Investigations on the dynamics of water in the macrostructural dentine

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National University of Singapore Biophotonics Laboratory Faculty of Dentistry Singapore 119074, Republic of Singapore Abstract. The purpose of this study was twofold: (1) to investigate the nature and degree of water loss at 21°C, 60% relative humidity (dehydration) and at 105°C (desiccation), and to relate these findings with (2) the strains produced in the dentine structure during dehydration and rehydration processes. In stage 1, digital moiré interferometry (DMI) was used to study the strain distribution pattern during dehydration and rehydration at 21 °C. In stage 2, the nature and degree of water loss was determined using gravimetric analysis and nuclear magnetic resonance spectroscopy. DMI showed that dehydration produced strains in the dentine structure after an initial latent period. Gravimetric analysis showed that dentine exhibited an initial rapid water-loss phase followed by a slow and steady water-loss phase. Though the major portion of water loss occurred in the initial 2 h of dehydration (rapid water-loss phase), no obvious strains were produced during this period. Rehydration lead to the major reversal of dehydration-induced water loss and strains in dentine. Heating at 105°C resulted in further substantial loss of water from dentine. These experiments highlighted that the free water in the dentine surface, porosities and tubules are lost rapidly and constitute the major water lost when dehydrated at 21°C. © 2006 Society of Photo-Optical Instrumentation Engineers. [DOI: 10.1117/1.2360257]

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1 Introduction

Dentine is said to be a complex, hydrated, porous, biological composite structure consisting of a hydrated matrix of mainly type I collagen (organic phase) that is reinforced by poorly crystalline carbonated hydoxyapatite. The bulk of dentine consists of 50 vol % inorganic phase, 30 vol % organic components, and 20 vol % fluids. An understanding of the structural stability of dentine is imperative to study the effects of endodontic treatment and various restorative procedures on the integrity of the tooth structure. In spite of decades of research, very few studies have been conducted on the association of water with dentine and the role of different types of water in the biomechanical behavior of dentine. The mechanical properties and the structural behavior of dentine are still the least understood aspects in dental biomechanics.

The water content of dentine is believed to vary with location and is typically in the range of 8 to 16% by weight. The general conjecture is that there are two types of water in dentine. One type is associated with the mineral (apatite crystal) and the organic (collageneous and noncollageneous extracellular matrix proteins) fractions and is mostly "tightly"

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bound in nature. The second type is the free or "unbound" water, and this type of water fills the dentinal tubules and other porosities in the dentine matrix. The free water is associated with the inorganic ions such as calcium and phosphate and aids in their transport within the dentine matrix. Free water can be removed by heating at 100°C, but bound water can be substantially removed only by heating at 600°C. However, the precise distribution and effect of different types of water on dentine structure is not yet understood.

Some studies have attributed the increase in fracture susceptibility of endodontically treated teeth to a decrease in moisture content of dentine. This hypothesis was first conceived by G. V. Black, and later confirmed by Helfer et al. (1972), who reported that the moisture content of dentine from endodontically treated teeth was about 9% less than their vital counterparts. Plowever, there are other studies that contradict this view. Papa and Messer (1994) reported an insignificant difference in the moisture content between endodontically treated teeth and vital teeth and emphasized the importance of conserving the bulk of dentine to maintain the structural integrity of postendodontically restored teeth. In another study, Jameson et al. reported that dehydration of dentine at 20°C (50% relative humidity) brought about 30% loss

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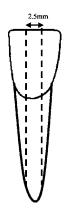


Fig. 1 Schematic diagram to highlight the region of the tooth used as a specimen.

of moisture, and this moisture loss resulted in a significant decrease in the toughness and increase in the stiffness of dentine bars. ^{12,13} The conflicting evidence in these studies was mostly attributed to the difficulty in establishing appropriate controls, comparable test procedures, and lack of direct comparison of root-filled teeth with vital teeth. ^{12,14}

