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APPLICATION AND ANALYSIS OF THE RISSECH ACETABULAR ADULT AGEING METHOD IN A COLOMBIAN SAMPLE

Vanessa Muñoz-Silva¹, Cesar Sanabria-Medina^{2,3}, Carme Rissech⁴

- 1- Facultad de Ingeniería y Ciencias Básicas, Fundación Universitaria del Área Andina, Bogotá, Colombia.
- 2- Facultat de Medicina, Universidad Antonio Nariño, Bogotá, Colombia.
- 3- Unidad de Búsqueda de Personas Desaparecidas (UBPD) de Colombia
- 4- Facultat de Medicina i Ciències de la Salut, Universitat Rovira i Virgili, Reus, Spain.

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Corresponding author:

Carme Rissech

Dept. de Ciències Mèdiques Bàsiques
Facultat de Medicina i Ciències de la Salut
Universitat Rovira i Virgili
Carrer de Sant Llorenç, 21
43201 Reus (Tarragona)
Spain

Carme.Rissech@gmail.com

Tel: 661142408

ABSTRACT

The classical age indicators of the innominate have been the pubic symphysis and auricular surface. However, recently, the acetabulum has been highlighted as an indicator of adult age, with applicability in young, middle-aged and older adults. The Rissech acetabular method was developed in a Portuguese population and tested in European and European-Americans, giving estimates within 10 year of age in more

than 89% of the sample. The main goal of this paper is to test the Rissech acetabular method in a modern South American sample. The material used for the study was 184 women and 378 men from a Colombian documented skeletal collection. The obtained morphological scores from the acetabulum were analysed through the IDADE2 web page, a Bayesian statistical program that estimates a relative likelihood distribution for the target individuals, produces age estimates, and provides 95% confidence intervals. Results showed this method useful in the modern Colombian population with an average absolute error of 10.63 years in females and 9.44 years in males. These errors are similar to those obtained in other European and North American samples when this method was performed and similar or lower than those obtained when the 3 classical ageing methods (Suchey-Brooks, Buckberry-Chamberlain and Lovejoy) were applied in the same collection (absolute error: 10,29y ♀ and 9,05y ♂ in Suchey-Brooks, 12.5y ♀ and 12.17y ♀ in Buckberry-Chamberlain and 13.54y ♀ and 10.99y ♂ in Lovejoy). Although Rissech's method was developed in a Western European sample, the results of this study indicate its applicability in modern Colombian samples with reasonable accuracy.

Keywords: adult age indicator, age estimation, aging process, acetabulum.

INTRODUCTION

Estimating the age-at-death of adult skeletal remains is one of the most difficult tasks in forensic anthropology. The methods for estimating age in adult skeletal individuals are based on morphological changes in bones and teeth throughout life. The rate and degree of change are determined by a complex set of interactions among genes, culture, and environment that contribute to each individual life history [1,2]. The core to the effective application of a method is the comprehension of the accuracy (correct) and precision (refined) of the method, and its repeatability when applied to

unknown individuals outside of the original reference sample [3,4]. However, the reference samples on which many of the original methods were based are among very few known age-at-death skeletal collections of sufficient sample size for testing purposes [5,6,7]. Documented human skeletal reference samples are scarcer outside of the USA. In addition, the variability observed in the different age indicators increases during age and continues to increase throughout life, which is called the Trajectory Effect [8]. The error committed when applying the existing methods for adult age estimation in the current Colombian population is unknown. This unknown error increases in importance if the high degree of miscegenation of this population is taken into account. The error in age estimation can be quantified only when a method is tested on a contextualized osteological collection or on individuals of known chronological age. A contextualized collection includes known demographic data (sex, age, year of birth, and geographical area) as well as the socioeconomic and temporal context in which the individuals lived [9]. Recently, a Colombian documented osteological collection has been created in the National Institute of Legal Medicine and Forensic Sciences (Instituto Nacional de Medicina Legal y Ciencias Forenses) in Bogota. This collection is the Human Bone Collection of Colombian Reference (Colección Ósea Humana de Referencia Colombiana) which is a documented sample constituted by individuals from the current Colombian population [10]. The existence of this collection opens the door to test current standards in the modern Colombian population.

The most popular methods for adult age estimation are those based on the three articulations of the innominate (pubic symphysis, auricular surface and acetabulum). The acetabulum is the newest age indicator of the three. This new age indicator was proposed at the beginning of the 21st century together with Bayesian inference [11], being both the acetabulum and Rissech's method promising in the adult age estimation field. Rissech's method was developed in a sample of 242 male

individuals from the documented collection of Coimbra in Portugal [11]. This method uses seven acetabular variables (1. acetabular groove; 2. rim shape; 3. rim porosity; 4. apex activity; 5. activity on the outer edge of the rim fossa; 6. activity of the acetabular fossa, 7. porosities of the acetabular fossa) for adult age estimation. Each of the seven acetabular variables is broken into different states describing the different morphological conditions of the acetabular region (e.g., acetabular groove can be scored as: no groove (0), groove (1), pronounced groove (2), and very pronounced groove (3)). The scored states coming from each of the seven variables result in a combination of seven numbers (e.g., 1,0,2,2,3,2,2) which describes the individual condition of the acetabulum and is used to predict adult age by Bayesian inference. These calculi were done through IDADE2 software, which was designed by Professor George Estabrook [11]. Currently, the old IDADE2 software has been re-written in R and presented as a web page, named IDADE2 web page (<http://bass.uib.es/~jaume/IDADE2/https/index.html>) [12], offering an easy calculation and age estimation because it is now based on Microsoft Windows operating system [12].

