



Detection and Quantification of Class I Caries with Laser Fluorescence Technique

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Abstract: The objective of the present study is to verify the actual carious lesion depth by laser fluorescence technique using 650 nm CW diode laser in comparison with the histopathological investigation. Five permanent molar teeth were extracted from adult individuals for different reasons (tooth impaction, periodontal diseases, and pulp infections); their ages were ranging from 20-25 years old. Different carious teeth with varying clinical stages of caries progression were examined. An experimental laser fluorescence set-up was built to perform the work regarding in vitro detection and quantification of occlusal dental caries and the determination of its actual clinical carious lesion depth by 650 nm CW diode laser (excitation wavelength ($\lambda_{\text{excit.}}$) = 669 nm). Five teeth were sent to histopathological examination to confirm the efficacy of laser fluorescence technique for the determination of actual carious lesion depth. The results are leading to the detection of carious lesions for different depths. The deepest carious lesions revealed high fluorescence intensity. Based on these findings; it was concluded that 650 nm CW diode laser ($\lambda_{\text{excit.}}$ = 669 nm 40 mW) is a suitable and a reliable tool for caries diagnosis and depth assessment. Histopathological findings for the estimation of actual carious lesion depth revealed a good correlation with that of laser fluorescence technique.

Introduction

Dental caries is a dynamic process taking place in the microbial deposits on the tooth surface. It results in a disturbance of the equilibrium between the tooth surface and the surrounding plaque fluid so that, over time, the net results may be a loss of mineral from tooth surface (Mohammad B. 2003).

Decay on occlusal surface accounts for the majority of lesions in the permanent dentition of adolescents and adults (Bastingn RT. 1999). The complicated anatomy of the permanent molar occlusal surfaces makes the hygiene difficult with increased caries risk. It has been suggested that the occlusal lesions are initiated on fissure walls and are therefore difficult to detect (Heinrich-Weltzien R, et al., 2002). An accurate diagnosis of enamel decay is more challenging on occlusal than on proximal surfaces. The diagnosis of occlusal decay and its

clinical depth are highly subjective, and there is considerable variation in opinion among dentists as to appropriate diagnosis and treatment of early carious lesions on occlusal surface (Ferreria Z. et al., 1998). As accurate diagnosis of occlusal caries and its actual clinical carious lesion depth are difficult, advanced diagnostic technologies are emerging to meet the challenge of diagnosing occlusal decay.

Such advanced techniques include measurements of the scattered light, fiber optic transillumination, endoscopically viewed fluorescence, electrical conductance and quantitative laser- or light-induced fluorescence. These advanced technologies quantify changes in the physical characteristics of enamel related to demineralization (Dorothy M. and Laura E. 2001). Laser fluorescence of caries was demonstrated in the early 1980's. Subsequently many workers have modified the technique, demonstrating differences in fluorescent light

emitted from sound and carious tooth surfaces (Derek J. et al., 2001). Different types of laser like (Nitrogen laser λ : 337 nm, Argon ion laser λ : 488 nm and diode laser λ : 655 nm) have been used for early detection and diagnosis of dental caries taking the advantage of tooth autofluorescence (Alfheidur A. 2003).

Quantitative light-induced is probably the most extensively researched in the area of dental caries diagnosis. This method is based on the principle that mineral loss, caused by carious destruction of the tooth enamel can be detected and measured as a change in fluorescence of the tooth substance when exposed to laser light (Sofia T. 2002).

Hibst and Gall (1998), found that fluorescence induced by red light could differentiate between sound and carious tooth tissue. Fluorescence spectroscopic investigation that was done by Hibst et al. (2001) revealed considerable contrast between sound and carious tissue when excited by red light, 655 nm. Fluorescence was found to be more intense in carious tissue compared with sound tissue (Mohammad S. 2005). As a consequence, fluorescence intensity of tooth structure was increased by increasing the carious lesion progression. CW diode laser (λ : 650 nm, $\lambda_{\text{excit}} = 669$ nm) is used in this work to correlate carious lesion depth which was detected by laser fluorescence technique (LF) with histopathological examination.

Materials and Methods

The material comprised of 6 samples of permanent molars extracted from adult individuals for different reasons (tooth impaction, periodontal diseases, and pulp infections). Their ages were ranging from 20 to 25 years old. The occlusal surfaces were either visually intact occlusal, had non-cavitated lesions, or cavitated carious lesions.

