Duration and dose of chemotherapy and dental development

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Abstract

Background. Given the susceptibility of developing tissues to drugs, even small doses of anticancer drugs may affect odontogenesis. Although any toxic effect is transient, the treatment regimens are based on repeated drug administration.

Objectives. The study aimed to establish the impact of antineoplastic therapy on the occurrence of long-term adverse dental effects in a dose-dependent manner in young survivors treated for cancer before 10 years of age.

Material and methods. In total, 37 cancer survivors treated with antineoplastic therapy before 10 years of age underwent a dental examination with a thorough analysis of panoramic radiographs. A total of 236 teeth with 243 different developmental abnormalities were revealed in 28 survivors. Agenesis, tooth size reduction, taurodontia, and enamel and root abnormalities were diagnosed. All survivors received multi-agent chemotherapy, with the most frequently used drugs being vincristine (VCR), doxorubicin (DXR), cyclophosphamide (CP), ifosfamide (IF), etoposide (VP-16), carboplatin (CBDCA), cisplatin (CDDP), and actinomycin-D (ActD). A detailed analysis of medical records was also performed to assess the relationship between the treatment duration as well as the cumulative drug dose administered and the occurrence of particular disturbances.

Results. When analyzing the treatment duration and the drug doses in the affected and non-affected participants, there were no statistically significant differences between the survivors with different disturbances within most of the specific drug groups. In some groups, the mean cumulative treatment dose was significantly higher in the non-affected patients. According to Spearman's rho, no significant relationships were observed.

Conclusions. In the present study, no significant differences in terms of treatment duration or drug doses were observed between the patients with particular abnormalities. The developmental stage of tooth formation during chemotherapy is likely the most important factor influencing dental changes. For future research with respect to different treatment protocols, an analysis of a more homogenous group of survivors is warranted.

Keywords: tooth abnormalities, dental development, chemotherapy

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Introduction

Anticancer treatment in children is typically very effective. However, the treatment can result in acute and late adverse effects, including effects on developing dental structures. Various standard treatment models have been established for the management of malignant diseases. Some antineoplastic drugs, such as vincristine (VCR), etoposide (VP-16) and cyclophosphamide (CP), are used relatively often in the treatment regimens for different cancers. Some of these drugs are dedicated to specific cancer types. However, they all have a low therapeutic index, and, through different mechanisms of action, ultimately result in arrested cell division or apoptosis. Vincristine and alternative microtubule-binding vinca alkaloids still remain the first choice of cytostatic agent for the therapy of solid tumors and blood cancers. They cause the cessation of cellular division in the M phase, followed by cell apoptosis.[1] Doxorubicin (DXR) stabilizes the complexes of double-stranded DNA and topoisomerase IIα, which helps the enzyme cut both DNA strands.[2] Cyclophosphamide and similar alkylating drugs, such as ifosfamide (IF), cross-link guanine bases in DNA.[3] Actinomycin D (ActD), like other antibiotics, self-inserts into DNA and causes damage to its structure, resulting in the inhibition of RNA polymerase and protein synthesis.[4] Cisplatin (CDDP) and carboplatin (CBDCA) represent a group of platinum-based cytotoxic drugs widely administered as part of therapy for various solid tumors. Their initial interaction with purine bases in DNA eventually results in cellular death.[5] Etoposide (VP-16) destroys the double-stranded structure of DNA by inhibiting type II topoisomerase and is used in the treatment protocols for many different cancers.[6]

Variability in treatment options and, for the same reason, possible differences in cytotoxic impact may be the deciding factors for the occurrence of long-term effects. However, the multi-drug nature of anticancer therapy makes it difficult to estimate the possible toxic impact of particular medications on odontogenesis. Considering the similarity of all developing tissues, we can surmise that the rapidly dividing dental cells are highly susceptible to toxic damage in the same manner as it is observed in cancer. Therefore, even a small dose of a drug may cause irreversible changes in odontogenesis. However, since the half-life of chemotherapeutic agents is short, their toxic effects could be transient, also in the case of fully developed cells.[7]

As far as developing dental tissues are concerned, chemotherapy-induced changes may be observed in either the rapidly dividing immature cells or fully developed hard tissue-forming cells. This has been well documented in vitro and in animal studies.[1,3] It has been reported that vinblastine (VBL) causes the dose-dependent inhibition of dentinogenesis in rat incisors.[8] Cyclophosphamide has been shown to inhibit cell division in the immature portion of the rat tooth germ, and to cause mutations in more developed dentin and enamel precursor cells when administered at low or high doses, respectively.[3] The treatment dose is likely to be the main factor influencing dental abnormalities. Another experimental study tested the effects of VCR on rat incisors.[9] The authors reported that after toxic drug administration, the observed mitotic cessation of young cell division, as well as changes in the secretory activity of mature odontoblasts were transient. The disturbances of odontogenesis occurred locally and were followed by the correct matrix secretion.[9] However, intensive chemotherapeutic regimens are based on repeated drug administration at standard treatment intervals. Therefore, a developing tooth germ is exposed to the regular toxic impact of multi-drug therapy. Repeated treatment may be another factor responsible for the dental sequelae of chemotherapy.

In the literature, dental disturbances induced by antineoplastic treatment are reported in approx. 70% of cancer survivors (62.29–71%).[10–13] The peak incidence of affected teeth has been reported in survivors who received chemotherapy at a young age.[3,7,11,14–19] Nevertheless, attempts to estimate the effects of individual drugs or their doses on human dental development are on-going.[12,20–22]

The present study aimed to establish the impact of antineoplastic therapy together with any dose-dependent relationship on the occurrence of long-term adverse dental effects in survivors treated for cancer before 10 years of age.

Material and methods

The study was approved by the Bioethics Committee at the Medical University of Silesia, Katowice, Poland, on February 25, 2013 and November 29, 2016 (KNW/0022/KB1/15/I/13 and KNW/0022/KB1/15/II/16, respectively). Dental examinations were carried out in cancer survivors who fulfilled the following inclusion criteria: anticancer therapy started before 10 years of age and was completed at least 2 years before the dental examination. The caregivers of 37 individuals aged 6–17 years provided informed written consent for dental examinations, including panoramic radiographs, and the processing of medical data concerning the details of anticancer treatment. In the examined survivors, a cancer diagnosis was established at 4 months of age at minimum, and at 8 years and 6 months at maximum. Twenty-eight patients were diagnosed with the following solid tumors: nephroblastoma, neuroblastoma, anaplastic ependymoma, medulloblastoma, sarcoma granulocyticum, teratoma malignum, embryonal rhabdomyosarcoma, primitive neuroectodermal tumor (PNET)/Ewing sarcoma (ES), yolk sac tumor, clear cell sarcoma, astrocytoma pilocyticum, hepatoblastoma, and infantile fibrosarcoma. The remaining 9 participants suffered from different types of hematological neoplasms, predominantly leukemia. All of the survivors received

multi-agent chemotherapy, with the most frequently used drugs being VCR, DXR, CP, IF, VP-16, CBDCA, CDDP, ActD, daunorubicin (DNR), dacarbazine (DTIC), cytarabine (ARA-C), methotrexate (MTX), and mercaptopurine (6-MP). Surgery was required in 27 patients and additional radiotherapy was performed in 11 participants (cranial radiotherapy in 4 cases). After a detailed analysis, the effects of radiotherapy, including cranial radiation, were considered to be negligible for the purpose of this study. Dental examinations supplemented with a thorough analysis of panoramic radiographs revealed a total of 236 teeth with 243 different developmental abnormalities. Special attention was paid to agenesis, microdontia, tooth size reduction, taurodontia, and enamel and root abnormalities. Enamel abnormalities, such as opacities, hypoplasia and marked perikymata, were found in the majority of teeth. Agenesis was the rarest abnormality in the study cohort. Table 1 shows the results of dental examinations, including the prevalence of particular developmental changes, in total and for each analyzed drug. The patients in whom all teeth showed a reduced size (microdontia and reduction) were additionally analyzed as a subgroup (M+R) due to the same mechanism of origin and the expected longer treatment duration. Some survivors presented with other dental abnormalities, such as root changes or supernumerary teeth. However, the small number of affected patients made statistical analysis impossible and these abnormalities were not further considered in this study.

