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

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ABSTRACT

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

***Objectives:** Validate LASER diode near-infrared transillumination (DIAGNOcam), as a diagnostic instrument in the early detection of noncavitated carious lesions.*

***Materials and Methods:** Thirty three extracted sound premolars were examined by (D1) EDX to determine Ca/P ions ratio (1.8≤), (D2) Periapical Digital Radiography, and (D3) LASER diode near-infrared transilluminated using DIAGNOcam. Any defect that indicated cavities by any of the mentioned methods discarded the tooth. To confirm that the selected sound teeth were intact, three teethwererandomly selected and invasively imaged by scanning electron microscopy. The remaining teeth weredivided into two groups (Group I & Group II) according to the time of immersion in a demineralizing solution (48 hours & 72 hours). Then, the teeth were then re-examined using D1 (EDX), D2 (X-RAY)&D3 (DIAGNOCAM), as well as imaged afterwards by scanning electron microscopy.*

***Results:** Pearson's Chi square test as well as validity tests showed that there is a statistically significant difference between D2 (X-RAY) and the other diagnostic means (D1 (EDX) and D3 (DIAGNOCAM)). SEM showed evidence of enamel demineralization in the three randomly selected teeth.*

***Conclusion:** LASER diode near-infrared transillumination (DIAGNOcam) is a radiation free, reliable and valid method that helps in the early detection of enamel carious lesions allowing us to diagnose and treat the affected surfaces instead of fill them.*

Keywords: *DIAGNO cam, LASER diode near-infrared transillumination, early detection, noncavitated lesions.*

INTRODUCTION

Dental caries is the most prevalent dental diseases in the world. It is a multi-factorial disease formed by a complex interaction between acid-producing bacteria and fermentable carbohydrate (1).The accumulation of microbial plaque to the enamel surface, initiates the carious surface dissolution which in turn proceeds to subsurface demineralization (2). As intervention and treatment should become with a micro-minimally invasive approach(3).Ideally, caries detection methods should capture the whole caries process, from the beginning of early demineralization through the cavitation stage. It should be accurate and easy to apply for all surfaces of teeth, also for lesions adjacent to restorations (4). Detection of carious lesions on neighboring approximal surfaces of posterior teeth is also a challenge (5). The sensitivity of dental radiographs (Periapical, Panoramic & Posterior Bitewing) in detecting evidence of dental caries is lower than expected, rarely showing more than 60% of the lesions (6). Which means, by using only conventional clinical and radiographic methods, the dentist will detect only cavities (3).In general, cavitated lesions are the ones being recorded; however, there is an understanding among researchers that the detection of the caries process has progressed far beyond the point of confining the evidence for dental caries at the cavitation level involving enamel or both enamel and dentin. Hence, recording carious lesions only at the cavitation level is no longer acceptable by researchers (4). 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 cavitation (7). Sound enamel is comprised of modified hydroxyapatite crystals that are closely packed, producing a semi transparent structure. The color of teeth is strongly affected by the dentin shade lying underneath. In presence of demineralization, enamel is disrupted and scatters the penetrating photons of light which results in an optical disruption(8).A new LASER diode near infrared transillumination digital video camera known as (DIAGNOcam) records the image and displays it live on a computer screen, using a computer 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 DIAGNOcamindiagnosing early enamel demineralization before cavitation, enabling the practitioner to treat instead of fill the tooth.

Materials and Methods

Thirty three sound human premolars were extracted for orthodontic indications visually free from any carious lesions, then 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.

Selection criteria: All the thirty three were visually inspected to detect any discoloration, cracks, white spots or anything that looked to interfere with normal enamel and were subjected to the following diagnostic tools and named (D1 (EDX), D2 (X-ray)&D3 (DIAGNOcam)) accordingly.

