viscosity. Further studies comparing the viscosity patterns of these solutions would be beneficial. In the apical third, C-DW showed a better penetration ability than C-AA. C-DW was prepared with low-molecular-weight chitosan. The lower weight may provide a deeper diffusion ability, thus enabling the irrigant to penetrate deeper into the narrow tubules involved in the apical third.

The 0.2% chitosan solution prepared with medium-molecular-weight chitosan and acetic acid was evaluated for the smear layer removal effect. A chitosan solution prepared with low-molecular-weight chitosan and distilled water has not been evaluated as an irrigation solution in the literature. The C-DW solution manifested similar penetration ability, as well as the microhardness values at the 500-µm and 1,000-µm depths, as the C-AA solution in each third. However, the lowest microhardness values at the 1,000-µm depth were observed for the C-AA solution. Therefore, the C-DW solution can be considered an alternative to the C-AA solution.

Conclusions

All the tested irrigation solutions showed a similar tubular penetration ability in the apical and middle thirds. The microhardness values were also similar for the 500-µm depth for each third. At the 1,000-µm depth in the apical third, the fumaric acid solution provided an increased microhardness value as compared to the C-AA solution.

Ethics approval and consent to participate

The study was approved by the ethics committee of Ankara Yıldırım Beyazıt University, Turkey (research code: 2020-149; decision date and No. of approval: 09.07.2020/39).

Data availability

The datasets supporting the findings of the current study are available from the corresponding author on reasonable request.

Consent for publication

Not applicable.

Use of AI and AI-assisted technologies

Not applicable.

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References

1. Caron G, Nham K, Bronnec F, Machtou P. Effectiveness of different final irrigant activation protocols on smear layer removal in curved canals. *J Endod*. 2010;36(8):1361–1366. doi:10.1016/j.joen.2010.03.037
2. Ricucci D, Siqueira JF Jr. Fate of the tissue in lateral canals and apical ramifications in response to pathologic conditions and treatment procedures. *J Endod*. 2010;36(1):1–15. doi:10.1016/j.joen.2009.09.038
3. Violich DR, Chandler NP. The smear layer in endodontics – a review. *Int Endod J*. 2010;43(1):2–15. doi:10.1111/j.1365-2591.2009.01627.x
4. Silva Antunes PV, Souza Flamini LE, Mazzi Chaves JF, Silva RG, da Cruz Filho AM. Comparative effects of final canal irrigation with chitosan and EDTA. *J Appl Oral Sci*. 2020;28:e20190005. doi:10.1590/1678-7757-2019-0005
5. Prado M, Gusman H, Gomes BP, Simão RA. Scanning electron microscopic investigation of the effectiveness of phosphoric acid in smear layer removal when compared with EDTA and citric acid. *J Endod*. 2011;37(2):255–258. doi:10.1016/j.joen.2010.11.011
6. van der Sluis LW, Gambarini G, Wu MK, Wesselink PR. The influence of volume, type of irrigant and flushing method on removing artificially placed dentine debris from the apical root canal during passive ultrasonic irrigation. *Int Endod J*. 2006;39(6):472–476. doi:10.1111/j.1365-2591.2006.01108.x
7. Saha SG, Sharma V, Bharadwaj A, et al. Effectiveness of various endodontic irrigants on the micro-hardness of the root canal dentin: An in vitro study. *J Clin Diagn Res*. 2017;11(4):ZC01–ZC04. doi:10.7860/JCDR/2017/24018.9472
8. Panighi M, G'Sell C. Effect of the tooth microstructure on the shear bond strength of a dental composite. *J Biomed Mater Res*. 1993;27(8):975–981. doi:10.1002/jbm.820270802
9. Arends J, ten Bosch JJ. Demineralization and remineralization evaluation techniques. *J Dent Res*. 1992;71 Spec No:924–928. doi:10.1177/00220345920710S27
10. Saleh AA, Ettman WM. Effect of endodontic irrigation solutions on microhardness of root canal dentine. *J Dent*. 1999;27(1):43–46. doi:10.1016/s0300-5712(98)00018-9
11. Perdigão J, Eiriksson S, Rosa BT, Lopes M, Gomes G. Effect of calcium removal on dentin bond strengths. *Quintessence Int*. 2001;32(2):142–146. PMID:12066675.
12. Nogo-Živanović D, Kanjevac T, Bjelović L, Ristić V, Tanasković I. The effect of final irrigation with MTAD, QMix, and EDTA on smear layer removal and mineral content of root canal dentin. *Microsc Res Tech*. 2019;82(6):923–930. doi:10.1002/jemt.23239
13. Jaiswal S, Patil V, Satish Kumar KS, Ratnakar P, Rairam S, Tripathi S. Comparative analysis of smear layer removal by conventional endodontic irrigants with a newly experimented irrigant-fumaric acid: A scanning electron microscopic study. *J Conserv Dent*. 2018;21(4):419–423. doi:10.4103/JCD.JCD_290_16
14. Ballal NV, Kandian S, Mala K, Bhat KS, Acharya S. Comparison of the efficacy of maleic acid and ethylenediaminetetraacetic acid in smear layer removal from instrumented human root canal: A scanning electron microscopic study. *J Endod*. 2009;35(11):1573–1576. doi:10.1016/j.joen.2009.07.021

15. Yazdi MR, Mrowietz U. Fumaric acid esters. *Clin Dermatol*. 2008;26(5):522–526. doi:10.1016/j.clindermatol.2008.07.001
16. Gkalpakiotis S, Arenberger P, Gkalpakioti P, Meluzinova E, Chandran D, Arenbergerova M. Management of psoriasis vulgaris and multiple sclerosis with fumaric acid. *J Am Acad Dermatol*. 2014;70(3):e60–e61. doi:10.1016/j.jaad.2013.10.031
17. Zhou H, Li Q, Wei L, Huang S, Zhao S. A comparative scanning electron microscopy evaluation of smear layer removal with chitosan and MTAD. *Niger J Clin Pract*. 2018;21(1):76–80. doi:10.4103/1119-3077.224798
18. Akncbay H, Senel S, Ay ZY. Application of chitosan gel in the treatment of chronic periodontitis. *J Biomed Mater Res B Appl Biomater*. 2007;80(2):290–296. doi:10.1002/jbm.b.30596
19. Aguilar A, Zein N, Harmouch E, et al. Application of chitosan in bone and dental engineering. *Molecules*. 2019;24(16):3009. doi:10.3390/molecules24163009
20. Silva PV, Guedes DF, Nakadi FV, Pécora JD, Cruz-Filho AM. Chitosan: A new solution for removal of smear layer after root canal instrumentation. *Int Endod J*. 2013;46(4):332–338. doi:10.1111/j.1365-2591.2012.02119.x
21. Guo Z, Xing R, Liu S, et al. Antifungal properties of Schiff bases of chitosan, N-substituted chitosan and quaternized chitosan. *Carbohydr Res*. 2007;342(10):1329–1332. doi:10.1016/j.carres.2007.04.006
22. Tan W, Li Q, Dong F, Wei L, Guo Z. Synthesis, characterization, and antifungal property of chitosan ammonium salts with halogens. *Int J Biol Macromol*. 2016;92:293–298. doi:10.1016/j.ijbiomac.2016.07.023
23. Panighi M, G'Sell C. Influence of calcium concentration on the dentin wettability by an adhesive. *J Biomed Mater Res*. 1992;26(8):1081–1089. doi:10.1002/jbm.820260809
24. Fuentes V, Toledano M, Osorio R, Carvalho RM. Microhardness of superficial and deep sound human dentin. *J Biomed Mater Res A*. 2003;66(4):850–853. doi:10.1002/jbm.a.10064
25. Mathew SP, Pai VS, Usha G, Nadig RR. Comparative evaluation of smear layer removal by chitosan and ethylenediaminetetraacetic acid when used as irrigant and its effect on root dentine: An in vitro atomic force microscopic and energy-dispersive X-ray analysis. *J Conserv Dent*. 2017;20(4):245–250. doi:10.4103/JCD.JCD_269_16
26. Silva PV, Costa Guedes DF, Pécora JD, da Cruz-Filho AM. Time-dependent effects of chitosan on dentin structures. *Braz Dent J*. 2012;23(4):357–361. doi:10.1590/s0103-64402012000400008
27. Akcay I, Sen BH. The effect of surfactant addition to EDTA on microhardness of root dentin. *J Endod*. 2012;38(5):704–707. doi:10.1016/j.joen.2012.02.004
28. Pimenta JA, Zaparolli D, Pécora JD, Cruz-Filho AM. Chitosan: Effect of a new chelating agent on the microhardness of root dentin. *Braz Dent J*. 2012;23(3):212–217. doi:10.1590/s0103-64402012000300005
29. Kawasaki K, Ruben J, Stokroos I, Takagi O, Arends J. The remineralization of EDTA-treated human dentine. *Caries Res*. 1999;33(4):275–280. doi:10.1159/000016529
30. Machado R, da Fonseca Roberti Garcia L, da Silva Neto UX, da Cruz Filho AdM, Silva RG, Vansan LP. Evaluation of 17% EDTA and 10% citric acid in smear layer removal and tubular dentin sealer penetration. *Microsc Res Tech*. 2018;81(3):275–282. doi:10.1002/jemt.22976
31. Kok D, Húngaro Duarte MA, Da Rosa RA, Wagner MH, Pereira JR, Reis Só MV. Evaluation of epoxy resin sealer after three root canal filling techniques by confocal laser scanning microscopy. *Microsc Res Tech*. 2012;75(9):1277–1280. doi:10.1002/jemt.22061
32. Akcay M, Arslan H, Mese M, Durmus N, Capar ID. Effect of photon-initiated photoacoustic streaming, passive ultrasonic, and sonic irrigation techniques on dentinal tubule penetration of irrigation solution: A confocal microscopic study. *Clin Oral Investig*. 2017;21(7):2205–2212. doi:10.1007/s00784-016-2013-y
33. Generali L, Cavani F, Serena V, Pettenati C, Righi E, Bertoldi C. Effect of different irrigation systems on sealer penetration into dentinal tubules. *J Endod*. 2017;43(4):652–656. doi:10.1016/j.joen.2016.12.004
34. Küçük M, Kermeoğlu F. Efficacy of different irrigation methods on dentinal tubule penetration of chlorhexidine, QMix and Irritrol: A confocal laser scanning microscopy study. *Aust Endod J*. 2019;45(2):202–208. doi:10.1111/aej.12309
35. Paqué F, Luder HU, Sener B, Zehnder M. Tubular sclerosis rather than the smear layer impedes dye penetration into the dentine of endodontically instrumented root canals. *Int Endod J*. 2006;39(1):18–25. doi:10.1111/j.1365-2591.2005.01042.x
36. Bukiet F, Soler T, Guivarch M, et al. Factors affecting the viscosity of sodium hypochlorite and their effect on irrigant flow. *Int Endod J*. 2013;46(10):954–961. doi:10.1111/iej.12086

Review

Prevalence of tooth agenesis in young cancer patients: A systematic review and meta-analysis

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Abstract

Childhood cancer survivors report many health issues related not only to the disease itself but also to post-treatment complications. Dental problems in these patients are irreversible, as they mostly concern the permanent dentition. This systematic review and meta-analysis is aimed at determining the prevalence of hypodontia in cancer survivors. The research strategy was implemented using multiple databases, such as PubMed®, Scopus, Web of Science, and Embase. The literature search was performed on February 21, 2023. A total of 576 articles were screened. Of those, 72 full-text articles were assessed for eligibility, and 31 articles were ultimately selected for inclusion in the meta-analysis. The prevalence of tooth agenesis in pediatric cancer patients was found to be 22% (random effects model; 95% confidence interval (*CI*): 14–25%, $p < 0.001$). Pooled analyses of 15 unadjusted relative risk estimates demonstrated a significantly higher prevalence of tooth agenesis in cancer patients compared to healthy individuals (unadjusted odds ratio (*OR*): 3.12; 95% *CI*: 2.01–4.83; $p < 0.00001$). Factors reported in the literature as contributing to the incidence of hypodontia include younger age at diagnosis, the utilization of multiple cytostatic drugs, high-dose radiotherapy (RTX), hematopoietic stem cell transplantation (HSCT), and the presence of other dental abnormalities. Patients who underwent cancer therapy during childhood are more prone to hypodontia.

Keywords: children, cancer, hypodontia, tooth agenesis

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Highlights

- Childhood cancer treatment is linked to a higher incidence of tooth agenesis.
- Children undergoing cancer treatment typically exhibit at least 2 dental disorders.
- The severity of dental abnormalities is primarily influenced by the child's age, cancer type and the specific treatment protocol used.

Introduction

Childhood cancer survivors suffer from many health problems related not only to the disease itself but also to post-treatment complications. These include cardiometabolic diseases,[1] chronic kidney impairments[2,3] and endocrine disorders.[4] It is estimated that around 10% of children who survive cancer will experience hearing loss within several years following the disease.[5]

The curative cancer therapy in children may affect most of the growing and developing tissues, including those of the head and face, such as the teeth. Long-term complications, including hypodontia, microdontia, impaired development of the tooth roots, or demineralization of enamel, may not pose a direct threat to the patient's life. However, they may adversely affect their health and aesthetics in the future.[6,7] Cancer survivors may also suffer from delayed or accelerated dental development,[8,9] which, in turn, influences the development of the jaws and dental occlusion. The cancer patients were more likely to report at least 1 dental health problem after controlling for socioeconomic factors, age at last follow-up and diagnosis, other treatment exposures, and access to dental services. Consequently, long-term orthodontic or prosthodontic treatment could be necessary.[7,10]

The formation of deciduous teeth begins at 4 months of pregnancy, while the first signs of mineralization of the first permanent tooth become apparent at the time of childbirth.[11] The cancer treatment can be initiated during the first months or years of the child's life, when the most active mineralization of permanent tooth buds occurs.[12] Therefore, the majority of dental complications become evident later in life of patients with permanent dentition. It has been proven that both chemotherapy (CT) and radiotherapy (RTX) may cause direct or indirect irreversible changes in developing tooth buds. Radiotherapy may directly interfere with the mitotic activity of odontoblasts in developmental patients, resulting in the formation of "osteodentin" rather than the normal dentin and indirectly affecting the process of enamel formation, leading to severe demineralization.[6] Cytostatics were also proven to disrupt the metabolic processes and cell cycle of ameloblasts and odontoblasts, thus directly influencing the processes of amelogenesis and dentinogenesis.[8,11]

Chemotherapeutic drugs applied in cancer therapy, namely vincristine, doxorubicin, cyclophosphamide, or actinomycin D, exert particularly harmful effects on tooth buds.[12] Some cytotoxic antibiotics administered to cancer patients may present relative risks of hypodontia.[13] There is evidence demonstrating a relationship between RTX and dental damage, indicating that the dose of RTX correlates with the severity of changes.[14] Other studies indicate a relationship between mutations of certain genes and the occurrence of cancer and tooth agenesis.[15]

Hypodontia, defined as a lower-than-normal number of permanent teeth, results from a complete devastation of tooth buds and is one of the most severe and frequent complications among dental abnormalities experienced by childhood cancer survivors.[6,13,16] Therefore, the aim of the study was to systematically review the literature to determine the prevalence of hypodontia in pediatric cancer patients and to compare it with the prevalence of the condition in healthy individuals. The null hypothesis stated that the prevalence of tooth agenesis would be comparable in childhood cancer survivors and healthy individuals.

Material and methods

The present systematic review and meta-analysis was conducted according to the PRISMA (Preferred Reporting Items of Systematic Reviews and Meta-Analyses) guidelines in order to follow a uniform and transparent methodology.[17] The study was registered with PROSPERO (registration No. CRD42022308068). The following PICOS (Population, Intervention, Comparison, Outcome, and Study design) framework was employed: Population – pediatric patients; Intervention – cancer patients; Comparison – healthy patients; Outcome – prevalence of hypodontia. The research question was: "What is the prevalence of hypodontia in pediatric cancer patients?"

Literature search

The systemic research strategy was implemented using multiple databases, namely PubMed®, Scopus, Web of Science, and Embase. The literature search was performed on February 21, 2023. The search strategy used in PubMed® and adapted in other database searches is presented in Table 1. After the search, all articles were imported into the Mendeley Desktop v. 1.17.11 software (Glyph & Cog, LLC, Petaluma, USA) to eliminate duplicates.

