ORIGINAL ARTICLE

In vitro comparison of laser fluorescence performance with visual examination for detection of occlusal caries in permanent and primary molars

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Abstract The aim of this study was to compare the performance of the DIAGNOdent 2095 with visual examination for occlusal caries detection in permanent and primary molars. The sample comprised 148 permanent human molars and 179 primary human molars. The samples were measured and visually examined three times by two examiners. After measurement, the teeth were histologically prepared and assessed for caries extension. Sensitivity, specificity, accuracy and area under the receiver operating characteristics (ROC) curve were calculated. Intra-class correlation (ICC), unweighted kappa and the Bland and Altman method were used to assess inter- and intra-examiner reproducibility. DIAGNOdent showed higher specificity and lower sensitivity than did visual examination. The ICC values indicated an excellent agreement between the examinations. Kappa values varied from good to excellent for DIAGNOdent but from poor to good for visual examination. In conclusion, the DIAGNOdent may be a useful adjunct to conventional methods for occlusal caries detection.

Keywords Primary teeth · Permanent teeth · Laser fluorescence · DIAGNOdent · Visual examination · Occlusal caries detection

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Introduction

Occlusal caries detection is a difficult task in dentistry. Occlusal incipient lesions have become difficult to be detected due to the widespread use of fluorides and its superficial remineralization potential that seems to delay cavitation. Several methods have been developed and recommended as diagnostic aids to identify and quantify early caries lesions on smooth and occlusal surfaces [1].

Quantitative laser/light-induced fluorescence (QLF) measures intrinsic fluorescence of the teeth and has been mainly used on smooth surfaces to quantify minor mineral changes [2, 3]. The laser fluorescence device (DIAGNO-dent 2095, KaVo, Biberach, Germany) was developed in order to support dentists in the detection of non-cavitated enamel as well as hidden dentin caries lesions [1, 4–9]. This device has the ability to emit red light at 655 nm wavelength and to capture the fluorescence emitted by oral bacterial metabolites. This information is translated to a numerical scale varying from 0 to 99. The higher the number, the deeper the caries lesion [10]. Therefore, a direct relationship between the numerical result and the lesion depth can be established.

Although much evidence has shown the promising results proposed by the DIAGNOdent [11], some potential confounding findings that complicate interpretation of the values have been pointed out. It has been demonstrated that plaque, calculus, toothpaste, dental materials, stains, hypomineralized non-carious teeth or incorrect calibration show false-positive results [9, 12–14].

The aim of this in vitro study was to compare the performance of the DIAGNOdent 2095, which is able to emit red light at a wavelength of 655 nm, with visual examination for occlusal caries detection in permanent and primary molars.

Materials and methods

This study was approved by the Ethics Committee of the School of Dentistry, São Paulo State University (UNESP), Araraguara, Brazil (report no. 85/04). One hundred fortyeight permanent human molars and 179 primary human molars, suspected to have initial caries lesions on their occlusal surfaces, were selected. An independent senior researcher, who did not take part in the study as an examiner, selected the sites exhibiting suggestive signs of caries lesions, such as white spots, stained fissures or surfaces and microcavities (small surface defects), by observing only the teeth. After prophylaxis with water/ pumice slurry the teeth were carefully washed for 15 s with a 3-in-1 syringe to remove any remnants inside the fissure that could influence the laser fluorescence readings. Then, the teeth were stored in thymol solution at 4°C [15]. Afterwards, the occlusal surfaces were photographed with a digital camera (Sony CyberShot 717) and the photographs were mounted onto cards. Only one site per tooth was selected for the laser fluorescence measurements and visual examination.

The laser fluorescence measurements were carried out with the DIAGNOdent 2095 (KaVo). The measurements as well as visual examination were performed three times by two calibrated examiners observing a 1-week interval between the evaluations. The probe tip selected for the DIAGNOdent was the cone-shaped one (tip A), indicated for pits and fissure areas. Dry, clean surfaces were inspected by direct visualization and under excellent illumination, without probing, and the visual examination was coded according to the criteria shown in Table 1. The teeth were not allowed to become dehydrated at any time.

Prior to the laser fluorescence measurements, for each individual tooth, the device was calibrated against a ceramic reference (standard calibration) according to the manufacturer's specifications, and, afterwards, the zero value of fluorescence was obtained from a sound part of the buccal surface. The probe tip was placed perpendicularly on the test site and moved until the maximum value (peak) was reached, and this value was recorded. From the peak value, the zero value fluorescence was subtracted. This peak value was then correlated with the definitions of a scale supplied by Lussi and Hellwig [8], which corresponds to the absence or presence of a carious lesion, as well as to its degree of progression, as follow: 0–7, sound; 7.1–14, caries in the enamel; 14.1–24, caries in the dentin–enamel junction; >24, caries in the dentine.