D1 (EDX)(Oxford Instruments plc, Abingdon, UK): Examination by energy dispersive X-ray spectrometry (EDX), which was performed to detect Calcium and Phosphorus ions concentration just below the contact areas of the mesial and distal surfaces of the teeth in order to calculate the Ca/P ions ratio. Any tooth with a ratio less than 1.8 on either of its sides was considered a demineralized carious tooth according to Ten Cateet al (9) and Jalevik et al (10).

D2 (X-ray) (Owandy, Croissy-Beaubourg, France): The same teeth from D1 (EDX), were examined using periapical digital radiography showing both mesial and distal surfaceswith a radiation dose of 0.19 s/mGy. Any tooth showing radiolucencies on either of its mesial or distal surfaces that indicated caries was replaced by another tooth and re-examined by D1 (EDX).

D3 (DIAGNOcam) (KaVo, Biberach, Germany): The same teeth from D1 (EDX) and D2 (X-ray) were imaged on both mesial and distal surfaces using LASER diode near infrared transillumination operating at 788 nm wavelength (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 using the KaVo KiD V 2.4 computer software, both taken from the occlusal view; one taken with 18° tilting towards the mesial surface and one with 18° tilting towards the distal surface. The degree of inclination was measured using iLevel LITE High Precision Clinometer v 1.0 mobile app. Any tooth showing defects by a dark area in the images that indicated demineralization or caries was replaced by another tooth and re-examined by D1 (EDX) and D2 (X-ray).

Scanning electron microscopy (JEOL, Akishima, Tokyo): (Descriptive images) three teeth were randomly selected out of the thirty three that were diagnosed as sound teeth in D1 (EDX), D2 (X-ray) and D3 (DIAGNOcam). They were imaged by scanning electron microscopy at 25KV and x1000 magnification to check the absence of micro porosities on the examined areas of the teeth.

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 just below the contact area of the proximal surface and the remaining tooth surfaces were covered with Hoffmann copal varnishPreparation of the demineralizing solution according to Murdoch-Kinch(11):50 mmol acetic acid, 2.2 mmol $Ca(NO_3)_2$, 2.2 mmol KH_2PO_4 and 0.1 ppm NaF. pH was adjusted to 4.5 by addition of KOH solution.

All the thirty teeth weredivided into two groups (Group I and Group II) and immersed for 48 and 72 hours accordingly in demineralizing solution. Then, the teeth were examined in the same sequence of diagnostic instruments explained earlierafter removal of the varnish using acrylic monomer(14) to allow better penetration of the LASER beam, thus better transillumination.

All (D1) EDX data obtained was statistically evaluated concerning their calcium and phosphorous content. All the images obtained by (D2) Digital radiography and (D3) DIAGNOcam were statistically analyzed using a qualitative criterion according to Maiaet al(15) to determine the presence of caries changes on each approximal surface in which: (0) sound enamel, (1) Enamel caries, (2) Caries reaching but not crossing the enamel–dentine junction, (3) Caries into dentine.

Statistical Analysis

The obtained data and images were statistically analyzed by SPSS version 20 software using Pearson's Chi square significance test. Also, validity test was executed, which is the extent to which a test measures what it is supposed to measure; in other words, it is theaccuracy of the test. Validity is measured by sensitivity, specificity, positive predictive value and negative predictive value.Sensitivity = (true positive) / (true positive + false negative)Specificity = (true negative) / (true negative + false positive)Positive predictive value = (true positive) / (true positive + false positive)Negative predictive value = (true negative) / (false negative + true negative)

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 (EDX): 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 (X-ray): All radiographic images obtained showed sound enamel (Score 0) on both their mesial and distal sides as shown in figures (1&2).