Table 1. Search strategy

Search No.	Keywords
1	cancer patients OR pediatric cancer survivors OR pediatric stem cell transplantation OR blood transplantation OR marrow transplantation OR radiotherapy adverse effects OR chemotherapy adverse effect OR cervico-facial irradiation OR colorectal polyposis OR cancer
2	dental agenesis OR tooth agenesis OR hypodontia OR oligodontia OR anodontia
3	#1 and #2

The 3 searches were implemented across all databases (PubMed®, Scopus, Web of Science, and Embase).

Study selection

The articles were imported into the Rayyan online tool,[18] and the titles and abstracts were initially screened to identify studies that potentially met the following eligibility criteria: human experimental studies (cross-sectional and longitudinal, retrospective, and prospective) investigating the prevalence and patterns of tooth agenesis in pediatric patients with cancer; studies with at least 3 subjects with dental anomalies per group. Only manuscripts published in the English language were considered. Case series, case reports, pilot studies, and reviews were excluded from the analysis. The full texts of the articles were reviewed, and a systematic methodology was employed to label all the relevant information for the exclusion or inclusion of individual papers. The decision process was performed by 2 independent reviewers (PP and MLS). In the case of disagreement between the authors, the final decision was made through consultation with a third reviewer (CECS), a senior experienced researcher.

Data extraction

The relevant data from the included studies was extracted independently by 2 authors (PP and MLS) using a Microsoft Excel spreadsheet (Microsoft Corporation, Redmond, USA). In instances where information was incomplete or unclear, the authors of the included reports were contacted via e-mail for clarification. The following data was recorded for each included report: study design and sample size; age of participants during examination; age at diagnosis; cancer type; length of therapy; prevalence of hypodontia in cancer patients; and other dental anomalies.

Risk of bias

The risk of bias for all the included clinical trials was assessed by 2 independent reviewers (PP and MLS), and discrepancies were resolved by discussion and in consultation with a third reviewer (CECS). All included studies were evaluated using specific tools for each experimental design: the ROBINS-I (Risk Of Bias In Non-randomized Studies – of Interventions) for non-randomized clinical trials; the Newcastle–Ottawa Scale (NOS) for cohort studies; and the Joanna Briggs Institute (JBI) critical appraisal tool for cross-sectional and case–control studies.[17]

Statistical analysis

The data regarding the prevalence of tooth agenesis was pooled, and the risk difference with a 95% confidence interval (*CI*) was used as the effect size. Subsequently, the inverse variance method was selected to calculate the pooled effect. When data from the control patients was available, information regarding the prevalence of tooth agenesis in both cancer and non-cancer patients was used to generate unadjusted odds ratios (*OR*s) and 95% *CI*s of the tooth agenesis for the cancer vs. non-cancer group. The heterogeneity (I^2) index and Cochran's Q test were used to examine the heterogeneity between the studies. For the Cochran's Q test, the *p*-value was significant at <0.05. All analyses were performed using the MedCalc® statistical software, v. 20.027 (MedCalc Software Ltd, Ostend, Belgium).

Results

Literature search

The literature search yielded a total of 917 records (Fig. 1). After removing duplicates, 576 articles were screened, resulting in the exclusion of 504 papers based on the eligibility criteria. A total of 72 full-text articles were assessed for eligibility. Of these, 39 were not selected for the qualitative analysis. Nine of them did not present complete data, 7 were based on studies conducted exclusively on adults, 6 were case reports, 4 were published in a language other than English, 4 did not provide the full text, 3 were performed on non-cancer patients only, 2 evaluated data on third molars only, 2 were reviews, one of the studies included patients who did not complete the treatment, and 1 was a pilot study. A total of 33 studies were included in the qualitative analysis. However, 2 additional articles were excluded: one due to missing data; and the second one because it employed the same sample as another article. Finally, 31 studies were included in the single-arm meta-analysis.[3,6,8,9,16,19–45] Only 14 studies presented data for a control group and were included in the proportion meta-analysis.[6,12,19–22,29–34,36,44,45]

The characteristics of the included articles are summarized in Table 2. Several types of clinical studies were included, such as cross-sectional, cohort and case–control studies. In the investigated groups, the number of cancer patients ranged from 10 to 9,308. The subjects suffered

Table 2. Characteristics of the included studies

Study	Type of study	Cancer patients, *n*	Cancer patients with hypodontia, *n* (%)	Age at examination	Age at cancer diagnosis
Alpaslan et al.[19] 1999	cross-sectional	30	15 (50.0)	4–15 years	ND
Atif et al.[20] 2022	cross-sectional	120	6 (5.0)	>12 years	<8 years
Bica et al.[8] 2017	cohort	36	12 (33.3)	10–12 years	1–6 years (*n* = 20); 7–12 years (*n* = 16)
Çetiner et al.[21] 2019	cohort	53/31 who underwent dental examination	21 (39.6)	10 years ±4 months	ND
Cubukcu et al.[45] 2012	case–control	37	6 (16.2)	ND	2.7 ±0.6 years
Estilo et al.[23] 2003	cohort	10	4 (40.0)	10 years ±4 months	4.5 years (10 months–19.5 years)
Flandin et al.[24] 2006	cohort	32 (TBI + CT); 30 (CT only)	TBI + CT: 1 (3.1); CT: 19 (63.3)	TBI + CT: 181 (130–240) months; CT: 198 (147–247) months	TBI + CT: *M*: 37 months; CT: *M*: 37 months
Hölttä et al.[43] 2005	cross-sectional	52	16 (31) patients without third molars; 77% (<3 years), 40% (3–5 years), and 0% (>5 years)	11.7 (4.7–25.7) years	10 years at the time of SCT
Immonen et al.[25] 2021	cross-sectional	178	1.4–3.8%	ND	5.0 (2.5–16.8) years
Jodłowska et al.[26] 2019	non-randomized clinical trial	37	5 (13.5)	<18 years	3 years and 2 months (range: 4 months–8 years and 6 months)
Kang et al.[3] 2018	cross-sectional	196	40 (20.4)	14.9 (4.6–33.9) years	4/7 years (0–16.4 years)
Kaste et al.[27] 1995	cross-sectional	22	11 (50.0)	ND	5 years and 1 month
Kaste et al.[28] 1998	cross-sectional	52	9 (17.3)	ND	1.5 years (range: 3 days–7.2 years; *M*: 1.9 years)
Kaste et al.[44] 2009	cross-sectional	8,522	698 (8.2)	ND	6.0 (0–20) years
Kılınç et al.[29] 2019	case–control	93	21 (22.6)	9.54 ±1.25 years (range: 8–13 years)	9 months–7 years
Krasuska-Sławińska et al.[30] 2016	non-randomized clinical trial	60	16 (26.7)	11.81 ±3.87 years	5.9 ±4.0 years
Lauritano and Petruzzi[31] 2012	non-randomized clinical trial	52	7 (13.5)	8–15 years	<15 months
Lopes et al.[32] 2006	cross-sectional	137	8 (5.8)	0–6; 6–12 years	5 years and 6 months
Nemeth et al.[33] 2013	non-randomized clinical trial	38	4 (10.5) without third molars; 18 (47.4) with third molars	12.2 ±0.5 years	31 months–6 years; *M*: 4.29 ±1.71 years
Oğuz et al.[34] 2004	non-randomized clinical trial	36	16 (44.4)	10.0 (4.2–17.6) years	7.1 years (range: 3.2–15 years)
Owosho et al.[35] 2016	cross-sectional	13	7 (53.8)	ND	5 years (range: 19 months–13 years)
Pedersen et al.[22] 2012	cohort	150	14 (9.3)	12–18 years	1–7 years
Proc et al.[6] 2016	case–control	61	19 (31.1)	5–18 years (56–213 months)	1–196 months
Quispe et al.[36] 2019	case–control	111	11 (9.9)	*M*: 160.1 months	<192 months; *M*: 83.2 months
Ruyssinck et al.[37] 2019	case–control	42	51.3%	ND	<12 years
Shum et al.[38] 2020	case–control	59	9 (15.3)	14–16 years; *M*: 14.9 ±0.80 years	<10 years; *M*: 4.1 ±2.9 years
Singh et al.[39] 2021	case–control	29	3 (10.3)	37.3 (24.2–219.5) months	2.9 (0.8–14) years
Sonis et al.[40] 1990	case–control	97	5 (5.2)	8 year and 1 month–16 years and 2 months	<10 years
Stolze et al.[16] 2021	cross-sectional	154	21 (14.3)	32.4 (16.8–56.6) months	5.2 (0.3–16.1) years
Tanaka et al.[41] 2017	cross-sectional	56	9 (16.1)	13.9 (4.6–32.7) years	1.9 (0.0–13.7) years
Welbury et al.[42] 1984	cross-sectional	64	12 (18.8)	3–20 years	ND

ALL – acute lymphoblastic leukemia; ANLL – acute non-lymphoblastic leukemia; AML – acute myeloid leukemia; CNS – central nervous system; CT – chemotherapy; HSCT – hematopoietic stem cell transplantation; *M* – mean; ND – no data; PNET – primitive neuroectodermal tumor; RMS – rhabdomyosarcoma; RTX – radiotherapy; SCT – stem cell transplantation; TBI – total body irradiation.

Cancer type	Follow-up period	Therapy received	Other dental anomalies evaluated
non-Hodgkin lymphoma (17); Hodgkin lymphoma (13)	17 months (3–58 months)	CT	enamel discoloration; hypoplasia; unerupted teeth; premature apexification
acute lymphocytic leukemia (54); Hodgkin lymphoma (24); retinoblastoma (10); sarcomas (4); acute myeloid leukemia (9); medulloblastoma (1); Langerhans cell histiocytosis (2); PNET (3); non-Hodgkin lymphoma (13)	ND	CT	developmental defects of enamel
ALL	ND	CT	tooth eruption disorders (71%); hypoplasia (17%)
Hodgkin lymphoma (10); non-Hodgkin lymphoma (36); neuroblastoma (2); Wilms tumor (1); retinoblastoma (2); RMS (1); nasopharyngeal carcinoma (1)	1–5 years (*M*: 2 years ±4 months)	CT	enamel discoloration; enamel hypoplasia; unerupted teeth
non-Hodgkin lymphoma (8); Wilms tumor (8); soft tissue sarcoma (4); medulloblastoma (3); optic glioma (1); neuroblastoma (4); Hodgkin lymphoma (3); retinoblastoma (2); Langerhans cell histiocytosis (2); other (hepatoblastoma and germ cell tumor) (2)	>5 years (*M*: 6.7 ±1.5 years)	CT and RTX	ND
RMS of the neck	12.2 years	CT and RTX	enamel defects; bony hypoplasia/facial asymmetry; trismus; velopharyngeal insufficiency; radiographically underdeveloped mandible; tooth agenesis; root agenesis; root stunting/tapering; arrested/incomplete root development
neuroblastoma	TBI + CT: 157 months CT: 145 months	CT, RTX of the head or neck, TBI	ND
neuroblastoma; ALL; AML; chronic myeloid leukemia; myelodysplastic syndrome; severe aplastic anemia; RMS, yolk sac tumor	7.4 (1.0–20.6) years	CT, RTX of the head or neck, TBI	ND
ALL	6.3 (3.0–11.6) years	CT, RTX of the head or neck, TBI	ND
solid tumor (29); leukemia (8)	24–36 months	CT and RTX	ND
ALL (71); AML (7); lymphoma (23); brain tumor (22); sarcoma (18); abdomen (37); others (18)	6.9 (2.1–22.5) years	CT and RTX	enamel hypoplasia
RMS of the head or neck	9 years and 5 months (5–16 years)	CT and RTX	severe facial deformity; severe malocclusion; extensive caries
neuroblastoma	5.0 (1.9–19.3) years (*M*: 6.4 years)	CT and RTX	enamel hypoplasia (17%); excessive caries (29%)
leukemia (2,910 (34.2%)); CNS tumor (1,076 (12.6%)); Hodgkin lymphoma (1,086 (12.7%)); non-Hodgkin lymphoma (628 (7.4%)); Wilms tumor (794 (9.3%)); neuroblastoma (575 (6.8%)); soft tissue sarcoma (750 (8.8%)); bone cancer (702 (8.2%))	22.0 (15–34) years	CT and RTX	enamel hypoplasia; gingivitis; xerostomia
lymphoproliferative tumors; leukemia; lymphoma; Langerhans cell histiocytosis; solid tumors; neuroblastoma; renal tumor; soft tissue sarcoma; germ cell tumor; hepatic tumor; CNS tumor; retinoblastoma	5–8 years	CT and RTX	enamel defects (22 (23.7%))
Burkitt's lymphoma (15.0%); nephroblastoma (13.0%); neuroblastoma (10.0%); histiocytosis (8.3%); RMS (6.7%); Ewing sarcoma (6.7%); medulloblastoma (5.0%); neurofibromatosis type I (5.0%); others (30.3%)	4.9 ±3.4 years	CT	root resorption (36 (60.0%)); enamel defects (53 (88.3%))
ALL (39); AML (13)	60 ±24 months	CT and RTX	enamel hypoplasia (9 (17.3%))
leukemia/lymphoma (61%); solid tumors (39%)	3–58 months (*M*: 17 months)	CT and RTX	microdontia (10 (7%)); taurodontism (19 (14%)); macrodontia (7 (5%)); blunted root (2 (2%)); tapered root (5 (4%))
ND	6.9 ±2 years	CT, RTX of the head or neck, TBI	macrodontia (2–2.6%); unerupted teeth (6–15.8%)
non-Hodgkin lymphoma	2.6 (1–6.2) years	CT	enamel discoloration (24 (66.7%)); enamel defects (20 (55.6%)); unerupted teeth (7 (19.4%)); premature apexification (2 (5.6%))
RMS	9 (1–13) years	CT	facial asymmetry and jaw hypoplasia; trismus and hyposalivation/xerostomia; enamel malformation
lymphomas and other reticuloendothelial neoplasms; CNS, intracranial and intraspinal neoplasms; sympathetic nervous system tumors; retinoblastoma; renal and hepatic neoplasms; bone and soft tissue sarcoma; gonadal neoplasms	ND	CT	ND
ALL; ANLL; B-cell non-Hodgkin lymphoma; PNET; germinal tumor; brain tumor; hepatoblastoma; neuroblastoma; RMS; Wilms tumor	4.9 years (58.9 ±4.3 months)	CT and RTX	ND
various	*M*: 18.3 months	CT, RTX of the head or neck, TBI	various but not significant
primitive neuroectodermal tumor (1); ALL (9); AML (2); juvenile myelomonocytic leukemia (2); neutropenia (severe, congenital) (1); neuroblastoma (9); Wilms tumor/nephroblastoma (2); anaplastic large cell lymphoma (1); juvenile metachromatic leukodystrophia (1); X-linked adrenoleukodystrophy (2); myelodysplastic syndrome (4); secondary myelodysplastic syndrome (1); chronic myeloid leukemia (2); aplastic anemia (2); thalassemia major (1); hemophagocytic lymphohistiocytosis (1); Burkitt's lymphoma (1)	>1 year (*M*: 7 years)	CT and TBI	ND
various	ND	CT and RTX	ND
neuroblastoma	ND	CT	hypocalcification of enamel; trismus
ALL	5 years	CT and RTX	enamel hypoplasia
hematological malignancy (111); brain tumor (7); solid tumor (36)	25.2 (15.9–48.8) months	CT, RTX of the head or neck, TBI	peg-shaped teeth; hypomineralization; persistent deciduous teeth
ALL (30 (53.6%)); AML (11 (19.6%)); juvenile myelomonocytic leukemia (1 (1.8%)); malignant lymphoma (4 (7.1%)); neuroblastoma (4 (7.1%)); Wilms tumor (2 (3.6%)); hepatoblastoma (1 (1.8%)); Langerhans cell histiocytosis (1 (1.8%)); retinoblastoma (1 (1.8%)); germinoma (1 (1.8%))	3 years from the completion of cancer treatment or 5 years from the time of the diagnosis	CT	enamel defects/hypoplasia (6 (10.7%))
leukemia (37); solid tumor (27)	ND	CT	hypoplastic teeth (36%)

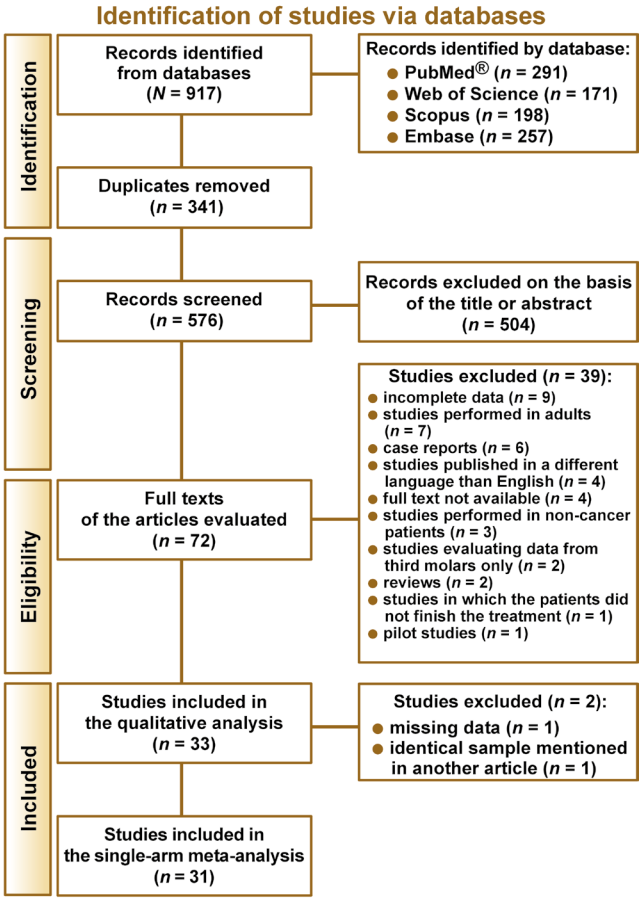


Fig. 1. Flowchart of the selection process

from various forms of cancer, including solid tumors, leukemias and lymphomas. They were most often treated with CT alone; however, some patients also received RTX (including head and neck RTX), total body irradiation (TBI) and hematopoietic stem cell transplantation (HSCT). At the time of diagnosis, the majority of patients were under 10 years of age, with the youngest subject being 1 month old.

