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ABRAF

ASSOCIAÇÃO BRASILEIRA DE ANTROPOLOGIA FORENSE

MENSAGEM DA PRESIDENTE DA ABRAF

É com extrema alegria que lançamos o Brazilian Journal of Forensic Anthropology & Legal Medicine (BJFA&LM), pois concretiza um sonho almejado por todos os que fazem a ABRAF.

Que essa Revista, fruto de muito esforço e dedicação, possibilite a ampla divulgação das pesquisas brasileiras em Antropologia Forense e promova o bom debate, que é essencial para o conhecimento científico.

Esperamos que todos sintam-se igualmente felizes e que enviem suas contribuições, a fim de que o conhecimento na área seja cada vez mais compartilhado.

Um grande e fraterno abraço a todos!

EVELYNE PESSOA SORIANO

Presidente da ABRAF

SUMÁRIO

Relato de Caso/Case Study

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EDITORIAL

A Antropologia Forense é uma ciência multidisciplinar dedicada a prover de conhecimentos científicos a sociedade no resgate de fatos pretéritos, geralmente criminosos, com implicações muito importantes.

Destaca-se a identificação de restos mortais, saber se são humanos ou não, lançando mão de todos os recursos disponíveis como parâmetros definidos de forma genérica, ou de forma bem específica, como as comparações *intra-vitae* e *post-mortem*. Entretanto, temporalidade, causas e circunstâncias também são objetivos da Antropologia Forense.

Como toda ciência, suas “certezas” são acompanhadas de graus de probabilidade e, no caso da Antropologia Forense, estes são muito relevantes, e deste fato decorre a necessidade de aglutinar a maior quantidade possível de meias-certezas, para se chegar a uma conclusão mais próxima da realidade possível.

Difícil definir a extensão da atuação da Antropologia Forense, que muitas vezes se soma à de outras áreas do saber, como Medicina Legal, Odontologia, Biologia, Arqueologia, entre tantas outras. Da mesma forma, os cenários são muito diversos: um único corpo putrefeito encontrado em local ermo, um acidente aéreo com múltiplas vítimas, fenômenos naturais como desabamentos e terremotos ou hipóteses de violações de direitos humanos como diversos corpos inumados clandestinamente em valas comuns.

Neste cenário de extrema dificuldade, atua a Associação Brasileira de Antropologia Forense - ABRAF, aglutinando os profissionais que atuam nesta área, propiciando um ambiente fértil para a troca de experiências.

E nesta quarta gestão, nossa Presidente Evelyne Pessoa Soriano, antecipando as comemorações de uma década da ABRAF, apresenta o Brazilian Journal of Forensic Anthropology & Legal Medicine, um instrumento que faltava alçar ainda mais a Antropologia Forense Brasileira no cenário científico internacional.

Será um espaço democrático, onde todos poderemos apresentar os resultados de nossas pesquisas e atuações, compartilhando saberes e experiências no formato digital, como forma de atingirmos o objetivo da ABRAF: congregar todas as pessoas interessadas no desenvolvimento científico da Antropologia Forense e apoiar a prática pela difusão do conhecimento.

Seja bem-vinda, BJFA&LM!

Malthus Galvão

Ex-presidente da ABRAF

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Original article

Forensic Anthropology

Can we still use cranial sutures to estimate age at death of individuals after age 50?

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ABSTRACT

Adults' age at death is one of the most problematic parameters in the assessment of biological profile, particularly in forensic contexts, despite a large number of methods that can be applied to the different parts of the skeleton. Regarding the estimation of age at death in adults through the skull, the method which still stands out for its popularity continues to be the one of cranial sutures' obliteration. However, the

accuracy of the results achieved is far from being consensual. The aim of this study was precisely to test its effectiveness and try to understand what information can be gained from the closure of the vault sutures combined with palatine sutures, and how its study can help us to more accurately estimate age ranges for adults, specifically middle-aged and elderly. For that purpose, we analyzed a sample of 200 identified subjects aged older than or equal to 50 years from two identified skeletal collections, chronologically spaced apart by a century and both belonging to the Department of Life Sciences of Faculty of Sciences and Technology of the University of Coimbra.

The results allowed us to conclude that there is a gradual increase in the degree of sutures closure until about age 70 and after that a slight decrease. The comparison between the two collections only revealed different results for the palatine sutures. We also noted that there are differences in the obliteration of the sutures between females and males, the males showing an earlier closure time than females. The correlation between the chronological age and the age estimated, the biological one, from the observed degree of obliteration revealed very low values due to large individual variability, which, in turn, lies on the cranial sutures obliteration. These results lead us not to advise the use of this method alone, yet not neglect its value when inserted in a global context as a general and secondary age indicator.

***Keywords*:** Age at death; Vault sutures; Palatine sutures; Forensic Anthropology.

Introduction

In recent years Forensic Anthropology has been showing exponential growth. Simultaneously, the presence of the forensic anthropologist in collaboration with the pathologist in the autopsy room takes on an increasing nature of strong complicity. Since the estimation of age at death is a parameter of extremely importance for assessing the biological profile of skeletonized corpses and/or human remains in different stages of decomposition and recognizing the limitations that estimation of age at death in adults over 50 years encloses, currently there are no virtual methods to estimate the age of the elderly. It is virtually impossible to discriminate whether the senior was 70, 80, or 90 years[1]. Therefore, the possibility of defining more specific age ranges for these people becomes a major concern.

The real challenge of forensic anthropologist earns the largest dimension when faced with an isolated skull, which in many instances appears toothless. Cunha (2008)[2] asserts that the skull is the part of the human skeleton most often sent to the forensic investigation. In these cases, the most popular method for the estimation of age at death has been the closure time of the cranial vault sutures (coronal, sagittal and lambdoid), but its character is controversial, and the debate keeps on[3,4]. Although Vesalius in 1542 possibly has been the first to realize the real relationship between age and synostosis of the cranial sutures[5-7], the idea that the sutures obliterate during life only appears in literature from the middle of the sixteenth century, with Fallopius[8]. Several studies have been developed over the years by researchers as Welcker, Picozzo, Dwight, Todd and Lyon, McKern and Stewart, Ascadi & Nemeskeri, highlighting the creation of the 5 degrees scale of Broca to classify the state of suture obliteration, the popular method developed by Masset (1982)[9] and the method of Meindl and Lovejoy (1985)[10] which improved the accuracy of the estimation of age through sutures closure as an age indicator and their utility when associated with other trustworthy osteological markers. In the 20th century, and early of the 21st stand out some of the papers published by Key et al., (1994)[11], Hershkovitz at al., (1997)[4], Nawrocki (1998)[12], Schmitt (2002)[6], Sahni at al., (2004)[13] among others who pursued this challenge.

On the other hand, authors like Franklin (2009)[14] point to the limited usefulness of the cranial sutures as an indicator of age at death, since the majority of studies show inaccurate results. Another problem concerns the large age ranges, which, within the forensic investigation, revealed little avail[6]. Despite these assumptions, why some investigators keep using sutures?

As the cranial vault sutures, the palatine sutures also provide an important source of interest. In 1987, Mann and collaborators[15] were pioneers in developing a method to estimate the age at death through the progressive obliteration of the palatine sutures. Over time have emerged further studies in this area, like Gruspier et al. (1991)[16], Ginter (2005)[17] and Beauthier et al. (2010)[7]. Can these sutures provide more reliable results, especially when combined with other methods, including the cranial sutures, in older age groups?

With this study, we intend to test the reliability of the method for estimating the age at death in individuals over 50 years through the analysis of cranial vault and palatine sutures. Furthermore, the aim is to compare the effectiveness between the ectocranial, endocranial and palatine sutures and ascertain, if, indeed, the palatine

sutures, which theoretically close later, are more efficient to estimate the age at death in older individuals. We also aim to verify whether the closure of sutures behaves similarly in females and males and confirm the existence, or not, of secular drift between two identified osteological collections with a chronological difference of a century. Finally, we want to advise or discourage, in a clear way, the use of the analyzed sutures to estimate the age at death of the elderly.

Materials and Methods

This study used two bone collections: The International Exchange Collection (CTI) dated from the late 19th and early 20th century, and the 21st Century Identified Skeletal Collection (CEI/XXI) century. Since this study aims to assess the reliability of age estimation obtained from observation of the cranial and palatine sutures in middle-aged and elderly adults, all who were aged below 50 years were automatically excluded.

Other exclusion parameters were situations that could compromise the results, like pathological conditions, taphonomic alterations, and metopic suture. It was intended to achieve a normal distribution, so that the composition of each age group considered to include the same number of individuals, whether male or female. Nevertheless, differences in the representativeness of particular age groups, namely the one with ages equal or larger than 90, clearly less, made this assumption not always allowed. A total of 200 skulls were examined, 136 belonging to CTI and 64 to CEI/XXI, with ages at death ranging from 50 to 109 years (tables 1 and 2).

Table 1 - Distribution of the total sample (N = 200), by 10 years age range groups.

Age Range (years)	CTI ♂	CTI ♀	CEI/XXI ♂	CEI/XXI ♀	Total
50-59	16	16	1	1	34
60-69	16	16	7	5	44
70-79	16	16	9	3	44
80-89	15	15	9	18	57
≥90	3	7	3	8	21
Total	**66**	**70**	**29**	**35**	**200**

Table 2 - Descriptive statistics of the total sample (N=200).

Collection	N	Mean Years	Standard Deviation	Minimum Age	Maximum Age
CTI	136	70,40	13,244	50	109
CEI/XXI	64	79.05	10,958	50	95
Total	**200**	**73,17**	**13,167**	**50**	**109**

The degree of suture obliteration was examined in the three main ones, coronal, sagittal, and lambdoid, both ecto- and endocranially. Each one was analyzed taking into account their different segments, C1, C2, and C3 for the coronal, S1, S2, S3, and S4 to the sagittal and L1, L2, and L3 for the lambdoid. In the case of coronal and lambdoid, both right and left sides were scored and the average scored between symmetrical segments was calculated. In total, we counted up 10 segments. This procedure was an adaptation of Masset´s (1982)[9] principle. Noteworthy that although each segment was taken into account, we only took into consideration the average value for each of the major sutures. We also examined, individually, the median and the transverse palatine sutures.

To classify the degree of obliteration of each portion was used the scale created by Broca (1875)[18], which was later reversed by Ribbe (1885)[19]. This scale classifies the degree of obliteration of each segment with scores between zero, fully opened, and four, fully closed, going through intermediate stages: 1 - 25% or less closed; 2 - approximately 50% closed, 3 - to about 75% closed[5,9].

The first step of the analysis of the cranial vault and the palatine sutures comprised an essay/training phase of the method, tested in fifteen skulls randomly selected. During this stage, the classifications attributed were compared with those of a second observer. The second step was to evaluate the intra-observer error, 30 skulls were randomly selected, and then two observations were made temporally spaced blindly. Afterwards, it was calculated the technical error of measurement (TEM), the coefficient of reliability (CR), and the absolute mean difference (AMD).

All the sutures were examined macroscopically with the support of a magnifying glass. Because endocranial sutures are difficult to access, we also used a flashlight in order to improve the observations of the different segments. Data records of personal

information such as age at death and sex of the individuals of our sample were only linked to them once the practical study was completed.

Subsequently to the observation of the sutures, values were collected in a database, namely the obliteration rates, the average of their sum, their age range and sex of each individual, for both collections. Afterwards, it was created a second database identical to the previous one, where were encoded the observed obliteration coefficients and the theoretically expected obliteration coefficients on SPSS® – Statistical Package for Social Sciences program.

For the ectocranial sutures was calculated the obliteration coefficient of the coronal, sagittal, and lambdoid individually and, later, for the total. In the case of endocranial sutures, only the total coefficient of the three was calculated. Towards palatine sutures was estimated the obliteration coefficient for the median and transverse palatine sutures individually and the sum of both.

The descriptive analysis of the mentioned variables allowed their characterization, obtained through their mean values, standard deviation, and minimums and maximums found for the five age ranges considered. In addition to these, outliers were also considered. The application of statistical tests took into account the distribution of the variable, thereby to the normal distribution variables we used the T-student test, and for the non-normal distribution variables we used the Chi-square and Mann-Whitney tests.

To test the results of our observations, we created a priori, a set of expected results. According to experience, it is expected that an individual with completely open sutures exhibits a coefficient equal to 0. Inversely, an individual with completely closed sutures must correspond to a maximum coefficient, equivalent to 4. This is the premise of the logic, which involved the creation of groups of tables that show the theoretically expected obliteration coefficients for each type of suture. According to that, the first ones to close will be the endocranial sutures, then the ectocranial ones, and finally palatine sutures (individually the first will be sagittal, then the coronal, and finally the lambdoid). Each of the following tables (3-5) reflects the obliteration coefficient values expected for each type of suture for each age range.

Table 3 - Age range and obliteration coefficient theoretically expected for lambdoid suture, the sum (∑) of ectocranial sutures, and palatine sutures.

Age Range	Obliteration Coefficient
≤ 49	0-0,9
50-59	1,0-1,9
60-69	2,0-2,9
70-79	3,0-3,9
≥ 80	4

Table 4 - Age range and obliteration coefficient theoretically expected for coronal and sagittal sutures.

Age Range	Obliteration Coefficient
≤ 49	0-0,9
50-59	1,0-2,9
60-69	3,0-3,9
≥ 70	4

Table 5 - Age range and obliteration coefficient theoretically expected for endocranial sutures.

Age Range	Obliteration Coefficient
≤ 49	0-2,9
50-59	3,0-3,9
≥60	4

Results

Table 6 shows the values of the technical error of measurement (TEM) and the absolute mean difference (AMD) to classify the obliteration degree of the sutures. In both categories, the error is less than 1, indicating that the observations have a low difference. The same table shows the values of the coefficient of reliability (CR); these are very close to 1, showing that the error has a very small influence on this variance.

Table 6 - Technical error of measurement (TEM), coefficient of reliability (CR), and absolute mean difference (AMD), to classify the obliteration coefficient of sutures.

Calculation	Ectocranial Sutures (n = 200)	Endocranial Sutures (n = 200)	Palatine sutures (n = 151)
TEM	*0.0532*	*0.05*	*0.1936*
AMD	0.0033	0.0033	0.05
CR	0.9999	0.9999	0.9999

For each variable was analyzed the basic statistics, towards ectocranial sutures, these values were calculated for the mean of the sum of the coronal, sagittal, and lambdoid (Cecto) and for each of them separately (Cc, Cs, and CL), table 7. The results show that in all cases, except for sagittal suture, there is an increase in the obliteration coefficient only between 50 and 69 years, thereafter, that coefficient tends to decrease gradually, although small variations are observed. The values of sagittal suture appear to be more constant; indeed, there is an increased obliteration coefficient with increasing age, but from around 90 years, this coefficient declines drastically. The sum of endocranial sutures shows that there is an increased obliteration coefficient between 50 and 79 years, decreasing slightly thereafter (Table 8).

For palatine sutures, we observed the average sum of the median and transverse sutures (CPalatine) and for each individually (CPM and CPT). Their behavior is very identical to that recorded for the endocranial sutures, except for the transverse palatine suture, which reveals less regular coefficients (table 9). Through general analysis of mean obliteration coefficients, it can be noted that for all age ranges the closure order of cranial sutures proceeds as follows: first endocranial

sutures, after the ectocranial and, as expected, the palatine last. When analyzed individually, the closure order of ectocranial sutures is: sagittal, coronal, and, finally, the lambdoid. Moreover, for the palatines, first to close is the median and later the transverse.

Table 7 - Obliteration mean, Standard Deviation, minimums, and maximums to the ectocranial sutures Cecto, CC, CS, and CL, according to age ranges.

Age Range	N	*Cecto*				*Cc*				*Cs*				*Cl*			
		Mean	S.D.	Min.	Máx.	Mean	S.D.	Min.	Máx.	Mean	S.D.	Min.	Máx.	Mean	S.D.	Min.	Máx.
50-59	34	2.0824	0.84260	0.50	3.50	1.9118	0.83508	0.17	4.00	2.5331	1.03596	0.25	4.00	1.6520	0.88608	0.00	3.00
60-69	44	2.4920	0.50887	1.05	3.50	2.4394	0.53807	1.50	3.67	2.8210	0.67793	0.25	4.00	2.1061	0.66384	1.00	3.33
70-79	44	2.4818	0.62804	1.20	3.70	2.4318	0.67755	1.00	3.67	2.8438	0.67132	1.00	4.00	2.0492	0.81140	0.67	3.67
80-89	57	2.5228	0.90243	0.35	3.90	2.4386	0.90338	0.33	3.83	2.8509	1.02819	0.13	4.00	2.1696	0.97462	0.17	4.00
≥90	20	2.2375	0.90203	0.50	3.90	2.2250	0.84686	0.67	4.00	2.4500	1.00770	0.63	4.00	1.9667	0.98021	0.17	3.67

Table 8 - Obliteration mean, Standard Deviation, minimums, and maximums to the endocranial sutures Cendo, according to age ranges.

Age Range	N	*Cendo*			
		Mean	S.D.	Min.	Máx.
50-59	34	3,6368	0,59764	1,90	4,00
60-69	44	3,8818	0,15741	3,20	4,00
70-79	44	3,9057	0,30466	2,10	4,00
80-89	57	3,6579	0,76227	1,00	4,00
≥90	20	3,7375	0,56913	1,85	4,00

Table 9 - Obliteration mean, Standard Deviation, minimums, and maximums to the palatine sutures Cpalatine, CPM, and CPT, according to age ranges.

Age Range	N	*Cpalatine*				*CPM*				*CPT*			
		Mean	S.D.	Min.	Máx.	Mean	S.D.	Min.	Máx.	Mean	S.D.	Min.	Máx.
50-59	24	1.3333	1.02858	0.00	3.50	1.5000	0.93250	0.00	4.00	1.1667	1.23945	0.00	4.00
60-69	37	1.5405	0.91574	0.50	3.50	1.6757	0.85160	1.00	3.00	1.4054	1.11703	0.00	4.00
70-79	37	1.5541	1.00543	0.00	3.00	1.7568	0.98334	0.00	3.00	1.3514	1.15989	0.00	4.00
80-89	38	1.3947	1.04078	0.00	3.00	1.5000	1.13304	0.00	4.00	1.2895	1.18340	0.00	3.00

≥90	15	1.5667	1.08342	0.50	3.50	1.7333	1.09978	0.00	4.00	1.4000	1.24212	0.00	4.00

The detection of outliers in this sample is essential for the interpretation of the method. For ectocranial sutures, only one case was reported; however, the graph shows that there is a wide dispersion of values, especially in the age ranges of 80-89 and ≥ 90 years (Fig. 1). The endocranial sutures showed the largest number of outliers, a total of 26 individuals (Table 10 and Fig. 2). This is related to the higher concentration of obliteration coefficient values between grades 3 and 4, and consequently, a smaller dispersion.

The age ranges that include individuals with 80 years or more seem to be the most problematic, there is the greatest concentration of outliers (14) and also where the largest discrepancy is observed. In these two classes, the minimum values are significantly lower and show extreme variability. The palatine sutures were the only group where there are no outliers. This fact is related to the greater dispersion of the obliteration coefficients distribution (figure 3).

Table 10 - Relation between real age and obliteration coefficients (C) for endocranial sutures outliers.

Age Range	N	♀		♂		Mean (CEndo)
		Real Age	CEndo	Real Age	CEndo	
50-59	6	50	2.10	59	1.90	3.6368
		52	2.70			
		54	2.40			
		54	2.70			
		57	2.90			
60-69	4	61	3.60	60	3.60	3.8818
		68	3.60	67	3.20	
70-79	2	75	2.10			3.9057
		75	3.25			
80-89	11	80	3.30	82	3.70	3.6579
		82	1.50			
		82	2.00			
		82	2.80			
		83	1.40			
		83	1.80			
		83	3.90			
		86	1.00			
		86	1.70			
		89	3.10			
≥ 90	3	90	1.85			3.7375
		96	2.70			
		98	2.10			

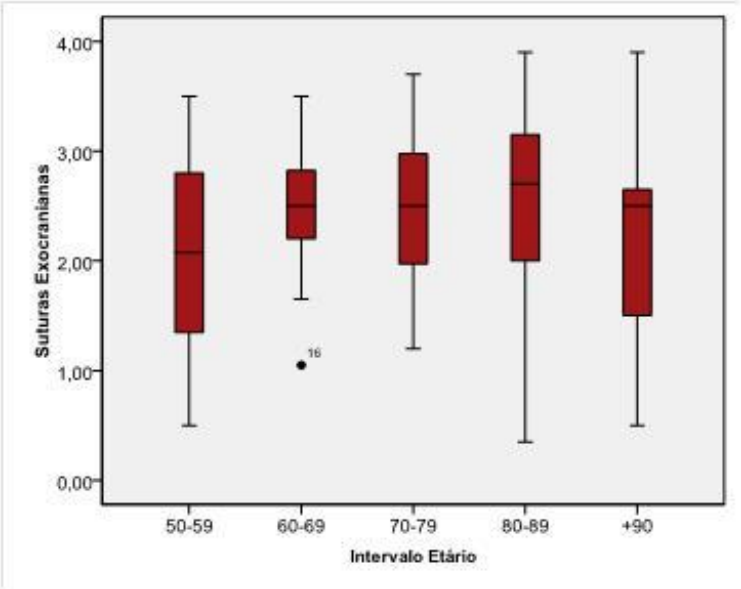

Fig. 1 - Graphical representation of outliers and obliteration coefficients dispersion of ectocranial sutures.

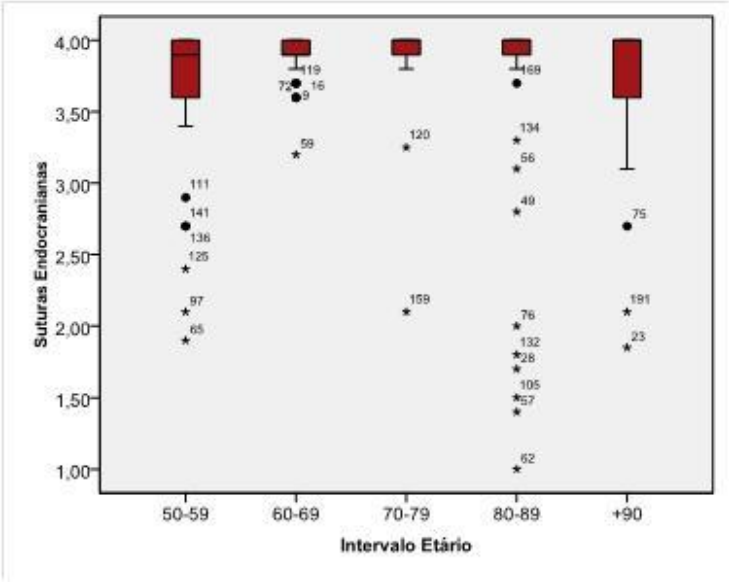

Fig. 2 - Graphical representation of outliers and obliteration coefficients dispersion of endocranial sutures.

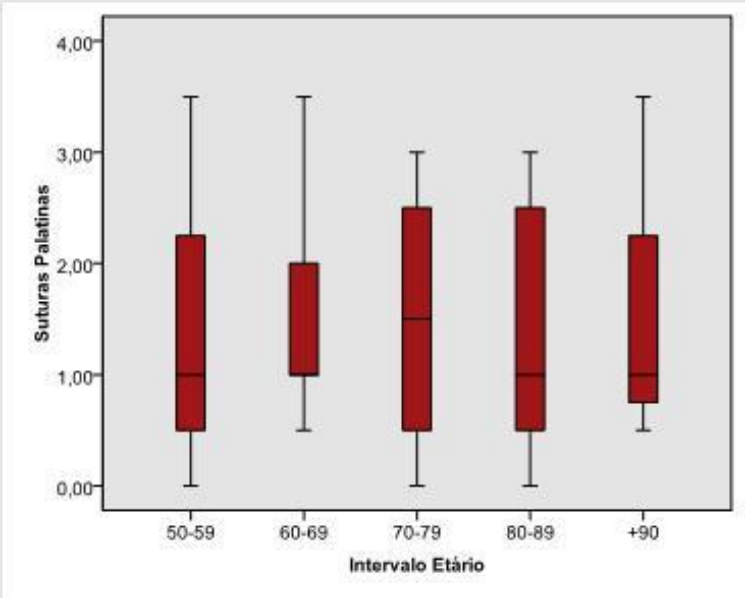

Fig. 3 - Graphical representation of outliers and obliteration coefficients dispersion of palatine sutures.

To assess the effectiveness of the estimated obliteration coefficients to age at death in adults, we compared the percentage of correct diagnoses with the incorrect ones, in each age range for the different sutures. In general, this method has extremely low efficiency, except for endocranial sutures, where the value of correct diagnosis (54.5%) was slightly above the wrong (45.5%) but still constitutes particularly low effectiveness. Regarding the differences between the five age ranges, the obliteration method of the ectocranial and palatine sutures seems to reduce its effectiveness significantly from 70 years on, where values are extremely low, reaching a correct diagnostic efficacy of 0, as in the case of the sum of ectocranial and palatine sutures and for individuals older than 80 years. Contrary, the endocranial sutures show a greater efficacy from 70 years. When individually analyzed, the sutures of the cranial vault and the palate provide results quite insufficient, with low reliability in the estimation of age at death in individuals aged from 50 years (Table 11).

Table 11 - Accuracy of the estimation of age at death through the ∑ ectocranial sutures, coronal, sagittal, lambdoid, ∑ endocranial, ∑ palatines, median palatine, and transverse palatine suture.

Age Range	Correct Diagnosis		Incorrect Diagnosis		Total	
	N	%	N	%	N	%
∑ Ectocranial	52	26	148	74	200	100
Coronal	35	17.5	165	82.5	200	100
Sagittal	45	22.5	155	77.5	200	100
Lambdoid	44	22	156	78	200	100
∑ Endocranial	109	54.5	91	45.5	200	100
∑ Palatines	22	14.6	129	85.4	151	100
Median Palatine	37	24.5	114	75.5	151	100
Transverse Palatine	22	14.6	129	85.4	151	100

The application of the t-student and the Mann-Whitney tests for independent variables allowed identifying the differences that may exist in the obliteration coefficients of the sutures between the two collections that have a difference in time of 100 years. The significance value found for the ectocranial sutures is higher than 0, 05, indicating that the obliterating coefficients scanned in two collections are not significantly different. In contrast, levels of significance assessed for the endocranial (0.017) and palatine (0.006) sutures, refer to the existence of statistical evidence to claim that their obliteration coefficients differ depending on the collection. When comparing the mean of endocranial sutures recorded for each collection, we find that there is practically no difference; however, the value of the Mann-Whitney test shows a significant value, this may be due to a bias caused by the large difference in the number of individuals of each collection (N = 64 and N = 136). Regarding the palatine sutures, there is actually a small discrepancy, indicating that these sutures obliterate earlier in the most recent collection CEI/XXI (tables 19 to 22).

Table 19 - The obliteration coefficients Mean and Standard Deviation, observed for ectocranial sutures on both collections.

Collection	Ectocranial Sutures		
	N	Mean	S.D.
CEI/XXI	64	2.3742	0.86212
CTI	136	2.4059	0.06319

Table 20 - T-Test for the relation between the coefficients of obliteration observed in ectocranial sutures between the two collections.

Type of Sutures	N	t-student	Degrees of liberty	p
Ectocranial	200	0.268	198	0.789

Table 21 - The obliteration coefficients Mean and Standard Deviation, observed for endocranial and palatine sutures on both collections.

Type of Sutures	CEI/XXI			CTI		
	N	Mean	S.D.	N	Mean	S.D.
Endocranial	64	3.7570	0.63651	136	3.7588	0.51261
Palatine	45	1.8333	1.03901	106	1.3255	0.94123

Table 22 - Mann-Whitney (u) for the relation between the coefficients of obliteration observed in endocranial and palatine sutures between the two collections.

