Detection of Early Carious Lesions Using Laser Diode Near-Infrared Transillumination (In Vitro Study)

Amir N El-Kholi¹, Samir A Koheil² and Mona M Ghoneim³

¹Department of Demonstrator Conservative, Alexandria University, Egypt
²Department of Conservative, Professor of Operative Dentistry, Alexandria University, Egypt
³Department of Conservative, Assistant Professor of Operative Dentistry, Alexandria University, Egypt

Received: August 07, 2017; Published: August 22, 2017

*Corresponding author: Amir N El-Kholi, Department of Demonstrator Conservative, Alexandria University, Egypt

Abstract

Introduction: The importance of early detection before the development of irreversible damage is now generally accepted [1]. Most studies do not report the presence of non cavitated lesions, though they have been shown to have predictive value [2].

Aim of the study: Validate DIAGNO cam (KaVo, Biberach, Germany) as a diagnostic instrument in the early detection of non cavitated carious lesions.

Materials and method: Thirty three extracted sound premolars were examined on both approximal surfaces by EDX to determine Ca/P ions ratio on (D1), imaged using Periapical Digital Radiography (D2), and transilluminated using DIAGNO cam (D3). Any defect that indicated cavities by any of the mentioned methods discarded the tooth. Three teeth were randomly selected and imaged by Scanning Electron Microscopy. The remaining thirty teeth were covered with acid resistant varnish except in 3x3mm in approximal areas and 15 teeth were immersed in a demineralizing solution for 48H (Group I), the other 15 teeth were immersed for 72H (Group II). Both groups were then re-examined using D1, D2 & D3. Six teeth of groups I & II were selected and imaged by Scanning Electron Microscopy.

Results: Group I: D1 detected (100%) Ca/P ions ratio less than 1.8 (demineralized), D2 (100%) sound enamel, while D3 (66.67%) black spots and (33.33. %) sound. Chi square test showed that D1 is more accurate than D2 and D3, while D3 is more accurate than D2 and the differences were statistically significant. Group II: D1 detected (100%) Ca/P ions ratio less than 1.8 (demineralized), detected (6.67%) with enamel radiolucency and (93.33%) sound, while D3 (80%) black spots and (20%) sound. Chi square test showed that there is a statistically significant difference between D2 and the remaining diagnostic means (D1 and D3). SEM in both groups I & II showed evidence of enamel demineralization in all randomly selected samples.

Conclusion: DIAGNOcam is a reliable and valid method in detecting early enamel caries demineralization.

Keywords: DIAGNOcam; Transillumination; Early Detection; Non Cavitated Lesions; DIFOTI

Introduction

Dental caries is the most prevalent dental diseases in the world. Ideally, caries detection methods should capture the whole caries process, from the beginning of early demineralization through the capitation stage. Studies proved that by using only conventional clinical and radiographic methods, the dentist will detect about half the lesions present [3]. Non-invasive treatment of early caries lesions by remineralization has become of major importance in clinical daily practice, where many studies in turn would prevent white spot lesions formation and further capitation; but instead, rematerialize the existing lesions eliminating the need to restore the tooth [4]. Therefore, the attempts to identify more advanced measures to detect early carious lesions increased. These technologies include Quantitative Laser or Light Fluorescence (QLF), Electrical Conductance Measurements (ECM), infrared Laser Fluorescence (LF), direct digital radiography and Digital Imaging Fiber-Optic Trans-Illumination (DIFOTI) [5,6]. Recent evidence showed that the Near Infrared (NIR) spectra (780 to 1550 nm), have the ability to penetrate the tissue more deeply. This deeper penetration is essential for the Transillumination (TI) [7].

