

Aim of the study: Chemotherapeutic treatment in children and adolescents carries a risk of congenital tooth disorders and dentinoma. Study objective is to assess the correlation between tooth abnormalities, early complications of multidrug chemotherapy, and chemotherapeutics used in different antineoplastic therapies in children and adolescents.

Material and methods: Enamel defects (developmental defects of enamel index – DDE index) and defects in tooth number, size, and structure were assessed clinically and radiologically in 60 patients who underwent chemotherapy on average 4.9 ± 3.4 years earlier (PCH), and 60 generally healthy subjects (control group – CG), aged 6–18 years. Höltta's defect index (Del) was calculated. Medical files provided information on neoplasm type, age at treatment start and chemotherapy duration, chemotherapeutic type and dose, vomiting, and mucositis (CTCAE v4.0). Statistical significance of differences between groups was assessed with the Mann-Whitney *U* test and the correlation between dental defects and chemotherapy with Spearman's rank correlation coefficient (significance $p \leq 0.05$).

Results: Enamel defects, tooth agenesis, microdontia, root resorption, taurodontism, and dentinoma occurred statistically significantly more often in the PCH group. A correlation was established between vincristine use and dose and all types of dental defects; cyclophosphamide, doxorubicin, and isophosphamide and hypodontia; microdontia, root resorption, and enamel defects; etoposide and cisplatin and microdontia, root resorption, and enamel defects; methotrexate root resorption and enamel defects; carboplatin and dentinoma and enamel defects. *Mucositis* and vomiting promoted root resorption, microdontia, and enamel defects.

Conclusions: Dental defects are related to both the use of respective chemotherapeutics, especially vincristine, cyclophosphamide, doxorubicin, and isophosphamide, and to early complications in multidrug chemotherapy – *mucositis* and vomiting. Vincristine and carboplatin use may promote dentinoma.

Key words: neoplasm, chemotherapy, children, enamel defects, agenesis, microdontia, congenital root abnormalities.

Antineoplastic chemotherapy and congenital tooth abnormalities in children and adolescents

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Introduction

Multidrug chemotherapy allows effective treatment of more than 70% of children with neoplasms, independently of the disease type or stage [1–9]. Unfortunately, chemotherapeutics are not selective and have a low therapeutic index, which results in damaging also healthy cells. To increase therapy effectiveness and, at the same time, minimise its side effects, several chemotherapeutic agents, with different mechanisms of action, at minimal therapeutic doses tolerated by the host, are used in multidrug chemotherapy. Unfortunately, even despite that, chemotherapy presents risks of side effects, i.e. early and late complications.

Children under antineoplastic treatment in early childhood present more often tooth aplasia, microdontia, root resorption, and congenital enamel defects than generally healthy children. Taurodontism, enlarged pulp chamber, and supernumerary teeth also occurred more often [10–43].

Congenital defects, which often occur in neoplasms or are early chemotherapy complications, may result from fever, metabolic disorders and hormonal imbalance, and malnutrition. Chemotherapeutics may also have a direct impact on dental tissues; e.g. vincristine, colchicine, and vinblastine temporarily impair odontoblast activity [7, 43–45]. Chemotherapy probably also delays the development of the Hertwig sheath [23].

Multidrug chemotherapy makes it more difficult to assess the impact of respective drugs on odontogenesis and tooth pulp. Up to now, most researchers have assessed the impact of chemotherapy on teeth in children with leukaemia. There is little information on the effects of chemotherapeutics in other neoplasm therapies, although brain and germ cell tumours are common under the age of five years.

The present study aims to assess the correlation between the prevalence of congenital defects in permanent teeth and early complications of multidrug chemotherapy and chemotherapeutics used in various neoplasms in children.

Material and methods

Patients

The study was approved by the Children's Memorial Hospital Commission for bioethics on May 12, 2010 (permit 95/KBE/2010), and patients/their legal guardians consented to participate in the research study.