D2: Before demineralization

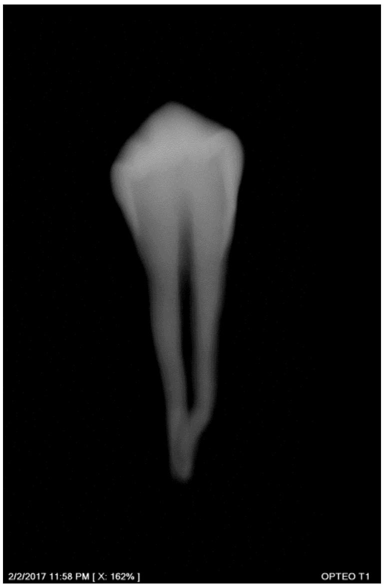


Figure1. *Digital radiographic image showing (Score 0) carious demineralization on both mesial and distal surfaces of the tooth.*

Figure2. *Digital radiographic image showing (Score 0) carious demineralization on both mesial and distal surfaces of the tooth.*

D3 (DIAGNOcam): All DIAGNOcam images obtained showed sound enamel (Score 0) on the mesial and distal surfaces of the teeth as shown in figures (3&4).

D3: Before demineralization

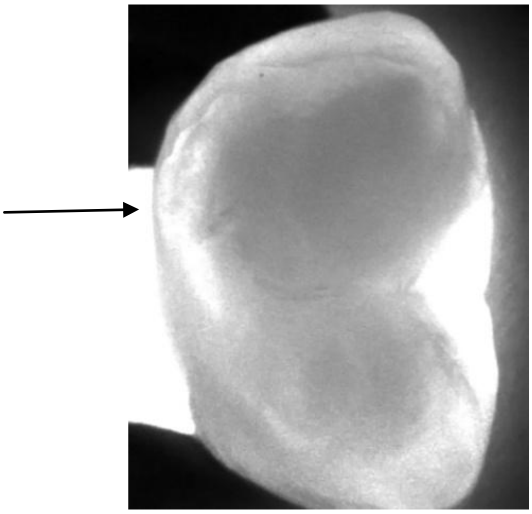


Figure3. *DIAGNOcam image showing (Score 0) carious demineralization on the mesial surface of an upper left 1st premolar.*

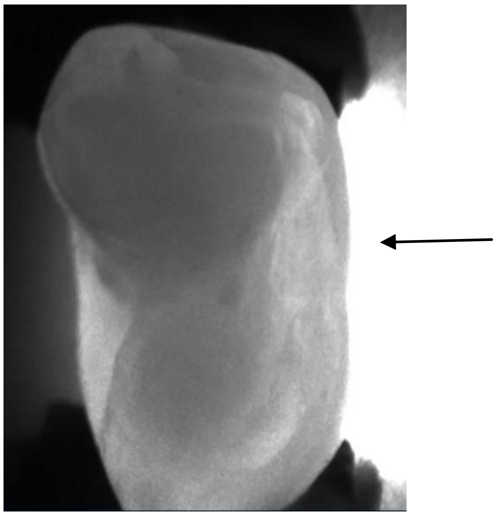


Figure4. *DIAGNOcam image showing (Score 0) carious demineralization on the distal surface of the same upper left 1st premolar.*

All scanning electron microscopy images examined randomly showed completely sound prismless intact enamel with no surface demineralization or porosities on both mesial and distal sides of the teeth as shown in figure (5).

