Qualitative Analysis of Teeth and Evaluation of Amalgam Elements Penetration into Dental Matrix Using Laser Induced Breakdown Spectroscopy

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Abstract:

Introduction: In this study, laser induced breakdown spectroscopy (LIBS) is used for qualitative analysis of healthy and carious teeth. The technique of laser ablation is receiving increasing attention for applications in dentistry, specifically for the treatment of teeth such as drilling of micro-holes and plaque removal.

Methods: A quality-switched (Q-switched) Neodymium-Doped Yttrium Aluminium Garnet (Nd:YAG) laser operating at wavelength of 1064 nm, pulse energy of 90 mJ/pulse, repetition rate of 2Hz and pulse duration of 6 ns was used in this analysis. In the process of ablation a luminous micro-plasma is normally generated which may be exploited for on-line elemental analysis via laser induced breakdown spectroscopy technique. We propose laser induced breakdown spectroscopy as a rapid, in situ and easy method for monitoring drilling process.

Results: The results of elemental analysis show the presence of some trace elements in teeth including P, Ca, Mg, Zn, K, Sr, C, Na, H, O and the permeability of some amalgam (teeth filling materials) elements including Hg, Ag, Cu and Sn into dental matrix.

Conclusion: This study addresses the ability of LIBS in elemental analysis of teeth and its feasibility in acute identification of healthy and carious teeth during drilling process for future clinical applications.

Keywords: spectroscopy; tooth; carious; amalgam.

Introduction

Nowadays laser applications in medicine have attracted much more attention and one of these techniques is laser-induced breakdown spectroscopy (LIBS). LIBS is a kind of atomic emission spectroscopic technique which is used for both quantitative and qualitative elemental analysis of any form of the samples.

LIBS techniques proposes plenty of advantages in comparison with other methods for analysis of different biological samples including very tiny amount of sample (about pico gram) is needed for analysis; the detection limits of this technique are in the limit of few tens of part per million (ppm); This technique can be used to analyze any form of material including solid, liquid and gaseous, and requires a slightly or no preparation of sample. LIBS analysis can be performed in situ or remote as an in vitro or in vivo analytic technique. Other methods including inductively coupled plasma mass spectrometry (ICP-MS), atomic absorption spectrophotometry (AAS), neutron activation analysis (NAA), x-ray fluorescence (XRF) and proton particle-induced x-ray emission.
These benefits made it appropriate for the analysis of plenty of biological tissues such as bacteria, hairs, nails, bones. Samek et al. offered the elemental analysis of tooth samples using LIBS technique for the first time. Other groups also reported such application of LIBS in analyzing teeth samples.

In this paper, laser induced breakdown spectroscopy is applied for qualitative analysis of healthy and carious teeth and also amalgam (teeth filling) materials. We propose laser induced breakdown spectroscopy as a quick, real-time and in-situ technique for investigating the proceeding of laser ablation in dentistry including micro-holes drilling and plaque removal. Elemental analysis of healthy and carious teeth and also amalgam materials is done and the penetration of some amalgam elements into dental matrix is investigated. The purpose of this study is to show the capability of LIBS in accurate recognition of healthy and carious teeth within the process of drilling for future clinical applications.

**Experimental setup**

The experimental setup used for teeth analysis experiments by laser induced breakdown spectroscopy is shown in Figure 1. Neodymium-Doped Yttrium Aluminum Garnet (Nd:YAG) laser pulses with 1064 nm wavelength, 6 ns temporal widths, 2 Hz repetition rate and 90 mJ energy are applied to produce plasma on teeth sample surfaces. The laser pulse is focused on teeth sample by a lens with focal length of 3.5 cm. The samples are located on a XYZ stage and are shifted in a plane perpendicular to the incident laser pulse. So, every laser pulse strikes to the new sample surface. After hitting the focused laser pulse to the sample surface, a hot plasma creates which emits light. The plasma radiation is accumulated by an optical fiber and is sent to the spectograph. For time resolved detection, an intensified charge-coupled device (ICCD) camera is attached to the spectograph. A section of laser beam is directed to a photodiode using a beam splitter and then a signal is directed to the delay generator. The optimum delay time of 1 µs and gate delay width of 20 µs are selected for the experiments. Therefore, ICCD is triggered using delay generator 1 µs after the initiation of plasma and every spectrum recorded in 20 µs.