A detailed analysis of the patients' medical records was also performed. The following treatment protocols were implemented in the study group: CWS 2006 Non-RMS-like HRG; CWS 2006 RMS-like; CWS 2002 (HR); SIOP 2001; 2002 PPGL; PPGGL SIOP December 2001; Euro-Ewing 99 PPGGL; Protocol I, III, IV PPGGL; Protocol II, III, IV PLGM recommended by PPGGL; TGM 95 PPGGL (HR); SIOPEL 3 (SR); ALL IC-BFM 2002 (SR, IR); ALL-REZ BFM 2002; and BFM Interim 2004 HRG. Among these protocols, VCR administration was the most common, followed by alkylating agent-containing anticancer therapy (CP, IF), anthracycline antibiotic-containing regimens (DXR, DNR), platinum-based chemotherapy (CBDCA, CDDP), and ActD. The characteristics of the study population and important treatment details are summarized in Table 1.

The next step was to analyze the mean treatment duration and the average cumulative dose for the most frequently used anticancer drugs, which were calculated separately for different abnormalities in relation to patients with or without the analyzed abnormality, and in relation to patients without dental abnormalities. The relationship between the treatment duration and the occurrence of particular disturbances for each drug was also assessed. The same procedure was applied for the cumulative drug doses administered during the first 10 weeks of treatment and for the entire duration of the therapy using the analyzed drug (Table 2).

Statistical analysis

The patient characteristics were analyzed using the non-parametric Mann–Whitney *U* test. Continuous variables were reported as the mean (*M*), standard deviation (*SD*), minimum (min), and maximum (max) values. The statistical analysis of the treatment period data involved the correlation between drug administration in the affected and/or non-affected survivors and the prevalence of long-term dental effects, and the impact of age at the onset of therapy and the treatment duration on the occurrence of abnormalities. Radiographically and clinically recorded dental outcomes, such as agenesis, microdontia, reduction in tooth size, the M+R status, enamel changes, and taurodontia, were assessed in this analysis. The values were reported in weeks and milligrams. To assess differences and the strength of the relationships between the chemotherapy-related variables analyzed in Table 2, the Mann–Whitney *U* test and Spearman's rho were used, respectively (Table 2 and Table 3). A p-value ≤0.05 was considered to be statistically significant. All statistical analyses were performed using the Statistica® software, v. 13.3 (StatSoft Polska, Kraków, Poland).

Results

The results are shown in Tables 1–3. All patients (100%) treated with CDDP were diagnosed with dental developmental changes. However, this rate was not significantly higher than that for other anticancer drugs. The prevalence of the affected patients treated with various medications analyzed separately was comparable to the prevalence noted in all treated participants (Table 1). The age at diagnosis for each drug or group of medications in the total population and in the affected cancer survivors ranged between 4 and 102 months, whereas the non-affected cancer survivors started their treatment at a minimum of 25 months and a maximum of 69 months (Table 1).

With regard to the administration of VCR, antibiotics and VP-16, treatment with alkylating and platinum-based agents was paradoxically longer in the non-affected patients than the affected ones (Table 2). In order to establish the treatment duration-dependency of different long-term dental effects, the mean duration of anticancer therapy was determined for 6 groups of survivors diagnosed with agenesis, microdontia, tooth size reduction, microdontia and/or tooth size reduction, enamel defects, and taurodontia. Due to the combined drug administration in some anticancer protocols, a few drug analogs were excluded from a detailed analysis. No significant differences in the treatment duration were noted between abnormalities within the affected and non-affected groups for each analyzed medication, except for patients with a reduction in tooth size and enamel changes in the CP affected group ($p = 0.05$) (Table 2). Furthermore, there were no significant differences in the treatment duration