Pashley has suggested that the fluid-filled dentinal tubules can function to hydraulically transfer and dissipate occlusal forces applied on the teeth. 15 Recently, Kishen and Asundi 4 have reported that dehydration at 24°C for 72 h produced obvious changes in the stress-strain behavior of the structural dentine to physiologically relevant compressive forces. They found a stress-strain response characteristic of a tough material in fully hydrated dentine, while dehydration at 24°C for 72 h resulted in a response characteristic of a brittle material in dentine.⁴ The purpose of this study was twofold: (1) to investigate the water-loss (dehydration)-and water-uptake (rehydration)-induced strain distribution patterns in dentine at 21°C using a digital moiré interferometry; (2) to study the nature and degree of water loss at 21°C (dehydration), water uptake at 21°C (rehydration), and water loss at 105°C (desiccation) in dentine. A gravimetric analysis and nuclear magnetic resonance (NMR) spectroscopy is used for this purpose.

2 Experiments

2.1 Experiment 1: Moiré Interferometric Analysis of Dehydration- and Rehydration-Induced Strains in Dentine Structure

2.1.1 Specimen preparation

Four freshly extracted noncarious mandibular central incisor teeth were collected and stored in phosphate buffered saline (PBS) solution (0.1 M phosphate buffer, pH 7.2). These specimens were transilluminated so that teeth with cracks or extraction damage could be excluded. Specimens were prepared by grinding the mesial and distal sides of the teeth specimens on wet emery paper of grit size 400, 800, and 1000, under constant irrigation to obtain midline buccolingual plane-sided, slab-shaped sections $(2.5\pm0.2 \text{ mm})$ thicknesses) of the teeth. These specimens represented the entire faciolingual bulk of the dentine and contained the pulp chamber and the root canal in the core region (Fig. 1).

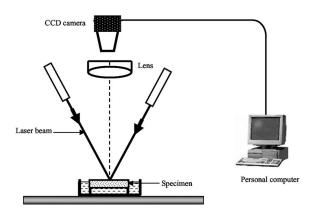


Fig. 2 Schematic diagram of the digital moiré interferometer.

2.1.2 Experiments

Moiré methods utilize diffraction gratings as a deformation sensing element. In moiré interferometry, a grating replicated specimen (specimen grating) was interrogated using a virtual reference grating formed by the interference of two mutually coherent beams incident on the specimen plane at a fixed angle. Moiré fringes result from the interference between the deformed specimen grating and the virtual reference grating. These fringes represent contours of in-plane displacement fields and were analyzed using image processing software as discussed previously. In order to prepare the specimen grating, a high-frequency cross-grating (f=1200 lines/mm) was replicated on a buccolingual surface of the tooth using epoxy adhesive at room temperature. The replication was done in such a way that the grating lines were parallel and perpendicular to the long axis of the tooth. The distance between the adjacent grating lines is referred to as the pitch of the grating, which in this case was $0.833 \mu m$.

The moiré interferometer consisted of two mutually coherent light beams from a diode laser (λ =670 nm) that were incident on the specimen grating at an oblique angle and generated a virtual reference grating of 2400 lines/mm [corresponding to a pitch (P) of 0.417 μ m]. This virtual reference grating interacted with the deformed specimen grating to produce the moiré fringe patterns. A high-resolution chargecoupled device (CCD) camera with a spatial resolution of $753(H) \times 244(V)$ pixels was used to digitize and record the fringe patterns obtained for further analysis (Fig. 2). Moiré fringes represent contours of displacement components in the direction perpendicular to the lines of the grating. ¹⁶ During experiments, (1) the fully hydrated specimen grating was positioned in the moiré interferometer and the real-time dehydration-induced surface deformations along the x axis, known as the U field and that along the y axis, known as the V field were recorded for a period of 12 h. Similarly, (2) a 72-h prior dehydrated specimen grating was positioned in the moiré interferometer and the real-time hydration-induced surface displacements along the x and y axes were recorded. The acquired digital moiré fringe patterns were used to determine the in-plane normal strains along different regions of interest on the cervical portion of the root dentine as shown in Fig. 3. Further details about the moiré interferometry and the calculations of normal stains are presented elsewhere. 17,18

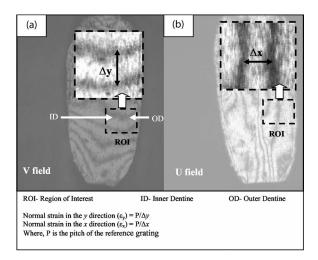


Fig. 3 Shows a typical (a) *V*-field and (b) *U*-field moiré fringe patterns and the calculation of normal strain from the region of interest.

