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Case Report / Приказ болесника

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Developmental hypomineralization of the enamel of the first permanent and the second deciduous molars – report of two cases

Развојна хипоминерализација глеђи првог сталног и другог млечног молара – приказ два болесника

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SUMMARY

Introduction Molar-incisor hypomineralisation (MIH) is a developmental defect of dental enamel that affects one to all four first permanent molars (FPM) and frequently permanent incisors. Enamel aberrations are observed as demarcated opacities of different colours (from white to brown) and as posteruptive enamel breakdown (PEB). Clinically similar pathological signs can also be present in deciduous molars.

Case outline Histology of an FPM and a second deciduous molar (SDM) was performed after extraction from two unrelated patients with MIH due to inflammatory complications. Tooth samples were analysed using a stereomicroscope (SM), light microscope (LM) and scanning electron microscope (SEM). Enamel thickness of both affected teeth was normal. An obvious distinction in enamel microstructure was observed between the normally developed and the MIH-hypomineralised enamel with SM, LM, and SEM.

Conclusion In MIH patients, regular dental visits enable early diagnosis of the disease and appropriate treatment of the patient as soon as possible, with included preventive measures.

Keywords: MIH, enamel, first permanent molar, second deciduous molar, histology

САЖЕТАК

Увод Хипоминерализација молара и инцизива (*molar-incisior hypomineralization – MIH*) развојно је оштећење зубне глеђи која погађа један до сва четири прва стална молара а често и сталне секутиће. Аберације глеђи виде се као ограничена замућења различитих боја (од беле до смеђе) или као постеруптивни губитак глеђи (*posteruptive enamel breakdown – PEB*). Слични клинички патолошки знакови могу бити присутни и на млечним моларима.

Приказ болесника Хистологија првог сталног молара и другог млечног молара изведена је након што су зуби екстраховани због запаљенских компликација код два болесника са *MIH*. Хистологија зуба анализирана је уз помоћ стерео микроскопа (*SM*), светлосног микроскопа (*LM*) и скенирајућег електронског микроскопа (*SEM*). Дебљина глеђи оба оболела молара била је нормална. Уочена је јасна разлика у микроструктури глеђи између нормално развијене и *MIH* – хипоминерализоване глеђи са *SM*, *LM* и *SEM*.

Закључак Код болесника са *MIH* редовни стоматолошки преглед омогућава рану дијагнозу болести и одговарајуће лечење болесника у најкраћем могућем року, уз укључене превентивне мере.

Кључне речи: *MIH*; глеђ; први стални молар; други млечни молар; хистологија

INTRODUCTION

Molar-incisor hypomineralization (MIH) is a distinct entity, with a typical clinical picture of developmentally impaired enamel. One to all four first permanent molars (FPM) are affected and, in many cases, permanent incisors [1]. On individual teeth, enamel hypomineralization can be expressed in a wide variety in each patient [2]. The area of insufficiently mineralized enamel is clearly delineated from normal enamel. Aberrant enamel of normal thickness can be of different colors, from whitish to brownish. Enamel mineralization can be insufficient to such an extent that the loss of enamel tissue occurs after the eruption of the tooth. Such areas of missing enamel are clinically identified as

posteruptive enamel breakdown (PEB). As a rule, PEB is present on FPM. The size and the shape of such defects differ from carious lesions. PEB defects are present on areas where the carious process is not normally expected [3]. Because of the atypical location of deteriorated hypomineralized enamel, the shape of the filling on the MIH tooth does not coincide with the shape of a demineralized lesion that would develop due to caries. In patients with missing FPM that do not coincide with the clinical picture (the remaining teeth are healthy, with no caries or with only minor carious pathology), or in the presence of hypomineralized signs of MIH and/or atypical fillings on the remaining FPMs, the possibility that FPM was extracted due to MIH should also be considered.