In the first study by Rissech and colleagues [11], results from Rissech's method on the Coimbra collection indicated the potential value of the method and applicability of the seven acetabular variables of this method (the difference between known age and estimated age was within 10 years in more than 89% of the individuals). In 2007, the same authors tested the method proposed in 2006 in four documented skeletal collections from Western Europe [2]. The sample consisted of 394 individuals aged between 15 and 99 years old coming from the Coimbra and Lisbon collections from Portugal, the UAB collection from Spain and the St Bride collection from England. Results showed differences between known age and estimated age within 10 years in between 80 to 100% of the individuals, depending on the biological distance between the test population and reference population used. Although there are some authors that have some concerns in the description of variables 6 and 7 [13,14] of Rissech's

method, different studies have demonstrated the applicability of the acetabulum [15-24] as adult age indicator and Rissech's method [20,25,26] to age estimation in young, middle-aged and older adults. In these last studies, Rissech's method performed much better (lower mean absolute errors) than the classical methods (Suchey-Brooks [27], Buckberry-Chamberlain [29] and Lovejoy [28]) and two newer methods (Osborne [30] and Calce [31]) based on the pubic symphysis (Suchey-Brooks), auricular surface (Buckberry-Chamberlain, Lovejoy and Osborne) and acetabulum (Calce) for both an Iberian sample [26] and a European-American sample [20,25]. That is to say, Powanda's study [26] showed mean absolute error (8.53 years) lower than that obtained when Lovejoy (13.88 years), Buckberry-Chamberlain (12.87 years), and Suchey-Brooks (13.98 years) methods were applied in two different Iberian samples. Miranker's study [25] demonstrated that the Rissech method was the most accurate method, with the smallest mean absolute error (8.61 years), compared to those obtained in Osborne et al (15.61 years), Suchey-Brooks (19.27 years), and Calce (13.18 years) methods when applied on the same European-American skeletal collection. At this point, it is necessary to clarify that although Calce's method is based on the acetabulum, it corresponds to a phase system method like that of Lovejoy. In addition, several studies have demonstrated that most of the aspects of acetabular aging correspond to natural aging physiological changes and that the seven acetabular variables are resistant to the effects of bone loss, diffuse idiopathic skeletal hyperostosis (DISH), obesity and physical activity [15,18,23,24,32,]. Thus, the acetabulum appears to be a good indicator of adult age, with applicability in young, middle-aged and older adults.

Therefore, taking advantage of the existence of the Human Bone Collection of Colombian Reference, the main goal of this paper is to evaluate the accuracy of Rissech's method and the acetabular ageing process in the current Colombian population. This method was selected due to its popularity in the forensic and bioarchaeological Colombian contexts and because it is recommended in Colombian

anthropological manuals [33]. In addition, Rissech's method has never been tested in a sample originating from South America, particularly from current Colombian population.

MATERIAL AND METHODS

The skeletal sample

The skeletal material used in the study comes from the Colombian Human Bone Reference Collection (COHRC) of the National Institute of Legal Medicine and Forensic Sciences of Colombia, located in the city of Bogotá in Colombia. This collection originates from three public cemeteries in the city of Bogotá (Central Cemetery, El Paraíso Cemetery Park, Jardines La Inmaculada Cemetery). The collection has a total of 600 individuals born in the 20th century (between 1907 and 1989 and deceased between 2004 and 2008), of whom 194 are female, with an average age of 59.04 years and 406 are male, with an average age of 52.25 years [10]. The age range of these individuals is 18 to 100 years old. These individuals are fully identified with their identity document of the National Civil Registry and the reports of the necropsies in cases of violent death (206 individuals) [10]. Figure 1 shows the birth year distribution in the Colombian Human Bone Reference Collection (COHRC) by sex.

To carry out the present study, females and males with fused acetabula were chosen from the COHRC. Individuals who showed pathologies affecting the acetabulum were excluded, but those with non-inflammatory osteoarthritis or DISH were included. These individuals were included because these conditions are related to age [34]. In total, 562 individuals (184 women and 378 men) aged between 18 and 97 years were analyzed (Table 1). This wide age range was used to illustrate all the morphological changes that occur in the acetabular area during the human life span. Following previous studies [2,11,14-18], the left innominate was scored. However, as differences between right and left acetabulum are negligible [16,19, 25], if the left

innominate was damaged, pathologic, taphonomically altered, or unavailable, the right innominate was evaluated.

Females and males were analysed separately using a sex specific reference sample. This is because, although females and males show the same acetabular ageing pattern [18], which allows us to use the same acetabular variables in both sexes, males age faster than females [18,19,32], because they have a higher ageing rate than females [18,19,32]. Therefore, the female and male Colombian samples were divided randomly into reference samples and test samples. The reference sample was always higher in number than the test sample, in order to catch the maximum information possible on ageing variability in the reference sample. Consequently, the 184 Colombian females were randomly distributed between 114 reference sample and 70 test sample individuals, and the 378 Colombian males were randomly distributed between 210 reference sample and 168 test sample individuals (Table 1).