2.2 Experiment 2: Gravimetric Experiments to Determine the Water Loss During Dehydration and Desiccation and Water Regain During Rehydration

2.2.1 Specimen preparation

Twelve freshly extracted single-rooted teeth were collected and stored in PBS solution. These teeth were transilluminated so that teeth with cracks or extraction damage could be excluded. Specimens were prepared by grinding the mesial and distal sides of the teeth specimens on wet emery paper of grit size 400, 800, and 1000, under constant irrigation resulting in the formation of midline buccolingual plane-sided, slabshaped sections $(2.5\pm0.2\text{-mm}$ thick). All the specimens had a thin layer of enamel on the coronal aspect, cementum on facial and lingual sides, and pulp chamber in the center (Fig. 1). The ground surface was polished to a final finish on an automatic polisher with 3- μ m particle size diamond paste. These tooth specimens were used for the gravimetric experiments.

2.2.2 Experiments

During experiments, twelve tooth specimens were allowed to dehydrate at 21°C and 60% relative humidity for 72 h. The rate of weight loss due to dehydration was monitored gravimetrically. The gravimetric measurements were recorded every 15 min for the first 8 h and subsequently after 24, 48, and 72 h intervals. The twelve samples subjected to dehydration were later divided into two groups of equal numbers. Group 1 was subjected to rehydration at 21°C, and group 2 was subjected to desiccation at 105°C. 7.8.19 The weight gain during the rehydration process (group 1) and the weight loss due to desiccation (group 2) were recorded every 15 min for the first 8 h and after 24, 48, and 72 h intervals.

2.3 Experiment 3: NMR Experiments to Quantify the Water Loss During Dehydration and Desiccation, and Water Regain During Hydration

2.3.1 Specimen preparation

Nine freshly extracted mandibular single-rooted teeth were collected and midline buccolingual plane-sided, slab-shaped

sections $(2.5\pm0.2\text{-mm}$ thickness) were prepared by grinding the specimens on wet emery paper of grit size 400, 800, and 1000. The enamel and cementum portions of these specimens were trimmed off using a tapering fissure diamond bur (Shofu, ISO no. 017).

2.3.2 Experiments

These specimens were divided into three equal groups for this study. The specimens in group 1 were subjected to room temperature dehydration at 21 °C (60% relative humidity) for 72 h. The specimens in group 2 were subjected to desiccation at 105°C for 72 h, (7, 8, 19) while specimens in group 3 were kept fully hydrated. The dentine specimens in group 1, group 2, and group 3 were pulverized to fine powder in a mechanical grinder. Total time spent in grinding one group was less than a minute. The fine powder was immediately transferred to a NMR tube and was tested by NMR spectroscopy. The NMR measurements were carried out with a 300-MHz Bruker ACF-300 (Bruker Analytik, GmbH, Rheinstetten, Germany), equipped with a BVT-3000 temperature controlled unit. A dedicated 1-H probe was used. Line width was used to measure the area under the curve. Spectra were collected with a radio frequency of 300.13 MHz, and 16 K data points were sampled in 3.4 s, corresponding to a spectral width of 2.4 kHz. During each measurement, 250 scans were used to record the data.