Scanning electron microscopy before demineralization

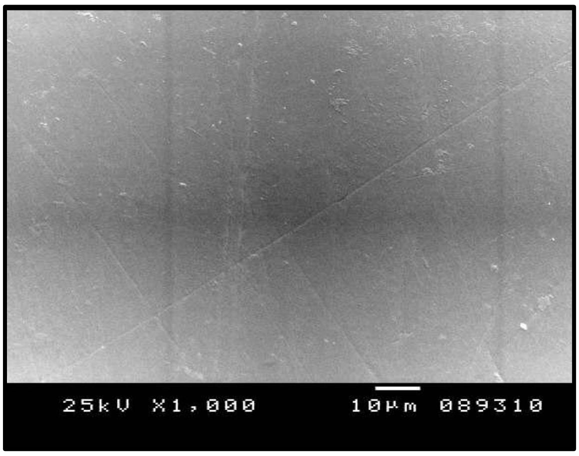
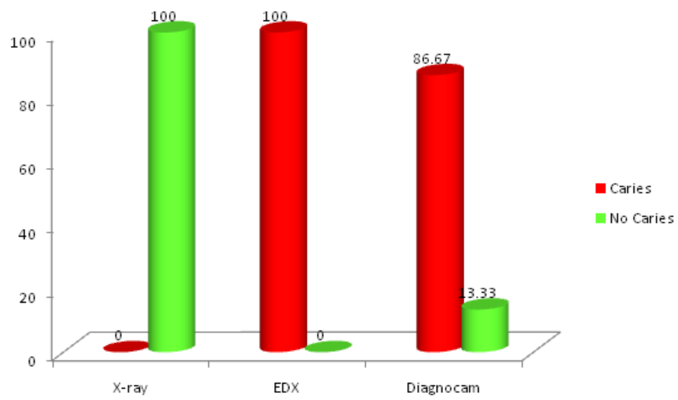


Figure5. *SEM image showing prismless intact enamel (x1000)*

For the 48 hours demineralization (Group I), the results were as the following as shown in bar chart (1):

Pearson's Chi square test of significance in Group I (48 hours)



Bar chart1. *Results obtained in Group I after 48 hours of demineralization.*

D1 (EDX): All the thirty EDX results obtained showed a mesial surfaces Calcium ions range between 58.2% and 53% with a mean percentage of 55.6%. Phosphorus ions ranged between 46.9% and 41.8% with a mean percentage of 44.35%. Ca/P ions ratio ranged between 1.3946 and 1.1257 with a mean ratio of 1.276. The distal surfaces showed a Calcium ions range between 57% and 52.8% with a mean percentage of 54.9%. Phosphorus ions ranged between 46.8% and 41.9% with a mean percentage of 44.35%. Ca/P ions ratio ranged between 1.3876 and 1.1209 with a mean ratio of 1.2501.

D2 (X-ray): The radiographic images showed (Score 0) carious demineralization on both mesial and distal sides of all the teeth as shown in figures (6&7).

D2: After 48 hours of demineralization

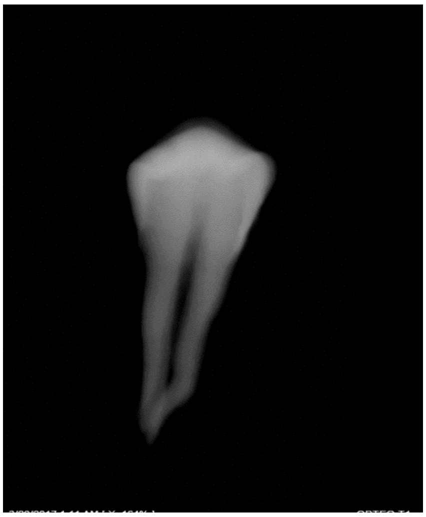


Figure6. *Digital radiographic image showing (Score 0) carious demineralization on both mesial and distal surfaces of the tooth.*

Figure7. *Digital radiographic image showing (Score 0) carious demineralization on both mesial and distal surfaces of the tooth.*

D3 (DIAGNOcam): DIAGNOcam images showed thirteen teeth with (Score 1) carious demineralization on both their mesial and distal surfaces and two teeth with (Score 0) carious demineralization, making a total of twenty six surfaces with (Score 1) carious demineralization as shown in figures (8&9).