The inclusion criteria for teeth in this study were apparent absence of:

1. Occlusal restorations.
2. Developmental defects (like enamel hypoplasia and hypoplastic pits).

Immediately following the extraction, the teeth were rinsed thoroughly using tap water and cleaned with pumice by a rubber cup and copiously washed with distilled water, and calculus was carefully removed with a scaler. Then they were numbered and stored under

room temperature in 10% formalin, in individual plastic containers.

CW diode laser (Dream laser, China) was used in this study with 650 nm wavelength and 40 mW output power ($\lambda_{\text{excit}} = 669$ nm), with a beam diameter of 3mm.

A specially designed experimental laser fluorescence set-up, as shown in Fig.1, was built in this study consisted of (650 nm CW diode laser system ($\lambda_{\text{excit}} = 669$ nm).

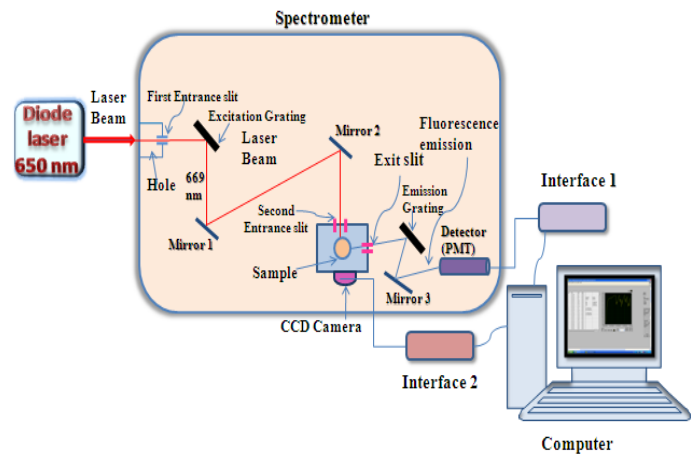


Fig. (1): Schematic drawing of the experimental set-up of laser fluorescence technique.

spectrometer, imaging CCD camera, PC with interface. This system was built to perform detection and quantification of occlusal dental caries and its actual clinical carious lesion depth.

The following experiment focused on the determination of the fluorescence spectra of carious teeth with different clinical grades of carious process progression in correlation with the actual clinical lesion depth. For that purpose, four teeth showing different clinical grades of progression of dental caries with one sound tooth were included in this experiment. To perform the work, this experiment was divided into two parts:

I. To differentiate between the fluorescence spectra of carious teeth, all of the four teeth were irradiated by using the diode laser with output power of 40 mW. The results with the fluorescence spectrum of sound tooth structure were compared.

II. To correlate the fluorescence spectra of carious teeth with the actual clinical lesion depth, all four carious teeth were sent to the laboratory for histopathological examination to estimate the clinical carious lesion depth of each one.

Histopathological Examination

Five teeth have been sent to laboratory for histopathological examination. To validate the results LF technique for estimation of actual carious lesion depth. Five teeth were sectioned for that purpose.

Regarding the histological preparation of teeth for ground section, all the studied teeth were embedded in acrylic for a distance occupied the whole length of the teeth. This was done to provide support for teeth to be sectioned longitudinally by minitome in a thickness of (50-60 μm). The sections were mounted on slides and examined under stereomicroscope at magnification of an X500.

Results and Discussion

The fluorescence spectra of carious teeth in correlation with that of sound tooth structure:

The fluorescence spectra of carious teeth with different clinical grades of carious process progression have demonstrated the following:

Figure 2 represents the fluorescence intensity in W/cm^2 as a function of the emitted wavelength in nanometers (nm).