3 Results

- **3.1** Experiment 1: Moiré Interferometric Analysis of Dehydration- and Rehydration-Induced Strains in Dentine Structure
- **3.1.1** Dehydration-induced strains in dentine structure

Figure 4(a) shows the strain distribution pattern in the dentine (in the direction perpendicular to the dentinal tubules) during dehydration. The fourth-order polynomial trend line was used to fit the data. Notice that the R^2 value is 0.9876 for inner dentine data and 0.9941 for outer dentine data, which is a good fit of the line to the data. The U-field moiré analysis (strain in the direction perpendicular to the dentinal tubules) showed a generalized increase in strains in dentine with dehydration. This dehydration-induced strain response occurred in three stages. In the initial stage, there was no obvious increase in strains with dehydration (stage 1). This stage (stage 1) lasted for the first 4 h, and subsequently, there was a rapid increase in strains with dehydration (stage 2). The stage 2 lasted until 9 h of dehydration. The outer dentine (or peripheral dentine adjacent to the cementum) produced higher and earlier strains in these stages. After 9 h, both inner (core dentine adjacent to the root canal lumen) and the outer dentine tended to behave uniformly with minor increase in strains with dehydration (stage 3).

Figure 4(b) shows the strain distribution pattern in the inner and the outer dentine (in the direction parallel to the dentinal tubules) during dehydration. The fourth-order polynomial trend line was used to fit the data. Notice that the R^2 value is 0.9889 for outer dentine data and 0.9871 for inner dentine data, which is a good fit of the line to the data. The V-field moiré analysis (strains in the direction parallel to the

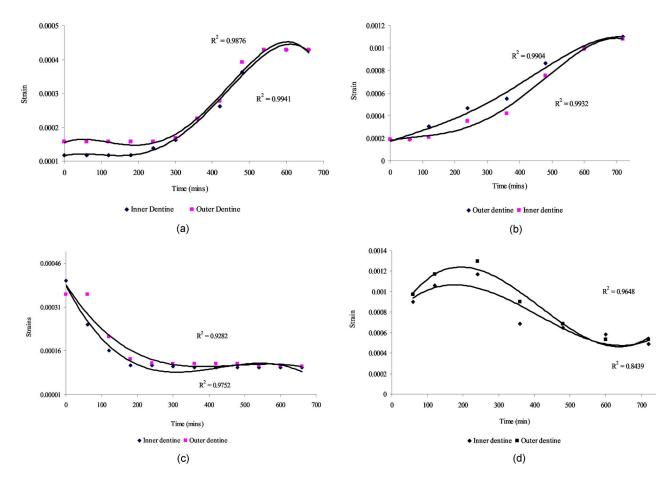


Fig. 4 (a) Dehydration-induced strains at the region of interest (direction perpendicular to the dentinal tubules) in the dentine structure. (b) Dehydration-induced strains at the region of interest (direction parallel to the dentinal tubules) in the dentine structure. (c) Rehydration-induced strains at the region of interest (direction perpendicular to the dentinal tubules) in the dentine structure. (d) Rehydration-induced strains at the region of interest (direction parallel to the dentinal tubules) in the dentine structure.

dentinal tubules) also showed an increase in strains with dehydration in three stages. There was no obvious increase in strains for the first hour of dehydration (stage 1), which was followed by a rapid increase in strains with dehydration until about 10 h (stage 2). During this period, the outer dentine exhibited higher strain than the inner dentine. There was only a minor increase in strains after 10 h of dehydration (stage 3).

3.1.2 *Rehydration-induced strains in dentine structure*

The U-field moiré analysis (strains in the direction perpendicular to the dentinal tubules) displayed a decrease in strains with rehydration, which occurred in two stages [Fig. 4(c)]. The third-order polynomial trend line was used to fit the data. Notice that the R^2 value is 0.9752 for inner dentine data and 0.9282 for outer dentine data. This indicates a good fit of the line to the data. In the stage 1, there was a rapid decrease in strains with rehydration for the first 3 h, which was followed by a steady state in which there was only a minor decrease in strains with rehydration (stage 2). The rate of decrease in strains with rehydration was higher in the inner than the outer dentine.

