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Detection of carious lesion by laser speckle analysis using the first order moment

Detecção de lesão de cárie por análise de laser speckle usando momento de primeira ordem

Detección de lesiones cariosas mediante análisis de láser speckle utilizando el momento de primer orden

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Abstract

Carious lesion is one of the most prevalent diseases in humankind, affecting nearly all human beings at least once in a lifetime. Early carious lesion (ECL, also known as white spot lesion) is part of the caries process in which the enamel surface loses mineral but the subsurface layer overlying the mineral-poor region remains intact. In its early stages, the detection of the carious lesion is currently still a major challenge because it is based on visual inspection, thus with an inherent subjectivity. In this work, we present a method to detect and assess the severity of ECLs using a computer-assisted laser speckle imaging technique. Forty-five polished and cut bovine incisors samples allow a paired t-test by exposing half of the surface while protecting the other half. The samples, included in a PVC tube, were immersed in 50ml of a solution (pH 5.0) containing 0.05M acetate buffer solution and 50% saturated hydroxyapatite enamel powder at 37°C. The etching time is 24h; 48h and 72h; each group contain 15 samples. We illuminate each sample with a laser and analyse the first momentum of the laser speckle images with a computer. Our results shows that the first momentum of the LSI of the sound region statistically differs from the decay region of the samples for all groups ($p<0.0001$). We also found a strong correlation between the acid etch duration (severity of the decay) and the shift in LSI contrast (Pearson's correlation coefficient $\rho=0.9989$, $R^2=0.9978$). Detecting decay in its early stages is still a challenge in the clinical practice, however this work demonstrates that the analysis of the statistical features of the laser speckle image in the spatial domain allows for the detection of microstructural changes in the enamel associated with the presence of the lesion even before any intervention is required.

Keywords: Laser speckle imaging; Diagnostic; Computer vision; Dental Caries.

Resumo

A lesão de cárie é uma das doenças mais prevalentes na humanidade, afetando quase todos os seres humanos pelo menos uma vez na vida. A lesão de cárie precoce (ECL, também conhecida como lesão de mancha branca) é parte do processo de cárie no qual a superfície do esmalte perde mineral, mas a camada subsuperficial que recobre a região pobre em minerais permanece intacta. Em seus estágios iniciais, a detecção da lesão cariosa ainda hoje é um grande desafio, pois é baseada na inspeção visual, portanto, com uma subjetividade inerente.

Neste trabalho, apresentamos um método para detectar e avaliar a gravidade de lesão de carie usando uma técnica de imagem por laser speckle assistida por computador. Quarenta e cinco amostras de incisivos bovinos polidos e cortados permitem um teste t pareado, expondo metade da superfície e protegendo a outra metade. As amostras, incluídas em tubo de PVC, foram imersas em 50ml de uma solução (pH 5,0) contendo solução tampão acetato 0,05M e pó de esmalte com hidroxiapatita saturada 50% a 37 ° C. O tempo de corrosão é de 24h; 48h e 72h; cada grupo contém 15 amostras. Iluminamos cada amostra com um laser e analisamos o primeiro momento das imagens do laser speckle com um computador. Nossos resultados mostram que o primeiro momento do LSI da região do som difere estatisticamente da região de decaimento das amostras para todos os grupos ($p < 0,0001$). Também encontramos uma forte correlação entre a duração da corrosão ácida (gravidade da deterioração) e a mudança no contraste LSI (coeficiente de correlação de Pearson $\rho = 0,9989$, $R^2 = 0,9978$). Detectar cáries em seus estágios iniciais ainda é um desafio na prática clínica, porém este trabalho demonstra que a análise das características estatísticas da imagem speckle a laser no domínio espacial permite a detecção de alterações microestruturais no esmalte associadas à presença de a lesão antes mesmo de qualquer intervenção ser necessária.

Palavras-chave: Imagem laser Speckle; Diagnóstico; Visão computacional; Cárie dentaria.

