THE USE OF MULTIPLE LINEAR REGRESSION METHOD TO ESTIMATE AGE BY MEASUREMENTS IN DRY ADULT HUMAN SKULLS

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Abstract

Methods for human identification through characteristics such as age have been developed throughout the years and are object of investigation of Forensic Anthropology and Legal Dentistry. Objective: this study aimed at proposing a new method for age prediction using human dry skulls. Material and Methods: seven linear measurements and one rectangular geometrical area were performed in 299 dry skulls (181 male and 118 female) of individuals with age ranging between 18 and 95 years. Measurements were performed by well trained examiners. Results: Qui-square test demonstrated statistical difference (p<0.01) regarding age proportion. Pearson Correlation Coefficient did not evidenced association between age and the other variables. However, association of age with 3 measurements (ANS-LPLMP, ANS-LPRMP e IMD) was observed (p<0.05), making possible to apply multiple linear regression model to predict age by means of cranial measurements. Conclusion: the method proposed in this study is not exact for age estimation. Moreover, it was not observed association regarding age and the variables examined.

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Introduction

The forensic anthropology and forensic dentistry use bones and teeth for estimating characteristics of unidentified persons. Individual identity is an immutable condition and can be used for court, civilian or criminal purposes. Identification should be practiced by means of scientific international established methods, considering the available resources, and staff efficiency and training. Due to circumstantial peculiarities inherent of anthropologic identification, these procedures should be guided by unity immutability and perenicity.[1]

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In the field of Forensic Dentistry, age determination is a relevant aspect for individual identification. Firstly is determined skull phase: infant, adult or senior. Then, age is prediction is refined by evaluation of teeth condition, alveolar bone and ossification stage of espheno-occipital sincondrose and cranial sutures. Considerations of mandibular development is also taken.[2,3]

Investigation of chronological stage of teeth eruption is a valuable tool for age prediction. Moreover, evaluation of other characteristics of craniofacial complex, such as, pulp chamber area, tooth crown and root mineralization and condition of sinphisis and sincondres of mandibular bone and metopic suture of frontal bone contribute for individual identification.[4,5]

Tooth formation evaluation is considered the best method for age prediction compared to other developmental variables. The relation between teeth occlusion development and individual maturity is object of interest of several study fields such as archeology, anthropology, forensic science, child dentistry and orthodontics.

Tooth development is less affected in cases of hormonal diseases or nutrition problems. Two methods are employed for dental age determination: radiography and clinical observation of tooth eruption. Tooth eruption is relatively a stable event and can be considered a valuable tool for individual age prediction.[6,7]

In order to effectively compare or establish individual characteristics for age estimation, it is necessary to considerably increase the number of measurements and new studies and methods are still anticipated.

Forensic science has improved investigation methodology through the years, providing valuable tools for human identification. In this context, methodological protocols are considered relevant for determination of animal identification, gender, age, stature biotype and particularities.[8]

There is a consensus in the literature about the importance of applying proper methodologies when examining distinct geographic regions, according to population standards. Considering that in Brazil, especially in Bahia state, there is no pure race, because of the intense racial miscegenation throughout 5 centuries, studies are anticipated for human identification in such regions. The present study aims at proposing a new method for age estimation using human dry skulls.

Material and Methods

The sample was composed of 299 dry adult human skulls (181 male and 118 female), with previously known ages ranging from 18 to 95 years. Skulls did not present any damage regarding the structures examined. The sample, properly identified and classified, comprised skulls belonging to the Human Anatomy Lab of Unime University and from the skull collection of the Department of Orthodontics of Bahia Federal University.

For the present study, seven linear measurements and two geometrical areas were performed. For method validation, calibration was set with two experienced examiners. Measurements were accessed 3 times with 15 days interval. For method calibration 20% of the sample was randomly selected and Intraclass Correlation Coefficient (ICC) applied. The examiners were blind regarding the variables measured. For standardization of measurements, a skull fixation device was constructed. The device consisted of skull immobilization screws and sliding magnifying glasses for improving landmark visualization and positioning (Fig. 1A).

For linear measurements obtainment, a digital caliper was used (King tools, China) (Fig. 1B & C). Measurements were obtained by measuring the distance between landmarks previously determined. After collected, measuremts were tabulated in Excel spreadsheet.

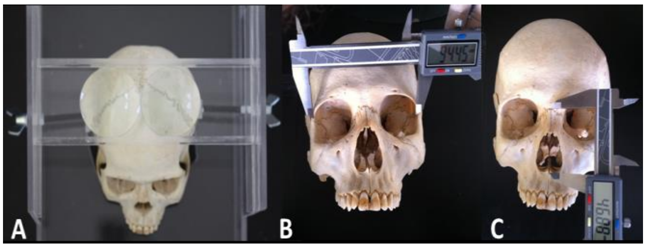

Figure 1. Skull fixation device with two magnifying glasses **(A)**; digital caliper used for measurements **(B & C)**.