Table 1. Study group characteristics

Drug/ group of drugs (prevalence of patients treated [%])	Number of patients			Mean age at the treatment onset [months] min–max			Mean duration of treatment with drug analyzed [weeks] *M* ±*SD* min–max			Mean duration of treatment with all medications [weeks] *M* ±*SD* min–max			Number of teeth affected						
	total *n* (%)	affected *n* (%) n_A/1	non-affected *n* (%)	total	affected	non-affected	total	affected	non-affected	total	affected	non-affected	total *n* (n_A)	agenesis n_A (%) *p*-value	microdontia n_A (%) *p*-value	reduction in tooth size n_A (%) *p*-value	enamel changes n_A (%) *p*-value	taurodontia n_A (%) *p*-value	root changes n_A (%) *p*-value
Total (100.00)	37 (100.00)	28 (75.68) 8.7/1	9 (24.32)	38.08 4–102	35.07 4–102	47.44 25–69	–	–	–	60.49 ±32.65 5–122	60.75 ±31.23 11–122	59.67 ±38.81 5–118	236 (243)	20 (8.23)	30 (12.35)	59 (24.28)	80 (32.92)	27 (11.11)	27 (11.11)
VCR (81.08)	30 (100.00)	22 (73.33) 9.3/1	8 (26.67)	36.40 4–91	32.45 4–91	47.25 25–69	38.10 ±20.06 5–83	38.50 ±23.09 6–83	37.00 ±20.38 5–63	64.77 ±32.80 5–122	66.27 ±30.10 11–122	60.63 ±41.38 5–118	198 (205)	18 (8.78)	19 (9.27)	53 (25.85)	67 (32.68)	21 (10.24)	27 (13.17)
DXR (35.14)	13 (100.00)	11 (84.62) 8.4/1	2 (15.38)	29.85 4–72	27.73 4–72	41.50 37–46	24.85 ±12.82 1–47	27.55 ±13.19 7–47	10.00 ±12.73 1–19	50.69 ±22.09 5–88	54.36 ±18.95 30–88	30.50 ±36.06 5–56	88 (92)	16 (17.39) 0.0300*	18 (19.57)	13 (14.13)	30 (32.61)	14 (15.22)	1 (1.09) 0.0060*
DXR/DNR (54.05)	20 (100.00)	16 (80.00) 8.0/1	4 (20.00)	34.70 4–91	32.25 4–91	44.50 37–48	26.20 ±11.74 1–47	27.31 ±10.85 7–47	21.75 ±15.86 1–38	69.65 ±34.17 5–122	69.38 ±30.88 30–122	70.75 ±51.25 5–118	124 (128)	20 (15.63) 0.0400*	19 (14.84)	32 (25.00)	30 (23.44)	16 (12.50)	11 (8.59)
CP (35.14)	13 (100.00)	11 (84.62) 9.8/1	2 (15.38)	34.85 11–91	32.55 11–91	47.50 47–48	28.46 ±6.02 20–40	28.91 ±6.43 20–40	26.00 ±1.00 25–27	81.08 ±32.46 39–122	75.64 ±32.29 39–122	111.00 ±9.90 104–118	103 (108)	8 (7.41)	21 (19.44)	28 (25.93)	32 (29.63)	9 (8.33)	10 (9.26)
IF (21.62)	8 (100.00)	7 (87.50) 8.9/1	1 (12.50)	38.00 4–88	38.14 4–88	37.00 37–37	23.88 ±14.01 4–49	20.29 ±10.44 4–32	49.00 ±0.00 49–49	59.38 ±25.84 17–88	59.86 ±27.87 17–88	56.00 ±0.00 56–56	60 (62)	9 (14.52)	6 (9.68)	12 (19.35)	11 (17.74) 0.0200*	7 (11.29)	17 (27.42) 0.0020*
CP/IF (56.76)	21 (100.00)	18 (85.71) 9.4/1	3 (14.29)	36.05 4–91	34.72 4–91	44.00 37–48	26.71 ±9.87 4–49	25.56 ±9.16 4–40	33.67 ±13.32 25–49	72.81 ±31.34 17–122	69.50 ±30.83 17–122	92.67 ±32.52 56–118	162 (170)	17 (10.00)	27 (15.88)	40 (23.53)	43 (25.29)	16 (9.41)	27 (15.88)
VP-16 (37.84)	14 (100.00)	12 (85.71) 8.6/1	2 (14.29)	30.31 4–102	34.42 4–102	41.50 37–46	21.71 ±12.00 1–39	23.33 ±11.88 1–39	12.00 ±9.90 5–19	52.36 ±24.54 5–82	56.00 ±22.18 17–82	30.50 ±36.06 5–56	100 (103)	8 (7.77)	23 (22.33) 0.0200*	17 (16.50)	29 (28.16)	10 (9.71)	16 (15.53)
CBDCA (35.14)	13 (100.00)	10 (76.92) 5.3/1	3 (23.08)	32.62 4–69	24.90 4–55	58.33 46–69	37.38 ±20.82 1–87	35.50 ±23.47 1–87	43.67 ±6.81 36–49	55.08 ±18.53 30–92	52.50 ±17.08 30–83	63.67 ±24.54 49–92	53 (53)	8 (15.09)	16 (30.19) 0.0020*	16 (30.19)	3 (5.66) 0.0001*	10 (18.87)	0 (0.00) 0.0200*
CDDP (18.92)	7 (100.00)	7 (100.00) 10.9/1	0 (0.00)	45 11–95	45 11–95	–	34.29 ±19.32 14–69	34.29 ±19.32 14–69	–	49.00 ±30.51 14–80	49.00 ±30.51 14–80	–	71 (76)	0 (0.00) 0.0200*	12 (15.79)	11 (14.47)	29 (38.16)	8 (10.53)	16 (21.05) 0.0200*
CBDCA/ CDDP (54.05)	20 (100.00)	17 (85.00) 7.6/1	3 (15.00)	36.95 4–95	33.18 4–95	58.33 46–69	36.30 ±19.85 1–87	35.00 ±21.22 1–87	43.67 ±6.81 36–49	52.95 ±22.79 14–92	51.06 ±22.72 14–83	63.67 ±24.54 49–92	124 (129)	8 (6.20)	28 (21.71) 0.0100*	27 (20.93)	32 (24.81)	18 (13.95)	16 (12.40)
ActD (32.43)	12 (100.00)	10 (83.33) 10.5/1	2 (16.67)	27.33 9–55	26.6 9–55	31 25–37	18.25 ±13.73 3–43	18.50 ±14.68 3–43	17.00 ±11.31 9–25	43.08 ±21.17 11–88	45.00 ±20.27 11–88	33.50 ±31.82 11–56	101 (105)	14 (13.33)	11 (10.48)	13 (12.38) 0.010*	54 (51.43) 0.001*	12 (11.43)	1 (0.95) 0.002*

n – number of patients/number of teeth affected; n_A – number of abnormalities (e.g., 2 abnormalities in 1 tooth); n_A/1 – number of abnormalities per 1 affected patient; min – minimum; max – maximum; *M* – mean; SD – standard deviation; VCR – vincristine; DXR – doxorubicin; DNR – daunorubicin; CP – cyclophosphamide; IF – ifosfamide; VP-16 – etoposide; CBDCA – carboplatin; CDDP – cisplatin; ActD – actinomycin-D;
* statistically significant difference between the drug group and the total group ($p \leq 0.05$; Mann–Whitney *U* test).

between the affected and non-affected patients within most groups of dental abnormalities for each drug, except for microdontia in the DXR group ($p = 0.04$) and a reduction in tooth size in the CP group ($p = 0.03$) (Table 2). However, CP therapy in the participants with a reduced crown size was paradoxically longer in the non-affected group, with a similar anomaly appearing in many groups of abnormalities regardless of the drug administered (Table 2). For the mean treatment duration, no significant correlations were observed in the VCR group. Strong significant positive correlations in the affected group were observed between microdontia and M+R in CP recipients ($p = 0.01$), M+R and the total group receiving VP-16 ($p = 0.01$), and taurodontia and the total group receiving CDDP ($p = 0.01$). Among the non-affected patients with a particular abnormality, the following strong significant positive correlations were noted: agenesis vs. reduction in tooth size in DXR recipients ($p = 0.0003$); agenesis vs. taurodontia in the CP group ($p = 0.0005$); agenesis vs. enamel changes in the IF and CBDCA groups ($p = 0.004$ and $p = 0.0003$, respectively); and M+R vs. agenesis ($p = 0.03$) and enamel changes ($p = 0.03$) in patients receiving CBDCA. A strong significant positive correlation was also established between the affected and non-affected patients with a particular abnormality within the DXR group in terms of reduction in tooth size ($p = 0.05$). Strong significant negative correlations were established only within the ActD group between the affected and non-affected patients in terms of reduction in tooth size and taurodontia ($p = 0.05$ and $p = 0.05$, respectively). Some groups of patients with disturbed odontogenesis were too small to analyze the relationships (Table 3).

To test the hypothesis that the observed shorter treatment intervals in the first 10 weeks would result in a higher cumulative treatment dose than in the remaining longer period of therapy, a separate calculation was performed for the initial 10 weeks and for the total duration of therapy. There were no statistically significant differences between patients with different disturbances in most specific drug groups when analyzing the treatment duration and the drug doses, except for VCR patients with agenesis and microdontia in the first 10 weeks of therapy. Moreover, a pattern was frequently observed such that the mean treatment dose in the affected group was lower than in the non-affected group, although no statistically significant differences were noted, except for the following groups: VCR (the first 10 weeks of therapy) presenting with microdontia ($p = 0.01$); IF exhibiting M+R (both the initial 10 weeks and the total duration of therapy) ($p = 0.02$ in both cases); and the CBDCA total group (both the initial 10 weeks and the total duration of therapy) ($p = 0.03$ and $p = 0.05$, respectively). In all of the above examples, the mean cumulative treatment dose was significantly higher in the groups non-affected with the analyzed abnormalities (Table 2). Due to the lack of differences in the treatment duration and dose, only the correlations between the survivors with different abnormalities, and between the survivors affected and non-affected with a particular abnormality could be established. Strong significant correlations are listed in Table 3.