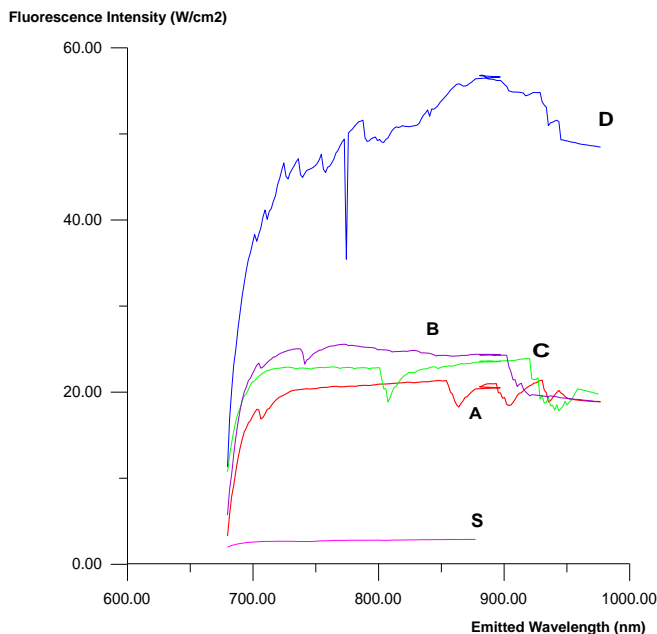


Fig. (2): Fluorescence spectrum of sound tooth (S) in comparison with the fluorescence spectra of different carious teeth showing different clinical grades of carious process progression (A, B, C, & D).

The results show that the fluorescence intensity level had increased with increasing the

extension of the clinical carious lesion depth within the tooth structure.

Brown spot lesions (B & C) revealed higher fluorescence intensity level than dull or shiny one (A). The highest fluorescence intensity level was recorded with tooth that was shown cavitation in opaque or discolored enamel exposing dentin (D). The fluorescence intensity levels of all carious teeth under investigation within this experiment (A, B, C, & D) were still higher than that of sound tooth (S).

On the basis of the fluorescence spectroscopic investigations that had been described before, red light of 650 nm CW diode laser ($\lambda_{\text{excit.}} = 669 \text{ nm}$) penetrated deeper into the tooth structure. This also helps to increase the depth that can be examined.

This comes in a good agreement with what was stated by of Hibst et al. (2001), Raimund H. and Robert P., (2001), as red light and IR fluorescence radiation are less absorbed and scattered by enamel than light of shorter wavelength. Increasing the fluorescence intensity level by increasing the clinical carious lesion depth can be attributed to the presence of diffusible bacterial metabolites fluorescing under red light excitation.

This hypothesis comes with an acceptable agreement with the results of an experiment that was done by Konig K. et al., (2001). According to their study, they tested the incubated bacteria from caries on blood agar and analyzed the grown colonies by fluorescence microscopy. Based on their results, not only the bacteria colonies showed fluorescence, but also the surrounding agar.

The fluorescence intensity levels of the four carious teeth were higher than that of the sound tooth. This indicates the presence of bacterial metabolites within the tooth structure which is considered to be the baseline of fluorescence. This finding is in a good agreement with that of Hibst et al. (2004). Based on their study, diode laser light induced fluorescence can differentiate between carious and non carious tooth tissue.

Histopathological Examination

After histopathological examination, one tooth (S) was sound as shown in Figures 3 and 4. Two teeth (A & C) had enamel caries as demonstrated in Figures 8, 9, and 10. Two teeth (B & D) had dentinal caries, as presented in Figures 5, 6, and 7.

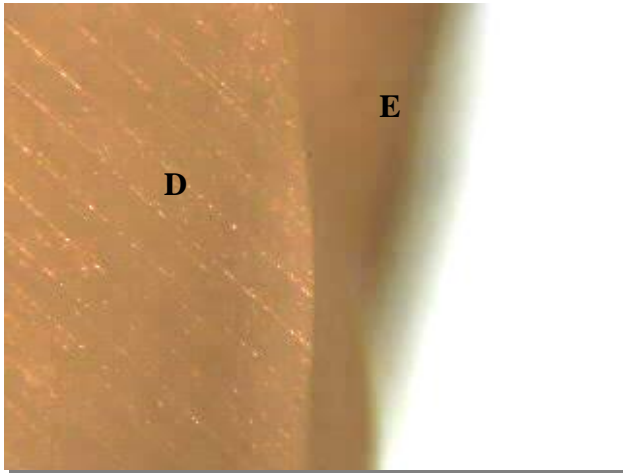


Fig. (3): Normal view for sound tooth showed enamel (E) and dentine (D). Ground section X500 (Tooth No. S)