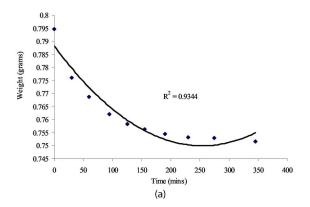
The V-field analysis (strains in the direction parallel to the dentinal tubules) showed strain response in the outer and in-

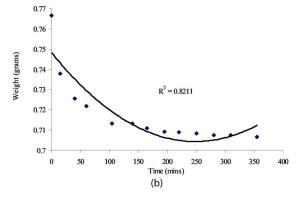
ner dentine upon rehydration in three stages. Figure 4(d) shows the strain distribution in the inner and the outer dentine (in the direction parallel to the dentinal tubules) during rehydration. The third-order polynomial trend line was used to fit the data. Notice that the R^2 value is 0.8439 for inner dentine data and 0.9648 for outer dentine data. This indicates a good fit of the line to the data. It was found that there was an increase in strain immediately upon rehydration (stage 1). This stage lasted for about 4 h, and subsequently, there was a rapid decrease in strains with rehydration for about 10 h (stage 2). Beyond 10 h of rehydration, the dentine structure showed only a minor decrease in strains with rehydration (stage 3).

3.2 Experiment 2: Gravimetric Experiments to Determine the Water Loss During Dehydration and Desiccation and Water Regain During Rehydration

3.2.1 Water loss during dehydration

Figure 5(a) shows the pattern of water loss in dentine when dehydrated at 21° C (Table 1). The second-order polynomial trend line was used to fit the data. Notice that the R^2 value is 0.9344, which is a good fit of the line to the data. Gravimetric





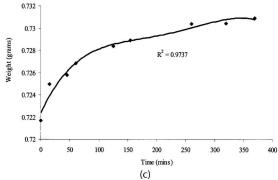


Fig. 5 (a) Dehydration-induced weight loss of the dentine specimen. (b) Desiccation-induced weight loss of the dentine specimen. (c) Rehydration-induced weight gain of the dentine specimen.

analysis of the fully hydrated dentine specimens showed that room temperature dehydration resulted in a biphasic water-loss response. An initial rapid weight-loss phase, which lasted for \sim 2 h, followed by the slow weight-loss phase, during

which the rate of water loss was minimal. These observations were consistent in all the samples (Table 1). The average weight loss due to dehydration after 72 h was 5.3% [standard deviation (SD): 0.61]. Further, it was noted that 81% of the dehydration-induced water loss occurred in the first 2 h of dehydration.

3.2.2 Water loss during desiccation

Figure 5(b) shows the pattern of water loss in dentine when desiccated at $105\,^{\circ}$ C (Table 1). The second-order polynomial trend line was used to fit the data. Notice that the R^2 value is 0.8211, which is a good fit of the line to the data. The high-temperature desiccation also produced a biphasic response in water loss. There was a rapid water-loss phase in the first 2 h of desiccation (stage 1), following which there was a gradual water-loss phase (stage 2). Desiccation of dentine specimens at $105\,^{\circ}$ C accounted for 7.73% (SD: 0.43) weight loss in 72 h. However, the initial rapid water-loss phase or the stage 1 accounted for 86% of the total weight loss.

3.2.3 Water regain during rehydration

Figure 5(c) shows the pattern of water regain in dentine when rehydrated at 21° C. Rehydration of previously dehydrated specimens showed a 99.1% (SD: 1.1) weight gain in 2 weeks (Table 2). The second-order polynomial trend line was used to fit the data. Notice that the R^2 value is 0.9737 and this is a good fit of the line to the data. Rehydration-induced weight gain also had a biphasic response. There was an initial rapid water-regain phase for the first 2 h of rehydration (stage 1). This phased resulted in about 96% of the water regain. The second phase showed a gradual regain of water with rehydration (stage 2).

3.3 Experiment 3: NMR Experiments to Quantify the Water Loss During Dehydration and Desiccation, and Water Regain During Hydration

NMR spectroscopy of the hydrated, dehydrated, and desiccated dentine samples showed a distinct difference in their water content. Though the spectral width, peak width, and the peak height was determined from each experiment, the peak height was used for this analysis. The average peak height of water for hydrated dentine specimens were at 7232, while that of dehydrated and desiccated dentine specimens were at 4972 and 1312, respectively (Fig. 6). If the water content in the hydrated dentine is considered to be 100%, then the water loss by dehydration process was calculated to be 31%, while the water loss by desiccation process was calculated as 50.61%.