D3: After 48 hours of demineralization

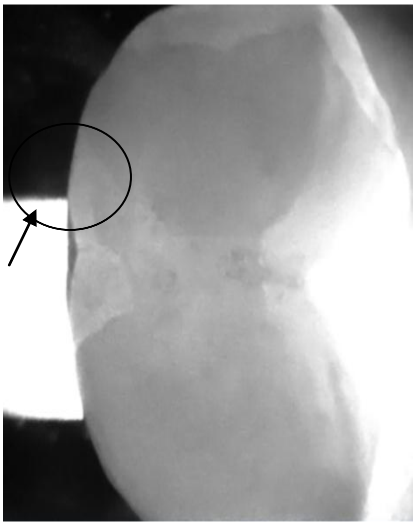


Figure8. *DIAGNOcam imageshowed (Score1) carious demineralization of the mesial surface of an upper left 1st premolar.*

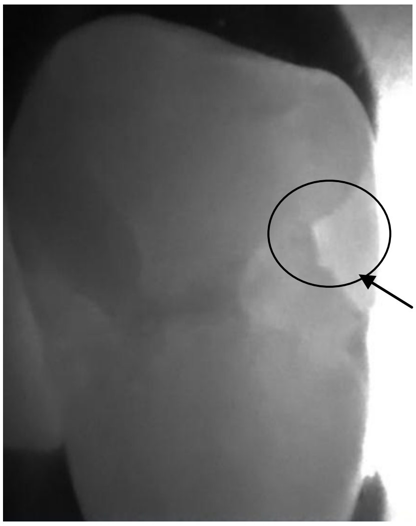


Figure9. *DIAGNOcam image showing (Score 1) carious demineralization of the distal surface of the same upper left 1st premolar.*

All scanning electron microscopy images showed demineralized carious enamel with irregular pattern of surface destruction on both mesial and distal sides of all the three teeth appearing as hap hazardous thin splits as shown in figure (10).

Scanning electron microscopy after 48 hours of demineralization

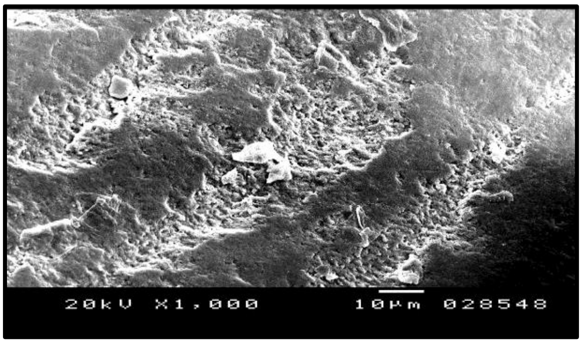
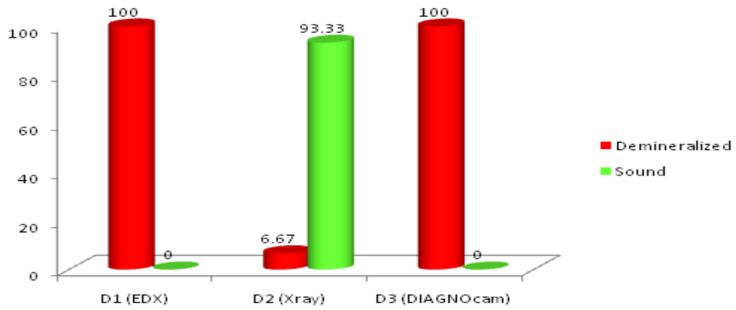


Figure10. *SEM image of Group I showing irregular pattern of surface destruction (x1000).*

For the 72 hours demineralization (Group II), the results were as following as shown in bar chart (2):

Pearson's Chi square test of significance in Group II (after 72 hours demineralization)



Bar chart 2: *Results obtained in Group II after 72 hours of demineralization.*

D1 (EDX): 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 (X-ray):Radiographic images showed only four teeth with radiolucencies extended to enamel only (Score 1) on one of their proximal surfaces from which two were on the mesial surfaces while the other two were on the distal surfaces. The remaining twenty six teeth showed sound enamel (Score 0) on both their mesial and distal surfaces as shown in figures (11&12).