Except for FPM and permanent incisors, signs similar to MIH are also described on the second deciduous molar (SDM), the second permanent molar and incisal part/cusps/tips of the permanent canine [3]. Garot et al. pointed out that children with MIH-like affected SDM have almost a five-fold higher likelihood of MIH presence in permanent dentition [4]. The study aimed to describe macro- and micro- aberrations in the enamel of the first permanent molar and the second deciduous molar obtained from two unrelated patients diagnosed with MIH.

CASE REPORT

Clinical examination of the patients

A nine-year-old boy was referred to the University Dental Clinic due to complications related to the endodontic treatment of a non-vital upper left FPM (tooth 26). The patient was otherwise healthy, with no metabolic, endocrine or any other systemic disease. The likelihood of dental fluorosis was excluded. The clinical status is described under Figure 1. A final interdisciplinary treatment plan was agreed. Based on the results of the analysis of occlusal relations, the poor long-term prognosis of both upper FPMs, and a possible midline shift, a decision to extract both upper FPMs was made. Tooth 16 (Fig. 1C) was analysed as described below.

The second patient, a nine-year-old girl, was referred to the clinic due to severe hypersensitivity of an upper right FPM (tooth 16). Her medical history also reported no systemic disease or medications (e.g., antibiotics, fluorides). The clinical status is described under Figure 2.

For the proposed treatment of the patients and further examination of the extracted teeth, informed consent was obtained from the patients and their parents. The study was approved by the Slovenian Committee for Medical Ethics (65/05/14).

Tooth samples

The analysed teeth were MIH-affected FPM and MIH-like SDM, as described above. Upon the extractions of the FPM and SDM, teeth were placed in an isotonic saline solution and then cut in the bucco-palatal direction. Both halves of each tooth were embedded in epoxy resin (Araldite, Ciba-Geigy, East Lansing, MI, USA) and polished after 24h, according to the established laboratory protocol. These prepared tooth specimens were examined in relation to histology with a stereomicroscope (SM), light microscope (LM), and scanning electron microscope (SEM).

Stereo microscopy, light microscopy, and scanning electron microscopy

Initially, the samples' enamel histology was observed with SM (Olympus SZ61, Olympus, Tokyo, Japan), LM (Olympus BX61, Olympus, Tokyo, Japan) at different magnifications. After histological examination with SM and LM was completed, the tooth samples were prepared for SEM according to the established laboratory protocol. Non-etched and later-etched enamel samples (37% phosphoric acid) were observed with SEM (Thermo Scientific QuattroS, Waltham, Massachusetts, USA).

Enamel histology

In both tooth samples, the thickness of the enamel was normal. However, both samples had areas with developmentary hypomineralised enamel, which extended almost all the way from the dento-enamel junction (DEJ) to the tooth surface (Fig. 3 A-D and 5 A-C). A thin layer of unaffected enamel on the tooth surface was lined with a normal aprismatic layer, while the majority of the bulk of the enamel was altered to varying extents. Under the LM, the hypomineralised areas appeared darker. As present in Figure 3 and 5, the surface of the MIH-affected tooth cusp could be preserved. In parts with regular development, prisms were normal and well defined (Fig. 4A and 6A). In the aberrant part of the enamel, the microstructure was deficient in most of its thickness, with prisms poorly defined and inadequately mineralised (Fig. 4B and 6B). In areas with poorly formed or even unrecognisable enamel prisms, different levels of porosity is anticipated as well as residual organic material, which had not been removed during amelogenesis. On etched samples, the difference between the normally formed enamel (with a well-formed etching pattern) and the hypomineralised enamel (with prism boundaries not clearly delineated) was even more apparent. The microstructure of the hypomineralised enamel showed poorer organisation of hydroksiapatite crystals within the prisms and wider sheath regions.

In the FPM and in the SDM, a clear demarcation between the normally developed and the developmentally affected enamel was observed under SM, LM, and SEM (Fig. 4C and 6C). Furthermore, in both samples, hypomineralised areas seemed to follow the incremental lines of Retzius. In the FPM as well as in the SDM, no changes in the structure of the dentine underneath the hypomineralised enamel was observable.