For validation of the results, the teeth were bisected buccolingually through the center of the carious lesion with a water-cooled diamond disk at low speed (KG Sorensen 7020). The deepest areas of the caries lesions were examined with the aid of a stereoscopic magnifying lens

Table 1 Criteria for visual examination

Score	Visual examination	Criteria
0	Sound (D ₀)	Normal texture of enamel
1	Caries lesion in enamel (D ₁ , D ₂)	Opacity or discoloration, rough surface, without loss of substance
2	Caries lesion in the outer half of dentin (D ₃)	Opacity or discoloration with microcavities and soft consistency and darkened dentin subsurface surrounding enamel fissure
3	Caries lesion in the inner half of dentin (D ₄)	Opaque and soft fissure. Usually there is a loss of substance

at \times 32 magnification (Carl Zeiss Jena GmbH, Jena, Germany). These analyses were carried out by an independent senior researcher (who was not involved as an examiner), according to the following scores: 0, no caries present; 1, demineralization involving the enamel; 2, demineralization involving the outer half of the dentin; 3, demineralization involving the inner half of the dentin. For histological criteria, the cut-off point between 1 and 2 was determined when the caries lesion reached the enamel– dentin junction.

The statistical analyses were performed with MedCalc for Windows, version 9.3.0.0 (MedCalc Software, Mariakerke, Belgium) and Excel 2002 (Microsoft Corporation, Redmond, WA, USA). The average of all six DIAGNOdent measurements was used for the calculation, and the significance level was set at P < 0.05. We assessed the reproducibility by calculating Lin's intra-class correlation (ICC) [16] and unweighted Cohen's kappa values for both intra- and interexaminer agreement. The ICC was assessed as poor when the values were below 0.40, fair for values between 0.40 and 0.59, good for values between 0.60 and 0.75, and excellent for values above 0.75. Kappa values above 0.75 denoted excellent agreement, while values between 0.40 and 0.75 indicated good agreement [17]. Furthermore, the receiver operating characteristics (ROC) method was used, and values of sensitivity, specificity, accuracy and area under the ROC curve were obtained, in accordance with the cut-off limits suggested by Lussi and Hellwig [8] to determine the presence or absence of occlusal caries. Additionally, optimal cut-off limits were obtained through the ROC curve analysis by the highest sum of sensitivity and specificity at each threshold $(D_1, D_2 \text{ and } D_3)$ as well for the primary as for the permanent teeth. For the DIAGNOdent values, we used the Bland and Altman method to identify systematic differences, and the 95% limits of agreement were calculated [17, 18]. For paired readings (intra- and inter-examiner), the difference between the measurements was plotted against their means (mean difference \pm 1.96 SD).

Results

For the 148 occlusal test sites of primary teeth analyzed in this study, the lesion depth measurements revealed that 12 of them were caries free (D₀), 96 had caries extending up to halfway though the enamel (D₁), 35 had caries extending into the inner half of the enamel (D₂), and five had caries in the dentin (D₃,D₄). Of the 179 permanent teeth, 12 were caries free (D₀), 105 had caries extending up to halfway though the enamel (D₁), 43 had caries extending into the inner half of the enamel (D₂) and 19 had caries in the dentin (D₃,D₄).

The values for specificity, sensitivity, accuracy and area under the ROC curve (A_z) are shown in Table 2, where it can be observed how accurate the DIAGNOdent and visual examination were, as well for primary teeth as for permanent teeth. The DIAGNOdent showed values of specificity varying from 0.72 to 0.94 and sensitivity from 0.20 to 0.69. The values for visual examination varied from 0.20 to 0.98 (specificity) and from 0.13 to 0.97 (sensitivity), on depending on the threshold analyzed. Due to this enormous variation, new cut-off limits were obtained for the primary teeth, which can be observed in Table 3.

The reproducibility assessed by calculating the ICC and unweighted Cohen's kappa is represented in Table 4. ICC values for DIAGNOdent varied from 0.90 to 0.92, as well for inter as for intra-examiner reproducibility, indicating an excellent agreement between the examinations. Kappa values varied from good to excellent for DIAGNOdent, but from poor to fair for visual examination, for both intra and inter-examiner reproducibility.

Finally, the Bland and Altman plot (Fig. 1) showed the limits of agreement, in which 95% of the laser fluorescence measurements could be repeated. The range between the

upper and the lower limits of agreement (\pm 1.96 standard deviation) was 25.8 and 23.5 for primary teeth and 31.2 and 28.8 for permanent teeth (both for intra- and inter-examiner reproducibility, respectively).