Measurement constancy

To carry out this study the acetabular adult ageing method of Rissech [11] was applied. The seven variables of this classical acetabular method are the following: (1) acetabular groove, (2) acetabular rim shape, (3) acetabular rim porosity, (4) apex activity, (5) activity on the outer edge of the acetabular fossa, (6) activity of the acetabular fossa, and (7) porosities of the acetabular fossa. The variables of this method were described, illustrated and evaluated extensively by Rissech et al. [2,11] and San-Millán et al. [19], particularly, the intra and inter-observer error [2,11,19]. These studies were particularly interested on the inter-observer error and the utility of the descriptions and photos of the seven variables. To carry out this last analysis, in each study 38 innominate [2,11,19] were observed, under identical conditions and using only the descriptions and photos by three different observers with different osteological experience [2,11,19]. One of the observers was a Ph.D student of Zooarchaeology, another held a Master's degree in Anthropology, and the third was a

Ph.D expert on innominate [2,11,19]. The intention was to evaluate how an untrained but osteologically competent person could score the traits using only the information given in descriptions and photos of the variables. The constancy among the three data sets was evaluated using Friedman's test [2,11] and Weighted Kappa statistic for ordinal data [19]. In all these studies, none of the seven variables showed significant intra- and inter-observer differences between different observers or times [2,11,19], indicating that the seven variables had states and descriptions that can be consistently observed by an untrained but osteologically competent person [2,11,19]. However, and in spite of these analyses, because the observations of the present study were exclusively undertaken by the first author of this paper (V.MS), and because the measurement error can be an important source of variation affecting age estimation and lead to biased estimations [35,36], she (V.MS) wanted to evaluate the measurement error committed during her assessment. Therefore 12 innominate were randomly chosen and observed twice, at different times, three months apart, by the same observer (V.MS). The constancy of observations was evaluated by the Wilcoxon's test, a non-parametric test useful to compare two related samples [37].

Age estimation based on Bayes inference

The age estimation was performed by applying Bayesian inference, as used by Rissech et al. [2,11]. Further details of this methodology are entirely explained by Lucy et al. [38] and Rissech et al. [11,12]. To carry out this age estimation we used the IDADE2 web page (<http://bass.uib.es/~jaume/IDADE2/https/index.html>), which is the original IDADE2 software re-written in R and presented as a web page freely accessible [12]. As in the original, this web page uses Bayesian inference to estimate the age of unknown individuals. As in the old IDADE2, in the new IDADE2, prior probability (the probability that the age at death of an unknown individual falls in an age class before any acetabula have been evaluated) is estimated as the fraction of individuals in the reference collection with known age at death in that age class. Posterior probability (the

probability that the age at death of an unknown individual falls in an age class after the acetabula have been evaluated) is based on conditional probability distributions of age (class) at death, given that a particular set of features has been observed in the test specimen [11,12,38]. These distributions were estimated based on the frequencies observed in the reference collection. The underlying assumptions of these calculi are: (1) different variables give independent information about age and (2) test individuals are at least 19 years old (the youngest individual of our sample) and are drawn from a population with similar survivorship to the reference collection. Results in age estimation are reported in a probability distribution over 5-year wide age intervals (15–19, 20–24, etc). In addition, a single-year estimate of age at death is calculated as the expected value of this distribution, attributing to each age class its central age.

Statistical analysis

To evaluate the success of Rissech's method performance in the Colombian collection, the average values of bias and absolute error of the age estimates were calculated and evaluated. Both parameters estimate the method's inaccuracy [39], considering the difference between the estimated age and the chronological age. Specifically, the bias was calculated as the average difference between the estimated age provided by the method and the chronological age ($\Sigma(\text{estimated age} - \text{chronological age})/n$). This identifies the direction of the difference between both the estimated and chronological ages indicating whether the age is over- (positive value) or underestimated (negative value) [7,40,41]. Absolute error was calculated as the average absolute difference between the estimated age and chronological age ($\Sigma|\text{estimated age} - \text{chronological age}|/n$). This identifies the degree of committed error. This parameter does not take into account the sign of the difference (positive or negative) between estimated age and chronological age. To analyse possible sex differences in bias and absolute error a Student-T test was applied for each age group

(< 40 years, 40–64 years, and > 65 years), following San-Millán methodology [18-20]. These age groups were used to follow the same methodological process followed by San-Millán et al. [18-20] in order to facilitate comparisons and to increase the number of individuals in the different age intervals for statistical purposes. All the statistical analyses were performed by SPSS 15.0 software.

RESULTS

None of the 7 variables showed significant intra-observer differences, suggesting a substantial agreement between both distant (three months) observations (V1: $Z=-0.753$, $p=0.450$; V2: $Z=-0.966$, $p=0.334$; V3: $Z=-0.447$, $p=0.655$; V4: $Z=-1.134$, $p=0.257$; V5: $Z=0.000$, $p=1.000$; V6: $Z=-0.333$, $p=0.739$; V7: $Z=-0.707$, $p=0.480$).

Table 2 shows detailed bias and absolute error values of each 10-years-age-intervals and the number of individuals “Not estimated” for Colombian females and males test samples when Rissech’s method was performed based on the Colombian sex reference samples. The individuals not estimated are those which showed a combination of the seven numbers which had an extremely low frequency in the reference sample, impeding the calculus of age probability of these individuals. This is the reason that reference samples need to be bigger than test samples when Bayes inference is utilised. The objective is to catch the maximum information possible on ageing variability in the reference sample. This table 2 also shows detailed results of 5, 10 and 20 years of absolute error, indicating the existence of overestimation (+1), infraestimation (-1) and perfect estimation (0). Results show that either underrepresentation or not representation for the first years intervals are observed, particularly in females (Table 2). This is due to the well-known scarcity of young individuals in documented collections. Results indicate that 27% (17/63) females and 39.2% (62/158) males were estimated with an absolute error equal or lower than 5 years. They also indicate the 53.1% (36/63) females and 66.5% (105/158) males were estimated with an absolute error equal or lower than 10 years (Table 2) and that the

88.9% (56/63) females and 91.1% (144/158) males were estimated with an absolute error equal or lower than 20 years (Table 2).