Type of Sutures	N	u	p
Endocranial	200	3518.000	**0.017**
Palatine	151	1725.000	**0.006**

We also evaluated the existence of differences between males and females in the total sample, the resulting values are significant, allowing us to conclude that the closure time of the sutures differs between sexes, happening earlier in males and later in females (tables 23 to 26).

Table 23 - The obliteration coefficients Mean and Standard Deviation, observed for ectocranial sutures between two sexes on the total sample.

Sex	Ectocranial Sutures		
	N	Mean	S.D.
Female	105	2.2043	0.89088
Male	95	2.6074	0.56058

Table 24 - T-Test for the relation between the coefficients of obliteration observed in ectocranial sutures between males and females, on the total sample.

Type of Sutures	N	t-student	Degrees of liberty	p
Ectocranial	200	3.784	198	**0.000**

Table 25 - The obliteration coefficients Mean and Standard Deviation, observed for endocranial and palatine sutures between two sexes on the total sample.

Type of Sutures	♀			♂		
	N	**Mean**	**S.D.**	**N**	**Mean**	**S.D.**
Endocranial	105	3.5910	0.71790	95	3.9432	0.11360
Palatine	79	1.0253	0.81216	72	1.9722	0.94530

Table 26 - Mann-Whitney (u) for the relation between the coefficients of obliteration observed in endocranial and palatine sutures between males and females, on the total sample.

Type of Sutures	N	u	p
Endocranial	200	3510.500	**0.000**
Palatine	151	1259.000	**0.000**

Given that differences were observed between sexes, we decided to investigate how these differences revealed themselves within each collection. Collection CEI/XXI shows that the significance value of ectocranial sutures (p = 0.279) indicates that there are no significant differences between men and women. Contrary, the endocranial sutures exhibit a premature synostosis value in females compared to males, inversely palatine sutures indicate a higher coefficient in men. For CTI, all values are significant, and this collection clearly shows a premature sutures closure in males relative to that observed in females (tables 27 to 34).

Table 27 - The obliteration coefficients Mean and Standard Deviation, observed for ectocranial sutures between two sexes on CEI/XXI.

Sex	Ectocranial Sutures		
	N	Mean	S.D.
Female	35	3.6071	0.13891
Male	29	3.9379	0.03194

Table 28 - T-Test for the relation between the coefficients of obliteration observed in ectocranial sutures between males and females, on CEI/XXI.

Type of Sutures	N	t-student	Degrees of liberty	p
Ectocranial	64	1.093	62	0.279

Table 29 - The obliteration coefficients Mean and Standard Deviation, observed for endocranial and palatine sutures between two sexes on CEI/XXI.

Type of Sutures	♀			♂		
	N	Mean	S.D.	N	Mean	S.D.
Endocranial	35	3.6071	0.82178	29	2.5034	0.52029
Palatine	23	1.4565	1.10693	22	2.2273	0.81251

Table 30 - Mann-Whitney (u) for the relation between the coefficients of obliteration observed in endocranial and palatine sutures between males and females, on CEI/XXI.

Type of Sutures	N	u	p
Endocranial	64	385.000	**0.041**
Palatine	45	150.000	**0.017**

Table 31 - The obliteration coefficients Mean and Standard Deviation, observed for ectocranial sutures between two sexes on CTI.

Sex	Ectocranial Sutures		
	N	Mean	S.D.
Female	70	2.1729	0.79831
Male	66	2.6530	0.57525

Table 32 - T-Test for the relation between the coefficients of obliteration observed in ectocranial sutures between males and females, on CTI.

Type of Sutures	N	t-student	Degrees of liberty
Ectocranial	4.003	134	**0.000**

Table 33 - The obliteration coefficients Mean and Standard Deviation, observed for endocranial and palatine sutures between two sexes on CTI.

Type of Sutures	♀			♂		
	N	**Mean**	**S.D.**	**N**	**Mean**	**S.D.**
Endocranial	70	3.5829	0.66621	66	3.9455	0.07683
Palatine	56	0.8482	0.57934	50	1.8600	0.98478

Table 34 - Mann-Whitney (u) for the relation between the coefficients of obliteration observed in endocranial and palatine sutures between males and females, on CTI.

Type of Sutures	N	u	p
Endocranial	136	1517.000	**0.000**
Palatine	106	550.000	**0.000**

Discussion

The present study sought to test the reliability of the method to estimate age at death in adults of both sexes, over than 50 years, by analyzing the obliteration of the cranial sutures. Should we, or not, continue to look for the sutures closure as an indicator of age? Moving away from the majority of the studies based on the closure of the cranial sutures, this study focuses on an elderly identified sample, which ages are between 50 and 109 years, with the particularity that these individuals are from two different osteological collections, separated by an interval of approximately 100 years.

The purpose of using these two collections was to establish the possible existence, or not, of the secular trend phenomenon that Masset (1982)[9], Perizonius (1984)[20], İşcan (1989)[21], Reichs and Bass (1998)[22] have linked to the sutures closure.

In addition to the more traditional cranial sutures (coronal, sagittal and lambdoid) were also examined palatine sutures. Because the skull is the anatomical segment more often sent for forensic analysis and that, in many situations, the expert has only this isolated piece (including skulls without teeth), the question that arises is to know how the cranial sutures may indicate a reliable age at death.

The descriptive statistics of the obliteration coefficients calculated for all sutures (Tables 7 to 9) allowed us to find, as expected, that there is an increase in the obliteration coefficient of most of the sutures to the age range of 70 years. However, with advancing age the evolution of this coefficient is far from being linear or gradual. Contrary to what would be expected this ratio decreased from 80 years, often reaching values significantly lower compared to inferior age ranges and then would be waiting. It should be kept in mind that the age of 90 years' interval comprises a smaller number of individuals, apart from the possible effect of sampling bias, the significant decrease of obliteration coefficient in the older group is an important finding.

Through a comparative analysis of obliteration, coefficients mean it was possible to realize that there is a sequential order of obliteration among several sutures. The first to enclose are endocranial sutures, after the ectocranial and lastly the palatine sutures. Sahni et al. (2004)[13], in his study of the closure of the cranial sutures based on a sample of Indians individuals of the XXI century also concluded that the obliteration starts earlier in the endocranial face than in ectocranial. According to the theoretical expectations, when individually analyzed, the ectocranial sutures revealed that the first to close is sagittal, then coronal, and by the end the lambdoid. It is also known that for the sagittal suture the first closing segment is S3 and for the coronal suture is the C3. Regarding the palatine sutures, we found that the first to obliterate is the median and then the transverse, these results agree with those found by Beauthier et al., (2010)[7] in their study of palatine sutures of 134 skulls identified and Mann et. al. (1987)[15].

For a better understanding of what is going on in each age range in relation to the progression of sutures obliteration the outliers were analyzed, they inform us of the dispersion of the results and help us to understand the differences that can be found in the sample. In the case of ectocranial sutures was detected only an individual whose obliteration coefficient moves away significantly by deficit from the other individuals in the group (Figure 1), the age range where it occurs is precisely that one which exhibits the smaller dispersion, in all other the dispersion of values recorded is significantly higher, mainly from 80s. For endocranial sutures it stands out the high number of

outliers (26 individuals). There is a large concentration of values between grades 3 and 4, which in turn, indicates a much smaller dispersion of the values found for the total sample of individuals. The age group of 80 years reflects this situation particularly well (Figure 2), and indicates a great variability in the obliteration coefficients for these individuals, demonstrating that, although most show higher obliteration degrees, we must keep in mind that lower coefficients do not necessarily mean a lower age. Key et al., (1994)[11] in their study of 183 skulls of Spitalfields collection of the late nineteenth century also found that after 50 years most individuals showed obliteration coefficients above grade 3, however several individuals obliteration coefficients persisted below, which means that there are always elderly individuals who persist with open sutures. Palatine sutures were the ones that did not register outliers, being the group, which exhibits a greater dispersion in all age ranges, reducing the possibility of individuals with more extreme coefficients.

In general, the efficiency of the method for estimating the age at death using the obliteration coefficients of the cranial sutures in older adults was very low, the more assertive values were achieved by endocranial sutures, and still, with only 54.5% of correct diagnosis, which is insufficient. All other sutures showed a higher percentage of incorrect diagnosis than correct ones, particularly the palatine sutures, which indicate an accurate obliteration coefficient for only 22% of subjects. Our results, somehow, come out to the Todd and Lyon (1925)[8] and Key et al., (1994)[11], who claim that the use of endocranial sutures reveals to be more assertive than the study of ectocranial sutures, adding that, the latter is more sensitive to inter-population variation. Beauthier et al. (2010)[7] claim that the palatine sutures seem not to be more effective than sutures of the cranial vault on the estimation age at death, which meets our results. As Gruspier et al., (1991)[16] state that the obliteration coefficients of palatine sutures assume values so broad that it would not be recommended for use it as an isolated method of estimating the age at death in forensic cases.

Hershkovitz et al. (1997)[4] studied a sample of 3638 skulls with known ages to establish the performance of the sagittal suture in age estimation, the results proved not to be able to establish any relationship between age and the real level of suture closure. Both these studies and the results that here obtained show that for each of the major sutures, whether used alone or altogether (the sum of ecto- and endocranial and also to the palatine sutures), the low percentage of correct diagnosis leads us to discourage the use of sutures to estimate the age at death as an isolated indicator. Through our results, we also realize that the ectocranial and palatine sutures show

percentages of correct diagnoses higher up the age range of 70 years and that this percentage decreased dramatically over this age. In contrast, endocranial sutures appear to be less effective to estimate the correct age at death in individuals aged up to 69 years, and more effective for older than 70 years (Table 11). We also claim, as Beauthier et al. (2010)[7], a tendency to underestimate the age of older adults.

Regarding the comparative analysis of the two collections, the results indicate a similar evolution of the ectocranial and endocranial sutures obliteration in CTI and CEI/XXI. Palatine sutures show a closure difference between the two collections (table 21 and 22), this occurs early in the new collection CEI/XXI. This latter result conflicts with what is advocated by authors such as Masset (1982)[9], Perizonius (1984)[20], Iscan (1989)[21], Reichs and Bass (1998)[22], which defend an existence of a secular trend that can manifest itself in the closure of the cranial sutures that seems to occur more slowly in modern populations.

When we examined the comparative behavior of sutures obliteration for both sexes, there are significant disparities in the rate of sutures closure for ectocranial, endocranial, and palatine sutures. In these three groups, the rate of sutures obliteration is faster for men than for women, the same conclusion was ascertained by Beauthier et al. (2010)[7], Ginter (2005)[17], Sahni et al., (2004)[13], Masset (1982)[9] and Key et al. (1994)[11].

Since the comparative study between sexes in the overall sample showed differences in obliteration degrees, it seemed to be a relevant question to see how these differences manifested themselves within each collection. The behavior of ectocranial sutures synostosis in the more recent collection (CEI/XXI) is identical in males and females, as opposed to endocranial sutures that perform prematurely in women. This last result is opposite to that seen in the total sample, which may be justified by the number of individuals, in CEI/XXI the number of women is half (N = 35) than its counterpart in the CTI (N = 70), leading that the results found in CEI/XXI when aggregated to the CTI decrease its influence. The palatine sutures behave differently between sexes of CEI/XXI coefficient foreclosure is higher in men than in women. The oldest collection (CTI) revealed differences between males and females present in ectocranial, endocranial and palatine sutures, the three-point that males show higher obliteration coefficients, leading us to conclude that in men the foreclosure process occurs at a faster pace than in women.

Why is there such a variable behavior on sutures? This is the never-ending question. Cranial sutures are affected by all intra- and interpersonal variations, which

in itself justifies some differences in the progression of their closure. Increasingly it is admitted that longevity may be closely linked to genetic factors. Given the difficulties that exist in executing cross-sectional studies on the sutures obliteration process, when confronted with an isolated skull, the forensic anthropologist should make a joint analysis of all the information that characterizes the cranium, using multi-factorial techniques rather than a single technique as well as necessarily use a specific standard to each population[6].

The most aged skulls reflect skeletal changes that evolve over the years, such as the bregma position. Another difference has to do with the fact that the vault tends to move downwards and backward relative to the central position of the cranium with the spine. Some individuals may present a large decrease in the thickness of the cranial vault and/or increasing the thickness thereof[23]. Also, the external surface of the cranial vault of elderly individuals displays a rougher and granular texture, with areas of salient muscle insertion particularly in the base and in the lateral zones, and a flattening of the parietal. The internal surface stresses a greater depth brand meningeal artery, and along the sagittal suture can be found pacchionian depressions[23]. In addition to these, we should still be evaluated degenerative joint disease (the occipital condyles and the mandibular fossa) and non-articular degenerative pathology (the external occipital protuberance, the palatine vault, and alveoli). The articulation of all these features, together with the observation of cranial sutures had to be the tools of the forensic anthropologist for the estimation of age at death in older adults, emphasizing its extreme importance, especially when dealing with toothless skulls.

Conclusion

This study examined the reliability of the method of estimating age at death in individuals over 50 years, through the cranial vault sutures and palatine sutures, in a sample composed of 200 skulls identified.

We concluded that:

a) The obliteration coefficients showed a gradual increase until about the age group of 70 years, registering a decline of values after that age;
b) The sutures revealed a sequential closure order: the first is endocranial, after the ectocranial, and lastly, the palatine sutures. Individually, the first to obliterate is the

sagittal suture, followed by the coronal and finally the lambdoid, in the case of palatine sutures, first closes the median and then transverse;

c) The comparison between the two collections (100 years spaced), only showed differences in obliteration coefficients calculated for the palatine sutures, which showed higher degrees of synostosis in CEI/XXI, the latest collection;

d) It was also concluded that the process of sutures obliteration occurs differently between males and females. Generally, men show higher degrees of synostosis than women, which means that the suture closure is faster in men. The only exception occurred in CEI/XXI, where females showed for the endocranial sutures higher obliteration values than males.

According to the earned results, and given to the low number of correct diagnoses, cranial sutures have not shown to be a credible method to estimate age at death in individuals over 50 years. There is great variability in the closure time of the cranial sutures, and each individual is influenced by internal and external factors that may partly explain the low reliability of this method.

The estimation of age at death in elderly adults through the cranial sutures in an isolated skull is not advised, but its interpretation should not be neglected, we must have an understanding of all degenerative manifestations that express themselves in the rest of the skull. Only through a global and contextual approach, the cranial sutures should be interpreted.

Industrialization and improvements in living and medical conditions have driven the increase in prolongation of longevity, however, many areas are yet to discern, like genetics, particularly in the aging phenomenon, and hence further studies in this area are required.

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Original article

Forensic Anthropology

The academic scenario of Forensic Anthropology in Brazil

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ABSTRACT

This research is aimed at mapping and analyzing scientific production within the area of Forensic Anthropology in Brazil. The bibliographic search was performed within the major online scientific databases (PubMed, Scopus, Scielo, Lilacs). Only publications originated in Brazil and published in scientific journals were included in the study. Collected data was introduced into an SPSS database and analyzed statistically. A total of 161 studies (1992-2017) fulfilled the inclusion criteria. A significant increase in the number of publications during the last ten years was observed. Seventy-one

percent of publications originated at the Faculties of Dentistry, and the University of São Paulo was the leading research center in this area. Almost 38% of scientific production was performed within a single institution, 45% included national collaboration, and 17% was conducted with international cooperation. Fifty-eight percent of research focused on the subject of Forensic Odontology and 42% on Forensic Anthropology. The main focus of the research was human identification (39%), followed by age estimation (21%) and sex estimation (15%). Thirty-five percent of the articles presented a new approach, and 28% focused on method evaluation. The rest was equally distributed (18.5%) between case reports and field evaluations. The number of scientific productions is relatively low but is quickly growing. The potential for collaboration could be better explored. The research is centralized and lead by the Faculties of Dentistry, with a major focus on Forensic Odontology.

***Keywords*:** Forensic Anthropology; Forensic Dentistry; Science, Technology, and Innovation Indicators.

Introduction

For the construction of a quality academic and professional field that would guarantee the homogeneity of training, regularization of professional performance and uniformity of procedures that would allow the best development of the area, it is necessary to know the profile of the discipline, its reality and its concept within a concrete context[1]. Given the recent growth of Forensic Anthropology and the opening of this field in Brazil, the lack of studies and scarce scientific production in this area, as well as the lack of unified training offered, among other circumstances, justify a detailed analysis of various aspects of this discipline in order to understand the challenges, needs, and perspectives of Forensic Anthropology in Brazil.

The productivity, development, and the state of art of a scientific discipline can be evaluated through the quantification of its scientific production. Statistical data – bibliometric indicators – obtained from the analysis of scientific publications allow the evaluation of the scale, extent, and trends in knowledge production of a particular field of science[2].

For this reason, the following research aims at evaluating the state of the art, mapping, and analyzing the scientific production within the area of Forensic Anthropology in Brazil to understand the advances, directions, and perspectives in knowledge production in this discipline. Furthermore, it seeks to comprehend the difficulties and challenges and to reflect on

the future development of this area that is becoming increasingly essential in the Medical-Legal and Human Rights contexts. We will focus mainly on the subject such as the trends in the production of knowledge, the main research centers, the potential of collaboration, and the focus of the research.

Materials and Methods

The research was based on a systematic bibliographic analysis of the scientific production in the last 26 years related to Forensic Anthropology in Brazil. Being a relatively new area and considering the historical context of Brazil and its implications, it was not expected that there would be much production and research in this field before the 1990s. Therefore, the period of research was established between 1992, when the Forensic Anthropology Group was created within the Office of Public Safety in Brasília[3] until the year 2017.

The data was collected by a thorough search and analysis of scientific articles indexed within the PubMed - Medline database (https://www.ncbi.nlm.nih.gov/pubmed). Additionally, local databases SciELO and Lilacs were analyzed together with the archives of the Revista Brasileira de Odontologia Forense (RBOL), which traditionally publishes research related to Forensic Odontology and Anthropology. The search was performed from March to August 2018.

For the bibliographic search, the following keywords were used: forensic anthropology, sex estimation, age estimation, stature estimation, height estimation, ancestry estimation, taphonomy, forensic analysis, forensic methodology, forensic methods, their possible combination, and always together with the word “Brazil.” The search was performed in English, Portuguese, and Spanish, where the keywords were translated accordingly.

The inclusion criteria for the articles were the following:

1. Scientific articles published between 1992 and 2017;
2. Use of both quantitative and qualitative methodology, as well as bibliographic reviews, field evaluation, and opinion;
3. The relationship with the field of Forensic Anthropology that included analysis of partially or totally skeletonized human remains with the aim of sex, age, stature, ancestry, time since death and possible death cause estimation, application or evaluation of existing methods, facial reconstruction and technical development

related to the previously mentioned aspects. The analysis also included the field of Forensic Odontology considering teeth being a part of a skeleton but only with the aim of human identification;

4. Papers written in Portuguese, English, and Spanish in both national and international scientific journals;
5. Articles indexed in PubMed (Medline), Scielo, Lilacs, and RBOL archives;
6. Works originated in Brazil, but which may include the participation of foreign researchers. The origin of the research was considered following the academic affiliation of the first author.

During the analysis, data regarding the number of publications, authors, type of journal, its impact factor, the language of publication, research center, existing national and international collaborations, as well as the main focus of the study were collected for each article in a PSAW18 database.

In order to analyze the focus of the research, each publication was attributed to the Forensic Anthropology or Forensic Odontology area depending on whether the object of the study was explicitly the dental arcade and the oral cavity (Forensic Odontology) or other parts of the body/skeleton (Forensic Anthropology). Each research was also attributed to a specific subject that included sex estimation, age estimation, stature estimation, ancestry estimation, human identification, facial reconstruction, technical development, multiple (if combining at least two topics) and other (if the research was based on the bibliographic review, opinion or field evaluation). The scope of the study was defined as the field in which the analysis was placed, and this could vary between a new approach, field evaluation, case report, and existing method evaluation. The statistical analysis included the examination of descriptive and frequency statistics.

Results

Number of publications

The keyword search delivered a total of 2,202 articles, out of which only 161 were selected for further analysis. The first articles that fulfill the inclusion criteria of this research were detected in 1999, what constitutes a 7-year time frame (1992-1999) without relevant publications in the area from the *a priori* established time 1992-2017. The total number of articles represents a relatively low activity in the production of

knowledge (~6 articles per year), considering that the research covers 26 years. If taking into account only the period in which the scientific production was identified – 19 years – the average amount of articles per year goes up to 8.5, which is still quite low. However, there was a strong tendency observed towards the increase in the number of publications related to the area of Forensic Anthropology or Odontology during the last ten years (Fig. 1), and the observed movement permits the prediction that this tendency will probably continue.

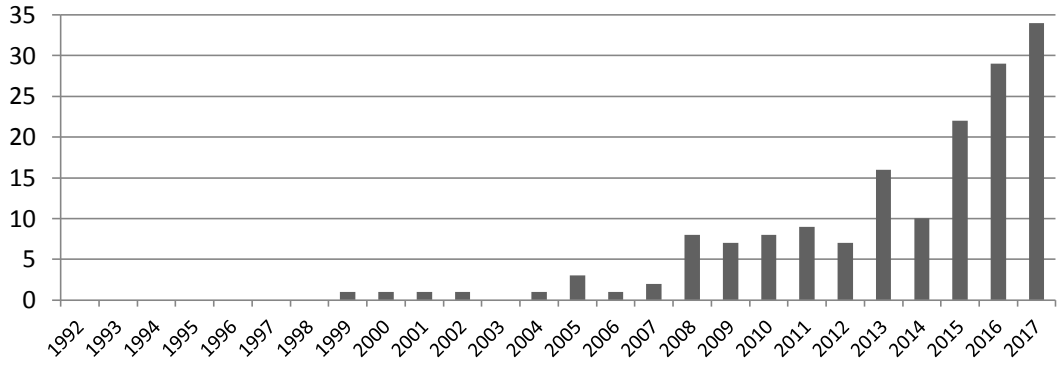

Fig. 1 Number of identified publications per year in the analyzed period (1992 – 2017)

Type of publication

Slightly more than half (56.52%) of the articles were published in international journals, the rest accounted for national publications. Ninety-four papers (58.39%) appeared in journals with Impact Factor, and these included both national and international publications. A vast majority of publications were written in English (111), 46 in Portuguese, and 4 in Spanish, which indicates that even national papers are often prepared and presented in English, which increases the international visibility of the research. Although the tendency of increasing the number of English-written publication was observed, in the year 2017, the last year included in this analysis, this proportion significantly dropped – 22 vs. 7 (English vs. Portuguese) in 2016 to 19 vs. 14 in 2017 (respectively).

Only 59% of analyzed research (94) was published in collaboration with at least two different institutions. Within this number, 74% of publications (70) were realized in partnership among Brazilian research centers, and 26% (24) had international participants, respectively representing 43% and 15% of the total production. International collaboration remains, therefore, quite a minor portion of Brazilian scientific production in the field of Forensic Anthropology and Archaeology. There was,

however, observed a significant increase in the number of publications prepared within the cooperation among at least two national institutions in the last ten years. Still, a big part of the research belongs to a single center.

Research center

The research identified 34 different institutions, among them the Ministry of Justice (2: Federal Police and State's Prosecutor's Office), the State Public Safety Office (13 – Medico-Legal Institutes) and independent practitioners (7), as centers where research in Forensic Anthropology or Odontology is being conducted in Brazil. The leading research center was USP (University of São Paulo), with almost 30% of the total count of analyzed publications. It was followed by UNICAMP (State University of Campinas) with 12.42% of analyzed scientific production, the State Public Safety Office with 8.07% of participation, and UFG (University of Goiás) with 7.45%. The remaining institutions delivered between 1 and 7 publications.

The faculties of Odontology are the leading centers of research in both Forensic Anthropology and Odontology[1] (111/161) (Fig. 2). The research is also conducted, however, to a significantly lower extent, in the Faculties of Medicine (12), Anatomy/Morphology (11), and Health (3). There exists an important contribution to the research (24) in non-academic institutions, mainly the Medico-Legal Institutes and private dental practitioners.

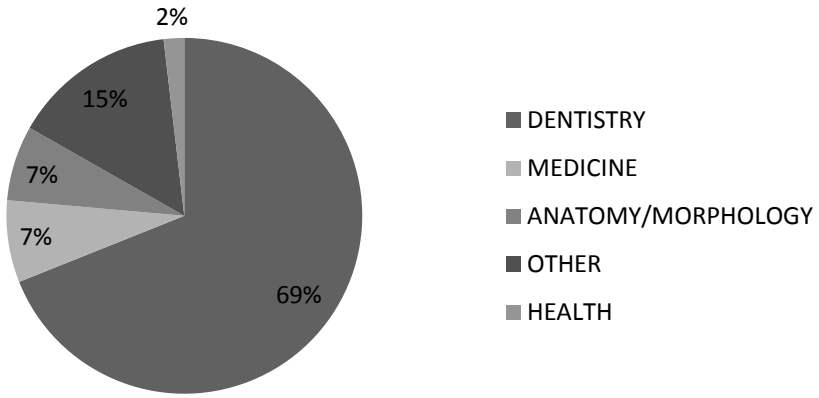

Fig. 2 Distribution of departments where research in Forensic Anthropology and Odontology is being conducted in Brazil (other = independent researchers, Medico-Legal Institutes etc.)

Research focus

[1] A common term, *Department of Odontology* is used here for various departments related to the science of Odontology (i.e. Dpt. of Social Odontology, Dpt. of Stomatology, Collective Health and Forensic Odontology, Dpt. of Clinic and Social Dentistry, Dept. of Odontology and Biomedical Service, Dpt. of Dentistry, Dpt. of Community Dentistry) as states in the affiliation provided by the authors of the articles.

Of the analyzed publications, 56.52% (91/161) were categorized as belonging to the Forensic Odontology area, and the remaining 43.48% were attributed to Forensic Anthropology. The most prevailing subject was human identification (61), followed by age estimation (32) and sex estimation (27). 8% of the research involved more descriptive work, such as field evaluation, bibliographic review, and historical background of the field of Forensic Odontology and Anthropology. Ten publications focused on facial reconstruction issue, and 9 proposed a technical development to existing methods. Six articles combined at least two categories, and none raised the subject of ancestry estimation. Regarding the scope of the publications, the highest percentage (33.54%) aimed at presenting a new approach to the analyzed subject (29), and these were mostly related to sex estimation and positive human identification. Nineteen percent of analyzed material focused on applying and evaluating existing methods on Brazilian samples, 21.74% were classified as literature review or field evaluation, and 15.53% were examples of a case report.

Discussion

The analysis of the trends in the field of Forensic Anthropology and Odontology in Brazil revealed that the production of knowledge in this area, until very recently was relatively low. There exists a tendency of fast growth, however. Only in the last five years (2013-2017), the number of scientific articles doubled in relation to all previous years in which scientific activity in this field was detected (1999-2012). This trend is consistent with the worldwide tendency observed for this discipline[4]. Nevertheless, it represents a relatively insignificant minority when compared to the international production. Although Gualdi-Russo and Fonti[4] note a certain slowdown in the rate of scientific production in the field of Forensic Anthropology during the last decade, this tendency is not present in the Brazilian context. It is hard to predict whether the intensification of production will continue in the upcoming years, but considering the growing interest for this field among Brazilian scholars, it is quite possible.

As Baraybar and Blackwell[5] notice, the countries of the Global South, Brazil included, developed the practice of Forensic Anthropology rather in response to a practical need and not that much out of academic interest. The practitioners of Forensic Anthropology quite often are not academics but rather technicians[6]. Therefore, they focus much more on the practical aspects of the discipline, rather than on its scientific development. As Cooley[6] states, the main task of the forensic

technician is the acquirement and application of a specific advanced technique or routine without the requirement of having problem-solving skills and experience in conducting research. Consequently, the interest of the majority of Forensic Anthropology practitioners in the development and advance of science is also limited. However, it is essential to notice that there exists a considerable contribution and effort of non-academic centers, especially the Medico-Legal Institutes, where a majority of the practitioners work, to the scientific production in the area of Forensic Anthropology in Brazil.