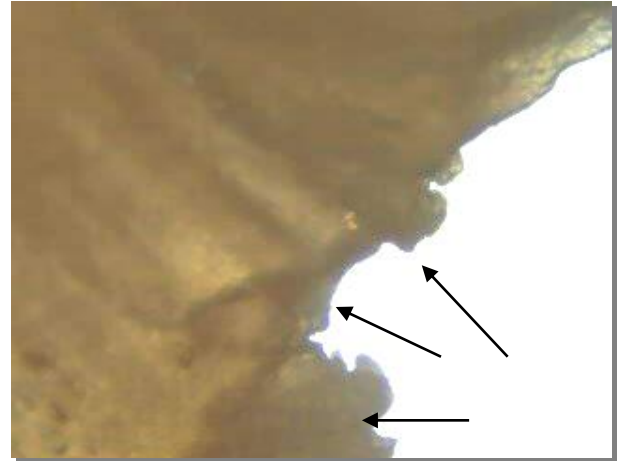


Fig. (6): Deep caries showed exposed dentinal tubules (arrows). Ground section X 500 (Tooth No. D)

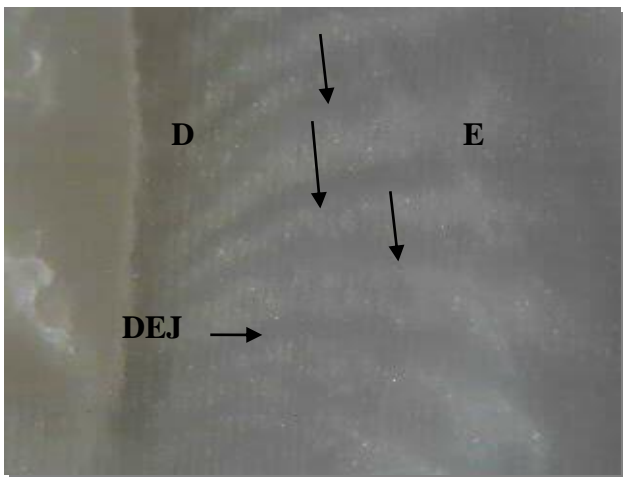


Fig. (4): Photograph view for normal sound tooth showed enamel (E) with Hunter Schenger band (arrow), dentine enamel junction (DEJ), dentine (D). Ground section X500 (Tooth No. S)

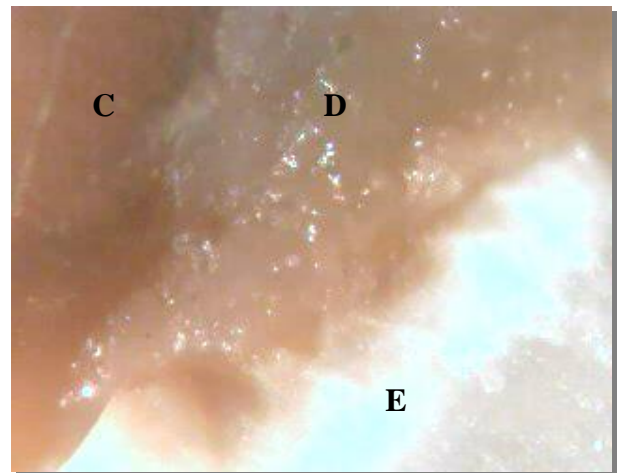


Fig. (7): Caries in dentine (C) showed changes in ideal histological feature of dentine. Enamel can be detected (E). Ground section X500 (Tooth No. D)



Fig.(5): Caries (C) extend beyond the Dentinoenamel junction (arrows). Ground section X500 (Tooth No. B)

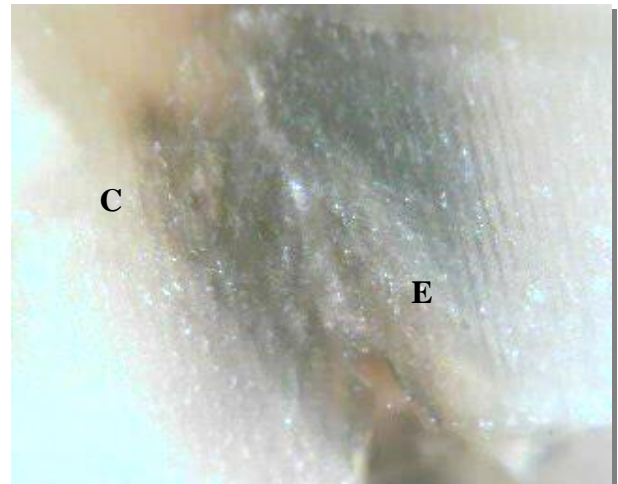


Fig. (8): Caries (C) as a lesion occupied whole thickness of enamel (E). Ground section X500 (Tooth No. C)