DISCUSSION

In this study, we observed poorly formed enamel prisms and insufficient mineralisation of macroscopically MIH-affected enamel. Hypomineralized areas spread from the DEJ towards the surface of the dental crown. The results coincide with a publication on the typical histology findings in MIH-affected FPMs [5]. Regarding the extent of hypomineralisation, aberrant areas may only be present in the inner layers of the enamel, at the DEJ, or may

include almost the whole enamel thickness [6]. In the yellow-brownish MIH-affected enamel, the entire thickness of the enamel is usually affected [7]. If the surface of the crown remains intact, the surface of the enamel is better mineralised compared to the deeper layers of enamel. This is attributed to the final mineralisation of the enamel after the eruption of the tooth [6]. The porosity of hypomineralised enamel enables the penetration of bacteria into the dentin, although the tooth surface is clinically intact. The bacteria in the dentinal tubules provoke an inflammatory reaction in the pulp, which consequently contributes to the hypersensitivity of MIH teeth [8]. Hypomineralised molars are also more prone to caries than those without developmental impairment, can cause serious restorative problems, and often even need to be extracted due to the extent of developmental disruption and treatment complications [9].

In this study, obtained results of chalky-like whitish patches on SDM also confirmed a poorer histological structure and enamel hypomineralisation. Similarly to the recently published article [10], we also observed a similarly aberrant histology of the affected enamel. In both specimens, the FPM and the SDM, there were clear demarcations between aberrant and normal enamel. However, the extent of hypomineralisation was more severe in the FPM in comparison to the SDM. This was not surprising since, as a rule, clinically MIH-like aberrations on SDMs are less severe than on FPMs [4]. A recently published systematic review of studies performed on extracted teeth diagnosed with MIH found a reduction in mineral quantity and quality in the MIH-affected enamel compared with unaffected enamel [11]. Further, MIH-affected enamel showed less dense prism structure, loosely packed crystals, more marked inter-prismatic space, wider sheath regions and abnormal etching pattern compared to normal enamel.

In conclusion, early diagnostics and proper treatment of the MIH-affected enamel is of utmost importance. Especially in the case of severe MIH, the failure to diagnose the disease early or delaying the necessary treatment may lead to additional complications that can result in the loss of tooth vitality or even of a tooth. The predictive factor for MIH disease of non-erupted FPMs can also be clinically detected in the MIH-like developmental impairment of SDMs; not only those with PEB but also with demarcated chalky hypomineralisation defects on the surface of its tooth crown.

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Conflict of interest: None declared.