Discussion

Occlusal caries is difficult to be detected due to the specific morphology of the pits and fissures and to the changes in the clinical patterns of the disease. Plaque retention and the difficulty of its removal have increased the prevalence of incipient lesions in occlusal surface. Furthermore, occlusal dentinal caries has been observed under a fissure which seems intact to the naked eye due to the use of fluorides [4], making the detection of such lesions difficult by conventional methods (visual examination and radiography).

An ideal diagnostic method should have high sensitivity and high sensitivity [19] and should also be reliable and valid, with good intra- and inter-examiner agreement [20]. In our study, the efficiency of the DIAGNOdent, which has been used for occlusal caries detection in several studies [8, 12, 21–24], was compared with that of visual examination in primary and permanent molars. The most recent investigations have shown that the DIAGNOdent seems to be a reliable method for occlusal caries detection [8, 22].

For both permanent and primary teeth, high values for specificity at the D_1 threshold were found. However, the small amount of sound surfaces observed could have influenced these values.

For primary teeth, although the DIAGNOdent presented high values of specificity, the sensitivity was low. This low value could be explained by the cut-off limits used for this calculation, once the area under the ROC curve had shown high values. The ROC curve analysis does not take a stipulated cut-off point and summarizes the results of several cut-off points [12]. For this reason, optimal cut-off limits were calculated considering all examinations of the

Table 2 Specificity, sensitivity, accuracy and area under the ROC curve (A_z) for occlusal caries detection using the cut-off limits suggested by Lussi and Hellwig [8]

Method		Specificity		Sensitivity		Accuracy		Az					
		D_1	D ₂	D ₃	D_1	D ₂	D ₃	D_1	D_2	D ₃	D_1	D ₂	D ₃
DIAGNOdent	Primary	0.92	0.93	0.94	0.24	0.20	0.20	0.30	0.73	0.92	0.71	0.71	0.82
	Permanent	0.92	0.75	0.89	0.53	0.39	0.16	0.56	0.63	0.81	0.83	0.64	0.68
Visual	Primary	0.20	0.86	0.98	0.95	0.31	0.13	0.89	0.71	0.95	0.59	0.60	0.78
	Permanent	0.40	0.79	0.98	0.97	0.56	0.30	0.93	0.71	0.91	0.75	0.70	0.75

 $D_1: D_0 =$ sound $D_1-D_3 =$ decayed

 D_2 : D_0, D_1 = sound; D_2, D_3 = decayed

 D_3 : D_0 - D_2 = sound; D_3 = decayed

Histology	Primary teeth		Permanent teeth		
	Cut-off limit	Highest sum of sensitivity/specificity	Cut-off limit	Highest sum of sensitivity/specificity	
D_0	0–2	_	0–5	_	
D_1	2-6	72.8/75.0	5–9	65.5/91.7	
D_2	7-11	57.5/77.8	9–12	66.1/65.8	
D _{3,4}	>11	80.0/77.6	>12	84.2/55.6	

Table 3 Optimal cut-off limits of the DIAGNOdent for primary teeth and permanent teeth

whole sample of primary teeth and the lesion depth measurements. Lower values of fluorescence were observed in the primary teeth, probably due to the different levels of mineralization and morphology of the enamel crystals. For this reason, optimal cut-off limits could be suggested for occlusal caries detection in primary teeth. In the same way, the optimal cut-off limits obtained for permanent teeth were lower than the values suggested by Lussi and Hellwig [8]. The storage method could have influenced the measurements in our study, once the teeth had been stored in thymol solution at 4°C [15].

Visual examination showed very low specificity but high sensitivity for caries detection in enamel, and the opposite for dentin, for both primary teeth and permanent teeth. This confirmed that changes in enamel were easily detected by visual examination, while many dentin lesions could have been covered up by the mineralized enamel.

In general, the DIAGNOdent showed higher specificity than did visual examination, in agreement with the findings of Reis et al. [25]. However, in a meta-analysis, Bader and Shugars [11] reported higher sensitivity and lower specificity for this device when it was compared with the visual examination, and Alwas-Danowska et al. [20] did not find any statistically significant difference between them. Lussi and Hellwig [8], who stored the teeth frozen and performed the histological analysis using rhodamine B, found higher values for sensitivity and lower ones for specificity. They found values of 0.96, 0.88 and 0.81 for sensitivity at threshold D_1 , D_2 and D_3 , respectively. However, the values for specificity were 0.69, 0.69 and 0.79 (lower than ours). A comparison between DIAGNOdent, visual and radiographic examination has recently been performed in vivo. The authors concluded that the DIAGNOdent device may be a useful supplement to visual examination, and its diagnostic performance seems to be good for occlusal caries detection [22]. It is important to point out that a high value of fluorescence may indicate caries as changes in the physical properties of the tooth structure, which could increase the value of sensitivity as a false-positive result. However, in our study, teeth with such alterations were not included in the sample.

