*Bull Group Int Rech Sci Stomatol Odontol*. 53(1): 33-36 (2016)

# ORIGINAL RESEARCH ARTICLES

## MINERAL DENSITY OF HYPOMINERALISED AND SOUND ENAMEL

**E. Garot[1,2], P. Rouas[1,2], E. d'Incau[1,2], N. Lenoir[3], D. Manton[4], C. Couture[2]**

[1]Univ. de Bordeaux, UFR des Sciences Odontologiques, Bordeaux, France [2]Univ. de Bordeaux, PACEA, UMR 5199, Pessac, France [3]PLACAMAT, UMS 3626 CNRS-Univ. de Bordeaux, Pessac, France [4]Melbourne Dental School, University of Melbourne, Melbourne, Victoria, Australia

**Key words**
Molar incisor hypomineralisation, X-ray microtomography, enamel mineral density, characterisation.

**Introduction**
The term Molar Incisor Hypomineralisation (MIH) was first proposed in 2001 by Weerheijm (1). MIH is a demarcated qualitative defect of enamel of systemic origin, affecting one or more first permanent molars, often affecting the permanent incisors (2). The affected teeth show clearly demarcated enamel opacities of normal thickness with a smooth surface and white, yellow or brown in colour (2). Internationally, the current MIH prevalence ranges between 3 and 44% with an average of 15%, which, considering its aesthetic and functional consequences, make it a major public health problem (3). Currently, the aetiology of MIH is not clearly elucidated, but researchers agree that it is likely to be multifactorial in origin (4). MIH consists of an alteration in the action of the ameloblasts during the maturation phase (5). Authors determined the mineral density of hypomineralised enamel in permanent teeth by means of X-ray MicroComputed Tomography (XMCT) (6-10). Here, from our preliminary results, we aimed to distinguish hypomineralised enamel from sound enamel by mean of XMCT with a sample including eight hypomineralised teeth.

**Materials and Methods**
The tooth material consisted of a convenience sample of eight permanent first molars, collected from seven patients with a clinical diagnosis of MIH, extracted as part of an orthodontic treatment plan at the Teaching Hospitals of Bordeaux. Diagnosis of MIH was made by one of two experienced paediatric dentists.
X-ray Micro-Computed Tomography (XMCT) was performed on the MIH teeth. The material was digitised using high resolution microtomography at the laboratory PLACAMAT (UMS 3626) in Bordeaux (Microtomograph X GE V/TOME/X S equipment). The scanning parameters were 120 kV, 147 $\mu$A for the x-ray tube, an exposure time of 500ms with four integrations per projection, 2550 projections/360° and a copper filter of 0.1 mm was placed on the source in order to reduce beam hardening artefacts. The voxel size was 7x7x7 µm3. The final volume was reconstructed in 16-bit. The microCT images were compiled using software Avizo® 7.0.1 (FEI, OR, USA). Measures were performed at the same coronal heights. This requirement is because in the permanent dentition the occlusal enamel is more mineralised than the cervical enamel (11, 12). Thus, the most apical plane of section through the cervix that shows a continuous ring of enamel was located, and the images were then recompiled according to this plane (13). Five

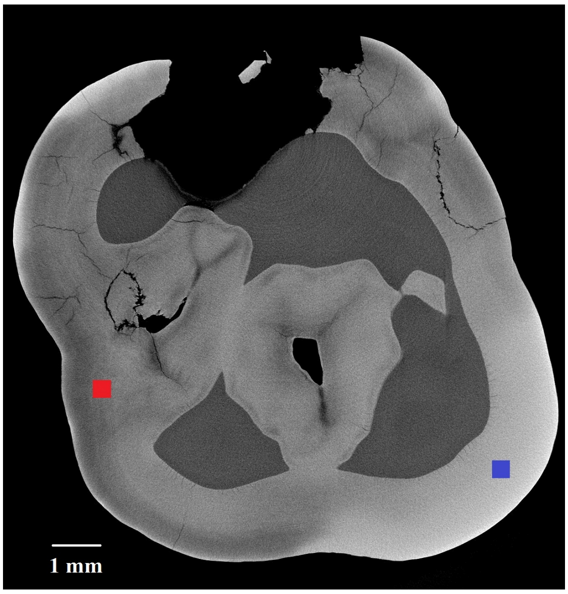


Figure 1: Image from micromotomographic analysis of a MIH first permanent molar. The grey level average was calculated for each square in hypomineralised enamel (dark area) and normal enamel (light area). A red square is placed in the centre of the hypomineralised enamel and a blue square is placed in normal enamel at the same distance from the enamel surface.

slides including an enamel defect were selected. The average of grey levels contained in a square (1 mm2) was calculated in hypomineralised and normal enamel for each slide. This square is placed in the centre of the defect and at the same distance to the enamel surface in normal enamel (Figure 1). This procedure was performed using Plot Profile ImageJ® 1.45 software (NIH, USA).

Non-parametric statistical tests (Wilcoxon test for paired samples) were carried out from Statistica® software Package Version 7.1 software (Statsoft® Dell, OK, USA) to compare data from these two areas (stained and normal).

The research protocol (DC-2015-2415) was registered at the "Comité de Protection des Personnes Sud-Ouest et Outre Mer III" and the "Ministère de la recherche". Data were anonymised.