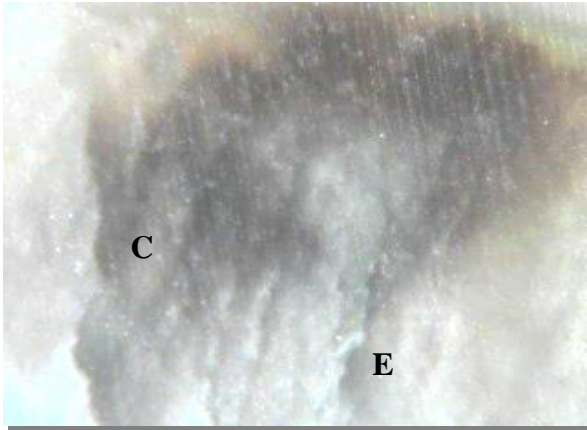


Fig. (9): Caries (C) destroying the histological feature of the enamel (E). Ground section X500 (Tooth No. C)

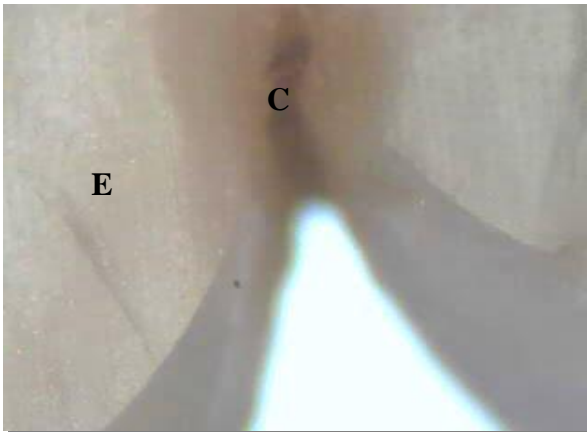


Fig. (10): Pit caries (C) showed in enamel surface (E). Ground section X500 (Tooth No. A)

The actual clinical carious lesion depth (histopathological depth) for each studied tooth was measured in millimeters, as demonstrated in Table 1. Histopathological carious lesion depth can be used to evaluate the results of the LF technique to estimate the actual clinical carious lesion depth for each studied tooth.

Table (1): The histopathological carious lesion depth

Tooth No.	Histopathological carious lesion depth (mm)
S	0
A	1.0 - 1.5 mm
B	6.4 mm
C	2.3 - 2.5 mm
D	8.5 - 9.0mm

The histopathological examination showed that the normal histological feature of the sound tooth revealed enamel, dentine, and DEJ; also Hunter Scherger bands could be detected in ground section, as shown in Figures 3 and 4.

Cariou tooth showed different feature, shape, and extension depending on caries itself and the procedure of treatment which was used.

As a consequence of that, caries could be identified in dentin, just beyond the DEJ, as shown in Figure 5 Deep caries can attack dentin and causes destruction to the dentinal tubules as in Figure 6, discontinuity and loss of normal curvature feature of the dentinal tubules can be distinguished.

Also caries can change the histological and characteristic features of dentine, as demonstrated in Figure 7, were mashed dentine, was destroyed by caries.

Caries could be identified by being occupying the whole thickness of enamel, as shown in Figure 8. Progressive loss of enamel structure could be distinguished by small regions where prisms worn away, as appeared in Figure 9.

Pit caries in enamel could be identified in deepest point of enamel grooves and extended for a short distance, as shown in Figure 10.

The results of the histopathological scoring showed that only one tooth is sound, three teeth showed enamel caries whether it is in inner or outer enamel. One tooth revealed caries in the outer dentine just beyond the DEJ, and one showed caries in the inner dentine just clearly beyond the DEJ, as demonstrated in Table 1.

The correlation between the results of LF technique, shown in Figure 2, and the histopathological scoring, demonstrated in Table1, that shows promising results regarding the assessment of carious lesion depth and both of them were not considerably different from one another. This result comes with a good agreement with what was reported by Shi et al. (2000), in which there was a considerable correlation between histopathology and the readings of KaVo DIAGNOdent.

