

657

Use of the Reflectance Confocal Microscopy to Measure Enamel Demineralization. IRENE PETROU\*, CYNTHIA M. MURPHY, and RICHARD J. SULLIVAN  
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A method was developed to quantify demineralization in dental enamel using confocal laser scanning microscopy (CLSM) in the reflection mode. A validation study was conducted to show that mineral loss measured by confocal microscopy correlates with measurements made by surface microhardness. Twelve bovine enamel blocks, approximately 5x5x1 mm, were polished until the front and back surfaces were parallel. The enamel surfaces were then covered halfway with an acid-resistant varnish to provide a sound and demineralized surface. The blocks were demineralized by immersing them in a hydroxyapatite-saturated, acetic acid demineralization solution for 0, 1, 3, 5, or 16 hours. The varnish was removed after demineralization, and then reflectance and microhardness measurements were made on both halves of each block, sound and demineralized. Enamel demineralization was expressed as mean loss in reflectance for CLSM and mean loss in hardness for microhardness in comparison to sound enamel. The results of the study showed that the loss in reflectance after 1, 3, 5, and 16 hours were 144.59 (a), 288.28 (b), 429.89 (b), and 496.93 (b), respectively. The corresponding losses in hardness were 27.29 (a), 20.86 (a), 58.70 (b), and 57.25 (b) for the same time points. (Different letters indicate statistically different groups.) A least squares analysis comparing demineralization measured by reflectance confocal microscopy and surface microhardness was consistent with a linear fit ( $r^2 = 0.86$ ). **In conclusion, the results of this study show that CLSM in the reflectance mode can be used to quantify enamel demineralization. This method was shown to correlate well with an established surface microhardness methodology for measuring demineralization.**

658

Influence of Enamel Thickness on Quantification of White-spot using Laser-fluorescence. M. ANDO\*, B.R. SCHEMORH, G.J. ECKERT, D.T. ZERO, G.K. STOOKEY  
(Indiana University School of Dentistry, Indianapolis, IN, USA).

The objective of this study was to evaluate the effect of enamel thickness on the quantification of enamel demineralization with laser-induced fluorescence technique. One hundred twenty 3-mm diameter human enamel specimens with flat polished surfaces were used. The enamel surfaces were gently blotted dry to remove excess moisture immediately before image acquisition using the Quantitative Light-induced Fluorescence system (QLF, QLF/clin 11007, Inspektor Research Systems B.V., the Netherlands). Specimens were divided into two groups and demineralized for 48 hrs (Group I) or 96 hrs (Group II). Images of the demineralized enamel were made using the same methods as described above. Fluorescence intensity (FI) of images for both sound (FI<sub>s</sub>) and demineralized enamel (FI<sub>d</sub>) was measured with QLF program using a standard area. One thin section (approximately 100 µm) was obtained from each specimen for the transverse microradiography (TMR) analysis. Integrated mineral loss (IML) and depth were recorded. Enamel and dentin thickness of the thin sections were measured under a stereomicroscope. Enamel thickness ranged from 0.50 to 1.76 mm and dentin thickness ranged from 1.57 to 2.80 mm. There was a good correlation between FI and enamel thickness (FI<sub>s</sub>:  $r = -0.36$ , FI<sub>d</sub>:  $r = -0.53$ ), i.e. the thicker the enamel, the lower the FI in both sound and demineralized enamel. However, a poor correlation was found between FI and dentin thickness (FI<sub>s</sub>:  $r = 0.00$ , FI<sub>d</sub>:  $r = 0.12$ ). There was a correlation between FI<sub>d</sub> and TMR values for Group I (IML:  $r = -0.32$ , depth:  $r = -0.46$ ), but in Group II, there was no significant correlation (IML:  $r = -0.12$ , depth:  $r = -0.18$ ). There were significant differences in the lesions between Group I and II for IML ( $p = 0.0001$ ), depth ( $p = 0.0001$ ), and FI<sub>d</sub> ( $p = 0.0001$ ). **Under the conditions of this study, it can be concluded that fluorescence intensity depends on enamel thickness for lesions of the same size, with fluorescence increasing with lower thickness. Fluorescence intensity was not affected by dentin thickness.** E-mail: mando@iupui.edu

