

Root malformation associated with a cervical mineralized diaphragm – a distinct form of tooth abnormality?

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Objective. Deviations in length and shape of tooth roots result from hard tissue resorption or occur as a developmental disorder. The purpose of this report is to present a type of root malformation which seems to have gone unreported so far.

Study design. Two patients showing severely dysplastic roots of all permanent first molars were evaluated using radiography, histology as well as scanning and transmission electron microscopy.

Results. Medical histories of the patients revealed significant, but diverse events in the first year after birth. Radiographically the pulp cavity floors of the affected molars in large part were occupied by ectopic mineralized plates. Microscopically these plates consisted of hard tissue, densely calcified globules, and a network of canals which contained large blood vessels and were lined by cementum and periodontal ligament.

Conclusions. We propose that the ectopic mineralized plate was derived from the dental follicle, had developed during crown formation around the vascular plexus at the base of the dental papilla, and represented a mechanical obstacle interfering with normal root development. (Oral Surg Oral Med Oral Pathol Oral Radiol 2014;117:e311-e319)

Short or absent tooth roots most frequently result from hard tissue resorption as a sequel to dento-periodontal traumas, orthodontic tooth movements, local inflammatory processes, tumors, and endocrine disorders.^{1,2} More rarely, however, radicular dysplasias can also occur as a developmental disorder involving either a primary disruption or premature arrest of normal root formation. Primary disruption of radicular development is associated with dentin dysplasia type I³⁻⁵ and regional odontodysplasia.^{6,7} In these conditions, the root dysplasia is generalized or affects a continuous section of a dental arch. Premature arrest of radicular development results from local events such as trauma, infection, radiation therapy, systemic influences like chemotherapy,⁸⁻¹⁰ and sometimes without any apparent reason.¹¹ Therefore, this type of dysplasia usually affects individual teeth or a specific group of teeth, the roots of which were forming at the time of the external insult.

The aim of this report is to present a type of root malformation detected in two unrelated patients, which does not fit in any of the above categories and seems to have gone unreported so far.

MATERIALS AND METHODS

Patients

Two patients presenting a radiographically very similar, severe root malformation of all four permanent first

molars were detected incidentally. The first patient, a boy born in Germany of Macedonian parents, was referred to a pediatric dentist at the age of 8 years 1 month for the treatment of deep caries of primary and permanent teeth. At that time, the boy was healthy, but the mother reported that at the age of 9 months he had been admitted to a hospital because of a severe infection of the left thigh. Medical records obtained from the clinic revealed an osteomyelitis of the left femur head with involvement of the soft tissues due to an infection with *Staphylococcus aureus*. The in-patient treatment consisted of intravenous administration of a combination of a cephalosporine (Cefuroxim; Sandoz Pharmaceuticals, Rotkreuz, Switzerland) for 5 days, a penicillin (Flucloxacillin; OrPha Swiss, Küssnacht, Switzerland) for 5 weeks, a lincosamide (Clindamycin; Pfizer, Zurich, Switzerland) for 5 weeks, and a glycopeptide antibiotic (Vancomycin; Pfizer) for 4 weeks. Recovery after dismissal from the hospital was free of complications, and several follow-up examinations over almost 2 years did not disclose any permanent damage of the hip joint.

In addition to bite-wing radiographs, the pediatric dentist exposed a panoramic radiograph to assure that all permanent teeth were present. The first permanent molars were extracted and the primary molars restored under general anesthesia. With informed consent of the parents, the extracted teeth were examined by X-ray microtomography (microCT), light microscopy (LM), backscattered scanning (BSE), and transmission electron microscopy (TEM).

The second patient, a girl of Swiss descent, was 10.5 years old when a panoramic radiograph was taken for orthodontic treatment planning disclosed the root malformation of all permanent first molars. The medical history obtained from the father revealed that

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a relatively advanced astrocytoma of the mother, diagnosed during the 34th week of pregnancy, had necessitated a premature delivery by a Caesarean section in the 36th week. Thereafter, treatment of the tumor with oral doses of a corticosteroid was initiated, but the mother insisted on breast-feeding her daughter. Two months later she had to stop breast-feeding, and when the child was 3 years old, the mother died. Further medical history of the girl only revealed relatively frequent middle ear infections which started at about 2 years of age and were mostly treated with a combination of amoxicillin and clavulanic acid (Augmentin; GlaxoSmithKline, Münchenbuchsee, Switzerland) orally for no longer than 1 week.