One hundred and twenty patients under the age of 18 years, including 60 who completed chemotherapeutic treatment for neoplasms at least one year earlier (PCH group; mean age 11.81 ± 3.87 years) at the Oncology Clinic of the Children's Memorial Hospital, and 60 generally healthy patients (CG – control group; mean age 12.22 ± 3.63 years), treated at the Paediatric Dental

Clinic of the Children's Memorial Hospital, were examined. Children with chronic diseases other than neoplasms, or undergoing or having undergone chronic treatments, or after radiation therapy in the head and neck region, were excluded from the research study. Patients from both groups were of similar socioeconomic status.

Methods

Retrospective analysis of medical files

Patient medical files were analysed for information on neoplasm type, age at treatment start, antineoplastic treatment duration, chemotherapeutic type and dose, and early chemotherapy complication prevalence and severity, i.e. vomiting and mucositis according to CTCAE v4.0 [46]. Accumulated doses that every patient received under treatment were calculated. All doses were then converted into mg/m².

Dentition

All tooth surfaces were clinically assessed in successive quadrants. Tooth number, congenital disorders, including anatomic crown size and shape, and enamel defects were assessed with the modified DDE index (opacities, hypoplasia, and combination of both) [47].

Pantomographs served to assess congenital disorder prevalence: no tooth buds (agenesis was diagnosed when there was no tooth/tooth bud after extraction was ruled out), hypodontia, microdontia (when tooth width was smaller or equal to half of the normal size), anatomic crown disorders, i.e. resorption, V-shape, U-shape, taurodontism (in the case of excessive vertical elongation of the tooth chamber with no tooth neck, and a lowered root bifurcation in multiradicular teeth) [23, 47–49]. Höllta's defect index (Del) was used [23].

No bud of the first premolar in children aged less than five years or of the second premolar and second molar in children under six years old, as well as poor visibility of teeth on the pantomograph, led to exclusion from the assessment. Furthermore, teeth with unfinished root development or with important root curves, teeth with attrition, abrasion or anatomic crown fracture, and third molars were also excluded from root assessment.

The assessed teeth were attributed the following codes:

- D0 – no congenital disorders; ratio R/C > 1.6,
- D1 – ratio R/C from 1.6 to 1.2 – mild root resorption,
- D2 – ratio R/C from 1.1 to 0.9 – severe root resorption,
- D3 – ratio R/C < 0.9 – severe root resorption,
- D4 – microdontia,
- D5 – no tooth (aplasia).

Root length assessment included the longest root in multiradicular teeth, and the longest buccal root in mandibular molars and premolars. Crown height was calculated with measurement points at the line crossing through the incisive side of front teeth and buccal cusps of premolars and molars. R/C, i.e. the ratio of root length (R) to crown length (C), was calculated.

The prevalence of congenital disorders in respective teeth was assessed, and Del was calculated with the formula (nD1 × 1) + (nD2 × 2) + (nD3 × 3) + (nD4 × 4) + (nD5

× 5), where n was the number of teeth with respective D1, D2, D3, D4, and D5 codes.

Statistical analysis

Results were statistically analysed with the nonparametric Mann-Whitney *U* test (after a preliminary analysis of the compatibility of numeric values distribution with real distribution with the Shapiro-Wilk test). The impact of chemotherapy-related factors on dentition was assessed with Spearman's rho. Statistical significance was set at $p \leq 0.05$.

Results

The mean age at chemotherapy start was 5.9 ± 4.0 years, and treatment duration was 1.3 ± 0.5 years. On average, 4.9 ± 3.4 years elapsed since chemotherapy completion. Chemotherapy was used to treat: Burkitt's lymphoma (15.0%), nephroblastoma (13.0%), neuroblastoma (10.0%), histiocytosis (8.3%), rhabdomyosarcoma (6.7%), Ewing's sarcoma (6.7%), medulloblastoma (5.0%), neurofibromatosis type I (5.0%), and others (19.7%). The treatment protocols were appropriate for the respective diagnosis. The drugs most often used in combination therapy included: vincristine (VCR), cyclophosphamide (CTX), doxorubicin (ADM), etoposide (VP-16), cisplatin (CDDP), ifosfamide (IF), and actinomycin (ACTD). Other drugs were used in < 15% of patients. Table 1 presents the antineoplastic treatments.