659

Evaluation of Several Methods for the Detection of Root Caries. E. DUNN\*, C. GONZALEZ-CABEZAS, M. ANDO, H. EGGERTSSON, D. GOMES, M. FONTANA, G.J. ECKERT, G.K. STOOKEY. (Indiana University School of Dentistry, Oral Health Research Institute)

Conventional diagnostic techniques detect root caries when it is relatively advanced and a significant amount of tissue has been lost. The purpose of this study was to determine the ability of Quantitative Light-induced Fluorescence (QLF), Visual Examination (VE), Electrical Conductance Method (ECM), and Laser Fluorescence (DIAGNOdent, Dd) for detecting early root surface caries. One hundred extracted teeth with root surfaces ranging from sound to definite clinical caries were marked (identifying the specific site for analysis) and used for the study. Three examiners independently characterized the marked sites with each of the four diagnostic techniques. Exams on 25% of the specimens were repeated. After the examinations, the teeth were sectioned through the selected site and analyzed by confocal laser scanning microscopy (gold standard). The repeatability of the Dd and ECM measurements within examiner was assessed using paired t-tests and intra-class correlation coefficients. The repeatability of the QLF and VE scores was assessed using kappa statistics. Agreement between examiners was assessed using the same methods used for within examiner repeatability. Sensitivity, specificity, and % correct were compared using generalized estimating equation methodology applied to logistic regression models. Areas under the ROC (Receiver Operating Characteristic) curves were calculated to assess the overall diagnostic abilities of the methods. Repeatability within examiner varied for examiner-method combinations, but generally was not strong. Agreement among examiners was also not strong. Based on the area under the ROC curves, the methods had equal diagnostic capabilities (QLF: 0.64, VE: 0.65, ECM: 0.67, Dd: 0.67). In terms of % correct there were no differences (0.62-0.74), but there were differences among some of the methods in terms of sensitivity and specificity. VE and ECM had higher specificity, while Dd had higher sensitivity. **In conclusion, the four techniques were equally accurate in diagnosing root caries and showed similar variations in agreement within and between examiners.** Further research is needed to determine the appropriate strategies to improve the accuracy and reproducibility of the methods for detecting root caries. Supported by NIDCR Grant DE07125-18.

660

Use of a Force Transducer During Clinical Caries Detection. P. HARRIS\*, C. STANFORD, J. WAGNER, G. THOMAS (University of Iowa, Iowa City, Iowa).

Previous studies of the forces used in caries detection have used data from "benchtop" studies. **Purpose:** The objective of this study was to use a force transducer to clinically measure the forces used during routine caries detection. **Methods:** The measurement system consisted of a force-detecting sleeve fitting over the handle of a #5 Shepherd's hook explorer. The sleeve housed 4 strain gauges (EA-06-125BZ-350) to detect forces in a vertical and lateral direction relative to the long axis of the explorer. Custom analog circuitry was designed to amplify and condition the output signal. Data recordings (50Hz) with a force vs. time signal were displayed using HP VEE software. Subjects (n=8) were recruited and IRB approved Informed Consent obtained. Treatment planned carious lesions were evaluated using the explorer and visual observation. Two recording sessions were made for each subject, one performed by the same operator for all subjects. A second operator (n=9) performed a second session in a blinded fashion. **Results:** One-way ANOVA analysis was performed with a Tukey's post-hoc test ( $\alpha = 0.05$ ). Average vertical force used in caries detection on 94 teeth (1252 exploring observations) was not significantly different ( $p > 0.05$ ) at 212.09 +/- 32.11gm while the average horizontal force was 91.53 +/- 30.95gm. Anterior teeth were explored with an average of 203.16 +/- 31.60gm as compared to 218.80 +/- 32.70 for posterior teeth ( $p > 0.05$ ). Left-handed clinicians averaged 289.67 +/- 35.25gm when exploring left quadrants and 288.11 +/- 37.79gm in right quadrants. Right-handed clinicians averaged 229.63 +/- 36.80 in right quadrants and 70.11 +/- 9.72 in left quadrants. Left-handed operators, (n=2) had consistently higher force values ( $p < 0.001$ ) than right handed operators on the left quadrant. **Conclusions:** There was no difference in the forces used by clinicians for caries detection in the anterior vs. posterior quadrants. There was an effect of the dominant handedness of the operator. Measured force values were consistent between 9 operators suggesting that a consistent but narrow range of force values are needed for the conventional detection of caries. P30-DE10126-09; 2000 AADR Research Fellowship.

