Wavelength comparison for laser induced breakdown spectroscopy caries detection

Marcello M. Amaral, Marcus P. Raele, Patrícia A. Ana, Sílvia C. Núñez, Claudia B. Zamataro, et al.

Marcello M. Amaral, Marcus P. Raele, Patrícia A. Ana, Sílvia C. Núñez, Claudia B. Zamataro, Denise M. Zezell, "Wavelength comparison for laser induced breakdown spectroscopy caries detection," Proc. SPIE 10473, Lasers in Dentistry XXIV, 104730K (8 February 2018); doi: 10.1117/12.2288373

Event: SPIE BiOS, 2018, San Francisco, California, United States

Wavelength comparison for Laser Induced Breakdown Spectroscopy caries detection

Marcello M. Amaral*[a], Marcus P. Raele[b], Patrícia A. Ana[c], Sílvia C. Núñez[a], Claudia B. Zamataro [b], Denise M. Zezell[b]

[a]Universidade Brasil, Rua Carolina Fonseca 235, São Paulo, SP, BR 00000-000; [b] Instituto de Pesquisas Energéticas e Nucleares, IPEN–CNEN/SP, Av. Prof. Lineu Prestes, 2242 – Cidade Universitária – CEP 05508-000, São Paulo – SP – Brasil; [c]Universidade Federal do ABC, CECS, Alameda da Universidade s/n, Bairro Anchieta, São Bernardo, SP, BR 09606-045

ABSTRACT

Laser Induced Breakdown Spectroscopy (LIBS) is a technique capable to perform elemental analyses of a variety of samples, independent of matter state. Other spectroscopy techniques may require a destructive and time-consuming sample preparation. On the other hand, LIBS is a less destructive technique with no (or considerably less) sample preparation, using a relatively simple experimental setup. LIBS also provides a multielement analysis into one single spectrum acquisition, applying a Nd:YAG short-pulsed laser to ensure the stoichiometry between the sample and the generated plasma. LIBS have been applied on the study of carious lesions using a Nd:YAG into its fundamental emission at 1064 nm. It was shown that ratio of P/Ca and Zn/Ca can be used to monitor the cariogenic process. Another minor elements, e.g. C and Cu, associated with bacteria biofilm were also measured with the Nd:YAG laser. The fundamental wavelength emission (1064 nm) of Nd:YAG is coincident with a hydroxyapatite transmission window and it may affect the result. In order to address this issue a study used the second harmonic of the Nd:YAG laser at 532 nm. It was show that it is also possible perform LIBS on carious lesion using the Nd:YAG at 532 nm. However, there is not a work direct comparing the LIBS at 532 nm and 1064 nm for carious lesion detection. So, the aim of this work was to investigate the influence of laser wavelength on the LIBS performance for carious lesion detection. In both cases the carious lesion was detected with the advantage of no interference with hydroxyapatite at 532 nm.

Keywords: Laser Induced Breakdown Spectroscopy, LIBS, caries detection, Nd:YAG

1. INTRODUCTION

Laser Induced Breakdown Spectroscopy (LIBS) is an analytical technique based on atomic emission spectroscopy that applies one or more high intense, short duration laser pulses focused on a sample using a lens, promoting its ablation with the formation of a plasma. The plasma is generated at high orders of atomic ionization and excited states. As electrons of the atoms on excited states return to its stead state the emits photons occur. The plasma emitted light is collected by a second set of lenses and transported to a high-resolution spectrometer. It allows the light components to be analyzed and consequently the sample atomic constituents.

The LIBS technique has the capability of simultaneous multi-element detection of all elements, simultaneous detection of major and trace elements and it requires little or no sample preparation allowing direct measurements. The analyses can be equally performed on gases, liquids and/or solids samples. Its simple setup allows a rapid or real-time analysis and a in situ analysis, requiring only optical access to the sample. For the analyses, LIBS remove a small sample portion and in many cases can be considered a non-destructive technique. LBS presents some drawbacks associated to a higher limit of detection and poor precision when compared to other atomic spectroscopic technique.[1–5]

*marcello.magri@universidadebrasil.edu.br; phone 55 11 99780-1966; universidadebrasil.edu.br

Lasers in Dentistry XXIV, edited by Peter Rechmann, Daniel Fried, Proc. of SPIE Vol. 10473, 104730K · © 2018 SPIE · CCC code: 1605-7422/18/$18 · doi: 10.1117/12.2288373

The human teeth are a matrix of calcified tissue composed mainly of hydroxyapatite ($Ca_{10}(PO_4)_6(OH)_2$). The main structures of the teeth are the enamel and dentine with some difference in its structure. The enamel is composed of 95% of hydroxyapatite, 4% water and 1% organic matter and is the hardest substance on the body. Dentine is not as hard as enamel and it consist of about 70% hydroxyapatite, 20% organic matter and 10% water.