Resumen

La lesión cariosa es una de las enfermedades más prevalentes en la humanidad y afecta a casi todos los seres humanos al menos una vez en la vida. La lesión cariosa temprana (ECL, también conocida como lesión de mancha blanca) es parte del proceso de caries en el que la superficie del esmalte pierde mineral, pero la capa subsuperficial que cubre la región pobre en minerales permanece intacta. En sus etapas iniciales, la detección de la lesión cariosa sigue siendo un gran desafío en la actualidad porque se basa en la inspección visual, por lo que tiene una subjetividad inherente. En este trabajo, presentamos un método para detectar y evaluar la gravedad de las ECL utilizando una técnica de imagen de moteado láser asistida por computadora. Cuarenta y cinco muestras de incisivos bovinos pulidos y cortados permiten una prueba t emparejada al exponer la mitad de la superficie mientras se protege la otra mitad. Las muestras, incluidas en un tubo de PVC, se sumergieron en 50 ml de una solución (pH 5,0) que contenía una solución tampón de acetato 0,05 M y polvo de esmalte de hidroxiapatita saturada al 50% a 37ºC. El tiempo de grabado es de 24 h; 48h y 72h; cada grupo contiene 15 muestras. Iluminamos cada muestra con un láser y analizamos el primer impulso de las imágenes de motas láser con una computadora. Nuestros resultados muestran que el primer impulso del LSI de la región de sonido difiere estadísticamente de la región de decaimiento de las muestras para todos los grupos (p <0,0001). También encontramos una fuerte correlación entre la duración del grabado ácido (severidad de la descomposición) y el cambio en el contraste LSI (coeficiente de correlación de Pearson $\rho = 0.9989$, $R^2 = 0.9978$). La detección de la caries en sus primeras etapas sigue siendo un desafío en la práctica clínica, sin embargo, este trabajo demuestra que el análisis de las características estadísticas de la imagen de moteado láser en el dominio espacial permite la detección de cambios microestructurales en el esmalte asociados con la presencia de la lesión incluso antes de que se requiera cualquier intervención.

Palabras clave: Imágenes de láser speckle; Diagnóstico; Visión por computadora; Caries dental.

1. Introduction

Although easily treated, carious lesion would affect all humans at least once in a lifetime making it one of the prevalent humankind diseases. A significant reduction in the prevalence, incidence, and severity of caries has been shown in the last three decades; however, this reduction is dashed by disparities because lower socio-economic groups are still at high risk of developing dental caries (WHO, 2003).

Human oral pH is normally below, yet very near, the point at which tooth mineral begins to dissolve. Therefore, a slight change in pH, due to acidic food for example, is exposing teeth to mineral loss. In contrast, some conditions favour the re-deposition of mineral and tooth repair Throughout this relentless irregular cycle of demineralization and remineralization, the net loss or gain in mineral over time ultimately determines whether tooth decay will advance, stabilize or regress (Langhorst, O'Donnell, & Skrtic, 2009) Thus, early caries lesions (ECL) are formed during alternating periods of demineralization and remineralization (Silverstone, 1977) where demineralization prevails. If this process continues, the result will be cavitation. Prior to cavitation, the subsurface lesion presenting partial demineralization can be remineralized (Arends and Christoffersen, 1986). In another words, an early detection of ECL and timely intervention prevent cavitation.

ECLs can be considered the stage of the caries process where mineral has been lost from the enamel subsurface, with an intact surface layer overlying the mineral-poor region (Torres *et al*, 2011). Clinical assessment would classify these lesions as either active or inactive (Nyvad, Machiulskiene and Baelum 1999) When these lesions are active, noncavitated demineralized areas appear chalky and "whitish", thus called "white spot lesions" (WSL) (Mosby, 2003, Ismail *et al*, 2007). The International Caries Detection and Assessment System (ICDAS) developed criteria to differentiate the various lesions stages (sound enamel, white spot lesions, and cavitated lesions), using visual-tactile assessment, to simplify the choice of an adequate therapy (Ismail *et al*, 2007). However, a visual clinical detection alone is far from unfailing because it depends on the operator eyes' quality. On the other hand, calibrating the operators is not a viable solution.