During the early stage of caries formation (demineralization), micropores are formed in the carious lesion due to partial dissolution of some individual mineral crystals. These small pores behave as scattering centers for both visible and near infrared (NIR) light. The optical properties of enamel become quantitatively described by the absorption and scattering coefficients [7]. Therefore, Near Infrared (NIR) technology can be used to obtain...
images of dental decay that is not detectable by the conventional means neither graphically nor by visual/tactile examination in both occlusal and approximal caries [7-9]. A new LASER diode near infrared trans illumination digital video camera known as (DIAGNocam) records the image and displays it live on a computer screen, using KaVo KID V 2.4 software demineralized lesions are displayed as dark shadows. The images recorded can be stored, allowing the determination of early demineralization without using a radiograph, thus significantly simplifying monitoring and patient communication. The purpose of this study is to determine the validity of that device for the diagnosis of early enamel demineralization before capitation, enabling the practitioner to treat instead of fill the tooth.

Materials and Method

Thirty three sound human premolars extracted for orthodontic indications at the department of Oral and Maxillofacial Surgery (Faculty of Dentistry - Alexandria University) with a written consent signed by the patients. Teeth were stored in a thymol saturated saline to prevent any bacterial growth, rinsed in 10% sodium hypochlorite solution for 20min, followed by rinsing in distilled water for 20min. The teeth were subjected to the following procedures and named (D1, D2 & D3) accordingly to make sure that all the specimen’s enamel was free from any demineralized carious lesions.

D1: Energy dispersive x-ray spectrometry (EDX) to detect Calcium and Phosphorus ions concentration on the middle thirds of the mesial and distal surfaces of the teeth in order to calculate the \( \frac{Ca}{P} \) ions ratio. The procedure started by dehydration of teeth by desiccators. Each tooth was introduced in the scanning electron microscopy and an area on the middle third of mesial surface of the tooth to be examined by EDX was determined and using ISIS computer software and a graph was obtained showing both Calcium and Phosphorus ions content on the area examined. The same procedure was performed for an area on the middle third of the distal surfaces as well. The resulting concentrations were used to calculate the Calcium and Phosphorous ions ratio and any tooth with \( \frac{Ca}{P} \) ions ratio less than 1.8 on any of its sides was considered dematerialized (ten Cate, JM et al 1982 & Jalevik, B. et al, 2001) [10, 11]. Hence, it was discarded, replaced by another tooth and examined as mentioned previously.

D2: Periapical digital radiography showing both mesial and distal surfaces. The teeth were fixed by soft wax at the tip of their roots in a vertical position with the digital sensor placed in a vertical manner lingual to them and the cone positioned from the distal surfaces as well. The resulting images were used to calculate the Calcium and Phosphorous ions ratio and any tooth with \( \frac{Ca}{P} \) ions ratio less than 1.8 on any of its sides was considered demineralized (ten Cate, JM et al 1982 & Jalevik, B. et al, 2001) [10, 11]. Hence, it was discarded, replaced by another tooth and examined as mentioned previously.

D3: LASER diode near infrared transillumination operating at 788 nm wavelength (DIAGNocam), which is a fluorescent LASER device for caries detection. DIAGNocam produces images reminiscent of X-ray images, but which are completely radiation free by means of a LASER beam that is especially adapted to this examination method, it is delivered with a computer-based image processing software for showing and saving the images. The tooth structures allow the passage of the LASER beam from the entry site to the camera. Areas that block LASER beam transmission (e.g. demineralized carious lesions) show up clearly as well delimited, dark areas. A digital camera captures the actual situation and makes it visible in real time on the screen. DIAGNocam images were obtained from an occlusal view in a dark room setting to simulate the situation inside the oral cavity. Each tooth had two images captured, all from the occlusal view; one taken from the occlusal aspect but with 8° tilting towards the mesial surface and one with 8° tilting towards the distal surface. Any tooth showing defects by a dark area in the images that indicated demineralization or caries was replaced by another tooth and re-examined in the previously mentioned sequences of D1 and D2.