Dentition

Enamel defects were observed statistically significantly more often in PCH than in controls (Table 2). PCH also presented higher mean numbers of teeth with opacities (6.316 ± 6.10 vs. 1.866 ± 2.64; $p = 0.000$) and hypoplasia (1.516 ± 3.61 vs. 0.15 ± 0.55; $p = 0.003$). There was no significant difference (0.533 ± 1.83 vs. 0.083 ± 0.38 respectively; $p = 0.06$) between mean numbers of teeth with a combination of enamel defects. Spearman's rho analysis revealed a positive correlation between enamel defect prevalence and age at chemotherapy start and its duration. Vomiting and the use and doses of vincristine, carboplatin, cyclophosphamide, doxorubicin, ifosfamide etoposide, and cisplatin were linked to the prevalence of opacities. Vincristine, methotrexate and mucositis showed a positive correlation with hypoplasia (Table 3).

Dental defects visible on pantomographs occurred statistically significantly more often in PCH than in controls (53.3% vs. 76.7%; $p = 0.0077$). More than one congenital defect ($p = 0.0000$) occurred in 46.7% of patients after chemotherapy, and only in 10% of controls ($p = 0.0000$). Root resorption was the most common defect (Table 2). The prevalence of short roots and the mean Del component related to this defect (D1 + D2 + D3) were statistically significantly higher in PCH than in CG. Severe and very severe root absorption (38.33% vs. 6.66%; $p = 0.0000$) occurred significantly more often in this group. In PCH the resorption occurred more often in first molar roots (in 21.6% of patients), and less often in incisors (15%), premolars (15%), and second molars (10%). In controls it most often (5.0% of subjects) occurred in first molars. Gener-

Table 1. PCH patient characteristics

Chemotherapeutic agent	n	Mean dose \pm SD (mg/m ²)
Vincristine (VCR)	53/88.0	10.46 \pm 8.0
Cyclophosphamide (CTX)	41/68.3	5287.18 \pm 12233.51
Doxorubicin (ADM)	40/66.5	167.41 \pm 152.43
Etoposide (VP-16)	39/65.0	1434.58 \pm 1712.87
Cisplatin (CDDP)	26/43.03	255.92 \pm 402.08
Ifosfamide (IF)	25/41.6	12511.67 \pm 21032.63
Actinomycin (ACTD)	18/30.0	3.06 \pm 8.54
Dacarbazine (DTIC)	17/28.3	1649.14 \pm 2757.64
Methotrexate (MTX)	13/21.6	3795.90 \pm 8195.43
Carboplatin (CBDCA)	11/18.3	1478.70 \pm 7148.41
Vinblastine (VBL)	8/13.3	15.33 \pm 56.24
Cytarabine (Ara-C)	8/13.3	1592.86 \pm 5498.39
Teniposide (VM-26)	7/11.6	155.67 \pm 446.76
5-Fluorouracil (5-FU)	3/5.0	83.33 \pm 457.95
Bleomycin (BLM)	2/3.3	3.00 \pm 17.06
Irinotecan (IRI)	2/3.3	38.33 \pm 247.72

Table 3. Statistically significant Spearman's rho for enamel defects and factors related to used anti-neoplastic treatment

