



Technical Note

Discrimination of human bodies from bones and teeth remains by Laser Induced Breakdown Spectroscopy and Neural Networks



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ABSTRACT

A fast and minimally destructive method based on Laser Induced Breakdown Spectroscopy (LIBS) and Neural Networks (NN) has been developed and applied to the classification and discrimination of human bones and teeth fragments. The methodology can be useful in Disaster Victim Identification (DVI) tasks. The elemental compositions of bone and teeth samples provided enough information to achieve a correct discrimination and reassembling of different human remains. Individuals were classified with spectral correlation higher than 95%, regardless of the type of bone or tooth sample analyzed. No false positive or false negative was observed, demonstrating the high robustness and accuracy of the proposed methodology.

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1. Introduction

The proper identification of human bodies is important from the legal and administrative point of view. However, this identification issue is sometimes extremely difficult, particularly in mass disaster situations or genocide crimes [1,2]. Adequate human body identification management goes through strict methodology and protocols. This requires adopting and applying standard operating protocols such as the Interpol Disaster Victim Identification (DVI) Standing Committee guidelines [3]. When the number of remains is high, such as in natural disasters, accidents and mass graves, finding the identity of an individual is required. The necessity to achieve an accurate identification is not only important from the humanitarian approach but is also important in lawsuits. The identification process involves a multidisciplinary group of experts and techniques, i.e. fingerprint analysis, forensic pathology, forensic odontology and DNA analysis. When the bodies are non-decomposed the methods used in DVI include physiognomic data analysis such as scars or marks, personal effects, matching of fingerprints and dentition pattern (provided premortem records are available) [2,4,5]. In many situations, these methods cannot be used due to either extensive putrefaction and destruction of the remains or unavailability of appropriate medical or dental records. Common methods for human identification are not sufficient in approximately

42% of cases, because of decomposition of the body and unavailability of pre-mortem data and therefore DNA identification was requested [6]. However, the unavailability of DNA from relatives, the complications in evidence collection and the degradation and contamination of DNA extracted from bone and teeth samples can hinder the identification process [7]. Moreover, in case of huge number of remains, even identification by DNA analysis becomes difficult, requiring more time and economical resources.

Therefore, a new methodology that provides simple, direct and cost-effective analysis is needed in forensic science. In recent years, Laser Induced Breakdown Spectroscopy (LIBS) has become a powerful analytical tool because of its ability to carry out a rapid qualitative and quantitative analysis of different samples, able to provide real time spectral fingerprint of the elemental composition of the sample [8–10]. The possibility to combine LIBS with chemometric methods allows extracting significant information to classify different samples, such as providing an acceptable classification of commercial pharmaceutical tablets using PCA and SIMCA [11].

Human bones and teeth are the typical remains that can be found in forensic scenes due to their resistance to degradation and, hence, are useful as evidences in anthropology, archeology and forensic science [12,13]. These remains are useful to classify and discriminate individuals in crime scenes, accidents or mass burial sites. Although differences in the elemental composition of these samples between individuals are negligible, some trace elements can be related to diet and environment [7], providing variations in the LIBS spectra. These differences allow the achievement of the discrimination when a chemometric analysis is done.

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Neural Network methodology has shown successful results in many areas of knowledge, due to its capacity to generalize and simplicity of implementation that make it useful for qualitative analysis [14–16].

The aim of this work is to develop a simple, direct and cost-effective method based on LIBS analysis and NN mathematical models to discriminate and reassemble individuals using bones and teeth remains.