661

Novel clinical design for the assessment of examiner repeatability and sensitivity for detection of caries progression. P.A. WALTERS\*, A.R. BIESBROCK, S.A. JACOBS, R.W. GERLACH, J.M. McCLARY, R.D. BARTIZEK, M.S. MAU, S.A. KELLY, G.S. STOOKEY (P&G, Cincinnati, OH; IUPUI, Indianapolis, IN)

Calibration of caries examiners for clinical studies is a difficult proposition given the slow nature of the disease. Assessment of repeatability is often readily achieved, but that does not necessarily translate into examiner sensitivity to detect caries progression. This novel examiner calibration study design was a longitudinal, 12 month exercise conducted to assess the proficiency of seven examiners to detect caries progression in the permanent teeth of children between the ages of 9 and 12, as well as their repeatability. This study was conducted in Connersville, IN, where the public water supply was not fluoridated. A total of 228 children were enrolled in this study. At baseline, caries examinations were performed by each of the seven examiners using a visual tactile examination method performed with the aid of an artificial light, mouth mirror, compressed air, and a dental explorer employing a modified Radike criteria. Importantly, white spot demineralization was not scored as caries. In addition, a subset of 15 subjects was examined twice by each examiner at baseline as a repeatability exercise. At year 1, 198 subjects were examined by six of the examiners, as the seventh examiner was only available to examine a subset of 91 subjects. All six of the examiners examining the entire returning cohort observed a statistically significant ( $p < 0.05$ ) positive caries increment over the one year study period as compared to baseline, with mean DMFS increments ranging from  $0.52 \pm 2.10$  to  $1.56 \pm 2.28$  surfaces. With respect to repeatability of the DMFS scores, the examiners had kappa scores ranging from 0.70 to 0.95, with four of the examiners having kappa scores  $> 0.90$ . **This 12 month model has promise for assessing both repeatability of examiner scores and examiner sensitivity to caries progression.**

662

Examiner calibration exercise for the assessment of radiographic caries. J.M. McCLARY\*, P.A. WALTERS, S.A. JACOBS, R.W. GERLACH, R.D. BARTIZEK, J.L. WINSTON, A.R. BIESBROCK (P&G, Cincinnati, OH)

The calibration of caries examiners for clinical studies often involves a clinical visual-tactile repeatability assessment and sometimes an assessment of radiographic repeatability. However, there are very few reports of structured radiographic calibration protocols in the literature. This novel examiner calibration study design was developed to assess the proficiency of eight examiners to detect caries presentation and caries progression in the permanent teeth of children between the ages of 9 and 12. A test set of bitewing radiographs was selected from a one year clinical caries study involving 150 subjects who had four radiographs taken at baseline and four at one year. The test set consisted of the matched baseline and one year radiograph sets from 20 subjects that were selected based on film quality and the diverse presentation of radiographic caries. The radiograph film sets were blinded with respect to subject identifier and visit period and randomly ordered. This set was independently scored for interproximal caries using an illuminated light box by a panel of three experienced clinicians whose consensus served as the gold standard. Overall, a total of 88 lesions were detected at baseline and at one year. The test set was then evaluated by eight caries examiners with varying levels of clinical experience, using their preferred methods of evaluation. Sensitivity of the eight examiners ranged from 66% to 100%, while the number false positive scores ranged from 2 to 99 representing a specificity ranging from 88% to 99%. Because of the matched nature of the radiograph sets, radiographic increment over one year could also be interpreted. Based on the increment observed by the standard panel, kappa analyses were performed for each of the eight examiners. The kappa scores ranged from a low of 0.042 to a high of 0.642. **This model has promise for assessing examiner sensitivity to radiographic caries detection.**