Moreover, as Guimarães *et al.*[3] point out, the political scenario under the military dictatorship in Brazil for decades has limited the investment and discouraged the interest in the development of Forensic Sciences, and this was strongly motivated by political reasons[3,7.] The situation did not improve after the end of the dictatorship in 1985. After a certain period of significant increase of public funds for research support, since 2013 the funding for science in Brazil has been systematically decreasing year by year[8]. This can be seen as a general austerity measure and systemic issue regarding national, federal, and state policies in Brazil regarding the investment in science. Considering Forensic Anthropology as a very specific and niche discipline, its lack of visibility and general low social interest, the resources destined to it were and continue to be scarce. Lack of systematic and prolonged funding results in limitations regarding the vacancies and salaries of the practitioners, as well as poorly equipped laboratories and infrastructure[6]. As a consequence, professionals are very often overloaded with work and lack the time, but not the will necessary for research[9].

It should also be noted that the development of science in general in Brazil is highly delayed in relation to other countries from the so-called Global North. It is commonly accepted that the creation of Instituto Oswaldo Cruz in Rio de Janeiro in 1900 represents the institutionalization of science in Brazil, and the inauguration of the University of São Paulo (USP) and the Faculty of Philosophy, Letters, and Sciences (FFCL) in 1935, is considered the turning point of the professional training of researchers[10]. Therefore, it cannot be expected that the scientific production in any discipline could be comparable with the one in countries with hundreds of years of scientific tradition.

Most definitions of Forensic Anthropology derive its origin in the methods developed and used by biological anthropologists. Research conducted in this area is also based on the principles of Biological Anthropology. It is therefore important to emphasize the fact that in Brazilian academia, there really does not exist a traditional

field of Biological Anthropology as a separate field of science, and it is virtually “absent from Brazil’s postgraduate system”[11]. The lack of specialized training and clearly defined institutionalized space does not, however, totally impede the research in this area[11]. Nevertheless, it severely limits the preparation of professionals with research background and with knowledge of bio-anthropological theory and methodology that could further specialize in the area of Forensic Anthropology and conduct research in this field.

As the results demonstrated, over two-thirds (69%) of scientific production in the area of Forensic Anthropology in Brazil is conducted in the departments of Odontology. It is not especially surprising, as the majority of practicing forensic anthropologists are dentists by training[9]. Forensic Anthropology, however, goes much beyond positive human identification[12], and Forensic Odontology, on the other hand, is a clearly defined and very limited part of forensic analysis[13-15], that can be treated as a part of the anthropological analysis or as an independent field of science[16]. And although some faculties of Odontology in Brazil created centers for studies in Forensic Anthropology and provide conditions for research in this area, they still do not offer in their curriculum sufficient training, knowledge, and understanding of evolutionary processes to perform high-quality research on topics outside the specific area of Forensic Odontology. And it is especially crucial in Latin America, Brazil included, where many of the consolidated Forensic Anthropology methods are producing biased and misleading results due to the discrepancy between the biological profile of the local population and the biological profile of the collections of reference[5,17-19].

As pointed out by Christensen and others[20], to understand and interpret the results of anthropological analysis of human remains, independently on the context, whether archaeological or forensic, professionals should have a profound and broad training, not only in technical aspects of the examination but also in the evolutionary, biological, biomechanical and cultural elements and mechanisms of human variation. It is crucial to understand the morphological changes of the human body along with the history and in relation to geographic origin, sex, or individual development, which is the core of Biological Anthropology as a discipline. It is probably even more crucial in the Brazilian context, where the biological diversity of its population is exceptionally high. Therefore, the lack of such training is highly limiting when we consider the broad possibilities of knowledge production in Forensic Anthropology in Brazil. It results in a high deficit of academic professionals that could extend the research in this discipline. For this reason, and to advance and progress the production of knowledge in Forensic

Anthropology, it is recommended to include or extend courses of Biological Anthropology in the academic curriculum either at the Faculties of Odontology, Medicine or Biological Sciences, which would fill the void of professionals in this discipline prepared to analyze and research the topics of osteology, human biological variation, and taphonomy, and thus form a holistic, bio-anthropological perspective.

Not surprisingly, there exists a certain dominance of research attributed to Forensic Odontology, in relation to research that focuses on areas other than oral cavity and dental arcade parts of the skeleton. However, the prevalence is not extensive. Although odontologists conduct the majority of the research, the focus of research goes beyond their typical object of study, and this brings a notion of interdisciplinarity that is somehow required due to the lack of professional biological anthropologists in the country.

Human identification, as the most common research topic, is coherent with one of the fundamental aims of Forensic Anthropology that is the identification of unknown skeletal remains[20]. Methods that allow the fastest, most accurate, and most precise identification and comparison with a possible known person are of utmost value in forensic analysis. Next to sex estimation, this topic was presented with the highest amount of proposals for new methodologies and techniques. As Gualdi-Russo and Fonti[4] indicate, new approaches to various topics covered by Forensic Anthropology could be the direction in which the discipline will continue developing. According to these authors, research on skeletal remains will continue being the main area of investigation. However, studies on living subjects would increase its contribution to the total production of knowledge in this area. In the Brazilian context, this could be a possible line of research, especially regarding the topic of age estimation, which was the second most common subject of research detected in this analysis.

The first substantial rise in the number of articles published in scientific journals is observed in 2008. This intensification is coherent with the general evolution of the quantity of published scientific articles in Brazil[21], as well as with the general trend observed within the field of Forensic Anthropology worldwide[4]. Apart from the overall development of science in Brazil, such increase could be also be attributed to the outburst of the so-called CSI effect[22,23], which although often questioned, the belief of affecting the police, jurors, judges, prosecutors, defendants, and victims[24] could also have a meaningful impact on students and researchers[25,26]. For as much as there has been studied and published on the effect of forensic shows on various sectors of the society, we are not aware of research that would analyze its possible influence on the

scientific community. Even if no such effect can be demonstrated, the shows that portray the work of forensic specialists certainly have brought more visibility to the area, promoting interests among both students and professionals[25].

The fact that more than half of publications appear in international and national journals with Impact Factor – those with greater reach and higher citation record - validates the quality of research that is being conducted in Brazil in the field of Forensic Anthropology and Odontology. Although the concept of Impact Factor is often questioned[27,28], it remains the principal indicator of the reach, and hence the quality of a scientific journal[29]. These results can, of course, be improved, and the prevalence of the English language, even in national publications, demonstrates an attempt and will to reach the biggest audience possible. A modest trend was observed in the increase of the proportion in the number of papers written in English in relation to those written in Portuguese, although during the last year included in this research, this proportion considerably dropped. It is difficult to predict the tendency for the next years, but it is expected that English-written publications will at least maintain their prevalence in the global production of knowledge.

Scientific collaboration is an inherent part of contemporary scientific production. It promotes the exchange of knowledge, development of science, integration among scholars, and the growth of the potential for complex problem solving[30]. Working together among academics offers the possibility of acquiring new skills, understanding different points of view, and expanding the scope and the range of research. It has been reported that effective co-authorship is often related to a publication in journals with higher Impact Factor, and the paper is more frequently and longer cited[30,31]. What is interesting, as Frenken and others[32] noted, is that an international co-author increases the citation rate to a greater extent than a national one. Diverse input of different co-authors contributes to various angles, perspectives, and backgrounds, which may increase the quality of the research, as the historical specialization of knowledge greatly narrows down the area of expertise of scientists[33].

Additionally, funding agencies, both national and international, foster, or even force such actions, as it becomes more common and probable to obtain funds when a project counts with the participation of several researchers from diverse institutions[34]. In this context, the production of knowledge in the field of Forensic Anthropology and Odontology in Brazil leaves much to be desired. Only a bit more than half of the total production was prepared in a collaboration of at least two different institutions. In the last decade, the amount of national co-authorship presented a significant growth, yet

the internationalization of research is hardly essential. Therefore, to increase the impact and quality of knowledge produced in Brazil in the area of Forensic Anthropology, and to increase the possibilities of funding, it is recommended that Brazilian researchers could better explore the options of international collaboration.

Conclusion

The research in the field of Forensic Anthropology and Odontology in Brazil is in the phase of development and growth. This delay in relation to other countries of the so-called Global North can be related to the general slower and delayed pace of scientific progress in Brazil, as well as to the particular socio-political context of this country. The numbers in terms of scientific production are generally relatively low, but the production is quickly growing. The fact that most of the research in this field is conducted at the Faculties of Odontology results in a particular prevalence of the topic of Forensic Odontology in relation to Forensic Anthropology. The general lack of biological anthropologists in the academic scenario of Brazil is probably responsible for the fact that some crucial issues related to the field are not being addressed. The potential of collaboration that could significantly increase the visibility and quality of the research could be better explored.

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Conflict of interest

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Original article

Forensic Anthropology

Retrospective analysis of forensic anthropology cases investigated by the Legal Medicine Institute of Rio de Janeiro State, Brazil

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ABSTRACT

Forensic Anthropology plays an essential role in forensic investigations. The application of its concepts, from the crime scene to the laboratory, is critical to avoid that traces be ignored or lost during a criminal investigation on human skeletal remains. A Forensic Anthropology Office was created in the State of Rio de Janeiro at

the end of 2010 with the purpose to help in criminal investigations. In 2011 and 2012, the Anthropology Office received 66 examination requisitions, comprising 74 human skeletal remains. The biological profile established after the study of the skeletal remains showed that the samples were constituted mainly of male subjects (80%), white (32%), and young adults ranging from ages 21-50 (54%), considered as the risk group. The injuries most frequently observed were blunt trauma (33,3%), followed by gunshot (31,4%). The most susceptible body regions were head and neck, affected in 42% of cases. Gunshots represented 52,1% of injuries found on the head and neck. Only 18,8% of the human remains under analysis contained more than 95% of all skeletal bones. Likewise, only 7 (10,1%) of 47 completely skeletonized remains had more than 50% of his bones recovered. The delays in locating human remains and their continuing decomposition poses a marked reduction in the number of bones recovered, confirming the necessity of a trace collection carried out by trained professionals. Lastly, 80% of incoming cases came from 18 of the 160 districts of Rio de Janeiro, indicating a significant prevalence of human remains found in conflict areas.

***Keywords*:** Forensic Anthropology; Forensic Dentistry; Forensic Medicine; Forensic Sciences.

Introduction

Rio de Janeiro is a large city, with around 6 million inhabitants and an area of 1,200 km2, according to the 2010 census carried out by the Brazilian Institute of Geography and Statistics (IBGE, Portuguese abbreviation). The tremendous social differences provide, for some, ideal ground for violence to germinate. The great land extension, presence of rivers, mounds, lakes, mangroves, and forests, as well as outcast areas where the State has only recently attempted to stand ground, may function as clandestine graves; the tropical weather and rich and abundant fauna accelerate decomposition, decreasing the time-lapse and viability for a conventional necropsy.

In the State of Rio de Janeiro, the Forensic Anthropology Office (SAFO, Portuguese abbreviation) was established in October 2010 within Afrânio Peixoto Legal Medicine Institute (IML-AP, Portuguese abbreviation), linked to the Technical-Scientific Police Department branch of the Civil Police of Rio de Janeiro State.

Therefore, as in most Brazilian States, the forensic examination is subordinated to the Public Security Secretary, that is, constitutes a police unit[6,10-12].

SAFO´s establishment generated demand for specialized professionals, which contributed to gather all field professionals in the country, and inspired the creation of the Brazilian Association of Forensic Anthropology (ABRAF, Portuguese abbreviation) in September 2012, encouraged and supported by the Forensic Anthropology Society of Europe (FASE) and, similar to the latter, aims to contribute to the specialize, homogenize and publicize FA concepts[13, 14].

The demand mentioned above for professionals to act in forensic institutions should be supplied by Universities, leading to the creation of careers and development of research that will offer back scientific basis and support in the future. Forensic and academic institutions are two sides of the same coin[2, 6].

Proper qualification and the adoption of incentive policies towards the scientific police contribute to the investigation process and helps decrease impunity, while its inappropriate functioning paves the way for a rise in violence. Rodrigo Garrido alerts to the difficulties generated by the distancing between forensic and academic institutions and states that their approximation and strengthening support and add consistency to court decisions, being fundamental to the confrontation of violence.

This process depends on the administrative and financial autonomy of forensic investigation organs[4, 6, 7]. The Nina Rodrigues Legal Medicine Institute, in Bahia, and the Center of Legal Medicine (CEMEL), in Ribeirão Preto, are well-succeeded examples of ongoing partnerships between institutions[2, 6, 8].

Forensic Anthropology is the science that brings together concepts of Physical Anthropology and Forensic Sciences[10]. Its routine includes the search for trace evidence during the examination of skeletal remains, aiming to identify the victim, analysis of traumatic injuries that may have occurred and reconstitute all events leading to death, including an estimate of the time of death, or postmortem interval (PMI)[5, 11, 13, 15].

The application of FA concepts is not restricted to skeletonized remains[12] and is applicable in cases of age estimation of live subjects without documents; identification and age estimation of criminals and crime victims of video recorded felonies, such as child pornography[13,15]; verification of age of majority of criminals is also a part of the anthropological analyses group[16]; and in the event of mass disasters, as well as in crimes against humanity and human rights violations[1, 11, 15, 17], mainly when clandestine graves are located.

The complete and detailed examination of the crime scene represents an element of prime importance to crime-solving. Every time an investigation involves buried corpses, it is mandatory that the search for remains be made using archeological excavation techniques, carried out by trained professionals, in a way that no trace may be lost or destroyed[1, 3, 5, 10, 11, 13, 15]. The employment of such excavation techniques tends to be very helpful in cases involving narcotraffic victims, serial crimes, massacres, political crimes, homicides, and others[1–3, 5–9].

It is doubtless that FA is a multidisciplinary science that embraces anthropologists, archeologists, medical doctors, dentists, biologists, biomedical professionals, among others, acting individually or as a team, provided they have been adequately trained[11]. Efforts are being directed towards enabling the SAFO to deploy to the field of human remains detection so that it can collaborate more efficiently with crime elucidation. As stated by Cattaneo, today FA lies way beyond calipers and osteometric tables[13].

In Brazil, FA lacks support to research to identify the physical peculiarities of the local population, and thus validate the techniques used in forensic examinations. The first step is to create an osteological collection that reflects the current population. For example, the Piracicaba Dental School (FOP-UNICAMP Portuguese abbreviation) is developing, with the technical support of the ABRAF, a skeletal collection consisting of 320 complete and cataloged specimens from the current, local population, so far.

Aware of the contribution FA offers to criminal case elucidation, and, consequently, to decrease crime[3], this study aimed to collect and organize data produced by the SAFO. The secondary objectives were to present its examination protocol and to identify the steps of action, where much effort is still necessary to increase the quantity and quality of traces recovered, so that the investigation efficiency may be improved.

Materials and Methods

The research was approved by the Ethics Committee on Research of FOP-UNICAMP, under registration number 040/2014, and authorized by the Technical-Scientific Police Headquarters (DGPTC, Portuguese abbreviation) from the State of Rio de Janeiro. The study comprised a retrospective analysis of information derived from individual examination reports generated by the SAFO of IML-AP in the years of 2011 and 2012.

Data were extracted from each report about the site of corpse location; time elapsed from death; biological profile: sex, age, ancestry, stature, laterality and additional elements that might contribute to identification; and trauma analysis, including search for cause of death and for any other traces that may help clarify its circumstances.

Two SAFO forensic examiners collected all data. Over these two years, 66 examination requisitions were received, and each generated a report. Of these 66 requisitions: 6 corresponded to non-human material; other 3 represented material without forensic implications, 2 consisted of anatomical material discarded from study institutes, and 1 consisted of archeological material; and another one corresponding to a complementary examination requisition for a previously located skeleton, already accounted for statistically.

The remaining 56 requisitions comprised skeletal remains of a total of 74 different individuals since in 9 cases there were bones from more than one person commingled.

Five out of 74 skeletal remains did not enter the SAFO laboratory because they had been positively identified by the Forensic Dentistry Office or through Papiloscopy, due to the presence of soft tissue remains. Therefore 69 skeletal remains were examined, and the resulting data compose the present study. All examination requisitions informed the site of skeletal recovery, so all 74 cases were accounted for statistically.

Results and Discussion

Taphonomy

Quantity of bones recovered

Out of 56 requisitions, 47 (83%) presented skeletal remains of single individuals, while, in other 9 (17%), the commingling of skeletal remains took place. These nine requisitions comprised a total of 27 individuals, although the single request with the highest number of subjects investigated presented skeletal remains of six different people. As previously mentioned, laboratory analyses were centered on 69 skeletal remains because 5 of 74 had been identified before entering the SAFO. Less than 18,8% of skeletal remains recovered comprised more than 95% of body bones (Table 1).

Table 1. Descriptive statistics of recovered bones.

Bones recovered per individual	2011	2012	Total	%
1- complete or almost complete	6	7	13	18,8
2- almost complete, most long bones and many small bones	1	3	4	5,8
3- the majority of long bones and small few or rare small bones	6	1	7	10,2
4- few long bones e few small bones	10	8	18	26,1
5- few long bones	13	9	22	31,8
6- bone fragments	1	4	5	7,3
Total	37	32	69	100

Stages of decomposition of recovered material (taphonomy)

It was observed that 38 (55,1%) remains examined showed signs of degradation, indicating they had been exposed to the environment for a long time before being located. Besides the skeletal remains, the laboratory received partially saponified corpses, partially mummified corpses, and even at advanced stages of decomposition, namely bloat and liquefying stages (Table 2).

A total of 47 (68,1%) of 69 skeletal remains were fully skeletonized, that is, presented no soft tissue remains or cartilages. Of these, only 7 (10,1%) had more than half of the bones recovered, proving that the longer it takes for remains to be located, and with its subsequent decomposition, there is a marked reduction in the number of bones recovered.

Table 2. Descriptive statistics of body taphonomy.

Stage of tissue decomposition at the moment of recovery	2011	2012	TOTAL	%
1- Degrading bones	18	20	38	55.1
2- Skeletonized	8	1	9	13
3- Partially skeletonized	9	1	10	14.5
4- Putrid – the presence of soft tissue	1	2	3	4.3
5- Calcined	0	4	4	5.8
6- Mumified/Skeletonized	0	4	4	5.8
7- Saponified/Skeletonized	0	1	1	1.5
Total	36	33	69	100

Partially skeletonized – the presence of cartilages, little soft tissue, putrid odor.
Skeletonized – the presence of entire bones, absence of cartilages or soft tissue, and lack of putrid odor.

Sex analysis

Of the 69 skeletal remains, 65 underwent sexual dimorphism analysis, and the anthropological examination concluded that 55 (80%) displayed male characteristics, and 10 (14%) displayed female characteristics. All skeletal remains that comprised at least the skull, pelvis, femur or humerus allowed for gender estimation. Only four skeletal remains (6%) did not present at least one of the cited bones and were, therefore, not considered for the examination.

Pelvic bones represented the first choice for analysis of sexual dimorphism, followed by the skull. Morphological characteristics were analyzed and organized into a decision table with twelve anatomical items to be examined on the pelvis[18] and fourteen on the skull. Landmarks were selected from the study by Buikstra, Ubelaker, and Walker[15, 19–21]. Each feature was observed and classified according to the characteristics found, as male, female, or undetermined. The method for sex estimation based on pelvic metric characteristics, known as DSP2, was also applied[15, 22].

Age analysis

As far as age was concerned, it was observed that fetuses or individuals with ages ranging from 0 to 10 years were not received. 10% of the sample consisted of individuals with age ranging from 11 to 20 years, 34% were included in the age ranging from 21 to 30 years, 20% were in the range between 31 to 50 years and, finally, 20% were included in the group above 50 years. Another 16% did not contain the bones needed for the exam. Individuals with age ranging from 21 to 30 years, represents the group with the highest associated risk, followed by the age range of 31 to 50 years.

As for age estimation, methods used in young adults were based on dental development tables developed by Nicodemo, Moraes e Medici Filho[23]; on Ubelaker´s chart cited by Couto[24], and on the epiphyseal plate, calcification described by Buikstra and Ubelaker[21]. When it came to adult skeletons, the analysis was based on the stage of development of the pubic symphysis, proposed by Suchey, Brooks[15, 24]; the first costal arch by DiGangi et al.[25, 26]; the fourth costal arch[27], and the auricular surface[28]. Lamendin´s analysis was applied as often as possible[15, 29, 30].

Ancestry analysis

In 31 (45%) of 69 cases, ancestry research could not be carried out because of missing skulls or due to their fragmentation. From the 38 human skeletal remains that allowed for an ancestry analysis, it was concluded that 22 (32%) displayed predominantly European characteristics, 14 (20%) showed predominantly African features, and 2 (3%) displayed mostly Asian characteristics.

The skull represents the main point of interest while investigating ancestry. The face, especially the nose area, were morphologically analyzed, as advised by Bass[31,32], Rhine[33], Gill[33–36], and Krogman and Mehmet[37]. The morphological analysis of the face is known as the most assertive means to evaluate geographical origin.

While examining the skull, a decision table of 17 anatomical characteristics was observed. The six craniometric indices proposed by Arbenz were also applied[23]. The postcranial skeleton was used only to confirm the findings of the skull, where tibiofemoral and radiohumeral indexes cited by França were applied[38].

Stature analysis

Stature estimation was based on the Trotter & Gleser table cited by Couto[24], and on the table proposed by Mellega [39]. The latter represents a study conducted on the Brazilian population, offering good results when reproduced. Height is a parameter that should be evaluated with restriction because, in Brazil, height is only measured for military service purposes around the age of 18. As often as possible, it is suggested that ratios be used based on photographs of the missing person and that additionally, measurements of siblings, should there be any, be taken as a frame of comparison.

Functional laterality analysis

This analysis was introduced to the formal examination protocol at the end of 2011 after being considered a relevant determinant that may aid in identification. Since then, 46 skeletal remains were examined, although only ten could undergo the analysis to determine the side of dominance because they comprised bones of the appendicular skeleton of both left and right sides. One of the exams was inconclusive, and the other nine suggested individuals were right-handed.

The protocol followed was the same that has been used and supported by CEMEL, in Ribeirão Preto, for more than 10 years[8]. It is based on a decision table that

analyzes and compares eight anatomical features on both sides of the upper appendicular skeleton.

Forensic Facial Approximation (FFR) and Craniofacial Superimposition

Between 2011 and 2012, two exams with craniofacial superimposition and three exams of FFR were carried out. Both craniofacial superimposition exams demonstrated coincidence between facial and craniometric landmarks, as did facial characteristics match. Later, DNA tests confirmed the results obtained by approximation.

The FFR was carried out digitally and still cannot function as a means of attracting the missing person´s family members because its results cannot be released to the general public. In one case where the family waited for the DNA result to confirm the identity of skeletal remains, the FFR was applied, and the family recognized the result presented as similar to the missing person´s appearance.

The techniques of overlapping images and forensic facial approximation were used eventually by the SAFO and are not part of the routine protocol. The entire process is usually concluded in a single day[40].

Identification

In its first two years functioning as a formal unit, SAFO voided one case of corpse identification that had been wrongly recognized by family members. In another case, the SAFO was responsible for the inclusion of skeletal remains as a possible match after pointing out coincidences between information obtained from family members and the victim´s biological profile. A DNA test confirmed SAFO´s findings.

The SAFO positively identified a case where the biological profile, the FFR, OI matched, and the anterior teeth could be observed, including the gaps between them. It should be noted that at least 29 of 69 skeletal remains examined by the SAFO presented characteristics that could contribute to the identification process, such as dental fillings, fractures and antemortem pathologies, characteristic anatomical variations such as septal and sternal foramina, bifid ribs, metopic suture, sacral bifid spine, sutural bones, ankylosis, unerupted teeth, and osteomas. Again, it is necessary

to bring attention to the fact that a missing person´s database could highly improve the statistics of positive identifications.

Analysis of trauma injury

In the first stage of trauma injury investigation, the triple distinction was made between antemortem, perimortem, and postmortem. From the 69 skeletal remains analyzed, 44 (63,8%) showed some skeletal trauma, namely: blunt force, burning, sharp force, gunshot, or an association between these. In 25 (36,2%) of 69 skeletal remains, no trauma injury was observed.

The most frequently observed injury was blunt trauma (33,3%), followed by gunshot (31,4%), sharp-blunt trauma (21,6%), and burning action (13,7%). Six out of 44 skeletal remains presented an association between more than one type of injury.

The head-neck region was affected in 42% of the 69 cases, representing the most frequently affected location, and having been subjected to all four types of trauma injury evaluated. The upper arms were affected in 13% of cases; the chest-abdomen region was affected in 27,5% of cases. Finally, the hip region was affected in 13% of cases and the lower members in 14,5%. In some cases, the five body regions were affected.

The highest correlation found between a type of trauma, and a specific body part was the gunshot action to the skull-neck region, corresponding to 27,3% of the cases. Of all gunshot injuries, 52,1% were located in the skull-neck area.

Correlation between the site of location and trauma

Of all cases investigated, 80% were located in one of 18 out of 160 districts found in the city of Rio de Janeiro, indicating a substantial prevalence concentrated over few areas and also showing that some specific regions display above average according to violence numbers.

An association between recovery sites and type of trauma injury could also be identified, indicating that criminals acting within a specific district display a modus operandi, as 15% of victims who suffered multiple sharp-blunt injuries, in this case, the scattering of body parts, were found in the surroundings of Ilha do Governador district, while gunshot injuries to the head and neck prevailed (25%) in Realengo district.

Trauma and biological profile correlation

The biological profile established by skeletal remains investigation evidenced that the sample was mainly comprised of men (80%), white (32%), young-adult with age range between 21-50 (54%), representing the risk group in 11,59% of studied cases.

The authors have presented the methods used in the examinations and the results obtained after two years of service. Results are still limited due to the reduced sample size, but the presentation of the method employed is significantly relevant because it exposes techniques that have proved efficient.

Final Considerations

We emphasize that the methodology presented in this work were used in the creation of the forensic anthropology section, in 2010, and all cases at the time culminated in success. Currently, the techniques used for the diagnosis of ancestry are based on Hefner (2009). For sex assessment, we use DSP2, Klales et al. (2012)[41], and Walker (2008).

Also, we understand that there is no metric methodology, in the literature, that can replace morphological methods. In this sense, we prioritize its uses due to the practicality, reliability, and success rate in obtaining data.

Conclusion

It was possible to point out the existence of a risk group, made up of young-adult, white males. It is noteworthy that a high number of skeletal remains were located in a few regions, and that patterns of trauma can be associated with specific districts, thus establishing a correlation between types of homicide and certain parts of town. Gunshot to the head and neck was the most common combination of trauma and body injury.

The establishment of biological profiles and trauma injury analysis must be carried out by trained and experienced professionals in the forensic anthropology field, which can optimize the trace investigation.

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Conflict of interest

There are no known conflicts of interest associated with this publication, and there has been no significant financial support for this work that could have influenced its outcome.

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Original article

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Sexual dimorphism of the mandibular basis: A logistic regression analysis

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ABSTRACT

The human identification process can be a complex role because whole bodies are not always found in a suitable preservation state. In these bodies, sex estimation is necessary. Sex estimation has a higher probability of success when all bones of the subject are examined. However, some bones present a more pronounced sexual characteristic, namely: pelvis, skull, mandible, and femur. The present study analyzed the efficiency of metric criteria on the mandible base to estimate sexual dimorphism in Brazilian adults. The measurements of 214 mandibles were studied; there were 111 males and 103 females aged over 22 years old. The sample was gathered in the Santa Casa cemetery in the state of Rio Grande do Sul, Brazil. Thus, three landmarks

were selected on the mandible: Menton, Right-Gonion, and Left-Gonion; consequently, three measures were obtained using a digital precision pachymeter. Measurements were statistically analyzed using Stat 11.1 Software. All parameters showed very significant sexual dimorphism ($p<0.0001$). Five approaches of logistic regression were addressed to sexual prediction; 2 of them become more suitable for sex classification when compared with the others. The percentage of success was 81.39% to Multivariable Logistic Regression and 86.04% to Univariable Logistic Regression. In this work, the metric criterion through the Logistic Regression has been an important tool for sex estimation in the forensic area.