Caries is one of the most commons disease in the word, it is a demineralization process of the enamel that becomes a porous material in a few days. During the demineralization process the calcium bounded on the hydroxyapatite is ionized, depressed from the tooth and replaced by other elements such as magnesium or zinc. Besides that, organic minor elements on can be increased by the presence of bacteria biofilm by the presence of carious infection.

Since its beginning, LIBS have been applied to study carious lesions and its ability to differentiate carious lesion from normal tissue has demonstrated. Different laser sources have been applied to perform LIBS analyses, among the laser sources, a number of studies applies Nd:YAG as the laser source for the plasma generation. The Nd:YAG laser besides being a turn on easy to use laser technology, have been applied on odontology during endodontic procedures to remove bacterial biofilm and smear layer, e.g., so it is a reality in same clinics with increase potential.

The Nd:YAG laser on its fundamental emission at 1064 nm have been applied to study carious lesion[6,7]. As described earlier the demineralization process changes the elements proportion on the tooth matrix, thus by monitoring the ration between phosphorous to calcium (P/Ca) and zinc to calcium (Zn/Ca) it was possible to monitor the cariogenic process. In this work it was used the 215.29 nm transition for P I, 211.27 nm transition for Ca II and 206.42 nm transition for Zn II.[7]

The increase of minor elements associated with bacteria biofilm, such as carbon and copper, were also measured using Nd:YAG laser on its fundamental emission (1064 nm)[6].

One of the biggest concern of the odontologists is that the fundamental wavelength emission of Nd:YAG (1064 nm) is in a transmission window region of the hydroxyapatite and it may affect the LIBS result. In order to avoid this possible interference of the laser wavelength on the LIBS result, some of study used the second harmonic[7] (532 nm) and third harmonic[8,9] (355 nm) of the Nd:YAG laser. In these works it was show that it is also possible perform LIBS on carious lesion using the Nd:YAG at these wavelength.

Although there is results for LIBS approach using 532 nm and 1064 nm, we didn't find a work direct comparing the performance of LIBS at 532 nm and 1064 nm for carious lesion detection directly. The aim of this work was to investigate the influence of laser wavelength on the LIBS performance for carious lesion detection.

2. MATERIAL AND METHODS

2.1 Sample preparation

Six teeth, three normal and three carious, were obtained from dental clinic. All the experiments were performed after the protocol approval by an ethical committee CEP CAAE 49461415.0000.5594. The teeth were kept in saline solution to maintain its integrity.

The carious teeth used in this study presented an advanced lesion and the teeth had a deep cavity. This deep cavity could difficult the signal acquisition due to the emission shadowing. To avoid this sample interference of the experiment the teeth were cut in their half by a cutting machine. It results in two portions that facilitates the access to the lesion and tooth structures.

2.2 LIBS Experimental setup

In this work, the emission of Nd:YAG laser (Brilliant, Quantel) was focalized by 90 mm focal length lens into the sample, so that the laser reached the sample generating plasma. The generated plasma emissions were collected by a telescope, positioned at approximately 30° angle. An optical fiber coupled to the telescope guided the plasma emission to an Echelle Spectrometer (Butterfly, LTB – Lasertechnik, Berlin). The Figure 1 shows schematically the experimental setup used for the LIBS measurements.

To compare the effect of different wavelength on caries detection using LIBS technique a Nd:YAG actively Q-switched laser (Quantel Brilliant) operated at its fundamental emission (1064 nm wavelength) and at its second harmonic (532 nm wavelength), 5 ns pulse width. On a Q-switched laser the energy is related to the time delay between the flashlamp and the Q-switch, on this experiment we set 335 μs. It leads to 35 mJ per pulse at 1064 nm. To obtain the Nd:YAG second harmonic, a frequency doubler is connected to the laser, it leads to a decrease (10 mJ) on the energy per pulse at 532 nm. For booth cases the experimental setup was the same as described.

The spectrum acquisition was performed with 1 μs of delay between the laser pulse and the spectral acquisition and 1.5 μs of gate width controlled by the echelle spectrometer software.

