Optical Hardness Meter for Diagnosis of Dental Root Caries

Hisanao Hazama*
Graduate School of Engineering, The University of Osaka,
2-1 Yamadaoka, Suita, Osaka, Japan 565-0871

ABSTRACT

A method to quantitatively evaluate the progress of root caries in a clinical setting is strongly desired. The root caries could be diagnosed by measuring hardness because dentin becomes softer as the caries progresses. Vickers hardness, which has been customarily used as an indicator of tooth hardness, cannot be measured *in vivo* because the teeth must be polished and dried prior to measurement to make the indentation. A hardness meter using an indenter with light for tooth monitoring (HAMILTOM) is proposed to measure hardness of teeth in a wet condition. An area appearing black without a total internal reflection at an indenter of HAMILTOM increases as a dentin becomes softer. Bovine dentin samples with different hardnesses were used, and the correlation between the dark areas measured by HAMILTOM and the Vickers hardness measured by the Vickers hardness tester was evaluated. Twenty bovine dentins were demineralized by a lactic acid solution with different times and divided into groups 1 and 2 of 10 samples each. Group 1 was used to obtain a calibration curve to calculate Vickers hardness from the dark area. Group 2 was used to validate the calibration curve. The Vickers hardness of group 2 calculated by the dark areas of group 2 and the calibration curve obtained in group 1 and the Vickers hardness of group 2 measured by the Vickers hardness tester were strongly correlated with a determination coefficient of 0.99. The results demonstrate that HAMILTOM may be a suitable alternative to the conventional method.

Keywords: dentin, hardness, root caries, demineralization, light-emitting diode, total internal reflection

1. INTRODUCTION

The world's population is simultaneously increasing and aging. According to the WHO, one in six individuals will be 60 years or older by 2030, and the world's population of people more than 60 years of age will reach 2.1 billion. Japan is a country with a remarkable aging population. Currently, more than one in four people are 65 years or older, and this is expected to increase to one in three by 2030. As life expectancy increases, people retain their teeth for longer. Root caries is a dental disease that affects more than one in three persons in the geriatric population. Root caries is commonly found on the exposed root surfaces or the margin of the cementoenamel junction. Root caries is defined as a cavitation below the cementoenamel junction not including the adjacent enamel. It is usually discolored, softened, ill-defined, and involves both cementum and underlying dentin. Enamel is stronger and more acid-resistant than any other dental tissue since it contains about 90 % minerals. In comparison, cementum and dentin are composed of about 45–50 % and 70 % inorganic materials, respectively. In addition, the higher content of magnesium and carbonate makes cementum and dentin more soluble than enamel. Therefore, dentin on a root surface is not only more likely to develop and progress into caries than enamel but also more likely to cause tooth loss. The management of root caries is an important issue in the aging society because the risk of caries is higher as the number of untreated teeth increases. Root caries is diagnosed by inspection and palpation. Indicators include the color, surface texture, and hardness of the lesion. Although the diagnosis is based on inspection and palpation, there is no clear color change in the initial root caries, and palpation is performed using a probe. Hence, the diagnosis is qualitative. Hardness testing is an indirect method to track changes in the mineral content of dentin. As a caries progresses, dentin becomes softer, but the diagnosis using a probe depends on the dentist's sensitivity to pressure changes when inserting and removing the probe. In fact, the kappa statistics for the inspection and palpation for root caries are as low as 30–51 %. Therefore, a method to objectively and quantitatively evaluate the activity and progress of root caries in a clinical setting is strongly desired. A quantitative evaluation of the hardness of dentin may realize a more accurate assessment of the progression of root caries. Vickers or Knoop hardness has been customarily used as an indicator of tooth hardness. The Vickers hardness and Knoop hardness tests evaluate hardness by measuring the size of indentations after pressing an indenter of a quadrangular pyramid with a constant load on the target object. However, the sample must be dried prior to measurement because an indentation does not remain in samples with a large elastic deformation such as a

* hazama-h@see.eng.osaka-u.ac.jp; phone 81 6 6879 7895

carious tooth. Practically, a hardness measurement cannot be performed on an *in vivo* tooth as the tooth must be extracted prior to the evaluation.

A device is proposed to easily measure the hardness of *in vivo* teeth using a light-emitting diode (LED).[1–3] The device is named "HAMILTOM," which stands for "hardness meter using indenter with light for tooth monitoring." HAMILTOM quantifies the hardness of dentin from the contact projection area (dark area) between the indenter and dentin when the indenter is pressed into the dentin. Here the basic principle of HAMILTOM is demonstrated using bovine dentin samples with different demineralization times. The dark areas are measured by HAMILTOM, and the correlation with the Vickers hardness measured by a conventional Vickers hardness tester was evaluated.