As regards reproducibility, the ICC calculated for the DIAGNOdent showed an excellent correlation for both intra- and inter-examiner agreement for primary teeth and for permanent teeth. Additionally, kappa values confirmed the good reproducibility of the DIAGNOdent. These results are in agreement with those found by Kühnisch et al. [23], who observed excellent agreement using the DIAGNOdent and the new DIAGNOdent *pen*. Lussi and Hellwig [8] also observed excellent values for the ICC (>0.98) for both devices, and kappa values varying from 0.83 for intra-examiner reproducibility, in agreement with those of this study. High ICC values were also observed by Alwas-Danowska et al. [20], who assessed the reproducibility of the device. The good reproducibility means that the device could be used for monitoring the carious process [8].

The Cohen's kappa values for visual examination found in this study were low for both intra- and inter-examiner reproducibility, although they were considered as a good agreement as well (save for the inter-examiner agreement for primary teeth, which was poor). It should to be considered that these results involve subjective aspects

Table 4 Unweighted kappa values and ICC for inter and intra-examiner reproducibility of DIAGNOdent and visual examination for occlusal caries detection

		Inter-examiner		Intra-examiner		
		Kappa	ICC	Kappa	ICC	
DIAGNOdent	Primary	0.77 (excellent)	0.92 (excellent)	0.67 (good)	0.91(excellent)	
	Permanent	0.59 (good)	0.90 (excellent)	0.56 (good)	0.91(excellent)	
Visual	Primary	0.19 (poor)	-	0.45 (good)	_	
	Permanent	0.30 (fair)	-	0.65 (good)	-	

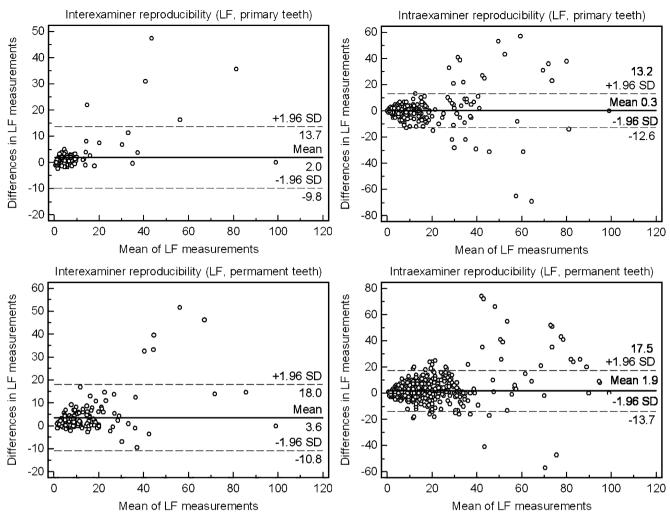


Fig. 1 The Bland and Altman method for assessing intra- and inter-examiner reproducibility. The range between the upper and the lower limits of agreement (*dashed lines*, mean difference \pm 1.96 SD) corresponds to the interval in which 95% of all measurements were reproducible

such as knowledge and clinical experience of the examiners, which can affect the intra- and inter-examiner agreement [22, 26]. Therefore, the difference found between these results could be due to the subjectivity of this examination as well as the different clinical experiences of the examiners.

The Bland and Altman method was used also to assess the reproducibility of the DIAGNOdent. This method has been indicated and readily incorporated to enhance the quality of research reports in dentistry [27]. This statistic should not have any systematic deviation (mean of differences = 0), and there should be only a small range between the upper and the lower limits of agreement [24]. Through this graph a range between ± 1.96 SD from the mean of the difference in the values can be observed. Kühnisch et al. [23] suggest that the deviation should not be greater than \pm 20 laser fluorescence units (range of 40 units), which is in agreement with the values found in our study. It can also be observed that most of the points are closer to each other and were obtained for the lowest values of fluorescence, confirming the good reproducibility of this device.

Finally, the good performance of the DIAGNOdent could be confirmed for permanent teeth, but association with the conventional methods seemed to be a better condition to detect those lesions. For primary teeth, considering their differences from the permanent dentition, further in vitro and in vivo studies should be carried out to test this device, using new cut-off limits, comparing it with other methods. In conclusion, the DIAGNOdent may be a useful adjunct to conventional methods for the detection of occlusal caries.

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