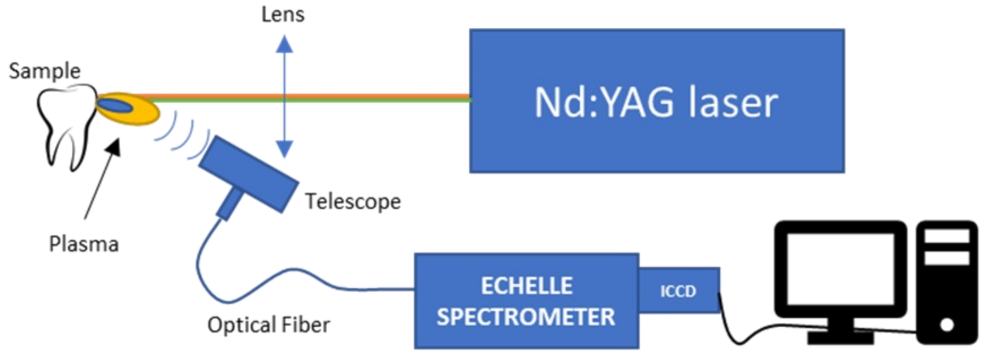

Figure 1. Experimental setup of LIBS spectral measurement of teeth.

It was acquired five spectral per tooth, totalizing 60 spectra for Normal and 60 spectra for Caries teeth, each of them is an average of 2 acquisition accumulated by 15 laser shots.

2.3 Data analysis

The obtained spectra of Normal and Caries enamel were analyzed using the proprietary software of the Echelle Spectrometer for an initial and exploratory determination of the elements corresponding to the observed transitions. After this step, the spectra were analyzed using the OriginPro 8 (OriginLab Corp.) and using the NIST Atomic Spectra Database[10] to identify the spectral lines and elements on the samples.

3. RESULTS

The obtained 120 LIBS spectra for enamel of normal end carious teeth using the 532 nm and 1064 nm were averaged to express the mean behavior of the sample. These results are presented on Figure 2.

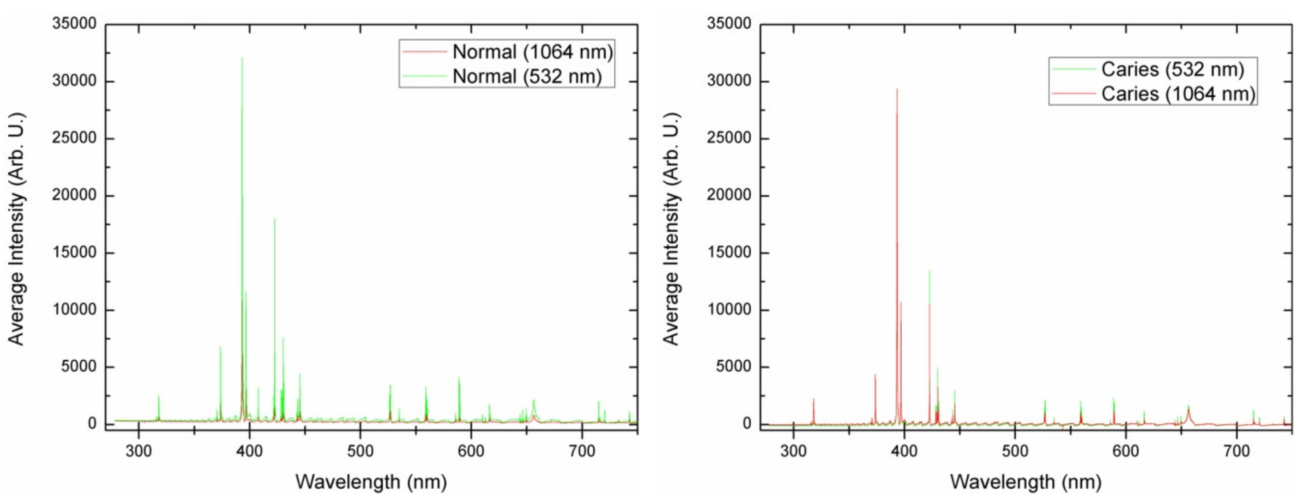

Figure 2. Average LIBS spectra of Normal and Caries acquired using a Nd:YAG laser on 1064 nm and 532 nm.

The most part of the observed emissions on booth spectra are Ca transition. Ca is the one of the major elements of hydroxyapatite and it has persistent lines on the region of the observed spectra. Another minor elements were observed, such as Cu, Mg and Mn. The increase of these elements has been associated to the presence of bacterial contamination. At first place, it was expected a great quantity of phosphorus (P) transitions lines. Although the P on the measured region are weak transition and only one transition were observed (558.83 nm).