Discussion

The toxic effects of chemotherapy on odontogenesis have been well documented in animal models.[9,23,24] Cell susceptibility to toxic factors varies depending on the developmental stage during the course of treatment, and is related to different side effects, from the death of immature precursors, through transient development cessation, to the disturbed metabolic function of fully formed elements. Phenotypically, the toxic effects of chemotherapy can be observed as the lack of or reduced germs, abnormal tissue structure, or shorter dental roots.[19,25–28]

Clinical risk factors, such as age at diagnosis, the stage of dental development at the onset of treatment and the type of therapy, are still areas of active research.[10,14,20,25] A young age at diagnosis is hypothesized to be the most important risk factor. It is widely documented that the earlier anticancer therapy is started, the higher probability of dental abnormalities. The most affected patients have been reported to be treated before 3–5 years of age and they tended to show the most severe abnormalities, such as agenesis and microdontia.[3,7,11,14–19] In our study, only one survivor started treatment before 3 years of age, and presented with no abnormalities and a relatively short therapy duration. This patient received 8 courses of VCR and 3 cycles of ActD during 11 weeks of chemotherapy. However, as shown in an earlier study, agenesis and a reduction in crown size were also diagnosed in patients not treated at a very young age.[25] If therapy overlaps with the early odontogenesis of third molars, which develop at a later age, agenesis or microdontia may also occur.[25] Some investigators reported no significant relationship between tooth impairment and age at the onset of therapy, although some noticed an association between early exposure to chemotherapy and agenesis or microdontia.[7,11,17] Proc et al. reported that microdontia was observed in patients who started therapy before 42 months of age; patients treated between 43 and 61 months of age had no abnormalities, and patients aged >61 months presented with disturbed third molars.[11] In our study, the use of drug-dependent group division randomly created various age groups of patients. Agenesis was most commonly diagnosed (more often than the average prevalence of 8.23%) in younger affected patients, aged between 24.90 and 38.14 months at the onset of anticancer treatment. Combined M+R was more common (vs. the average prevalence of 36.63%) when therapy was administered to patients aged between 24.90 and 34.72 months. A higher than average (11.11%) prevalence of root disturbances was observed in the older group of patients who started

Table 2. Characteristics of the groups affected and non-affected with particular abnormalities in terms of treatment duration and dose administered for each analyzed drug

Drug administered (group analyzed)	Mean treatment duration [weeks] *M ±SD*							Mean cumulative drug dose 0–10 weeks [mg] *M ±SD*							Mean cumulative drug dose 0–90 weeks [mg] *M ±SD*						
	total	agenesis	microdontia	reduction in tooth size	M+R	enamel changes	taurodontia	total	agenesis	microdontia	reduction in tooth size	M+R	enamel changes	taurodontia	total	agenesis	microdontia	reduction in tooth size	M+R	enamel changes	taurodontia
VCR (affected)	38.50 ± 23.09	25.20 ± 9.96	39.50 ± 23.63	39.93 ± 21.55	41.18 ± 22.95	28.20 ± 21.34	46.00 ± 22.95	5.28 ± 2.41	**5.90 ± 2.06[d]**	**3.71 ± 2.09[de]**	5.09 ± 2.50	4.85 ± 2.48	4.19 ± 2.68	4.72 ± 2.72	12.47 ± 8.24	9.56 ± 3.64	10.87 ± 10.29	10.92 ± 5.19	11.81 ± 7.76	7.47 ± 1.55	12.32 ± 9.01
VCR (non-affected)	37.00 ± 20.38	40.68 ± 23.02	37.59 ± 22.02	36.50 ± 23.07	34.08 ± 21.05	40.08 ± 22.08	34.71 ± 21.33	6.55 ± 3.65	5.56 ± 2.94	**6.40 ± 2.78[e]**	6.07 ± 3.02	6.61 ± 2.94	5.70 ± 2.86	6.00 ± 2.79	12.81 ± 7.87	13.16 ± 8.54	13.17 ± 7.19	13.99 ± 9.79	13.54 ± 8.53	13.58 ± 8.40	12.66 ± 7.77
VCR (total)	38.10 ±20.06							5.60 ±2.76							12.56 ±7.89						
DXR (affected)	27.55 ± 13.19	27.60 ± 11.26	**34.80 ± 8.56[a]**	20.75 ± 6.08	29.29 ± 11.73	35.67 ± 17.10	26.40 ± 12.03	71.32 ± 48.12	68.94 ± 50.86	45.97 ± 34.16	79.92 ± 52.12	65.41 ± 46.69	104.07 ± 74.42	44.51 ± 38.15	153.28 ± 68.04	155.48 ± 68.51	159.64 ± 54.34	153.08 ± 66.44	157.46 ± 50.16	194.8 ± 66.34	113.52 ± 71.30
DXR (non-affected)	10.00 ± 12.73	23.13 ± 16.28	**18.63 ± 13.76[a]**	26.67 ± 16.64	19.67 ± 16.12	21.60 ± 12.39	24.25 ± 15.73	69.20 ± 54.87	72.28 ± 47.50	86.64 ± 48.44	67.03 ± 46.86	77.52 ± 50.25	61.08 ± 34.70	87.55 ± 45.76	123.20 ± 131.24	144.39 ± 80.98	141.79 ± 86.40	146.69 ± 80.39	138.38 ± 98.77	134.81 ± 72.99	170.61 ± 70.40
DXR (total)	24.85 ±12.82							71.00 ±46.71							148.65 ±73.62						
CP (affected)	28.91 ± 6.43	30.00 ± 7.94	31.17 ± 5.19	**25.29 ± 5.22[bc]**	27.80 ± 5.96	**35.00 ± 4.36[c]**	29.25 ± 6.18	885.85 ± 500.29	940.00 ± 225.17	650.40 ± 361.21	850.00 ± 339.12	775.24 ± 346.89	885.80 ± 995.61	520.60 ± 343.59	2719.21 ± 1286.68	3300.00 ± 1530.88	2741.47 ± 1251.62	2207.14 ± 626.78	2484.88 ± 1080.91	3070.43 ± 1891.51	2012.20 ± 735.66
CP (non-affected)	26.00 ± 1.00	28.00 ± 6.11	26.14 ± 6.52	**32.17 ± 5.56[b]**	30.67 ± 8.14	26.50 ± 5.44	28.11 ± 6.64	375.00 ± 530.33	770.74 ± 583.43	946.43 ± 618.54	762.90 ± 709.44	925.00 ± 1023.78	787.00 ± 369.08	938.33 ± 546.99	2200.00 ± 141.42	2441.13 ± 1087.41	2551.79 ± 1230.35	3143.55 ± 1538.93	3154.17 ± 1655.69	2510.00 ± 1008.37	2918.06 ± 1281.39
CP (total)	28.46 ±6.02							809.8 ±518.89							2639.33 ± 1191.34						
IF (affected)	20.29 ± 10.44	26.00 ± 1.41	17.33 ± 14.05	28.33 ± 3.51	21.00 ± 11.18	25.00 ± 0.00	19.33 ± 11.37	10.89 ± 7.96	8.40 ± 1.70	7.75 ± 1.10	7.85 ± 1.00	**7.56 ± 0.82[f]**	7.20 ± 0.00	14.92 ± 12.06	22.83 ± 12.20	34.20 ± 12.73	14.91 ± 10.59	23.35 ± 4.85	**17.56 ± 8.71[h]**	25.20 ± 0.00	22.08 ± 10.12
IF (non-affected)	49.00 ± 0.00	23.17 ± 16.50	27.80 ± 13.92	21.20 ± 17.71	28.67 ± 19.55	23.71 ± 15.13	26.60 ± 15.95	16.20 ± 0.00	12.60 ± 8.67	13.83 ± 9.13	13.77 ± 9.19	**18.20 ± 9.75[f]**	12.17 ± 8.00	9.53 ± 3.86	45.00 ± 0.00	22.73 ± 13.89	32.01 ± 11.74	26.95 ± 17.69	**39.00 ± 8.88[h]**	25.65 ± 14.85	27.71 ± 16.27
IF (total)	23.88 ±14.01							11.55 ±7.61							25.60 ±13.75						