Keywords**:** Forensic Sciences, Forensic Anthropology, Human Identification, Sex Dimorphism, Logistic Regression.

Introduction

The forensic dentistry identification process tries to determine the biological profile of the subject when skeletal remains are found and human nature confirmed; information such as sex, age, biotype, and height are essentials[1,2]. Sex estimation in the forensic anthropological context has a higher probability of success when a complete exam on all bones of the subject is performed. Sometimes bones are missing or destroyed; in these cases, the analysis should be performed on specific bones because the sexual dimorphism is more evident, namely: (a) the pelvis and the skull[2–8], (b) the mandible, morphological[3–5,9,10] and metrically [1,2,17–20,6,7,11–16], and (c) the long bones[3,5,14]. Therefore, the analysis of single bones and skeletal fragments have an essential role in sex estimation.

Morphological indicators through visual analysis depend on the ability and experience of each specialist[15,21]. This method is frequently used for all kinds of populations but is more susceptible to errors due to the subjective criteria of each researcher[3]. The intra/inter-observer errors can be decreased using metric evaluations, which are more accurate, reproducible, and standardizable[21,22].

Several populations have significant skeletal differences, which take into account the metric evaluation of size and proportions. Thus, specific data to each population are required. Different measurements on mandibular bone for sex estimation have reported a high percentage of correct classification on samples from the following populations: *(a)* South-African, with 80-86%[23], 80-85 %[7] and 63.6-84%[1],

(b) French, with 87.3%[11], *(c)* American, with 85%[24], *(d)* Egyptian, with 83.6% for male and 84.2% for female[12], *(e)* Indian, with 81.5%[2], 80.2%[6], *(f)* Tibetan, with 88.2%[2], *(g)* German, with 88.6% for male and 79.5% for female (whole mandibles) and 72.5%-81.7% for male and 71.6%-79.5% for female (fragmented mandibles)[13], *(h)* Canary Islanders, with 71.2%-72.2%[14], *(i)* Chinese, with 80.5 to 84.2% with multivariable functions[16], and *(j)* Brazilian, with 93.33% for male and 94.74% for female[17], 76 to 86% with multivariable functions[19], and 76.47% for males and 78.13% for females[20].

This study aimed to estimate the sexual dimorphism on 214 mandibles of the Brazilian adult sample in Rio Grande do Sul state, using metric analysis, using the following measurements: Menton (Me) - Right Gonion (RGo), Menton (Me) - Left Gonion (LGo) and Right Gonion (RGo) - Left Gonion (LGo). The effectiveness of the metric criteria was analyzed on the mandible bone; functions with few variables were found and are ready to use when the mandible bone is available for forensic purposes.

Material and Methods

Data collection was gathered at Santa Casa cemetery, Porto Alegre (RS). The Research Ethics Committee of Piracicaba Dental School, University of Campinas (FOP-UNICAMP), approved this research under protocol number 138/2010. Statistical analysis and data processing were performed in the FOP-UNICAMP. The measurements of 214 adult mandibles were studied of both sexes, all of them from known origin, with absolute certainty about the sex; 111 males and 103 females over 22 years old in the moment of death. All measurements were performed on the premises of the cemetery. After research, the bones were returned in their entirety. Entire mandibles were selected without apparent external trauma (mainly in the mandible base), pathologies, anomalies, or skeletal malformations that could interfere with the measurements.

A Mitutoyo digital pachymeter was used to take the measurements. Three mandibular landmarks were established, as follows: (a) Menton (Me), the point on the middle of the mandible corpus, placed in the most prominent part of the mentonian eminence; the mandible is oriented in the Frankfort plane, (b) Right Gonion (RGo) and (c) Left Gonion (LGo), in which Gonio (Go) is the point on the mandible where the inferior margin of the mandibular corpus and the posterior margin of the ramus meet, i.e., the point on the mandibular angle which is directed most inferiorly, posteriorly, and laterally (Fig. 1). Two approaches were selected to sexual dimorphism study from Mandibular landmarks through the:(a) linear measurements: Me to RGo (Fig. 2a); Me

to LGo (Fig. 2b); RGo to LGo (Fig. 2c), (b) area of the triangle formed by the linear measurements (called a, b and c) and calculated using the Heron formula (Fig. 2d).

Fig. 1 - Top view of the mandible and landmarks: Right Gonion (RGo), Left Gonion (LGo), and Menton (Me).

Fig. 2a - Menton – Right Gonion.

Fig. 2b - Menton – Left Gonion.

Fig. 2c - Right Gonion – Left Gonion.

Fig. 2d - Area.

The measurements were repeated on 20% of the sample randomly, in two different situations, and not less than two weeks, in order to evaluate intra-observer error. The paired t-student test was used to detect possible intra-observer systematic errors. The independent t-student test was used to analyze the statistical significance of the data set related to sexual dimorphism. To the statistical tests, it was adopted a significance level of 5% ($p < 0.05$). The measurements were submitted to statistical analysis using Stata 11.1 software.

Functions to the correct sex classification were found through the Logistic Regression method; it allowed tuning the set of the independent variables to one categorical response variable. So, the sex was considered a dependent variable, where the male was associated with “1” value and female with “0” value. The predictor variables were RGo-LGo, LGo-Me, RGo-Me, and Area.

The sample comprised 214 mandibles divided randomly into two subsets; the first one with 171 mandibles (79.90%) for estimation of the logistic models, and the second one

with 43 mandibles (20.09%) for validation of the logistic models. Several variable associations were tested in order to identify the best configurations of sex classification. Altogether five logistic functions were calculated; one multivariable and four simples.

The logistic model calculates the probability of the mandibles to be male or female using the equation (1):

$$P(x) = \frac{1}{1+e^{-logit(pi)}}\ , \qquad (1)$$

Where the *logit* can be calculated by equation (2):

$$logit(pi) = \alpha + \sum X_i \beta_i . \qquad (2)$$

The α and β coefficients are found by the model, *X* is the measure on the mandible, so the *logit* calculated in the equation (2) is substituted on the equation (1), if the result is higher than 0.50 the prevision is "mandible of the male sex," otherwise "mandible of the female sex."

Results

The p-value for the paired t-student test to evaluate intra-observer error was higher than 0.05; no significant differences were observed, as shown in Table 1. which demonstrates the researcher's calibration, because of the random measurements made in week one and week two had no significant differences.

Table 1 - Paired t-test for intra-examiner error

Measure	1fst Measure		2nd Measure		p-value
	Mean	Variance	Mean	Variance	
RGo – LGo	95,681	46,413	95,688	47,278	0,415 *
RGo – Me	84,574	27,236	84,599	27,2	0,186 *
LGo – Me	84,249	22,868	84,231	23,258	0,279 *

RGo (Right Gonion), LGo (Left Gonion), Me (Menton)
*(p-value > 0,05) difference not statistically significant

The p-value of the independent t-student test to analyze the statistical significance of sexual dimorphism can be observed in Table 2. For all measurements,

the p-value was less than 0.0001; the difference between both sexes (male and female) is statistically very significant.

Table 2 - Comparison of male and female mandibular measurements using the independent t-student test.

	Male			**Female**				
Measure	**Mean (sd)**	**Min**	**Max**	**Mean (sd)**	**min**	**Max**	**t-value**	**p-value**
RGo – Lgo (mm)	98,228 (6,922)	83,61	113,33	91,644 (5,867)	76,14	107,12	7,542	< 0,0001
RGo – Me (mm)	85,974 (5,739)	73,2	101,98	81,958 (5,036)	69,61	97,36	6,094	< 0,0001
LGo – Me (mm)	85,892 (5,562)	73,02	102,86	81,492 (4,472)	71,19	93,37	7,071	< 0,0001
Area (mm^2)	3452,409 (377,688)	2648,59	4262,74	3092,929 (300,298)	2356,76	3912,02	8,841	< 0,0001

RGo (Right Gonion), LGo (Left Gonion), Me (Menton) (p-value < 0,0001) difference statistically very significant.

The arithmetic mean of the parameters is shown in Fig. 3. The difference between the sexes is evident, and a confidence interval of 95% was adopted.

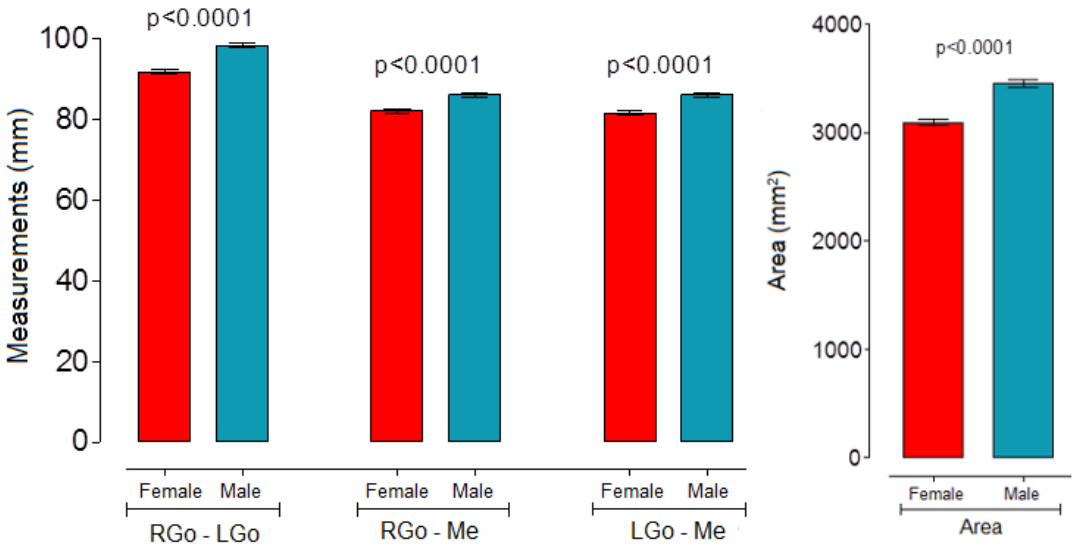

Fig. 3 - Difference between male and female mandible measurements.

Five logistic functions were calculated; one multivariable and four simples. Table 3 shows the logistic functions found by the software from the data set.

Table 3 - Logistic Regression found

No.	Logistics Regression	Description
1	Logit Pi = -19.3399 + (0.1039 X_1) + (0.0375 X_2) + (0.0769 X_3)	Multivariable: X_1=BGo, X_2=RGo-Me, X_3=LGo-Me
2	Logit Pi = -12.4921 + (0.1324 X_1)	Simple: X_1=RGo-LGo
3	Logit Pi = -11.3688 + (0.1368 X_1)	Simple: X_1=RGo-Me
4	Logit Pi = -13.3458 + (0.1610 X_1)	Simple: X_1=LGo-Me
5	Logit Pi = -8.8583 + (0.0027 X_1)	Simple: X_1=Area

RGo (Right Gonion), LGo (Left Gonion), Me (Mentonian), BGo (RGo-LGo).

To calculate the probability of a mandible being male or female, once the value of the *Logit Pi* shown in table 3 has been obtained according to the variables of interest, it will be replaced in equation (1) as indicated in material and methods. Table 4 shows the success percentage of the sex classification for the different logistics regressions.

Table 4 - Validating of the Logistic Models

Logistic Regression	Male		Female		Total
	Successes (%)	Failures (%)	Successes (%)	Failures (%)	(%)
1	19 (86.4)	3(13.6)	16 (76.2)	5(23.8)	81.39
2	19 (86.4)	3(13.6)	18(85.7)	3(14.3)	86.04
3	16(72.7)	6(27.3)	11(52.4)	10(47.6)	62.79
4	20(90.9)	2(9.1)	12 (57.1)	9(42.8)	74.42
5	16(72.7)	6(27.3)	16(76.2)	5(23.8)	74.42

The multivariable logistic function No. 1 was found from the BGo, RGo-Me, and LGo-Me variables with a success percentage of 86.4% for males and 76.2% for females. The simple logistic functions No. 2, 3, and 4 were established from BGo, RGo-Me, and LGo-Me independent variables, respectively. Logistic function No. 2 shows the RGo-LGo measure as the most dimorphic variable for classification, with a performance percentage of 86.4% for males and 85.7% for females. Logistic function No. 3 had an acceptable rating for males 72.7% and low for females 52.4%, while logistic function No. 4 had a high-performance percentage for males 90.9% and a low percentage for females 57.1%. The simple logistic function No. 5 was obtained from the area variable, which was calculated. The performance percentage was 72.7% for males and 76.2 for females.

Discussion

The growth patterns of the human skeleton have a faster evolution of the neural dimensions in childhood and slower evolution in the diameter of the large bones. This development sequence shows an early growth of the neural structures followed by the middle face and part of the skull base, in contrast with the more gradual development of masticatory structures[3,25]. The highest relative growth is seen in the mandible from

childhood to adulthood, followed by the maxilla, upper face, skull base, and skull height, respectively[26]. The skeleton parts that present early growth are less dimorphic than the areas with slow growth, from sexual viewpoint[2,4], so the mandible is a bone highly dimorphic.

The mandible is one of the hardest and most durable skull bones[5,15]; it is commonly preserved in anthropological[1–3,6,10,27,28] and archaeological[3,14,27] contexts, due to the high resistance to adverse taphonomic processes[1,2,29]. A human mandible has been evaluated for its usefulness in the classification of human groups describing differences between modern forms and fossils, and in particular, in determining sexual dimorphism patterns. It is a useful element in forensic sciences such as dentistry, anthropology, and legal medicine. Multiple factors influence the features found on the mandible within the framework of sex determination, namely: patterns of late growth[2,4,25], sex hormones[3–5,9,18,30] and muscle attachments with forces that differ between the sexes[3,5,9,10]; some of which were detected in this work showing the mandible highly dysmorphic for both sexes.

The skull-mandibular sex estimation is not reliable until the secondary sexual characters appear in adulthood[2,4,6], so only mandibles with over 22 years were part of the sample. The mandible has shown a satisfactory performance morphologically[3–5,9,10] and metrically[1,2,17–20,6,7,11–16]. The sex estimation defines a starting point to the biological profile, it is one of the most important components due to the fact age, and height estimation follow different patterns in men and women[6,26]. In this research conducted in the Rio Grande do Sul state, Brazil, a metrical sexual dimorphism was found that at best reached a success rate of 81.39% with RL multivariable and 86.04% with RL simple, considering this bone element useful for forensic purposes.

Morphological indicators are more susceptible to errors due to subjective criteria of each researcher[3,21]; disadvantages have been shown when compared with metric evaluations, which are more accurate, reproducible, and standardizable, decreasing intra/inter-observer errors[21,22], and that is the main reason why this method was chosen in this work. In sex estimation on skeleton remains, the most important thing is the accuracy of the available patterns and their suitability on badly maintained bone elements[1]. Morphological studies can be applied on burned or fragmented remains, showing advantages when compared with metric methods[26], but metric studies applied to bony fragments are shown to be a viable alternative[6,13]. This paper explores measurements in the mandible base, showing the application of the functions found on mandibles with broken ramus.

The mandible shape and size vary in accordance with the cultural habits and adaptations to local environmental conditions; hence their features vary between different ethnic groups[3,23,31]; it related to the specific biomechanical needs, which has a functional and evolutionary meaning[4,6,9]. The significant differences of the skeleton through populations require specific data that fit each one[1,2,30,7,8,12,14,16,19,20,23]. Thus, the objective of this work was to obtain a metric criterion for sex estimation in a Brazilian population sample for forensic identification cases.

The Bigonial breadth (RGo-LGo) measure and the mandibular body length (Gonion-Menton) show good results in different populations with p-value to Bigonial breadth of 0.01[2,14,17], 0.001 significant[1,2,7,23,] 0.0001 very significant[15,16,19], and to mandibular body length of 0.001 significant[1,2,24], 0.0001 very significant[11,15,16]. The Brazilian population of Rio Grande do Sul has shown a p-value of 0.0001 for all metric variables evaluated in this research. Several measurements have been reported on mandibular bone to sex determination, showing success percentage between 63.6 – 94.74%[1,2,19,20,23,24,6,7,11–14,16,17]. In this work, the success percentage varied between 74.42 - 86.04%, showing a high degree of sexual dimorphism.

Currently, techniques with advanced technological resources to sex determination are used; computed tomography scanners allow getting 3D images, and computational tools allow posterior digital processing[8,12,16,17,31,32], proving efficiency and decreasing errors in both morphological and quantitative methods; but the costs are exponentially increased. Craniometry, on the other hand, is a practical method with lower cost and which has proven reliable[19,20,32], ratified in this work.

Conclusion

Sex estimation was presented through logistic regressions on the mandible base; success percentage varies between 74.42 - 86.04%. Therefore, sex estimation by quantitative analysis through mandible or their fragments have been accurate and objective. The method can be used as support to current techniques for sex determination, enabling quantitative comparisons. On the possibility of experts provide only the mandible base (even without the presence of the ramus), this method explores metric variables establishing their usefulness in sex determination for forensic purposes.

The mathematical model employed in this work using logistic regression contributes to strengthening the traditional metric resources available to sex

determination, particularly in populations that express sexual dimorphism, as shown. The success of the adjustment to logistic regression models depend on the quality of the measurements; it was verified using a hypothesis test in which all measurements showed values with very significant differences.

Conflict of interest

There are no known conflicts of interest associated with this publication, and there has been no significant financial support for this work that could have influenced its outcome.

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Original article

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Evaluation of frontal sinus in computed tomography for human identification: A pilot study

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ABSTRACT

This study aimed to evaluate the frontal sinuses making a comparison between the measurements of the frontal sinuses and comparing the measurements obtained with height, sex, age, and body mass index body of the examined individuals in CT scans**.** Thirty patients of both sexes, who sought the radiological clinic of the University Hospital Oswaldo Cruz (University of Pernambuco) for diagnosis over four months.

The sex, age, and height of each individual were collected at the same time the patient underwent a CT scan. Ages were grouped into age groups of 18-39; 40-59 and ≥60 years of age. No significant values for the age group difference ($p > 0.05$) were observed. The other variables showed no significant difference when related to the height of individuals ($p > 0.05$). This study concluded that there were significant differences in the frontal sinus measures on men and women and individuals with greater height.

Keywords: Frontal sinus, Tomography, X-Ray Computed, Body height.

Introduction

Identification corresponds to a set of several procedures to individualize a person or object. Personal identification is crucial for forensic medicine, whether for legal or humane reasons, and is often conducted even before the determination of the cause of death. There are several human identification methods, such as dactyloscopy, the most common, and DNA analysis, also very common. Alternative methods include palatoscopy, cheiloscopy, dental arch analysis, and frontal sinus imaging. Through identification, people may secure their rights and be liable for their civil or criminal acts[1].

Several imaging techniques may be used for human identification, such as dental radiography, facial radiography, and computerized tomography (CT), which can be traditional or tridimensional. CT has several advantages over conventional radiography - it is free of structure overlapping, allows the individualization of the plan of interest, and the visualization of small differences in tissue density[2].

Frontal sinus pattern characterization is a well established personal identification technique in forensic anthropology. Variations in size, shape, symmetry, outer borders, presence, and the number of septa and cells are analyzed using radiographs and tomographies *antemortem* and *postmortem*. The frontal sinus is well accepted in the identification due to its uniqueness - its septal structure and boundaries vary between individuals, allowing a safe identification[3].

These pneumatic cavities are located in the frontal bone, which is frequently intact in fragmented skulls or isolated bones. Its role in sex prediction through radiographic analysis can benefit forensics since the accurate diagnosis can contribute to the post-mortem identification of mortal remains. However, there are some

contradictions in the literature involving these data. Some works report sex-based differences and show that male frontal sinuses are two times larger than the female sinuses[4]. On the other hand, studies analyzing anteroposterior view radiographs failed to find significant differences between male and female frontal sinuses in computerized tomographies[5]. However, Uthman et al.[6] were able to identify sex-based differences using the same radiographic exam. Therefore, considering the inconsistencies found in the literature and the easiness of frontal sinuses analysis with forensic purposes, this study aims to analyze the frontal sinuses through tomographic exams and to assess height, sex, and age influences on frontal sinuses dimensions.

Materials and Methods

This study is in accordance with Resolution 466/2012 CNS-CONEP, which concerns research ethics involving human subjects. The project was applied to Plataforma Brasil and submitted to the Universidade de Pernambuco Research Ethics Committee and approved under the process number 31173514.9.0000.5207.

This work consists of a cross-sectional study since the data concerning the variables of interest were collected simultaneously; prospective since the data were obtained from patients in the imaging service; and observational since the study aims to describe specific parameter distribution[3].

The sample consisted of 30 patients from both sexes using the radiology clinic of the University Hospital Oswaldo Cruz at the Pernambuco University for diagnosis, during four months, referred by neurology, maxillofacial, and otolaryngology. Therefore, the patients were not submitted to radiation for the sole purpose of this study. Patients with signs of trauma, younger than twenty years old, congenital pathologies involving the region of interest, previous surgery, or that presented artifacts in the tomography were excluded from the sample.

The parameters height, sex, and age for each individual were recorded at the same time as the tomographic exam. The individuals were divided according to the age range as 20 to 39 years old, 40 to 59 years old, and above 60 years old. The data were registered in an appropriate spreadsheet. Also, all participants who agreed to participate in the study signed an informed consent form.

Computerized tomography technique

The images were taken in a 4-channels *multislice*/GE computerized tomographer (General Electric, Nova York, USA), with a slice thickness of 1.25 mm and 1-mm increment. This exam produces sliced images over three planes and along the three dimensions of an object. The images were visualized with Invesalius® (CTI, Brasil), which allowed the adjustment of position and orientation of the head plans and the analysis of skull and frontal sinuses.

Frontal sinus measurements

The frontal sinus measures and volume were estimated with the program measurement tools. The measures were taken directly from the screen according to Lee et al.[3], using the tomography console cursor with 0.01 mm precision. All measures were taken by an independent trained evaluator who was not involved in height, weight, and sex data collection. All data were registered in an appropriate spreadsheet.

Horizontal and vertical lines were defined on the sagittal plane of CT for the measurements (Figures 1 and 2). The base of the skull was defined as the horizontal reference plane and referred to as the H line. Perpendicular to it, the vertical reference line was set and referred to as the V line.

Still, in the sagittal plane, line A defines the forehead inclination, determined by the glabella curvature. The forehead inclination was given as the angle (ANV) between line A and the vertical reference (V line).

Line B is the effective slope and was measured as the angle (BNV) between line B and the vertical reference (V line).

Line C represents nose inclination, and the nasofrontal angle (BNC) is, therefore, defined as the angle between Lines B and C.

Fig. 1 - Representation of line A (from nasio (point N) to the point right above the supraorbital edge); line B (from nasio (point N) the anteriormost point of the supraorbital edge); line C (from nasio (point N) to the inferior most point of the nasal bone). (Source: MATTHEW et al., 2010)

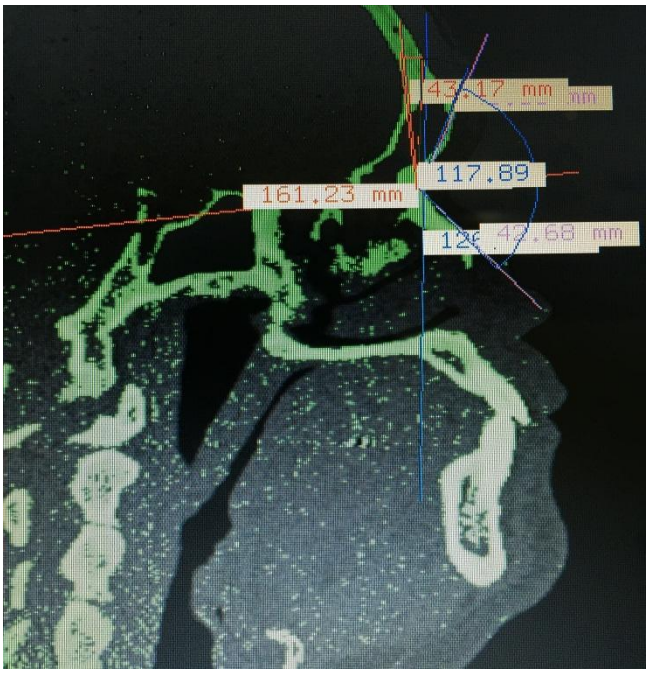

Fig. 2 - Representation of lines A, B, and C, and angles ANV, BNV, and BNC obtained from patients CT with software Invesalius.

In the coronal plane, the frontal cavity height was measured at the midline and 10 mm, 20 mm, and 30 mm of it at both sides. On the sagittal view, the anterior and anteroposterior (AP) table of the frontal sinus thickness and depth of the sinus were measured at the most protruding level of the supraorbital edge.

These measurements were also taken at the axial plane and 10, 20, and 30 mm of the midline at both sides (Figures 3 to 7). The width of the midline was measured at both sides to evaluate the left and right variations. Finally, the width of the glabella,

established as the midline of the area beyond the forehead's natural curvature, was also taken at the most protruding level of the supraorbital edge.

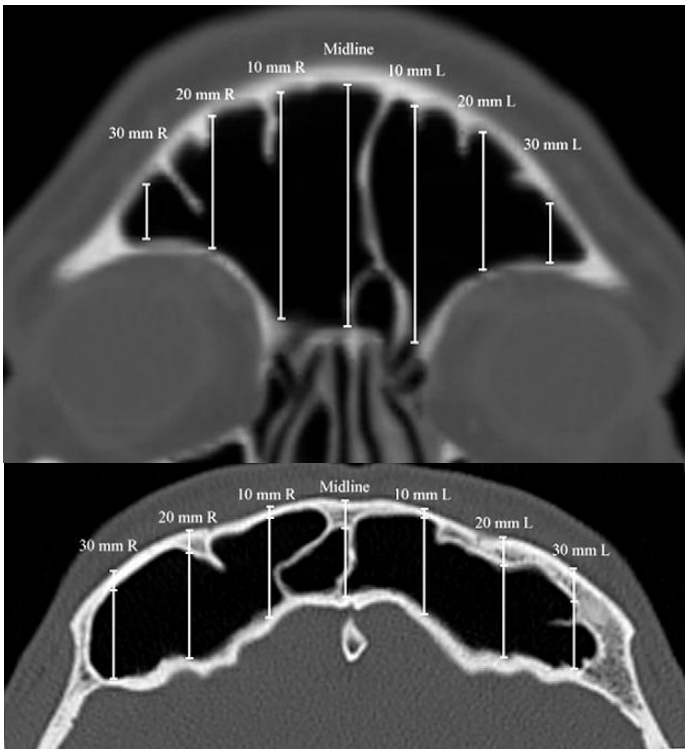

Fig. 3 – A and B: Frontal sinus height and width in axial and coronal planes. (Source: Matthew et al., 2010)

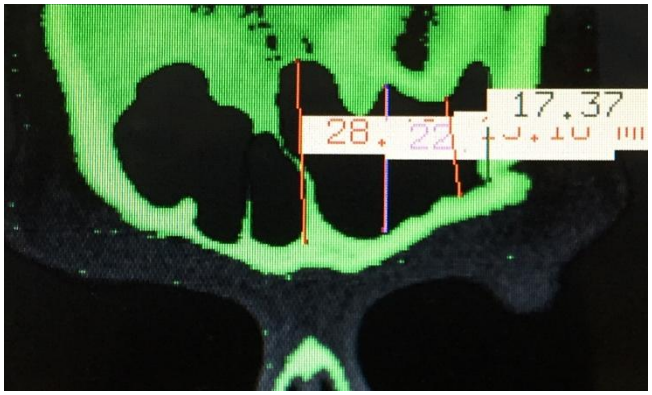

Fig. 4 - Frontal sinus height at the midline and 10 mm, 20 mm, and 30 mm of it.