D2: After 72 hours of demineralization

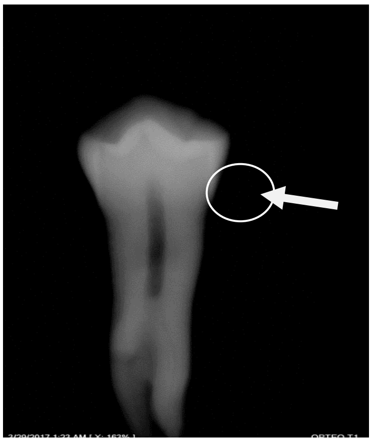


Figure11. *Digital radiographic image showing (Score 1) carious demineralization on the distal surface of a lower right 2nd premolar.*

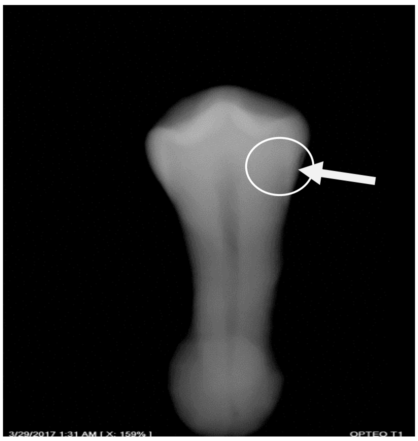


Figure12. *Digital radiographic image showing (Score 1) carious demineralization on the mesial surface of a lower left 2nd premolar.*

D3 (DIAGNOcam): DIAGNOcam images showed all the thirty teeth with carious demineralization (Score 1) on both their mesial and distal surfaces, making a total of sixty surfaces with demineralized carious enamel (Score 1) as shown in figures (13&14).

D3: After 72 hours of demineralization

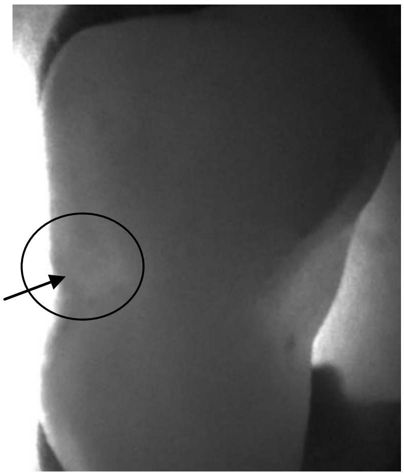


Figure13. *DIAGNOcam image showing (Score 1) carious demineralization of the mesial surface of an upper left 1st premolar.*

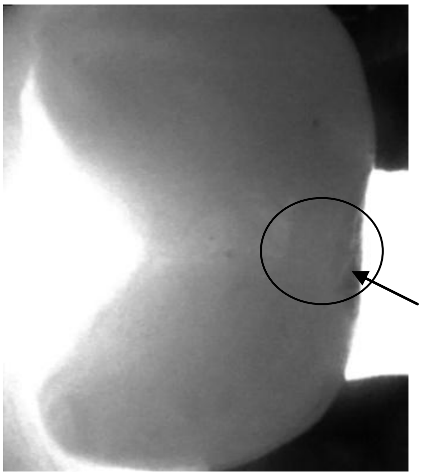


Figure14. *DIAGNOcam image showing (Score 1) carious demineralization of the mesial surface of the same upper left 1st premolar.*

All scanning electron microscopy images showed demineralized carious enamel with irregular pattern of surface destruction on both mesial and distal sides of all the three teeth appearing as hap hazardous thin splits as well as the typical honeycomb appearance indicating prismatic pattern destruction where the prism cores have been destroyed with remaining interprismatic substance which is less affected as shown in figure (15).

