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Prenatal factors associated with the neonatal line thickness in human deciduous incisors

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ABSTRACT

The neonatal line (NNL) is used to distinguish developmental events observed in enamel which occurred before and after birth. However, there are few studies reporting relationship between the characteristics of the NNL and factors affecting prenatal conditions.

The aim of the study was to determine prenatal factors that may influence the NNL thickness in human deciduous teeth. The material consisted of longitudinal ground sections of 60 modern human deciduous incisors obtained from full-term healthy children with reported birth histories and prenatal factors. All teeth were sectioned in the labio-lingual plane using diamond blade (Buechler IsoMet 1000). Final specimens were observed using scanning electron microscopy at magnifications 320×. For each tooth, linear measurements of the NNL thickness were taken on its labial surface at the three levels from the cemento–enamel junction.

The difference in the neonatal line thickness between tooth types and between males and females was statistically significant. A multiple regression analyses confirmed influence of two variables on the NNL thickness standardised on tooth type and the children's sex (*z*-score values). These variables are the taking of an antispasmodic medicine by the mother during pregnancy and the season of the child's birth. These two variables together explain nearly 17% of the variability of the NNL. Children of mothers taking a spasmolytic medicine during pregnancy were characterised by a thinner NNL compared with children whose mothers did not take such

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medication. Children born in summer and spring had a thinner NNL than children born in winter. These results indicate that the prenatal environment significantly contributes to the thickness of the NNL influencing the pace of reaching the post-delivery homeostasis by the newborn's organism.

Introduction

The neonatal line (NNL), observed for the first time by Rushton in 1933, is present on all teeth undergoing mineralisation process in the prenatal period (Antoine et al., 2009; Molnar and Ward, 1975). This line is present on all deciduous teeth and on 10% of permanent first molars (Skinner and Dupras, 1993). The neonatal line is the first hypomineralised layer of enamel laid down after birth, corresponding to a stria of Retzius (Hillson, 2014; Skinner, 1992). It is characterised by a marked change in the direction of enamel prisms. It extends from the enamel–dentine junction in the cervical part of the tooth through the whole thickness of the enamel and it separates the enamel formed prenatally from that formed postnatally (Smith and Avishai, 2005; Whittaker and Richard, 1978). The average width of NNL in children born naturally, observed with optical or scanning microscopy, is about 12 μm (Eli et al., 1989; Sabel et al., 2008). In contrast, Skinner (1992) reports that the assessment of normal width NNL varies from 30 μm while using a light microscope to 10–16 μm using microradiography (Whittaker and Richard, 1978).

Formation of the neonatal line is closely associated with perinatal stress understood as a change of conditions of the living environment resulting from the "transition" from the intrauterine environment to the external environment and, as a result of the change in the quality and manner of nutrition (Molnar and Ward, 1975; Szpringer-Nodzak, 2005). The perinatal mechanism of the neonatal line formation is related to the levels of calcium ions and of 1,25-$(OH)_2$ vitamin D_3 (calcitriol) available for the mineralising enamel (Chan et al., 1978; Norén, 1983; Ranggård, 1994; Seow, 1991).

Determination of the position of NNL is the first step in all analyses regarding characteristics of the enamel formed in the prenatal and postnatal parts of ontogeny (Antoine et al., 2009; Beynon et al., 1998; Birch and Dean, 2009, 2014; Eli et al., 1989; Mahoney, 2011, 2012; Sabel et al., 2008; Seow et al., 2005; Smith and Avishai, 2005; Żądzińska et al., 2013).

Therefore, the presence of the neonatal line is used as a simple marker of a child's live birth and his/her survival of a post-birth period, necessary for the formation of a postnatal enamel layer permitting an observation of the neonatal line as a border between the prenatal and postnatal enamel (Scheuer and Black, 2000; Whittaker and Richard, 1978). Usually 6–10 days of postnatal life are necessary to form NNL (Smith and Avishai, 2005). For that reason, the neonatal line is most often used for forensic purposes in order to determine a child's age at the moment of death and to assess whether a child was stillborn or was possibly the victim of infanticide (Janardhanan et al., 2011; Lewis, 2007; Skinner and Anderson, 1991; Skinner and Dupras, 1993). The presence and assessment of the NNL thickness is also of crucial importance in bioarchaeological and palaeoanthropological contexts for the interpretation of birth conditions as well as for the assessment of pre- and perinatal environments of past populations (FitzGerald et al., 2006; Macchiarelli et al., 2006; Schwartz et al., 2010; Zanolli et al., 2012).

Besides articles reporting the fact of existence of NNL in the tooth enamel there are also analyses focused on the explanation of NNL variability in relation to prenatal factors and perinatal conditions, such as the state of health of the pregnant women (Norén et al., 1978; Norén, 1984), the newborn's birth weight (Norén, 1983), the duration of pregnancy (Skinner and Dupras, 1993; Zanolli et al., 2011), and the type of delivery (Eli et al., 1989). Results of those analyses are often contradictory, and are usually based on observations of preterm, small for gestational age and/or low-birth-weight children.

Although intrauterine disturbances (malnutrition, vitamin D and vitamin A deficiencies, medicines such as tetracyclines, antimalarian or antiepileptic drugs used by pregnant women) were found to affect human deciduous enamel (e.g. Avery, 2002; Jacobsen et al., 2013; Kiukkonen, 2006; Mobley and