Radiographs can be associated to ICDAS to improve the detection outcome. However, radiographs cannot see ECL unless there is at least 30-40% mineral loss, meaning that the tooth decay moved already to another stage (White & Pharoah, 2009). No existing techniques allows a timely detection of ECL. The diagnosis is typically successful for late states only when an invasive treatment is required with significant hard tissue removal. To avoid the invasive treatment due to significant mineral loss, a reliable method for early detection is imperative (Estellano, 2007).

Previous studies (Bakhmutov *et al*, 2004) showed that the interaction between light and teeth (absorption, scattering, and fluorescence) is related to the constitution of the dental tissue. Carious lesion induces mineral loss and, hence, changes in the optical properties of the affected tissue, so the study of these properties may produce novel, non-invasive, non-destructive methods for the early diagnosis of the carious lesion (Kishen *et all*, 2008, Karlsson, 2010).

Several diagnostic techniques based on the optical properties of dental tissue have been developed for this purpose, such as quantitative light-induced fluorescence (QLF), optical coherence tomography (OCT) and fiber optic transillumination (LF) (Karlsson, 2010). However, these aforementioned techniques present some limitations regarding intraoral use, and some methods show high cost, which may be unavailable for clinical practice (Araújo *et al*, 2020; Cruz *et al*, 2020).

When a coherent light illuminates a rough surface, it causes a diffuse reflection, which generates an optical phenomenon called laser speckle. By capturing the reflected light of the surface, it is possible to quantify this speckle pattern using statistical theory. Previous studies that compare optical speckle patterns generated by healthy and injured dental tissues showed the efficacy of detection of white spot lesions by speckle coherent light scattering (Deana *et al*, 2013). However, it is interesting to go beyond and detect subsurface lesions with a subtle mineral loss, and not only in a substantial mineral loss as occurs in the white spot lesions.

Either so, even if demineralization is not visible, dry or wet, it is important that initial demineralization be detected in all possible areas to the finalize choice of treatment.

Our previous studies demonstrate that laser speckle image analysis is a non-invasive, non-destructive, real-time, and cost-effective technique capable of enhancing the contrast between sound dental tissue and a carious lesion in the early stage, but with a more clinically visible white spot lesion (Deana *et al*, 2013, Koshoji *et al* 2015A, Koshoji *et al* 2015B, Koshoji *et al* 2016),.The application of first-order spatial statistics to laser speckle imaging allows the detection of early carious lesions based on the optical properties of the enamel of eroded samples (Deana *et al,* 2013). and *ex-vivo* human samples (Koshoji *et al* 2015A, Koshoji *et al* 2015B, Koshoji *et al* 2016) In this work, we demonstrate the usage of this approach to a more comprehensive and controlled set of artificially induced early carious lesions.

2. Metodologia

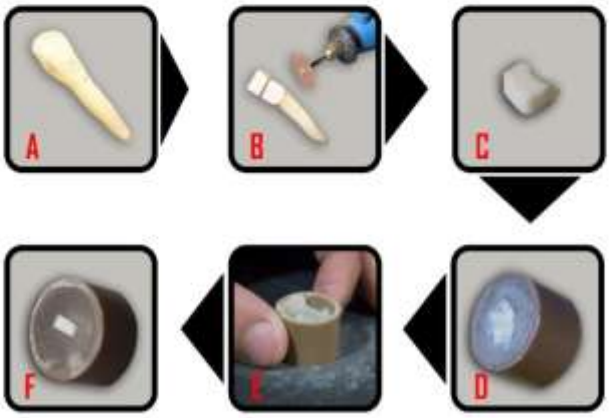
This is a quantitative experimental research

2.1 Sample preparation

A fragment of the vestibular surface of bovine incisor of 4 x 6 mm² was cut and included in a PVC tube with acrylic resin, exposing a flat and parallel surface of dental enamel. Each sample was polished using sandpapers with different grits, which range from 600 to 2000, in belt orbital sander followed by a liquid polishing (3M - n ° 3).

Half of the surface of the sample was protected by adhesive covering tape and the remaining surface was exposed to an acid etch. The demineralization was achieved by immersing the sample in 50 ml of a 5.0-pH solution containing 0.05 M acetate buffer solution and 50% saturated hydroxyapatite enamel powder for 24, 48 and 72 h at 37 ° C (Araujo and Sfalcin 2013). After the demineralization process, the tapes were removed, and the samples were washed with Milli-Q water and stored for further analysis. Figure 1 summarizes the sample preparation.