To compare the performance of the two laser wavelengths on the detection of spectral difference between normal and caries teeth subsets of the spectrum were analyzed. Figure 3 presented the phosphorous transition at 558.83 nm for Normal and Caries enamel using Nd:YAG laser at 1064 nm (Figure 3a and c) and 532 nm (Figure 3b and d) for all the measured teeth samples.

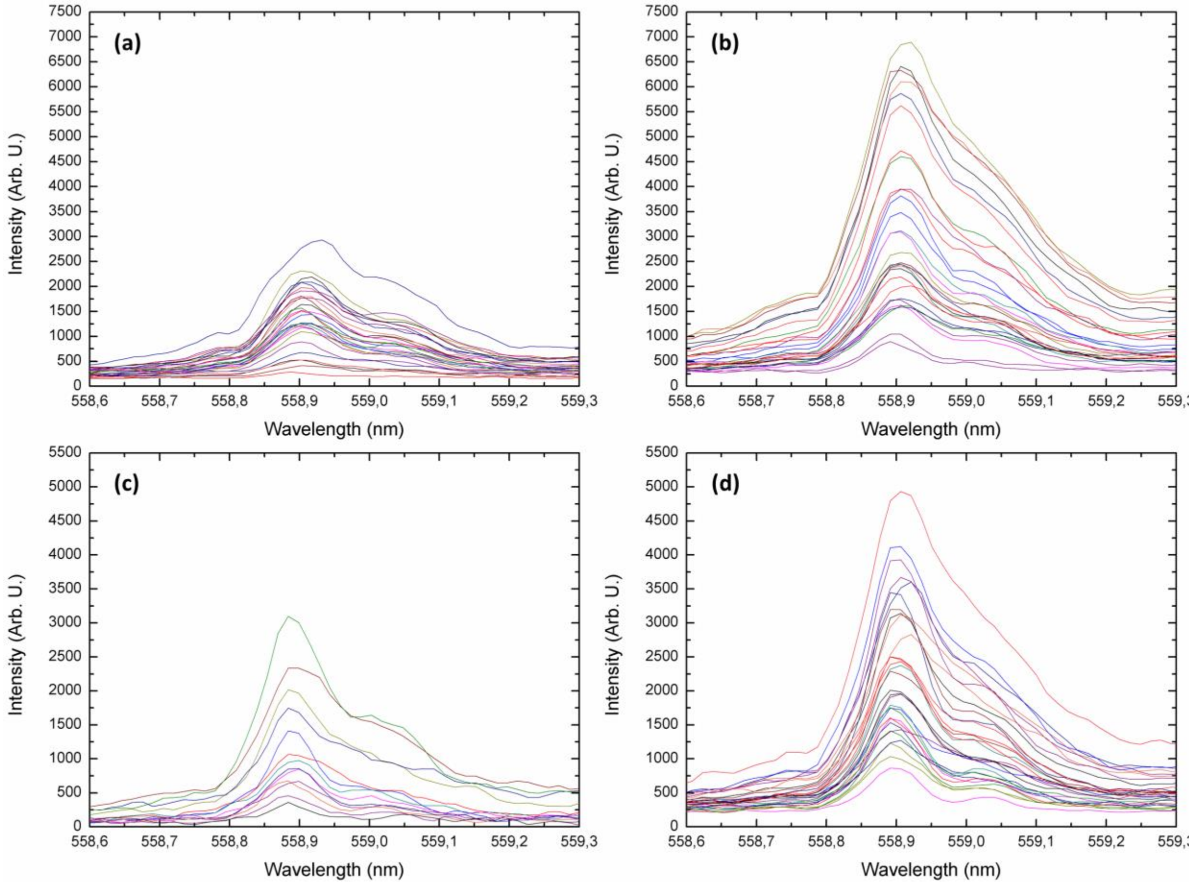

Figure 3. Phosphorous I transition at 558.83 nm of Normal enamel (Nd:YAG laser at (a) 1064 nm and (b) 532 nm) and Caries enamel (Nd:YAG laser at (c) 1064 nm and (d) 532 nm).

Comparing the laser wavelength, it is possible notice that at 532 nm the phosphorus line is more clearly observed. It is also possible to notice that the phosphorus spectral line is weaker for caries than to normal. This behavior can be associated to the changes occurring on tooth during the demineralization process were, as previous described, the phosphorus is substituted to Zn.