Three teeth were randomly selected out of the thirty three that were diagnosed as sound teeth in D1, D2 and D3. They were gold sputtered and invasively imaged on the middle thirds of both their mesial and distal surfaces by scanning electron microscopy at 25KV and x1000 magnification to check the absence of micro porosities on the examined areas of the teeth and serve as a gold standard for the work. The remaining thirty teeth were mounted in acrylic cylindrical blocks with their crowns and the coronal third of their roots visible both measuring 16 mm, leaving the rest of the root in the acrylic block to facilitate the use of DIAGNocam and allow proper imaging of the approximal areas. A window of 3x3 mm was identified in the middle third of the proximal surface by a ruler and the crown of each tooth was covered with Hoffmann copal varnish (Hoffmann, Berlin, Germany). A demineralization solution was prepared containing 50 mmol acetic acid, 2.2 mmol Ca (NO3)2, 2.2 mmol KH2PO4 and 0.1 ppm NaF. PH was adjusted to 4.5 by addition of KOH solution. (Murdoch-Kinch, C. et al) [12].

All the thirty teeth were further grouped into two groups 15 teeth each according to the time of immersion in the demineralization solution. Group I teeth were immersed for 48 hours in demineralizing solution and examined afterwards in the same sequence of diagnostic instruments explained earlier (D1, D2 & D3). Group II teeth were immersed for 72 hours in demineralizing solution and examined afterwards in the same sequence of diagnostic instruments explained earlier (D1, D2 & D3). Three teeth were randomly selected out of each group and invasively imaged by scanning electron microscopy on the middle third of both mesial and distal surfaces to confirm demineralization and to serve as a gold standard. All the data obtained was statistically evaluated concerning their calcium and phosphorous content. All the images obtained were statistically analyzed using a qualitative criterion (Maia, A. et al) [13] to determine the presence of caries changes on each approximal surface in which:

a. sound enamel
b. Enamel caries
c. Caries reaching but not crossing the enamel-dentine junction
d. Caries into dentine.
Results

The selected teeth prepared for the study were diagnosed as sound by all diagnostic means used. Visually all teeth looked sound without discoloration, cracks, white spots or anything that looked to interfere with normal enamel.

D1: The mesial surfaces showed a Calcium ions range between 84.3% and 64.9%, a Phosphorus ions range of 35.1% and 15.7% and a Ca/P ions ratio range between 5.3758 and 1.849. The average Ca/P ions ratio is 2.4044. The distal surfaces showed a Calcium ions range between 84.3% and 64.9%, Phosphorus ions range between 35.6% and 28.8% and a Ca/P ions ratio range between 2.8949 and 1.809. The average Ca/P ions ratio is 2.1546. D2: All radiographic images obtained showed sound enamel (Score 0) on both their mesial and distal sides. D3: All DIAGNOcam images obtained showed sound enamel (Score 0) on the mesial and distal surfaces of the teeth. All scanning electron microscopy images examined randomly showed completely sound prism less intact enamel with no surface demineralization or porosities on both mesial and distal sides of the teeth.

Group I (48 hours demineralization): D1: All the thirty EDX results obtained of the mesial and distal surfaces showed a mesial surface Calcium ions range between 58.2 % and 53 %, a Phosphorus ions range between 46.9 % and 41.8 % and a Ca/P ions ratio range between 1.3946 and 1.1257. The average Ca/P ions ratio is 1.276. The distal surface Calcium ions range is between 57 % and 52.8 %, Phosphorus ions range between 46.8 % and 41.9 % and the Ca/P ions ratio range between 1.3876 and 1.1209. The average Ca/P ions ratio is 1.2501. D2: Radiographic images showed sound enamel (Score 0) on both mesial and distal sides of the teeth. D3: DIAGNOcam images showed eight teeth with carious demineralization (Score 1) on both their mesial and distal surfaces, four teeth with carious demineralization (Score 1) on one surface only and three teeth were sound (Score 0). Nine out of fifteen mesial surfaces showed carious demineralization extended to enamel only (Score 1), while eleven out of fifteen distal surfaces showed carious demineralization extended to enamel only (Score 1), making a total of twenty surfaces with demineralized carious enamel (Score 1). All scanning electron microscopy images examined randomly from both mesial and distal surfaces of the three teeth showed demineralized carious enamel with irregular pattern of surface destruction appearing as haphazardous thin splits.