Figure 1. a) bovine sample tooth. B) and C) Samples cute to 4 x 6 mm² in size. D) Sample included in a PVC tube with acrylic resin. E) Polishing the samples with sandpaper and F) finished sample ready for the demineralization process.



Source: Authors.

2.2 Experimental Setup

To obtain the laser speckle image, each sample was illuminated with a 650 nm red diode laser with a maximum power of 200 mW using a setup similar to the one proposed by Deana (2013). The laser was placed approximately 12 cm from the sample and a lens was positioned to expand the beam area, achieving a spot of Ø = 10 mm with a visible and uniform illumination throughout the region of interest in the sample — both healthy and lesion region were illuminated homogeneously. The laser speckle image was captured by a color CMOS sensor (with 23.7 mm X 15.3 mm, 4752 x 3168 pixels; pixel pitch = 4.99 μm). The digital camera (Canon EOS Rebel T3i) was fitted with a Canon 100 mm macro lens approximately 30 cm of the sample. The photometric parameters were 1/4000 s shutter speed, lens aperture at F5.6, and ISO100. Also, the camera performed no data binning. Figure 2 shows a schematic diagram of the imaging setup.

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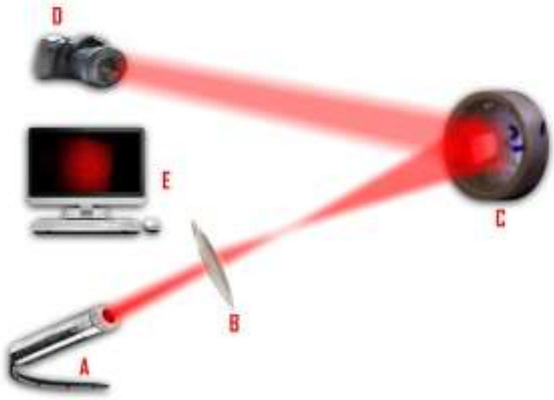
Figure 2. schematic diagram of the laser speckle imaging setup: A) Laser semiconductor, B) lens biconvex, C) healthy-lesion sample D) digital camera E) software analysis.



Source: Authors.

2.3 Image Segmentation

Figure 3 shows an example of a speckle image acquired from a representative sample. As a pre-processing step, each image was manually trimmed to a size containing only the region of interest (ROI), as shown in Figure 3. To analyse of the first momentum of the images, (see Equation 1) we developed a custom software written in Python Language.

The sample size of segmented speckle image varies usually from N = 4 x 4 up to N = 7 x 7 pixels. However, our previous studies demonstrated that for the LSIs of tooth, the sample size of N = 4 x 4 presents a good compromise between statistical accuracy and spatial resolution (Deana *et al*, 2013, Koshoji *et al* 2015A, Koshoji *et al* 2015B, Koshoji *et al* 2016), since the spatial resolution of the processed image is inversely proportional to the sample size(Briers 2001)

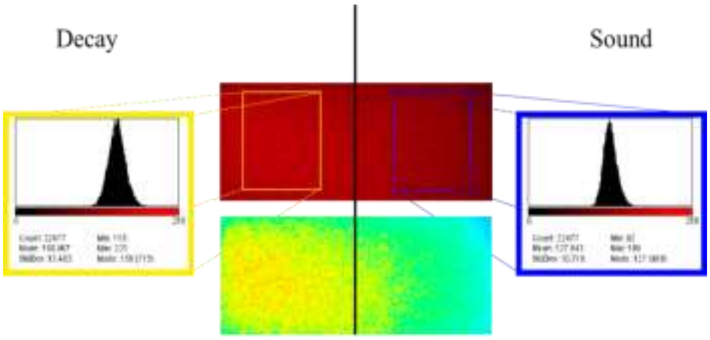
The value is calculated by:

$$< I >= \frac{\sum_{i=1}^{n} I_i}{n} (1)$$

Where $< I >$ is the average of I, which is the intensity of the pixel, and n is size of the sliding window.