REFERENCES

1. Weerheijm KL, Jalevik B, Alaluusua S. Molar-incisor hypomineralisation. Caries Res 2001; 35(5): 390-1. DOI: 10.1159/000047479. PMID: 11641576.
2. Sahlstrand P, Lith A, Hakeberg M, Norén JG. Timing of mineralization of homologues permanent teeth--an evaluation of the dental maturation in panoramic radiographs. Swed Dent J 2013; 37(3): 111-9. PMID: 24341164.
3. Weerheijm KL, Duggal M, Mejare I, Papagiannoulis L, Koch G, Martens LC, et al. Judgement criteria for molar incisor hypomineralisation (MIH) in epidemiologic studies: a summary of the European meeting on MIH held in Athens, 2003. Eur J Paediatr Dent 2003; 4(3): 110-3. PMID: 14529329.
4. Garot E, Denis A, Delbos Y, Manton D, Silva M, Rouas P. Are hypomineralised lesions on second primary molars (HSPM) a predictive sign of molar incisor hypomineralisation (MIH)? A systematic review and a meta-analysis. J Dent 2018; 72: 8-13. DOI: 10.1016/j.jdent.2018.03.005. PMID: 29550493.
5. Fagrell TG, Salmon P, Melin L, Norén JG. Onset of molar incisor hypomineralization (MIH). Swed Dent J 2013; 37(2): 61-70. PMID: 23957140.
6. Fearne J, Anderson P, Davis GR. 3D X-ray microscopic study of the extent of variations in enamel density in first permanent molars with idiopathic enamel hypomineralisation. Br Dent J 2004; 196(10): 634-8; discussion 25. DOI: 10.1038/sj.bdj.4811282. PMID: 15153976.
7. Al-Azri K, Melita LN, Strange AP, Festy F, Al-Jawad M, Cook R, et al. Optical coherence tomography use in the diagnosis of enamel defects. J Biomed Opt 2016; 21(3): 36004. DOI: 10.1117/1.JBO.21.3.036004. PMID: 26968386.
8. Fagrell TG, Lingstrom P, Olsson S, Steiniger F, Noren JG. Bacterial invasion of dentinal tubules beneath apparently intact but hypomineralized enamel in molar teeth with molar incisor hypomineralization. Int J Paediatr Dent 2008; 18(5): 333-40. DOI: 10.1111/j.1365-263X.2007.00908.x. PMID: 18328044.
9. Ivanovic M, Zivojinovic V, Sindolic M, Markovic D. [Molar incisor hypomineralisation in the first permanent teeth]. Srp Arh Celok Lek 2007; 135(7-8): 472-7. PMID: 17929543.
10. Alifakioti E, Arhakis A, Oikonomidis S, Kotsanos N. Structural and chemical enamel characteristics of hypomineralised second primary molars. Eur Arch Paediatr Dent 2020. DOI: 10.1007/s40368-020-00557-3. PMID: 32865711. [Epub ahead of print].
11. Elhennawy K, Manton DJ, Crombie F, Zaslansky P, Radlanski RJ, Jost-Brinkmann PG, et al. Structural, mechanical and chemical evaluation of molar-incisor hypomineralization-affected enamel: A systematic review. Arch Oral Biol 2017; 83: 272-81. DOI: 10.1016/j.archoralbio.2017.08.008. PMID: 28843745.

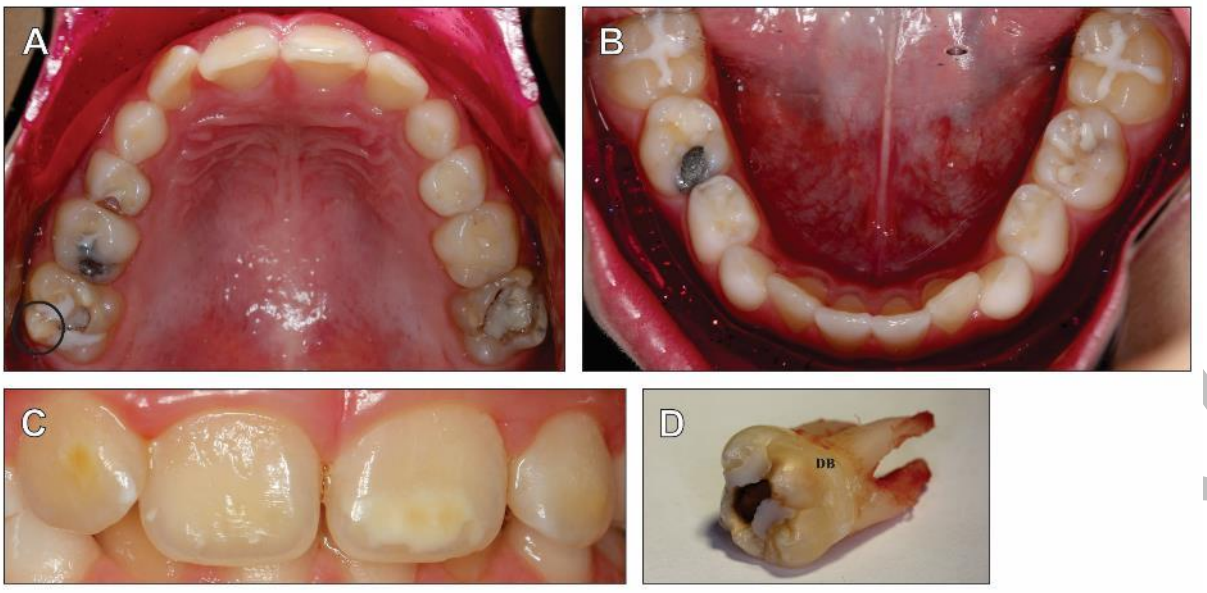

Figure 1. A) Upper dental arch of the 9-year-old boy with severely affected enamel of both upper FPMs; on Tooth 16, a hypomineralized disto-buccal tooth cusp (present in Figure 3) is marked with a circle; B) lower dental arch with healthy FPMs without hypomineralizational aberration; C) MIH-affected permanent upper incisors; D) the extracted upper right FPM (tooth 16) with posteruptive enamel breakdown (PEB) due to exceptionally poor mineralization of the enamel; DB: disto-buccal side of the tooth