The obtained data and images were recorded, statistically analyzed by SPSS version 20 software using Pearson’s Chi square significance test. At the mesial sides, of group I, D1 detected 15 surface (100%) out of 15 surfaces with Ca/P ions ratio less than 1.8 (demineralized), D2 detected 15 surfaces (100%) out of 15 surfaces with sound intact enamel surfaces free from any radiolucency, while D3 detected images of 11 surfaces (73.33%) out of 15 surfaces with black spots and images of 4 surfaces (26.67%) out of 15 surfaces that showed sound enamel surfaces. Statistical comparison of the three diagnostic means D1, D2 and D3 showed that D1 is more accurate than D2 and D3, while D3 is more accurate than D2 and the differences are statistically significant between the three methods in detection of early carious demineralization ($X^2=30.54, p=0.000*$). At the distal sides, of group I, D1 detected 15 surface (100%) out of 15 surfaces with Ca/P ions ratio less than 1.8 (demineralized), D2 detected 15 surfaces (100%) out of 15 surfaces with sound intact enamel surfaces free from any radiolucency, while D3 detected images of 11 surfaces (73.33%) out of 15 surfaces with black spots and images of 4 surfaces (26.67%) out of 15 surfaces that showed sound enamel surfaces. Statistical comparison of the three diagnostic means D1, D2 and D3 showed that D1 is more accurate than D2 and D3, while D3 is more accurate than D2 and the differences are statistically significant between the three methods in detection of early carious demineralization ($X^2=32.98, p=0.000*$).

Group II (72 hours demineralization): D1: All the thirty EDX results obtained of the mesial and distal surfaces showed a mesial surface Calcium ions range between 56.5 % and 49.4 %, a Phosphorus ions range between 50.6 % and 43.5 % and a Ca/P ions ratio range between 1.3006 and 0.9761. The average Ca/P ions ratio is 1.1404. The distal surface Calcium ions range is between 56.3% and 44.9%, Phosphorus ions range between 55.1 % and 43.7 % and the Ca/P ions ratio range between 1.2682 and 0.9481. The average Ca/P ions ratio is 1.0774. D2: Radiographic images showed only two teeth with radiolucencies extended to enamel only (Score 1) on one of their proximal surfaces from which one was on the mesial surface while the other was on the distal surface. The remaining thirteen teeth showed sound enamel (Score 0) on both their mesial and distal surfaces. D3: DIAGNOcam images showed eleven teeth with carious demineralization (Score 1) on both their mesial and distal surfaces, two teeth with carious demineralization (Score 1) on one surface only and two teeth were sound (Score 0). Eleven out of fifteen mesial surfaces showed carious demineralization extended to enamel only (Score 1), while thirteen out of fifteen distal surfaces showed carious demineralization extended to enamel only (Score 1), making a total of twenty four surfaces with demineralized carious enamel (Score 1).

All scanning electron microscopy images examined randomly showed demineralized carious enamel with irregular pattern of surface destruction on both mesial and distal sides of all the three teeth appearing as haphazardous thin splits as well as the typical honeycomb appearance indicating prismatic pattern destruction where the prism cores has been destroyed with remaining interprismatic substance which is less affected. The obtained data and images were recorded, statistically analyzed by SPSS version 20 software using Pearson’s Chi square significance test. At the mesial
Discussion

Detection of carious lesions on neighboring approximal surfaces of posterior teeth is a challenge, and the inaccessibility of clinical visual and tactile methods is the reason that use of ionizing radiation for bitewing radiographs [14] which are unable to identify the initial demineralization of the tooth, resulting in low sensitivity, with decalcification ranging from 40% to 60% necessary to produce a radiographic image of caries, and this might result in false-negative tests [15,16]. A reliable detection method is therefore required to be agreed upon as routine to detect caries at its earliest stages in order to remineralize instead of restoring teeth. Thirty three sound teeth were selected for the study and stored in a thymol saturated saline to prevent any bacterial growth, rinsed in 10% sodium hypochlorite solution for 20 min, followed by rinsing in distilled water for 20 min, that solution did not affect the surface of the teeth which was in agreement with (Astwaldsdottir, L. et al 2012) [1]. These teeth were selected for comparing the detection of early caries demineralization using EDX, X-ray and DIAGNOcam. Many studies compared X-ray and near-infrared Transillumination (NIR-TI). However, there was no recorded study comparing the three methods combined in early detection of enamel caries demineralization.