Table 1 Shows the dehydration- and dessciation-induced weight loss in the dentine specimens.

	Dehydration at 21°C and 60% relative humidity			Desiccation at 105°C in a desiccator			
Specimens	Water loss after 2 h (%)	Water loss after 24 h (%)	Water loss after 72 h (%)	Water loss after 2 h (%)	Water loss after 24 h (%)	Water loss after 72 h (%)	Total
Average	4.33	5.31	5.30	6.72	7.48	7.73	13.03
Standard deviation	0.40	0.60	0.61	0.29	0.44	0.43	0.645

Table 2 Shows the rehydration-induced weight gain in the previously dehydrated dentine specimens.

Specimens	Water gain after 2 h (%)	Water gain after 24 h (%)	Water gain after 48 h (%)	Water gain after 72 h (%)	Water gain after 2 weeks (%)
Average	96.35	97.3	97.8	98.2	99.1
Standard deviation	0.5	0.4	0.1	0.3	1.1

The total water loss by dehydration and desiccation was then 81% of the total water content. Even after 72 h of desiccation at 105°C, 18.14% of the total water content was found to remain in dentine.

4 Discussion

Gravimetric analysis of the fully hydrated dentine specimens showed that dehydration caused a biphasic response in water loss. There was an initial phase of rapid water loss, followed by a phase in which slower water loss occurred. The rapid water loss phase occurred in the first 2 h of dehydration and accounted for 80% of the total water loss during dehydration. Further substantial water loss occurred only when the specimens were subjected to high-temperature desiccation at 105°C. Desiccation at 105°C also showed a biphasic response in water loss. Biphasic trends in the pattern of water loss under similar experimental conditions were also reported by Jameson et al. 13 However, in their experiment, 85% of the total water loss during dehydration took place in the first 30 min. This decrease in the time taken to achieve peak water loss can be attributed to the type of specimens employed. The specimens employed in their experiments were thin dentine bars with a significant number of patent dentinal tubules, while in our study we used larger slab-shaped faciolingual sections with thin impermeable enamel and cementum surrounding the facial and lingual surfaces. It was suggested that the initial rapid water loss was due to evaporation of water from the dentinal tubules and the dentine surface, while the

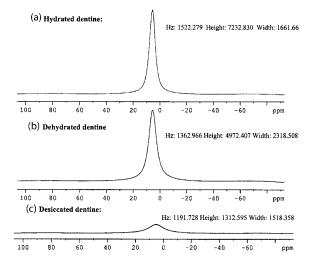


Fig. 6 Typical NMR spectra obtained from (a) hydrated dentine, (b) dehydrated dentine, and (c) desiccated dentine.

slow water loss in the second phase occurred when movement of water to the surface becomes a rate-limiting factor. ¹³

Moiré interferometry is a highly sensitive, whole-field nondestructive optical technique that can be used to study moisture-loss-induced deformations on dentine.¹⁸ It was found from this analysis that the water-loss-induced strains occurred in the direction perpendicular and parallel to the dentinal tubules in three stages. In stage 1, there was minimal increase in strain with dehydration. This was followed by stage 2, in which there was a rapid increase in strain with dehydration, and finally in stage 3, there was no obvious increase in strain with dehydration. When the findings from the gravimetric analysis and the moiré interferometry were compared, it was interesting to note that the maximum water loss in dentine occurred in the first 2 to 4 h (rapid water-loss phase), while the water-loss-induced strains were produced in the dentine after 1 h (in the direction parallel to the dentinal tubules) and after 4 h (in the direction perpendicular to the dentinal tubules). This difference in the rate of water loss and nature of water-loss-induced strains indicates that the initial rapid water loss during dehydration may be due to the loss of free water from the dentinal tubules and the dentine surface, and this water loss did not produce any obvious strains in the dentine structure. However, after the major loss of such free water, there will be movement of free water from the dentine matrix to the surface resulting in strains in dentine matrix. This effectively means that it is the movement and loss of free water from the intertubular dentine matrix that brings about increase in strains in dentine structure. Further, the initial onset and larger strains were produced in the direction parallel to the dentinal tubules. This may be because of the large dehydration-induced water movement and water loss in this direction. Similar, moisture-loss-induced strain gradients and compressive deformation of the dentine sections were reported previously. 16,18