2. PRINCIPLE

Figure 1 shows a schematic illustration of HAMILTOM. HAMILTOM is a device that measures the hardness of dentin from the dark area, which is the contact area between the indenter and the dentin. HAMILTOM includes an optical system composed of an LED with a 455-nm center wavelength, a film diffuser (#17-682, Edmund Optics, USA), a beam splitter (#47-007, Edmund Optics), a transparent conical glass indenter with a 90-deg apex angle (#49-397, Edmund Optics), a lens with a 30-mm focal length (#45-134, Edmund Optics), and a CMOS camera (ID1MB-MDL-U, iDule, Japan) in a lens barrel. It should be noted that the purchased glass indenter originally had an aluminum mirror coating, and the coating was removed by immersing the indenter in hydrochloric acid before use. The LED light passing through the film diffuser was incident on the indenter. The reflected light was reflected toward the camera by the beam splitter, and the image of the tip of the glass indenter was projected on the CMOS camera using a lens with about threefold magnification. Out of the lens barrel, a capacitive load sensor with an 8-mm diameter and 0.3-mm thickness [SingleTact S8-1N, Pressure Profile Systems (PPS), USA], a load sensor extension cable (SingleTact Tail-Extender, PPS), and an I^2C digital interface board (SingleTact Standard Electronics, PPS) were placed in a handpiece housing. By applying a load to the tip of the indenter, the lens barrel rotates around the rotation axis. Then the lens barrel and the housing applied the load to the load sensor. The load applied to the sensor was continuously monitored on a tablet PC (Surface Go 2, Microsoft, USA) through the I^2C digital interface board and a microcontroller (PSoC 5LP, Cypress Semiconductor, USA). An image of the indenter when the load reached the set value was acquired automatically by the CMOS camera using in-house software.

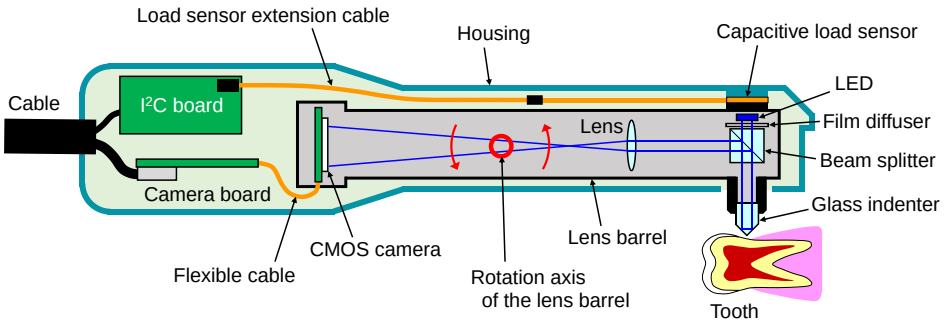

Figure 1. Schematic illustration of the HAMILTOM handpiece. Image of the tip of the glass indenter is projected onto the CMOS camera with a threefold magnification using light from the LED. By applying a load to the tip of the indenter, the lens barrel rotates around the rotation axis, and the lens barrel and the housing apply the load to the load sensor. The CMOS acquires an image of the indenter when the load reaches the set value.

Figure 2 shows the measurement principle of HAMILTOM. When the indenter is not in contact with the dentin, the indenter appears bright because a total internal reflection occurs at the boundary between the glass indenter and the air. In contrast, when the indenter comes into contact with the dentin, a total internal reflection does not occur. Thus the indenter at the contact area appears dark because the refractive index of N-BK7 (1.52), which is the material of the glass indenter, and that of dentin (1.54) are close. For a constant contact load, the dark area should be larger when the dentin is softer. Therefore, the hardness of the dentin can be assessed by measuring the dark area when a constant load is applied to the tip of the indenter. Dentin becomes softer as caries progresses. Hence the dark area should indicate the degree of caries progression because the dark area should increase compared to sound dentin. It is necessary to emphasize that the dark areas are not the size of the indentation and the influence of the water exuded by the contact of the indenter is significant, and it is considered that the indentation does not remain due to the elastic recovery of the dentin after removing the indenter.