	Opacities	Hypoplasia	Combination of defects
Age at chemotherapy start	0.2955*	0.1845*	0.1568*
Treatment duration	0.1943*	0.1980*	0.1459
VCR	0.2536*	0.1513*	0.0689
Dose	0.2401*	0.1186	0.0837
CBDCA	0.1504*	0.0164	0.0148
Dose	0.1493*	-0.0089	-0.0057
CTX	0.1834*	0.1136	0.0459
Dose	0.2034*	0.1198	0.0761
ADM	0.1937*	0.1219	0.0287
Dose	0.2042*	0.0834	0.0239
IF	0.1874*	0.0355	-0.0249
Dose	0.1720*	0.0339	-0.0201
MTX	0.0557	0.1546*	0.0535
Dose	0.0543	0.1568*	0.0524
VP-16	0.3239*	0.0696	0.0208
Dose	0.2907*	0.0724	0.0058
CDDP	0.2436*	0.0915	0.0772
Dose	0.2536*	0.0612	0.0517
Mucositis	0.1785*	0.1246	0.1395
Grade 2	0.1391	0.2099*	0.2023*
Vomiting	0.2550*	0.0246	0.0271

Table 2. Congenital disorders and Del in patients after anti-neoplastic treatment and in the control group

Congenital disorders	PCH	CG	p
	n/%		
Enamel defects	53/88.3	24/40.0	0.000*
opacities (DDE index: 1, 2, 5)	34/56.6	19/32.3	0.006*
hypoplasia (DDE index: 3)	7/11.6	1/1.6	0.001*
combination of lesions (DDE index: 6, 7)	12/20	4/6.6	0.068
Root resorption (D1 + D2 + D3)	36/60.0	25/41.6	0.0450*
V-shaped	18/30.00	0/0.0	0.0000*
Microdontia (D4)	12/21.67	0/0.0	0.0003*
no tooth bud (D5)	16/26.67	4/6.66	0.0035*
Del > 0	45/75.00	2643.33/	0.0004*
Del	mean \pm SD		
	12.48 \pm 13.16	2.24 \pm 3.84	0.0000*
Del component D1 + D2 + D3	5.65 \pm 6.92	1.81 \pm 3.03	0.0009*
Others	n/%		
taurodontism	12/20.00	3/5.00	0.0135*
mesiodens	1/1.67	0/0.0	0.3254
Dentinoma	18/30.0	9/15.0	0.0505
Impacted teeth	6/10.00	2/3.33	0.1466

*statistically significant differences; $p \leq 0.05$

ally healthy patients did not present any important premolar root resorption or V-shaped roots. The analysis of Spearman's rho revealed a positive correlation between dental root resorption and age at treatment start, and the use of vincristine, cyclophosphamide, ifosfamide, cisplatin P, and their doses. Early chemotherapy complications had no impact on this congenital defect (Table 3). A similar analysis of severe and extremely severe root resorption revealed a positive correlation also with doxorubicin, etoposide, teniposide, and vomiting (Table 3). The D1 + D2 + D3 component of Del was correlated with vincristine, cyclophosphamide, doxorubicin, ifosfamide, etoposide, and cisplatin use, age at treatment start, sometimes treatment duration, and grade 3 mucositis (Table 3).

Tooth agenesis occurred also more often in patients after chemotherapy than in controls (Table 2). Hypodontia (< 6 missing teeth) occurred in 15 PCH patients and 5 controls; oligodontia (8 missing teeth) in one. In PCH on average 2.87 \pm 1.6 teeth were missing, and in CG 1.6 \pm 0.55 (statistically significant difference; $p = 0.0023$). Premolars were most often missing in PCH (12/16 patients); second molars (4/16) and mandibular incisors (2/16) were missing less often. For one patient from that group both premolars and second molars were affected, while for another one, premolars, second molars, and incisors were affected. In controls, premolars (3/5) and maxillary lateral incisors (2/5) were missing. Maxillary teeth were missing more often than mandibular ones. Five PCH patients had both maxillary and mandibular teeth missing. Spearman's rho established a positive correlation between the absence of

Table 4. Statistically significant Spearman's rho determining the correlation between tooth congenital disorders and the used antineoplastic treatment, age at treatment start, treatment duration, and early complications