## Results

The mean of five measures per area (hypomineralised and normal) for each tooth are presented in Table 1. The relative grey scale values were markedly higher in normal enamel compared to hypomineralised enamel with an average difference of 9148 (SD=2455). There was a mineral density reduction of 19%. The Wilcoxon test for paired samples was conducted for 80 measures in hypomineralised and normal enamel. The test showed that mineral

| a) | MIH area | | Normal area | | Difference |
|---|---|---|---|---|---|
| | Mean (GS) | SD (GS) | Mean (GS) | SD (GS) | |
| MIH_1 | 40879 | 2081 | 51060 | 155 | 10181 |
| MIH_2 | 28616 | 189 | 34797 | 698 | 6182 |
| MIH_3 | 38503 | 1161 | 52253 | 619 | 13751 |
| MIH_4 | 47150 | 194 | 53278 | 883 | 6128 |
| MIH_5 | 47454 | 710 | 55260 | 735 | 7806 |
| MIH_6 | 42847 | 888 | 56133 | 485 | 13287 |
| MIH_7 | 47533 | 678 | 54190 | 539 | 6657 |
| MIH_8 | 26546 | 626 | 35743 | 1198 | 9197 |

| b) | | |
|---|---|---|
| | Mean | 9148 |
| | SD | 2455 |
| | p value | 3.57E-08* |

Table 1: a) Mean and standard deviation (SD) of five measures performed per area (normal and hypomineralised) for each MIH tooth. b) Results from the Wilcoxon test for paired samples performed (p value) between the 80 measures calculated in the hypomineralised and normal enamel.(GS : Grey Scale; * : statistically significant).

density was significantly lower in hypomineralised enamel than it was for normal enamel ($p<0.0001$;Table 1).

## Discussion

The present microtomographic analyses of hypomineralised enamel showed a reduction (mean 19%) in mineral density. To our knowledge, the mineral density of MIH enamel has been the subject of five studies using back-scattered electron or x-ray microcomputed tomography (6, 7, 9, 10, 14-16). The samples of reviewed studies included from 2 to 15 teeth affected by MIH. In all studies, a decrease in mineral density was shown in hypomineralised enamel versus unaffected enamel (6-10, 14, 16). From these five studies, on average, a reduction in mineral density of 18% was calculated. Studies by Farah et al. included the largest sample set analysed by microcomputed tomography (10 MIH teeth) (9, 15). Parameters vary depending on studies, for example, previous studies used a voxel size of 15$\mu$m3 whereas in our study a higher resolution was obtained (7$\mu m^3$). Currently, authors perform analyses in 2D, however 3D analyses should be used. 2D data analysis leads to a loss of dimensional information compared to 3D (17). A standardisation of the methodology with clearly defined parameters is necessary to perform meta-analyses.

## Conclusions

The present study of hypomineralised teeth has shown a significantly decrease in mineral density in hypomineralised enamel compared to normal enamel.

## Acknowledgements

This work was supported by the Institut Français de Recherche Odontologique (IFRO).

## Bibliography

(1) Weerheijm KL, Jalevik B, Alaluusua S. Molar-incisor hypomineralisation. Caries Research 2001;35:390-1.

(2) 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. European Journal of Paediatric Dentistry 2003;4:110-3.

(3) Elfrink MEC, Ghanim A, Manton DJ, Weerheijm KL. Standardised studies on Molar Incisor Hypomineralisation (MIH) and Hypomineralised Second Primary Molars (HSPM): a need. Eur Arch Paediatr Dent 2015;16:247-55.

(4) Alaluusua S. Aetiology of Molar-Incisor Hypomineralisation: A systematic review. Eur Arch Paediatr Dent 2010;11:53-8.

(5) Jalevik B, Dietz W, Noren JG. Scanning electron micrograph analysis of hypomineralized enamel in permanent first molars. International Journal of Paediatric Dentistry 2005;15:233-40.

(6) Fagrell T. Molar incisor hypomineralization. Morphological and chemical aspects, onset and possible etiological factors. Swedish Dental Journal Supplement 2011:5, 11-83.

(7) Fagrell TG, Salmon P, Melin L, Noren JG. Onset of molar incisor hypomineralization (MIH). Swedish Dental Journal 2013;37:61-70.

(8) Farah RA, Monk BC, Swain MV, Drummond BK. Protein content of molar-incisor hypomineralisation enamel. Journal of dentistry 2010;38:591-6.

(9) Farah RA, Swain MV, Drummond BK, Cook R, Atieh M. Mineral density of hypomineralised enamel. Journal of Dentistry 2010;38:50-8.

(10) 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. British Dental Journal 2004;196:634-8; discussion 25.

(11) Theuns HM, van Dijk JW, Jongebloed WL, Groeneveld A. The mineral content of human enamel studied by polarizing microscopy, microradiography and scanning electron microscopy. Archives of Oral Biology 1983;28:797-803.

(12) Wilson PR, Beynon AD. Mineralization differences between human deciduous and permanent enamel measured by quantitative microradiography. Archives of Oral Biology 1989;34:85-8.

(13) Olejniczak AJ, Smith TM, Feeney RN, Macchiarelli R, Mazurier A, Bondioli L, et al. Dental tissue proportions and enamel thickness in Neandertal and modern human molars. Journal of Human Evolution 2008;55:12-23.

(14) Mahoney EK, Rohanizadeh R, Ismail FS, Kilpatrick NM, Swain MV. Mechanical properties and microstructure of hypomineralised enamel of permanent teeth. Biomaterials 2004;25:5091-100.

(15) Farah R, Drummond B, Swain M, Williams S. Linking the clinical presentation of molar-incisor hypomineralisation to its mineral density. International Journal of Paediatric Dentistry 2010;20:353-60.

(16) Crombie FA, Manton DJ, Palamara JE, Zalizniak I, Cochrane NJ, Reynolds EC. Characterisation of developmentally hypomineralised human enamel. Journal of Dentistry 2013;41:611-8.

(17) Cocker D. An investigation into the characteristics of dentine associated with Molar Incisor Hypomineralisation [thesis]: University of Melbourne; 2014.