Drug administered (group analyzed)	Mean treatment duration [weeks] *M* ±*SD*							Mean cumulative drug dose 0–10 weeks [mg] *M* ±*SD*							Mean cumulative drug dose 0–90 weeks [mg] *M* ±*SD*						
	total	agenesis	microdontia	reduction in tooth size	M+R	enamel changes	taurodontia	total	agenesis	microdontia	reduction in tooth size	M+R	enamel changes	taurodontia	total	agenesis	microdontia	reduction in tooth size	M+R	enamel changes	taurodontia
VP-16 (affected)	23.33 ± 11.88	22.00 ± 18.19	26.00 ± 9.74	24.83 ± 9.50	24.10 ± 9.62	32.00 ± 11.27	28.25 ± 9.74	261.25 ± 285.57	210.00 ± 187.35	330.00 ± 315.56	180.00 ± 173.12	277.50 ± 301.19	240.00 ± 226.50	232.50 ± 179.37	1121.88 ± 731.84	1380.00 ± 923.69	1193.44 ± 505.69	785.00 ± 338.97	1040.25 ± 553.17	1660.00 ± 905.15	864.38 ± 268.14
VP-16 (non-affected)	12.00 ± 9.90	21.64 ± 10.99	16.00 ± 13.15	19.38 ± 13.72	15.75 ± 16.72	18.91 ± 11.01	19.10 ± 12.23	817.50 ± 10.61	376.36 ± 359.47	355.00 ± 381.68	461.25 ± 379.20	498.75 ± 396.37	368.18 ± 358.65	384.00 ± 374.98	1222.50 ± 562.15	1069.77 ± 655.11	1060.00 ± 935.29	1399.69 ± 788.76	1376.25 ± 1024.43	993.41 ± 594.89	1245.00 ± 788.48
VP-16 (total)	21.71 ±12.00							340.71 ±331.38							1136.25 ±691.98						
CBDCA (affected)	35.50 ± 23.47	21.33 ± 17.62	34.00 ± 9.90	42.33 ± 26.27	39.63 ± 22.77	37.00 ± 0.00	48.50 ± 27.97	**444.60 ± 528.48[g]**	240.00 ± 207.85	333.20 ± 433.63	621.00 ± 614.62	510.75 ± 566.36	–	684.00 ± 671.10	**2532.10 ± 1783.00[i]**	1800.00 ± 1298.00	2137.20 ± 1202.58	2720.17 ± 2111.01	2670.13 ± 1796.87	3600.00 ± 0.00	2827.75 ± 2292.78
CBDCA (non-affected)	43.67 ± 6.81	42.20 ± 19.94	39.50 ± 25.96	33.14 ± 15.70	33.80 ± 19.18	37.42 ± 21.75	32.44 ± 16.36	**1358.33 ± 562.86[g]**	780.10 ± 690.08	856.88 ± 701.99	685.00 ± 723.82	887.00 ± 768.86	710.08 ± 645.27	723.13 ± 678.74	**4816.67 ± 1172.36[i]**	3437.10 ± 1939.38	3635.63 ± 2094.61	3350.00 ± 1818.51	3682.00 ± 2100.81	3014.25 ± 1979.18	3467.50 ± 1716.10
CBDCA (total)	37.38 ±20.82							655.46 ±648.43							3059.31 ±1901.87						
CDDP (affected)	34.29 ± 19.32	–	41.75 ± 21.72	26.50 ± 17.68	36.20 ± 22.53	41.00 ± 2.83	31.40 ± 12.05	138.79 ± 155.05	–	93.13 ± 58.32	94.00 ± 56.57	101.30 ± 53.71	45.25 ± 12.34	135.10 ± 189.48	432.13 ± 344.22	–	333.48 ± 248.33	192.00 ± 8.49	303.98 ± 224.95	228.95 ± 43.77	427.38 ± 379.93
CDDP (non-affected)	–	34.29 ± 19.32	24.33 ± 12.34	37.40 ± 20.96	29.50 ± 12.02	31.60 ± 22.94	41.50 ± 38.89	–	138.79 ± 155.05	199.67 ± 239.35	156.70 ± 184.00	234.00 ± 330.93	176.20 ± 172.91	148.00 ± 19.80	–	432.13 ± 344.22	563.67 ± 466.40	528.18 ± 370.60	752.50 ± 470.23	513.40 ± 385.16	444.00 ± 364.87
CDDP (total)	34.29 ±19.32							138.79 ±155.05							432.13 ±344.21						
ActD (affected)	18.50 ± 14.68	21.25 ± 12.34	14.25 ± 19.21	26.00 ± 15.85	18.67 ± 16.74	10.00 ± 8.37	21.75 ± 20.22	1538.40 ± 585.94	1361.25 ± 476.45	1425.00 ± 695.52	1391.25 ± 659.02	1415.00 ± 623.30	1543.50 ± 754.73	1248.75 ± 570.97	3506.40 ± 2207.54	4121.25 ± 1642.50	2010.00 ± 820.76	4428.75 ± 2706.25	3455.00 ± 2603.28	2466.00 ± 1474.66	2493.75 ± 1276.34
ActD (non-affected)	17.00 ± 11.31	16.75 ± 14.94	20.25 ± 11.15	14.38 ± 11.70	17.83 ± 11.57	22.38 ± 14.41	16.50 ± 10.50	799.00 ± 1129.96	1442.13 ± 808.83	1410.25 ± 739.31	1427.13 ± 753.24	1415.33 ± 816.75	1351.00 ± 704.09	1498.38 ± 768.46	2374.00 ± 1097.43	2915.88 ± 2243.49	3971.50 ± 2233.50	2762.13 ± 1595.63	3180.33 ± 1617.96	3743.50 ± 2277.99	3729.63 ± 2337.29
ActD (total)	18.25 ±13.73							1415.17 ±692.68							3317.67 ±2071.46						

M+R – microdontia + reduction in tooth size subgroup. The same superscript letters indicate a statistically significant difference ($p \leq 0.05$; Mann–Whitney *U* test).