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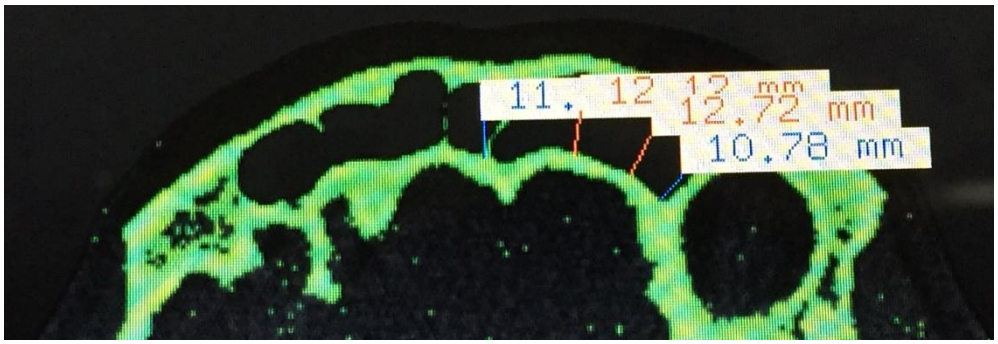

Fig. 5 - Frontal sinus thickness at the midline and 10 mm, 20 mm, and 30 mm of it.

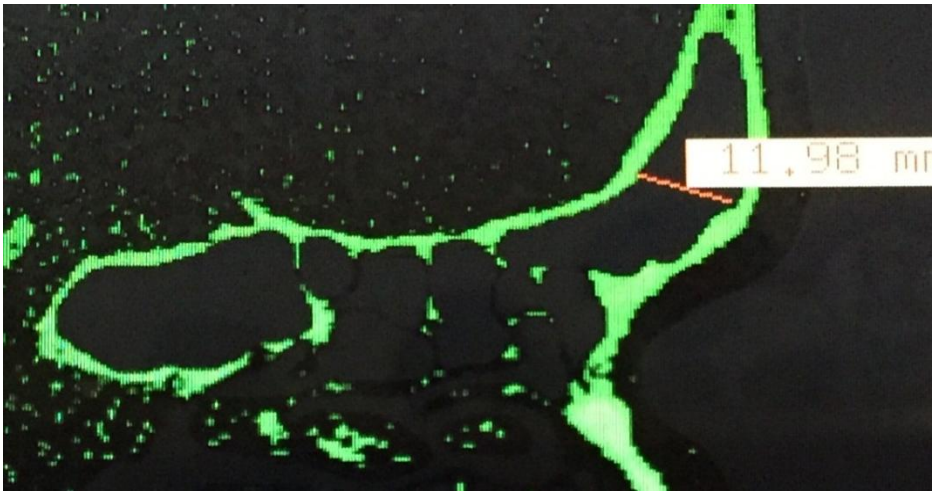

Fig. 6 - Sinus anteroposterior distance at the most protruding point.

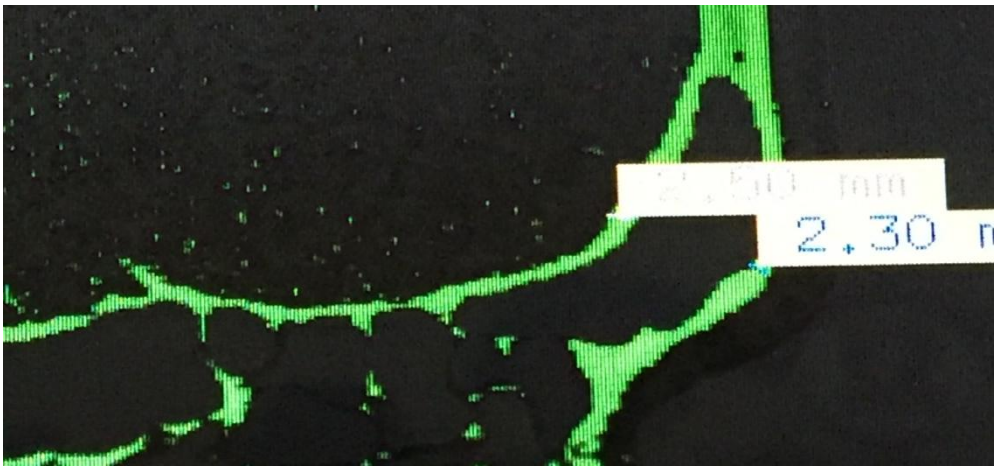

Fig. 7 - Frontal sinus anterior and posterior cortical thickness at the most protruding point

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In addition to these measurements, the frontal sinuses were evaluated according to Tatlisumak et al.[7] methodology, which allows the assessment of the following features:

- F (presence or absence of the sinus);

- S (septum);
- S (scalloping).

The frontal sinus measurements were coded according to Tatlisumak *et al*. (2007) methodology:
The following scores were given to height and width
1 - 0 mm< x ≤ 15 mm
2 – 15 mm< x ≤ 30 mm
3 – 30 mm< x

For anteroposterior length, the following scores were used:
1 – 0 mm< x ≤ 10 mm
2 – 10 mm< x ≤ 20 mm
3 – 20 mm< x

For total width and distance between the highest sinuses points, the following scores were used:
1 – 0 mm< x ≤ 30 mm
2 – 30 mm< x ≤ 60 mm
3 – 60 mm< x

The distance between the highest right sinus point and the maximum right lateral border and the distance between the highest left sinus point and the maximum left lateral border were scored according to:
1 – 0 mm< x ≤ 10 mm
2 – 10 mm< x ≤ 20 mm
3 – 20 mm< x

Tomography evaluation

A calibrated examiner evaluated the measurements using the software Invesalius® and the Evaluation Sheet (Appendix A). It is worth mentioning that at most, ten images were evaluated per day to avoid visual fatigue and compromising the evaluation.

Statistical analysis

The data were described through absolute and percentage distributions of the categorical variables and the statistical measures: mean, median, and standard deviation for the numerical variables. Data were differentially analyzed through the statistical tests: F (ANOVA) or Kruskal-Wallis for numerical variables comparing three categories and t-Student with equal and unequal variances or Mann-Whitney. The association between the categorical variables was assessed with Pearson's chi-square test or Fisher's exact test when the conditions for the application of chi-square were not met. The degree of association between the numerical variables was given by Pearson correlation and by Spearman correlation when normality was not found. The null hypothesis was tested with a specific test.

It is worth stressing that the tests F (ANOVA) or t-Student were chosen when normality was verified for each category and Kruskal-Wallis or Mann-Whitney otherwise.

Normality was verified through the Shapiro-Wilk test and variance equality through Levene's F test. All tests had a margin of error of 5.0%.

The data were organized in an EXCEL sheet, and the statistical calculations were done in SPSS (Statistical Package for the Social Sciences), version 21.0.

Results

Participants' ages ranged from 20 to 86 years old, with an average of 45.67, a standard deviation of 19.48, and a median of 42.50 years. Of the 30 patients analyzed, 12 (40.0%) were between 20 to 39 years old, 12 (40.0%) were between 40 to 59 years old, and 6 (20.0%) were 60 years old or older. From this group, 15 (50%) were male, and 15 (50%) female.

Except for ANV, variability, as given by the standard deviation, was not high since the measure remained below half the mean value of the respective variable (Table 1).

Table 1 – Statistics of the studied variables.

Variable	Statistics			
	Average ± DP	**Median**	**Minimum**	**Maximum**
• **Average sinus thickness**				
Right	7,34 ± 2,48	7,55	3,49	15,96
Left	6,98 ± 2,96	6,39	3,51	14,53
Total	7,67 ± 2,72	6,66	4,20	16,52
• **Frontal sinus total width**	47,48 ± 15,97	49,61	11,23	68,78
• **Mean sinus height**				
Right	13,10 ± 5,18	13,13	4,21	28,53
Left	13,40 ± 5,99	12,76	4,40	30,79
Total	16,35 ± 7,30	13,61	6,68	38,69
• **Anterior cortical**	3,23 ± 1,00	2,96	1,66	5,46
• **Posterior cortical**	3,88 ± 1,75	3,11	1,96	8,29
• **Anteroposterior distance**	8,79 ± 3,31	8,41	2,06	15,70
• **Angle**				
ANV (frontal inclination)	10,07 ± 6,08	11,52	0,00	20,67
BNV (Declive frontal)	24,05 ± 9,99	24,16	4,67	48,31
BNC (nasofrontal angle)	125,88 ± 11,15	126,75	96,21	144,00

Table 2 shows that, except for the variables "Posterior cortical", “ANV” and “BNC”, which had higher averages in females, all other variables were higher in average in males; except for the variables: “frontal sinus total width”, “left sinus mean height”, “Anterior cortical”, “Posterior cortical” and “BNC”, all other variables were significantly different between males and females ($p < 0.05$).

Table 2 – Statistics of the studied variables according to sex.

Variable	SEX		
	Male	**Female**	**Value of p**
	average ± DP (Median)	average ± DP (Median)	
• **Average of sinus thickness**			
Rght	8,39 ± 2,63 (8,10)	6,20 ± 1,77 (6,27)	$p^{(1)} = 0,017^*$
Left	8,20 ± 2,59 (8,19)	5,57 ± 2,80 (4,61)	$p^{(1)} = 0,001^*$
Total	9,18 ± 2,66 (8,77)	6,17 ± 1,85 (5,46)	$p^{(1)} < 0,001^*$
• **Frontal sinus total width**	51,19 ± 13,89 (50,50)	43,77 ± 17,49 (48,76)	$p^{(2)} = 0,208$
• **Mean sinus height**			
Right	15,50 ± 5,07 (14,77)	10,51 ± 4,05 (10,40)	$p^{(2)} = 0,009^*$
Left	15,18 ± 6,39 (15,91)	11,34 ± 4,96 (11,19)	$p^{(2)} = 0,091$
Total	18,90 ± 7,54 (18,97)	13,80 ± 6,30 (13,31)	$p^{(1)} = 0,049^*$
• **Cortical anterior**	3,40 ± 1,04 (3,00)	3,05 ± 0,96 (2,91)	$p^{(2)} = 0,346$
• **Cortical posterior**	3,83 ± 1,74 (2,81)	3,93 ± 1,83 (3,35)	$p^{(1)} = 0,943$
• **Anteroposterior distance**	10,21 ± 3,79 (10,68)	7,37 ± 1,99 (7,45)	$p^{(2)} = 0,016^*$
• **Angle**			
ANV	8,39 ± 5,68 (10,66)	11,74 ± 6,19 (13,43)	$p^{(1)} = 0,038^*$
BNV	27,78 ± 11,58 (30,44)	20,32 ± 6,53 (22,86)	$p^{(3)} = 0,003^*$
BNC	119,99 ± 12,34 (117,64)	131,76 ± 5,58 (132,48)	$p^{(2)} = 0,137$

In Table 3, it is possible to notice that most of the patients showed presence of sinus with 93.3% on each side; presence of right (73.3%) and left (60.0%) intersinus septa; one or more scalloping on each side, with 46.7% to 60.2% of patients with two or more scallopings and 13.3% to 16.7% of patients with one scalloping.

It is possible to calculate that the highest percent differences appear in patients with two or more right scallopings, with higher values for males (73.3% x 46.7%) and one left scalloping with higher values for females (26.7% x 6.7%); however, no significant association was found between sex and the variables analyzed on the table ($p > 0.05$).

Table 3 – Presence of right, left and central sinuses, and of right and left intersinus septa according to sex.

Variable	Gender						Value of p
	Male		Female		Total group		
	n	%	n	%	N	%	
TOTAL	**15**	**100,0**	**15**	**100,0**	**30**	**100,0**	
• **Pesence of right sinus**							
Yes	14	93,3	14	93,3	28	93,3	$p^{(1)} = 1,000$
No	1	6,7	1	6,7	2	6,7	
• **Presence of left sinus**							
Yes	15	100,0	13	86,7	28	93,3	$p^{(1)} = 0,483$
No	-	-	2	13,3	2	6,7	
• **Presence of central sinus**							
Yes	14	93,3	14	93,3	28	93,3	$p^{(1)} = 1,000$
No	1	6,7	1	6,7	2	6,7	
• **Presence of right intersinus septa**							
Yes	12	80,0	10	66,7	22	73,3	$p^{(1)} = 0,682$
No	3	20,0	5	33,3	8	26,7	
• **Presence of left intersinus septa**							
Yes	9	60,0	9	60,0	18	60,0	$p^{(1)} = 1,000$
No	6	40,0	6	40,0	12	40,0	
• **N° of right scallopings**							
No	3	20,0	5	33,3	8	26,7	$p^{(1)} = 0,380$
Um	1	6,7	3	20,0	4	13,3	
Dois ou mais	11	73,3	7	46,7	18	60,0	
• **N° of left scalloping**							
No	6	40,0	5	33,3	11	36,7	$p^{(1)} = 0,489$
one	1	6,7	4	26,7	5	16,7	
Two or more	8	53,3	6	40,0	14	46,7	

Discussion

The computerized tomography allows better visualization of the anatomical structures and tridimensional images, which are not possible with conventional radiographs, offering another possibility for identification when conventional ways are limited. This technique, combined with tomographies, is not common in the literature, and this due to the recent inclusion of this exam in the routine of health services and diagnosis. Also, softwares that allow the visualization and measurement in various planes of the tomography have only recently been available.

Conventional radiographs are challenged due to the use of simple linear measurements, such as height and width of the frontal sinus. In addition to this,

Studies using computerized tomographies with the availability of information in three dimensions seeks to eliminate the limitations of radiographs using a digital sinusal trace for a qualitative improvement in information to increase the technique reliability and confirmation of the frontal sinus uniqueness[8].

Populations with a lesser degree of anatomic differentiation, such as European and Asian, have already been studied about the frontal sinus morphology aiming at individual identification. Concerning the Brazilian population, characterized by a high degree of variation due to racial miscegenation, there are still few studies on the frontal sinus morphology. The results showed here agree with those by Camargo Jr et al.[9] that also showed a significant difference in morphology for sex identification in a group of caucasian subjects.

Studies seek differences in the frontal sinuses based on parameters as sex and age, which are the most studied and most common association. The search for new methods and variables is essential for the corroboration of the method for identification.

For this group, composed of 50% males and 50% females, the measures posterior cortical, ANV (frontal inclination), and BNC (nasofrontal angle) were significantly higher in females, and the other measures were higher in males, except for total sinus width, mean left sinus and anterior cortical that were not significantly different. Thus, the variables studied here showed great importance and accuracy for sex estimation. This finding corroborates a previous study with an Indian population and conventional radiographs[10].

Distance between anterior cortical and posterior cortical is equivalent to the sinus anteroposterior distance and is well visualized in sagittal views. This measurement varies according to the individual height, with a significant Pearson correlation ($p=0.001$). The results obtained here agree with those where bidimensional radiographs of Indian subjects were analyzed, confirming that the anterior and posterior bone walls delimit the frontal sinus, the thickness between them are not commonly variable, and are presented as two thin cortical layers that are part of the frontal bone structure, unique to each individual[11].

The angles measured correspond to the forehead inclination that, in turn, is determined by the glabella curvature, given by ANV. The results were significant, with a range of 6.08 to 10.07 in mean, the only statistically significant variable. The measurement of the effective forehead declivity and its inclination, as well as BNC,

which measures nasal and forehead inclination, present no significant variation, suggesting that the cranial profile variation is not relevant for sex identification.

Identification methods using frontal sinus imaging is of paramount importance since it allows the use of measurements with a significant variation for individual identification, as shown in the studies. Also, the use of more modern technologies, such as computerized tomography, can increase accuracy and improve the available techniques for human identification through the frontal sinuses measurements.

This work faced some challenges related to the high incidence of pathologies and fractures associated with the frontal sinus since our sample was composed of patients using the oncology and maxillofacial diagnosis services. These conditions could compromise the analysis of the anatomical morphology, and many potential subjects were, thus, excluded from the sample. Another obstacle was the scarcity of studies using computerized tomographies for the identification with the frontal sinuses. Most of the studies use only conventional radiographs.

Conclusion

It is possible to conclude that the frontal sinus may be helpful in human identification once there were significant differences in the frontal sinus measures on men and women and individuals with greater height.

Conflict of interest

There are no known conflicts of interest associated with this publication, and there has been no significant financial support for this work that could have influenced its outcome.

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Original article

Forensic Anthropology

The *arcus senilis* as a parameter for age estimation

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ABSTRACT

This study aimed to evaluate the presence of *Arcus senilis* in adults, aged 40 years old or more, of both sexes. This cross-sectional study encompassed 294 Brazilian participants. Direct observation of the eyes was performed for evaluating the presence or absence of the Arcus Senilis. Data collection on sex, age, the auto-declared color of the skin, and the presence or not of the *Arcus senilis*, as well as the history of

hypercholesterolemia, was conducted. The presence of the Arcus Senilis was identified in 20% (n = 59) of the total sample. It was observed that this phenomenon occurred more in men than in women (62.7 and 37.3%, respectively), and more often among participants of 60 years old or more, with the brown color of the skin and brown eyes. Thirty-eight participants reported having been submitted to blood screening in the past six months. Of these, 14 presented higher levels of LDL cholesterol. Among the patients with corneal arch, 32 (54.24%) had reports of high cholesterol levels in the family history. The accuracy of the presence of the Arcus Senilis as an age estimation indicator was 68%. Although present in older people, the Arcus Senilis itself should not be used alone as a parameter for the age estimation for this population, once it doesn't provide a satisfactory accuracy.

***Keywords*:** Arcus Senilis; Age Estimation; Forensic Anthropology.

Introduction

The medico-legal identification uses somatoscopic and somatometric examinations to establish information such as ancestry, gender, and age[1, 2]. Obtaining the chronological age of individuals is a matter of great social interest, in terms of criminal matters, in the identification of skeletonized corpses or skulls, for example, as well as in the civil law, in adoption processes and cases of proof of majority[3].

Individual aging can be distinguished in chronological aging, which results exclusively from age, and biopsychological aging that is a reflection of chronological aging but is not restricted to the years lived[4]. In estimating age, many elements are analyzed, such as appearance, skin, hairs, and eyes[1].

The arcus senilis or arcus corneae consists of a whitish ring at the perimeter of the cornea, being common in normal aging, with no pathological significance5. It represents the most common peripheral opacity of the cornea and is not associated with tissue decomposition but with lipid deposition[6]. It may also be an indicator of systemic alterations, such as familial hypercholesterolemia, characterized by elevated levels of plasma cholesterol, resulting in cholesterol deposits in the extravascular tissues[7].

It's more prevalent in men than in women, increasing with advancing age. In addition to being associated with hypercholesterolemia, it's been also demonstrating

the relation of the arcus seniles with xanthelasma, alcohol abuse, increased blood pressure, cigarette smoking, diabetes, age, and coronary disease[6].

Because it is mentioned in the forensic literature as one of the elements for age estimation1, arcus senilis should be better investigated, to find the answer whether or not it has value for the age estimation mainly for adults and elderly individuals. In other words, it is desired to know the real utility of the evaluation of *arcus cornae* for expert practice.

However, it has been observed that there is a lack of scientific publications regarding the investigation of the presence of the senile arch and its validity as auxiliary information for the examinations that aim at the estimation of age.

Thus, to find an answer to this question, the present study proposed to evaluate the presence of the *arcus senilis* in people over 40 years old, verifying the reliability of its use as an auxiliary indicator for the estimation of age.

Materials and Methods

The research was approved by the Ethics Committee on Research of the University of Pernambuco (UPE), under registration number 1.050.112. The study was carried out in the city of Camaragibe, located in the Metropolitan Region of Recife, State of Pernambuco, Northeast of Brazil. According to data from the last census, the resident population of the municipality aged 40 years or more corresponds to 46,680 people (21,121 men and 25,559 women).

The population studied consisted of individuals of 40 years and over, of both sexes, with self-declared skin color (white, black, yellow, brown, or indigenous), according to the standards of the Brazilian Institute of Geography and Statistics (IBGE).

The minimum acceptable sample size was 245 participants. A correction factor of 1.2 was applied to avoid a smaller sample than required (minimum participation rate = 80%). Thus, a sample of 294 people was obtained to reach the desired precision. The sample calculation was performed using the online OpenEpi software.

Due to the differences between the number of components from one age group to another, a coefficient of proportionality was used, so that the number of individuals selected per range was proportional to the percentage represented by this range in relation to the universe of residents in the municipality, which fit the criteria of the study.

Persons less than 40 years of age who had cognitive problems, which impaired their understanding and free consent for participation in the research, as well as individuals with a history of previous surgeries of eyes, corneal transplants, or ophthalmological pathologies were excluded from the study. The approach to the participants was carried out personally by the researcher, between passers-by on public roads with a large circulation of people, to ensure that all age groups were taken into account.

Results

All the 294 participants were examined. One hundred and forty-eight individuals were women and 146 men, observing to the maximum the amount established for each age group. Regarding the self-reported skin color, 163 participants (55.4%) reported having a brown skin color, 90 (30.6%), white, and 32 (10.9%) black. Only 7 (2.4%) reported as having yellow skin color, and 2 (0.7%) were classified as indigenous (red skin). There was no statistically significant difference between skin color and the presence of arcus senilis (p = 0.271).

When asked if blood tests were performed in the last six months, and if cholesterol levels were requested, 145 (49%) participants said they did not have any tests. Among the individuals who did the blood check, 62 reported having had a change in cholesterol level. Of the total number of participants, 162 reported having a history of familial hypercholesterolemia.

The *arcus senilis* was observed in 59 participants of the study. Males presented more *arcus senilis* in the sample studied. When the Chi-square test was applied, it was verified that there were statistically significant differences between these two variables (p = 0.025).

Table 1. Presence of the arcus senilis according to sex.

Arcus senilis	**Sex**		**TOTAL**
	Males	**Females**	
	N (%)	**N (%)**	**N (%)**
Absent	109 (46.4)	126 (53.6)	235 (79.9)
Present	37 (62.7)*	22 (37.3)	59 (20.1)
TOTAL	146 (49.7)	148 (50.3)	294 (100.0)

* Chi-square test. *p* value: >0.05.

When the *arcus senilis* was analyzed in relation to the age group, a higher concentration of cases was observed among people over 60 years old (Table 2).

Statistical analysis was performed, and it was found that there was a statistically significant association between these two variables (p = 0.000).

Table 2 - Presence of the *arcus senilis* according to the participants' age group.

CATEGORIZED AGE	ARCUS SENILIS		TOTAL N(%)
	Absent N (%)	Present N(%)	
From 40 to 49 years	120(96.0)	5(4.0)	125(42.5)
From 50 to 59 years	69(84.1)	13(15.9)	82(27.9)
From 60 to 69 years	29(58.0)	21(42.0)	82(17.0)
From 70 years	17(45.9)	20(54.1)	37(12.6)
TOTAL	**235(79.9)**	**59(20.1)**	**294(100.0)**

We re-categorized the age variable into people "aged 60 or more years" and performed a linear regression analysis, using the backward model. It was observed that age was a risk factor, i.e., people aged 60 or more years was 8.9 times (OR) more likely to present an arcus senilis (Table 3).

Table 3 – Linear regression analysis for the "Age" variable.

Model	Variable	Non-Standardized Coefficients		Standardized Coefficients	T	Sig.	95%CI to an Exp (B)	
		B	S.E.	Beta			Minimum	Maximum
1	(Constant)	0.853	0.045		19.007	0.000	0.765	0.942
	Categ_Age_60_or_more_years	0.174	0.020	0.455	8.740	0.000	0.135	0.213
2	(Constant)	0.941	0.063		14.904	0.000	0.817	1.066
	Categ_Age_60_or_more_years	0.176	0.020	0.460	8.856	0.000	0.137	0.215

In order to verify the accuracy of the arcus senilis for the age estimation, the value of 0.200 described in the Brazilian literature was considered as a prior probability. In the present study, the true positives presented a value of 0.201, while the genuinely negative had a value of 0.799. After the analysis, an accuracy equal to 68% was obtained.

Discussion

It is essential to highlight that the literature on the observation of the *arcus senilis* for forensic purposes of age estimation is very scarce. In Brazil, we only found an explicit citation of this parameter in França (2015)[1]. For this reason, we decided to conduct the present study to investigate whether there is a viable relationship between the presence of the *arcus* and the age of individuals. The importance of this analysis lies in the fact that there are few age estimation methods for adults and older people when compared to subadult individuals, and it is well known that the longer individuals grow older, the less anthropological information will be available to be described.

In the present study, the prevalence of arcus corneae in the sample was 20,1%, and 70% of the cases in which arcus senilis was observed were of individuals aged 60 years or older. This same result was also observed by Raj et al. in 2015[8]. The presence of the *arcus corneae* increases with age. In our study, older people (60 or more years) had 8.9 times (OR) more chance of having *arcus senilis*.

Regarding the sex, in 2014 Hashemi et al.[9] found that the arcus senilis was more prevalent in men than in women, and it increased according to the progression of age, in the same way that occurred in the present study. The results of the survey of Hashemi et al.[9] showed that men were 2.2 times more prone to have *arcus senilis* than women.

Some other factors, such as smoking, systolic hypertension, and high level of cholesterol, are pointed out as being conditions that play a role in the presence of the *arcus corneae*. Chen et al.[10] showed that people who had higher levels of total cholesterol, low-density lipoprotein (LDL-C) cholesterol, non-high-density lipoprotein cholesterol, and total cholesterol / HDL ratio had increased chances of presenting *arcus senilis*.

It is important to note that in our study there was no blood collection from the participants to verify if there was an individual and/or family history of hypercholesterolemia. This information was asked directly to the participants at the time of data collection. However, the authors would like to inform that even knowing that the data collected through an interview may present some information bias, it was not possible to perform blood tests in the participants at the moment of the interview. A randomized clinical trial is being planned for further investigation.

Regarding the accuracy of the observation of the *arcus senilis* for the age estimation, it was verified that the percentage obtained was not statistically satisfactory or sufficient for this parameter to be used as a forensic parameter for the age estimation in adults and older people. Therefore, this observation should be considered in conjunction with others that may be used when estimating age.

There was great difficulty in finding studies that had related the *arcus senilis* with age for forensic purposes, both in Brazil and in other countries. Thus, there were difficulties in comparing the information of this study with those of other studies, which indicates the need for more research to be carried out to investigate the *arcus senilis* as a parameter for the age estimation in forensic practice.

Conclusion

It was concluded that the *arcus senilis* was found in 20.1% of the total sample studied. The most affected age groups were 60 to 69 years and 70 years or more. Among the studied variables, only age was associated with the occurrence of the arcus senilis. The accuracy obtained for the method was considered not statistically satisfactory for forensic purposes.

Conflict of interest

There are no known conflicts of interest associated with this publication, and there has been no significant financial support for this work that could have influenced its outcome.

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Literature review

Legal Medicine

Tortura, prova pericial e quesitos

Torture, expert evidence, and questions

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RESUMO

Este trabalho tem como objetivo discutir aspectos éticos e profissionais fundamentais ao médico legista para realizar um trabalho consistente na perícia de casos de tortura alegada e propor um quesito compatível à sua atribuição. Trata-se de revisão da literatura utilizando os descritores DeCs (tortura, prova pericial, medicina legal) e os MeSH terms (torture, expert testimony, forensic medicine) com consultas realizadas

nas bases de dados bibliográficas: Biblioteca Virtual em Saúde, Medline, SciELO e Lilacs, Periódicos CAPES e PubMed. Também utilizamos como referencial teórico a legislação aplicada, os tratados ratificados pelo Brasil, o Protocolo de Istambul e o Protocolo Brasileiro de Perícia Forense no Crime de Tortura. Os protocolos a respeito do assunto trazem recomendações importantes para avaliação física e psíquica em perícias para avaliação de tortura, sobretudo no que diz respeito ao nexo de causalidade. A tipificação do crime de tortura é uma tarefa que compete a autoridades judiciais, mediante o processo *in totum*. É relevante discutir a perícia médico legal em casos de tortura e fomentar a pesquisa sobre esse assunto. Propõe-se uma mudança dos quesitos atualmente vigentes, com ênfase na discussão dos principais achados periciais. A discussão permitirá que o perito, diante das informações presentes no exame físico, documentos, histórico e alegação, com fundamento na literatura, possa analisar o grau de consistência entre o exame clínico e o relato do periciado.