Regarding the bias and taking into account the total test sample (Table 2), females have a mean positive value slightly higher than 2 years (2.5 years, overestimation) and males have negative mean value close to -1 (-0.89 years, underestimation), see Table 2. The results in bias through the 10-years-age-intervals, in females present a mixed pattern of over and underestimation, showing stages of overestimation (20-29y and 40-79y) and underestimation (30-39y and $\geq 80y$), see Table 2. In males, this bias pattern is simpler; it shows overestimation until 29 years of age and underestimation between 30-39 and ≥ 50 years of age. However, a deeper and more detailed look at the values of bias for each female (Figure 2) and male (Figure 3) shows that, in general, overestimation predominates in females up to approximately 74 years and in males up to approximately 65 years of age. From these ages (74 in females and 65 in males) underestimation predominates in both sexes.

Regarding the mean absolute error, taking into account the total test sample, females have a value of 10.63 years and males a value of 9.44 years (Table 2). Figures 4 and 5 show the absolute error values for each female (Figure 4) and male (Figure 5). This parameter in females seems more constant (Figure 4) compared to males (Figure 5). In males this parameter has a tendency to increase with age (Figure 5). The females with best estimates were those younger than 39 years of age (Table 2) and those older than 90 years of age, followed by those between 70 and 79 years of age, with absolute errors lower than 9 years. The females with worst estimates were those between 40 and 69 years old and between 80 and 89 years old, with absolute errors higher than 10 years (Table 2). In males, this pattern of age estimation observed in females is also repeated, where the individuals with the best estimate of age are those younger than 39 years old and those who are between 70 and 79 years old, with absolute errors lower than 6 years (Table 2). However, in this case, the males older

than 90 years of age are also poorly estimated, with an absolute error of 16.16 years (Table 2).

Tables 3 and 4 show the results on sex differences of bias and absolute error obtained in the performance of Rissech's method in the Colombian test sample based on the Colombian reference sample. In this analysis we considered the age intervals proposed by San-Millán et al. [20]. As has been explained previously in the Material and Methods section, these intervals consist of young individuals (18-39 years), matures (40-64 years) and senile (≥ 65 years). They are mainly useful for comparative and statistical purposes, increasing the number of individuals per age interval. As in the previous analysis of this study, in this section, results indicate that males have more precise age estimations than females (Tables 3 and 4), showing lower values of bias (considering the total sample: 2.5y in females and -0.89y in males) and absolute error (considering the total sample: 10.63y in females and 9.44y in males) than females. These differences are true, except in the age interval of senile individuals (≥ 65 y), where females (bias: -2.57y; absolute error: 10.99y) have lower values of bias and absolute error than males (bias: -9.10y; absolute error: 11.29y). However, in bias, these differences are only statistically significant in senile individuals (≥ 65 y), see Table 3. In absolute error these differences are significant in individuals younger than 39 years of age, where the value of the "p" is near to the significance "0,052" (Table 4). When the total sample is considered, these sexual differences are not statistically significant neither in bias nor in absolute error (Tables 3 and 4).

DISCUSSION

Although the acetabular method of Rissech was developed based on a Portuguese male sample, the age estimations performed in the females and males coming from the Colombian collection (COHRC) were much more accurate (mean absolute error: 10.63y in females and 9.44y in males) than those obtained by the two classical age markers (pubic symphysis and auricular surface) through the methods of

Suchey-Brooks [27], Lovejoy [28] and Buckberry-Chamberlain [29] when applied in the same Colombian collection (Suchey-Brooks: 12.62y in females and 9.17y in males; Lovejoy: 13.54y in females and 10.99y in males and Buckberry-Chamberlain: 12.15y in females and 12.17y in males) by the same first author [42] of this paper. In addition, the results of the present study are also more or equal accurate than those obtained by these three classical methods when applied by other authors in other populations such as Great Britain (Buckberry-Chamberlain: 10.56y in females and 9.67y in males in [29]; 9.8y for both sexes in [39]), Italy (Buckberry-Chamberlain: 13.9y in females and 11.8 y in males in [43]. Lovejoy: 11.6y in both sexes in [44]; and 12.8y in females and 11.4y in males in [7]. Suchey-Brooks: 10.5y in both sexes in [44]; 13.8y in females and 13.6y in males in [7]), Spain (Buckberry-Chamberlain: 11.24y for both sexes in [4]. Suchey-Brooks: 16.04y in females and 12.87y in males in [4]; 13.8y in females and 13.6y in males in [7]), Greek (Buckberry-Chamberlain: 11.18y in females and 11.72y in males in [45]), European American (Buckberry-Chamberlain: 12.38y in females and 13.39y in males in [46]. Suchey-Brooks: 17.57y in females and 20.3y in males in [25]) and Thai (Lovejoy: 16.4y in females and 14.2y in males in [47]. Suchey-Brooks: 16.4y in females and 14.2y in males in [47]). One might think that the results of the present study were to be expected for a Colombian sample due to the historical and biological relationship between Colombia and the Iberian Peninsula. Nevertheless, both populations are quite morphologically and genetically different due to the strong native (Amerindian) and a slightly Sub-Saharan components in the Colombian population, and the geographic distance between Colombia (South America) and the Iberian Peninsula (Western Europe), mainly constituted by Portugal and Spain.