Table 3. Strong correlations according to Spearman's rho with regard to the study results depicted in Table 2

Drug administered	Value analyzed	Group tested	Therapy duration [weeks]	Group of patients with the same adverse effect						
				agenesis (A)	microdontia (M)	reduction in size (R)	M+R	enamel changes (E)	taurodontia (T)	total
VCR	treatment duration	affected	X	T: $r_s = -0.80$ R: $r_s = 1$	T: $r_s = -0.69$*	–	T: $r_s = 0.65$*	–	total: $r_s = -0.65$*	$r_s = 1$
	dose	affected	0–10	R: $r_s = -0.90$*	–	–	T: $r_s = 0.82$*	–	–	$r_s = 1$
			0–90	T: $r_s = 0.80$	–	–	T: $r_s = 0.93$*	–	–	$r_s = 1$
		non-affected	0–10	total: $r_s = 0.71$*	–	–	E: $r_s = -0.57$*	–	total: $r_s = -0.78$*	–
			0–90	–	$r_s = -0.76$*	–	–	–	–	–
DXR	treatment duration	affected	X	E: $r_s = 1$	–	M+R, total: $r_s = 0.83$	–	T: $r_s = -0.86$	–	–
		non-affected	X	R: $r_s = 0.94$* E: $r_s = 0.71$* T: $r_s = 0.70$*	–	–	–	T: $r_s = 0.75$*	–	–
		affected/non-affected	X	–	–	$r_s = 0.94$*	–	–	–	–
	dose	affected	0–10	R: $r_s = -0.80$	R: $r_s = 0.80$ E: $r_s = 0.86$	–	E: $r_s = -1$	–	–	$r_s = 1$
			0–90	–	–	total: $r_s = 0.80$	T: $r_s = 0.80$	–	–	$r_s = 1$
		non-affected	0–10	M+R: $r_s = -0.82$*	–	–	–	–	–	–
			0–90	T: $r_s = 0.68$*	–	–	–	–	–	–
		affected/non-affected	0–10	–	–	–	$r_s = 0.80$	–	$r_s = 0.80$	–
CP	treatment duration	affected	X	M, M+R: $r_s = -0.86$ E: $r_s = 1$	M+R: $r_s = 0.89$* E: $r_s = -0.86$	E: $r_s = 0.86$	E: $r_s = -0.86$ total: $r_s = 0.62$*	–	–	$r_s = 1$
		non-affected	X	M+R: $r_s = -1$ T: $r_s = 0.91$*	M+R: $r_s = -1$	–	E: $r_s = 1$	–	–	–
		affected/non-affected	X	$r_s = 0.86$	–	–	–	–	–	–
	dose	affected	0–10	E: $r_s = 0.86$ total: $r_s = -1$	E: $r_s = -0.86$	E: $r_s = -0.86$	E: $r_s = -0.86$	T, total: $r_s = -0.86$	–	$r_s = 1$
			0–90	M, M+R: $r_s = 1$ E: $r_s = -1$	M+R: $r_s = 1$ E: $r_s = -1$ T: $r_s = -0.80$	T: $r_s = 1$	E: $r_s = -1$ T: $r_s = -0.80$	–	–	$r_s = 1$
		non-affected	0–10	M+R: $r_s = 0.86$	–	M+R: $r_s = -0.86$	T: $r_s = 0.86$	–	–	–
			0–90	–	M+R: $r_s = -0.86$ E: $r_s = -0.78$*	M+R: $r_s = -1$	T: $r_s = -1$	–	–	–
		affected/non-affected	0–10	–	–	–	$r_s = 0.86$	–	$r_s = -0.86$	–
			0–90	–	–	–	$r_s = 1$	–	$r_s = 0.80$	–

Drug administered	Value analyzed	Group tested	Therapy duration [weeks]	Group of patients with the same adverse effect						
				agenesis (A)	microdontia (M)	reduction in size (R)	M+R	enamel changes (E)	taurodontia (T)	total
VP-16	treatment duration	affected	X	M, E, T: $r_s = 1$ R, M+R, total: $r_s = -1$	M+R, total: $r_s = -0.77$* E, T: $r_s = 1$	M+R, total: $r_s = 1$ E: $r_s = -1$ T: $r_s = -0.80$	E: $r_s = -1$ T: $r_s = -0.80$ total: $r_s = 0.81$*	T: $r_s = 1$ total: $r_s = -1$	total: $r_s = -0.80$	$r_s = 1$
		non-affected	X	–	M+R: $r_s = -0.80$	–	–	–	–	–
		affected/non-affected	X	$r_s = -1$	–	–	–	$r_s = -1$	$r_s = -0.80$	–
	dose	affected	0–10	R, M+R, total: $r_s = 1$	T: $r_s = 1$	M+R, total: $r_s = 1$	total: $r_s = 0.66$*	–	–	$r_s = 1$
			0–90	M, T: $r_s = 1$	T: $r_s = 1$	M+R, total: $r_s = 0.82$* E: $r_s = -1$	E: $r_s = -1$ total: $r_s = 0.64$*	total: $r_s = -1$	–	$r_s = 0.97$*
		non-affected	0–10	–	–	–	E: $r_s = 0.94$* T: $r_s = -0.94$*	–	–	–
			0–90	–	E: $r_s = 0.82$*	–	–	–	–	–
		affected/non-affected	0–10	$r_s = 1$	–	–	–	$r_s = -0.86$	–	–
			0–90	–	–	–	–	$r_s = -1$	–	–
IF	treatment duration	affected	X	–	T: $r_s = 1$	M+R, total: $r_s = 1$	total: $r_s = 1$	–	–	$r_s = 1$
		non-affected	X	E: $r_s = 0.94$*	T: $r_s = 1$	–	–	–	–	–
	dose	affected	0–10	–	–	M+R, total: $r_s = 1$ T: $r_s = -1$	T: $r_s = -1$	–	total: $r_s = -1$	$r_s = 1$
			0–90	–	–	–	T: $r_s = -1$	–	total: $r_s = -1$	$r_s = 1$
		non-affected	0–10	M+R: $r_s = -1$	M+R: $r_s = 1$	–	E: $r_s = -1$	–	–	–
			0–90	–	M+R: $r_s = -1$	–	–	–	–	–
		affected/non-affected	0–10	–	–	–	$r_s = -1$	–	–	–
			0–90	–	$r_s = -1$	$r_s = 1$	–	–	$r_s = 1$	–
CBDCA	treatment duration	affected	X	R, total: $r_s = -1$	T: $r_s = 0.80$	total: $r_s = 1$	–	–	–	$r_s = 1$
		non-affected	X	M+R, E: $r_s = 0.90$*	–	–	E: $r_s = 0.90$*	–	–	–
		affected/non-affected	X	$r_s = 1$	–	–	–	–	–	–
	dose	affected	0–10	M, T: $r_s = 0.86$	–	M+R, total: $r_s = 1$	total: $r_s = 0.89$*	–	–	$r_s = 1$
			0–90	R, M+R, total: $r_s = -1$	T: $r_s = 0.80$	M+R, total: $r_s = 1$	total: $r_s = 0.97$*	–	–	$r_s = 1$
		non-affected	0–10	total: $r_s = 0.86$	–	total: $r_s = 0.86$	–	total: $r_s = 0.86$	–	–
			0–90	E: $r_s = 0.75$*	–	–	–	–	–	–
		affected/non-affected	0–10	–	$r_s = 0.87$*	–	–	–	–	$r_s = 0.86$
			0–90	$r_s = -1$	–	–	–	–	–	–