Various teeth were affected by agenesis. Most often missing teeth were second premolars, second molars and lower incisors.[6,8,16,19,21,22,28,30,35–37,43] Risk factors associated with a higher incidence of agenesis in cancer survivors were: younger age at diagnosis or treatment (1–7 years)[3,8,16,19,20,23,27,29,32,34–37,40,43,44,46–48]; use of multiple (>4) classes of chemotherapeutic agents, particularly alkylating agents in high doses, and prolonged duration of therapy[3,16,30,37,38]; use of heavy metal compounds in CT[2]; RTX dosage greater than or equal to 2,200 cGy[32,44]; head and neck radiation therapy (RTX)[23,35,40]; history of HSCT[3,37,38,43]; and the presence of other dental anomalies.[16]

Risk of bias

For cross-sectional studies, the average quality score ranged between 4 and 6 (Table 3). The criteria that exhibited the highest failure rate pertained to the identification of confounding factors. For cohort studies, the quality score ranged between 4 and 6 (Table 4).

Table 3. Assessment of the quality of studies using the Joanna Briggs Institute (JBI) critical appraisal tool for cross-sectional studies

Study	Q1	Q2	Q3	Q4	Q5	Q6	Q7	Q8	Score
Alpaslan et al.[19] 1999	N	Y	U	Y	N	N	Y	Y	4
Atif et al.[20] 2022	Y	Y	Y	Y	N	N	Y	Y	6
Hölttä et al.[43] 2005	Y	Y	Y	Y	N	N	Y	Y	6
Immonen et al.[25] 2021	Y	Y	U	Y	U	N	Y	Y	5
Kang et al.[3] 2018	U	U	Y	Y	N	N	Y	Y	4
Kaste et al.[27] 1995	Y	Y	Y	Y	N	N	Y	N	5
Kaste et al.[28] 1998	Y	Y	Y	Y	N	N	Y	N	5
Kaste et al.[44] 2009	Y	Y	Y	Y	N	N	Y	Y	6
Lopes et al.[32] 2006	Y	Y	Y	Y	N	N	Y	Y	6
Owosho et al.[35] 2016	N	Y	Y	Y	N	N	Y	N	4
Stolze et al.[16] 2021	Y	Y	Y	Y	N	N	Y	Y	6
Tanaka et al.[41] 2017	Y	Y	Y	Y	N	N	Y	Y	6
Welbury et al.[42] 1984	N	Y	Y	Y	N	N	Y	N	4

Y – yes; N – no; U – unclear.

Table 4. Assessment of the risk of bias using the Newcastle–Ottawa Scale (NOS) for cohort studies

Study	Selection				Comparability	Outcome			Total
	representativeness of the exposed cohort	selection of the non-exposed cohort	ascertainment of exposure	outcome of interest not present at the start of the study	comparability of cohorts on the basis of the design or analysis	assessment of outcome	duration of follow-up	adequacy of follow-up	
Bica et al.[8] 2017	1	1	1	1	0	0	0	0	4
Çetiner et al.[21] 2019	1	1	1	1	1	0	0	0	5
Estilo et al.[23] 2003	1	1	1	1	0	0	1	0	5
Flandin et al.[24] 2006	1	1	1	1	0	0	1	1	6
Pedersen et al.[22] 2012	1	1	1	1	1	0	0	0	5

The studies under review failed to complete the independent blind assessment. Non-randomized clinical trials were catalogued as having a high risk of bias in domains of confounding and selection of participants into the study (Table 5). For case–control studies, the quality score ranged from 5 to 8. However, all studies failed to meet the criteria related to the identification and management of confounding factors (Table 6).

Meta-analysis

Figure 2 presents the results of the single-arm meta-analysis, which revealed that the prevalence of tooth agenesis in pediatric cancer patients was 22% (random effects model; 95% *CI*: 14–25%, $p < 0.001$). Pooled analyses of 15 unadjusted relative risk estimates demonstrated a statistically significant 2.94-fold increase in the prevalence

Table 5. Assessment of the risk of bias using the ROBINS-I (Risk Of Bias In Non-randomized Studies – of Interventions) tool for non-randomized studies

Study	Bias due to confounding	Bias in selection of participants into the study	Bias in classification of interventions	Bias due to deviations from intended interventions	Bias due to missing data	Bias in measurement of outcomes	Bias in selection of the reported result
Jodłowska et al.[26] 2019	high	high	low	some concerns	low	low	low
Krasuska-Sławińska et al.[30] 2016	high	high	low	high	low	low	low
Lauritano and Petruzzi[31] 2012	high	high	low	high	low	low	low
Nemeth et al.[33] 2013	high	high	low	high	low	low	low
Oğuz et al.[34] 2004	high	high	low	some concerns	low	low	low

Table 6. Assessment of the quality of studies using the Joanna Briggs Institute (JBI) critical appraisal tool for case–control studies

Study	Q1	Q2	Q3	Q4	Q5	Q6	Q7	Q8	Q9	Q10	Overall score
Cubukcu et al.[45] 2012	Y	Y	Y	Y	Y	N	N	Y	Y	Y	8
Kılınç et al.[29] 2019	Y	Y	Y	Y	Y	N	N	Y	Y	Y	8
Proc et al.[6] 2016	Y	Y	U	Y	U	N	N	Y	Y	Y	6
Quispe et al.[36] 2019	Y	Y	Y	Y	Y	N	N	Y	Y	Y	8
Ruyssinck et al.[37] 2019	Y	Y	U	Y	U	N	N	Y	Y	Y	6
Shum et al.[38] 2020	Y	Y	U	Y	U	N	N	Y	Y	Y	6
Singh et al.[39] 2021	Y	Y	U	Y	U	N	N	Y	Y	N	5
Sonis et al.[40] 1990	Y	Y	U	Y	U	N	N	Y	N	Y	5

of tooth agenesis in cancer patients compared to non-cancer patients (unadjusted *OR*: 3.12; 95% *CI*: 2.01–4.83; $p < 0.00001$) (Fig. 3). Dental abnormalities were found to be more common among cancer patients than in healthy controls in most of the reviewed studies.[6,19,21,30–31,33,44,45] All the details are provided in Table 2.

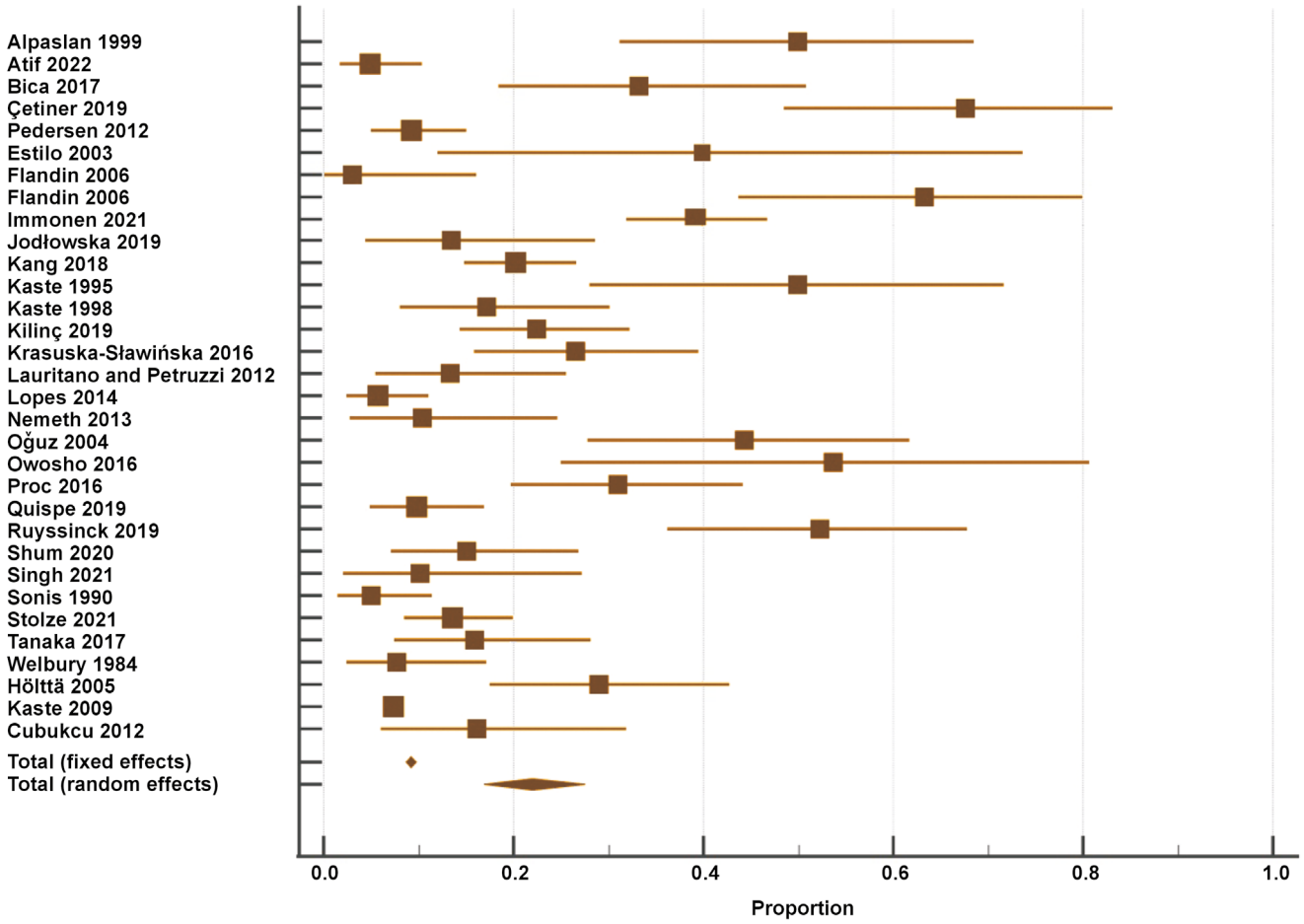
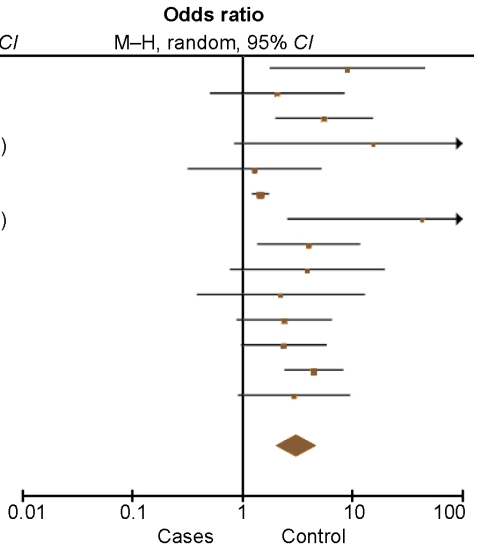


Fig. 2. Results of the single-arm meta-analysis presenting the prevalence of tooth agenesis in pediatric cancer patients

The study by Flandin et al. involved 2 types of patients: patients in the first group received radiation therapy, and the second group received chemotherapy.

Study or subgroup	Cases events	Cases total	Control events	Control total	Weight	Odds ratio M–H, random, 95% *CI*
Alpaslan 1999	15	30	2	20	5.0%	9.00 (1.77, 45.79)
Atif 2022	6	120	3	121	6.0%	2.07 (0.51, 8.48)
Cetiner 2019	21	3	11	40	8.5%	5.54 (1.99, 15.42)
Cubukcu 2012	6	37	0	37	2.0%	15.48 (0.84, 285.57)
Jodłowska 2019	5	37	4	37	6.0%	1.29 (0.32, 5.24)
Kaste 2009	698	9,308	149	2,831	15.4%	1.46 (1.22, 1.75)
Kilinc 2019	21	93	0	72	2.1%	43.00 (2.56, 723.35)
Krasuska-Slawinska 2016	16	60	5	60	8.1%	4.00 (1.36, 11.77)
Lauritano 2012	7	52	2	52	5.0%	3.89 (0.77, 19.70)
Nemeth 2013	4	38	2	40	4.5%	2.24 (0.38, 12.98)
Oguz 2004	16	36	9	36	8.7%	2.40 (0.88, 6.53)
Peddersen 2012	14	150	8	193	9.5%	2.38 (0.97, 5.83)
Proc 2016	19	61	48	521	12.0%	4.46 (2.40, 8.27)
Quispe 2019	11	111	4	111	7.4%	2.94 (0.91, 9.54)
Total (95% *CI*)	**859**		**247**		**100.0%**	**3.12 (2.01, 4.83)**
Total events		10,164		4,171		



Heterogeneity: $\tau^2 = 0.32$; $\chi^2 = 33.40$, $df = 13$ ($p = 0.001$); $I^2 = 61\%$
Test for overall effect: $Z = 5.09$ ($p < 0.00001$)

Fig. 3. Forest plot for the prevalence of tooth agenesis in cancer patients compared to non-cancer patients

CI – confidence interval; *df* – degrees of freedom.

Figure 4 presents an exemplatory panoramic radiograph of a 15-year-old male patient diagnosed with neuroblastoma at the age of 3. The patient underwent a 21-month treatment regimen consisting of combination therapy, which included high-dose and conventional CT, bone marrow transplantation and RTX. The patient suffers from hypodontia, short roots of teeth and microdontia.

Discussion

This systematic review was aimed at assessing the prevalence of tooth agenesis in childhood cancer survivors and healthy individuals. The findings revealed that the occurence of hypodontia was higher in children who had undergone cancer treatment compared to their healthy peers. The null hypothesis stating that childhood cancer survivors and healthy individuals would have the same prevalence of tooth agenesis was rejected. The presence of defects depended on various factors, including both individual characteristics of the child and the applied treatment. According to the peer-reviewed articles, hypodontia was estimated to affect between 1.4% and 66.42% of cancer patients.[3,6,8,16,19,20–39,40–46] In the healthy group, the prevalence of hypodontia ranged from 0% to 25%.[29,34] The number of missing teeth in the cancer groups ranged from 6 to 69.[6,19,27,33,35,36,46] It was also found that 15–85% of third molars were missing in cancer patients.[19,21]

The teeth most frequently affected by agenesis were second premolars and second molars. In healthy individuals, the most often missing teeth were lateral incisors. The prevalence of specific groups of microdontic teeth depended on the time of the treatment and the conditions of the most active mineralization.[13] A similar trend was observed with respect to the prevalence of hypodontia in particular tooth groups; however, the difference was not statistically significant. This phenomenon can be explained by the observation that, when exposed to particularly strong external factors, tooth buds undergo complete degradation, irrespective of their development stage.