663

Incidence and prevalence of dental caries in children in five cities throughout Guatemala. L. ARCHILA\*, P.A. WALTERS, R.D. BARTIZEK, R.W. GERLACH, J.M. McCLARY, S.A. JACOBS, A.R. BIESBROCK (UTSA, San Antonio, TX; P&G, Cincinnati, OH)

The caries epidemiology within the country of Guatemala is poorly understood. This work reports the cross-sectional incidence and prevalence of caries in the permanent teeth of children between the ages of 6 and 12 in five different urban cities within Guatemala and relates the caries experience to fluoride levels in the communal drinking water in each of these cities. These cities were selected because they represent geographically distinct regions of Guatemala, where there is no artificial fluoridation of public drinking water. Fluoride concentrations from drinking water, collected from three water spigots, at the elementary schools in each city were determined using a fluoride ion selective electrode. The fluoride concentration in water was 0.05 ppm, 0.23 ppm, 0.14 ppm, 0.50 ppm, and 0.60 ppm for Coban, Solola, Guatemala City, Chiquimula, and Estanzuela, respectively. A total of 1160 children were enrolled in this study, with 228, 236, 236, 230, and 230, enrolled at Coban, Solola, Guatemala City, Chiquimula, and Estanzuela, respectively. Caries examinations were performed by a calibrated examiner using a visual tactile examination method performed with the aid of an artificial light, mouth mirror, compressed air, and a dental explorer employing a modified Radike criteria. The cross-sectional mean DMFS in children 6-8 year old was 4.61, 4.34, 3.74, 3.52, and 2.71 for Coban, Solola, Guatemala City, Chiquimula, and Estanzuela, respectively. The cross-sectional mean DMFS in children 9-12 year old was 10.96, 10.50, 8.67, 8.72, and 6.02 for Coban, Solola, Guatemala City, Chiquimula, and Estanzuela, respectively. The prevalence of caries in children between the ages of 9-12 was  $> 98\%$  in all cities, with the exception of Estanzuela (90%). **Collectively, these data demonstrate that the high prevalence and incidence of caries in Guatemala appears to be directly correlated to levels of fluoride in the community drinking water.**

664

Fluoride concentration in mineral water from Lindóia and Serra Negra, Brazil. F.M. LEVY\*, T.T.C. RIBEIRO, M.A.R. BUZALAF, J.R. de M. BASTOS (Bauru Dental School, Univ. of São Paulo, Brazil).

The maintenance of optimal fluoride concentration in mineral water and its presence in water from natural sources is fundamental to assure the efficiency of this method in relation to public health. The use of systemic fluoride must be controlled in respect to risks and benefits, once low dosage won't bring benefits against caries and overdosage is associated with dental fluorosis. The aim of this study was to evaluate fluoride concentrations in natural sources and bottled mineral water in the cities of Águas de Lindóia, Lindóia and Serra Negra, in São Paulo State, Brazil, region called "Water Circuit". It were analysed 20 samples of water from natural sources collected in various points and 9 samples of bottled mineral water commercially available. Fluoride was analysed by the direct method, after addition of Tisab III (Orion), using a fluoride ion specific electrode (Orion 9609), previously calibrated with standards containing known concentration of fluoride. It were observed fluoride concentrations ranging from 0.02 to 0.46 ppm. **It was concluded that all samples had fluoride concentrations below the limits considered beneficial to prevent dental caries at the region analysed. It's necessary to make these data public to population and to health professionals too, once the consumption of these kinds of water has been increasing in the last years in the whole country. Furthermore, local population, by cultural reasons, only drinks water that comes from the natural sources, and then doesn't receive the benefits of fluoride present in public water supply.**