In the present study, in general, underestimation in females and over estimation in males (Table 2 and 3) is observed, coinciding with the observations of San-Millán et al. [20]. However, a more detailed observation, through the age ranges (Table 2) and the graphs showing the bias for each individual (Figure 2 and 3), shows that, in general, there is a tendency to overestimate the younger individuals and underestimate

the older ones. In the analysed sample, this change in the bias behaviour (overestimation to underestimation) occurs later in females (~ 74 years) than in males (~ 65 years), coinciding with the observations of other authors [3,4,20,44]. Bias gives us information regarding the error direction of age estimation. This helps us to understand the limitations of the method and the ageing process of the age marker used, in this case the acetabulum. As other authors have demonstrated in different age indicators (for example, Martrielle et al. [44], Rissech et al. [3], San-Millán et al. [4], San-Millán et al. [20]), the bias of the age estimates tends to show positive values (tendency of overestimation in younger individuals) to negative values (tendency of underestimation in older individuals) throughout the life of each individual. The age-point that marks the change between positive to negative values of bias is usually younger in males than in females [18-20], this difference is possibly related to the higher aging rate observed in males in relation to females [32,18,19,20].

The Rissech's original method is based exclusively on males from the Iberian Peninsula [11], mainly Portuguese (Coimbra and Lisbon). In the present study, the age in the Colombian sample has been estimated in both sexes, using the same definitions of the 7 variables described in the male sample by Rissech et al. [11]. However, the calculation of probabilities and age estimation were performed independently for the two sexual series, following the recommendations of San-Millán et al. [18,19] and Mays [32]. According to these two authors, the acetabulum follows the same pattern of ageing in both sexes [18,19], but males have a higher rate of ageing than females [32,18-20]. This implies that the variables of the acetabulum can be applied indistinctly in both sexes, but the estimation of the age should be carried out separately (using separate sex reference samples for female and male test samples) to have better estimations. However, San-Millán et al [19] also demonstrated that even though the sex separation gives absolute errors (7.09y in females and 7.28y in males) lower than those obtained using mixed reference samples (7.15y in females and 7.35y in males), the error differences between both analysis are small (from 7.09y to 7.15y in females

and from 7.28y to 7.35 in males) [19]. In addition, these absolute errors based on mixed reference samples continued to be lower [19] than those obtained when Suchey-Brooks and Buchberry-Chamberlain were applied to the same population i.e., considering both sexes Suchey–Brooks (12.38y in [48]; 14.42y in [26]) and Buckberry–Chamberlain (11.24y in [48]; 14.12 y in [26]). This indicates, that although it is preferable to use reference samples separated by sex, the acetabular method seems to be robust enough to use mixed reference samples for age estimation, if necessary. In addition, the good results (expressed in lower absolute error values than other ageing methods), obtained during the age estimation of the Colombian sample indicate the applicability of Rissech's method in the Colombian population.

The results of the present study show that, in general terms, estimates in the Colombian female series are less accurate than in the Colombian male series, probably due to high morphological variability observed in females [18]. However, it is also necessary consider the possible influence of the smaller sample size of the female series on the accuracy of the estimates compared to the male series. In the present study, sex differences are only statistically significant in bias, specifically in individuals equal or older than 65 years of age. Nevertheless, in both bias and absolute error, there are some age intervals which have "p" values close to significance, indicating possible sexual differences. In particular, these age intervals for bias are 40 to 64 years ($p = 0.059$) and for the absolute error 18 to 39 years of age ($p = 0.052$). These results are in accordance with different authors [4,7,25,29,43,44,47,49] who have indicated the existence of this sexual bias in the accuracy that favours males when applying age estimation methods based on the pubic symphysis, auricular surface and the acetabulum, and that mostly occurs in mature ages.

Despite the geographical distance between the Colombian and Portuguese populations, the results of the present study in the male series are similar to those obtained in the first evaluation of the Rissech's method performed by the authors of the original method on males from 4 Western European populations (Coimbra, Lisbon,

UAB and St Bride), particularly in those obtained in the St Bride collection, an English sample [2]. In fact, the English sample was the most biologically distant of the four samples analysed in this study (Coimbra and Lisbon are from Portugal and UAB is from Spain). In the present study, 66.46% of Colombian males had an absolute error equal or lower than 10 years. In the original study 78% of the sample had an absolute error equal or lower than 10 years when the reference sample used for the estimation was different from that of the test [2]. Specifically, 56% of English males had an absolute error equal or lower than 10 years when the reference sample used for the estimation was from Iberian Peninsula [2]. There are also similarities, related to bias values and the over and underestimation, between the graph of the bias considering each of the Colombian males obtained in the present study and the graph of the bias of English males in the original study (compare Figure 3 of the present study and Figure 4 of the original study [2]). In the Iberian populations (Coimbra, Lisbon, UAB) of the original study, as we have seen, the bias was slightly lower than that obtained for the English sample, as expected. In addition, the mean absolute error values obtained in the Colombian females (10.63y.) and males (9.44y.) of the present study are similar to those obtained during the performance of Rissech's method on the European-American population of USA (San-Millán et al. [20] - 10.53 years in females and 8.14 years in males) or better (Miranker, [25] - 13.78 years in females and 12.8 years in males). Even with worse accuracy for the Colombian female series, the results obtained in this study are reasonable enough to be Rissech's method applied in both archaeological and forensic contexts, especially when we compare them with the results of other authors when applying classical methods based on pubic symphysis and auricular surface age markers of the innominate in European-American population of USA [25,44,46,], England [29,39], Italy [7,43], Spain [4], Greece [45], and Thailand [47]. It is notable that the Rissech's method, when applied to a population different from that used to develop the method, provides equal or better estimates than those provided by classical methods based on the pubic symphysis and auricular surface.