Descritores: Tortura; Prova Pericial; Medicina Legal.**ABSTRACT**

ABSTRACT

This research aimed to discuss ethical and professional main aspects in order to perform consistent forensic medical work in the investigation of alleged cases of torture and propose a compatible legal questioning. Descriptors DeCs (torture, expert evidence, legal medicine) and MeSH terms (torture, expert testimony, forensic medicine) in the bibliographic databases: Virtual Health Library, Medline, SciELO and Lilacs, CAPES journals and PubMed were used. We also used as theoretical reference the applied legislation, the ratified treaties by Brazil, the Istanbul Protocol, and the Brazilian Protocol of Forensic Expertise in the Crime of Torture. The protocols on the subject bring valuable recommendations for physical and psychic evaluation, especially concerning the causal link. The typification of the crime of torture is a task that is incumbent upon judicial authorities through the *in totum* process. It is relevant to discuss medico-legal expertise in cases of torture and to promote research on this subject. We propose a change of the current legal questioning, with emphasis on the discussion of the main forensic findings. The discussion will allow the expert to analyze the degree of consistency between the clinical and psychic examination and the claim of the victim.

***Keywords*:** Torture; Expert Testimony; Forensic Medicine.

Introdução

A definição mais aceita mundialmente para atos de tortura e outros tratamentos ou penas cruéis, desumanos e degradantes, é a contemplada pelo Artigo 1º da Convenção contra a Tortura, adotada pela Assembleia Geral das Nações Unidas em 10 de dezembro de 1984: “Para fins da presente Convenção, o termo ‘tortura’ designa qualquer ato pelo qual dores ou sofrimentos agudos, físicos ou mentais, são infligidos intencionalmente a uma pessoa a fim de obter, dela ou de terceira pessoa, informações ou confissões; de castigá-la por ato que ela ou terceira pessoa tenha cometido ou seja suspeita de ter cometido; de intimidar ou coagir esta pessoa ou outras pessoas; ou por qualquer motivo baseado em discriminação de qualquer natureza; quando tais dores ou sofrimentos são infligidos por funcionário público ou por outra pessoa no exercício de funções públicas, ou por sua instigação, ou com o seu consentimento ou aquiescência. Não se considerará como tortura as dores ou sofrimentos que sejam consequência unicamente de sanções legítimas, ou que sejam inerentes a tais sanções ou delas decorram”[1].

Nos tratados e nas convenções internacionais o crime de tortura é um crime próprio, ou seja, decorrente de ato de determinadas pessoas especificadas, praticado pelo funcionário público, na condição de representante do Estado.

A lei brasileira que tipifica o crime de tortura traz um conceito amplo e caráter bifronte. Ou seja, a lei 9.455/97 alcança também os atos perpetrados por particulares e movidos por outros. Quando é praticado por agentes públicos, considera-se um agravante[2]. A Lei do Crime de Tortura no Brasil define que:

> “Constitui crime de tortura: constranger alguém com emprego de violência ou grave ameaça, causando-lhe sofrimento físico ou mental: com o fim de obter informação, declaração ou confissão da vítima ou de terceira pessoa; para provocar ação ou omissão de natureza criminosa; em razão de discriminação racial ou religiosa; submeter alguém, sob sua guarda, poder ou autoridade, com emprego de violência ou grave ameaça, a intenso sofrimento físico ou mental, como forma de aplicar castigo pessoal ou medida de caráter preventivo. (...) § 1º Na mesma pena incorre quem submete pessoa presa ou sujeita a medida de segurança a sofrimento físico ou mental, por intermédio da prática de ato não previsto em lei ou não resultante de medida legal. § 2º Aquele que se omite em face dessas condutas, quando tinha o dever de evitá-las ou apurá-las, incorre na pena de detenção de um a quatro anos. § 3º Se resulta lesão corporal de natureza grave ou gravíssima, a

> pena é de reclusão de quatro a dez anos; se resulta morte, a reclusão é de oito a dezesseis anos. § 4º Aumenta-se a pena de um sexto até um terço: se o crime é cometido por agente público; se o crime é cometido contra criança, gestante, portador de deficiência, adolescente ou maior de 60 (sessenta) anos; (Redação dada pela Lei nº 10.741, de 2003[4]); se o crime é cometido mediante sequestro. § 5º A condenação acarretará a perda do cargo, função ou emprego público e a interdição para seu exercício pelo dobro do prazo da pena aplicada. § 6º O crime de tortura é inafiançável e insuscetível de graça ou anistia. § 7º O condenado por crime previsto nesta Lei, salvo a hipótese do § 2º, iniciará o cumprimento da pena em regime fechado. Art. 2º O disposto nesta Lei aplica-se ainda quando o crime não tenha sido cometido em território nacional, sendo a vítima brasileira ou encontrando-se o agente em local sob jurisdição brasileira[2]."

Considerado pelos tratados internacionais, como o Estatuto de Roma, norma supralegal, um crime imprescritível, a tortura não está listada como crime imprescritível pela Constituição Federal Brasileira e nem na lei 9.455/97. O parecer do STF sobre proposta de revisão da OAB (ADPF nº 153) à Lei da Anistia, reitera anistia ampla, geral e irrestrita, refutando revisão por maioria[3,4].

No Brasil, sítio da terceira mais numerosa população carcerária do mundo[5], de imigração em massa de venezuelanos em penúria[6], de grandes desigualdades socioeconômicas, da política antidrogas, da violência associada ao narcotráfico[7] e de 65.602 assassinatos anuais[8], novos desafios à preservação dos direitos humanos se interpõem diante de nós cotidianamente.

Hoje, a tortura continua presente no mundo, seja no emprego de intenso sofrimento psíquico, físico ou mesmo sob alguma forma de omissão. A materialização desse crime, composta inclusive pela perícia médico legal, é fundamental para coibi-lo e preveni-lo[9].

Revisão da Literatura

I- Exame Médico Legal

O Código de Processo Penal obriga a realização do exame de corpo de delito quando há vestígios de violência[10]. A perícia é a modalidade de prova que se destina

a atestar os vestígios físicos e psíquicos deixados pelo delito, ou, em outras palavras, compor a materialidade.

Os sinais relativos ao crime de tortura são bastante desafiadores pelas próprias práticas e pela possibilidade de lapso temporal entre a alegada tortura e o exame médico legal. De acordo com o Protocolo de Istambul, cabe ao médico legista avaliar se há nexo de causalidade entre o relato da vítima e os sinais clínicos médico-legais. Raramente são executados, apesar da grande relevância, exames no local e dos objetos utilizados na prática de tortura, deixando a avaliação médico legal ainda mais desafiadora[11].

A materialidade da tortura pode ser sutil e difícil de ser evidenciada. Exames especializados que, por exemplo, possam determinar sequelas psíquicas, exigem pessoal capacitado e recursos materiais adequados, muitas vezes não disponíveis nos serviços médico-legais. Dessa forma, a atenção é dada, prioritariamente, às lesões físicas externas e visíveis. Dois dos mais importantes componentes do corpo do laudo pericial, que são a discussão e a conclusão, não devem ser subutilizados. Por esse motivo, é fundamental investir em formação, qualificação e capacitação de médicos legistas e peritos criminais para que realizem perícias, em casos de tortura alegada, cumprindo protocolos e orientações norteadores[12, 13].

O Protocolo de Istambul, denominado “Manual para Investigação e Documentação Eficazes da Tortura e de outras Formas Cruéis, Desumanas ou Degradantes de Castigo ou Punição”, apresentado ao Alto Comissariado das Nações Unidas para os Direitos Humanos, em 9 de agosto de 1999, é um documento de referência e deve nortear a conduta pericial[13]. O treinamento em Protocolo de Istambul também deve ser contemplado nos curso de formação de peritos oficiais. Apesar de grande importância, foge do escopo deste trabalho tratar da perícia necroscópica. Na perícia tanatológica em casos de tortura duas referências importantes são o Protocolo de Minnesota e o Protocolo Brasileiro de Perícia Forense no Crime de Tortura[14].

Os exames periciais em casos de tortura devem, preferencialmente, ser realizados por uma equipe especializada nesse tipo de exame. A avaliação pericial nos casos de suspeita de crime de tortura, assim como em qualquer outra perícia, deve ser realizada de forma objetiva e imparcial, com base em fundamentos médico-legais[11].

O laudo pericial deve contemplar, minimamente, os seguintes aspectos: relato, história pregressa, patologias pregressas, exame pericial, documentação fotográfica

(quando aplicável), discussão e conclusões[9]. As vítimas de tortura podem se apresentar psicologicamente afetadas, em face das situações a que foram submetidas. O acolhimento e a confiança são requisitos para que o periciado consiga relatar a sua vivência.

A confiança entre o perito e o periciado deve ser estabelecida de forma clara, sendo indicado colher o consentimento por escrito sobre o que o periciado autoriza que seja colocado no laudo, ao final da perícia[11]. Os *experts* devem manter sigilo das confidências relatadas e somente divulgá-las com a devida autorização formal.

O exame deve acontecer em um ambiente adequado, sem a presença de outros indivíduos, especialmente de pessoas que possam ser responsáveis, direta ou indiretamente, pela violação alegada[11]. É permitido ao periciado a escolha de quem o examinará, por exemplo: um médico legista do sexo masculino ou feminino. Se a vítima for estrangeira, também há o direito de se escolher livremente um intérprete para acompanhá-la e orientá-la durante o exame pericial[11].

É essencial que todos os profissionais envolvidos sejam imparciais e obtenham um treinamento para realizar o exame clínico pericial adequadamente[15].

Algumas lesões físicas não deixam lesões macroscopicamente visíveis ou sequelas aparentes. Diversas modalidades de suspensão, com duração de quinze minutos a várias horas, podem causar lesões definitivas ao plexo braquial. Para além da fase aguda pode haver parestesia e dores ao levantar os braços[13]. Outros padrões de tortura física frequentes são: falanga (espancamento dos pés), contusões diversas, golpes nas orelhas (telefone), violação sexual, empalamento, choques elétricos, tortura dentária, mutilações, ingestão de insetos, arrancamento das unhas, asfixia, suspensão em barra de metal (pau-de-arara) e outras formas de tortura posicional[11,16]. Apesar da exemplificação clássica de algumas variedades de tortura corporal, cabe destacar que as agressões podem ser físicas a partir da violência explícita, podem ser morais com a realização de hostilização, intimidações e ameaças, podem ser sexuais e podem ser omissivas com a privação de boas condições ambientais, negligência alimentar e desmazelo com a higiene[15].

Essa perícia demanda a coleta detalhada do que se passou durante o ato de tortura e a expertise para avaliar a consistência quanto ao nexo de causalidade. Quando disponível, a ressonância magnética é o método radiológico mais indicado para o diagnóstico das lesões associadas a todas as formas de tortura posicional[11].

De acordo com Goulart, muitas vezes os ferimentos e lesões desaparecem até a data da realização da perícia e ensejam a realização de exame indireto de corpo de

delito. Tantas outras vezes, atos violentos, apesar de caracterizarem tortura, são desferidos, propositadamente, de maneira a não deixarem vestígios ou deixam apenas leves escoriações na pele da vítima[17]. Nesse contexto, o histórico deve ser o mais completo e detalhado possível, incluindo informações sobre uso de medicamentos, doenças e traumas pregressos, tratamentos médicos, cirurgias, aspectos psíquicos e padrão de sono. Deve-se colher um relato, minucioso, sobre como a tortura ocorreu, por quanto tempo e de que forma[15].

As manifestações psicológicas podem compor evidências determinantes e significativas de que uma pessoa foi torturada. É crucial ter em mente que a ausência de alterações psicossomáticas não é sinônimo de que não existiu tortura. A violência psíquica, não deixa sequela aparente e sua apresentação varia de indivíduo para indivíduo[11,15,16].

As alterações passíveis de serem encontradas dividem-se em desordens afetivas (fobias, depressão), desordens comportamentais (irritabilidade, isolamento, impulsividade), desordens psicossomáticas (cefaléia, pesadelos, insônia, tremores) ou sintomas intelectuais (desorientação, perda de memória)[15].

O exame de um caso de tortura deve buscar o conjunto e natureza dos achados clínicos e psicopatológicos. A composição do corpo de delito deverá integrar equipe multidisciplinar com profissionais especializados em Psicopatologia Forense, Medicina Legal em casos de tortura e nas diversas áreas da Criminalística (odontologia, balística, toxicologia, exame de local, DNA e outros)[18].

Quanto à conclusão do exame médico legal, o Protocolo de Istambul propõe múltiplas respostas que podem auxiliar a tomada de decisão quanto ao nexo de causalidade[13], fornecendo ao perito alguns termos específicos indicando uma graduação. O termo “inconsistente” significa que o sinal/sintoma não pode ser causado pelo trauma alegado. O termo “consistente” indica que o sinal/sintoma pode ter sido causado pelo trauma alegado, entretanto há outras causas mais prováveis. O termo “altamente consistente” indica que o sinal/sintoma pode ter sido causado pelo trauma alegado, havendo poucas outras causas mais prováveis. O termo “típico de” significa que o sinal/sintoma é geralmente associado ao trauma alegado, mas há outras causas possíveis. O termo “diagnóstico de” indica que o sinal/sintoma não poderia ter sido causado em nenhuma outra circunstância, que não a relatada[11].

A literatura propõe recomendações específicas para os peritos forenses com a finalidade de realização de exame de lesão corporal e manifestações psicológicas em casos de tortura. Dentre elas estão: descrição detalhada do exame, ilustração por

meio de esquemas corporais e fotografias, exame radiológico completo das regiões afetadas e exame cuidadoso das vestes e evidências encontradas junto delas[18].

II- Quesitos

O artigo 160 do Código de Processo Penal reza que “Os peritos elaborarão o laudo pericial, onde descreverão minuciosamente o que examinarem, e responderão aos quesitos formulados”. Dessa forma a quesitação oficial e a quesitação complementar devem ser cuidadosamente elaboradas pelos doutos julgadores, uma vez que se trata de atribuição do médico legista responder aos mesmos[19].

Os quesitos oficiais são vinculados aos tipos penais previstos no Código de Penal Brasileiro[19]. De acordo com os Procedimentos Operacionais Padrão (POP) da Secretaria de Segurança Pública do Ministério da Justiça (SENASP/MJ) de 2013 a maior parte dos estados utiliza a seguinte estrutura para os exames de corpo de delito envolvendo pessoas vivas[20]:

- “1º Houve ofensa à integridade corporal ou à saúde do (a) periciando(a)?
- 2º Qual instrumento ou meio que a produziu?
- 3º A ofensa foi produzida com o emprego de veneno, fogo, explosivo, tortura ou outro meio insidioso ou cruel?
- 4º Resultou perigo de vida?
- 5º Resultou incapacidade para as ocupações habituais por mais de trinta (30) dias?
- 6º Resultou debilidade permanente de membro, sentido ou função, ou aceleração do parto? (Resposta especificada).
- 7º Resultou incapacidade permanente para o trabalho, ou enfermidade incurável, ou perda ou inutilização de membro, sentido ou função, ou deformidade permanente, ou aborto? (Resposta especificada)”[20].

O Protocolo Brasileiro de Perícia Forense no Crime de Tortura sugere a adoção de quatro quesitos diferentes sobre a tortura[18]:

1º) Há achados médico-legais que caracterizem a prática de tortura física?

2º) Há indícios clínicos que caracterizem a prática de tortura psíquica?

3º) Há achados médico-legais que caracterizem a execução sumária?

4º) Há evidências médico-legais que sejam característicos, indicadores ou sugestivos de ocorrência de tortura contra o(a) examinando(a) que, no entanto, poderiam excepcionalmente ser produzidos por outra causa? Explicitar a resposta.

Estas sugestões foram incorporadas pelo Conselho Nacional de Justiça (CNJ) na Recomendação Nº 49/2014, que também faz referência explicitamente ao Protocolo de Istambul, assim como pelo Conselho Nacional do Ministério Público (CNMP) na Recomendação nº 31/2016. Estas recomendações indicam que os magistrados, diante de notícias concretas e fundamentadas da prática de tortura, sempre devem solicitar exames de corpo de delito com os mesmos quesitos previstos no Protocolo Brasileiro[11,18,22,23].

Os quesitos referentes aos crimes de maus tratos e tortura, esses citados acima, arrolam a tipificação de um delito ao médico legista, o que ultrapassa sua atribuição[21].

Discussão

A tipificação do crime de tortura, cabe ao douto julgador. Ao médico-legista cabe avaliar o grau de consistência entre a alegada tortura e as lesões e sequelas apuradas durante seu ato pericial.

O 3º quesito que questiona se “A ofensa foi produzida com o emprego de veneno, fogo, explosivo, tortura ou outro meio insidioso ou cruel?” atribui aos médicos-legistas uma função exclusiva do Poder Judiciário: a de determinar a tipificação de crimes.

Ao pedir que profissionais da Medicina respondam se uma lesão “foi produzida com emprego de tortura”, é o mesmo que perguntar se havia:

- autoridade, guarda, ou vigilância sobre a vítima;
- finalidade de obter informação, declaração ou confissão da vítima ou de terceira pessoa;
- grande sofrimento físico ou mental;
- discriminação racial ou religiosa;
- finalidade de castigo pessoal ou medida de caráter preventivo.

A tipificação de um delito depende de estudos que fogem à capacidade médica do perito, extrapolando sua competência. Alguns elementos não são passíveis de

comprovação somente num exame médico, como, por exemplo qual a intencionalidade e a finalidade do agressor.

Entendemos que os quesitos existentes extrapolam as atribuições dos médicos-legistas, uma vez que as perguntas apresentam elevado grau de taxatividade para as respostas. Perguntar se “há achados médico-legais que caracterizem a tortura” demanda uma avaliação de cunho categórico sobre uma relação de causa e efeito cuja resposta se considera inadequada para avaliações médico-legais.

Exigir dos médicos legistas a tipificação do crime de tortura não é razoável. A tipificação é uma tarefa que compete a autoridades judiciais, mediante investigações imparciais e garantias de devido processo.

Mais importante do qualquer tipo de quesito é mister que os laudos médico-legais sigam os parâmetros de análise de consistência previstos no Protocolo de Istambul, com especial atenção à indicação ao grau da consistência entre os relatos das vítimas e os achados clínicos (inconsistente, consistente, altamente consistente, típico ou diagnóstico).

Por fim, em casos de suspeita de tortura, sugerimos aos magistrados, a propositura de uma quesitação, com resposta especificada com base no Protocolo de Istambul: **qual o grau de consistência entre os achados periciais e o relato do periciado?**

A resposta a esse quesito permitirá que o perito - diante do histórico, das informações extraídas do exame forense, dos exames complementares, dos documentos, da discussão e da literatura - possa chegar a uma conclusão sobre o grau de consistência entre os achados médico legais em relação ao relato do periciado.

Conclusão

A tortura está presente no mundo, assim como no Brasil. A vítima deve ser acolhida, ter sua privacidade respeitada e seu consentimento deve ser colhido. O exame de corpo de delito multidisciplinar e especializado é indicado para composição da prova material desse tipo de crime. É atribuição do médico forense avaliar o grau de consistência entre os sinais e sintomas e o histórico, com fundamento no Protocolo de Istambul. Diante disso, uma alternativa aos quesitos vigentes seria: **qual o grau de consistência entre os achados periciais e o relato do periciado?**

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Conflito de Interesse

Não há conflitos de interesse conhecidos associados a esta publicação e não houve apoio financeiro significativo para este trabalho que pudesse ter influenciado seu resultado.

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Literature review

Forensic Anthropology

Isótopos aplicados à Antropologia Forense

Isotopes applied for Forensic Anthropology

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RESUMO

Análises isotópicas vêm sendo cada vez empregadas com finalidades forenses para compreensão do deslocamento geográfico, de modo a refinar os critérios para identificação do indivíduo, se tornando cada vez mais eficazes, sobretudo com o

desenvolvimento de novas técnicas e a confecção de *isoscapes* (mapas preditivos). Para além disso, o emprego de radioisótopos tem sido desenvolvido como um bom critério para se avaliar tempo decorrido desde a morte. O objetivo do presente artigo é apresentar o potencial dos isótopos estáveis e instáveis para fins forenses, tanto dentro de estudos acadêmicos, como com exemplo do papel das análises isotópicas dentro de investigações criminais e, por fim, apontar o futuro dos isótopos estáveis forenses no Brasil.

Descritores: isótopos forenses; isoscapes; radioisótopos; Antropologia Forense.

ABSTRACT

Isotopic analyzes have being increasingly used for forensic purposes to understand geographic displacement, in order to refine criteria for estimating the biological profile, becoming effective, especially with the development of new techniques and confection of isoscopes (predictive maps). In addition, the use of radioisotopes has been developed as a good criterion for assessing the postmortem interval (PMI). The objective of this article is to present the potential of stable and unstable isotopes for forensic purposes within academic studies and their role within criminal investigations. Therefore, we also aim to point out the future of stable forensic isotopes analyses in Brazil.

***Keywords*:** Forensic isotopes; Isoscapes; Radioisotopes; Forensic Anthropology.

Introdução

Em 2000, um crânio humano foi localizado dentro de um plástico, em uma cova rasa, às margens do Great Salt Lake, nos EUA[1]. Além disso, a polícia averiguou a existência de mais doze ossos humanos, além de pertences pessoais, como uma meia branca, uma camiseta tamanho grande e um colar de trançado azul. Contudo, nem a análise da dentição nem os pertences localizados possibilitaram a identificação da vítima. Por falta de maiores informações, e conscientes de que a análise de DNA sem uma amostra comparativa em nada ajudaria na identificação da vítima, o caso foi arquivado em 2003.

Passados os anos, com o avanço de métodos e tecnologia científica, a polícia demandou a uma equipe do IsoForensics, Salt Lake City, que respondesse a partir de

dados isotópicos de oxigênio e hidrogênio informações sobre a vítima como seu local de origem, possíveis mudanças regionais e, se positivo este mapeamento, as localizações geográficas pelas quais ela teria passado.

Realizada as primeiras análises para fins forenses desse tipo pelo grupo em 2012, os resultados mostraram que a assinatura isotópica de Oxigênio-18 (18O) obtidas a partir do cabelo - em comparação com o mapeamento regional da água de torneira -, apontavam para uma mobilidade constante nos dois últimos anos de vida da vítima, por regiões consistentes com Salt Lake City e Intermountain West e, ainda, um terceiro lugar mais a noroeste (Fig. 1).

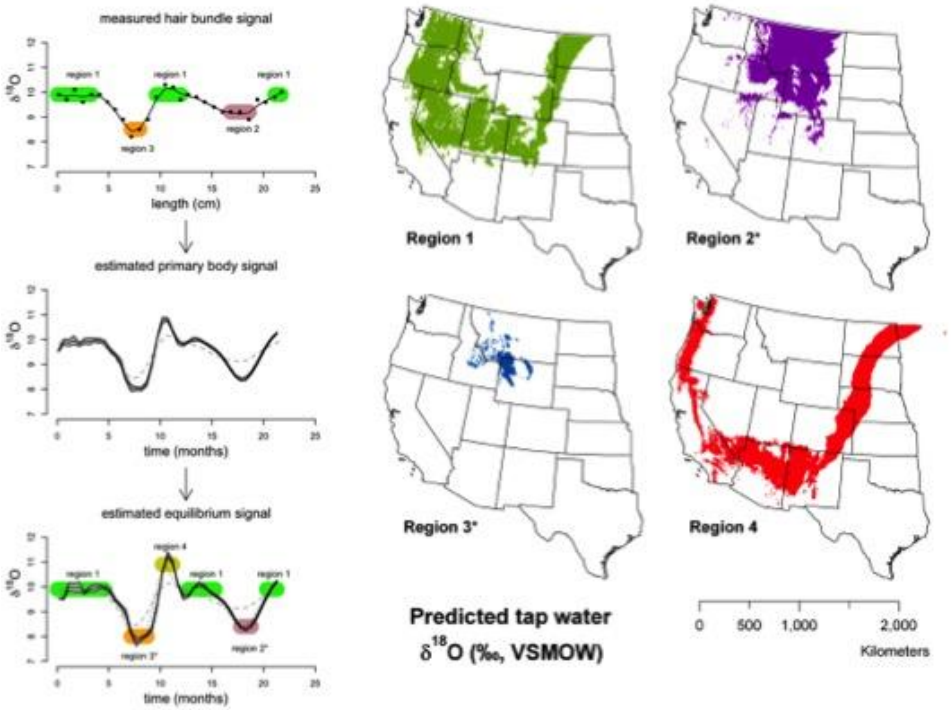

Fig. 1 - Mapeamento preditivo de água de torneira nos EUA. Fonte: Ehleringer et al., 2015[1].

Nesse ano, com os dados obtidos por meio do cruzamento dos valores isotópicos de 18O do cabelo e da água com dados provenientes do relatório de pessoas desaparecidas, foi possível para a polícia identificar, com base no padrão de deslocamento geográfico, uma possível vítima. Apoiado nessas informações, e tendo a partir desse momento um perfil genético em mãos para ser comparado, a análise fez sentido, e um teste de DNA foi realizado confirmando o perfil identificado pelos dados isotópicos.

Nikole Bakoles era uma jovem de 20 anos de idade que teria sido vista pela última vez em março de 2000, época em que residia em um hotel no subúrbio de Salt

Lake City com o namorado e uma criança. Nos anos anteriores, ela teria viajado para Seattle algumas vezes para visitar seus familiares, conforme a composição isotópica de oxigênio de seus cabelos já havia apontado.

Esse caso é um exemplo do potencial informativo dos isótopos estáveis quanto a proveniência e deslocamento geográfico de uma pessoa. Não obstante, análises dentárias e genéticas tenham potencial mais categóricos para a identificação de indivíduos, muitas vezes esses métodos não são suficientes para resolver casos de maior complexidade e a aplicabilidade de diferentes métodos nas Ciências Forenses vêm adicionando dados que permitem um cruzamento de informações que permitam melhor traçar o perfil do indivíduo investigado, possibilitando, por vezes, o uso de métodos com maior precisão identitária.

Entendendo os tecidos humanos

As investigações isotópicas, embora já comuns na área de arqueologia para compreensão de permanências e migrações populacionais e hábitos alimentares no passado, a partir de ossos e dentes, mais recentemente vem sendo empregadas na antropologia forense para obtenção de dados acerca das origens biológicas, química e geográfica que possam indicar lugar de origem e proveniência como dado auxiliar para identificação do indivíduo, porém, com aplicabilidade em uma gama mais ampla de tecidos humanos: osso, dente, cabelo e unha.

Ossos e dentes são compostos por frações orgânicas e inorgânicas, sendo respectivamente fibras de colágeno e hidroxiapatita (ou bio-apatita)[2,3].

É amplamente conhecido que o osso é constantemente remodelado ao longo da vida, porém, em ritmos e em taxas distintos, com tempos mais curtos de renovação óssea na juventude em comparação em idosos. Ademais, uma costela pode fornecer dados de dieta e proveniência geográfica dos últimos 5 a 10 anos de vida, enquanto um fêmur pode fornecer um registro dos últimos 20 a 25 anos de vida[3,4,5].

A hidroxiapatita (apatita) presente nos ossos e esmalte dentário é formada a partir do bicarbonato dissolvido no sangue. Esse componente biomineral aponta a assinatura isotópica de carboidratos, lipídios e proteínas não utilizados na síntese tecidual de colágeno ósseo (ou seja, a apatita reflete a dieta de maneira mais global)[6].

Os dentes também são analisados para obtenção dos isótopos estáveis, mas a partir do colágeno da dentina e, diferentemente do osso, os resultados isotópicos apresentados com base nos dentes indicam o período de sua da formação, a infância,

e não de outro período posterior, uma vez que os dentes não se remodelam ao longo da vida[7].

Por toda a influência que o organismo recebe em diferentes fases da vida por conta do consumo de alimentos disponíveis no ambiente, e também por escolhas culturais, os tecidos humanos não são apenas resultados do histórico genético, mas também da expressão cultural dos alimentos.