	Root resorption			Tooth agenesis	Microdontia	Del
	D1, D2, D3	D2 and D3	Del D1 + D2 + D3			
Age at start	0.1844*	0.3600*	0.2823*	0.1981*	0.1050	0.3664*
Treatment duration	0.1520	0.3089*	0.2459*	0.2415*	0.3099*	0.4276*
Time since treatment completion	0.2459*	0.4413*	0.3462*	0.3726*	0.4801*	0.5802*
VCR	0.2517*	0.4049*	0.3746*	0.2777*	0.3748*	0.5487*
Dose	0.2327*	0.3427*	0.3517*	0.2756*	0.3322*	0.4962*
CTX	0.2866*	0.4054*	0.4029*	0.2724*	0.3702*	0.5047*
Dose	0.2796*	0.4032*	0.3992*	0.2632*	0.3678*	0.4852*
ADM	0.1601	0.3931*	0.2850*	0.2148*	0.3025*	0.4385*
Dose	0.1504	0.3722*	0.2632*	0.1990*	0.2549*	0.3942*
DTIC	0.1673	0.1817*	0.1784	0.1389	0.1036	0.2228*
Dose	0.1758	0.1843*	0.1807*	0.1544	0.1296	0.2290*
IF	0.2257*	0.4115*	0.2989*	0.2111*	0.2394*	0.3491*
Dose	0.2135*	0.4046*	0.2902*	0.2024*	0.2062*	0.3339*
MTX	0.1111	0.1530	0.1717	0.1491	0.0741	0.2111*
Dose	0.1091	0.1505	0.1737	0.1401	0.0709	0.2068*
VP-16	0.1791	0.3625*	0.2999*	0.1763*	0.1911*	0.3597*
Dose	0.1977*	0.3631*	0.3062*	0.1917*	0.1919*	0.3636*
CDDP	0.1847*	0.2641*	0.2053*	0.1009	0.3078*	0.3193*
Dose	0.1949*	0.2784*	0.2030*	0.1346	0.3497*	0.3370*
5-FLU	-0.1601	-0.0863	-0.1544	-0.0716	0.3025*	0.0942
Dose	-0.1302	-0.0701	-0.1255	-0.0582	0.3906*	0.1843*
VM-26	0.1778	0.2916*	0.1714	0.1749	0.1541	0.2176*
Dose	0.1795*	-0.0701	0.1733	0.1748	0.1481	0.2166*
Mucositis	0.0934	0.1649	0.1050	0.0626	0.2489*	0.1303
Grade 2	0.0556	0.0865	0.0149	0.0745	0.2593*	0.0877
Grade 2, 3	0.1601	0.1694	0.2328*	0.0716	0.1245	0.2252*
Vomiting	0.1618	0.2010*	0.1767	0.0362	0.2967*	0.2379*

*statistical significance; $p \leq 0.05$

tooth buds and the age at treatment start, and sometimes also its duration, and the use of vincristine, cyclophosphamide, doxorubicin, ifosfamide etoposide, and their doses (Table 4). Furthermore, the analysis revealed that for each of these drugs the number of missing teeth increased when the used dose (correlation coefficient for vincristine: 0.2911, cyclophosphamide: 0.2795, doxorubicin: 0.2043, ifosfamide: 0.2058, etoposide: 0.2041) and treatment duration were increased (correlation coefficient: 0.2526).

Microdontia occurred only in PCH (Table 2). The number of teeth with smaller dimensions ranged between 1 and 8 (mean 4.0 ± 2.26). In three patients microdontia occurred only in maxillary teeth, and in one only in mandibular teeth. Microdontia of premolars and second molars was most common (in 9/12 and 7/12 respectively, including in 4 patients in both premolars and second molars). One patient presented a reduction in tooth size in maxillary premolars and mandibular incisors. A positive correlation between microdontia and chemotherapy duration, chemotherapeutic use and its doses (VCR, CTX, ADM, IF, VP-16,

CDDP, and 5-FU), vomiting and mucositis (Table 4) was established. The same factors had an impact on the number of microdontic teeth, which increased with chemotherapy duration (correlation coefficient 0.3109) and the dose of the aforementioned drugs (coefficients for vincristine: 0.3309, cyclophosphamide: 0.3617, doxorubicin: 0.2577, IF: 0.2010, etoposide: 0.1851, cisplatin: 0.3435, 5-fluorouracil: 0.4099).