Figure 4 shows the Calcium I (Ca I) transition (422.67 nm, 428.3 nm, 428.93 nm, 429.89 nm, 430.25 nm, 430.77 nm) and Manganese II (Mn II) transition (421.55 nm and 431.85 nm) observed for Normal and Caries enamel using Nd:YAG laser at 1064 nm (Figure 3a and c) and 532 nm (Figure 3b and d) for all the measured teeth samples.

It is possible to observe that the Ca transition are clearly observed for all the laser wavelengths, however the Nd:YAG at 532 nm a sharper and more intense spectrum compared to the 1064 nm. Moreover, on normal teeth sample the use of a 532 nm laser provided more clearly the presence of the Mn.

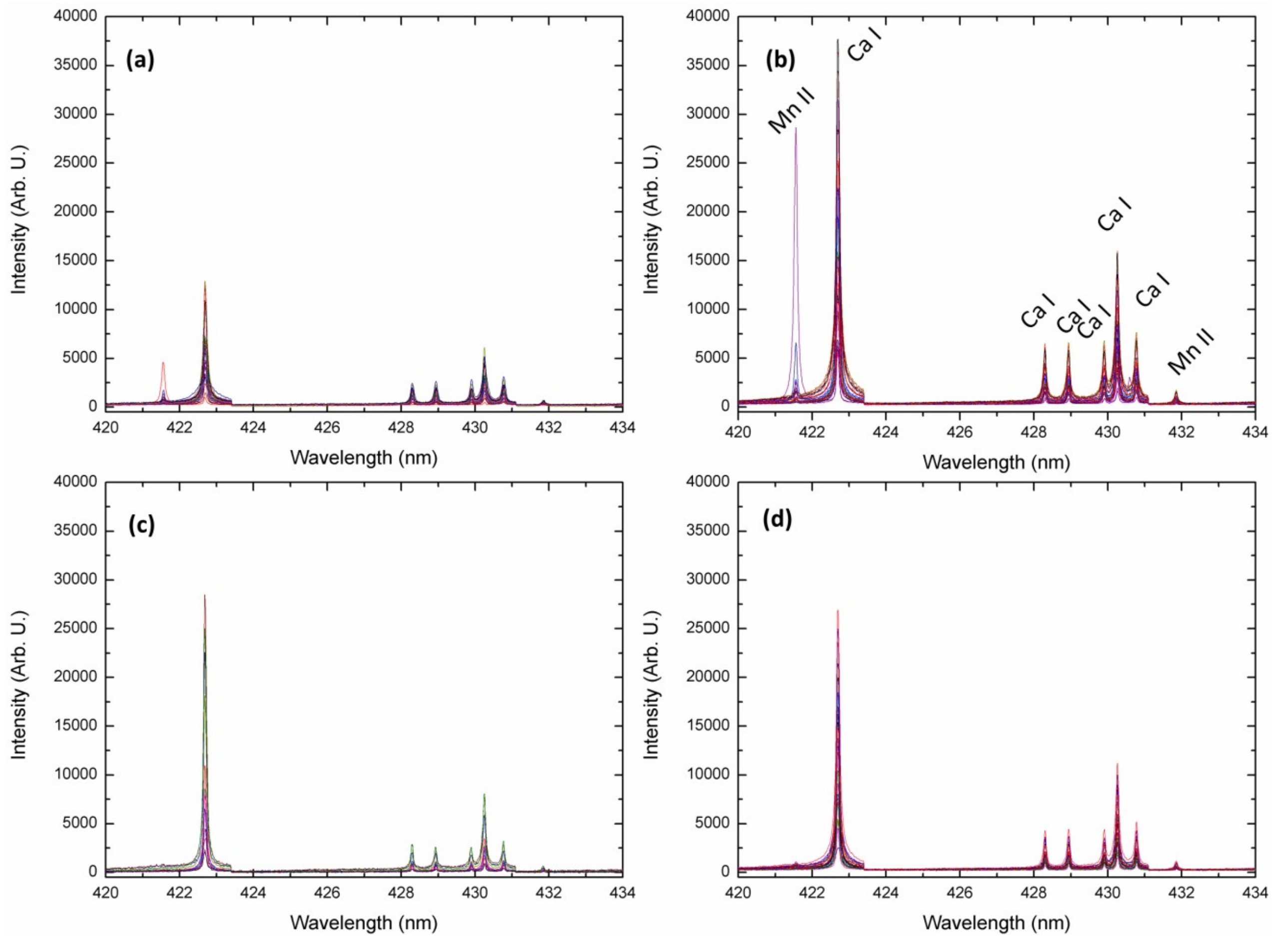

Figure 4. Calcium I transition (422.67 nm, 428.3 nm, 428.93 nm, 429.89 nm, 430.25 nm, 430.77 nm) and Manganese II transition (421.55 nm and 431.85 nm) of Normal enamel (Nd:YAG laser at (a) 1064 nm and (b) 532 nm) and Caries enamel (Nd:YAG laser at (c) 1064 nm and (d) 532 nm).