On the basis of the correlation between the results of histopathological examination and those of LF technique, a LF scoring system can be utilized in correlation with the results of histopathological examination, as demonstrated in Table 2. The LF score represents the fluorescence intensity in W/cm².

Table (2): Laser fluorescence scoring system for the carious lesion depth

LF score	Carious lesion depth
1-3	Sound
4-24	Enamel caries
≥ 25	Shallow dentinal caries
≥ 40	Deep dentinal caries

In the present study, LF scores (1-3) and (4-24) for sound and enamel caries respectively are not quite different in comparison with the cut-off values that were suggested by Lussi et al. (2001). Based on their preliminary study that a score of (0-7) was suggested for sound tooth, and a score of (8-21) was recommended for enamel caries. A score of (> 21) was suggested by Lussi et al. (2001) for the detection of shallow dentinal caries (caries in the outer half of dentin). This cut-off value is not quite different from what was reported in the current study as it is ≥ 25. On the basis of the results of the present study, the LF score of (≥ 40) was suggested for the detection of deep dentinal lesions. This score is not slightly different from what was proposed by Heinrich-Weltzien et al. (2003), which was > 37 for deep dentinal caries. As a consequence of that, the detection of a carious lesion and the estimation of its depth are considered to be important parts in the diagnostic process. This is because if the lesion is active. This is often instrumental in determining the choice of intervention (Cortes D. F. et al., 2000, Denise M. Z. et al., 2007 and Karlsson L. et al., 2009). According to the present study, a correlation of the LF scoring with the possible intervention can be done, as demonstrated in Table 3. This is in a good agreement with what was stated by Young DA (2002) (Young DA. 2002) regarding the management of dental caries.

Table (3): Correlation of LF scoring with possible interventions

LF score	Intervention
1-3	no intervention
4-24	record and monitor/ sealant application/ preventive resin application
≥ 25	restoration
≥ 40	Deep dentinal caries

Conclusions

CW diode laser ($\lambda = 650 \text{ nm}$, $\lambda_{\text{excit.}} = 669 \text{ nm}$, 40 mW output power) is a reliable tool for clinical caries diagnosis. The histopathological results proved a good accordance of the carious lesion depth measurement by laser fluorescence technique.

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الكشف الكمي للتسوس من النوع الأول باستخدام تقنية التألق بواسطة الليزر

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الخلاصة

الهدف من هذه الدراسة هو التحقق من مدى عمق تسوس الأسنان بواسطة تقنية التألق الإنبعائي لليزر باستخدام ليزر داوود (650 نانومتر) ذو الموجة المستمرة بالمقارنة مع التشخيص النسيجي. خمسة سنًا قُطعت من أشخاص بالغين تتراوح اعمارهم من 25 الى 45 سنة، لأسباب مختلفة (انحشار السن، امراض اللثة، التهابات لب السن). لقد تمَّ صنع منظومة مختبرية للطيف الإنبعائي لليزر من أجل اتمام متطلبات العمل للكشف الكمي عن التسوس الإطباقى للإنسان خارج الجسم بواسطة ليزر داوود (650 نانومتر) ذو الموجة المستمرة وبطول موجي تحفيزي (669 نانومتر). خمسة أسنان أُرسلت الى الفحص النسيجي للتأكد من فعالية الطرق التشخيصية المستخدمة في هذه الدراسة في تحديد العمق الفعلي للتسوس. أدت هذه النتائج الى الكشف عن التسوس وبأعماق مختلفة. لقد أظهرت الأسنان المصابة بالتسوس بعمق كبير شدة عالية للطيف الإنبعائي لليزر. استناداً الى هذه النتائج، يُستنتج أنّ ليزر داوود (650 نانومتر) ذو الموجة المستمرة وبطول موجي تحفيزي (669 نانومتر) وبقدرة خارجية مقدارها 40 ملي واط يُعتبر اداة مناسبة وموثوقة لتشخيص التسوس وتقييم عمقه. أظهرت نتائج الفحص النسيجي لتحديد عمق تسوس الأسنان ارتباطاً جيداً مع نتائج تقنية الطيف الإنبعائي لليزر.