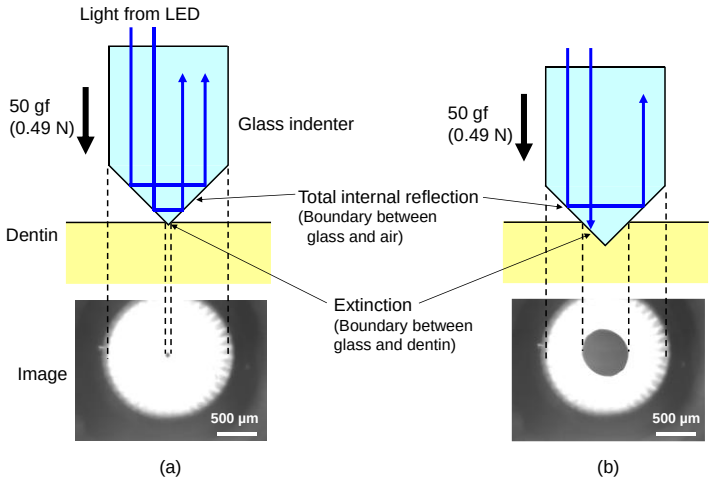

Figure 2. Principle of measuring the dark area between the glass indenter and dentin. When the indenter comes into contact with dentin, a total internal reflection of LED light does not occur, and the indenter at the contact area appears dark because the refractive indices of the glass indenter and dentin are similar. (a) Dark area for sound dentin, which is hard, is small at a constant load of 50 gf (~0.49 N). (b) The dark area for demineralized dentin is larger for a given load because the demineralized dentin is softer than sound dentin.

3. MATERIALS AND METHODS

3.1 Sample preparation

Twenty sound bovine dentins provided from the Osaka Dental University were used to evaluate the correlation between dark areas measured by HAMILTOM and Vickers hardness. Simulated models with various hardnesses were prepared by demineralizing sound dentin samples for different times to create caries. This study was performed after obtaining approval from the Animal Care and Use Committee, Osaka Dental University (Approval No. 19-12001). Preparation includes cutting, the sample embedding in epoxy resin, curing for at least 24 h, and mirror polishing of the sample surface. First, the extracted bovine tooth was cut perpendicular to the running direction of the dentinal tubules. The cut dentin samples were ~20 × 10 × 1 mm^3. To protect the dentinal tubules from the epoxy resin solution, a manicure was applied to the back and all sides of the dentin samples prior to embedding. Second, the dentin samples were embedded in an epoxy resin (Crystal Resin, NISSIN RESIN, Japan) using silicone molds. The samples were cured for at least 24 h, and then they were removed from the molds. Third, the sample surfaces were mirror polished under water injection using waterproof abrasive papers #400, 800, and 2000 (Kohnan, Japan) and a lapping film #8000 (LF1D, Thorlabs, USA) to create a horizontal hardness measurement surface. The sample tilt was adjusted to 1 deg or less during polishing for the sample orientation of 90 deg to the indenter. In addition, ultrasonic cleaning was performed for 2 min using a desktop ultrasonic cleaner (B2210, Branson Ultrasonic, USA) to remove the resin pieces and the inorganic components of dentin generated by polishing from the dentinal tubules. The 20 dentin samples were divided into two groups of 10 samples each (groups 1 and 2). The caries models were prepared by artificial demineralization. The dentin samples were immersed in a lactic acid solution. Lactic acid (20006-75, Nacalai Tesque, Japan) and distilled water were mixed in a beaker to prepare 1 L of 0.1 M demineralization solution. The beaker was placed in a constant temperature water bath (TM-3A, AS ONE, Japan) to hold the solution temperature at 37 °C. The solution was stirred with a stirrer (HE-16GA, KPI, Japan) at the rotation speed of 1000 rpm to maintain a pH of 2. Next, each dentin sample was immersed in the solution for a predetermined time. The demineralization time was set to 0, 0.25, 0.5, 0.75, 1, 2, 4, 6, 12, or 24 h. After demineralization, the samples were washed with tap water and stored in saline (Otsuka Pharmaceutical, Japan) at 4 °C.

3.2 Measurement with HAMILTOM

The dark areas were measured using HAMILTOM for the dentin sample at each demineralization time. Prior to each measurement, the dentin sample was immersed in saline for at least 24 h to make it moist. HAMILTOM was fixed so that the tip of the indenter was oriented vertically downward. The excess surface moisture was blown off with air, and the dentin sample was placed on a stage so that it was located directly below the indenter. The threshold of the load sensor when calculating the dark area was set to 50 gf (~0.49 N). The stage was manually raised in the vertical direction. The