For linear measurements and geometrical areas, the following landmarks were determined (Fig. 2):

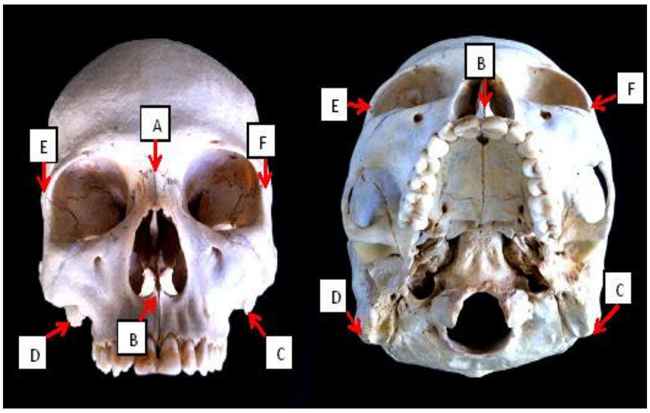

Figure 2. Landmarks used for linear measurements and geometrical area.

A) Nasion (N) - landmark located between internasal and frontonasal suture.
B) Anterior Nasal Spine (ANS) - bony projection located in most Antero-posterior portion of intermaxillary suture.
C) Lower portion of left mastoid process (LPLMP) - located at the lower portion of left mastoid process.
D) Lower portion of right mastoid process (LPRMP) - located at the lower portion of right mastoid process.
E) Orbital right fronto-zigomatic point (ORFZP) - union between frontal bone with zigomatic bone in the most anterior region of fronto-zigomatic suture.
F) Orbital left fronto-zigomatic point (OLFZP) – union between frontal bone and zigomatic bone in the most anterior region of fronto zigomatic suture.

From these landmarks, the following linear measurements were performed:

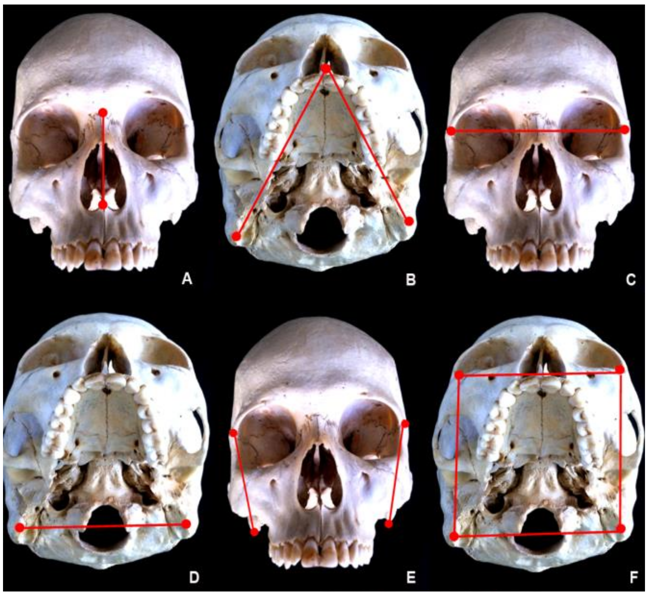

Figure 3. Distance between nasion and anterior nasal spine. **(A)**; Distance between inferior portion of mastoid process and anterior nasal spine (left and right) **(B)**; Distance Inter frontozigomatic **(C)**; Distance between right and left portion of mastoid processes **(D)**; Distance between lower portion of mastoid process and orbital frontozigomatic (left and right) **(E)**; Retangular geometrical area **(F)**.

1) Distance between nasion and anterior nasal spine (N-ANS) (Fig. 3A).
2) Distance between inferior portion of mastoid process and anterior nasal spine (Fig. 3B).
3) Distance between orbital frontozigomatic points (right and left) (Fig. 3C)
4) Distance between right and left portion of mastoid processes (Fig. 3D)
5) Distance between inferior portion of mastoid process and orbital frontozigomatic point (Fig. 3E)

A geometrical area was also determined as follows:

A=Measurement of rectangular geometrical area.

For rectangular geometrical area determination, the following mathematical formula was applied:

$$A = [(B + C) / 2] * [(D + E) / 2]$$

In this formula, the B, C, D, E measurements correspond to:

B=orbital interfrontozigomatic distance

C=distance between right and left inferior portion of mastoid processes

D= distance between left and right orbital frontozigomatic points

E= distance between left orbital fronto zigomatic point and left inferior portion of mastoid process

For the measurement of a two different side rectangular area (A), mean of the two parallel sides were obtained and then multiplied.