The same behavior was observed for the Mg and Cu transition lines. In this study we don't observed Zn transitions as expected for a demineralization process. Due to experimental setup limitation we acquired all spectra ranging from 278 nm to 750 nm limiting the analysis and the elements transitions that can be observed. The use of different optical elements could provide a clearer presence of P and Zn, e.g., when measuring it on the ultraviolet region.

Besides these modifications in the experimental setup, for future works we intends to use chemometrics techniques for tissue classification and development of a diagnostic technique.

For all cases studied the carious lesion was clearer detected, the transitions were more clearly differentiated, when the Nd:YAG operating at its second harmonic (532 nm). The no transmission of this wavelength on the hydroxyapatite

4. CONCLUSIONS

The use of Nd:YAG on its fundamental (1064 nm) and second harmonic emission (532 nm) demonstrated to be able to produce the laser ablation on teeth sample, despite of the transmission window of the hydroxyapatite on the infrared. However, this work shown that the use of 532 nm provided a better performance on the identification of the elements transition leading to a better identification of caries teeth.

Acknowledgment

This work was supported by FAPESP/CEPID proc. 05/51689-2, Capes/PROCAD proc. 88881.068505/2014-01; CNPq INCT proc. 573.916/2008-0 and CNPq proc. PQ 312397/2013-5.

REFERENCES

[1] Pasquine, C., Corttez, J., Silva, L.M.C., e Gonzaga, F.B., “Laser-induced breakdown spectroscopy”, Journal of Brazilian Chemical Society 18(3), 463 (2007).

[2] Fortes, F.J., Moros, J., Lucena, P., Cabalín, L.M., e Laserna, J.J., “Laser-induced breakdown spectroscopy”, in Anal. Chem. 85(2), p. 640–669 (2013).

[3] Hahn, D.W., e Omenetto, N., “Laser-induced breakdown spectroscopy (LIBS), part II: Review of instrumental and methodological approaches to material analysis and applications to different fields”, in Appl. Spectrosc. 66(4), p. 347–419 (2012).

[4] Michel, A.P.M., “Review: Applications of single-shot laser-induced breakdown spectroscopy”, in Spectrochim. Acta - Part B At. Spectrosc. 65(3), p. 185–191 (2010).

[5] Barbieri Gonzaga, F., e Pasquini, C., “A compact and low cost laser induced breakdown spectroscopic system: Application for simultaneous determination of chromium and nickel in steel using multivariate calibration”, Spectrochimica Acta - Part B Atomic Spectroscopy 69, 20–24 (2012).

[6] Borisova, E., Uzunov, T., Penev, D., Genova, T., e Avramov, L., “ Laser-induced breakdown spectroscopy of dental lesions: diagnostic and therapeutic monitoring tool ”, 2017, 1022616.

[7] Hamzaoui, S., Nouir, R., e Jaidene, N., “The Study of Carious Teeth by Laser-Induced Breakdown Spectroscopy”, Journal of Applied Spectroscopy 84(1), 82–86 (2017).

[8] UNNIKRISHNAN, V.K., CHOUDHARI, K.S., KULKARNI, S.D., NAYAK, R., KARTHA, V.B., SANTHOSH, C., e SURI, B.M., “Biomedical and environmental applicationsof laser-induced breakdown spectroscopy”, Pramana 82(2), 397–401 (2014).

[9] Unnikrishnan, V.K.., Nayak, R.., Bhat, S.., Mathew, S.., Kartha, V.B.., e Santhosh, C.., “Biomedical applications of Laser-Induced Breakdown Spectroscopy (LIBS)”, in Prog. Biomed. Opt. Imaging - Proc. SPIE 9332 (2015).

[10] Kramida, A., Ralchenko, Y., Reader, J., e NIST, A.T., “NIST Atomic Spectra Database (ver. 5.5.1)”, National Institute of Standards and Technology, Gaithersburg, MD.