A correlation was also established between Del and age at treatment start, treatment duration, the use and dose of vincristine, cyclophosphamide, doxorubicin, ifosfamide, methotrexate, etoposide, cisplatin, and grade 3 mucositis and vomiting (Table 4).

Other congenital disorders most often included taurodontism, which occurred statistically significantly more often ($p = 0.0135$) in PCH than in CG (20.0% vs. 5.0%). Hypertaurodontism was diagnosed in one PCH patient, and mesotaurodontism in three of them. Hypotaurodontism occurred in other patients after chemotherapy and in controls. A positive correlation between taurodontic teeth and

child age at treatment start (correlation coefficient 0.3000; $p = 0.0009$), and vincristine treatment (coefficient 0.2156), and its doses (coefficient 0.2096), was established.

Upon analysis of respective factors related to antineoplastic therapies, it turned out that vincristine treatment and its doses were related to all observed congenital disorders. Treatment with cyclophosphamide, doxorubicin, ifosfamide, and their doses, related to hypodontia, microdontia and root resorption, and enamel defects, also had a strongly negative impact on odontogenesis. Etoposide and cisplatin treatments were related to microdontic teeth, root resorption, and enamel defects; methotrexate use was related only to root resorption and enamel defects; and carboplatin use was related only to dentinoma prevalence and enamel defects. Early complications, such as mucositis and vomiting, promoted root resorption, reduction in tooth size, and enamel defects.

Discussion

The present study confirmed that chemotherapeutics could perturb odontogenesis, which also reflected the findings of other quoted studies.

Contrary to its results, some researchers did not discover any differences in enamel hypoplasia prevalence in children treated for neoplasms with chemotherapy or chemotherapy combined with radiotherapy and in the general population [13, 27, 50]. According to other researchers, the prevalence of enamel congenital disorders is higher in children who have undergone antineoplastic treatment [38, 51–53].

According to Oğuz *et al.*, some chemotherapeutics may cause congenital enamel defects [36], including vincristine, vinblastine, and cyclophosphamide. In animal research (hamsters 1997), Lyaruu *et al.* demonstrated that actinomycin D had a negative impact on amelogenesis [26]. In tests on rats receiving intravenous vincristine and vinblastine together with other chemotherapeutics, Greaves noted the presence of growth lines in teeth, correlated with chemotherapeutic treatment duration [54]. However, Marec-Berard *et al.* did not establish any correlation between drug use and dose and congenital enamel defects [27]. The present study indicates a correlation between hypoplasia and the use of vincristine and methotrexate, and opacities and the use of vincristine, cisplatin, methotrexate, doxorubicin, ifosfamide, and carboplatin. Näsman *et al.* noted white enamel opacities in 68% of children under chemotherapy and in 29% of controls [48]. Similarly, Nunn *et al.* observed higher prevalence of opacities and hypoplasia in the PCH group than in controls, but those differences were not statistically significant [55]. In the Hutton study (2008), 60.2% of children in a group of 120 undergoing chemotherapy presented enamel defects. Enamel hypoplasia did not occur as a single defect in any patient [11].

Oğuz *et al.* established a correlation between child age at treatment start and enamel hypoplasia, which reflects the results of the present study [36]. However, Marec-Berard *et al.* established, upon observing 27 children treated for different stages of neuroblastoma, and contrary to the results of the present study, that treatment duration had no impact on the prevalence of congenital disorders [27].