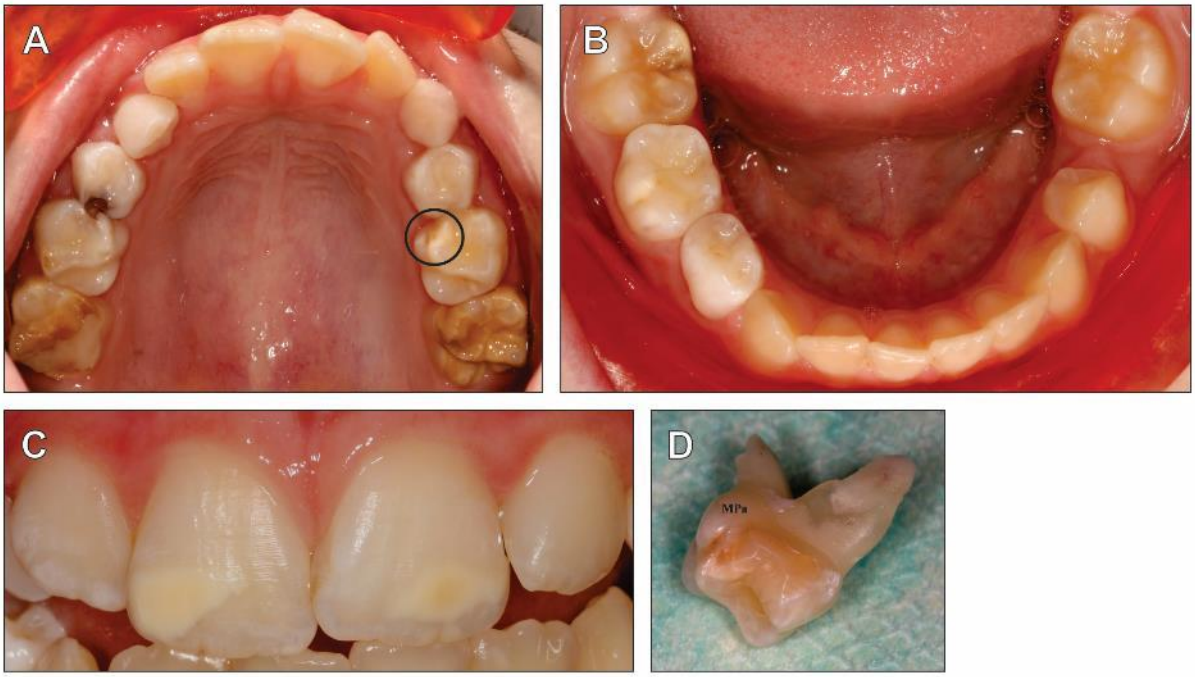

Figure 2. A) Nine-year-old girl with mixed dentition and severely affected enamel of both upper FPMs; the right FPM with an atypical restoration and the left FPM (tooth 16) with a posteruptive enamel breakdown (PEB); on Tooth 65, a hypomineralized mesio-palatal tooth cusp (present in Figure 5) is marked with a circle; B) on the lower right FPM (tooth 46), insufficiently mineralised enamel is sharply demarcated from a normally developed enamel; deciduous molars have chalky-whitish patches (teeth 54, 55, 65, 84 and 85); two deciduous molars are missing due to premature extractions; C) both upper central incisors are also MIH-affected; D) the hypomineralized upper left SDM (second deciduous molar) (tooth 65), extracted due to recurrent spontaneous pulpitic pain and further histologically examined; MPa: mesio-palatal side of the tooth

