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ARTICLE INFO

Article history: Received 12 September 2014 Received in revised form 20 November 2014 Accepted 22 November 2014 Available online 29 November 2014 Keywords: Aging Dentin Dynamic mechanical analysis (DMA) Elastic modulus Nanoindentation

ABSTRACT

An experimental evaluation of human coronal dentin was performed using nanoscopic dynamic mechanical analysis (nanoDMA). The primary objectives were to quantify any unique changes in mechanical behavior of intertubular and peritubular dentin with age, and to evaluate the microstructure and mechanical behavior of the mineral deposited within the lumens. Specimens of coronal dentin were evaluated by nanoDMA using single indents and in scanning mode via scanning probe microscopy. Results showed that there were no significant differences in the storage modulus or complex modulus between the two age groups (18–25 versus 54–83 yrs) for either the intertubular or peritubular tissue. However, there were significant differences in the dampening behavior between the young and old dentin, as represented in the loss modulus and tan δ responses. For both the intertubular and peritubular components, the capacity for dampening was significantly lower in the old group. Scanning based nanoDMA showed that the tubules of old dentin exhibit a gradient in elastic behavior, with decrease in elastic modulus from the cuff to the center of tubules filled with newly deposited mineral.

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^{*}The authors acknowledge support from the National Institutes of Health (NIDCR R01 DE016904 and NIDCR R01 DE015306-10) and the National Science Foundation (BES 0521467).

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http://dx.doi.org/10.1016/j.jmbbm.2014.11.021 1751-6161/© 2014 Elsevier Ltd. All rights reserved.

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1. Introduction

Dentin occupies the majority of each tooth by both weight and volume, and exhibits a complex hierarchical structure that consists of both organic and inorganic components (Nanci, 2008). Mature dentin is composed of approximately 70% mineral (i.e. carbonated apatite), 20% organic materials (primarily type I collagen) and 10% water by weight (Summitt and dos Santos, 2006). The most distinct feature of its microstructure is the network of tubules that extend outward from the pulp towards the Dentin-Enamel Junction (DEJ). Each tubule lumen is surrounded by a peritubular cuff, which consists of a hyper-mineralized collagen-poor region (approx. 0.5 to 1 µm in thickness) of apatite crystals (Nanci, 2008; Xu and Wang, 2012). Intertubular dentin occupies the region between the tubules and consists of an organic matrix (collagen fibrils) reinforced by nanoscopic apatite crystals similar to that of peritubular dentin (Marshall et al., 1997; Weiner et al., 1999; Kinney et al., 2003). Due to the differences in composition between the peritubular and intertubular components, and striking presence of the dentin tubules in microscopic evaluations, dentin is often considered a biological composite.

There are two characteristics of human dentin that are clearly evident in microscopic evaluations of the microstructure. First, there are spatial variations in the tubule density; the density decreases from roughly 60k lumens/ mm² near the pulp to 10k lumens/mm² at the DEJ (Pashley, 1989). Second, the microstructure of human dentin is dependent on age of the individual. There is a reduction in the tubule lumen diameter (and increase in thickness of the peritubular cuffs) with increasing age. The process begins in the third decade of life and appears to progress at a constant rate until the complete occlusion of the lumens (Arola, 2008). After the majority of tubule lumens are filled with mineral, the tissue appears translucent and is regarded as "sclerotic" (Nanci, 2008). As a result of the aforementioned process, human dentin undergoes an increase in mineralization with age (Weber, 1974; Vasiliadis et al., 1983; Kinney et al., 2005; Porter et al., 2005). This is directly opposite to that occurring in human cancellous bone, which undergoes a decrease in mineral content with age (Smith et al., 1975; Kanis et al., 1994). The comparison to cortical bone is more difficult as it undergoes an increase in mineral content with age during development to adulthood, and then either decreases with further aging (Boskey and Coleman, 2010) or develops a plateau (Akkus et al., 2003). Variations in findings appear to be associated with the exact location of bone being evaluated and what age range is being considered.

Owing to the spatial variations in microstructure of dentin within the tooth crown, the hardness (Pashley et al., 1985), strength (Giannini et al., 2004; Ryou et al., 2011) and resistance to crack growth (Ivancik et al., 2011; Ivancik and Arola, 2013) all decrease with increasing proximity to the pulp. And according to the changes in microstructure of dentin with age, similar changes would be expected in its mechanical behavior. Indeed, there is a decrease in the strength of dentin with patient age. That process appears to begin near the end of the third decade of life, and proceeds at approximately 20 MPa per decade (Arola and Reprogel, 2005). There is also a substantial reduction in the fatigue strength (Kinney et al., 2005; Ivancik et al., 2011), fatigue crack growth resistance (Bajaj et al., 2006; Ivancik et al., 2012) and fracture toughness (Koester et al., 2008; Nazari et al., 2009) with age. In comparing the fatigue crack growth behavior, there is nearly a 100-fold increase in incremental growth rate within coronal dentin from young adulthood to an age of \geq 50 years (Bajaj et al., 2006).

Spatial variations in strength and toughness of dentin do not appear to be reflected in the properties of the individual constituents. Kinney et al. (1996) reported that while the hardness of the intertubular dentin is substantially lower near the pulp than the DEJ, the hardness and elastic modulus of the peritubular dentin are essentially independent of depth. They attributed previous gradients in micro-hardness from the pulp to the DEJ to arise from the changes in intertubular dentin, not the tubule density. Nevertheless, that study was conducted on dehydrated tissue, and as expected, the indentation responses were found to be absent of viscous (i.e. dampening) behavior. Similar discrepancies are found in nanoscopic evaluations of the constituent properties and the influence of aging. For example, Balooch et al. (2001) evaluated the hardness and elastic modulus of the intertubular and peritubular dentin of normal and sclerotic tissue using atomic force microscopy. No significant difference was noted between the properties of normal and sclerotic tissue. Yet, Zheng et al. (2005) reported that the hardness of the sclerotic dentin beneath the cusps in old teeth was significantly greater than that of sound dentin from young teeth. Similarly, Senawongse et al. (2006) found that 'aged' dentin exhibited higher hardness and elastic modulus than young dentin, but in the mantle region only (within $5 \mu m$ from the DEJ). The two latter studies were conducted on moist sections, but not hydrated, which could influence the capacity for dampening behavior. The aforementioned studies did not evaluate the properties of the peritubular and intertubular dentin independently. Thus, it is not possible to know which of the two constituents was responsible for the unique mechanical behavior of old dentin. Nanoscopic dynamic mechanical analysis (nanoDMA) has been proven effective for characterizing the mechanical properties of hydrated dentin (Balooch et al., 1998; Ryou et al., 2012), but has not yet been applied to understand the effects of aging on the structural behavior.

In this investigation, nanoDMA was used to evaluate the mechanical behavior of human dentin, and quantify the changes that take place with aging. In the present study, the complex, storage and loss moduli were evaluated for the intertubular and peritubular dentin of teeth from a group of senior donors. These responses were compared to those from a similar evaluation of dentin that was conducted on a group of young donor teeth. The primary objective of the investigation was to identify if there are significant changes to the mechanical behavior of intertubular and peritubular dentin with age. The test null hypothesis was that there are no differences in the mechanical behavior of inter- and peritubular dentin with age.

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