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# Comparison of demineralisation rates in pre- and postnatal enamel and at the neonatal line<sup>☆</sup>

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## ABSTRACT

**Aims:** The neonatal line, which is an exaggerated incremental layer line, separates pre- and postnatal enamel. It has been suggested that this layer may be a barrier to the progress of a carious lesions. The objective was to measure the rate of demineralisation in pre- and postnatal enamel and within the neonatal layer using scanning microradiography (SMR). Permanent enamel and compressed permeable hydroxyapatite samples were used as controls.

**Methods and results:** Enamel specimens from deciduous incisors were cut into mesiodistal blocks of 2 mm thickness without altering the labial surface and located within SMR cells. Permanent enamel and hydroxyapatite specimens were similarly prepared. Artificial caries-like lesions were created by exposing the specimens to 0.1 mol l<sup>-1</sup> acetic acid (pH 4.0) within the SMR cells. SMR was used to measure the rate of mineral loss at 10 points either side of and at the neonatal line in the deciduous enamel, and 20 points across in the control specimens.

The rate of demineralisation was almost the same in pre- and postnatal enamel ((6.0–8.0) × 10<sup>-4</sup> g cm<sup>-2</sup> h<sup>-1</sup>), but much lower in the vicinity of the neonatal line (2.0 × 10<sup>-4</sup> g cm<sup>-2</sup> h<sup>-1</sup>). The rate of demineralisation was lower in permanent (5.0 × 10<sup>-4</sup> g cm<sup>-2</sup> h<sup>-1</sup>) than in deciduous enamel, and even lower in the permeable hydroxyapatite specimen (2.5 × 10<sup>-4</sup> g cm<sup>-2</sup> h<sup>-1</sup>).

**Conclusions:** This study showed no difference in the rate of demineralisation between pre- and postnatal enamel, but a reduced rate within the region that contained the plane of the neonatal line. This supports the hypothesis that the neonatal line may act as a barrier to the propagation of carious lesions.

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## 1. Introduction

Enamel comprises a mineral phase and an organic matrix. The organic matrix is secreted first, but, unlike the other calcified

tissues, begins to mineralise within minutes to produce partially mineralised, soft, immature enamel. Mineral content increases by thickening the initial crystallites: this occurs continuously, both during and after the periods in which more

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tissue layers are added. When mature, the fully formed enamel attains greater than 96% of mineral content by weight. All calcified tissues lose water, which is the space which can be occupied by mineral, but a further striking difference between enamel and the other mineralised tissues is that over 90% of the initially secreted protein matrix is lost during calcification, and that which remains forms envelopes around individual crystals. There is, finally, a higher residual content of organic matter at places where an abrupt change in crystal orientation occurs<sup>1</sup>: these boundary discontinuities form at the junctions of the walls with the floor of the pit made by each ameloblast<sup>1–4</sup>: they are most commonly and incorrectly called the ‘prism sheath’, a term which is correctly applied to a structure which develops around the boundary when enamel is partially demineralised.<sup>4</sup>

Most of human dental enamel is one continuous material: it is only partially subdivided by the boundary discontinuities<sup>1–3</sup> and these have to join by disruption of the continuous juxta-parallel-crystallite phase to create separate ‘prisms’ – the latter not existing as such in the intact and entire tissue.<sup>2</sup> Human enamel belongs to Pattern 3, in which the imaginary ‘prisms’ have a ‘keyhole’ pattern (Pattern 3) in transverse section. The boundaries reflect the path of movement of the individual secretory ameloblasts during the time course of the formation of the tissue. Because formative ameloblasts move relative to one another within the sheet of cells, the (prism) boundaries follow contrasting curved paths in traversing from the enamel–dentine junction (EDJ) to the surface of enamel. The overall path of one ‘prism’ resembles a flattened helix.<sup>3</sup>

In other mammals, most of the enamel may be truly subdivided into separate ‘prisms’ by a continuous interprismatic phase (of interameloblastic pit wall origin). This is obvious in cross sections of Pattern 1 enamel where the prism boundary discontinuities form closed cylinders. To cleave this type of enamel, it is again necessary to break a continuous juxta-parallel-crystallite phase. In Pattern 2 enamel, rows of prisms are separated by continuous inter-row sheets of interprismatic enamel: this type of enamel can break by failure within the boundary discontinuity planes.<sup>5</sup>