For the statistical analysis, the following methods were employed:

1-Descriptive study, contingency table and qui-square test: Descriptive study allowed identification of data characteristics and possible problems before the application of other analytical methods. For the descriptive study, contingency tables were constructed correlation qui-square test was employed.

2-Partial correlation coefficient, multiple linear regression model and stepwise method: After application of qui-square test, partial correlation coefficient was calculated aiming at measuring the association between age and cranial measurements obtained in order to test the hypothesis that the association is valid for the whole population (not casually observed in the present data).

After the correlation tests, linear regression model was performed, obtaining the variable “age” as the answer, cranial measurement as the predictors and gender as the co-variable. The application of this method aimed at developing a model for age prediction based on a optimal set of predictor measurements, selected by the stepwise method.

Results

The present study is based on the collection of data from a sample that it is believed to well represent the population. The method employed in the present study used age category, aiming at evaluating the relative composition of the several studied ages (Table 1)

Age	Frequency	Percentage
18 |– 24	44	14.72
24 |– 30	25	8.36
30 |– 36	23	7.69
36 |– 42	13	4.35
42 |– 48	19	6.35
48 |– 54	26	8.70
54 |– 60	31	10.37
60 |– 66	28	9.36
66 |– 72	33	11.04
72 |– 78	16	5.35
78 |– 84	25	8.36
84 |– 90	10	3.34
90 |– 95	6	2.01

Table 1. Frequency, percentage and qui-square test for proportional equality according to the age range.

Data shown in table 1 indicate strong evidences of proportional differences ($p<0.01$) between ages. Considering positive association between the variables measured and age, Pearson Partial correlation Coefficient was calculated and is shown in Table 2.

Correlation	Pearson (r)	p-value $H_0: \rho = 0$
Rectangular Area	0.07914	0.1730
N-ANS	0.04306	0.4590
ANS-RLPMP	-0.17696	0.0026
ANS-LLPMP	-0.11799	0.0418
IOFZD	0.09969	0.0858
IMD	0.11483	0.0477
LPRMP-OLFZP	-0.00394	0.9460
LPLMP-OLFZP	0.00905	0.8464

Table 2. Pearson partial correlation coefficient and null populational correlation hypotheis test between age and other variables (n=299).

According to data shown in table 2, no evidence of association between age and the five analyzed variables were observed.

In the case of IOFZD, despite no evidence of association with age, p-value (0.0858) was slightly above the significance level preliminary adopted for this study. In this context, it was not possible to assume specifically that existed significant correlation, however, special attention can be attributed to this measurement, since others studies can possible detect it within a significant range.

In addition, it was observed evidences ($p<0.05$) of association between the measurements ANS-RLPMP, ANS-LLPMP and IMD with age.

Once verified the positive linear association with age, it was possible to adjust the multiple linear regression model, allowing age prediction using cranial measurements.

Stepwise method was used for the variable selection. By using this method, the variables that did not contributed satisfactorily for age prediction were abandoned.

Equation	Index
$Age_{Male} = 120.2607 + 0.01027 \times \text{rectangular area} - 1.44201 \times \text{ANS-LPRMP}$	(1)
$Age_{Female} = 126.8585 + 0.01027 \times \text{rectangular area} - 1.44201 \times \text{ANS-LPRMP}$	(2)

Table 3. Equations that compose the multiple linear regression model developed based in the stepwise process of variable selection.

Previous information about gender showed to be important for age prediction accuracy. In this way, the prediction model presented two constants: male (equation 1) and female (equation 2)

ANS and LPRMP presented results statistically significant ($p<0.05$). Moreover, stepwise process showed strong evidence of positive relation between rectangular area and ANS-LPRMP with age.

The determination coefficient (R2) indicated that the model was not exact, representing variation of only 9.77% of age in relation to the studied variables. It is anticipated a method with increased reliability (determination coefficient close to 80%).

Low result of determination coefficient indicated that close to 90% of the age variation was not associated to the selected variables for this study. It is thus concluded that age variation occurred mainly by unknown causes.

Figure 4 illustrates the adjust of the prediction model in relation to the ages examined in the study.

The predicted ages followed a trend; however, the age variation was mainly around the predicted ages. Preferably, the results should be close center to the of the estimated model, thus generating a higher determination coefficient.