Scanning electron microscopy after 72 hours of demineralization

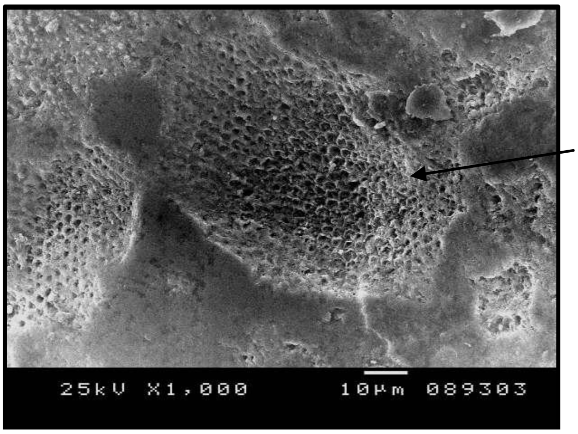


Figure15. *SEM image of the mesial side showing irregular pattern as well as prismatic pattern of destruction with the typical honeycomb appearance in a demineralized carious enamel (x1000)*

Statistical analysis showed the following:

Group I:

Accuracy of D1 (EDX) vs. D2 (X-ray): At both mesial and distal sides, statistical comparison of the two diagnostic means D1 (EDX)& D2 (X-ray) showed that D1 (EDX) is more accurate than D2 (X-ray) in detection of early carious demineralization and the differences are statistically significant. (X^2=60.00, p=0.000*)

Accuracy of D1 (EDX) vs. D3 (DIAGNOcam): At both mesial and distal sides, statistical comparison of the two diagnostic means D1 and D3 showed that D1 and D3 have a close accuracy in detection of early carious demineralization, and the differences are statistically insignificant. ($X^2_{(Y)}$=2.41, $p_{(Y)}$=0.120 NS).

Accuracy of D2 (X-ray) vs. D3 (DIAGNOcam): At both mesial and distal sides, statistical comparison of the two diagnostic means D2 and D3 showed that D3 is more accurate than D2 in detection of early carious demineralization and the differences are statistically significant. ($X^2_{(Y)}$=42.42, $p_{(Y)}$=0.000*).

Statistical comparison regarding the validity in detection of early carious demineralization showed that the overall accuracy of D3 (86.67%) is significantly higher than the accuracy of D2 (0.00%) within 95% confidence interval.

Group II:

Accuracy of D1 (EDX) vs. D2 (X-ray): At both mesial and distal sides, statistical comparison of the two diagnostic means D1 (EDX) & D2 (X-ray) showed that D1 (EDX) is more accurate than D2 (X-ray) in detection of early carious demineralization and the differences are statistically significant. ($X2(Y) = 48.82$, $p(Y) = 0.000$*).

Accuracy of D1 (EDX) vs. D3 (DIAGNOcam): At both mesial and distal sides, statistical comparison of the two diagnostic means D1 (EDX) and D3 (DIAGNOcam) was not applicable as both methods showed exact match in their results in detection of early carious demineralization. (NA).

Accuracy of D2 (X-ray) vs. D3 (DIAGNOcam): At both mesial and distal sides, statistical comparison of the two diagnostic means D2 (X-ray) and D3 (DIAGNOcam) showed that D3 (DIAGNOcam) is more accurate than D2 (X-ray) in detection of early carious demineralization and the differences are statistically significant. ($X2(Y) = 48.82$, $p(Y) = 0.000$*)

Statistical comparison regarding the validity in detection of early carious demineralization showed that the overall accuracy of D3 (DIAGNOcam) (100.00%) is significantly higher than the accuracy of D2 (X-ray) (6.67%) within 95% confidence interval.

Discussion

Detection of carious lesions on neighboring approximal surfaces of human teeth is a challenge (5), conventional methods of detecting caries are underestimating, causing some caries to go undetected until it has reached more advanced stages, requiring surgical intervention that destruct the tooth structure in order to restore it(14).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 cavitation, but instead, remineralize the existing lesions eliminating the need to restore the tooth(7). There is a need for a diagnostic tool that can clinically detect the earliest carious lesions in order to reach the goal of not restoring a tooth and instead treat and remineralize it.