After noting the abnormal roots, cone beam computed tomography (CBCT) scans of all permanent first molars were taken at the age of 13 years 3 months. Using a Veraviewepocs 3D device (J. Morita MFG Corp., Dietzenbach, Germany), four cylindrical volumes with a diameter of 42.625 mm and a height of 43.0 mm were scanned at a slice thickness of 1.5 mm and a slice interval of 1.0 mm. Three-dimensional reconstructions were made at a voxel size of 0.125 mm using the proprietary i-Dixel One Volume Viewer V2.0 (J. Morita MFG Corp., Dietzenbach, Germany). In addition, image data of individual slices in bmp-format were imported in the program VGStudio Max (Volume Graphics, Heidelberg, Germany), which was used to manually segment the various hard tissues and, thus, to isolate the teeth and view their roots.

MicroCT evaluation and histologic processing

All four permanent molars removed from patient 1 were fixed in a mixture of 4% paraformaldehyde and 0.2% glutaraldehyde in 0.1 M phosphate buffer (pH 7.2). Following a rinse in 0.185 M Na-cacodylate buffer (pH 7.2), they were photographed using a M420 microscope (Leica Microsystems, Heerbrugg, Switzerland) and scanned in a μ CT 40 microtomograph (Scanco Medical, Brüttisellen, Switzerland). Slices of 20 μ m in thickness were imaged at a resolution of 1024 \times 1024 px. After image data in raw-format had been imported in VGStudio Max, the radiographs were manually segmented, distinguishing enamel, dentin, pulp cavity, and root canals as well as an ectopic hard tissue plate at the level of the cemento-enamel junction (CEJ). By applying color codes to and varying the transparency of these components, it was possible to visualize their natural spatial relationship.

Following the microCT evaluation, teeth were divided in two halves using a diamond band saw (EXAKT, Norderstedt, Germany). Maxillary molars were cut in a bucco-lingual and mandibular molars in a mesio-distal direction. One half of each specimen was left fully mineralized and processed for examination in

the scanning electron microscope (SEM), whereas the other half was decalcified for 2 months in 10% EDTA containing 0.2% glutaraldehyde and processed for examination in the LM or TEM.

LM evaluation

Decalcified specimens from patient 1 were dehydrated in ethanol, transferred to xylol, and embedded in methyl-methacrylate (MMA). Serial sections were cut at a thickness of 5-7 μ m using a Leica RM2255 microtome (Biosystems, Nunningen, Switzerland) and tungsten-carbide knives. Sections were deplasticized and stained with toluidine blue, resorcin-fuchsin, or alcian blue (pH 2.5)-nuclear fast red. Overview micrographs were recorded with the M420 microscope and a ProgRes C14 + camera (Jenoptik, Jena, Germany), while a DM 6000 B microscope equipped with a DFC 420 C camera (Leica Microsystems) served for preparing detail micrographs.

SEM evaluation

Fully mineralized tooth halves from patient 1 were dehydrated in ethanol and embedded in Technovit 7200 VLC (Heraeus Kulzer, Wehrheim, Germany). Light-polymerized blocks were mounted on aluminum stubs, polished, and coated with a 10-15 nm thick layer of carbon.¹² Thereafter, they were examined using a Tescan VEGA TS5316 XM SEM (Tescan, Brno, Czech Republic) operated in BSE mode. Micrographs were recorded at 20 kV and a working distance of 23 mm. In addition, mineral densities of the ectopic hard tissue plates and, as an internal control, of enamel and dentin were estimated based on BSE signal intensities.¹² Finally, the elemental composition of the same sites was analyzed with the aid of energy-dispersive X-ray spectroscopy (EDS). A Si(Li) detector (Oxford Instruments, Wiesbaden, Germany) served for recording EDS spectra using an accelerating voltage of 7 kV, a working distance of 23 mm, and a counting time of 100 s. For the quantitative evaluation of these spectra, the Inca energy software (Oxford Instruments) was used. Both mineral densities and elemental compositions obtained from the various locations were compared using analyses of variance with repeated measurements contained in the SPSS software V20 (IBM, Armonk, NY, USA).

TEM evaluation

Decalcified tooth halves from patient 1 were further cut into smaller pieces comprising particularly the ectopic hard tissue plate. These blocks were postfixated in 1.33% Os-tetroxide in 0.067 M s-collidine buffer for 2 h at room temperature. Thereafter, they were dehydrated in ethanol, transferred to propylene oxide, and embedded in Epon 812 (Fluka, Buchs, Switzerland). From the resin blocks, thin sections of 80-100 nm in thickness

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