Particularly, if we take into account the existence of population differences and that the classic methods are frequently applied during anthropological routines [50]. Specifically, the mean absolute error of the present study (10.63 years for females and 9.44 years for males) is equal to that obtained by the Buckberry-Chamberlain method by these same authors [29] (10.56 years for women and 9.67 years for men) and similar to that obtained by Falys [39] (9.8 years for both sexes), both methods based on the auricular surface. In addition, the mean absolute errors obtained with the Buckberry-Chamberlain method are higher than those obtained in the present study when the Buckberry-Chamberlain method is applied to populations other than the English population (the population in which this method was developed), such as Mulhern & Jones [46] (12.38 years for females and 13.39 years for males), Hens & Belcastro [43] (13.9 years for females and 11.8 years for males), San-Millán et al. [4] (11.24 years for both sexes), Moraitis et al. [45] (11.18 years for females and 11.72 years for males). Continuing with the auricular surface, and taking into account the Lovejoy method [28], the error of the present study is lower than that obtained by Martrille et al. [44] (11.6 years in both sexes), Hens et al. [7] (12.8 years for females and 11.4 years for males), and Schmitt [47] (18.2 years for females and 13.8 years for males). Regarding the Suchey-Brooks method based on the pubic symphysis [27], the error of the present study is lower than that obtained by Miranker [25] (17.57 years for females and 20.3 years for males), Martrille et al. [44] (10.5 years in both sexes), Hens et al. [7] (13.8 years for females and 13.6 years for males), San-Millán et al. [4] (16.04 years for females and 12.87 years for males), and Schmitt [47] (16.4 years for females and 14.2 years for males). It is also notable that the error of the present study is also lower than that reported by Miranker [25] when she applied Calce's phase system acetabular method [31] (13.78 years for females and 12.8 years for males). All of these facts highlight the utility of the acetabular method of Rissech [11] for age estimation.

The poor preservation of the pubic symphysis due to post-depositional processes decreases its utility in age estimation, enhancing the methods based on the

auricular surface and the acetabulum. On the other hand, the Lovejoy method was developed before the recommendation of using 95% confidence age intervals. He defined a phase system based on the premise of the existence of a constant rate of morphological change related to age and in the existence of little variability in the aging process of the auricular surface between individuals and populations. Calce's method [31] is based on the same premises used by Lovejoy (a constant rate of morphological change related to age), however in this case, using the acetabulum as adult age marker. The methods of Buckberry-Chamberlain and Suchey-Brooks do use 95% confidence age intervals, but these are very wide. For example, phases IV (29–81 years) and V (29–88 years) in the Buckberry-Chamberlain method have amplitude of 52 and 59 years, respectively, covering almost all adult life, leading to very imprecise age estimates [3,4,45,].

One of the greater difficulties in adult age estimation is the estimation of elderly individuals. With the objective of improving the estimates and reducing the effects of the variability of the aging process on the methods of Lovejoy and Buckberry-Chamberlain, Osborne et al. [30] and Falys et al. [39] reduced the number of phases and stages of these two methods. Osborne and collaborators [30] collapsed the eight Lovejoy phases into a six phase system. Falys and collaborators [6] also reduced the seven Buckberry-Chamberlain stages to three. In this way, both authors achieved an increase in the accuracy of both methods, particularly at older ages. However, these two new proposals give very wide age intervals and reflect the general low quality of the information on the aging process contained in the human skeleton. For example, the phase III age range proposed by Falys and colleagues is 21 to 91 years, and the phase 5 and 6 age ranges proposed by Osborne are 24–82 years and 29–89 years, respectively. These two new proposals, together with Suchey-Brooks and Buckberry-Chamberlain, are based on wide age intervals with ranges that include most of the adult ages, facilitating that the chronological age falls within the estimated interval. Thus, these methods sacrifice precision for accuracy. However, both accuracy and

precision are basic to individual identification, and both need to be improved. Acetabulum and Bayesian inference has the potential to improve adult age estimation by providing higher accuracy and precision with smaller estimated age intervals, even for older individuals [2,11,17,19,20, 24]. Another benefit of using the acetabulum as age indicator is its resistance to postdepositional processes [2]. In addition, the features of the acetabular rim, acetabular apex and the acetabulum in general are particularly resistant to medical interventions, obesity, bone loss and physical activity, which is especially important in older individuals [15,24,32]. The changes observed in this joint are mainly physiological changes due to age rather than merely degenerative changes, what makes the acetabulum relevant to adult age estimation.

The Rissech acetabular method offers the user the possibility of choosing the reference sample, allowing a selection of standards that, as far as possible, resemble the material under study in terms of environmental and genetic factors that can affect the indicator-age association. Therefore, the methodology grants method flexibility and greater applicability to diverse populations. All this has been highlighted even more since the IDADE2 web page [12] has existed, which due to its easy use (because is based on Microsoft Windows operating system), facilitates the estimation of age through Bayesian inference, giving a specific estimated age and a confidence interval. The data in this study provides a good argument for increasing the use of acetabular methods in daily routines for adult age estimation in forensic and archaeological laboratories.