We applied a false color algorithm to the momentum images to enhance the visualizations (Figure 3) and then obtained the contrast of the sound and decayed regions of each image.

Figure 3. Processed representative image.



Source: Authors.

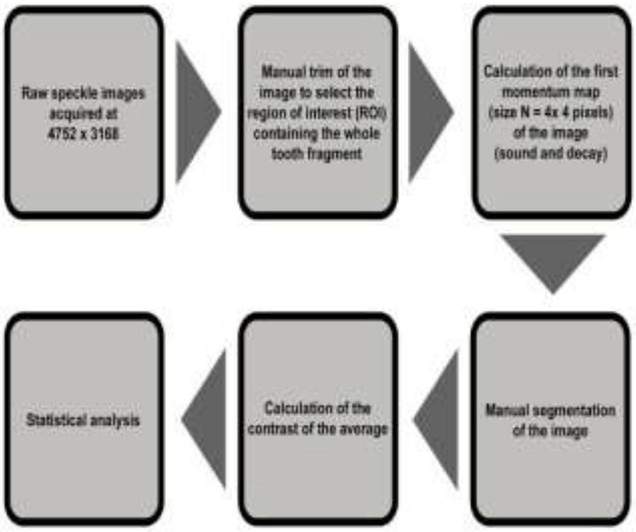
The image contrast was also obtained by equation (2) adapted from Koshoji,(2015A).

$$VR = 1 - \frac{Csound}{Cdecay} \qquad (2)$$

Where VR is the relative value, Csound is the average intensity of the sound region and Cdecay is the average intensity of the decay region (Koshoji *et al* 2015A).

Since biological samples present subject – to –subject variations and, in the case of the teeth, within subjects' variations, the analysis of the contrast provides value that normalizes the sound region with the decay region of the same sample. The resultant data represents the variation of the speckle pattern due to the demineralization process within the sample resulting in a more comparable data, regardless of the bit-depth of the image or of the photometric parameters of the camera. Figure 4 shows the summary of key image processing steps used analysing and segmenting the speckle images

Figure 4. Summary of key image processing steps used analysing and segmenting the speckle images.



Source: Authors.

2.4 Statistical Analysis

For normality analysis, the Shapiro-Wilk test was used. All data presented a normal distribution thus the comparison between the paired groups was performed using the paired Student's t-test. In addition, for the correlations analysis, the R^2 value and Pearson's correlation coefficient were used. The level of significance was set at $\alpha = 0.05$.

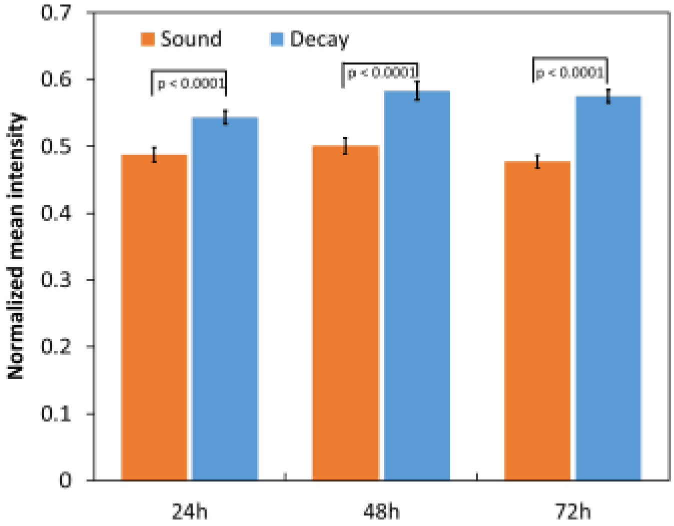
3. Results and Discussion

With the naked eye, after a careful drying process, it is possible to observe some alterations to the enamel surface of the samples nevertheless such changes are incipient even for the group that was in contact with the acid etch for 72h. A trained odonatologist classified the lesions at the 72h group as ICDAS level 1, thus the lesions in the groups 24h and 48h were sub-clinical, *i. e.* it would hardly be detected in a routine examination by visual inspection.