Kinirons *et al.* examined 54 children a long time after they had completed chemotherapy for acute lymphoblastic leukaemia and noted that treatment duration did not have any impact on the number of teeth with congenital enamel defects. They also established that the increase in the number of permanent teeth with enamel opacities was related to remission extension [56].

Dođan *et al.* assessed the correlation between early complications and enamel hypoplasia (2001). Nine among 85 children treated for acute lymphoblastic leukaemia suffered from mucositis, and five presented enamel hypoplasia [52]. In the present study, complications presented as vomiting and were correlated with opacities, and as grade 2 mucositis correlated with enamel hypoplasia. These complications could lead to malnutrition in children and present secondary amelogenesis disorders.

There are few publications on the impact of chemotherapy itself, without radiation therapy, on tooth bud development and congenital abnormality development. Since chemotherapy protocols include a couple of chemotherapeutics, there is no information on the impact of the respective drugs on odontogenesis. The available research presents all the types and prevalence of dental congenital abnormalities [14, 21–24, 57]. Maciel *et al.* found congenital abnormalities in 80.4% of patients treated for lymphoblastic leukaemia [14]. Höltta *et al.* reported that 100% of children after chemotherapy presented congenital abnormalities (Del>1) vs. 25% of controls [21]. Cubucku *et al.* (2012) compared the Del in children under chemotherapy (27 children) and in those under chemo- and radiation therapy (10 children) to Del in controls. According to their study, Del> 1 also occurred in 100% of patients and in 12.9% of controls [57]. In the present study, congenital tooth abnormalities, visible on pantomographs, occurred more often in patients after chemotherapy than in controls. However, the percentage of PCH patients (76.7%) with congenital tooth abnormalities was lower than the ones assessed by Minicucci *et al.* (82.9%) [28] and by Maciel *et al.* (80.4%) [14], but higher than the one assessed by Kaste *et al.* (60.5%) [38].

Researchers present various Del, which is probably related to different multidrug chemotherapy protocols in different neoplasms. For Höltta *et al.* [21], the Del was 15.3 ± 9.3 in patients under chemotherapy for lymphoblastic leukaemia and 1.8 ± 3.9 in controls. These index values were similar to the one in the present study [21]. Mean Del for Hisieh *et al.* (children treated for lymphoblastic leukaemia) was 24.7 ± 17.8 [25], and for Cubucku *et al.* (children treated for lymphoblastic leukaemia) 10.8 ± 11.2 [57].

In the present study, root resorption, occurring in 60% of patients after chemotherapy, was the most common congenital abnormality observed on pantomographs. In the Cubucku *et al.* study, congenital root abnormalities occurred more often (86.4%), but patients under chemotherapy and radiation therapy represented 27% of the treatment group [57].

Tooth agenesis or microdontia occurred slightly less often than root congenital abnormalities. The prevalence of these defects varied. Höltta assessed microdontia prevalence, in 55 children treated with high chemother-

apy doses before they turned ten years old, at 44% and agenesis at 46%. According to Marec-Berrard *et al.*, these percentages were 18% and 7% respectively [27]; to Oğuz *et al.*, 44% and 3% [36]; to Maciel *et al.* [14], 50% and 25%; and to Cubucku *et al.*, 16.2% and 13.5% [57]. In the present study, agenesis occurred in 26.67% of patients after chemotherapy, and microdontia in 21.67%. These defects most often occurred in premolars and second molars. According to Höltta *et al.* and Nishimura *et al.*, that is related to child age at treatment start, and at the same time to the stage of tooth development [21, 24, 58].

According to Nishimura *et al.*, there existed a strong correlation between age at treatment start and the prevalence of congenital tooth abnormalities. Root resorption occurred most often in treated patients between the age of zero and 11.8 years. In the present study microdontia or tooth agenesis was not reported in patients aged eight years or older. Tooth agenesis/microdontia occurred in 66.7% of patients under the age of four and treated with standard chemotherapy and in 100% of those treated with high drug doses. In the < 4 < 8 year group, tooth agenesis and microdontia occurred respectively in 18.2 and 25% of patients [58].