Moreover, the overall dental development, as expressed by dental age, varied in cancer survivors and depended on the type of cancer and the implemented therapy.[5] In the majority of cases, the dental age of cancer survivors was accelerated, predominantly due to premature closure of root apices. The dental age was significantly delayed in patients with familial adenomatous polyposis (FAP)-associated hepatoblastoma. However, the changes in dental age were independent of sex, age, or the duration of treatment.[9,49]

Numerous factors can influence the occurrence of hypodontia, with the most prevalent one being the age of the patient at the time of diagnosis and therapy. The younger the age of the child at the time of diagnosis, the earlier the stage of tooth development and the greater the risk of more serious dental defects. The significant age limit varied in different publications, although it was consistently below 7 years of age.[3,8,16,19,20,23,27,29,32–37,40,43,44,46,47] This is in line with the time of the most active development of tooth buds, which is considered to be the age between 6 and 8 years.[6]

The age at diagnosis correlated not only with the frequency but also with the severity of dental abnormalities. The patients in the youngest group presented with tooth agenesis

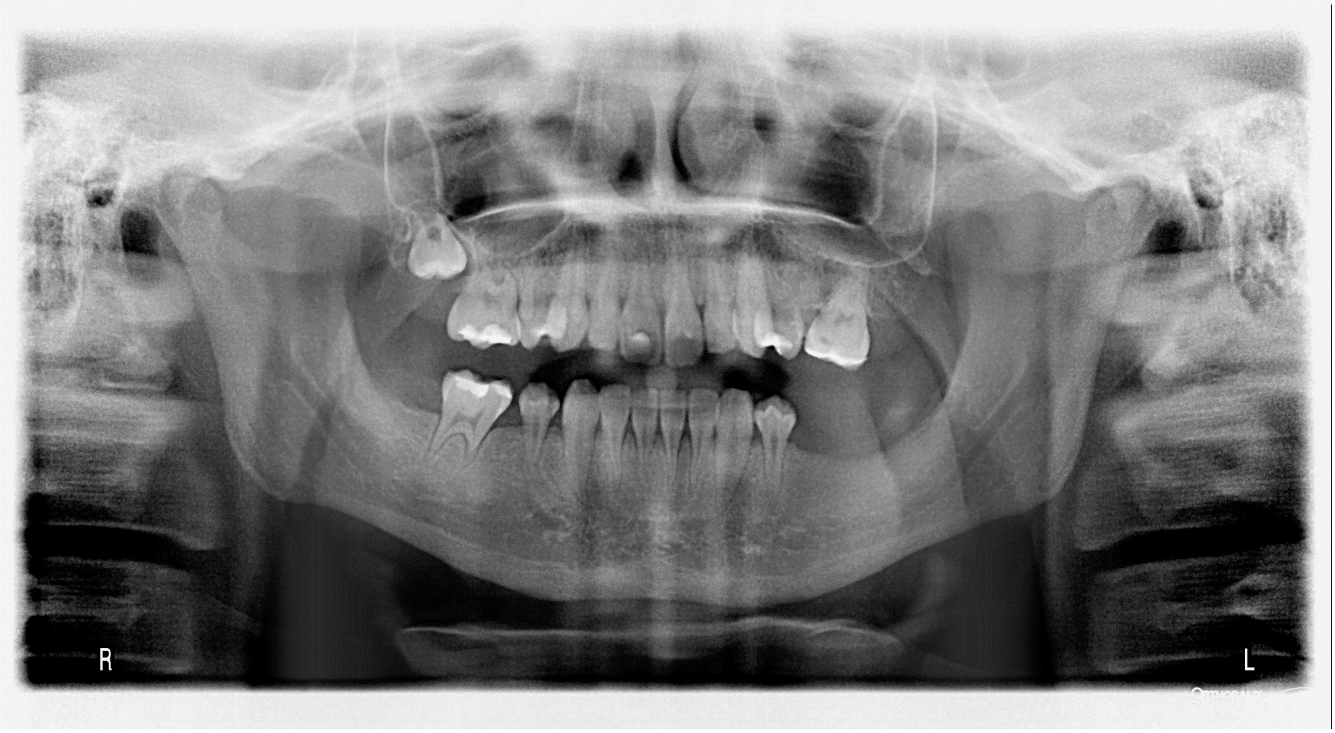


Fig. 4. Panoramic radiograph of a 15-year-old male patient diagnosed with neuroblastoma at the age of 3 years, who suffers from hypodontia, short roots of teeth and microdontia

or microdontia, while those in the oldest group demonstrated the most prevalent occurrence of abnormal root development.[6,20,23,26,27,29,34] Additionally, the prevalence of combined disturbances was significantly lower in the youngest group compared to the other groups.[2] The co-occurrence of different dental defects was frequently observed, as most of the cancer survivors suffered from more than one type of abnormality.[5] Apart from hypodontia, the most frequently reported complications were microdontia, root deformation with premature apexification, enamel discoloration, and unerupted teeth. Patients with rhabdomyosarcoma of the head or neck who underwent treatment, including RTX, suffered from oral diseases, i.e., bony hypoplasia/facial asymmetry, trismus, velopharyngeal insufficiency, radiographically underdeveloped mandible, severe malocclusion, caries, hyposalivation/xerostomia, and gingivitis.[23,27,35] On the other hand, factors like malocclusion, trauma, severe pain stimuli, parafunctional activities, and psychological elements, including stress, anxiety and depression can lead to temporomandibular disorders (TMD).[50]

It is worth noting that the dose, type and number of cytostatic drugs administered were identified as risk factors for hypodontia and other dental defects. The use of more than 4 different chemotherapeutic agents and heavy metals has been identified as a significant risk factor for severe dental disturbances.[2] Additionally, chemotherapeutic drugs such as vincristine, cyclophosphamide, doxorubicin, ifosfamide, etoposide, and cisplatin significantly increased the risk of tooth agenesis.[5] Interestingly, it has been reported that equivalent doses of cyclophosphamide above 8,000 mg/m^2 are associated with a higher number of teeth missing due to agenesis.[6]

Total body irradiation is performed in cancer patients to suppress the immune system and prevent the rejection of bone marrow transplantation (BMT).[7] The side effects of TBI are most pronounced in terms of height and weight delay, while other complications of TBI include hypothyroidism, cataracts and a high incidence of secondary tumors.[6] However, dental complications, such as tooth agenesis, were not found more frequently in the group of patients who had undergone TBI treatment.[24,43] As for patients treated with TBI, agenesis was more frequent in individuals receiving busulfan (63.2%) than in those treated with other chemotherapeutic agents (37.5%).[7]

On the other hand, some studies have documented a significantly higher prevalence of tooth agenesis in children treated with HSCT (similarly to BMT).[2] The prevalence of agenesis and microdontia affecting at least 1 permanent tooth in cancer patients who had undergone HSCT treatment was much higher when compared to the controls. Moreover, 92.3% of children aged ≤3 years old at the time of HSCT treatment exhibited tooth agenesis.[5] The condition manifested more prominently in certain tooth groups, including first and second premolars in the maxilla and mandible, as well as second molars in the mandible (all p-values <0.001).[6]

The relationship between the application of head and neck RTX and the occurrence of dental changes was also investigated.[23,35,40] The radiation exposure of ≥20 Gy to the dentition was significantly associated with an increased risk of 1 or more dental abnormalities.[6] After RTX, the frequency of dental changes reached from 80% up to 100% among children under 5 years of age.[23,40]

Impaired tooth development constitutes a complication that arises subsequent to cancer treatment. Tumor-induced osteomalacia has been widely described in patients ranging in age from 9 months to 90 years, with a broad spectrum of tumor types. In adults, the primary concern is a decreased level of serum phosphate, while in children (aged <18 years), it is a low or improperly circulating concentration of 1,25-dihydroxyvitamin D.[51] The 1,25-dihydroxyvitamin D, in turn, belongs to the group of interacting circulating hormones and their key receptors that regulate the state of calcium homeostasis.[52] Calcium and phosphate play a key role in the mineralization of teeth and bones. Disturbances in the levels of these minerals during the developmental phase of an organism may partially account for the increased occurrence of dental defects in childhood cancer survivors.

Tooth agenesis is more prevalent among cancer survivors in comparison to healthy controls. There are several factors related to cancer and its treatment that contribute to the occurrence of agenesis. Given the high risk of complications in cancer patients, increased dental attention and care are required.

Conclusions

Patients who underwent childhood cancer treatment may experience dental complications more frequently compared to the general population. The dissemination of knowledge on this subject among clinicians is necessary to ensure the provision of specialized dental care to such patients, thereby facilitating their recovery and enhancing their quality of life.

Trial registration

The study was registered with PROSPERO (registration No. CRD42022308068).

Ethics approval and consent to participate

Not applicable.

Data availability

The datasets generated and/or analyzed during the current study are available from the corresponding author on reasonable request.

Consent for publication

Not applicable.

Use of AI and AI-assisted technologies

Not applicable.

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References

1. Luongo C, Randazzo E, Iughetti L, et al. Cardiometabolic risk in childhood cancer survivors. *Minerva Pediatr (Torino)*. 2021;73(6):588–605. doi:10.23736/S2724-5276.21.06544-7
2. Dieffenbach BV, Liu Q, Murphy AJ, et al. Late-onset kidney failure in survivors of childhood cancer: A report from the Childhood Cancer Survivor Study. *Eur J Cancer*. 2021;155:216–226. doi:10.1016/j.ejca.2021.06.050
3. Kang CM, Hahn SM, Kim HS, et al. Clinical risk factors influencing dental developmental disturbances in childhood cancer survivors. *Cancer Res Treat*. 2018;50(3):926–935. doi:10.4143/crt.2017.296
4. van Iersel L, Mulder RL, Denzer C, et al. Hypothalamic-pituitary and other endocrine surveillance among childhood cancer survivors. *Endocr Rev*. 2022;43(5):794–823. doi:10.1210/endrev/bnab040
5. Strebel S, Waespe N, Kuehni CE. Hearing loss in childhood cancer survivors. *Lancet Child Adolesc Health*. 2021;5(5):e17. doi:10.1016/S2352-4642(21)00099-7
6. Proc P, Szczepańska J, Skiba A, Zubowska M, Fendler W, Młynarski W. Dental anomalies as late adverse effect among young children treated for cancer. *Cancer Res Treat*. 2016;48(2):658–667. doi:10.4143/crt.2015.193
7. Patni T, Lee CT, Li Y, et al. Factors for poor oral health in long-term childhood cancer survivors. *BMC Oral Health*. 2023;23(1):73. doi:10.1186/s12903-023-02762-0
8. Bica C, Chincesan M, Esian D, et al. Dental development in children after chemotherapy. *Rev Chim*. 2017;68(6):1397–1400. doi:10.37358/rc.17.6.5681
9. Proc P, Szczepańska J, Zubowska M, Zalewska-Szewczyk B, Młynarski W. The broad variability in dental age observed among childhood survivors is cancer specific. *Cancer Res Treat*. 2021;53(1):252–260. doi:10.4143/crt.2020.275
10. Feghali S, Vi-Fane B, Picard A, Kadlub N. Dental and orthodontic follow-up in nevoid basal cell carcinoma syndrome patient with odontogenic keratocystic tumors. *J Stomatol Oral Maxillofac Surg*. 2022;123(3):e57–e61. doi:10.1016/j.jormas.2021.07.009
11. Qin W, Wan QQ, Ma YX, et al. Manifestation and mechanisms of abnormal mineralization in teeth. *ACS Biomater Sci Eng*. 2023;9(4):1733–1756. doi:10.1021/acsbiomaterials.1c00592
12. Jodłowska A, Postek-Stefańska L. Systemic anticancer therapy details and dental adverse effects in children. *Int J Environ Res Public Health*. 2022;19(11):6936. doi:10.3390/ijerph19116936
13. Rabassa-Blanco J, Brunet-Llobet L, Marcote-Sinclair P, Balsells-Mejía S, Correa-Llano MG, Miranda-Rius J. Prevalence of, and risk factors for, dental sequelae in adolescents who underwent cancer therapy during childhood. *Oral Dis*. 2024;30(2):604–614. doi:10.1111/odi.14317
14. Halperson E, Matalon V, Goldstein G, et al. The prevalence of dental developmental anomalies among childhood cancer survivors according to types of anticancer treatment. *Sci Rep*. 2022;12(1):4485. doi:10.1038/s41598-022-08266-1
15. Jensen JM, Skakkebæk A, Gaustadness M, et al. Familial colorectal cancer and tooth agenesis caused by an AXIN2 variant: How do we detect families with rare cancer predisposition syndromes? *Fam Cancer*. 2022;21(3):325–332. doi:10.1007/s10689-021-00280-y
16. Stolze J, Vlaanderen KCE, Holtbach FCED, et al. Long-term effects of childhood cancer treatment on dentition and oral health: A dentist survey study from the DCCSS LATER 2 Study. *Cancers (Basel)*. 2021;13(21):5264. doi:10.3390/cancers13215264
17. Page MJ, McKenzie JE, Bossuyt PM, et al. The PRISMA 2020 statement: An updated guideline for reporting systematic reviews. *Syst Rev*. 2021;10(1):89. doi:10.1186/s13643-021-01626-4
18. Ouzzani M, Hammady H, Fedorowicz Z, Elmagarmid A. Rayyan – a web and mobile app for systematic reviews. *Syst Rev*. 2016;5(1):210. doi:10.1186/s13643-016-0384-4
19. Alpaslan G, Alpaslan C, Gögen H, Oğuz A, Cetiner S, Karadeniz C. Disturbances in oral and dental structures in patients with pediatric lymphoma after chemotherapy: A preliminary report. *Oral Surg Oral Med Oral Pathol Oral Radiol Endod*. 1999;87(3):317–321. doi:10.1016/s1079-2104(99)70215-5
20. Atif M, Mathur VP, Tewari N, Bansal K, Rahul M, Bakhshi S. Long-term effect of anticancer therapy on dentition in childhood cancer survivors: An observational, cross-sectional study. *Indian J Pediatr*. 2022;89(4):327–332. doi:10.1007/s12098-021-03818-1
21. Çetiner D, Çetiner S, Uraz A, et al. Oral and dental alterations and growth disruption following chemotherapy in long-term survivors of childhood malignancies. *Support Care Cancer*. 2019;27(5):1891–1899. doi:10.1007/s00520-018-4454-0
22. Pedersen LB, Clausen N, Schrøder H, Schmidt M, Poulsen S. Microdontia and hypodontia of premolars and permanent molars in childhood cancer survivors after chemotherapy. *Int J Paediatr Dent*. 2012;22(4):239–243. doi:10.1111/j.1365-263X.2011.01199.x
23. Estilo CL, Huryn JM, Kraus DH, et al. Effects of therapy on dentofacial development in long-term survivors of head and neck rhabdomyosarcoma: The memorial sloan-kettering cancer center experience. *J Pediatr Hematol Oncol*. 2003;25(3):215–222. doi:10.1097/00043426-200303000-00007
24. Flandin I, Hartmann O, Michon J, et al. Impact of TBI on late effects in children treated by megatherapy for Stage IV neuroblastoma. A study of the French Society of Pediatric Oncology. *Int J Radiat Oncol Biol Phys*. 2006;64(5):1424–1431. doi:10.1016/j.ijrobp.2005.10.020
25. Immonen E, Nikkilä A, Peltomäki T, Aine L, Lohi O. Late adverse effects of childhood acute lymphoblastic leukemia treatment on developing dentition. *Pediatr Blood Cancer*. 2021;68(9):e29200. doi:10.1002/pbc.29200
26. Jodłowska A, Sobol-Milejska G, Postek-Stefańska L. A critical look at prevalence assessment of dental abnormalities after chemotherapy. Clinical research. *J Stoma*. 2019;72(3):95–105. doi:10.5114/jos.2019.87522
27. Kaste SC, Hopkins KP, Bowman LC. Dental abnormalities in long-term survivors of head and neck rhabdomyosarcoma. *Med Pediatr Oncol*. 1995;25(2):96–101. doi:10.1002/mpo.2950250209
28. Kaste SC, Hopkins KP, Bowman LC, Santana VM. Dental abnormalities in children treated for neuroblastoma. *Med Pediatr Oncol*. 1998;30(1):22–27. doi:10.1002/(sici)1096-911x(199801)30:1<22::aid-mpo8>3.0.co;2-2
29. Kılınç G, Bulut G, Ertuğrul F, et al. Long-term dental anomalies after pediatric cancer treatment in children. *Turk J Haematol*. 2019;36(3):155–161. doi:10.4274/tjh.galenos.2018.2018.0248
30. Krasuska-Sławińska E, Brożyna A, Dembowska-Bagińska B, Olczak-Kowalczyk D. Antineoplastic chemotherapy and congenital tooth abnormalities in children and adolescents. *Contemp Oncol (Pozn)*. 2016;20(5):394–401. doi:10.5114/wo.2016.64602
31. Lauritano D, Petruzzi M. Decayed, missing and filled teeth index and dental anomalies in long-term survivors leukaemic children: A prospective controlled study. *Med Oral Patol Oral Cir Bucal*. 2012;17(6):e977–e980. doi:10.4317/medoral.17955
32. Lopes NNF, Petrilli AS, Caran EMM, França CM, Chilvarquer I, Lederman H. Dental abnormalities in children submitted to antineoplastic therapy. *J Dent Child (Chic)*. 2006;73(3):140–145. PMID:17367030.
33. Nemeth O, Hermann P, Kivovics P, Garami M. Long-term effects of chemotherapy on dental status of children cancer survivors. *Pediatr Hematol Oncol*. 2013;30(3):208–215. doi:10.3109/08880018.2013.763391
34. Oğuz A, Cetiner S, Karadeniz C, Alpaslan G, Alpaslan C, Pinarli G. Long-term effects of chemotherapy on orodental structures in children with non-Hodgkin's lymphoma. *Eur J Oral Sci*. 2004;112(1):8–11. doi:10.1111/j.0909-8836.2004.00094.x