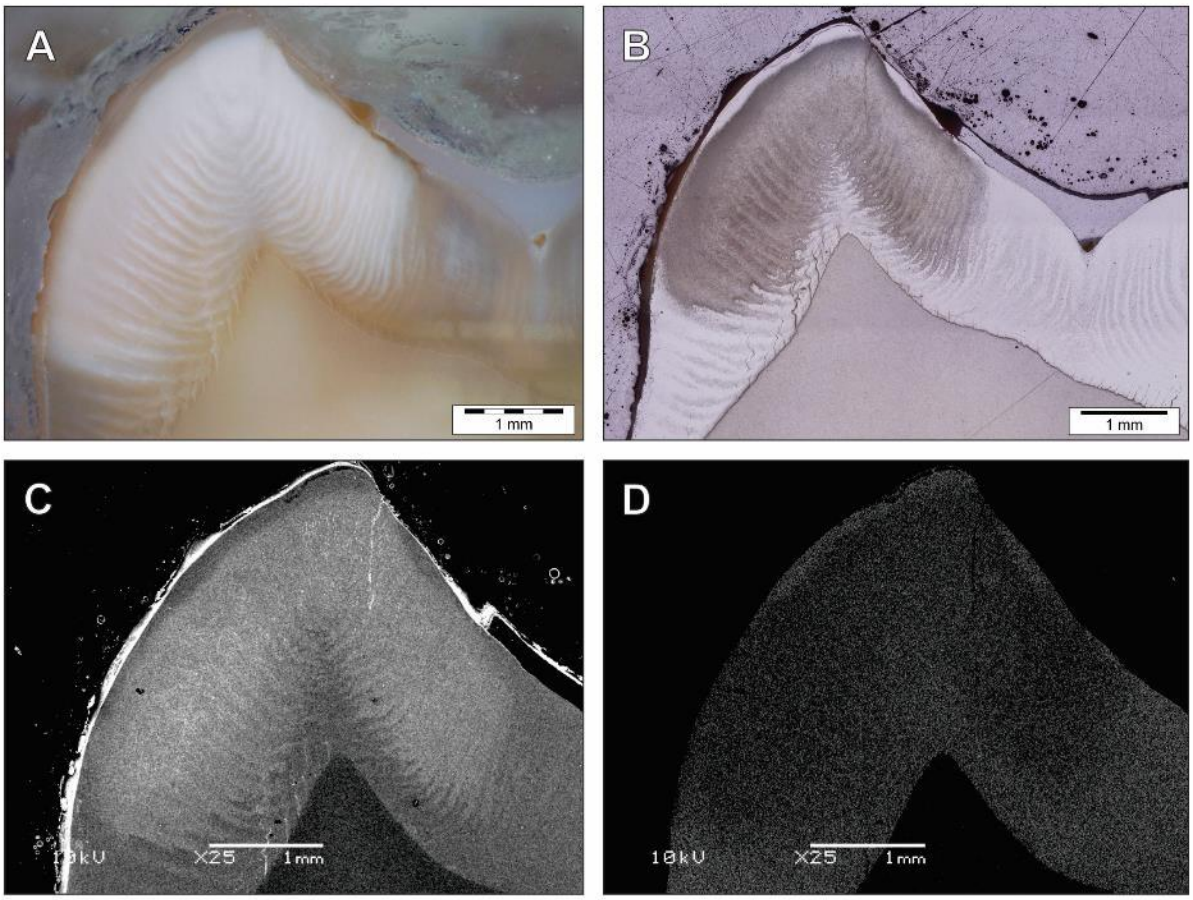

Figure 3. A) Stereo microscopy (SM) and B) light microscopy (LM) of the longitudinally cut upper-right FPM (Tooth 16) crown showed clearly less mineralized enamel through most of its thickness of an MIH-affected disto-buccal tooth cusp; between the porous and normally mineralized enamel, a clear delineation is visible; C, D) the same distobuccal tooth cusp observed with a scanning electron microscope (SEM); C) on the secondary electron image (SEI) directly below the surface, a thin layer (of some ten micrometers in its width) of appropriately mineralized enamel and a ribbon of normally mineralized enamel near the DEJ are visible; hypomineralized areas of enamel are brighter compared to apparently normal enamel; D) on the backscattered electron image (BSE) hypomineralized areas of enamel appear darker, and the borders are not as distinct as on the SEI image