Rehydration of dentine specimens also displayed changes in strain distribution in the directions parallel and perpendicular to the dentinal tubules. Although strains in the direction parallel to the dentinal tubules were lower, there were three stages in the strain response. In stage 1, there was an increase in strain with rehydration for the initial 4 h. Following which, there was a rapid decrease in strains with rehydration (stage 2). In stage 3, there was no obvious decrease in strains with rehydration. In the direction perpendicular to the dentinal tubules, there were only two stages in the strain response. There was an initial rapid decrease in strains for the first 3 h of rehydration (stage 1), following which, there was no obvious decrease in strains (stage 2). The initial increase in strains in the direction parallel to the tubules could be due to the swelling of previously dehydrated collagen fibrils. Moreover, it

was observed that more than 96% of water is regained in the first 4 h of rehydration, and the moiré interferometry showed conspicuous reduction in strains after 4 h of rehydration. Rehydration for 3 weeks resulted in 99% water regain, and this observation corresponded with earlier investigations. ¹³

In this study, NMR spectroscopy was conducted to quantify the water content found in the hydrated, dehydrated, and desiccated dentine samples. It was found from NMR spectroscopy that a total of 80% of the water content was lost from dehydration and desiccation together. In which, 30% of the water loss occurred during dehydration, while 50% of water loss occurred during desiccation. Also, 19% of water was found to remain in the dentine matrix even after desiccation for 72 h. When 13% of the absolute water loss observed in the gravimetric analysis was compared with 80% of the relative water loss observed in the NMR analysis, the total water content of the dentine can be assumed as $\sim 16.25\%$ by weight, and this value falls within the reported range of 8 to 16% by weight. Based on the gravimetric analysis, the water loss by dehydration was 5.3% of dentine weight, which was 32% of the total water present in the bulk dentine. This amount of water loss during dehydration corresponded to the value suggested in the previous work.¹³ Furthermore, the gravimetric analysis showed that the water loss due to desiccations was 7.73% of dentine weight, which was about 47% of the total water present in the bulk dentine. These values of water loss concurred well with the percentage of water loss determined from the NMR spectroscopy.

These experiments highlighted that 30% of the free water from dentine was lost rapidly, as soon as the dentine was exposed to 21 °C and 60% relative humidity. This early water loss did not induce obvious strains in the dentine. Once the major portion of the free water was lost, further movement and loss of water occurred from the intertubular dentine matrix and this process produced strains in the dentine matrix. Our earlier photomechanical experiments conducted on similar samples (planoparallel sagittal sections) have demonstrated reduction in the toughness of dentine after 72 h dehydration.⁴ The present investigation, which is a continuation of the earlier work, suggests that only 30% of the total water content was lost during this process of dehydration and this water is lost mainly from the dentinal surface, porosities, and dentinal tubules. Prior experiments have demonstrated the difference between the denaturation temperature of collagen in nonmineralized and mineralized tissues, indicating the protective role of minerals on the denaturation of collagen. ²⁰ Fourier transfer infrared spectroscopic analysis also did not show any obvious difference in the molecular structure between the hydrated and the dehydrated dentine specimens.²¹ Accordingly, it can be inferred from these experiments that the loss of free water from the dentinal surface, porosities, and dentinal tubules can compromise the fracture resistance of bulk dentine.

It is understood that the dental pulp tissue is made up of a connective tissue system consisting of cells and fibers both embedded in the extracellular matrix. The proteins of the extracellular matrix have very high water-holding properties. The water content of the pulp is approximately 90%. ²² During root canal treatment, the pulp tissue is extirpated, and the root canal is disinfected and dehydrated before obturation. The

loss of water-rich pulp tissue may be responsible for the loss of mechanical integrity in endodontically treated teeth. However, further experiments are indicated to understand the role of specific type of water on the mechanical characteristics of bulk dentine.

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