35. Owosho AA, Brady P, Wolden SL, et al. Long-term effect of chemotherapy-intensity-modulated radiation therapy (chemo-IMRT) on dentofacial development in head and neck rhabdomyosarcoma patients. *Pediatr Hematol Oncol*. 2016;33(6):383–392. doi:10.1080/08880018.2016.1219797
36. Quispe RA, Rodrigues ACC, Buaes AMG, Capelozza ALA, Rubira CMF, Santos PSS. A case–control study of dental abnormalities and dental maturity in childhood cancer survivors. *Oral Surg Oral Med Oral Pathol Oral Radiol*. 2019;128(5):498–507.e3. doi:10.1016/j.oooo.2019.07.005
37. Ruyssinck L, Toulouse K, Bordon Cueto de Braem V, Cauwels R, Dhooge C. Impact of hematopoietic stem cell transplantation on dental development. *Biol Blood Marrow Transplant*. 2019;25(1):107–113. doi:10.1016/j.bbmt.2018.08.027
38. Shum M, Mahoney E, Naysmith K, et al. Associations between childhood cancer treatment and tooth agenesis. *N Z Med J*. 2020;133(1523):41–54. PMID:33032302.
39. Singh A, Modak S, Solano AK, et al. Mandibular metastases in neuroblastoma: Outcomes and dental sequelae. *Pediatr Blood Cancer*. 2021;68(4):e28918. doi:10.1002/pbc.28918
40. Sonis AL, Tarbell N, Valachovic RW, Gelber R, Schwenn M, Sallan S. Dentofacial development in long-term survivors of acute lymphoblastic leukemia. A comparison of three treatment modalities. *Cancer*. 1990;66(12):2645–2652. doi:10.1002/1097-0142(19901215)66:12<2645::aid-cncr2820661230>3.0.co;2-s
41. Tanaka M, Kamata T, Yanagisawa R, et al. Increasing risk of disturbed root development in permanent teeth in childhood cancer survivors undergoing cancer treatment at older age. *J Pediatr Hematol Oncol*. 2017;39(3):e150–e154. doi:10.1097/MPH.0000000000000788
42. Welbury RR, Craft AW, Murray JJ, Kernahan J. Dental health of survivors of malignant disease. *Arch Dis Child*. 1984;59(12):1186–1187. doi:10.1136/adc.59.12.1186
43. Hölttä P, Alaluusua S, Saarinen-Pihkala UM, Peltola J, Hovi L. Agenesis and microdontia of permanent teeth as late adverse effects after stem cell transplantation in young children. *Cancer*. 2005;103(1):181–190. doi:10.1002/cncr.20762
44. Kaste SC, Goodman P, Leisenring W, et al. Impact of radiation and chemotherapy on risk of dental abnormalities: A report from the Childhood Cancer Survivor Study. *Cancer*. 2009;115(24):5817–5827. doi:10.1002/cncr.24670
45. Cubukcu CE, Sevinir B, Ercan I. Disturbed dental development of permanent teeth in children with solid tumors and lymphomas. *Pediatr Blood Cancer*. 2012;58(1):80–84. doi:10.1002/pbc.22902
46. van der Pas-van Voskuilen IGM, Veerkamp JSJ, Raber-Durlacher JE, et al. Long-term adverse effects of hematopoietic stem cell transplantation on dental development in children. *Support Care Cancer*. 2009;17(9):1169–1175. doi:10.1007/s00520-008-0567-1
47. Aarnoutse R, de Vos-Geelen JMPGM, Penders J, et al. Study protocol on the role of intestinal microbiota in colorectal cancer treatment: A pathway to personalized medicine 2.0. *Int J Colorectal Dis*. 2017;32(7):1077–1084. doi:10.1007/s00384-017-2819-3
48. Tanem KE, Stensvold E, Wilberg P, Skaare AB, Brandal P, Herlofson BB. Oral and dental late effects in long-term survivors of childhood embryonal brain tumors. *Support Care Cancer*. 2022;30(12):10233–10241. doi:10.1007/s00520-022-07405-8
49. Hölttä P, Hovi L, Saarinen-Pihkala UM, Peltola J, Alaluusua S. Disturbed root development of permanent teeth after pediatric stem cell transplantation. Dental root development after SCT. *Cancer*. 2005;103(7):1484–1493. doi:10.1002/cncr.20967
50. Minervini G, Franco R, Marrapodi MM, et al. Correlation between temporomandibular disorders (TMD) and posture evaluated through the Diagnostic Criteria for Temporomandibular Disorders (DC/TMD): A systematic review with meta-analysis. *J Clin Med*. 2023;12(7):2652. doi:10.3390/jcm12072652
51. Bosman A, Palermo A, Vanderhulst J, et al. Tumor-induced osteomalacia: A systematic clinical review of 895 cases. *Calcif Tissue Int*. 2022;111(4):367–379. doi:10.1007/s00223-022-01005-8
52. Matikainen N, Pekkarinen T, Ryhänen EM, Schalin-Jäntti C. Physiology of calcium homeostasis: An overview. *Endocrinol Metab Clin North Am*. 2021;50(4):575–590. doi:10.1016/j.ecl.2021.07.005

Review

Effect of melatonin as an adjunct to non-surgical periodontal therapy in the treatment of periodontitis: A systematic review and meta-analysis

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Abstract

Gingivitis is defined as the inflammation of the gums. The condition may spread to other parts of the periodontium, including the periodontal ligament and alveolar bone, and lead to bony defects. Melatonin has a positive impact on the bone healing process, a phenomenon attributable to its antioxidant properties, as well as its capacity to regulate bone cells and promote angiogenesis. The present meta-analysis aimed to evaluate the effect of melatonin as an adjunct to non-surgical periodontal therapy (NSPT) in the treatment of periodontitis. A thorough electronic search of the PubMed®/MEDLINE and Google Scholar databases was conducted, in addition to a manual search of the reference lists of archived articles published until May 2023. Among the 8 reviewed articles, 3 studies that evaluated probing depth (PD) and had a 6-month follow-up period were considered for the meta-analysis. After extracting the relevant information, the risk of bias was estimated. A summary of the estimates for standardized mean differences (*SMD*s) from fixed-effects and random-effects models was obtained based on the mean treatment differences reported in the selected studies. The results demonstrated the overall estimated effect from the fixed-effects model ($SMD = 0.862$, 95% confidence interval (95% *CI*): 0.517–1.207, $p < 0.001$) and the random-effects model ($SMD = 0.869$, 95% *CI*: 0.499–1.238, $p < 0.001$), with minimal inconsistency as indicated by the Q statistic and I^2, respectively. Thus, it can be concluded that melatonin may be used as an adjunctive medication with NSPT for the management of periodontitis.

Keywords: melatonin, periodontitis, periodontal disease, bone defects, chronic gingivitis

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Highlights

- Melatonin, with its antioxidant and bone-regenerative properties, shows potential as an adjunct to non-surgical periodontal therapy (NSPT).
- Adjunctive use of melatonin with NSPT significantly improved probing depth reduction, with both fixed- and random-effects models showing consistent outcomes and minimal heterogeneity.
- Melatonin may enhance periodontal healing and serve as a supportive therapy in periodontitis management.

Introduction

Periodontitis, a chronic inflammatory disease that affects the supporting structures of the teeth, poses a significant oral health challenge worldwide. The condition is characterized by the destruction of the periodontal ligament and alveolar bone, leading to tooth loss if left untreated.[1] In recent years, researchers have been exploring alternative therapeutic approaches to complement conventional periodontal treatments. One such avenue of investigation involves the role of melatonin, a hormone primarily known for regulating sleep–wake cycles,[2] in the management of periodontitis.

Melatonin, a hormone synthesized by the pineal gland in the brain, exhibits diverse physiological functions beyond sleep regulation.[3] It possesses powerful antioxidant, anti-inflammatory and immunomodulatory properties, making it an attractive candidate for periodontal therapy.[4] Periodontitis is a multifactorial disease, and as it is also driven by an imbalanced host immune response[5] and oxidative stress,[6,7] targeting these pathological factors is crucial for effective treatment.

Understanding the role of melatonin in periodontal health and disease progression may pave the way for innovative therapeutic strategies that target the underlying mechanisms of periodontitis. Melatonin can potentially enhance the outcomes of conventional periodontal treatment, that is, non-surgical periodontal therapy (NSPT), and contribute to improved oral health outcomes in individuals affected by this prevalent chronic inflammatory condition.

Melatonin can be administered systemically or locally, depending on the desired therapeutic effect and the specific needs of the patient.[8]

Systemic administration involves the oral ingestion of the hormone, typically in the form of tablets or capsules. Upon ingestion, melatonin is absorbed into the bloodstream and distributed throughout the body. Systemic administration of melatonin enables its potential systemic effects, including its antioxidant and immunomodulatory properties.[5,6] Moreover, it may influence sleep patterns, which can indirectly impact the overall health and healing processes in individuals with periodontitis.[9]

On the other hand, local administration of melatonin involves applying the hormone directly to the affected periodontal tissues. This objective can be achieved through the use of melatonin-containing mouthwashes, gels, or local drug delivery systems.[10] Local administration allows for targeted delivery of melatonin to the site of inflammation and infection in the periodontal pockets. Consequently, the hormone may exert its anti-inflammatory and tissue regenerative effects directly in the periodontal tissues, potentially enhancing the efficacy of conventional periodontal treatments.

By maximizing the potential of melatonin as an adjunctive therapy, we can improve patient outcomes, reduce the progression of periodontal disease, and promote overall oral health.

The objective of this systematic review and meta-analysis was to evaluate the effectiveness of both systemic and local administration of melatonin in the management of periodontitis. The study aimed to provide a comprehensive overview of the current understanding of the effect of melatonin in periodontitis. A review of studies that have explored the potential benefits of melatonin as an adjunctive treatment in periodontitis has been conducted.

Material and methods

The PRISMA (Preferred Reporting Items for Systematic reviews and Meta-Analyses) guidelines[11] implemented by the Cochrane Collaboration[12] were used for reporting the present analysis. The protocol of this study was registered in PROSPERO, the International Prospective Register of Systematic Reviews (ID: CRD42023438265).

Focused question

The research question for this systematic review and meta-analysis was specifically framed based on the PICO criteria, as follows: Population (P) – patients with chronic periodontitis; Intervention (I) – systemic and local administration of melatonin; Control (C) – administration of placebo; Outcome (O) – probing depth (PD).[13,14]

Information sources and search strategy

A comprehensive literature search of the PubMed®/MEDLINE and Google Scholar databases was performed to identify relevant studies. In addition, a manual search of the reference lists of archived articles was conducted. The search strategy included a combination of the following keywords: "periodontitis"; "periodontal diseases"; "non-surgical periodontal therapy"; "melatonin"; "randomized controlled trial"; "RCT"; and "clinical trials". The search was limited to studies published up to May 2023.

Inclusion criteria

The inclusion criteria encompassed randomized controlled trials (RCTs) and controlled clinical trials investigating the use of melatonin as an adjunct to NSPT in patients with periodontitis, which reported clinical parameters such as PD.

Exclusion criteria

Studies written in languages other than English, as well as in vitro and animal studies, case reports, case series, reviews, and conference abstracts were excluded from the analysis.

Selection process

Three reviewers (PAK, SR and PR) independently screened the titles and abstracts of the articles based on the established inclusion and exclusion criteria. Subsequently, the abstracts of all relevant studies and their possible significance were independently reviewed by the investigators. To resolve any discrepancies, an additional reviewer (SU) was consulted. The full-text articles of potentially eligible studies were obtained and assessed for inclusion. At this stage, discrepancies among the reviewers were resolved through discussion and consensus. Studies were selected based on the pre-defined inclusion and exclusion criteria, as stated earlier.

Outcome measures

Probing depth, referred to as pocket depth if periodontal disease is present, is defined as the distance from the gingival margin to the apical portion of the gingival sulcus.[15] The present meta-analysis analyzed PD reported in the included articles.

Software for meta-analysis

The present meta-analysis was performed in the MedCalc® statistical software v. 20.118 (MedCalc Software Ltd, Ostend, Belgium).

Data extraction

The data from the included studies was extracted using a standardized data extraction form. The extracted data included characteristics of the study (authors, year of publication, study design), characteristics of the participants (sample size, demographics), intervention details (dosage, duration), and outcome measures (PD). Disagreements in data extraction were resolved through discussion or by consulting the 3rd reviewer (SU).

Assessment of the methodological quality and risk of bias

The quality of the included studies was evaluated independently by 2 reviewers (PAK and SR) using the Cochrane Risk of Bias tool for RCTs. The tool assessed the risk of bias in the domains of random sequence generation, allocation concealment, blinding, incomplete outcome data, selective reporting, and other biases. During the judgment process, each "yes" or "no" indicated a low or high risk of bias, respectively, while "unclear" meant an uncertain risk of bias. The study was grouped as "low risk of bias" when all the aspects were deemed to be of a low risk of bias. Conversely, it was classified as "high" or "uncertain risk of bias" when one or more aspects indicated a high or unclear risk of bias. Discrepancies in the quality assessment were resolved through discussion or involvement of the 3rd reviewer (SU).

Data synthesis

The meta-analysis was performed using appropriate statistical methods. In 2 of the 3 included studies, melatonin was administered systemically,[16,17] while in the remaining study, local administration of the hormone was implemented.[18] The effect sizes (mean difference, odds ratio) and the respective 95% confidence intervals (95% *CI*s) were calculated for the outcome measure. The statistical heterogeneity of the included studies was assessed using the I^2 statistic. In instances where substantial heterogeneity was detected, a random-effects model was used; otherwise, a fixed-effects model was applied. Subgroup analyses and sensitivity analyses were conducted as appropriate. The presence of publication bias was assessed using funnel plots and statistical tests (Egger's test and Begg's test).

Data interpretation and reporting

The findings of the systematic review and meta-analysis were interpreted and discussed. The strengths, limitations and implications of the included studies were considered. The results were reported following the PRISMA guidelines.

Several studies have been conducted to evaluate the effectiveness of different interventions in the treatment of patients with periodontitis, particularly involving the use of melatonin. These studies provide evidence on the effectiveness of various interventions in treating periodontitis patients, with outcomes such as PD, clinical attachment level (CAL), bleeding on probing (BOP), plaque index (PI), gingival index (GI), biochemical parameters, and bone fill being evaluated at different time points ranging from baseline to 8 weeks or 6 months.[16–23] The present meta-analysis was performed on only 3 studies with the same control group, a 6-month follow-up period, and PD as the outcome variable.