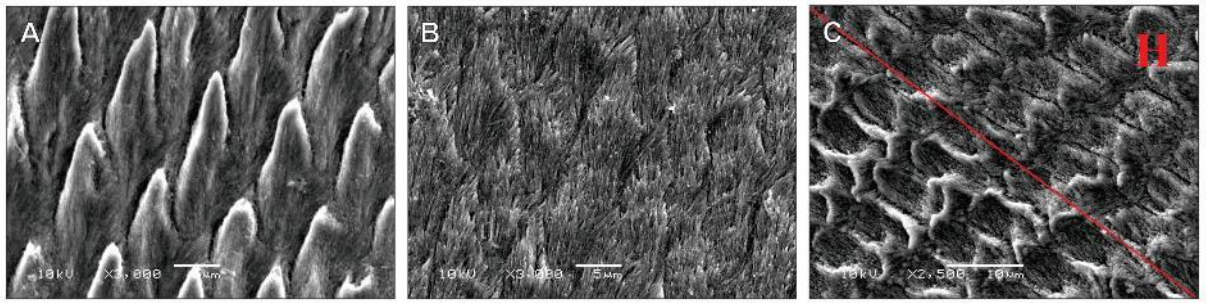

Figure 4. Scanning electron microscopy (SEM) of an etched sample of the affected FPM presenting A) normal and B) aberrant areas of the enamel; A) on parts where the course of amelogenesis was normal, the appearance of enamel prisms is typical, with nicely arranged enamel prisms; hydroxylapatite crystals are closely packed and correctly oriented (SEI, ×3000); B) conversely, on hypomineralized parts of the enamel, enamel prisms are less prominent; hydroxylapatite crystals are not packed together so tightly (SEI, ×3000); C) image presents the sharp delineation (delimited with red line) between the porous (i.e., hypomineralized-marked with H and normally mineralized enamel (SEI, ×2500)