However, it is clear that it is necessary to delve more deeply in order to comprehend how the methods based on the acetabulum perform in each different population and why the observed differences between populations occur when they are performed. This will help us to understand the different factors that can influence this age marker. All of this means that it is necessary to delve deeper into the acetabular ageing process. Nevertheless, it is not only necessary to do this with the acetabulum, but also with the other age markers. At the beginning of the 20th century, adult ageing

methods began to be applied in different populations, thinking that the ageing process of the different age markers was universal and that the different ageing methods could be applied homogeneously. However, by the end of the 20th century, it evidenced that the ageing process of the different age indicators were different between populations and individuals. Anthropologists also realized that the life history of an individual can affect the observations of his/her age markers. Therefore, it is necessary to increase our knowledge in adult ageing processes of the different age indicators. In addition, adult age estimation is not a straightforward matter. It needs a great capacity of observation, because the age-related morphological changes of the human skeleton are subtle. These morphological changes are usually small modifications such as porosities, striations, granulations, ridges, furrows, undulations, and roughness, which appear or disappear more or less (much less than more) gradually with age [15]. However, due to bone plasticity, there is an influence of environmental, cultural, and genetic factors that modify the combination of traits and also the morphology of these traits in an individual. They can increase or diminish or create a new expression of some features, which in turn affects estimated age [15]. This means, to have an accurate adult age estimation, morphological age changes need to be precisely and meticulously evaluated by the forensic anthropologist. In fact, as anthropologists, we need specific training in age estimation to be competent in these techniques [51]. We only have to recall the first time that we wanted to apply the classic methods for adult ageing, which at the beginning seemed almost incomprehensible to us. Therefore, it is necessary to increase precisely and meticulously our knowledge of the adult age indicators and their ageing process, and apply the ageing methods consciously.

CONCLUSIONS

The Results of testing Rissech's acetabular method in a documented skeletal sample (184 females and 378 males) of Colombian origin indicated that this method is applicable to the modern Colombian population, in spite of it having been developed on

a Western European population. This method provided better age estimates than the other two age markers (pubic symphysis and auricular surface) of the innominate, currently used in anthropological laboratories. The obtained results are better than or similar to the results obtained by this method in Western European and European-American populations, indicating that the acetabulum and the Rissech's acetabular method can be useful tools for age estimation in different populations. Even though new tests on other populations are necessary, these results point toward a wider applicability of the acetabulum and this methodology, which make the acetabulum a powerful age reference indicator.

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Authors' contributions The study was designed by the three co-authors. The data acquisition, analysis and interpretation of the results was done by Vanessa Muñoz-Silva. The manuscript was written by corresponding author based on a previous version written by the three co-authors, who critically revised it for important intellectual content. All of them approved the version to be published and agree to be accountable for all aspects of the work in ensuring that questions related to the accuracy or integrity of any part of the work are appropriately investigated and resolved.

COMPLIANCE WITH ETHICAL STANDARDS

Conflicts of interest The authors declare that they have no conflict of interest.

Ethical approval The use of skeletal specimens from documented skeletal collections for research upon anonymization is in accordance with local ethical standards and

regulations at the National Institute of Legal Medicine and Forensic Sciences in Bogotá (Colombia).

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FIGURE LEGENDS

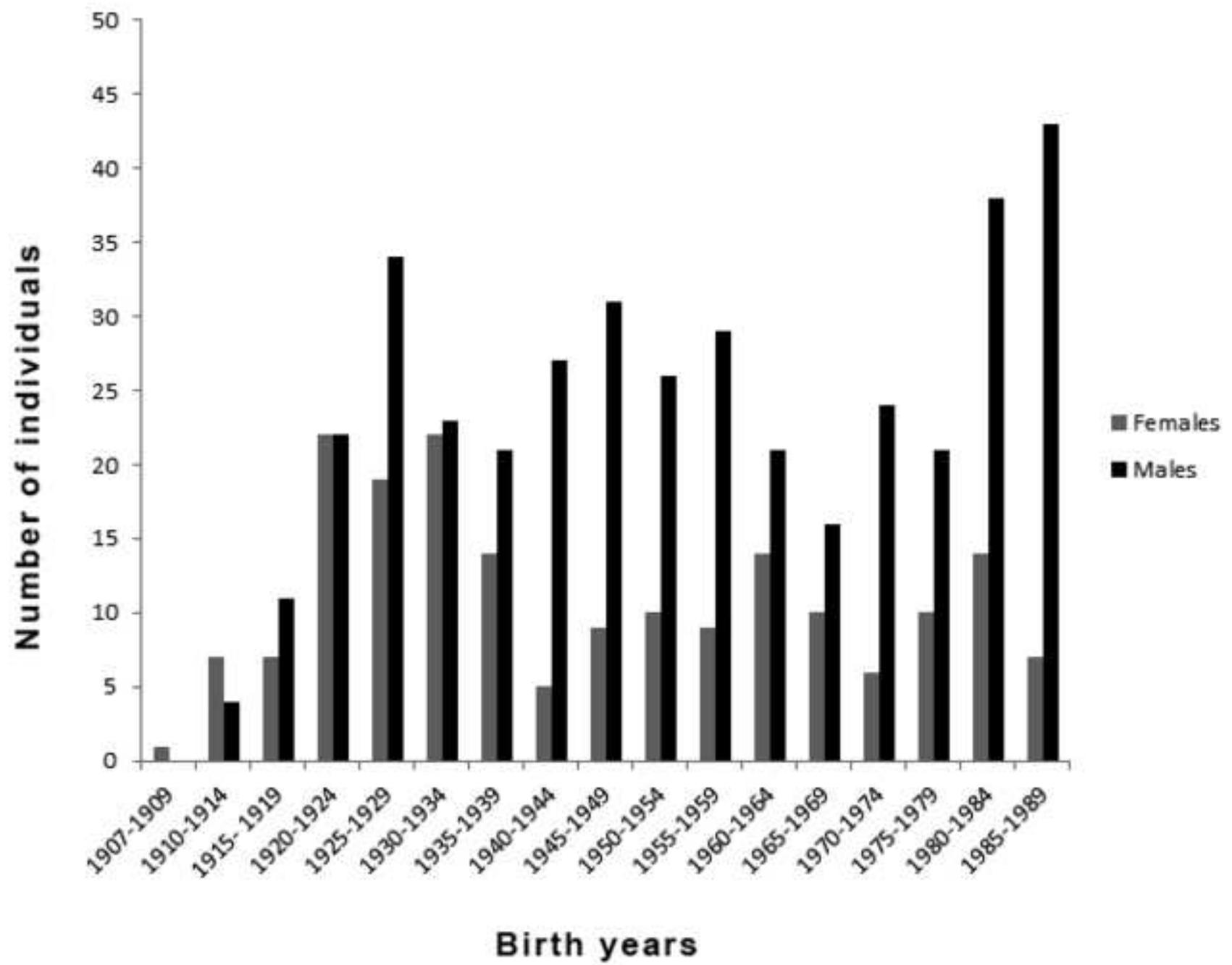
Figure 1. Birth year distribution in the Colombian Human Bone Reference Collection (COHRC) by sex,