Results

Study selection

The initial search yielded a total of 513 studies, of which 405 were duplicates and were thus removed (Fig. 1). The titles and abstracts of the 108 studies were screened by 2 independent reviewers who removed 100 articles deemed to be irrelevant. The papers excluded from the analysis encompassed animal studies, case series, review articles, and studies published in languages other than English. The systematic review was conducted based on 8 full-text articles. Five of these papers were excluded due to insufficient data available. The number of subjects included in these studies exhibited variability in terms of assessment parameters, the presence of a control group and the duration of the follow-up. Finally, 3 studies analyzing PD were included in the meta-analysis.

Study characteristics

Table 1 presents a comprehensive overview of the 8 studies considered for this systematic review. Three studies were included in the meta-analysis (Table 2). The data from 70 test group participants from 3 RCTs was compared to that of 68 patients in the control groups. The minimum and maximum numbers of participants per group were 10 and 38, respectively. In the test groups, melatonin was administered either locally in the form of a gel or systemically as capsules. In the control groups, a placebo drug was supplied. A 6-month follow-up period was designated in all 3 studies.

Type of interventions

The 3 studies included the same test (scaling and root planing (SRP) + melatonin) and control (SRP + placebo) groups in the experimental design. In one of these articles, melatonin was administered locally, while in the remaining studies, participants were given oral capsules.

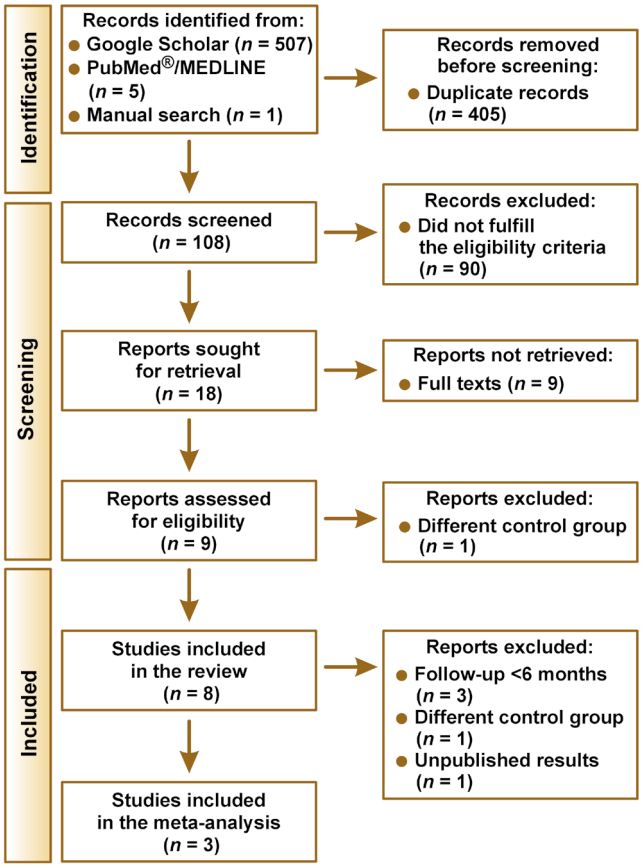

Fig. 1. PRISMA (Preferred Reporting Items for Systematic reviews and Meta-Analyses) flow diagram

Melatonin and PD

The estimated overall effect (i.e., pre–post reduction in PD) expressed as standardized mean difference (*SMD*) for the aggregate sample of 70 subjects from 3 studies was 0.862 for the fixed-effects model and 0.869 for the random-effects model (Table 2).

Both estimates were comparable, with overlapping 95% *CI*s. However, the estimate for the fixed-effects model was reported due to the non-significant heterogeneity ($p > 0.331$) and minimal inconsistency, as indicated by the Q statistic and I^2, respectively (Table 3). The forest plot presents individual and overall estimated *SMD* values for fixed-effects and random-effects models, along with respective 95% *CI*s (Fig. 2).

Publication bias

In the present meta-analysis, both Begg's funnel plot and the Egger's test were used to evaluate the publication bias. The funnel plot revealed no effect of the publication bias on the summary measure (i.e., *SMD* in PD from baseline to 6 months) plotted versus respective standard errors from all 3 studies (Fig. 3). The results of the Egger's test ($p = 0.823$) and Begg's test ($p = 0.602$) were non-significant ($p > 0.05$), indicating an absence of the publication bias (Table 3).

Table 1. Characteristics of the studies included in the systematic review

Study	Population	Test group	Control group	Outcome variables	Follow-up
Chitsazi et al. 2017[19]	periodontitis patients	1. SRP + melatonin capsules (orally, 2 mg/day for 4 weeks) 2. SRP + melatonin capsules + vitamin C (orally, 60 mg/day for females and 75 mg/day for males for 4 weeks)	SRP	PD, CAL, GI	baseline, 3 and 6 months
Bazyar et al. 2019[20]	patients with diabetes and chronic periodontitis	SRP + melatonin tablets (orally, two 250-mg tablets/day containing 3 mg of net melatonin each for 8 weeks)	SRP + placebo tablets	PD, CAL, BOP, PI, biochemical parameters	baseline and 8 weeks
El-Sharkawy et al. 2019[16]	periodontitis patients	SRP + melatonin capsules (orally, 10 mg/day before bedtime for 2 months)	SRP + placebo capsules	PD, CAL, BOP, biochemical parameters	baseline, 3 and 6 months
Tinto et al. 2020[17]	periodontitis patients	SRP + melatonin capsules (orally, 1 mg/day for 30 days)	SRP + placebo capsules	PD, PI, BOP	baseline and 6 months
Ahmed et al. 2021[21]	periodontitis patients	SRP + 5% melatonin gel	SRP + placebo gel	PD, CAL, PI, GI, biochemical parameters	baseline and 3 months
Anton et al. 2021[22]	patients with diabetes and chronic periodontitis	SRP + melatonin capsules (orally, two 250-mg tablets/day containing 3 mg of melatonin each for 8 weeks)	SRP + placebo capsules	PD, CAL, BOP, biochemical parameters	baseline and 8 weeks
Gonde et al. 2022[18]	periodontitis patients	SRP + 1% melatonin gel	SRP + placebo gel	PD, CAL, PI, mSBI, bone fill	baseline and 6 months
Dhande et al. 2024[23]	periodontitis patients	PRF + 1% melatonin gel	PRF	PD, CAL, DV, HPD, bone fill	baseline and 6 months

SRP – scaling and root planing; PD – probing depth; CAL – clinical attachment loss; GI – gingival index; PI – plaque index; mSBI – modified sulcus bleeding index; DV – defect volume; HPD – horizontal probing depth; PRF – platelet-rich fibrin.

Table 2. Results of the meta-analysis

Study	Study group, *n*	Control group, *n*	Total, *n*	*SMD*	*SE*	95% *CI*	*t*	*p*-value	Weight [%]	
									fixed	random
El-Sharkawy et al. 2019[16]	38	38	76	0.697	0.234	0.231–1.164	–	–	55.66	53.17
Tinto et al. 2020[17]	10	10	20	0.696	0.442	–0.233–1.625	–	–	15.59	16.94
Gonde et al. 2022[18]	22	22	44	1.271	0.326	0.614–1.928	–	–	28.75	29.89
Total (fixed-effects model)	70	70	140	0.862	0.175	0.517–1.207	4.938	<0.001*	100.00	100.00
Total (random-effects model)	70	70	140	0.869	0.187	0.499–1.238	4.644	<0.001*	100.00	100.00

* statistically significant ($p < 0.05$); *SMD* – standardized mean difference; *SE* – standard error; *CI* – confidence interval.

Table 3. Evaluation of the heterogeneity and publication bias

Variable			Result
Heterogeneity test		*Q*	2.2109
		df	2
		p-value	0.331
		I^2 (inconsistency)	9.54%
		95% *CI* for I^2	0.000–96.971
Publication bias	Egger's test	intercept	0.9407
		95% *CI*	–41.038–42.919
		p-value	0.823
	Begg's test	Kendall's Tau	0.3333
		p-value	0.602

df – degrees of freedom.

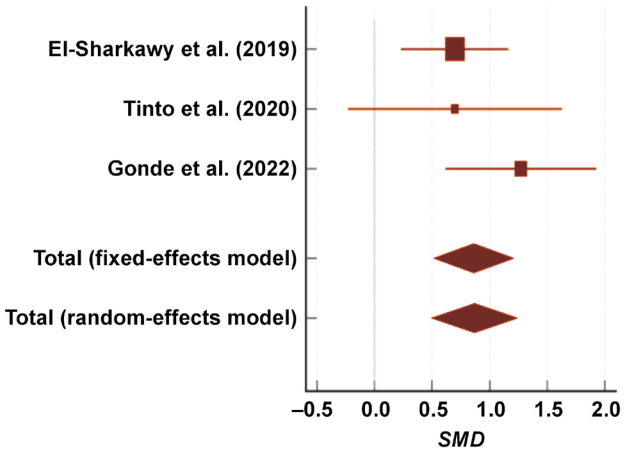

Fig. 2. Forest plot presenting individual and overall estimated standardized mean difference (*SMD*) values for fixed-effects and random-effects models along with respective 95% confidence intervals (95% *CIs*)

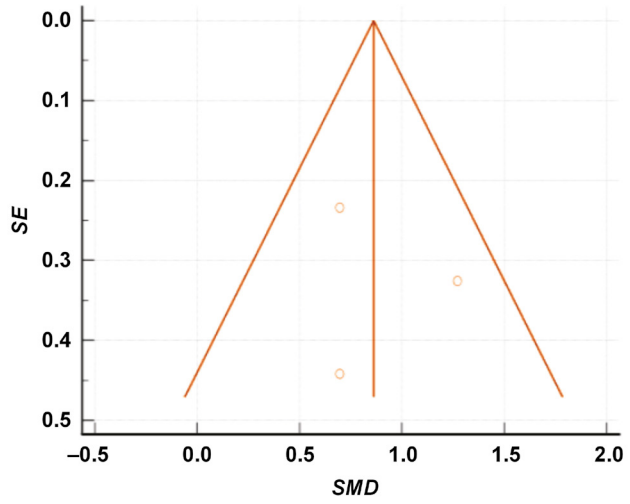

Fig. 3. Funnel plot for the assessment of publication bias
SE – standard error.

Discussion

Periodontitis is a chronic inflammatory disease that affects the supporting structures of the teeth, including the gingiva, the periodontal ligament and the alveolar bone.[1] The primary treatment approach for periodontitis is NSPT, which includes scaling and root planing. However, the use of adjunctive therapies to enhance the outcomes of NSPT has been explored in recent years. Melatonin, a hormone primarily involved in regulating sleep–wake cycles, has been identified as a promising adjunctive therapy due to its anti-inflammatory and antioxidant properties.[2]

The present systematic review and meta-analysis aimed to evaluate the effect of melatonin as an adjunct to NSPT in the treatment of periodontitis. The meta-analysis included 3 studies that measured periodontal parameters at baseline and after 6 months of treatment, comparing a melatonin-treated group to a control group.

The included papers showed variation in the patient population, as well as in the dosage and route of administration of melatonin. However, all studies assessed the clinical parameters of periodontitis, including PD, as well as biochemical parameters.

The baseline or pre-treatment PD measurements of the included studies ranged from 3.4 mm to 7.45 mm in the test group and from 3.00 mm to 6.95 mm in the control group. After 6 months of treatment, the PD measurements ranged from 2.3 mm to 3.95 mm in the test group and from 2.67 mm to 4.55 mm in the control group. The mean differences in PD between baseline and 6 months varied across the studies, with the test groups demonstrating generally greater reductions in comparison to the control groups.

A meta-analysis of the mean differences in PD between baseline and 6 months revealed that the use of melatonin as an adjunct to NSPT was associated with statistically significant improvements in periodontal parameters. El-Sharkawy et al. reported a mean difference of 2 mm in PD reduction in the test group compared to 1.4 mm in the control group.[16] Tinto et al. demonstrated a mean difference of 1.37 mm in the test group compared to 0.73 mm in the control group.[17] Gonde et al. noted a mean difference of 3.5 mm in the test group compared to 2.4 mm in the control group.[18]

These findings suggest that the adjunctive use of melatonin in NSPT may contribute to better clinical outcomes in terms of PD reduction. The anti-inflammatory and antioxidant properties of melatonin are believed to play a role in reducing periodontal inflammation and promoting tissue healing.[4] Additionally, the positive effect of melatonin on sleep quality may indirectly contribute to better periodontal health, as poor sleep has been associated with increased susceptibility to periodontal disease.[2,3]

It is important to note that the studies included in this meta-analysis had some limitations. The sample sizes were relatively small in some studies. Additionally, the heterogeneity of the study designs and patient populations makes it challenging to draw definitive conclusions. Well-designed RCTs with larger sample sizes are required to validate the findings of this meta-analysis and determine the optimal dosage and route of melatonin administration in periodontal therapy.

Based on the available evidence, the adjunctive use of melatonin in NSPT appears to have a positive effect on periodontal parameters, particularly in terms of PD reduction. However, further research is necessary to establish the optimal protocol for melatonin administration and to further explore its mechanisms of action in periodontal disease.

Limitations

The current evidence is derived from a limited number of RCTs and controlled clinical trials. Additional high-quality studies with larger sample sizes and longer follow-up periods are warranted to further validate these findings and determine the long-term effects of melatonin in periodontal therapy. Future research should focus on elucidating the optimal dosage, duration and timing of melatonin administration, as well as investigating its effects on other relevant clinical and inflammatory parameters.

Conclusions

The findings of this systematic review and meta-analysis provide evidence that the adjunctive use of melatonin in NSPT has a beneficial effect in the treatment of periodontitis. The analysis of the 3 included studies demonstrated that the addition of melatonin to NSPT resulted in significant improvements in PD compared

to NSPT alone. The results of the study suggest that the incorporation of melatonin into the treatment regimen has the potential to enhance the outcomes of NSPT and contribute to the management of periodontal disease.

Ethics approval and consent to participate

Not applicable.

Data availability

The datasets generated and/or analyzed during the current study are available from the corresponding author on reasonable request.

Consent for publication

Not applicable.

Use of AI and AI-assisted technologies

Not applicable.

