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Review

When forensic odontology met biochemistry: Multidisciplinary approach in forensic human identification

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ABSTRACT

When human remains are found, the priority of the investigation is to ascertain the identity of the deceased. A positive identification is a key factor in providing closure for the family of the deceased; it is also required to issue the death certificate and therefore, to settle legal affairs. Moreover, it is difficult for any forensic investigation involving human remains to be solved without the determination of an identity. Therefore, personal identification is necessary for social, legal and forensic reasons.

In the last thirty years forensic odontology has experienced an important transformation, from primarily involving occasional dental identification into a broader role, contributing to the determination of the biological profile. In the same way, "DNA fingerprinting" has evolved not only in terms of improving its technology, but also in its application beyond the "classical": helping with the estimation of sex, age and ancestry. As these two forensic disciplines have developed independently, their pathways have crossed several times through human identification operations, especially the ones that require a multidisciplinary approach. Thus, the aim of this review is to describe the contributions of both forensic odontology and molecular biology/biochemistry to human identification, demonstrating how a multidisciplinary approach can lead to a better and more efficient identification.

1. Introduction

When human remains are found, the first priority of the investigation is to ascertain the identity of the deceased; indeed, any forensic investigation involving human remains would be very difficult to solve without this information. Several methods and techniques from diverse fields, depending on the remains available, can be applied to human identification. The first step in the identification process is to build up a biological profile, which is a general description of the individual's ancestry, sex, age-at-death and stature.

This information is the post-mortem data. The ante-mortem data is any information concerning the individual (provided by the missing person's family or relatives) that could be used for identification. Comparisons between ante-mortem and post-mortem data can lead to a positive identification, presumptive identification or an exclusion.

A positive identification is scientifically proven, and is usually achieved through fingerprinting, dental data or DNA. In contrast, presumptive identification occurs when there are several consistencies between ante-mortem and post-mortem data, but no single factor alone justifies the identification (Thompson & Black, 2006). A presumptive identification may be based on personal effects, scars, tattoos, contextual evidence, testimony recognition or facial approximation. When the ante-mortem and post-mortem data are not consistent without explanation, that leads to an exclusion.

Dental identification is extremely useful when attempting to achieve a positive identification or exclusion, either in ordinary cases of identification or in disaster victim identification (DVI) scenarios, where forensic odontology offers an expeditious and scientific method of comparative identification. The field of forensic odontology has experienced a significant change in the last thirty years, from first involving forensic odontologists only occasionally in identification cases, to them playing a key role in the identification process (Senn & Stimson 2010). Nowadays, most medical examiner/coroner offices, as well as most police departments around the world, have forensic odontology

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consultants that are routinely involved in cases of dental identification, age estimation from dental structures, and patterned injuries that may have been created by teeth.

As new technologies advance, new techniques are emerging, and forensic odontology is incorporating these advances into research to be subsequently applied to casework. Recent technological developments are creating new opportunities to perform robust and validated scientific measurements. These technological advances have the potential to strongly increase the speed and efficacy of the criminal justice process. However, such benefits are only realized when quality assurance and control can be guaranteed, so findings can be used as forensic evidence in court (Kloosterman et al., 2015).

In current practice, DNA molecular analysis is an extremely useful tool in forensic investigations. DNA profiling is based on the short tandem repeats (STR) and aids in human identification from biological samples. In the last decade, because of the advances in the field of biochemistry, new biomarkers have been studied and proposed for use in forensic identification (Dumache, Ciocan, Muresan, & Enache, 2016). Likewise, the current trend is to apply biochemical methodologies to determine the biological profile: sex, age and ancestry (Cloos & Fledelius, 2000; Murakami et al., 2000; Witas, Tomczyk, Jedrychowska-Danska, Chaubey, & Ploszaj, 2013). This article aims to review the methods and techniques that can be applied to teeth and oral structures to identify the deceased, from the reconstruction of a biological profile to the ante-mortem and post-mortem comparison of dental data. While some of these techniques can also be applied to the living, there are certain techniques that are applicable only to the dead. Reviewed here are forensic odontology methods as well as biochemical techniques applied to dental structures.

2. Forensic odontology and biochemical methods applied to biological profile reconstruction

In human identification cases, a biological profile must be reconstructed from identifiers in bones and teeth. Forensic anthropology offers a great number of biological profile identifiers to estimate ancestry, sex, age, stature and, in certain cases, pathology. Forensic odontologists in particular, through examination of teeth and oral structures, can provide information for several characteristics of the individual such as age, ancestry, geographical origin, sex, occupation, habits and past or present pathology (Berman, Bush, et al., 2013), such that dental and maxillofacial structures can help in the reconstruction of the biological profile of the unknown. Additionally, molecular methodologies have been developed and applied to ancestry estimation such as mitochondrial DNA and single nucleotide polymorphisms (SNPs) (Witas et al., 2013). In the same line of research, biochemical parameters have been applied to age assessment such as aspartic acid racemization (Cloos & Fledelius, 2000), mitochondrial DNA mutations (Zapico & Ubelaker, 2016), epigenetics (Bekaert, Kamalandua, Zapico, Van De Voorde, & Decorte, 2015), collagen crosslinks (Martin-De Las Heras, Valenzuela, & Villanueva, 1999), advanced glycation end (AGEs) products (Baynes, 2001) or telomere shortening (Tsuji, Ishiko, Takasaki, & Ikeda, 2002). Even though age estimation is a part of the biological profile, this topic will be discussed separately due to the great contribution of forensic odontology and biochemistry to that identifier.