Drug administered	Value analyzed	Group tested	Therapy duration [weeks]	Group of patients with the same adverse effect						
				agenesis (A)	microdontia (M)	reduction in size (R)	M+R	enamel changes (E)	taurodontia (T)	total
CDDP	treatment duration	affected	X	–	–	–	–	–	total: $r_s = 0.90$*	–
		non-affected	X	E: $r_s = -0.80$ T: $r_s = 1$	R: $r_s = -1$	–	–	total: $r_s = -0.80$	–	–
		affected/non-affected	X	–	–	–	–	–	–	$r_s = 1$
	dose	affected	0–10	–	M+R, total: $r_s = 0.80$	–	–	–	–	$r_s = 1$
			0–90	–	T: $r_s = 1$	–	total: $r_s = 1$	–	–	$r_s = 1$
		non-affected	0–10	M: $r_s = 1$	R: $r_s = -1$	E: $r_s = 0.90$*	–	–	–	$r_s = 0.86$
			0–90	–	–	E: $r_s = 1$	–	–	–	–
		affected/non-affected	0–10	–	$r_s = 1$	–	–	–	–	–
ActD	treatment duration	affected	X	M, E: $r_s = -1$ M+R: $r_s = -0.80$	M+R: $r_s = 0.80$ E: $r_s = 1$	–	E: $r_s = 0.80$	–	total: $r_s = 1$	$r_s = 1$
		non-affected	X	$r_s = -0.80$	–	$r_s = -0.94$*	–	–	$r_s = -0.94$*	–
	dose	affected	0–10	M, M+R: $r_s = -0.94$* T, total: $r_s = 0.80$	M+R: $r_s = 0.83$ E: $r_s = 0.88$ T, total: $r_s = -0.94$*	–	T: $r_s = -0.94$* total: $r_s = 0.94$*	–	total: $r_s = 1$	$r_s = 1$
			0–90	–	E: $r_s = 0.94$	M+R: $r_s = 1$ E: $r_s = 0.80$	E: $r_s = 0.80$	–	total: $r_s = 1$	$r_s = 1$
		non-affected	0–10	M: $r_s = 0.79$* E: $r_s = 0.70$*	–	–	–	–	–	–
			0–90	–	R: $r_s = 0.79$*	E: $r_s = 0.80$*	–	–	–	–
		affected/non-affected	0–10	$r_s = -0.94$*	$r_s = 0.94$*	–	–	–	–	–

r_s – strong correlations between the abnormalities analyzed (Spearman's rho); *statistical significance ($p \leq 0.05$).

therapy at 32.45–45.00 months. Enamel changes occurred in participants aged 26.60–45.00 months during chemotherapy, and taurodontia, which was predominantly diagnosed in permanent first molars, in patients aged 24.90–38.14 months (Table 1). Although these observations do not account for the impact of particular drugs, and thus their potential individual toxicity, it is interesting that the results described above correspond with the age which is expected to be a particularly susceptible period in tooth development. Similar observations about the average age at the onset of treatment were reported by Proc et al.; patients with 1 missing tooth had an average age of 23.40 ±13.50 months, patients with >1 missing tooth had an average age of 31.71 ±22.07 months, patients with 1 microdens had an average age of 17.00 ±0.00 months, patients with >1 microdens had an average age of 35.75 ±27.63 months, and patients with root abnormalities had an average age of 32.83 ±21.27 months.[11] In addition, Wilberg et al. noticed a significant association between the age at diagnosis ≤5 years and microdontia and enamel hypoplasia.[15]

In some previous investigations, the type of anticancer agent, the treatment dose or duration were considered possible risk factors for dental sequelae.[15,20,21] The current study revealed no differences in the prevalence of affected patients treated with all of the analyzed medications. The prevalence of affected patients evaluated separately for each drug was higher than the average for the total study group (75.68%), except for patients receiving VCR (73.33%) (Table 1). Some differences appeared when the prevalence of particular abnormalities was analyzed. For agenesis, no hypodontia was diagnosed in the group of patients treated with CDDP, whereas this abnormality accounted for 17.39% of all dental changes among participants receiving DXR. Such a difference in prevalence as compared to the total study group was statistically significant. However, this observation cannot be interpreted without taking into account the average age at treatment initiation. In the CDDP group, despite the fact that the therapy duration was relatively long (34.29 weeks), the affected participants were treated at a later age (an average of 45.00 months), when agenesis or tooth germ aplasia is usually not expected. In contrast, the younger average age of the affected survivors from the DXR group (27.73 months) might explain the significantly higher prevalence of hypodontia. Similarly, a younger average age at the onset of therapy (24.90 months) is the probable reason for the relatively higher number of teeth reduced in size (M+R) in the CBDCA group (60.38%). Moreover, CBDCA therapy had a longer duration (an average of 37.38 weeks), which is only slightly shorter as compared to that for VCR. However, the lowest prevalence of teeth with changes in crown size (22.86%) was found in the ActD group with younger participants, aged on average 26.60 months. This could be explained by the fact that treatment with ActD was the shortest in the total study cohort, with the average duration of 18.25 weeks. However, the fact that the largest number of teeth in the ActD group had enamel abnormalities, which can occur over a wide age range, is non-negligible and may randomly disrupt reliable observations. On the other hand, the highest prevalence of taurodontia and root disturbances found for DXR and CBDCA, and for IF and CDDP, respectively, corresponded with the period of the potential developmental susceptibility of the analyzed dental tissues (Table 1). A separate evaluation of the impact of individual drugs without a thorough analysis of other treatment factors cannot explain all important correlations and may lead to faulty conclusions. Some previous authors attempted to evaluate the drug-dependency of dental abnormalities. According to Wilberg et al., treatment with anthracyclines was significantly associated with the individual defect index (IDeI).[15] Krasuska-Sławińska et al. reported positive correlations between hypodontia, age at the onset of treatment and the use of VCR, DXR, CP, IF, and VP-16.[20] A positive correlation was also observed between microdontia and treatment with VCR, DXR, CP, IF, and VP-16, and between taurodontia and VCR administration. The same analysis using Spearman's rho revealed positive correlations between dental root resorption, age at diagnosis and therapy with VCR, CDDP, CP, IF, VP-16, and DXR. The prevalence of enamel defects was positively correlated with age at the onset of treatment and the use of VCR, platinum agents, CP, DXR, IF, and VP-16. However, no concomitant analysis for a higher number of variables was performed. Moreover, Krasuska-Sławińska et al. noted that VP-16, CDDP and IF had no proven toxic dental activity.[20] In our study, a relatively higher prevalence of affected patients was noted in groups receiving these drugs (85.71%, 100.00% and 87.50%, respectively). Furthermore, a comparable number of dental abnormalities per affected patient was diagnosed for almost all drug groups (ranging from 8.4 to 10.5), except for the CBDCA group (an average of 5.3 abnormalities per affected patient). Unexpectedly, group receiving CDDP, which is a CBDCA analog, presented with the highest number of abnormalities per patient (10.9). Further studies are thus needed due to the multi-drug nature of anticancer therapy. For example, the ActD group had the shortest treatment with this drug and the shortest duration of the entire therapy (18.25 weeks and 43.08 weeks, respectively), but the highest number of abnormalities per affected survivor, apart from CDDP. It is possible that a young age at the onset of chemotherapy was a more significant risk factor in this group (Table 1).