The present study also established that the longer the antineoplastic treatment was, the more frequent and severe were the congenital abnormalities. Every congenital abnormality was strongly correlated to the duration of antineoplastic treatment.

Cubucku *et al.* [57], and Nishimura *et al.* [58] did not establish any correlation between conventional chemotherapy duration and odontogenesis disorders. However, Höltta *et al.* did not analyse the impact of antineoplastic treatment duration on the prevalence of congenital dental disorders [21, 25].

The correlation between microdontia and the number of teeth reduced in size, and early complications of antineoplastic therapies, including vomiting and mucositis, is an interesting one. It has only been assessed by Olczak-Kowalczyk and Dembowska-Bagińska; however, the studies focused on children treated with chemotherapy combined with radiation therapy [59–61].

Congenital tooth abnormalities are said to result from the direct impact of the drug on odontoblasts and from its indirect impact, including early complications of chemotherapy. Höltta assessed, in his PhD thesis, the impact of different drugs on odontoblasts and ameloblasts in hamster teeth [24]. He established that the studied drugs impaired cellular functions only when they were in use; after treatment completion ameloblast and odontoblast functions were restored.

Numerous researchers [21, 25, 27, 28] have also attempted to assess the impact of chemotherapeutics on odontogenesis and tooth formation. Heirh *et al.* found that most Del values in patients treated with a combination of drugs, including cyclophosphamide, were higher than in those treated without that drug [25]. In their tests on animals, Lyaruu *et al.* confirmed that actinomycin D had an impact on tooth enamel and dentine formation, causing pre-odontoblast loss [28]. The assessment of the impact of doxorubicin on human dental pulp cells and fibroblasts

(varying drug doses) established that this chemotherapeutic caused a considerable reduction of live cells within the dental pulp and fibroblasts [24]. Some researchers [24, 62] suggest that methotrexate does not cause any dental congenital abnormalities. Five-fluorouracil causes mild perturbations in dental structure only during drug administration; once it is no longer administered odontogenesis resumes [24]. There is no information on the impact of ifosfamide and cisplatin on tooth development.

The present study established a positive correlation between VCR, CTX, IF, and CDDP and their doses and root resorption, including its most severe forms. Vincristine, cyclophosphamide, doxorubicin, and isophosphamide presented a correlation with hypodontia, which was confirmed by Höltta's study on hamsters.

Apart from drugs presenting a positive correlation with hypodontia, other chemotherapeutics, such as etoposide, cisplatin, and 5-fluorouracil, had an impact on microdontia. These results were similar to the ones obtained by other researchers [21, 24, 48]. However, etoposide, cisplatin, and ifosfamide were not listed among the drugs whose adverse reactions could cause dental abnormalities, and therefore further studies seemed necessary.

Maciel *et al.* noted higher prevalence of taurodontic teeth in patients after treatment with solely chemotherapeutics, compared to controls (7.1% vs. 5.3%). However, these differences were not statistically significant [14]. In the present study, the prevalence of taurodontism was statistically significantly higher in patients after chemotherapy than in controls (20% vs. 5%). A positive correlation between taurodontic teeth and child age at treatment start and vincristine treatment and dose was established. No information is available on the impact of respective chemotherapeutics on the formation of dentinoma and taurodontic teeth. The aforementioned impact of vincristine on pre- and odontoblasts, leading to perturbations in dentin formation, and on ameloblasts explains the results of the present study.

In conclusion, antineoplastic chemotherapy in children and adolescents promotes dentinoma and congenital tooth abnormalities, especially hypodontia, microdontia, root resorption, taurodontism, and congenital enamel defects. Multidrug chemotherapy including cyclophosphamide, doxorubicin, cisplatin, and vincristine has a particularly negative impact on teeth, which increases with the increase of the dose, treatment duration, and the severity of early complications, such as mucositis and vomiting.

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