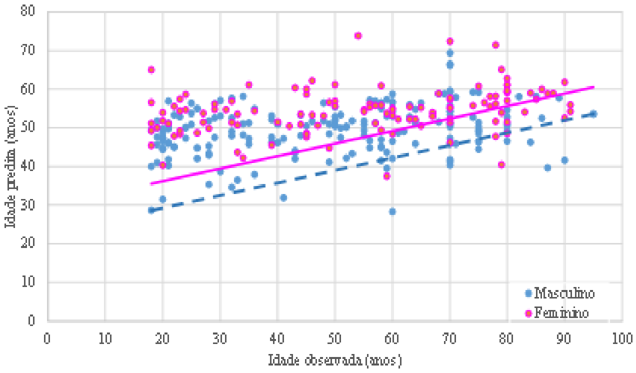

Figure 4. Superimposition of the observed ages (points) and predicted (lines) by multiple regression model.

All calculations were performed using SAS and it was adopted significance level of 5% for all statistical tests. For intraclass (ICC) interpretation, Weir (2005) criteria was used. This criteria is believed to be well accepted with no divergence. Results are shown in table 4.

Measurement	ICC (2.1)	Interpretation
N-ANS	0.99642	Excelent
ANS-RLPMP	0.99902	Excelent
ANS-LLPMP	0.99902	Excelent
FZOD	0.99877	Excelent
IMD	0.99920	Excelent
RPMP – RIZO	0.99851	Excelent
LRMP – LIZO	0.99879	Excelent

Table 4. Intraclass correlation coefficient of the examined variables.

ICC data indicated excellent reliability of the seven measurements examined (Table 4).

Discussion

Over the history, human society has searched for identification of animals and human beings for several purposes.[9] Actually, human identification became fundamental and the contribution from anthropology, legal medicine and legal dentistry studies are crucial, specially as an attestation in court reports[5,10,11,12,13,14]

The methods employed for human identification has varied substantially in terms of complexity. Menezes (2009) has pointed out the importance of microscopic techniques, such as the evaluation of sexual chromatin and Barr corpuscle from dental pulp cells stained by eosin-hematoxilin. Some methods using molecular biology employ DNA markers, gene analysis and Y chromosome markers. Rosing et al. (2007), however, have alerted that sophisticated techniques are not always the ideal method, since the choice depends on the integrity and morphology of the skeleton. For example, molecular techniques are compromised in the presence of fungus, bacteria and substances that can alter the DNA structure.

In the present study, a low cost methodology was tested. The results evidenced a satisfactory degree of reliability compared to previous studies.

The present study followed previous reports of Daruge Jr et al. (1975) and Konigsberg et al. (2009) using bony structures and dental structures for evaluation and application of the suggested method for individual identification, due to the high resistance over time. The present study was performed using dry human adult skulls. It is believed that the structures chosen are resistant and thus reliable for evaluation.

Studies that investigated the relation between gender and age for age prediction using linear measurements of cranial variables are scarce in the literature. Despite not directly related to the methodology used in the present study, Niels (2001) studied the thickness of cranial bones and the possible association of age and gender. The author did not find any significant statistical difference in this regard.

The findings of the present study corroborate with the report from Tomorimitsu et al. (2014).

The authors tested the physical and biomechanical properties of human dry skulls and the association of structural changes that possible occurs over time in both genders. The analytical method used computer tomography of 358 skulls and measured cancellus and cortical bone and used a scale of radiodensity. As reported by Niels (2001), the results did not evidenced statistical significant correlation of the radiodensity and age, or cortical thickness and age.

Willians & Rogers (2006), similarly to the method presented in the present study, evaluated 21 cranial morphologic characteristics considered of high representativeness and did not find any statistical significance association of the variables measured and age or age range. The study sample, however, is considerd small (50 skulls) Similarly, the present study did not find positive association of the trthe rectangular area or ANS(mm), RLPMP-RFZOP (mm) and LLPMP-LFZOP (mm) linear measurements with age. In this context, for this measurements mentioned above, probably it is not reasonable to establish positive association with age. The measurement IFZOD showed the worst results regarding association with age ($p>0.05$).

On the other hand, some studies using the alveolar bone, teeth, espeno-occipital suture, cranial vault and mandible have found positive association of changes in these structures and age. According to the studies, these structures are reliable for age estimation.[2,3]

The accessed literature has evidenced the importance of research in forensic science and that the result of forensic work is still the basis for a sort of investigations. Despite the several existing methods available, employed depending the aspect and conditions of the sample, still remains doubts regarding human identification. In this context, new methods of age estimation are still anticipated, especially in cases of sample diversity. The present study aimed at bringing new insights in terms of identification methodology.

Conclusion

It is thus concluded that the method presented in this study is not accurate for age estimation and that were not observed evidences ($p>0.10$) of positive association between age and variables tested.

Declaration of Interest

The authors report no conflict of interest and the article is not funded or supported by any research grant.

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