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References

1. Listgarten MA. Pathogenesis of periodontitis. *J Clin Periodontol*. 1986;13(5):418–430. doi:10.1111/j.1600-051x.1986.tb01485.x
2. Pevet P, Challet E, Felder-Schmittbuhl MP. Melatonin and the circadian system: Keys for health with a focus on sleep. *Handb Clin Neurol*. 2021;179:331–343. doi:10.1016/B978-0-12-819975-6.00021-2
3. Karasek M, Winczyk K. Melatonin in humans. *J Physiol Pharmacol*. 2006;57 Suppl 5:19–39. PMID:17218758.
4. Muñoz-Jurado A, Escribano BM, Caballero-Villarraso J, et al. Melatonin and multiple sclerosis: Antioxidant, anti-inflammatory and immunomodulator mechanism of action. *Inflammopharmacology*. 2022;30(5):1569–1596. doi:10.1007/s10787-022-01011-0
5. He L, Liu L, Li T, et al. Exploring the imbalance of periodontitis immune system from the cellular to molecular level. *Front Genet*. 2021;12:653209. doi:10.3389/fgene.2021.653209
6. Sczepanik FSC, Grossi ML, Casati M, et al. Periodontitis is an inflammatory disease of oxidative stress: We should treat it that way. *Periodontol 2000*. 2020;84(1):45–68. doi:10.1111/prd.12342
7. Permuy M, López-Peña M, González-Cantalapiedra A, Muñoz F. Melatonin: A review of its potential functions and effects on dental diseases. *Int J Mol Sci*. 2017;18(4):865. doi:10.3390/ijms18040865
8. Balaji TM, Varadarajan S, Jagannathan R, et al. Melatonin as a topical/systemic formulation for the management of periodontitis: A systematic review. *Materials (Basel)*. 2021;14(9):2417. doi:10.3390/ma14092417
9. Ball J, Darby I. Mental health and periodontal and peri-implant diseases. *Periodontol 2000*. 2022;90(1):106–124. doi:10.1111/prd.12452
10. Reiter RJ, Rosales-Corral SA, Liu XY, Acuna-Castroviejo D, Escames G, Tan DX. Melatonin in the oral cavity: Physiological and pathological implications. *J Periodontal Res*. 2015;50(1):9–17. doi:10.1111/jre.12176
11. Arya S, Kaji AH, Boermeester MA. PRISMA Reporting Guidelines for Meta-analyses and Systematic Reviews. *JAMA Surg*. 2021;156(8):789–790. doi:10.1001/jamasurg.2021.0546
12. Cumpston M, Li T, Page MJ, et al. Updated guidance for trusted systematic reviews: A new edition of the Cochrane Handbook for Systematic Reviews of Interventions. *Cochrane Database Syst Rev*. 2019;10(10):ED000142. doi:10.1002/14651858.ED000142
13. Stone PW. Popping the (PICO) question in research and evidence-based practice. *Appl Nurs Res*. 2002;15(3):197–198. doi:10.1053/apnr.2002.34181
14. da Costa Santos CM, de Mattos Pimenta CA, Nobre MRC. The PICO strategy for the research question construction and evidence search. *Rev Lat Am Enfermagem*. 2007;15(3):508–511. doi:10.1590/s0104-11692007000300023
15. Listgarten MA. Periodontal probing: What does it mean? *J Clin Periodontol*. 1980;7(3):165–176. doi:10.1111/j.1600-051x.1980.tb01960.x
16. El-Sharkawy H, Elmeadawy S, Elshinnawi U, Anees M. Is dietary melatonin supplementation a viable adjunctive therapy for chronic periodontitis?–A randomized controlled clinical trial. *J Periodontal Res*. 2019;54(2):190–197. doi:10.1111/jre.12619
17. Tinto M, Sartori M, Pizzi I, Verga A, Longoni S. Melatonin as host modulating agent supporting nonsurgical periodontal therapy in patients affected by untreated severe periodontitis: A preliminary randomized, triple-blind, placebo-controlled study. *J Periodontal Res*. 2020;55(1):61–67. doi:10.1111/jre.12686
18. Gonde NP, Rathod SR, Kolte AP. Comparative evaluation of 1% melatonin gel in the treatment of intrabony defect: A randomized controlled clinical trial. *J Periodontol*. 2022;93(12):1878–1888. doi:10.1002/JPER.21-0515
19. Chitsazi M, Faramarzie M, Sadighi M, Shirmohammadi A, Hashemzadeh A. Effects of adjective use of melatonin and vitamin C in the treatment of chronic periodontitis: A randomized clinical trial. *J Dent Res Dent Clin Dent Prospects*. 2017;11(4):236–240. doi:10.15171/joddd.2017.041
20. Bazyar H, Gholinezhad H, Moradi L, et al. The effects of melatonin supplementation in adjunct with non-surgical periodontal therapy on periodontal status, serum melatonin and inflammatory markers in type 2 diabetes mellitus patients with chronic periodontitis: A double-blind, placebo-controlled trial. *Inflammopharmacology*. 2019;27(1):67–76. doi:10.1007/s10787-018-0539-0
21. Ahmed E, Shaker OG, Yussif N, Ghalwash DM. Effect of locally delivered melatonin as an adjunct to nonsurgical therapy on GCF antioxidant capacity and MMP-9 in stage II periodontitis patients: A randomized controlled clinical trial. *Int J Dent*. 2021;2021:8840167. doi:10.1155/2021/8840167
22. Anton DM, Martu MA, Maris M, et al. Study on the effects of melatonin on glycemic control and periodontal parameters in patients with type II diabetes mellitus and periodontal disease. *Medicina (Kaunas)*. 2021;57(2):140. doi:10.3390/medicina57020140
23. Dhande SK, Rathod SR, Kolte AP, Lathiya VN, Kasliwal PA. Clinicoradiographic comparative evaluation of 1% melatonin gel plus platelet-rich fibrin over platelet-rich fibrin alone in treatment of Grade II furcation defects: A randomized controlled double-blind clinical trial. *J Periodontol*. 2024;95(8):707–717. doi:10.1002/JPER.23-0282

Review

Analysis of the reporting characteristics of gingival recession treatment trials registered in ClinicalTrials.gov

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D – writing the article; E – critical revision of the article; F – final approval of the article

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Abstract

Gingival recessions are a common disease and one of the most frequently used method for managing them is the tunnel technique (TUN). Although more and more articles are addressing the problem, few are adequately structured, usually without following the standard protocols. Many checklists have been developed for this purpose. Among them, the Template for Intervention Description and Replication (TIDieR) checklist is commonly used. Therefore, the present review analyzed trials based on it. The completeness of registration of the trial protocol data was assessed using the TIDieR checklist with regard to the coronally advanced tunnel technique (CAT) with subepithelial connective tissue graft (sCTG) (CAT + sCTG) and CAT alone. The review also investigated the consistency of the research description in the published articles.

A total of 37 records were gained from ClinicalTrials.gov and the corresponding publications, including 32 studies employing CAT + sCTG and 5 studies using only CAT.

The description of intervention in ClinicalTrials.gov was inadequate for all analyzed trials, and there were also differences between the registered and published studies.

The present analysis showed that the description of the registered trials is often incomplete and that detailed data is not pro-vided. It is essential that clinical research is well documented and properly described, so that the findings can be easily implemented in daily clinical practice without confusion.

Keywords: checklist, reporting, database, gingival recessions

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Highlights

- Many gingival recession treatment trials lacked complete descriptions of the interventions.
- Notable inconsistencies were found between the registered and published versions of studies, particularly regarding participant numbers and the reported outcomes.
- These discrepancies contribute to the lack of consistency across research studies, undermining the reliability of findings.
- Poor documentation and missing details reduce the ability to effectively translate research findings into clinical practice.
- The analysis emphasizes the need for thorough and accurate reporting in clinical trials to enhance usability and trust in clinical settings.

Introduction

Gingival recession is described as an apical displacement of the gingival margin, which can be associated with root caries, dentin hypersensitivity and esthetic outcomes.[1] It is a very common condition that can affect people regardless of the level of oral hygiene, and its incidence increases with age.[2] This disorder can be a tough issue to treat and manage.[3]

Over the years, several surgical techniques have been developed to treat gingival recessions.[4,5] Evidence shows that the coronally advanced flap (CAF) method and the tunnel technique (TUN) lead to satisfying results.[6] Many modifications to those procedures have been introduced to improve the final outcome.[7]

The tunnel technique is known as a minimally invasive procedure to treat gingival recessions. This method eliminates vertical releasing incisions, leaving the interdental papillae intact.[8] Therefore, this treatment leads to a greater blood supply and faster healing, and reduces post-operative morbidity.[9] Over the years, TUN has gained popularity due to its high effectiveness in treating multiple gingival recessions.[10]

The short- and long-term outcomes in terms of gain of keratinized tissue depend on using subepithelial connective tissue graft (sCTG).[11,12] Although potential alternatives to harvesting a graft from the palate have been proposed, such as enamel matrix derivatives,[13] collagen porcine dermal matrix,[14] concentrated growth factors,[15] or titanium-prepared platelet-rich fibrin (T-PRF),[16] sCTG is still considered the gold standard.[17] On the other hand, research confirms that in many cases, using the coronally advanced tunnel technique (CAT) alone results in the complete coverage of the recession and is stable over time.

In order to properly apply a specific method in practice, scientific research should be adequately described in detail. Various checklists have been developed for this purpose, for example the Standard Protocol Items: Recommendations for Interventional Trials (SPIRIT) 2013 statement,[18] the Consolidated Standards of Reporting Trials (CONSORT) 2010 statement[19] and the Template for Intervention Description and Replication (TIDieR).[20]

The TIDieR checklist regarding the description of intervention[21–23] should be implemented not only in the published articles, but also in trial registries, since usually, all the information about the conducted trials is included there.

The present review is the first to analyze published articles on gingival recession treatment in terms of consistency and transparency. Moreover, it advises researchers to adhere to established guidelines when preparing scholarly articles. It is important for young scientists to write according to certain standards to maintain consistency and clarity in their articles.

So far, there has been no research on the use of specific standards while writing articles on the coverage of gingival recessions. This is an important issue, taking into account the prevalence of gingival recessions and the increasing number of studies conducted on this topic.

The aim of the present study was to assess the completeness of the description of gingival recession treatment procedures in the clinical trial protocols registered in ClinicalTrials.gov, as well as the consistency of the research description in the published articles. It can be a useful guide for inexperienced researchers to include necessary details in the conducted studies.

Material and methods

Sample and inclusion criteria

The search for clinical trials registered in ClinicalTrials.gov was conducted on October 20, 2023. It was decided to analyze the research posted in ClinicalTrials.gov, as this online database provides information about the results of clinical research studies. The term "gingival recession treatment" was used in advanced search. Then, all trials regarding gingival recession treatment were reviewed, and studies using the CAT method, with or without sCTG, were selected for analysis. Then, another screening

of studies was conducted based on the inclusion criteria: articles that referred to gingival recession treatment in one or more fields in ClinicalTrials.gov (Brief Title, Official Title, Brief Summary, or Intervention); those with a National Clinical Trial (NCT) number; described as completed, recruiting, withdrawn, active non-recruiting, or terminated; and registered in October 2023 or earlier. Trials that used other techniques to treat gingival recession than CAT were excluded.

Data extraction

In order to fully register trials, the World Health Organization (WHO) prepared a checklist – WHO Trial Registration Data Set – with 24 items. Of these, the following 21 items were taken into account in our research: the trial identifying number (ID); the NCT number, the source(s) of monetary or material support; primary and secondary trial sponsors; the public title; the scientific title, the countries where the trial was conducted; the health condition(s) studied; the intervention(s); the inclusion criteria; the study type; the date of first enrollment; the sample size; the recruitment status; the primary outcome; the key secondary outcomes; the date of study completion; summary results; and the individual participant data (IPD) sharing statement. Contact for public queries, contact for scientific queries and the ethics review were excluded.

To assess the completeness of the description for each intervention in gingival recession treatment, we used the 12-item TIDieR checklist.[20] The trials included in the analysis referred to either CAT alone or CAT + sCTG. The interventions employing other techniques than CAT were excluded from the analysis and need further investigation. The trials from ClinicalTrials.gov were verified, and key data from a random 10% sample were independently extracted by an experienced periodontologist and a general dentist (PhD student). Some unclear studies were identified and reviewed in collaboration with another reviewer, a final-year dental student working under supervision. At the end of the analysis, all authors reviewed the included studies and resolved any disagreement through discussion.

All the information was obtained from the Descriptive Information section in ClinicalTrials.gov. None of the papers provided data related to points 9, 10 and 12 of the checklist, so we referred to 9 items only in the analysis. The interventions were checked for data consistency in the PubMed® and Google Scholar databases. The information from ClinicalTrials.gov was compared with the corresponding publications. After checking 157 interventions, we excluded 119 of them, since they used other type of technique to treat gingival recessions. Most data regarded CAF coronally advanced flap,[6] as it leads to excellent results. Fifteen studies did not mention the method used, and 3 papers did not provide the name of the method, but the description indicated the use of CAF.

Results

The study included 37 trials retrieved from ClinicalTrials.gov. on October 20, 2023, as shown in Fig. 1. As many as 32 (86%) reported using CAT + sCTG, and 5 (14%) used CAT alone.

The registration numbers of the trials qualified for analysis, using CAT with sCTG: NCT05688293; NCT04291963; NCT02632240; NCT05270161; NCT03791554; NCT02642887; NCT03657706; NCT05122468; NCT04133298; NCT06000228; NCT03163654; NCT04028037; NCT04104087; NCT05819515; NCT05568732; NCT04561947; NCT05045586; NCT03354104; NCT03162016; NCT06044870; NCT04966208; NCT05823415; NCT03676088; NCT06030947; NCT02916186; NCT02814279; NCT04016493; NCT04198376; NCT05976451; NCT02774967; NCT03690635; and NCT05436002.

The registration numbers of the trials qualified for analysis, using CAT without sCTG: NCT04513041; NCT04225351; NCT0565247; NCT03619096; and NCT04802473

At first, the general research characteristics were analyzed for both group of trials (Table 1). We collected data about the status – unknown, recruiting, active not recruiting, or completed. Most of the papers (n = 34; 92%) did not provide any data on the study phase, 3 trials (8%) declared phase 2 (n = 1; 3%) or 4 (n = 2; 5%).

Almost all trials (n = 36; 97%) were described as interventional, and only one paper (3%) as observational. The majority of articles (n = 34; 92%) applied randomized allocation, 1 trial used non-randomized allocation, and 2 studies (5%) stated "not applicable". The most frequently used interventional model was parallel assignment (n = 33; 89%). Trials also applied a single-group model (n = 3; 8%), crossover assignment (n = 1; 3%) and 1 paper (3%) used the term "other", but did not mention the type.

The included trials applied different types of masking: none (open-label) (n = 5; 14%); single-blind (n = 12; 32%); double-blind (n = 10; 27%); triple-blind (n = 9; 24%); and quadruple-blind (NCT02916186; n = 1; 3%). The majority

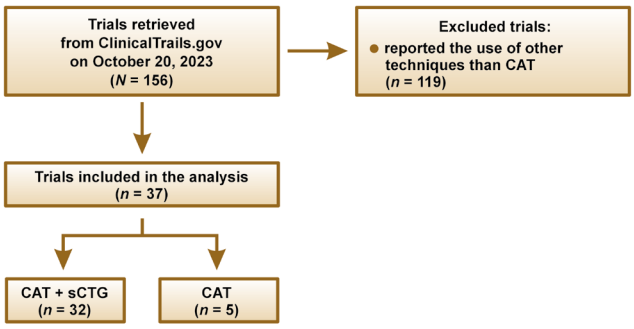

Fig. 1. Flowchart of the study

CAT – coronally advanced tunnel technique; sCTG – subepithelial connective tissue graft.

Table 1. Characteristics of 37 trials on gingival recession treatment using the coronally advanced tunnel technique (CAT), registered in ClinicalTrials.gov

Research characteristics		Trials *n* (%)	NCT number
Status	unknown	10 (27.0)	–
	recruiting	5 (13.5)	–
	active non-recruiting	7 (18.9)	–
	completed	15 (40.5)	–
Phase	phase 2	1 (2.7)	NCT06044870
	phase 4	2 (5.4)	NCT03690635 NCT04104087
	not applicable	34 (91.9)	–
Study type	interventional	36 (97.3)	–
	observational	1 (2.7)	NCT04016493
Allocation	randomized	34 (91.9)	–
	non-randomized	1 (2.7)	NCT06000228
	not applicable	2 (5.4)	NCT04513041 NCT04802473
Interventional model	single-group	3 (8.1)	–
	crossover assignment	1 (2.7)	–
	parallel assignment	33 (89.2)	–
Masking	none (open label)	5 (13.5)	–
	single-blind	12 (32.4)	–
	double-blind	10 (27.0)	–
	triple-blind	9 (24.3)	–
	quadruple-blind	1 (2.7)	NCT02916186
Primary purpose	treatment	36 (97.3)	–
	not provided	1 (2.7)	–

NCT – National Clinical Trial.

of trials mentioned treatment as the primary purpose (n = 36; 97%); only 1 study did not provide the aim of the study (Table 1).

The next step was the comparison of the participant characteristics (Table 2). All studies included volunteers of both genders, provided their age and enrollment. Most of the trials accepted healthy participants (n = 25; 68%). Fifteen papers (41%) did not apply the IPD sharing, 10 studies (27%) stated "undecided" and 12 trials (32%) claimed "not provided".

Quality of description of the main interventions in ClinicalTrials.gov

All the 32 trials using CAT + sCTG briefly described the intervention, and provided information about materials, procedures and the location where the intervention was applied (Table 3).

The majority of these studies (n = 30; 94%) mentioned the goal of treatment, which was determining the effect of root coverage. A total of 19 included trials (59%) reported a periodontist as a person responsible for treatment, 3 studies (8%) provided unclear information and 10 papers (31%) did not mention the investigator.

Table 2. Characteristics of participants in 37 trials on gingival recession treatment using the coronally advanced tunnel technique (CAT), registered in ClinicalTrials.gov

Participants' characteristics		Trials *n* (%)
Gender	male	0 (0.0)
	female	0 (0.0)
	both	37 (100.0)
Minimum age [years]	provided *Me*: 18 range: 18–45	33 (89.2)
	not provided	4 (10.8)
Maximum age [years]	provided *Me*: 60 range: 40–75	24 (64.9)
	not provided	13 (35.1)
Enrollment	provided *Me*: 30 range: 6–68	37 (100.0)
Conditions	provided	37 (100.0)
Healthy volunteers accepted	yes	25 (67.6)
	no	12 (32.4)
IPD sharing statement	yes	0 (0.0)
	no	15 (40.5)
	undecided	10 (27.0)
	not provided	12 (32.4)

Me – median; IPD – individual participant data.