The width of the ameloblastic pit in the surface of the developing enamel tissue varies with time of day at the expense of the intervening interameloblastic pit wall phase: this gives the boundaries their varicosities<sup>4</sup> as the ‘prism’ gets wider and narrower along its length. The wider parts correspond to faster and the constrictions to the slower phases of amelogenesis: i.e., during the slower phases, the ‘prism body’ (that part secreted by the end of the ameloblast contacting the floor of the pit) gets thinner at the expense of its tail (the interameloblastic pit wall phase). Cross striations are the corresponding periodic features seen by light microscopy at regular intervals of approximately 4 µm (range 2–8 µm). As such, they reflect the diurnal rhythmicity in prism formation, and the organisation of crystallites within the rod, and matching expansion and contraction of the prism ‘body’ (pit floor origin) and prism ‘tail’ (interameloblastic origin, interprismatic) regions and differences in the refractive index of the enamel mineral due to variations in its net composition, especially with respect to the concentration of carbonate.<sup>6</sup>

Exaggerated varicosities or cross striations are formed at periods of relative quiescence during rhythmic deposition.

These form incremental lines (or striae of Retzius) which can be seen, for example, by optical microscopy of ground sections as lines. They reflect variations in structure – principally in the location of the boundary discontinuity – and mineralisation. Regular Retzius lines in more superficial enamel recognised by a slight kinking of the prism boundary discontinuity are normal. Better marked lines which form a different pattern within every individual but the same pattern in all of the enamel forming in the same individual at the same time reflect systemic disturbances of enamel formation. All incremental lines demonstrate the plane of the developing enamel surface at a given moment in the secretory history of the tooth. Because primary teeth have relatively extensive developmental surfaces, the striae in primary enamel are almost parallel with the enamel surface. Perikymata or imbrication lines are the sites where the striae reach the completed tooth surface.<sup>1</sup>

All structural differences within enamel affect the rate of progress of carious lesions, and become more marked as a consequence.<sup>7</sup> The primary carious attack appears to be along the boundary discontinuities (‘the long axis of the prisms’) but is channelled laterally and along the direction of the striae by the more and less porous and more and less resistant layers of the striae of Retzius complex. The striae of Retzius frequently appear to constitute relative barriers to the advance of the carious process towards the centre of the enamel.<sup>7</sup>

### 1.1. The neonatal line

The enamel of all the deciduous teeth and first permanent molars develops partly before and partly after birth. The boundary between the two fractions of enamel is marked by a prominent and exaggerated incremental plane known as the neonatal line, first described by Rushton<sup>8</sup> who named it the birth line. The term neonatal line or ring was used by Schour<sup>9</sup> and he, and later Rushton,<sup>10</sup> considered the line to be a pronounced incremental line formed at birth or shortly afterwards. It clearly appears to be the result of an abrupt change in the environment and nutrition of the newborn infant. Its presence of course makes it possible to distinguish prenatal from postnatal enamel. In teeth from normal infants, the neonatal line extends from the enamel–dentine junction in the cervical third of the deciduous central incisors towards the enamel surface. In the lateral incisors and the canines, the neonatal line is positioned more to the incisal part of the enamel. Attempts to determine the structural nature of this and other incremental lines have produced slightly conflicting results. Rushton<sup>8</sup> suggested that the ‘interprismatic substance’ in the line is either more or less highly mineralised with respect to the prisms than elsewhere, but later opinion was that the enamel prisms became more highly mineralised as they crossed the neonatal line.<sup>10</sup> Sognnaes,<sup>11</sup> working on demineralised sections of teeth, concluded that the neonatal line is due to thickening of the organic prism boundaries, resulting in a beaded appearance of the prisms. Gustafson and Gustafson<sup>12</sup> considered the line to be a type of Retzius line produced by bending of the prisms or disturbances in their formation. The neonatal line is a region of hypomineralisation. Using microradiography, Crabb,<sup>13</sup> and later Allan,<sup>14</sup> reported radiolucency of the line. Weber and Eisenmann,<sup>15</sup>

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