A queratina do cabelo e das unhas, que comumente são preservados em esqueletos recém esqueletizados, fornece assinatura isotópica em relação tanto da dieta quanto de residência das últimas semanas e meses de vida.

Entretanto, se deve considerar que fatores como idade e condição de saúde influenciam as taxas de crescimento da queratina. Desse modo, segmentos de fios de cabelo e unhas devem ser segmentados para se precisar qual período cronológico da vida do indivíduo está sendo analisado. De acordo com Baterlink & Chesson[5], atualmente se tem mais compreensão do ritmo do crescimento do cabelo do que da unha, sendo, portanto, o cabelo um marcador mais confiável de análise.

Entendendo os isótopos

Os átomos possuem núcleos que contém próton (positivo) e nêutron (neutro). O número de prótons é o que dá a identidade química do elemento a partir do seu número atômico, uma vez que seu valor se mantém inalterado no mesmo elemento.

Os isótopos de determinados elementos se distinguem pela quantidade de nêutrons que ele apresenta[5]. Eles se dividem entre os estáveis e os instáveis (radioativos). Os primeiros sendo aqueles que não se deterioram em outros elementos, porque as suas combinações individuais de prótons e nêutrons são estáveis. Já os isótopos instáveis, por sua vez, possuem núcleos atómicos instáveis perante o balanço entre nêutron e prótons, e emitem alguma forma de radiação em período de desintegração e, dessa forma, possuem decaimento em períodos previsíveis.

Isótopos estáveis (IE) são formas diferentes, não radioativas e estáveis do mesmo elemento químico que diferem no número de nêutrons em seu núcleo e, portanto, em sua massa atômica. As IE de um elemento reagem quimicamente de maneira semelhante, mas essa pequena diferença de massa faz com que sua distribuição não seja aleatória. Moléculas contendo isótopos pesados (por exemplo, 18O) reagem mais lentamente e formam ligações mais fortes do que aquelas com

leves (por exemplo, 16O). Esse processo leva a uma separação ou fracionamento que determina amplamente a abundância relativa (18O / 16O) do IE em diferentes sistemas.

Ao trabalhar com o IE, é usada a notação δ ("delta"), que representa diferenças relativas na taxa de IE de uma amostra em relação a um material padrão e é formulada (exemplo para oxigênio) como: δ18O = (Ramostra / Rpadrão -1) × 1000, onde R representa a taxa do isótopo pesado em relação ao isótopo leve (18O/16O) e é expresso em unidades de partes por mil, ‰.

Como os isótopos podem ser usados nas Ciências Forenses?

A diversidade de isótopos que pode ser usada nas Ciências Forenses ocorre de acordo com a finalidade: crimes contra o meio ambiente, poluição e contaminação de águas, falsificação de alimentos, proveniência de drogas ilícitas, origem de animais silvestres, identificação de ossos humanos versus não humanos, zoologia forense, entre outros. Porém, são cinco os bioelementos aplicados em tecidos humanos como método auxiliar da Antropologia Forense, sendo ele os não metálicos, Hidrogênio (H), Carbono (C), Nitrogênio (N), Oxigênio (O), e Enxofre (S) e mais dois metálicos, o Chumbo (Pb) e o Estrôncio (Sr).

A análise de isótopos estáveis (AIE) é uma ferramenta particularmente útil para investigações forenses, pois é capaz de fornecer informações que sugerem a origem geográfica, biológica e / ou química de um material em observação e pode distinguir entre materiais aparentemente idênticos[8,1]. Em essência, o AIE é um meio muito valioso de fornecer análise comparativa de materiais de interesse no trabalho de casos forenses. Além disso, os processos de incorporação de isótopos estáveis em tecidos humanos têm sido bem descritos e modelos mecanicistas podem ser postulados que permitem estabelecer previsões para casos em que não há oportunidade de comparação direta[9,1,8].

A AIE pode ser especialmente útil durante investigações forenses envolvendo restos humanos não identificados, particularmente quando as técnicas tradicionais de investigação, como impressões digitais ou análise de DNA, não foram bem-sucedidas. Um grande número de publicações demonstrou a capacidade de isótopos estáveis de tirar conclusões sobre movimentos e migrações humanas usando os valores isotópicos de diferentes tecidos, incluindo ossos, dentes, unhas e cabelos que preservam informações sobre as condições ambientais e de saúde do indivíduo, bem como sobre seus alimentos e bebidas[10-15].

Diferenças nas taxas de crescimento de tecidos (ossos, cabelos, unhas, etc.) permitem o uso da AIE para reconstruir uma cronologia e documentar os movimentos geográficos de uma pessoa[16]. Embora A AIE não possa fornecer identificação direta, sem dúvida pode fornecer novas pistas e ajustar as opções de pesquisa (por exemplo, excluindo regiões geográficas específicas)[16,17]. A AIE aumenta o conhecimento do perfil biológico de um indivíduo, fornecendo informações adicionais sobre sua história, incluindo região de nascimento ou infância, a última década de vida ou as semanas e meses antes da morte. A AIE ajuda a responder perguntas como: O indivíduo se mudou ou viajou antes da morte? A pessoa mudou sua dieta antes da morte? A viagem e a dieta sugerem que o indivíduo era residente de uma região específica? O histórico de viagens e a região de origem do indivíduo correspondem ao de outra pessoa?

Apesar de uma rica história de estudos isotópicos para desvendar migrações humanas no passado[13,18-20], aplicação da AIE para pesquisar as origens dos migrantes modernos tem sido escassa na investigação forense humanitária (pelo menos como registrado em publicações até agora). Juarez (2008)[21] analisou a capacidade da AIE de determinar a região de origem das pessoas mortas durante sua tentativa de atravessar a fronteira entre o México e os Estados Unidos. Regan[22] e Holland et al.[23] usaram a AIE para identificar soldados mortos na Guerra do Vietnã e um piloto de avião no Laos, respectivamente. Bartelink et al.[24] usa o AIE para estudar a possível origem de indivíduos não identificados recuperados de diferentes regiões da Califórnia.

Existem poucos casos publicados nos quais isótopos estáveis levaram diretamente à resolução de um caso criminal, embora possa haver muitos casos em que ainda não há resolução e, portanto, eles não foram publicados. Rauch et al.[25] descrevem a AIE de vários elementos (C, N, H, Sr e Pb) em diferentes tecidos (ossos, dentes, cabelos e unhas) recuperados de um indivíduo do sexo masculino que foi encontrado enterrado perto de uma rodovia na Alemanha. Em 2008, Meier-Augenstein e Fraser[17] apresentaram uma combinação de dados isotópicos de H, O, C e N em restos mutilados de um homem descoberto em 2005 em Dublin, na Irlanda. Obviamente, o caso "Saltair Sally" mencionado acima também foi publicado em revistas científicas[1,9,26]. Em todos esses casos, a AIE foi crucial para fornecer informações que levassem à identificação da vítima e, nos dois casos europeus, levaram às prisões e acusações dos assassinos.

A abundância natural de isótopos estáveis de hidrogênio e oxigênio da precipitação (chuva ou neve) varia com a geografia. O principal reservatório de água e a principal fonte de água da chuva (ou neve) são os oceanos, que por definição têm valores de δ2H e δ18O iguais a zero[27-29]. Diferentes processos de fracionamento isotópico dos oceanos para o local onde chove ou neva causa diferenças geográficas na composição isotópica da água. Esses processos dependem, entre outros fatores, da temperatura e umidade, que estão, em média, globalmente relacionadas à latitude e elevação[27,28,30,31]. A tendência geral é de valores mais altos de δ18O em direção a baixas latitudes, baixa altitude e regiões costeiras e valores mais baixos de δ18O em direção a altas latitudes, altitude mais alta e regiões do interior dos continentes. O padrão geográfico dos valores de δ2H é semelhante aos valores de δ18O, embora locais altos de evaporação possam alterar sua correlação. Além disso, a variação espacial ou geográfica dos isótopos da precipitação se traduz em água de superfície, água da torneira e outras fontes de água, como água engarrafada, refrigerante e bebidas alcoólicas[32,33].

Moléculas de oxigênio e hidrogênio em tecidos humanos e animais são adquiridas diretamente da água potável, vapor de água ambiente, dieta e O2 atmosférico. A variação geográfica dos valores de δ2H e δ18O na água é traduzida em tecidos humanos com uma alta correlação porque o sinal isotópico do tecido é dominado pelo componente líquido da água bebido diretamente ou adquirido através do cozimento de alimentos[15,34-37]. Além disso, das outras fontes de variação isotópica, o alimento é o único com potencial para mascarar o sinal geográfico local se os indivíduos consumirem alimentos importados de outras regiões.

Os isótopos estáveis de carbono (δ13C), nitrogênio (δ15N) e enxofre (δ34S) nos tecidos humanos são derivados exclusivamente da dieta e suas proporções refletem as dos produtos consumidos; portanto, às vezes são chamados de "isótopos alimentares"[29,38]. Os valores de δ13C refletem de perto as proporções dos isótopos de carbono da fonte alimentar original e foram usados como indicadores da proporção de plantas C3 ou C4 na dieta humana, consumidas diretamente como alimento básico ou indiretamente através da alimentação animal[29,39-42]. Além disso, em certas regiões onde existe um grande contraste entre plantas C3 e alimentos de origem marinha, os valores de δ13C têm sido utilizados para diferenciar o consumo de recursos de origem terrestre e costeira[29,39,40,41,43]. As moléculas de nitrogênio são obtidas apenas pelo consumo de proteínas, e os valores de δ15N nos tecidos de um consumidor são maiores em relação à sua dieta[38,41,43,44]. Isto é devido a uma retenção preferencial do

isótopo mais pesado (15N) durante a excreção, o que gera um aumento nos valores de δ15N nos consumidores para cada nível trófico de uma maneira previsível. Isso permite que os valores de δ15N sejam utilizados como um indicador da proporção de proteínas animais e vegetais em uma dieta onívora[4,29,44].

Para além do C e N, o Enxofre (S) tem sido menos utilizado para identificação de dieta e local de proveniência geográfica, embora ele seja indicativo de ambientes marinhos versus terrestres, sendo os valores δ34S marinhos mais altos do que aqueles observados em ambientes terrestres, valores δ34S mais altos de um tecido humano são tipicamente interpretados como indicativos da dependência ou proximidade de um indivíduo com recursos marinhos. Contudo, os fatores que afetam as composições isotópicas de enxofre dos tecidos humanos ainda não são totalmente compreendidos. Além do mais, a poluição dos ambientes modernos afeta os valores de δ34S de ecossistemas modernos[5].

Nos últimos anos, os pesquisadores geraram informações substanciais sobre a variabilidade isotópica e a variação nas populações humanas modernas[45,47-50]. Por exemplo, os pesquisadores conseguiram mostrar diferenças nos valores de isótopos "dietéticos" estáveis entre e dentro de países desenvolvidos que permitiriam distinguir indivíduos de diferentes regiões. Valenzuela et al.[51] observaram homogeneidade nos valores de isótopos estáveis de C e N em cabelos nos EUA, porém encontraram diferenças geográficas nos valores de δ34S com valores mais baixos para o centro do país e mais altos para as costas. Essa descoberta levou à construção de um isoscape (mapa preditivo, ver adiante subitem) desse marcador para seu uso potencial na atribuição de região de origem. Valenzuela et al.[42] relataram diferenças notáveis entre os EUA e a Europa Ocidental na composição isotópica de C e S no cabelo, bem como correlações dos valores de δ13C e δ34S com variáveis geográficas na Europa. Mais recentemente, foi demonstrado que o IE do cabelo difere entre os grupos socioeconômicos nas cidades dos EUA e Colômbia[50,52].

O uso de isótopos estáveis para estudos de movimentos e atribuição à região de origem depende de um conhecimento profundo de sua distribuição espacial. Portanto, o sucesso das previsões de origem com base na medição de isótopos nos tecidos depende da qualidade dos dados isotópicos que foram utilizados para construir os mapas preditivos ou isoscopias da linha de base. Embora a comparação entre amostras recuperadas na mesma região (banco de dados) possa fornecer uma indicação da presença de imigrantes, a criação de mapas que representam a distribuição espacial de isótopos e capturam a variabilidade local oferece a

possibilidade de gerar probabilidades de atribuição incorporando índices de variabilidade e incerteza[53].

Se as análises para investigação de dieta para se saber sobre proveniência e mobilidade e migrações se baseiam em isótopos de O, C, N e S há ainda outro espectro de isótopos de traços de elementos metais como o Estrôncio (Sr) e o Chumbo (Pb)[54].

Radioisótopos para análise de tecidos humanos

Menos usual na Antropologia Forense por questões técnicas e metodológicas mais complexas, estão o Sr e o Pb. No entanto, no que concerne ao Sr, seu potencial é grande para uso forense como complemento para compreensão do C e N, pois ele pode informar sobre onde um indivíduo viveu sua infância.

No ambiente, ações ocasionadas por intemperismo acarretam na liberação de Sr da rocha no ambiente local, onde ele é primeiramente absorvido pelas plantas e, posteriormente, pelos animais em seus alimentos. Dessa forma, a variação isotópica de Sr no ambiente está relacionada a cronologia e sua suscetibilidade ao clima em que está inserido o depósito rochoso[55]. O isótopo radiogênico 87Sr é um produto do decaimento radioativo de 87Rb. Os depósitos de base com altas concentrações iniciais de 87Rb e / ou depósitos antigos normalmente apresentam taxas 87Sr / 86Sr mais altas do que depósitos jovens ou depósitos com baixas concentrações de 87Rb[54].

Semelhante ao Sr, o método do isótopo de Pb aproveita a deterioração radioativa, o tempo e as concentrações relativas de elementos de Pb, Th e U no ambiente. Os isótopos radiogênicos 206Pb, 207Pb e 208Pb são derivados do decaimento radioativo de 238U, 235U e 232Th, respectivamente. Algumas variações nas proporções de Pb podem se alterar por fatores cronológicos (208Pb / 204Pb, 207Pb / 204Pb e 206Pb / 204Pb) e as atividades antropogênicas tais como a mineração ou a introdução de tetraetiléter na gasolina na década de 1920 como um agente "antiderrapante" para motores[56].

O 14C e seu decaimento

A análise 14C é a técnica tradicionalmente conhecida pelo seu emprego em estudos arqueológicos para datação de amostras e remanescentes ósseos e

dentários que datem entre aproximadamente 400 e 50.000 anos. Nesse caso, o isótopo se refere a uma taxa relativamente constante de produção de 14C na atmosfera superior que é absorvido por plantas verdes e toda a cadeia alimentar subsequente, ou seja, de animais que se alimentaram dessa planta e dos animais carnívoros[57].

Ao longo da vida de um organismo, ele retém valor de 14C em equilíbrio, porém, a partir de sua morte, deixa de assimilar o 14C e, entrando em um processo de decaimento do nível desse isótopo (de acordo com a meia-vida de aproximadamente 7.340 anos).

Como é bem conhecido na arqueologia, a datação radiocarbônica apresenta um intervalo de erro que é variável conforme o ponto da curva de decaimento da datação, como se trata de uma curva exponencial, nas partes mais recente e mais antiga a tendência é apresentar uma maior margem de erro.

Assim sendo, na investigação forense, a datação por 14C, por estar em épocas muito recente, cujo processo de decaimento ainda é muito próximo, não ajuda a calcular a data de morte do indivíduo. Por exemplo, se o indivíduo morreu em 1985 e tivermos um erro de aproximadamente + 50 anos, o método mais atrapalha do que auxilia no estabelecimento de algum parâmetro do período de morte do indivíduo.

O emprego do 14C na investigação de tecidos humanos forense

A estimativa do tempo decorrido desde a morte (intervalo *postmortem*) de tecidos ósseos humanos representa um componente crítico da investigação antropológica forense. Os investigadores precisam saber se os remanescentes datam do período moderno de interesse médico-legal (geralmente nas últimas décadas).

Infelizmente, uma extensa pesquisa e experiência de casos demonstraram que essas estimativas realizadas a partir de observações de preservação de tecidos humanos são extremamente imprecisas devido à grande variedade de fatores que influenciam o processo de decomposição. Na maioria dos casos, na ausência de informações contextuais de diagnóstico, os indicadores morfológicos não permitem distinguir restos modernos de antigos e / ou estimativas confiáveis de tempo desde a morte.

Ademais, sobre a idade do indivíduo no momento da morte, aspecto importante para traçar o perfil do indivíduo para cruzar com o perfil de desaparecidos, os métodos da antropologia forense conseguem dar um maior grau de precisão no caso

de remanescente juvenis do que para os adultos maduros, onde o espectro de estimativa de idade é bem mais largo e menos acurado.

Embora o método tradicional de datação por 14C proveniente da arqueologia não nos auxilie compreender a cronologia dos ossos humanos, Douglas Ubelaker[58] observou que o uso de radiocarbono de bomb pulse oferecia uma solução para o problema de estimativa do tempo desde a morte (Fig. 2).

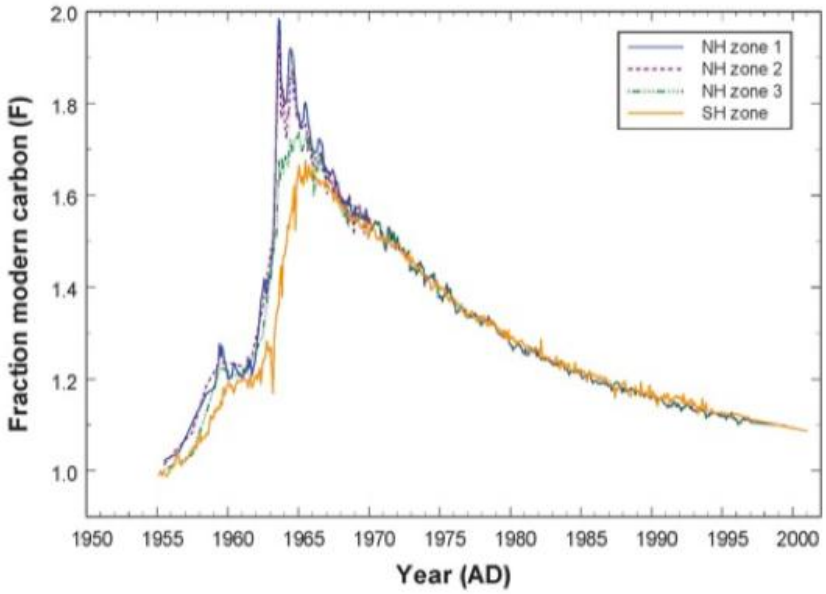

Fig. 2 - Representação de curva de radiocarbono para 04 diferentes zonas da Terra. Fonte: Hua & Barbetti, 2004[59].

Os níveis de 14C na atmosfera permaneceram inalterados até 1955 quando, a partir de então, devido aos mais de 1000 testes realizados com bomba nucleares durante o 1945 a 1962, período que engloba a Guerra Fria, a quantidade de 14C presente na atmosfera aumentou gradualmente, chegando em seu pico em 1963, ano em que foi assinado o Tratado de Interdição Parcial de Ensaios Nucleares, no qual diversos países signatários se comprometeram com a cessação dos testes com bombas nucleares, com o objetivo de retardar a corrida armamentista e conter a excessiva liberação de cinzas nucleares na atmosfera.

Após a cessação dos principais testes termonucleares no início dos anos 1960, os níveis de radiocarbono atmosférico atingiram um pico em cerca de 1963 e, subsequentemente, começaram a entrar em um lento declínio.58

Dessa forma, tanto a introdução desse elemento na atmosfera como sua incorporação pelas plantas resultaram em um gráfico no qual é possível conhecer, em determinado ano, a concentração desse elemento no ar e vice-versa.

Na análise forense de tecidos humanos, elevados níveis de radiocarbono acima dos níveis anteriores a 1950 provam que a pessoa representada estava viva após 1950. Como os anos seguintes a 1950 correspondem aproximadamente ao período de interesse médico-legal, o radiocarbono de bomb pulse oferece informações importantes em relação à estimativa do intervalo *postmortem*.

Se forem descobertos valores elevados (modernos) de radiocarbono, o objetivo forense passa a estimar quando, durante o período moderno, ocorreu a morte. Com um único valor moderno de radiocarbono, geralmente não é possível determinar se ele se relaciona com os níveis atmosféricos / dietéticos no aspecto de ascensão precoce da curva ou na parte posterior da curva posterior a 1963.

Ubelaker et al.[60] demostraram ser possível estimar a data da morte por meio da análise a partir do tecido ósseo, apontando que análises de radiocarbono de dois tecidos que refletem diferenças na formação e / ou taxas de remodelação sendo, portanto, a comparação dos valores desses dois tecidos adequados para melhor posicionamento na curva.

Por exemplo, o osso trabecular do corpo de uma vértebra lombar remodela mais rapidamente que o osso cortical denso da diáfise femoral. Assim, dentro de um único esqueleto adulto, os valores de radiocarbono derivados do osso trabecular cruzariam a curva da bomba em um ponto mais próximo da data da morte do que os derivados do osso cortical. A partir da perspectiva de bomb pulse de 14C, foi constatado ser possível estimar o ano de nascimento e de morte de um indivíduo por conta do decaimento em taxas conhecidas do 14C.

Ubelaker e Parra[61] apresentaram um importante estudo realizado em quatro indivíduos provenientes dos Andes peruano (16, 27, 44 e 56 anos de idade). Em relação ao indivíduo mais jovem, a análise 14C do esmalte permitiu a determinação precisa do ano de nascimento e da cortical e as atividades do osso trabecular, cujos resultados foram de valor de 14C consistentes com a data da morte. Para os três indivíduos mais velhos os resultados apontaram atividades de esmalte dentário de 14C consistentes com os anos de nascimento. No entanto, para esses três indivíduos, houve descompasso mínimo entre a formação óssea trabecular e a morte. Quanto a idade de morte para os três indivíduos mais velhos, o método apresentou discrepância (≥ 11 anos) entre o ano da morte e o valor de 14C do osso cortical

analisado. Por essa razão, faz-se ainda necessário ampliar os estudos em ossos em remanescentes de indivíduos identificados para balizar a taxa de remodelamento ósseo em relação as atividades de 14C.

Ainda assim, o método se mostrou promissor desde o início e já tem se demonstrado eficaz, nos últimos anos outros pesquisadores passaram a tentar estimar as atividades de 14C a partir de medições realizadas em dentes únicos, ou na comparação de dentes formados em momentos diferentes da infância do indivíduo, podendo fornecer estimativas do ano de nascimento entre aproximadamente um ou dois anos do real[57].

Baseados no gráfico do bomb pulse, Spalding et al.[62] apresentaram um estudo no qual foi demonstrado o sucesso da determinação do ano de nascimento de um indivíduo por meio da análise quantitativa de 14C em esmalte de dentes humanos de adultos. A estimativa da data de nascimento é baseada na obtenção da quantidade de 14C contida no esmalte no período correspondente na curva de 14C atmosférico associando-se à verificação da cronologia de mineralização do elemento dentário em questão em tabelas validadas em outros estudos.

Com o objetivo de obter solução para a questão de ambiguidade de qual lado do pico os resultados se relacionavam, foram medidos os índices de 14C em dois dentes que se formam em momentos diferentes. Cook et al.[57] e Cook & MacKenzie[63] propuseram a determinação de um ano preciso de nascimento do indivíduo a partir de análises 14C a partir de dois materiais distintos, o esmalte e o colágeno da dentina e cemento combinados. Dessa forma, os autores concluíram que o resultado de 14C do esmalte da coroa, formado antes da raiz, combinado com o 14C da dentina / cemento apontar um índice maior, a idade deve estar na curva ascendente e, se for menor, a idade deve estar na curva descendente.

Wang et al.[64] realizaram um teste a partir da seleção de esmalte próximo ao colo do dente, para redução do erro causado pela diferença entre o tempo de formação da amostra e o tempo considerável para a formação de todo o componente do esmalte. Por sua vez, Kondo-Nakamura et al.[65] precisaram idades em dentes únicos medindo 14C no esmalte de duas regiões, oclusal e cervical, à medida que se formam em diferentes momentos.

Os isótopos estáveis na Arqueologia Forense / Local de Crime

A partir de recursos metodológicos tradicionais da arqueologia, o arqueólogo com experiência em estudos forenses pode trabalhar em conjunto com equipes que trabalham em cenas de crime com o objetivo de reconstruir as atividades em locais de crimes mais recentes, a partir da contextualização das evidências e objetos *in loco*.

Nesse momento uma série de protocolos focados para interpretação de diversas áreas das Ciências Forenses devem ser aplicados de modo a não perder informações relevantes para a compreensão dos episódios que se sucederam, bem como preservação das evidências. Dentro desse cenário, ter em foco a possibilidade de interpretação de um caso de antropologia forense por meio de isótopos estáveis é essencial já em campo.

Como visto anteriormente, a base da incorporação dos isótopos nos tecidos humanos tem origens ambientais, geográficas e culturais e, em campo, o profissional deve estar atento para observar elementos que possam ser indicativos de fonte e origem de isótopos estáveis para recomendação de análises.

Vale lembrar que, embora tratado como um dado absoluto, como o DNA ou a impressão digital, os números dos isótopos não são precisos, e podem sofrer alterações a partir da sua fonte de origem, dependendo dos processos que possam alterar sua composição, e o investigador tem que ter em mente essa precaução na hora da interpretação de seus dados[3].

Para ser útil a assinatura isotópica de um osso, é necessário que se diferencie as alterações que ocorreram em momentos antemortem, perimortem dos postmortem, resultantes por exemplo da exposição ao solo e às águas subterrâneas. Os dados *antemortem* são indicativos de atividades durante a vida da pessoa que pode ajudar em sua identificação, os dados *perimortem* auxiliarão a entender a cena do crime enquanto os dados *postmortem* auxiliarão a compreender atividades que possam tentado ocultar o crime ou, ainda, processos diagenéticos que podem ser cruciais para a boa interpretação dos eventos.

De modo geral, a diagênese não atua em tecidos duros de corpos recém esqueletizados, pois eles são resistentes à alteração química a curtos intervalos de tempo. Porém, em tecidos humanos, sobretudo em cabelos e unhas, expostos e em contato com elementos que levam a ações diagenéticas, pode ocorrer uma degradação do isótopo significativa em curto prazo de tempo, e mais lentamente, nos ossos e dentes.

A diagênese acontece a partir de mudanças químicas e estruturais que ocorrem nas matérias à medida que são expostas a diferentes condições de modo a afetar, inclusive, os isótopos presentes nos tecidos e em que proporções.

O modo como a diagênese e os isótopos atuam em diferentes matérias dependem do tipo e grau de exposição a que foi submetido e, portanto, influenciam suas assinaturas isotópicas.

Por meio do contato da água subterrânea, por exemplo, a diagênese pode alterar a hidroxiapatita de superfícies ósseas ou permeando no interior do osso através de canais, canais de Haversian e Volkmann ou rachaduras *postmortem*. Ademais, evidências de que o ataque microbacteriano pode alterar a fração orgânica do osso, o colágeno, que é de especial relevância para a datação por radiocarbono[54].

Dentre os problemas de fatores diagenéticos a serem considerados na análise de isótopos estáveis recai especialmente no isótopo de Estrôncio (Sr), pois este elemento é mais suscetível a trocas diagenéticas com elementos do ambiente onde o um indivíduo enterrado ou está exposto, o que pode alterar o sinal de Sr/Ca Processos que levem a troca de Sr podem ser ainda mais intensos se facilitados pela ação d'água no local. Desta forma, a presença deste elemento no osso não permite saber se essa concentração foi diageneticamente alterado ou não. Na tentativa de contornar o problema, por muito tempo foi usado a dentina como fonte mais confiável para essa análise, levando, no entanto a se interpretar o início da vida, quando então o dente foi formado, restando a lacuna do período final da vida adulta do indivíduo analisado[66].