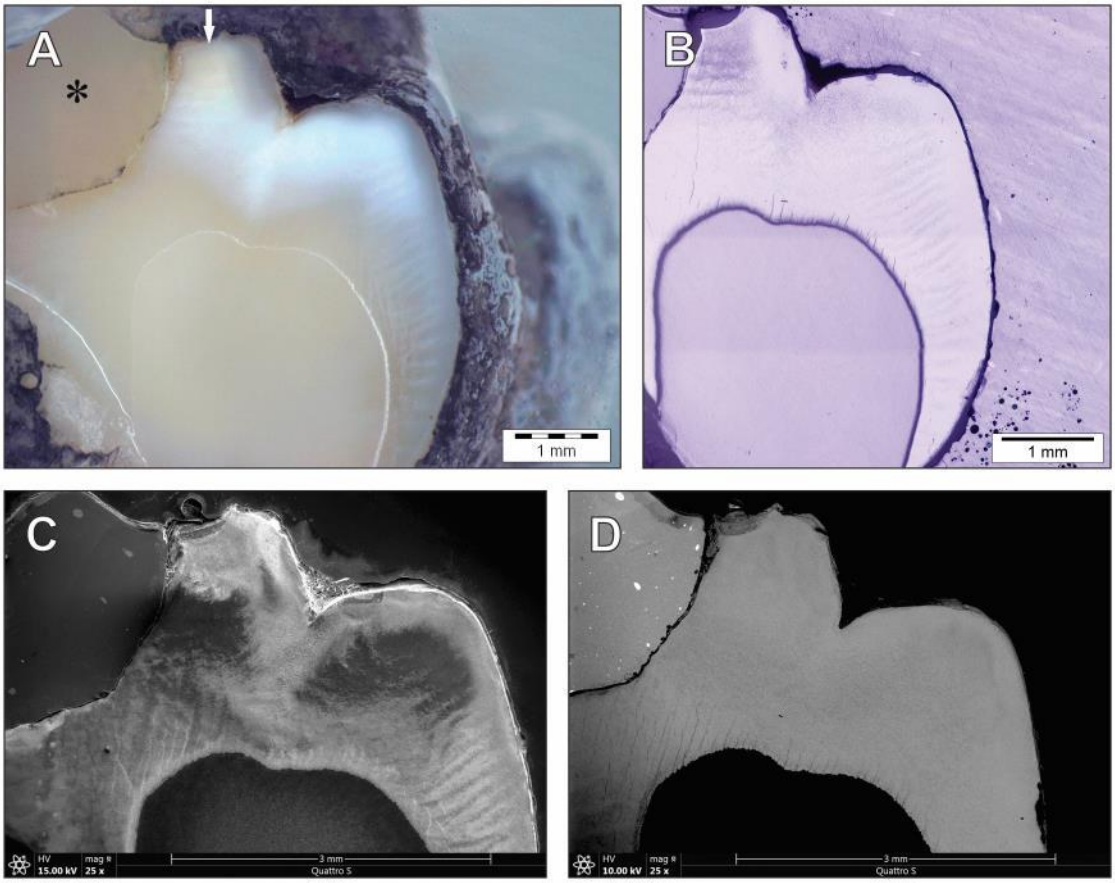

Figure 5. Longitudinally cut upper-left second deciduous molar (SDM; Tooth 65), observed as an unetched specimen with A) stereo microscopy (SM) and B) light microscopy (LM), revealed insufficiently mineralized enamel through the most of enamel thickness of the affected mesio-palatal tooth cusp; note a clear delineation between the insufficiently and normally mineralized enamel; the majority of the opacity is whitish, more porous hypomineralized enamel adjacent to the filling (marked with an asterisk) is cream colored (indicated by an arrow); the same tooth cusp, observed with a scanning electron microscope (SEM), after previous etching for 20 seconds, shows C) two layers of normally mineralized enamel: a layer directly below the enamel surface and a layer near the DEJ; the layer near the DEJ is slightly wider than the layer near the DEJ observed in the FPM (SEI); D) observation with backscattered electrons (BSE) exposed no obvious differences in the enamel structure.

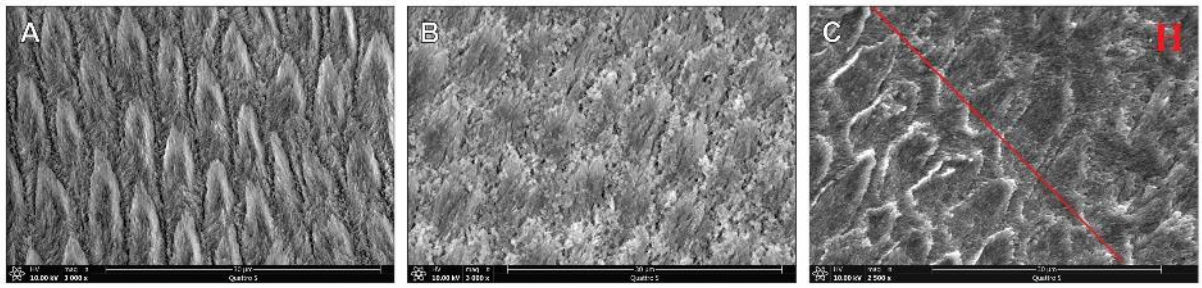

Figure 6. Scanning electron microscopy (SEM) of the etched sample of the affected SDM present A) typical etching pattern of properly arranged and well-formed enamel prisms in an area with normally developed enamel; hydroxylapatite crystals are closely packed and correctly oriented (SEI, ×3000); B) conversely, in the cream-colored area of the enamel, the prisms are disorganized (SEI, ×3000); C) the red line on an image shows clear demarcation in the histological structure of porous (H) and normally mineralized enamel (SEI, ×2500); H; hypomineralized enamel