The duration of cytotoxic therapy seems to be another important risk factor. Although there were no significant differences in the prevalence of affected survivors between the drug groups, the mean treatment duration ranged widely from 18.25 ±13.73 to 38.10 ±20.06 weeks (Tables 1,2). Moreover, the mean duration of individual drug therapy in relation to particular late effects was not always compatible with the results for the mean therapy duration investigated for the total group. For instance,

a longer treatment duration was noted in patients without agenesis who received VCR. For microdontia, the same observation was made in patients treated with ActD; for reduction in tooth size, it was observed for DXR, CP and CDDP; for enamel defects, it was observed for VCR and ActD; and for taurodontia, it was observed for CDDP (Table 2). Based on these results, no relationships between the drug and the treatment duration and adverse dental effects could be confirmed. According to our previous research, treatment-induced dental abnormalities are likely in all patients who receive chemotherapy during tooth formation.[25] Other researchers agree with the observation that the number of dental abnormalities does not differ with regard to the therapy duration.[11,12,22] However, Krasuska-Sławińska et al. reported positive correlations between enamel changes, microdontia, agenesis, and DeI and the chemotherapy duration.[20]

It is necessary to consider whether the treatment duration is proportional to the cumulative dose received. In general, the longer the duration of therapy, the higher the cumulative dose administered. The size of a single dose depends on the patient's weight or body surface at the treatment time. Although weight and the body surface increase with age, the susceptibility of tooth germinative cells does not change. Notably, most cytostatic drugs have a short elimination half-life in human plasma. After a single intravenous dose of 0.7 mg/m^2 VCR in dogs, the distribution half-life and the plasma elimination half-life were 21.5 ±6.9 min and 47.6 ±14.2 min, respectively.[29] The plasma VCR level was observed to decrease significantly up to 10% within 30 min after administration and about 99% of the plasma drug content was cleared within 5 h after administration. Moreover, 120 min after intravenous injection, VCR was not detectable in plasma.[29] The dose-limiting neurotoxicity and short elimination half-life of plasma VCR can reduce its anticancer activity.[30] Regardless of the aforementioned data, VCR is toxic to the dividing immature cells, and even more so when its administration is repeated at regular intervals. Intensive therapy with weekly VCR may thus make the regeneration of dental immature cells impossible. Moreover, humans exhibit a lower rate of CP metabolism as compared to dogs, cats or mice.[31] In fully developed dental tissue-secreting cells, polarized microscopy has revealed regular incremental lines in dentin corresponding to intravenous chemotherapy injection.[28] This abnormality, although observable, does not change tooth morphology and cannot be radiologically diagnosed. Similarly, the transient cessation of dental root development may be difficult to differentiate. Another question is the relation of the weight- or body surface-dependent doses of cytotoxic drugs to the immature tooth germ. Developing dental tissues have similar susceptibility to toxins, but chemotherapy doses differ significantly. The obvious expectation is that the higher the dose administered, the more potential damage to developing cells. Some authors reported dose-dependent dental abnormalities. Krasuska-Sławińska et al. observed dose-dependency between all possible congenital disturbances and VCR administration, as well as between hypodontia, microdontia, root resorption, and enamel defects and therapy with DXR, CP and IF.[20] Kaste et al. revealed dose-dependency between the risk of occurrence of at least one tooth abnormality and alkylating agent therapy.[21] Furthermore, they reported a dose-dependent association between alkylating agents and ≥1 tooth anatomical abnormality, abnormal roots and crown shape, a reduction in size, and ≥1 missing tooth in cancer survivors diagnosed at <5 years of age. They also noted a striking association between the cumulative dose of CP and an increased risk of dental sequelae.[21] Another study revealed dose-dependency between CP and Holtta's defect index (HDI); specifically, the administration of a CP dose ≥7.5 mg/m^2 increased the HDI score by 13 as compared to patients not treated with CP.[32] However, other prospective studies failed to confirm these observations.[12,22] The authors of the present study also did not note any pattern regarding the cumulative chemotherapeutic dose and the type of dental sequelae. Additionally, it is worth pointing to the hypothesis that the initial 10 weeks of treatment concerns the intensive part of anticancer therapy due to shorter intervals between toxic drug administration in that period. However, the drug doses during the initial 10 weeks relative to the cumulative drug doses over the entire treatment period corresponds to the percentage ratio – 10 weeks to the average whole duration of treatment with the analyzed medication ×100%. During the initial 10 weeks (26.25% of the average total treatment duration), only VCR was used at a relatively higher (44.59%) mean dose as compared to the mean value for the entire treatment period. A striking dose-dependent significant positive association in the affected patients was found between M+R and taurodontia in the VCR group, reduction in tooth size vs. M+R and the total group in the VP-16 group, M+R and the total group in CBDCA recipients, and M+R and the total group in the ActD group. Strong dose-dependent significant negative correlations in the affected patients were revealed between agenesis and reduction in tooth size in VCR recipients, as well as agenesis vs. microdontia and M+R, microdontia vs. taurodontia and the total group, and M+R vs. taurodontia in the ActD group (Table 3). In the VCR group, the patients with agenesis received a significantly higher mean drug dose as compared to the patients affected with microdontia in the first 10 weeks. By contrast, no significant dose differences were observed between the affected patients treated with ActD, although the mean total cumulative dose administered to the patients with hypodontia was more than two-fold higher than the dose administered to the patients with microdontia (Table 2).

In the present study, the age range of patients treated with all the analyzed medications was wide (from 4–11 months to 55–102 months at the onset of therapy). However, there was no relationship between age

and the treatment duration. In oncology, the required treatment protocols are typically introduced according to recommendations for the type and severity of the disease rather than age at diagnosis. Kaste et al. showed no increased frequency of developmental issues beyond the age of 5 years at diagnosis, although patients were exposed to the highest levels of alkylating agents.[21] Similarly, Minicucci et al. noted a higher prevalence of dental sequelae in a group of younger patients, even though these patients received longer-lasting chemotherapy with less intensive protocols.[3] According to Proc et al., there was no relationship between age at the onset of anticancer treatment and the prevalence of missing teeth in their study, where agenesis was diagnosed in each tooth group mineralizing in different age periods.[11]

Limitations

A substantial limitation of this study is the small number of participants treated with different protocols. The multi-drug nature of anticancer treatment enables decreasing its toxicity to normal human cells. However, this makes the evaluation of the toxicity of a single therapeutic agent difficult.

Conclusions

In the present study, there were no significant differences between the groups of cancer survivors with particular dental abnormalities in terms of treatment duration and drug doses. This result indicates that there is some other reason for late adverse effects. The developmental stage of tooth formation during chemotherapy is likely to be the most important factor determining dental changes in children. It is believed that even a small dose of each anticancer drug can affect immature tissues. Given the different treatment protocols used and other confounding factors, such as a small sample size, and varying age and therapy duration, an analysis of a more homogenous group of survivors seems necessary.

Ethics approval and consent to participate

The study was approved by the Bioethics Committee at the Medical University of Silesia, Katowice, Poland, on February 25, 2013 and November 29, 2016 (KNW/0022/KB1/15/I/13 and KNW/0022/KB1/15/II/16, respectively). The caregivers of the participants provided informed written consent prior to the investigations.

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.

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