Atualmente, muitos laboratórios já possuem metodologia adequada para remover o Sr contaminado de amostras analisada. Recentemente, um estudo realizado por Snoeck et. al.[67] em amostras de esmalte dentário e osso calcinado que foram expostas a 87Sr durante o período de um ano. As amostras foram lavadas com ácido acético e ultrassom. As taxas de 87Sr / 86Sr foram mensuradas antes e após o tratamento em um espectrômetro de massa de plasma. As razões isotópicas de estrôncio de todas as amostras imersas na solução foram fortemente modificadas, mostrando que quantidades significativas de estrôncio foram adsorvidas ou incorporadas.

Após o pré-tratamento, as amostras de esmalte ainda continham quantidades significativas de contaminação enriquecida com 87Sr, enquanto os fragmentos ósseos calcinados não. Os resultados, contudo, demonstram que as amostras de esmalte dentário ainda preservaram quantidades significativas de contaminação enriquecida

com 87Sr mesmo após a limpeza do material, enquanto que o osso calcinado não. Assim, o osso calcinado manteve preservada a assinatura isotópica antemortem, se demonstrando mais resistente à troca postmortem do que o esmalte do dente.

Isoscapes

Isoscapes são mapas georreferenciados onde são descritos nível de processo de fracionamento ou distribuição de isótopos elementares para melhor interpretação de padrões espaciais[68].

Esses mapas preditivos podem ser gerados para diferentes matérias como ossos, água e solo e vários elementos isotópicos, C, N, O etc, sendo extremamente úteis para correlacionar com dados de perfis isotópicos, com a finalidade de em conjunto com o perfil biológico e outras informações de identificação do esqueleto, fornecer dados que possibilitem restringir possíveis regiões geográficas pelas quais o indivíduo esteve durante sua vida.

Esses mapas são extremamente necessários para possibilitar cruzamento de dados confiáveis em quantidade e qualidade suficientes para apoiar a interpretação das composições isotópicas dos tecidos humanos. A falta desse tipo de mapeamento, pode ser um empecilho na utilização de dados provenientes de isótopos para finalidade forense em um tribunal.

No Brasil, a possibilidade de criação de isoscapes em isótopos estáveis forenses é um projeto da Polícia Federal que está em fase de implementação do seu Programa Nacional de Isótopos Forenses com a criação do Laboratório de Isótopos Forenses da PF (Lanif), com o objetivo de desenvolver e análises isotópicas para exames periciais de vestígios criminais de diversas origens, inclusive de tecidos humanos.

Em 2019, por realização da Polícia Federal, foi promovido o I Workshop de Isótopos Forenses, no Instituto Nacional de Criminalística, Brasília. O evento reuniu peritos criminais federais e estaduais, acadêmicos e demais profissionais atuantes no ramo da ciência isotópica com a finalidade de pensar estratégias e protocolos de pesquisas de modo a desenvolver e integrar os temas de isótopos forenses em uma rede nacional.

Dentre os Grupos de Trabalho que foram formados para a promoção de protocolos de coleta, preparação, armazenamento e análises isotópicas, com o

objetivo de se recolher dados para se criar diversos isoscapes, foi consolidado o GT Tecidos Humanos, ao qual integram os três autores desse artigo.

Essa iniciativa, se devidamente implantada, gerará informações importantes para as investigações forenses, sobretudo de interface com as análises antropológicas forenses, possibilitando uma maior quantidade de filtros que restrinjam o leque de possibilidades para identificação de indivíduos, devendo, portanto, beneficiar todos os Estados brasileiros.

Conflict of interest

There are no known conflicts of interest associated with this publication, and there has been no significant financial support for this work that could have influenced its outcome.

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Case Study

Forensic Anthropology

Identificação positiva de corpo carbonizado através dos arcos dentais

Positive identification of a burned body using dental analysis

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RESUMO

Este estudo tem como objetivo relatar um caso de identificação humana de um corpo carbonizado, através de análise odontológica. As observações foram feitas em um

corpo não identificado pertencente ao Instituto de Medicina Legal de São Luís / MA, Brasil. O corpo masculino carbonizado foi encontrado dentro de um veículo após um acidente de trânsito. O corpo foi severamente carbonizado, com partes calcinadas e volume reduzido. O registro dentário, constituído de uma história clínica, datada de 2015/2016, que continha algumas intervenções odontológicas realizadas, além de radiografia periapical dos dentes do pré-molar superior do lado direito, foi fornecido pelos familiares. Considerando que foram encontradas várias informações e características quantitativas e qualitativas coincidentes entre a documentação odontológica apresentada pelos familiares e o exame do cadáver, e pela singularidade e especificidade desses dados, e também porque nenhuma discrepância exclusiva foi detectada durante no exame, foi possível realizar a identificação positiva por meio de análise odontológica.

Descritores: Antropologia Forense; Identificação humana; Odontologia Forense.

ABSTRACT

This study aims to report one case of human identification of a burned body using dental analysis. The observations were made in an unidentified body belonging to the Institute of Legal Medicine of São Luís / MA, Brazil. The charred male body was found inside a vehicle after a traffic accident. The body was severely carbonized, with calcined parts and reduced volume. The dental record, consisting of a clinical history, dated 2015/2016, which contained some dental interventions performed, as well as periapical radiography of the upper premolar teeth on the right side, was provided by family members. Considering that several pieces of information and coincident quantitative and qualitative characteristics were found between the dental documentation presented by the family members and the examination of the corpse, and because of the uniqueness and specificity of these data, and also because no exclusive discrepancy was detected during the examination, it was possible to perform the positive identification using dental analysis.

***Keywords*:** Forensic Anthropology; Human Identification; Forensic Dentistry.

Introdução

A identificação pessoal é essencial tanto por razões legais como humanitárias[1], sendo a base de investigações criminais e um pré-requisito para declarar oficialmente a morte de uma pessoa[2,3]. Nessa perspectiva, a Odontologia Legal desempenha papel relevante a serviço da justiça, aplicando os conhecimentos técnicos odontológicos no processo de identificação humana *post-mortem*[2,4,5].

O fundamento da perícia odontológica de identificação está no fato de as características e a constituição dental serem únicas a cada pessoa, individualizando-as[6,7,8]. Em situações em que o reconhecimento visual está prejudicado e as impressões digitais não estão preservadas, como nos casos de corpos esqueletizados, putrefeitos ou carbonizados, a metodologia odontológica mantém-se eficaz, visto que os dentes são estruturas duráveis e resistentes[6,9], assim como os materiais restauradores usados na clínica odontológica, podendo manter-se na cavidade bucal mesmo quando submetidos a altas temperaturas[10].

Essa característica é especialmente importante, uma vez que diversos eventos podem levar à carbonização de corpos, como explosões, bombardeios, acidentes aéreos e acidentes de trânsito. Ademais, existe ainda a possibilidade de o fogo ser usado na tentativa de destruir provas periciais e impedir a identificação da vítima, em casos criminais[11].

O exame odontolegal de identificação humana é uma técnica simples, de baixo custo e classificada como comparativa, por relacionar informações odontológicas *ante-mortem* e *post-mortem* da suposta vítima[5,6,12,13].

A primeira etapa consiste no levantamento e análise das características odontológicas verificadas no exame do cadáver, tais como presença e ausência dental, cáries, restaurações, tratamentos endodônticos, forma e posicionamento dental, anomalia óssea e dental, presença de próteses, entre outras. Em seguida, faz-se o estudo das informações contidas no prontuário odontológico produzido antes da morte do indivíduo, composto por ficha clínica, radiografias, modelos em gesso e fotografias. Por fim, realiza-se o estudo e confronto detalhado dos dados obtidos nas etapas anteriores no intuito de verificar, qualitativa e quantitativamente, as coincidências e divergências porventura existentes[6,14].

Neste contexto, o presente trabalho teve como objetivo evidenciar a importância da documentação odontológica no trabalho pericial, por meio do relato de um caso de identificação positiva de um corpo severamente carbonizado, proporcionada pelo exame odontolegal.

Relato do caso

No ano de 2018, deu entrada no Instituto Médico Legal (IML) de São Luís/MA um corpo carbonizado, do sexo masculino, encontrado no interior de um veículo, vítima de acidente de trânsito. O corpo apresentava-se severamente carbonizado, com partes calcinadas e reduzido de volume, impossibilitando o reconhecimento visual, assim como a verificação de sinais/características particulares e a coleta das impressões digitais para exame papiloscópico (Fig. 1).

Fig. 1. Registro do cadáver carbonizado, ao ser recebido para exame pericial.

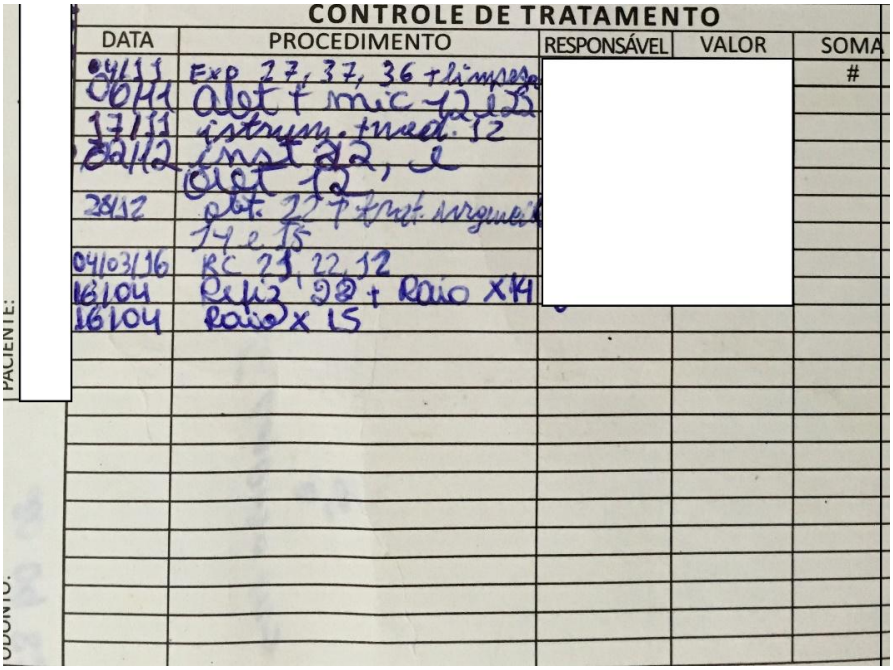
Apesar do alto grau de destruição dos tecidos pela ação do fogo, constatou-se a presença de elementos dentais, viabilizando a escolha da metodologia odontológica no processo de identificação. Para realização do exame pericial odontológico, procedeu-se a enucleação da maxila e da mandíbula de forma cuidadosa com o intuito de não danificar ainda mais sua estrutura.

Na maxila foi constatada perda de substancia óssea na região posterior esquerda e a ausência da coroa de vários elementos dentais, estando presente suas respectivas raízes no interior dos alvéolos, em decorrência da ação do fogo (Fig. 2). Na mandíbula verificou-se fratura na região do corpo à esquerda e presença de alguns dentes íntegros (Fig. 3). Em seguida, foram realizadas incidências radiográficas periapicais na maxila, na região anterior e posterior direita.

Fig. 2. Maxila do cadáver carbonizado recebido para exame pericial.

Fig. 3. Mandíbula do cadáver carbonizado recebido para exame pericial.

Familiares compareceram ao IML reclamando o corpo e informaram que a suposta vítima estava na presença da família quando entrou em seu veículo e sofreu um acidente a poucos quilômetros de distância. Desse modo, foi-lhes solicitada a documentação odontológica da suposta vítima que resultou na apresentação de prontuário odontológico composto de ficha clínica com datas de atendimentos referentes aos anos de 2015/2016 (Fig. 4), na qual constava algumas intervenções odontológicas realizadas e radiografia periapical da região dos dentes pré-molares superiores do lado direito (Fig 5).



CONTROLE DE TRATAMENTO

DATA	PROCEDIMENTO	RESPONSÁVEL	VALOR	SOMA
				#
04/11	Exp 27, 37, 36 + limpeza			
06/11	abt + mic 12, 13			
17/11	instrum. tmed. 12			
22/12	inst 12, e			
	abt 12			
28/12	abt. 27 + inst. urgência			
	14 e 15			
04/03/16	RC 21, 22, 12			
16/04	Rx 13, 22 + Raio X 14			
16/04	Raio x 15			

PACIENTE:

ODONTO:

Fig. 4. Ficha odontológica fornecida para confronto.

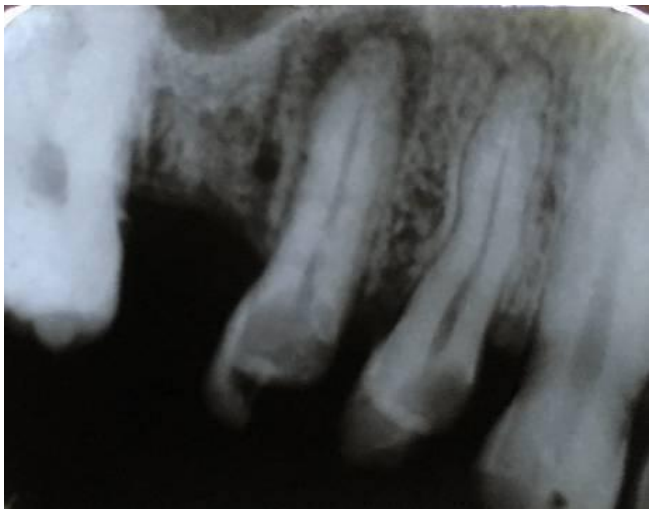


Fig. 5. Radiografia periapical *ante-mortem.*

Para melhor visualização do levantamento das informações e particularidades odontológicas verificadas no exame da documentação *ante-mortem* (AM) e no exame pericial *post-mortem* (PM) do cadáver, foi elaborado o Quadro 1.

Quadro 1. Confronto odonto-legal das informações *ante-mortem* (AM) e *post-mortem* (PM) (IML/São Luís/MA, 2018).

Dente	Informação *ante-mortem* (AM)	Informação *post-mortem* (PM)
12	Tratamento endodôntico Restauração de resina composta	Canal radicular obturado Ausência da coroa dental em decorrência da ação do fogo
21	Restauração de resina composta	Ausência da coroa dental em decorrência da ação do fogo
22	Tratamento endodôntico Restauração de resina composta	Canal radicular obturado Ausência da coroa dental em decorrência da ação do fogo
14	Tratamento de urgência. Indicação de tratamento endodôntico.	Lesão periapical (área radiolúcida adjacente ao ápice radicular)
15	Tratamento de urgência. Indicação de tratamento endodôntico.	Lesão periapical (área radiolúcida adjacente ao ápice radicular)
16	Ausente	Ausente
27	Exodontia	Prejudicado (perda de substância óssea)
37	Exodontia	Ausente
36	Exodontia	Ausente

Discussão

A Odontologia Legal contribui de forma significativa para o processo de identificação humana, sendo um método seguro, eficaz e vantajoso também quando aplicado em corpos carbonizados[15]. Contudo, por ser comparativo, o sucesso da técnica depende da disponibilidade e qualidade dos registros odontológicos *ante-mortem* da suposta vítima que possam ser comparados com os dados coletados no exame *post-mortem*[6,14].

No presente caso, os familiares apresentaram ficha clínica com descrição de tratamento realizado e radiografia periapical. As radiografias odontológicas são documentos importantes na perícia devido a quantidade de informações capaz de serem extraídas da imagem, como por exemplo tamanho e formato da coroa e raiz dental, posicionamento e forma da crista óssea alveolar e do seio maxilar, assim como a presença de material restaurador ou obturador[1].

Na ficha clínica odontológica foi verificada a informação de tratamento endodôntico realizado nos incisivos laterais superiores direito e esquerdo (dentes 12 e 22). Essa informação coincidiu com o achado na radiografia periapical realizada durante o exame necroscópico que evidenciou os condutos radiculares dos referidos elementos dentais com a presença de material obturador (Fig. 6).

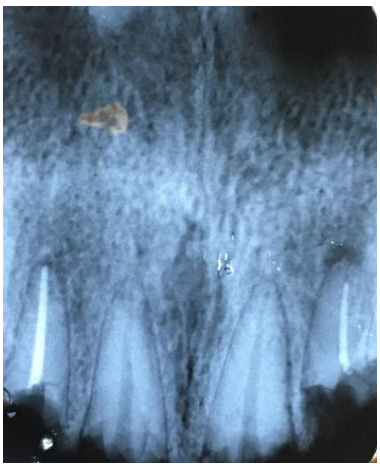


Fig. 6. Radiografia periapical post-*mortem* evidenciando os dentes 12 e 22 com condutos radiculares obturados.

Os dentes 14 e 15, segundo consta na ficha clínica, foram submetidos a tratamento de urgência e receberam indicação para tratamento endodôntico em 2015. A imagem radiográfica *ante-mortem* desses dentes evidencia o comprometimento pulpar dos mesmos. Esses dados vão ao encontro das características retratadas na imagem radiográfica PM que mostrou área radiolúcida adjacente ao ápice radicular desses dentes, sugestiva de lesão periapical. O aumento da lesão periapical verificado na radiografia *post-mortem* é justificado pela evolução da lesão que não recebeu o tratamento indicado há 03 anos (Fig. 7).

Fig. 7. Radiografia periapical ante-*mortem* (A) e *post-mortem* (B) da região dos dentes 14 e 15.

A análise e comparação das radiografias AM e PM da região dos dentes 14 e 15, permitiu ainda verificar outras particularidades importantes compatíveis em ambas as imagens, tais como, a ausência do dente 16, o espaço entre os dentes 15 e 17 e o formato do contorno do seio maxilar e o contorno sinuoso da raiz do dente 14 (Fig 8).

Fig. 8. Radiografia periapical ante-*mortem* (A) e *post-mortem* (B) da região dos dentes 14 e 15 evidenciando o contorno sinuoso da raiz do dente 14.

Algumas informações contidas na ficha clínica, como as restaurações de resina composta realizadas nos dentes 12, 21 e 22 e a exodontia do dente 27 não puderam ser confirmadas no exame *post-mortem* em decorrência da ausência das coroas dos dentes anteriores superiores e da perda de substancia óssea na porção posterior esquerda da maxila. Salienta-se que mudanças estruturais, encolhimento, fragmentação, alguma alteração de forma, presença de fissuras, o enegrecimento de raízes, a perda e/ou pulverização da coroa são alterações dentárias comuns e gradativas, decorrentes da exposição do corpo a altas temperaturas[6,11] (Fig. 2). Já os

dentes 36 e 37 estavam ausentes no exame PM da mandíbula, corroborando com a descrição de exodontia realizada em vida (Fig. 3).

Importante ressaltar que não foram encontradas divergências excludentes no confronto dos dados. Pequenas discordâncias observadas nas imagens radiográficas AM e PM são justificadas por diferenças entre os padrões radiográficos (incidência, intensidade, posicionamento, entre outros) aplicados nos registros AM e PM. Outrossim, a obtenção de registros radiográficos em corpos carbonizados é complicada devido à fragilidade da estrutura óssea, à dificuldade de posicionamento do corpo[1] e a mudanças estruturais que posam ter ocorrido.

Considerando que foram encontradas várias informações e características quantitativas e qualitativas coincidentes entre a documentação odontológica apresentada pelos familiares e o exame do cadáver, e diante da unicidade e especificidade desses dados, e, ainda, por não ter sido detectada nenhuma discrepância excludente no exame, concluiu-se como positiva a identificação pelo método odonto-legal.

Salienta-se que não existe um número mínimo de pontos concordantes necessários para se alcançar uma identificação positiva, bastando que as informações verificadas tenham qualidade suficiente para individualizar a vítima[6,16].

Conclusão

A análise das características odontológicas consiste em método eficaz e seguro de identificação, capaz de ser aplicado em corpos severamente carbonizados. Para tanto, faz-se necessária a existência de documentação odontológica *ante-mortem* produzida durante a clínica diária. A conscientização dos cirurgiões-dentistas quanto a importância desse documento na esfera pericial é imprescindível para que realizem o registro e o arquivamento adequado do prontuário do paciente que poderá servir de base para a perícia de identificação humana *post-mortem*.

Conflito de Interesse

Não há conflitos de interesse conhecidos associados a esta publicação e não houve apoio financeiro significativo para este trabalho que pudesse ter influenciado seu resultado.

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Case Study

Osteology

Sphere-shaped foreign bodies found in the spongy bone of axis vertebra

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ABSTRACT

Among the various areas that compose forensic medicine, forensic anthropology stands out in the determination of human remains. Forensic anthropology makes it possible to determinate the bioanthropological profile of human skeletons in the following categories: sex, age, stature, population affinity. Plus, it is possible to find, occasionally, bone markets connected to cause of death, such as gunshot wounds. This study aimed to describe the analysis of spheres encrusted in the spongy bone of

the axis vertebra from a male skeleton belonging to the Contemporary Human Bones Collection of the Forensic Anthropology and Osteology Laboratory (LAOF) at UFPE. For the analysis were used an electronic magnifier and a T100 Canon digital camera. The physicochemical tests were performed at the Physicochemical Laboratory of Professor Armando Samico Institute of Criminalistics (ICPAS) of Pernambuco. It was observed an atypical morphology of the axis vertebra – which has not shown his odontoid process, injuries on the skull compatibles with gunshot wounds, and small spheres lodges in the spongy bone of the vertebra. After the physicochemical tests, it was concluded that those foreign-bodies had physicochemical characteristics of hydroxyapatite, an organic product.

***Keywords*:** Forensic Anthropology; Axis, Cervical Vertebra; Hydroxyapatite.

Introduction

Forensic anthropology is one of the fields of forensic medicine that has a notably social role by presenting a significant contribution to the different criminal sciences. This field consists of a practical application of techniques for the determination of unknown human remains[1].

In the study of human skeletons, it is essential to recognize the morphological peculiarities of each human bone. Scientifically speaking, the human bone is composed of two tissues: compact bone and spongy bone. These two types of tissues have specific functions; for example, the spongy tissue is responsible for the cell-matrix[2].

The vertebrae, classified as irregular bones, present a generous volume of spongy bone tissue at the vertebral body, covered with a slight layer of compact bone tissue[3]. Occasionally, the spongy bone tissue is susceptible to contain foreign bodies and particles due to present cavities inside. This paper intends to report a case of spheres-shaped foreign bodies detected at the odontoid process spongy bone tissue of axis cervical vertebra.

Case Report

The following report treats about an analysis of a human skeleton belonging to the *Contemporary Human Bone Collection (LAOF/UFPE)*, whose skull presented foramina consistent with gunshot wounds[4]. When assembling and analyzing the skeleton, we observed that except the coccyx and pisiform bones, all the other bones were present. It is also noteworthy that the individual´s sacrum bone is not completely fused, which presents us whith bone characteristics of youthfulness. After a more careful approach, it was found that the bone head had fragmented and with the presence of orifices, which are consistent with a perforating instrument, in addition to which the mandible had a a metal plate possibly used for surgical approach. When evaluating the cervical vertebrae we found peculiar features in the second vertebra, also known as the axis. Presenting a coronal (and oblique) section at the level of the odontoid process we observed the exposure of its spongy tissue (Fig. 1) By using an electronic magnifier, dark and solid spheres were discovered in the spongy bone of the odontoid process, followed by the capture of the unknown material (Fig. 2).

Fig. 1 – Axis cervical vertebra obliquely sectioned. Superior view.

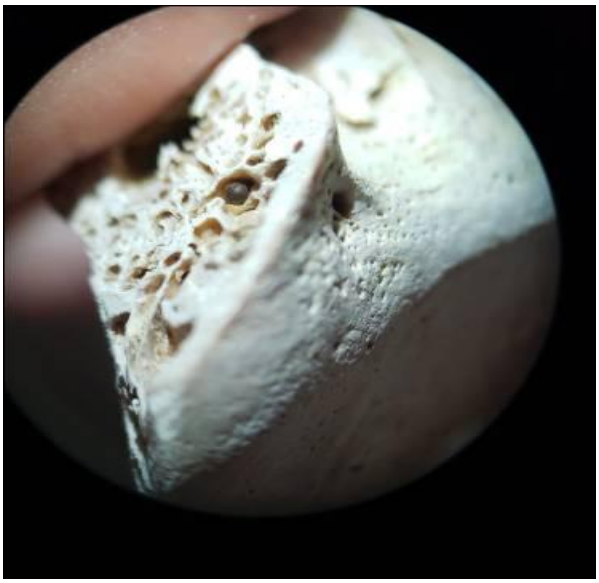

Fig. 2 – Sphere-shaped foreign body.

At *LAOF*, the diameter of these spheres was analyzed with a caliper, obtaining a diameter of 1.12mm. At *ICPAS*, the spheres were analyzed for physicochemical characteristics. After Raman spectroscopy, the results of the sample came back negative for Raman scattering for the two wave-lengths available at the device; thus, infrared spectroscopy was performed. The results of the second test showed that the sample's absorption spectrum was very similar to the absorption spectrum of hydroxyapatite. This similarity occurs due to the presence of typical hydroxyapatite functional groups in the sample, such as phosphate, calcium, and hydroxyl[5] (Fig. 3)

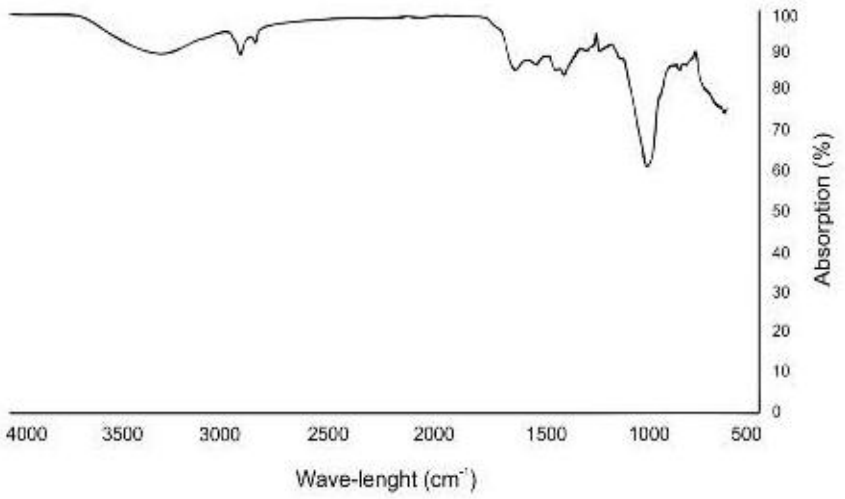

Fig. 3 – Sample’s infrared spectrum.

Discussion

The hydroxyapatite is a natural mineral constituent found in the body, where it represents 30 to 70% of the mass of bones and teeth[6]. According to Urist[7], calcium is the first ion that connects the protein, in the mineralization hypothesis, forming this way the calcified matrix, followed by phosphate uptake, until the formation of calcium hydroxyapatite crystals. The connection between hydroxyapatite crystals and collagen is responsible for bone rigidity and hardness[10].

Hydroxyapatite is abundantly distributed in the spongy bone tissue[6] and is also a typical product of bone neogenesis, a process where the hydroxyapatite degenerates[8]. According to White and Shorts[9], the degradation of hydroxyapatite can occur by reabsorption mediated by osteoclasts or chemical dissolution[8].

The calcifications of hydroxyapatite crystals found in the spongy bone of the axis cervical vertebra probable had formed over the years after the individual's death. This scenario relies on the fact that the protein present in bones is also degraded across time, resulting only in hydroxyapatite - a mineral product that will eventually be reduced to powder[11].

Conclusion

The present study aimed to report a case where classifieds foreign bodies were observed inside the spongy bone tissue of an axis cervical vertebra of a skeleton from the Contemporary Human Bone Collection (LAOF/UFPE). Morphological characteristics of skeletons are regularly employed to discover critical variables of an individual in criminal investigations, such as sex and height, and as teaching tools. Individualizing characters are weapons widely adopted by forensic areas and are extremely important for solving crimes since they help to define characteristics of the biological profile. Besides, the detailed analysis of the skeleton and the foreign bodies found are of great value for the forensic sciences, as they attach extra theoretical and scientific information to professionals of the field.

Conflict of interest

There are no known conflicts of interest associated with this publication, and there has been no significant financial support for this work that could have influenced its outcome.

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