EDX having the ability to detect the number of Calcium and Phosphorus ions in enamel can in fact determine whether the surface is demineralized or not, unfortunately, it cannot be used clinically as the specimen should be dehydrated first then introduced in a vacuum chamber, which is not possible in vivo. EDX is defined as a quantitative, semi quantitative or qualitative method for identification of chemical elements in a wide variety of samples. The technique sensitivity of this method depends on the atomic number of the element to be identified, the atomic number of all elements present in the sample and the technique used during sample preparation for microanalysis [17]. Ca/P ions ratio was measured to determine the health of enamel, where a ratio equal to or above 1.8 was normal healthy enamel according to (ten Cate, JM et al 1982 & Jalevik, B. et al, 2001) [18,19], while according to (de Sant’Anna, G. et al, 2009) [20] 1.67 was enough for the tooth to be considered healthy. A dark room setting was used during DIAGNOcam detection to simulate the conditions of the oral cavity however, the attempts to establish an appropriate in vitro set-up have failed to replicate the same degree of teeth Transillumination because the photo-optical properties of the different embedding materials are not comparable with periodontal anatomy (eg, the oral mucosa, alveolar bone, periodontal ligament, pulp and blood) as agreed with (Kühnisch, J et al 2015) [21].

A demineralizing solution was prepared in order to demineralize the exposed areas of the specimens’ enamel, it was prepared according to (Murdoch-Kinch, C. et al 2003) [22] the teeth were immersed in it and grouped accordingly to Group I (48 hours) and Group II (72 hours) where each group contained fifteen teeth. In contrast, many other studies used different demineralizing agents and different periods of immersion, such as: 6 per cent hydroxyethyl cellulose gel for 1, 3 or 7 days (Ferguson, S. et al 1988) [23], conventional lactic acid-Carbopol solution for varying periods of time between 0 and 24 h (Ando, M. et al 1997) [24] 48, 72 and 96 h (Ando, M. et al 2001) [25] and calcium phosphate (2 millimolars per liter) in an acetate (0.075 moles per liter) buffer) with 4.8 pH at 37 °C for 14 weeks (Young, D. et al 2005) [26]. The results are in agreement with; (Schneiderman, A. et al 1997) [27] that found that for approximal caries demineralization, the sensitivity of DIFOTI is more than twice as high for radiography (Vaarkamp, J. 2000) [28]. That made a review which included studies throughout 13 years about the efficiency of bitewing radiography versus optical fiber Transillumination and concluded in that the X-ray diagnosis accuracy is much lower for the incipient enamel lesions (Maia, A. et al 2011) [29].
That concluded that near-infrared Transillumination was more reliable than analogue (conventional) radiographs and that enamel caries surfaces were better identified using this method (Astvaldsdottir, A. et al 2012) [1]. Stated that the DIFOTI method showed better correlation with actual lesion depth than conventional and digital radiography (Marinova-Takorova, M. et al 2014) [17]. Concluded that visual examination seriously underestimated enamel lesions, while, there was a good correlation between near-infrared Transillumination and bitewing X-ray data where NIR showed higher sensitivity. (Giriligeriu, L. et al 2015) [30,31] that compared clinical examination, near-infrared Transillumination (DIAGNOcam) and radiography in proximal caries (Simon, J. et al 2016) [32]. Whose results showed that near-infrared imaging was significantly more sensitive than radiography in detecting early carious lesions on both occlusal and interproximal surfaces? (Marinova-Takorova, M. et al 2016) [33] That compared mirror and a dental explorer; under x6.4 magnification using a Leica M320 dental microscope, then DIAGNOcam, and bitewing radiographs. Their conclusion was that near-infrared Transillumination is an effective method for diagnosis of lesions both involving only the enamel and involving the enamel and dentin for both occlusal and proximal caries lesions and it could eventually substitute radiographic bitewings, especially in children and pregnant women, due to its efficiency as a diagnostic tool and the absence of radiation (Russotto, F. et al 2016) [34]. Concluded that near-infrared Transillumination performed significantly better than radiography as an interproximal caries detection tool. (Yu, J.L. et al 2017) [35]. Suggested that the DIAGNOcam may be used to assess the depth of caries cavity as a useful tool in diagnosis and treatment. There was disagreement with; (Young, D. et al 2005) [36] whose results showed that DIFOTI was not able to measure the depth of a lesion in any of the samples. It was, however, able to show surface changes associated with early demineralization as early as two weeks. The depth of a lesion measured using radiographic film was not statistically different from the depth of a lesion measured with PLM histologic analysis (P > .05) (Jost, F. et al 2015) [37]. That studied the reproducibility of near-infrared light Transillumination (NIR-TI) device (DIAGNOcam) for interproximal enamel caries and concluded that agreement with NIR-TI was worse compared to Bitewing digital radiography (BW). They owed this for the missing calibration of their examiners and shadow artifacts in the margins of the teeth in some NIR-TI images. The results demonstrate the potential of the DIAGNOcam for detection of early caries demineralization in enamel with high sensitivity relative to digital radiography. In particular, it was found that:

i. DIAGNOcam provides clear image of the location and dimensions of enamel demineralization.

ii. DIAGNOcam can indicate the presence of early enamel caries demineralization when digital radiography fails to show they exist.

This could be attributed to fact that the incipient carious lesion is characterized by the initial demineralization extending over 200-300 nm depth, and using radiology investigation, lesions can be detected only if they are at least 500 nm deep [38]. This novel and straightforward method, exploits the high transparency of dental enamel and the strong scattering and weak absorption of the underlying dentin to deliver a uniform distribution of diffuse NIR light underneath the transparent enamel of the crowns to facilitate high contrast. NIR imaging and detect hidden lesions beneath the enamel surface. NIR studies have demonstrated that stains are less visible in the NIR, since the organic molecules responsible for pigmentation absorb poorly in the NIR, making it easier to identify areas of demineralization [39].

However, studies proved that near-infrared Transillumination at 830-nm offers significantly improved image contrast over the visible range, but less image contrast than at 1310-nm [40]. Wavelengths longer than 1400-nm have better performance for the Transillumination of occlusal caries lesions while 1300-nm appeared best for the Transillumination of proximal surfaces [41]. It was suggested by (Bühler, C. et al 2005) that the cost of NIR technology will become available in the near future or alternative systems can be developed operating at 830-nm or 1550-nm, balancing cost, sensitivity and performance [39]. Many studies were made on other diagnostic methods/devices that were compared with NIR-TI such as; (Marinova-Takorova, M. et al 2014) [42] that compared visible tactile method, DIAGNOcam and laser fluorescence device (DIAGNO dent). Then X-rays were taken and finally, Fuschine caries detection dye in detection of occlusal caries. Their results showed that DIAGNO dent and DIAGNOcam were very close, but DIAGNO cams’ data was better correlating with the clinical results. (Simon, J. et al 2017) [43] concluded that near-IR imaging methods have greater potential than cross-polarization optical coherence tomography (CP-OCT) for improving the early diagnosis of occlusal caries demineralization. However, there were no studies comparing NIR-TI with cone beam computed topography, ultrasound and LED reflection and refraction.

Conclusion

a. Near-infrared LASER diode transillumination (DIAGNO cam) is a reliable and valid method in detecting early enamel caries demineralization.

b. Near-infrared LASER diode transillumination (DIAGNO cam) replaces the need for radiography and could be used as a routine method for diagnosing early enamel caries demineralization.

c. Near-infrared LASER diode transillumination (DIAGNO cam) could become a stronger preventive measure used to allow remineralization of teeth instead of restoring them.

References