Only 6 studies (19%) provided information with regard to TIDieR item 6, specifying that the participants received verbal or written instructions. All the trials provided the time frame of the study, and 24 papers (75%) also mentioned the number of sessions.

In our article we included 5 studies using CAT to treat gingival recessions (Table 4). All papers provided a short description, and information about materials and procedures. Most of the studies (n = 4; 80%) mentioned the aim of the study. The expertise provider as well as the investigation center were mentioned in 100% trials. Similar to the intervention reporting in the trials using CAT with sCTG, 20% papers described indications for volunteers.

For only one trial out of 32 (3%; NCT03676088), we identified accordance with the CONSORT criteria.

Comparison of the registry data and the corresponding publications

For the 37 registered papers analyzed in the study, we were able to identify 10 corresponding publications, searching PubMed and Google Scholar. The number of participants, and the primary and secondary outcomes were compared (Fig. 2). In 6 cases, the number of volunteers in the published article was different from that in the registry.

The primary outcomes were the same in ClinicalTrials.gov. and in the published articles. In the case of 1 trial

Table 3. Quality of reporting 12 items from the Template for Intervention Description and Replication (TIDieR) checklist for the coronally advanced tunnel technique (CAT) with subepithelial connective tissue graft (sCTG)

TIDieR item	Intervention description component	Trials *n* (%)
1. Brief name	provided	32 (100)
	not provided	0 (0)
2. Why	noted precisely	30 (94)
	noted, but insufficiently described	1 (3)
	not provided	1 (3)
3. What (material)	provided	32 (100)
	not provided	0 (0)
4. What (procedure)	provided	32 (100)
	not provided	0 (0)
5. Who provided expertise	periodontist	19 (59)
	unclear data provided	3 (9)
	not provided	10 (31)
6. How	provided*	6 (19)
	not provided	26 (81)
7.Where	recruitment/investigator institution noted	32 (100)
	not provided	0 (0)
8. When and how much	provided	24 (75)
	not provided	8 (25)
11. How well (planned)	provided**	1 (3)
	not provided	31 (97)

N = 32; * NCT02814279, NCT03676088, NCT04016493, NCT04028037, NCT05568732, NCT06000228; ** NCT03676088 (Consolidated Standards of Reporting Trials (CONSORT)).

Table 4. Quality of reporting 12 items from the Template for Intervention Description and Replication (TIDieR) checklist for the coronally advanced tunnel technique (CAT)

TIDieR item	Intervention description component	Trials *n* (%)
1. Brief name	provided	5 (100)
	not provided	0 (0)
2. Why	noted precisely	4 (80)
	noted, but insufficiently described	0 (0)
	not provided	1 (20)
3. What (material)	provided	5 (100)
	not provided	0 (0)
4. What (procedure)	provided	5 (100)
	not provided	0 (0)
5. Who provided expertise	periodontist	5 (100)
	unclear data provided	0 (0)
	not provided	0 (0)
6. How	provided*	1 (20)
	not provided	4 (80)
7.Where	recruitment/investigator institution noted	5 (100)
	not provided	0 (0)
8. When and how much	provided	5 (100)
	not provided	0 (0)
11. How well (planned)	provided	0 (0)
	not provided	5 (100)

N = 5; * NCT04802473.

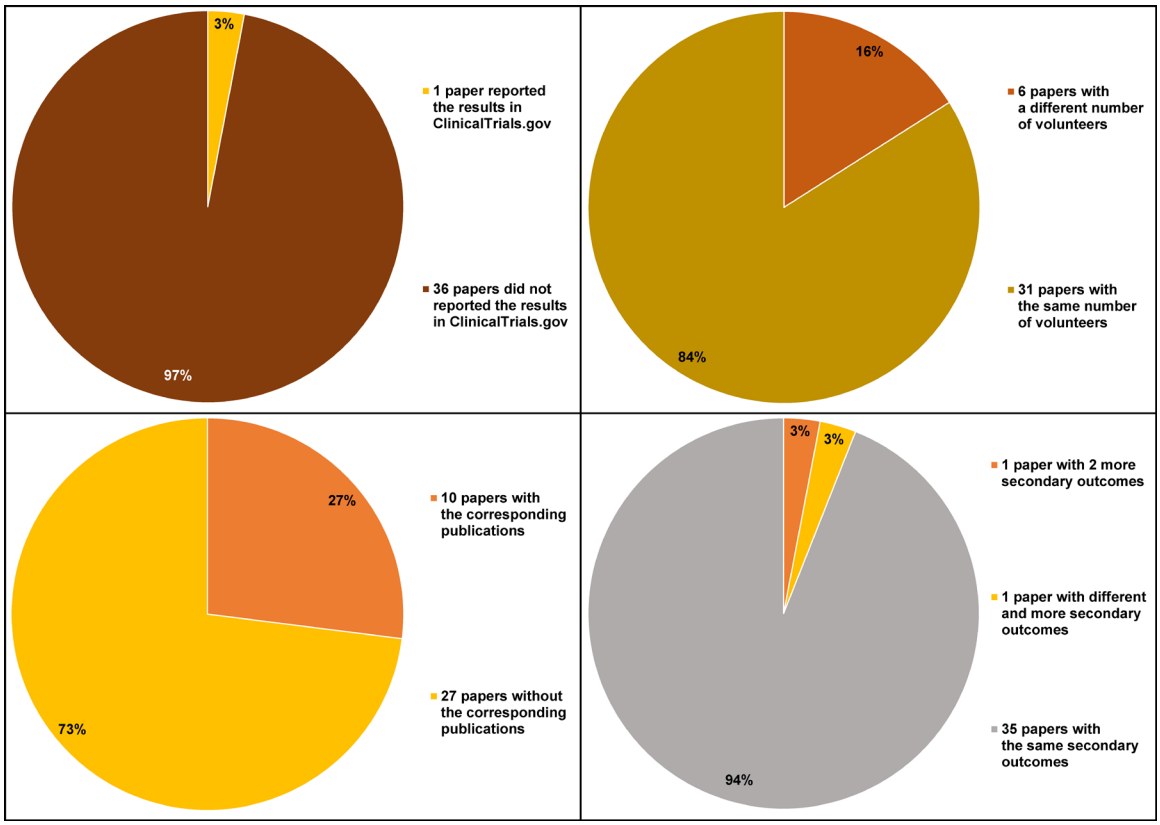

Fig. 2. Comparison of the registry data and the corresponding publications

(3%) (NCT02916186), there were 2 more secondary outcomes provided in the released paper, i.e., patient esthetic satisfaction and postoperative pain on a visual analog scale (VAS). Another published article (NCT03163654) contained different and more secondary outcomes in comparison with the registry data, which was the root coverage esthetic score, keratinized tissue width and gingival thickness gain, whereas in the corresponding trials, gingival recession width, keratinized tissue width, gingival thickness, and clinical attachment level (CAL) were provided.

Only one included study (NCT02814279) reported the results in ClinicalTrials.gov.

Discussion

The tunnel technique is recognized by periodontal specialists as one of the easiest and at the same time effective methods of covering gingival recessions, having many advantages in terms of dentin hypersensitivity reduction and esthetics, producing a significant gain in CAL and decreasing gingival recession.[1] However, with regard to clinical trials, there is a need to establish the most predictable surgical approach for the treatment of this condition. The present study shows that the description of the registered trials is often incomplete and that detailed data is not provided. It is concluded, after comparing the registered papers with the published ones that there exist substantial differences between the provided information and instructions. Discrepancies concern mainly the number of participants and the reported outcomes.

A key limitation of our analysis is that over 60% of the trials posted in ClinicalTrials.gov were not linked to the corresponding publications, and only one study reported the results in ClinicalTrials.gov. Study data should be shared via ClinicalTrials.gov, as usually this is the only source of information on ongoing research. Moreover, the information should be transparent, detailed and easily available to the public; only then studies have a scientific value. For this reason, it is recommended to post the obtained results in ClinicalTrials.gov.[24]

Considering the high prevalence of gingival recessions, it is essential that clinical trials are well documented and properly described, so that the findings can be easily implemented in the treatment. Moreover, nowadays significantly more clinical trials are being carried out in periodontology over the years, wherefore all those studies should be adequately reported.[25] In order to properly apply a specific method in practice, scientific research should be properly described in detail. Scientists should use the TIDieR guidelines to adequately report the studies.

Gingival recession treatment became predictable, with satisfactory results. Therefore, in our opinion, the ongoing research should be carried out according to specific guidelines, e.g., SPIRIT, CONSORT or TIDieR.[18–20] To improve the quality of interventions and to allow adequate translation into clinical practice, clinical researchers and also journal editors should concentrate on precisely described papers.

The analysis focused on one of the most frequently used methods, i.e., TUN.[6] To provide complete data on the ongoing trials regarding gingival recession treatment, we plan to analyze other methods, for example CAF.

Conclusions

Our review showed that the descriptions of intervention in gingival recession treatment trials were incomplete, and there were also differences between the registered and published studies. The main discrepancies concerned the number of participants, and the primary and secondary outcomes. This leads to the lack of consistency in research, and can also reduce the value of translation of the procedures into clinical practice.

Ethics approval and consent to participate

Not applicable.

Data availability

The datasets supporting the findings of the current study are available from the corresponding author on reasonable request.

Consent for publication

Not applicable.

Use of AI and AI-assisted technologies

Not applicable.

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References

1. Górski B, Górska R, Wysokińska-Miszczuk J, Kaczyński T. Tunnel technique with enamel matrix derivative in addition to subepithelial connective tissue graft compared with connective tissue graft alone for the treatment of multiple gingival recessions: A randomized clinical trial. *Clin Oral Investig*. 2020;24(12):4475–4486. doi:10.1007/s00784-020-03312-6
2. Rios FS, Costa RS, Moura MS, Jardim JJ, Maltz M, Haas AN. Estimates and multivariable risk assessment of gingival recession in the population of adults from Porto Alegre, Brazil. *J Clin Periodontol*. 2014;41(11):1098–1107. doi:10.1111/jcpe.12303
3. Jati AS, Furquim LZ, Consolaro A. Gingival recession: Its causes and types, and the importance of orthodontic treatment. *Dental Press J Orthod*. 2016;21(3):18–29. doi:10.1590/2177-6709.21.3.018-029.oin

4. Keceli HG, Sengun D, Berberoğlu A, Karabulut E. Use of platelet gel with connective tissue grafts for root coverage: A randomized-controlled trial. *J Clin Periodontol*. 2008;35(3):255–262. doi:10.1111/j.1600-051X.2007.01181.x
5. Cairo F, Nieri M, Pagliaro U. Efficacy of periodontal plastic surgery procedures in the treatment of localized facial gingival recessions. A systematic review. *J Clin Periodontol*. 2014;41(Suppl 15):S44–S62. doi:10.1111/jcpe.12182
6. Cortellini P, Prato GP. Coronally advanced flap and combination therapy for root coverage. Clinical strategies based on scientific evidence and clinical experience. *Periodontol 2000*. 2012;59(1):158–184. doi:10.1111/j.1600-0757.2011.00434.x
7. Zuhr O, Fickl S, Wachtel H, Bolz W, Hürzeler MB. Covering of gingival recessions with a modified microsurgical tunnel technique: Case report. *Int J Periodontics Restorative Dent*. 2007;27(5):457–463. PMID:17990442.
8. Fahmy R, Taalab MR. Modified tunnel technique for management of gingival recession in esthetic zone using acellular dermal matrix versus connective tissue graft. *Future Dent J*. 2018. doi:10.1016/J.FDJ.2018.12.001
9. Santamaria MP, da Silva Neves FL, Silveira CA, et al. Connective tissue graft and tunnel or trapezoidal flap for the treatment of single maxillary gingival recessions: A randomized clinical trial. *J Clin Periodontol*. 2017;44(5):540–547. doi:10.1111/jcpe.12714
10. Tavelli L, Barootchi S, Nguyen TV, Tattan M, Ravidà A, Wang HL. Efficacy of tunnel technique in the treatment of localized and multiple gingival recessions: A systematic review and meta-analysis. *J Periodontol*. 2018;89(9):1075–1090. doi:10.1002/JPER.18-0066
11. Chambrone L, Tatakis DN. Periodontal soft tissue root coverage procedures: A systematic review from the AAP Regeneration Workshop. *J Periodontol*. 2015;86(Suppl 2):S8–S51. doi:10.1902/jop.2015.130674
12. Evginer MS, Olgun E, Parlak HM, Dolgun AB, Keceli HG. Comparison of two techniques in gingival recession treatment: A randomized one-year clinical follow-up study. *Dent Med Probl*. 2022;59(1):121–130. doi:10.17219/dmp/137621
13. Sculean A, Cosgarea R, Stähli A, et al. The modified coronally advanced tunnel combined with an enamel matrix derivative and subepithelial connective tissue graft for the treatment of isolated mandibular Miller Class I and II gingival recessions: A report of 16 cases. *Quintessence Int*. 2014;45(10):829–835. doi:10.3290/j.qi.a32636
14. Cieślik-Wegemund M, Wierucka-Młynarczyk B, Tanasiewicz M, Gilowski Ł. Tunnel technique with collagen matrix compared with connective tissue graft for treatment of periodontal recession: A randomized clinical trial. *J Periodontol*. 2016;87(12):1436–1443. doi:10.1902/jop.2016.150676
15. Korkmaz B, Balli U. Clinical evaluation of the treatment of multiple gingival recessions with connective tissue graft or concentrated growth factor using tunnel technique: A randomized controlled clinical trial. *Clin Oral Investig*. 2021;25(11):6347–6356. doi:10.1007/s00784-021-03935-3
16. Bhattacharya HS, Gummaluri SS, Rani A, Verma S, Bhattacharya P, Rayashettypura Gurushanth SM. Additional benefits of titanium platelet-rich fibrin (T-PRF) with a coronally advanced flap (CAF) for recession coverage: A case series. *Dent Med Probl*. 2023;60(2):279–285. doi:10.17219/dmp/141919
17. Bakhishov H, Isler SC, Bozyel B, Yıldırım B, Tekindal MA, Ozdemir B. De-epithelialized gingival graft versus subepithelial connective tissue graft in the treatment of multiple adjacent gingival recessions using the tunnel technique: 1-year results of a randomized clinical trial. *J Clin Periodontol*. 2021;48(7):970–983. doi:10.1111/jcpe.13452
18. Chan AW, Tetzlaff JM, Gøtzsche PC, et al. SPIRIT 2013 explanation and elaboration: Guidance for protocols of clinical trials. *BMJ*. 2013;346:e7586. doi:10.1136/bmj.e7586
19. Schulz KF, Altman DG, Moher D; CONSORT Group. CONSORT 2010 statement: Updated guidelines for reporting parallel group randomised trials. *BMJ*. 2010;340:c332. doi:10.1136/bmj.c332
20. Hoffmann TC, Glasziou PP, Boutron I, et al. Better reporting of interventions: Template for intervention description and replication (TIDieR) checklist and guide. *BMJ*. 2014;348:g1687. doi:10.1136/bmj.g1687
21. Anderson JM, Stafford A, Jellison S, Vassar M. Intervention reporting of published trials is insufficient in orthopaedic surgery journals: Application of the Template for Intervention Description and Replication checklist. *Arthrosc Sports Med Rehabil*. 2021;3(3):e619–e627. doi:10.1016/j.asmr.2020.09.019
22. Palmer W, Okonya O, Jellison S, et al. Intervention reporting of clinical trials published in high-impact cardiology journals: Effect of the TIDieR checklist and guide. *BMJ Evid Based Med*. 2021;26(3):91–97. doi:10.1136/bmjebm-2019-111309
23. Torgerson T, Johnson AL, Jellison S, et al. Reporting of clinical trial interventions published in leading otolaryngology-head and neck surgery journals. *Laryngoscope*. 2020;130(9):E507–E514. doi:10.1002/lary.28404
24. Zarin DA, Tse T, Williams RJ, Califf RM, Ide NC. The ClinicalTrials.gov results database – update and key issues. *N Engl J Med*. 2011;364(9):852–860. doi:10.1056/NEJMsa1012065
25. Niederman R, Chen L, Murzyn L, Conway S. Benchmarking the dental randomised controlled literature on MEDLINE. *Evid Based Dent*. 2022;3:5–9. doi:10.1038/sj.ebd.6400095

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