**Sample collection and preparation**

The tooth sample is a kind of hard and calcified tissue. Hydroxyapatite is the main matrix material of all calcified tissue. Hydroxyapatite is a crystalline calcium phosphate and its chemical formula is $\text{Ca}_{10}(\text{PO}_4)_6(\text{OH})_2$, but they exhibit some distinct differences. Two different parts of tooth are enamel and dentin (Figure 2). The hardest part of body, enamel, consists of 95% mineral, 4% water and 1% organic material. The elementary mineral component of enamel is hydroxyapatite. Dentin is the material which is located between enamel and the chamber of pulp. It consists of 70% hydroxyapatite, 10% water and 20% organic material. So we expect that we will see the intense lines of calcium and phosphorus in the enamel and dentin spectrum, because they are the major element of the basic structure of the hydroxyapatite matrix.
of healthy, carious and amalgam filled teeth are shown in Figure 3.

Tooth samples are randomly chosen from elicited permanent tooth of patients of dental school of Shahid Beheshti University in the city of Tehran. No identification was made between the donor and the samples. For bacteria elimination, tooth samples are placed in the solution of 5% sodium hypochlorite for approximately five minutes. Then, for dryness prevention, all samples were placed in distilled water. Before experiments, the samples were dried in the room temperature. After all, they were placed in the experimental setup for LIBS measurements.

Result and discussion

In Figure 4, the tooth after LIBS experiments is shown. As it can be seen, laser pulses ablate very small area of tooth sample and it shows the advantage of this method for dental applications.

A. Elemental analysis of healthy teeth

The LIBS spectrum of healthy enamel is demonstrated in Figure 5. The spectrum is the average of three spectrum obtained from different points of healthy tissue of teeth. Emission lines are determined by NIST databases. The investigation of teeth spectrum shows that Ca and P are dominant elements and also small quantities of Mg, Na, K, Sr, C, H, Zn, and O can be observed. As it is expected, dominant ionic and atomic emitted lines of Ca and P are identifiers of hydroxyapatite structure of the teeth matrix.

B. Elemental analysis of carious teeth

The LIBS spectrum of a carious part of tooth is shown in Figure 6. The observed ionic and atomic emission lines of a healthy part of the tooth are also seen in the carious part. However, there are little differences between their intensities.

C. Elemental analysis of amalgam materials

Amalgam is a material which is used to fill the teeth. The LIBS spectrum of filled teeth with amalgam materials is shown in Figure 7. The main amalgam elements including Hg, Ag, Cu and Sn, emerged in teeth LIBS spectrum.

Figure 3. Pictures of a) healthy, b) carious and c) amalgam filled teeth.

Figure 4. Microscopic image indicating ablation locations in teeth after LIBS experiments: a) healthy b) amalgam filled tissue.
D. Penetration of amalgam elements into dental matrix

As it is mentioned, the main amalgam elements are Hg, Ag, Cu and Sn. Accumulation of some of these elements, for example Hg, has toxic effects in human tissue. As the LIBS method can analyze the samples spatially, we used it in order to investigate the penetration of amalgam elements into dental matrix. To do this, we cut the tooth in a cross-sectional way. By investigating the resulted spectrum from different points of this cross section, it can be seen that amalgam elements have remarkable...
penetration in other part of tooth matrix. By going away from filled center, the penetration of amalgam elements is reduced. The same results have been achieved when investigating one point of tooth by moving from surface to inside. As it is shown in Figure 8, the more laser pulse went into the tooth, the more reduced the existence of amalgam elements.

Conclusion

Laser induced breakdown spectroscopy (LIBS) technique is performed for qualitative analysis of healthy and carious parts of the teeth to investigate the possible utilization during the teeth treatment of micro-holes drilling and plaque removal. We propose laser induced breakdown spectroscopy as an in situ, quick and easy
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Technique for drilling process monitoring. The results of elemental analysis show the presence of some trace elements in teeth including P, Ca, Mg, Zn, K, Sr, C, Na, H, O. Elemental analysis of teeth filling material including Hg, Ag, Cu and Sn and the possibility of their penetration into the healthy tissue of tooth matrix is also studied. This study addresses the ability of LIBS in elemental analysis of teeth and its possibility in clinical utilization in valuable discrimination of healthy and carious teeth within the process of drilling and preventing from omitting some parts of carious tissue.

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References