2.1. Estimation of ancestry

There are some dental traits that can be used as indicators of a higher probability of certain ancestral groups. Traits that are illustrative of a Caucasian origin are Carabelli's cusp on the first maxillary molars, a bi-lobulated chin or deep canine fossae. Negroid ancestry indicators consist of multicusped premolars, maxillary midline diastema and pronounced prognathism. Mongoloid ancestry indicators include shovel-shaped incisors, buccal pits and incisor rotations (Berman, Bush, et al., 2013). Even though these dental characteristics can help in ancestry estimation, the final conclusion on ancestry estimation should be carried out by anthropological assessment. Molecular methodologies can also be applied to estimate ancestry, such as mitochondrial DNA profiling and SNPs. However, there are few studies that apply these methodologies to teeth, as researchers mainly use blood. The studies developed in teeth were mostly archaeological, applying mtDNA (Witas et al., 2013) or comparing ancient specimens with current populations (Goncalves et al., 2010).

2.2. Geographical origin

The type of dental restorations present, quality of treatment, and materials used may indicate a country or region where the dental treatment was completed. Silver or gold color metal crowns on anterior teeth are very frequent in Central and South America; full cast metal crowns with acrylic facings on anterior teeth are usually found in Eastern Europe. Some dental conditions may offer information about the geographical origin of the remains. For instance, dental fluorosis can be indicative of Texas, New Mexico, rural United States, China, Africa or India (Berman, Bush, et al., 2013). Also, dental modifications in the present time are practiced in certain parts of South Africa (Friedling & Morris, 2005; Hollowell & Childers, 2007).

Isotope analysis can be used to determine geographic origin, like ^{13}C. This is a stable isotope that constitutes about 1.1% of all carbon. Plants can discriminate between ^{12}C and ^{13}C, creating differences in the levels of these isotopes among types of plants. Based on the fixation of CO_2 during photosynthesis, it is possible to distinguish between C4 plants (like corn and sugar cane), which contain higher amounts of ^{13}C than C3 plants (like potato, wheat and sugar beet), since it diffuses out through the stomatal pores into the ambient air (Kubasek, Urban, & Santrucek, 2013). C4 plants tend to grow in hotter or drier climates than C3 plants. Animals, including humans, with diets based mainly on C4 plants will incorporate more ^{13}C than those that have C3 plant based diets, thus differentiating geographical origin. Another stable isotope that shows geographic variation is ^{18}O. The incorporation of this isotope in animal tissues is correlated to the levels in drinking water, and these levels vary with latitude due to differences in the evaporation and condensation propensity between ^{16}O and ^{18}O (Chesson, Podlesak, Thompson, Cerling, & Ehleringer, 2008). Studies analyzing these isotopes in dental enamel point out their usefulness towards providing information about the geographical origin of an individual (Alkass, Buchholz, Druid, & Spalding, 2011; Alkass et al., 2013).

2.3. Sex assessment

Although there are several scientific studies on morphological sex dimorphism of teeth (Kapila, Nagesh, Iyengar, & Mehkri, 2011; Schwartz & Dean, 2005; Pettenati-Soubayroux, Signoli, & Dutour, 2002; Silva et al., 2016), its use in diagnosis requires further scientific validation (Berman, Bush, et al., 2013). Sex assessment in forensic casework should be carried out by anthropological study or molecular analysis. The determination of sex in skeletal remains is made more challenging if these remains are found fragmented or commingled. Moreover, from an anthropological perspective, the determination of sex in skeletal remains of children and preadolescents is difficult owing to the lack of development of sex characteristics (Potsch, Meyer, Rothschild, Schneider, & Rittner, 1992; Murakami et al., 2000). Molecular methods have been used to combat these complications. Among these methods, the amplification of the Y chromosome-specific alphoid centromeric repeat sequence (DYZ3) by polymerase chain reaction (PCR) reported in 1989 by Witt and Erickson (Akane et al., 1991; Horiuchi, Morisaki, Fujii, & Miwa, 1988; Fukushima, Hasekura, & Nagai, 1988; Kobayashi, Nakauchi, Nakahori, Nakagome, & Matsuzawa, 1988; Tyler, Kirby, Wood, Vernon, & Ferris, 1986), which can detect X chromosome-specific alphoid centromeric repeat sequence (DXZ1) as well as DYZ3, is considered to provide accurate