Two laboratory (EDX and SEM) and two clinical (X-ray and DIAGNOcam) diagnostic tools used in our study. Calcium and Phosphorus ions ratios by EDX in all selected visually sound teeth were more than 1.80. This coincides with Ten Cateet al (9)andJaleviket al(10) who stated that a ratio equal to or above 1.8 was considered normal healthy enamel, while Shelliset al (15) and de Sant'Annaet al(16) stated that 1.67 was enough for the tooth to be considered healthy.

Digital radiography images obtained from the same previously visually examined sound teeth also showed images of sound enamel in all the specimens.

DIAGNOcam images obtained from the same previously visually examined sound teeth also showed images of sound enamel in all the specimens. Therefore, the thirty three teeth were accepted in the study. The SEM images obtained from the randomly selected specimens confirmed the results obtained from EDX, Digital X-ray and DIAGNOcam.

The same results were obtained by EDX coincided with those obtained by digital radiography, DIAGNOcam and SEM, this ranks DIAGNOcam as a diagnostic tool for detection of sound enamel.A dark room setting was used during DIAGNOcam detection to obtain reasonable images simulating the conditions of the oral cavity.

Our findings showed that; EDX used as a laboratory diagnostic tool for Calcium and Phosphorus ions concentration, detected all the changes in their concentrations, this may be due to the high sensitivity of this diagnostic tool, which coincides with Bloehaumet al(17) who stated that EDX measurements are used to obtain exact Ca/P ratios.

Statistical comparison between EDX and digital radiography showed that digital radiography had a much lower sensitivity in detecting radiolucency of early enamel demineralization and there is a statistically significant difference, this may be due to the fact that digital X-ray can't detect the earliest changes that occur on the surface of enamel, which coincides with Hintzeet al(18) who stated that radiography seemed to be of almost no value in the detection of the enamel approximal lesions.

DIAGNOcam images detected radiolucency of the early enamel demineralization with high sensitivity in comparison to EDX, that may be due to the penetration power of LASER beam at 780 nm, which has the ability to pass through the affected approximal enamel demineralization and image it. This was also confirmed by the results of the SEM, which may consider DIAGNOcam as sensitive as the EDX and SEM that are considered as gold standard. This coincides with Yuet al (19) who concluded that there was no significant difference between the depth of caries lesions checked with DIAGNOcam and the depth of the actual cavity. Also, Russottoet al (20) who concluded that near-infrared transillumination performed significantly better than radiography as an interproximal caries detection tool. Also, Marinova-Takorovaet al (21) who concluded 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. Also, Simonet al(22) who stated that near-infrared imaging was significantly more sensitive than radiography in detecting early carious lesions on both occlusal and interproximal surfaces, and Cirligeriuet al(23) who found that there is a good correlation between near-infrared transillumination and radiography but the former proved more accurate compared to the later. On the other hand Jostet al (24) disagreed and concluded that agreement with NIR-TI was worse compared to Bitewing digital radiography (BW). But, 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 SEM images obtained from the randomly selected specimens confirmed the results obtained from the EDX and DIAGNOcam, this may be due to the high sensitivity of DIAGNOcam in detection of early demineralized enamel.

Conclusions

1. Early demineralization of enamel carious lesions can't be detected by visual examination nor by digital radiography where digital near-infrared transillumination (DIAGNOcam) detection will help us to confirm it.
2. Near-Infrared Transillumination (DIAGNO cam) is a digital dental diagnostic device that will allow the application of the "rule not to restore" taking advantage of the depth of penetration of its LASER beam at 780 nm to detect the earliest stages of enamel demineralization to perform a true "micro-minimally invasive" line of treatment.
3. Near-Infrared Transillumination (DIAGNO cam) could provide us with a radiation free method that helps in the early detection of enamel carious lesions allowing us to diagnose and treat the affected surfaces instead of filling them.
4. Near-Infrared Transillumination (DIAGNO cam) can be used for monitoring the "corrective treatment" of the demineralized enamel carious lesions until sound enamel is obtained.

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