CMOS camera acquired an image once the load reached the specified value. The load was also measured by an electronic balance (ACS-5000, AS ONE, Japan) to confirm the accuracy of the load measured by the load sensor. Before the experiment, a coefficient to be multiplied to the measured value of the load sensor was adjusted to match the load measured with the electronic balance. After setting the dentin sample on the electronic balance, the electronic balance was reset to zero each time because the weight of each sample was slightly different. Images of the dark area were obtained at three different positions for each dentin sample. After each measurement, a lens cleaning paper (EK1546027S, Tiffen, USA) with ethanol was used to remove the water and deposits on the surface of the glass indenter. To calculate the dark area, a reference image was obtained before the indenter was brought into contact with the sample. When each reference image was acquired, the load measured with the load sensor was reset to zero to cancel the long-term drift of the sensor. Next, an image was acquired after the indenter was brought into contact with the sample. Then the reference was subtracted. A binarized image was created using a threshold of 50 % of the maximum brightness of the subtracted image. In the binarized image, the contact area appears as bright pixels. Finally, the dark area was calculated from the number of the bright pixels in the binarized image.

3.3 Measurement with Vickers hardness tester

The Vickers hardness was measured using a Vickers hardness tester (HMV-G30S, Shimadzu, Japan) for each sample, which had different demineralization times. Conventionally, the Vickers hardness is measured in dry conditions because indentations do not clearly remain on a sample with a large elasticity such as demineralized dentin. The dentin samples in this study were dried in a 23 °C indoor environment at 25 % humidity for 18 h before the hardness measurements. The test load was set to 500 gf (~4.9 N). The load holding time was 10 s, and indentations were made on the dentin samples. The Vickers hardness was calculated by measuring the length of the diagonal line of the indentation. The indentation was observed with the microscope of the hardness tester. The Vickers hardness of each sample was measured at three different positions. The measurement points were within a 5-mm diameter with respect to the measurement points with HAMILTOM.

4. RESULTS AND DISCUSSION

Figure 3 shows typical images of the reflected light from the indenter when measuring the dark areas of the samples in group 1. When the dark areas were measured, the load values measured by the electronic balance were 46 to 54 gf. The surface quality of the glass indenter did not change in the repeated measurement in this study because the threshold of the load sensor was set to 50 gf for reducing the burden on the indenter. The areas appearing black without a total internal reflection of the indenter increased as the demineralization time increased for both groups. In the images of the reflected light from the indenter, any dentinal tubules were not observed. It is considered that there was no influence of the porosity of the dentin to the dark area.

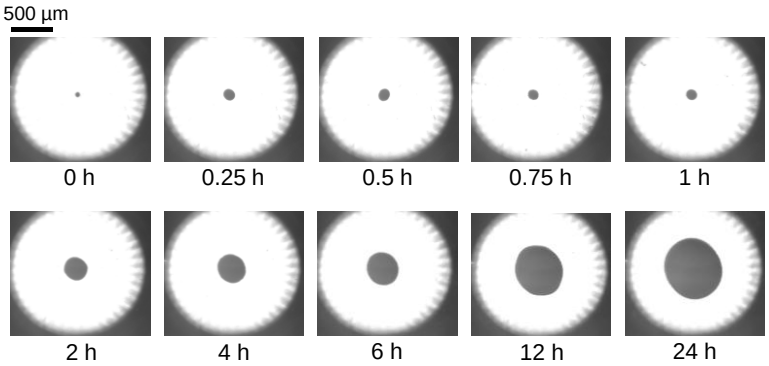

Figure 3. Typical images of reflected light from the glass indenter taken with the CMOS camera. Images for group 1 dentin samples with different demineralization times (0, 0.25, 0.5, 0.75, 1, 2, 4, 6, 12, or 24 h) are shown.

Figure 4 shows the relationship between the dark area A and the Vickers hardness H_V for group 1 with different demineralization times. As the demineralization time increased, the dark areas increased. Additionally, the Vickers hardness decreased to less than half of that of the sound dentin at a demineralization time of 6 h. Evaluating the correlation between the dark areas and the Vickers hardness using a power approximation gave a determination coefficient of 0.96.