2. Material and methods

2.1. LIBS setup

The LIBS technique and methodology used in the present work together with most significant experimental conditions have been previously described [17]. Briefly, experiments were performed using a Q-switched Nd:YAG laser (Quatel, Brio model) operating at 1064 nm, with a pulse duration of 4 ns full width at half maximum (FWHM), 4 mm beam diameter and 0.6 mrad divergence. The laser beam was focused onto the sample surface with a 100 mm focal-distance lens, producing a spot of 150 μm in diameter. This large working distance allowed easy sample manipulation and plasma light collection while the focusing provided by the lens enabled extremely precise placement of the beam over the target. The laser fluence was fixed to 20 J/cm² per pulse and the repetition rate was set to 1 Hz. The emission from the plasma was collected with a 4 mm-aperture fiber optic, (with a 1000 μm core diameter and 0.22 numerical aperture), coupled with a 7 mm focus fused silica collimator placed at 45° with respect to the surface normal, and at a distance of 5 cm from the sample, and then focused into an optical fiber which was coupled to the entrance of the spectrometer. The spectrometer system was a user-configured miniature single-fiber system (EPP2000, StellarNet, Tampa, FL, U.S.A.) with a gated CCD detector. A grating of 300 l/mm was selected; a spectral resolution of 0.5 nm was achieved with a 7 μm entrance slit. The spectral range from 200 to 1000 nm was recorded. The detector integration time was set to 100 ms, obtaining whole spectral information for ions, atoms and molecules. To prevent the detection of bremsstrahlung, the detector was triggered with a 4 μs delay time between the laser pulse and the acquired plasma radiation using a digital delay generator (Stanford model DG535). The spectrometer was computer-controlled using an interface developed with Matlab.

2.2. Bone and teeth samples

Twenty-five bones from five individuals and twelve teeth from four different individuals were collected from a local graveyard located in Segovia (Spain) by the permission of the local authorities. Samples were brushed to remove soft tissues and soil, washed with warm water without using soap to avoid the superficial chemical contamination and rinsed several times with distilled water. The samples were left to dry at room temperature before LIBS measurement. Tables 1 and 2 show the type of bones and teeth samples analyzed, respectively. The samples were designated with a letter and two numbers as follows: the letter represents the type of sample (B:bone, T:tooth), the first number corresponds to the individual and the second number to the bone or tooth sample of the individual.

2.3. LIBS measurements and spectral libraries

Bones and teeth samples were measured directly in air at atmospheric pressure. Each LIBS spectrum was acquired with six laser pulses: the first three laser pulses were used for cleaning the surface, whereas, the last three pulses were averaged to obtain the spectrum. Spectra were recorded by moving the sample stage about 0.25 mm to expose a fresh portion of the sample surface and avoiding areas irradiated by previous shots. A data-set of 50 spectra was collected in less than 2 min, taking into account the integration time of the spectrometer and the laser pulse frequency. In the case of the right femur bone and

Table 1
Type and ID of the bone samples analyzed.

Individual	Sample ID	Description
1	11B	Right femur
	12B	Second phalange
	13B	First right metatarsus
	14B	Ulna
	15B	First right rib
2	21B	Right femur
	22B	Radio
	23B	Scapula
	24B	First phalange
	25B	Ulna
3	31B	Right femur
	32B	Ulna
	33B	Left clavicle
	34B	First phalange
	35B	Right flue
4	41B	Right femur
	42B	Right ulna
	43B	Right tibia
	44B	Left femur
	45B	Left radio
5	51B	Right femur
	52B	Calcaneus
	53B	Right ulna
	54B	Foot metacarpus
	55B	Left clavicle

the incise tooth of each individual two data-sets were collected: the first data-set (training library) was used to train the neural network to estimate a suitable empirical model to discriminate each type of sample by supervised learning; whereas the second data-set (test library) was used, along with other bone and teeth samples, to validate the estimated NN models. In order to avoid data variations due to changes in the laser pulse energy, each spectrum was normalized by the intensity of one specific spectral line, i.e., Ca (393.37 nm).

2.4. Neural Network model

Home-made Neural Network software based on Matlab (Mathworks, 2010a) was specifically developed to deal with the problem of individual classification from LIBS data. The NN architecture was based on a multi-layer perceptron, feedforward, supervised network that consists of several neurons arranged in two or more layers (input, hidden and output layers), where each neuron receives information from all of the neurons in the previous layer. The neural connections are controlled by a weight value that modulates the output from each neuron before inputting its numerical content into a neuron in the next layer. This topology is widely used to model systems with a similar level of complexity [16].

The NN training process was based on a back-propagation (BP) algorithm based on the conjugate gradient method [14,18], one of the

Table 2
Type and ID of the tooth samples analyzed.

Individual	Sample ID	Description
6	61T	Incisive
	62T	Canine
	63T	Premolar
7	71T	Incisive
	72T	Canine
	73T	Premolar
8	81T	Incisive
	82T	Canine
	83T	Premolar
9	91T	Incisive
	92T	Canine
	93T	Premolar

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