The laser speckle approach allows the access of information on the microstructure of the enamel thus it is susceptible to minimal structural changes, such as the ones induced by the acid etch. Figure 2 shows the sample under laser illumination. The images were averaged and the 1st order moment was obtained (N = 4x4). A false-color algorithm was applied to enhance the visualization of the results.

Figure 5 shows the normalized average intensity of all samples as a function of the acid etch duration (24, 48 and 72 hours).

Figure 5. Normalized mean intensity of the samples for each group. The error bars represent the standard error.



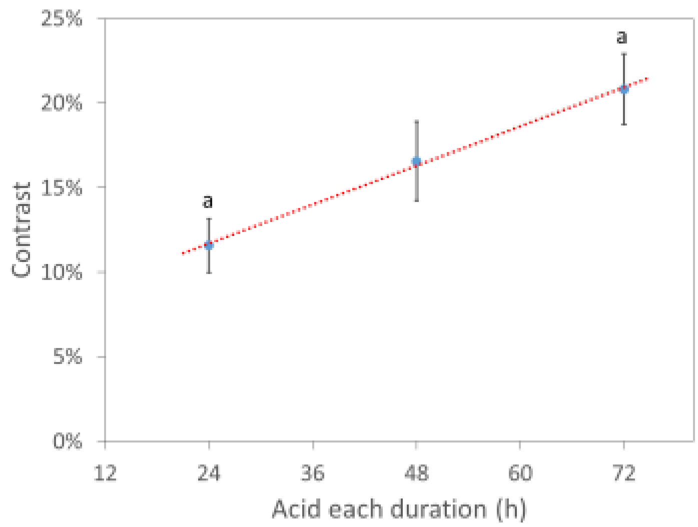
Source: Authors.

The normalized mean intensity of the samples increases with the duration of the acid etch, which is proportional to the severity of the lesion. All groups present statistically significant difference when comparing the sound and decay region (p <0.0001 paired t-test).

The demineralization process results in a material loss by the enamel. At first, the interprismatic material that is corroded exposing the prisms which ultimately results in increased heterogeneity (Briers 2001). If the demineralization conditions are kept for long periods, such corrosion manifests itself as a visual white spot lesion in the enamel. Nevertheless, the microstructural changes are already present even before being visually detectable. Such changes increase tissues' scattering properties resulting in a systematically higher laser speckle backscattering than the sound tissue as shown in Figure 5. The increase in the mean brightness (1st order moment) of the LSI results from the prevalence of constructive interference amongst the scattered waves in comparison to the sound region.

The average contrast (Equation 2) between the sound and decay region is shown in Figure 6

Figure 6. Average contrast of the LSI and the acid each duration. Error bar represents standard error. a: p = 0.0016, t-test.



Source: Authors.

The longer the sample is exposed to the acid etch the more the changes in the microstructure are detectable, therefore, Figure 6 shows a strong correlation between the acid etch duration and the shift in the laser speckle image intensity. (Pearson's correlation coefficient $\rho = 0.9989$, $R^2 = 0{,}9978$).

The contrast of the 24h of acid etch group shows 11.5 (15)% increase in the average brightness of the lesioned region compared to the sound region. The contrast systematically increases with the acid etch duration up to 20.8 (20)% for the 72h group, which significantly differs from the 24h group ($p = 0.0016$ t-test).

It is important to mention that the lesions induced by this work were incipient and even subclinical (undetectable by the naked eye in the clinical practice). Such lesions do not even reach level 1 on the ICDAS scale; however, the proposed method was sensitive enough to detect it. In addition, to detect the presence of microstructural changes in the enamel, the linear correlation between the laser speckle image and the acid etch demonstrates that this method may provide the severity of the lesion, thus provides an objective analysis of the disease's progression.

4. Conclusion

White spot lesions are highly prevalent in people of all ages; however, detecting it in its early stages is still a challenge in the clinical practice. The analysis of the statistical features of the laser speckle image in the spatial domain allows for the detection of microstructural changes in the enamel associated with the presence of the lesion even before any intervention is required, thus providing a valuable tool for prevention of the disease.

Future works will include the exploration of several other statistical features and artificial intelligence to detect the lesion.

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