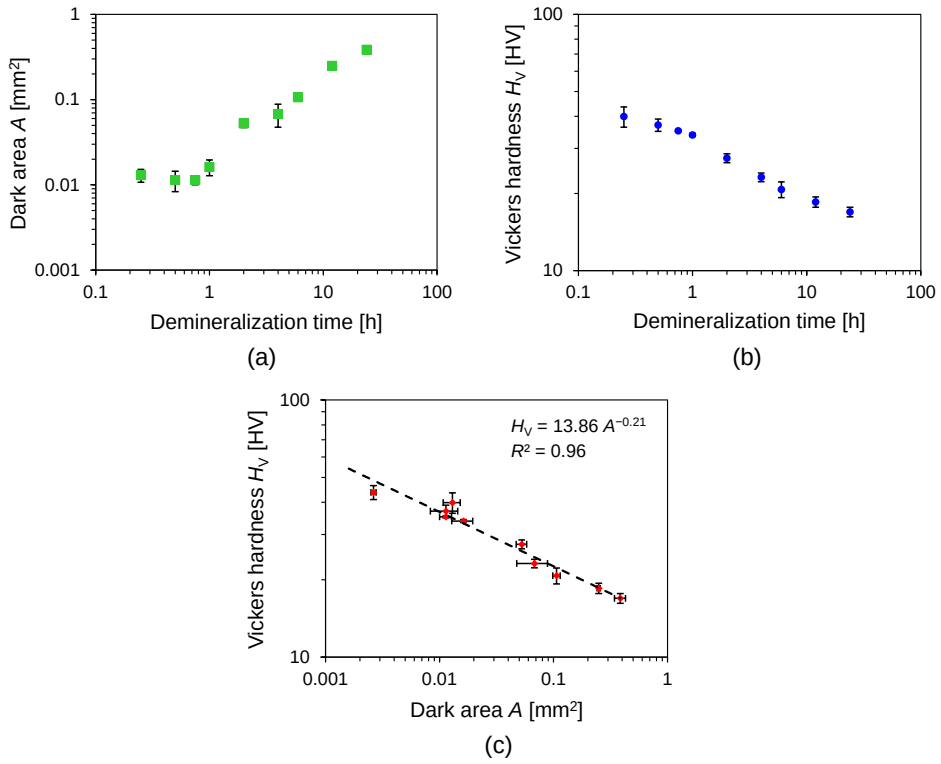

Figure 4. Dark area, Vickers hardness, and their relationship for the samples in group 1. Each graph shows the mean and standard deviation of three measurements. (a) Changes in the dark area with different demineralization times. (b) Changes in the Vickers hardness with different demineralization times. (c) Relationship between the dark area and the Vickers hardness. Dashed line shows the approximate expression. A determination coefficient is also shown.

Figure 5 shows the relationship between the Vickers hardness of group 2 calculated by the dark areas of group 2 and the calibration curve obtained in group 1 and the Vickers hardness of group 2 measured by the Vickers hardness tester. There is a strong correlation with a determination coefficient of 0.99. Furthermore, *p*-value of Pearson's correlation coefficient was 3×10^{-7}, and the *p*-value was smaller than the significance level of 5%. Therefore, it was suggested that the correlation shown in Fig. 5 was significant and the Vickers hardness can be calculated by measuring the dark areas using HAMILTOM.

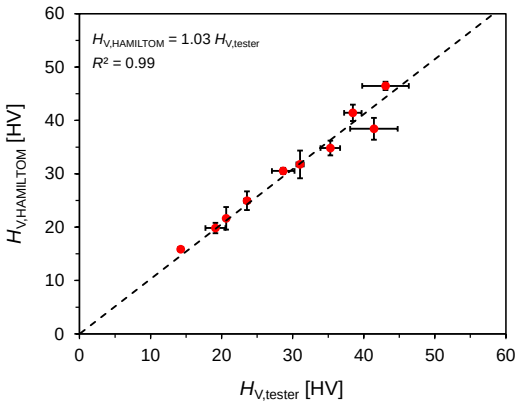

Figure 5. Relationship between the Vickers hardness $H_{V,tester}$ of the samples in group 2 measured with the Vickers hardness tester and the Vickers hardness $H_{V,HAMILTOM}$ calculated by the dark area of the samples in group 2 using the calibration curve obtained from the samples in group 1. The mean and standard deviation of three measurements, approximate expression (dashed line), and determination coefficient are shown.

5. CONCLUSION

The dark areas of bovine dentins with various demineralization times were evaluated using HAMILTOM. Next, the correlation between the dark area measured by HAMILTOM and the Vickers hardness measured by the Vickers hardness tester was evaluated. As the demineralization time increases, the areas appearing black without a total internal reflection of the indenter increase. Additionally, the Vickers hardness of group 2 calculated by the dark areas of group 2 and the calibration curve obtained in group 1 and the Vickers hardness of group 2 measured by the Vickers hardness tester are strongly correlated with a determination coefficient of 0.99. The results demonstrate that HAMILTOM may be a suitable alternative to the conventional method. Unlike the conventional method, which cannot be used for *in vivo* teeth, HAMILTOM holds potential to quantitatively evaluate the progress of caries in *in vivo* teeth. In the future, HAMILTOM may become a new hardness diagnostic method for root caries.

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