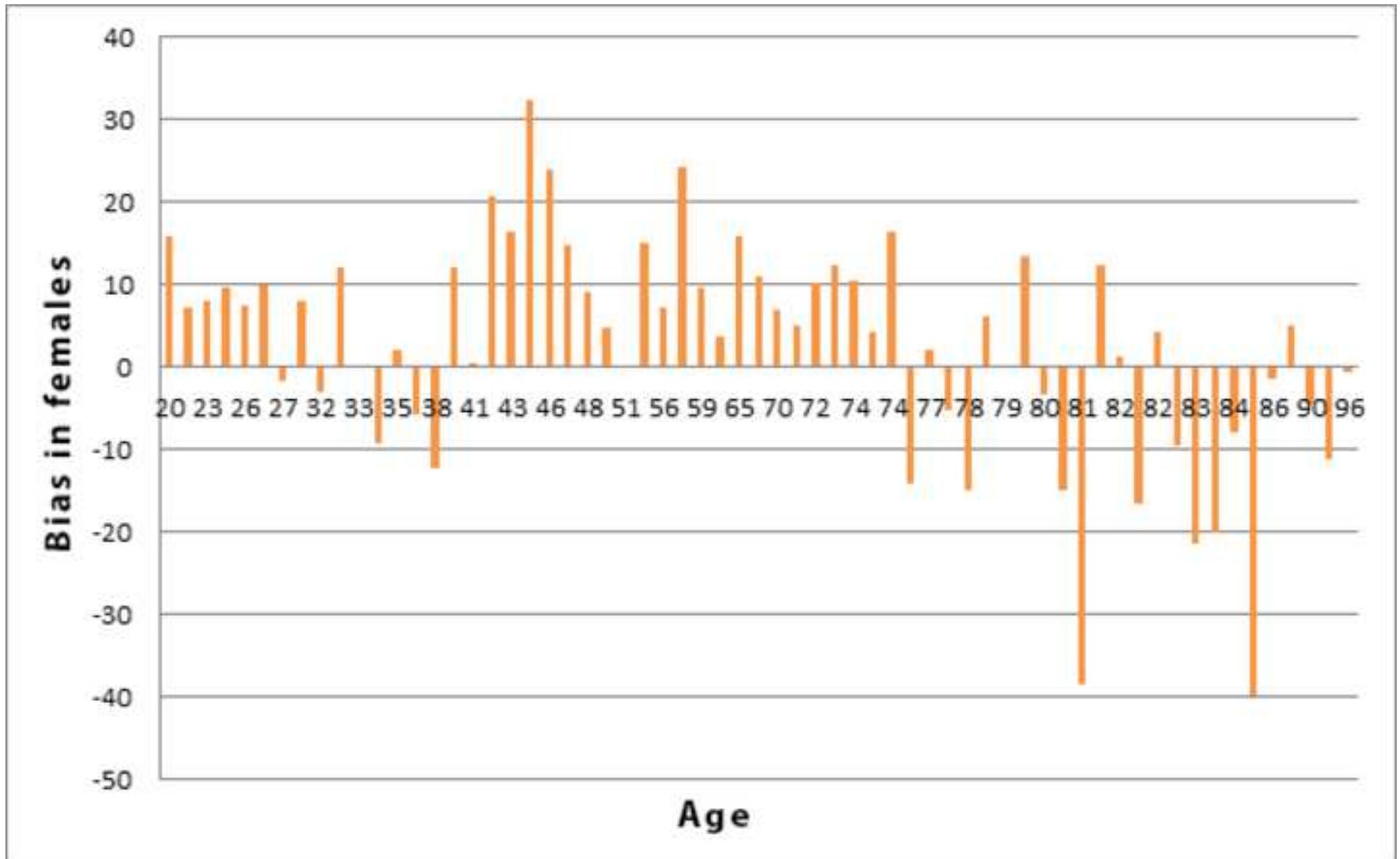
Figure 2. Age bias recoded for each female from the test sample when Rissech's method was performed.

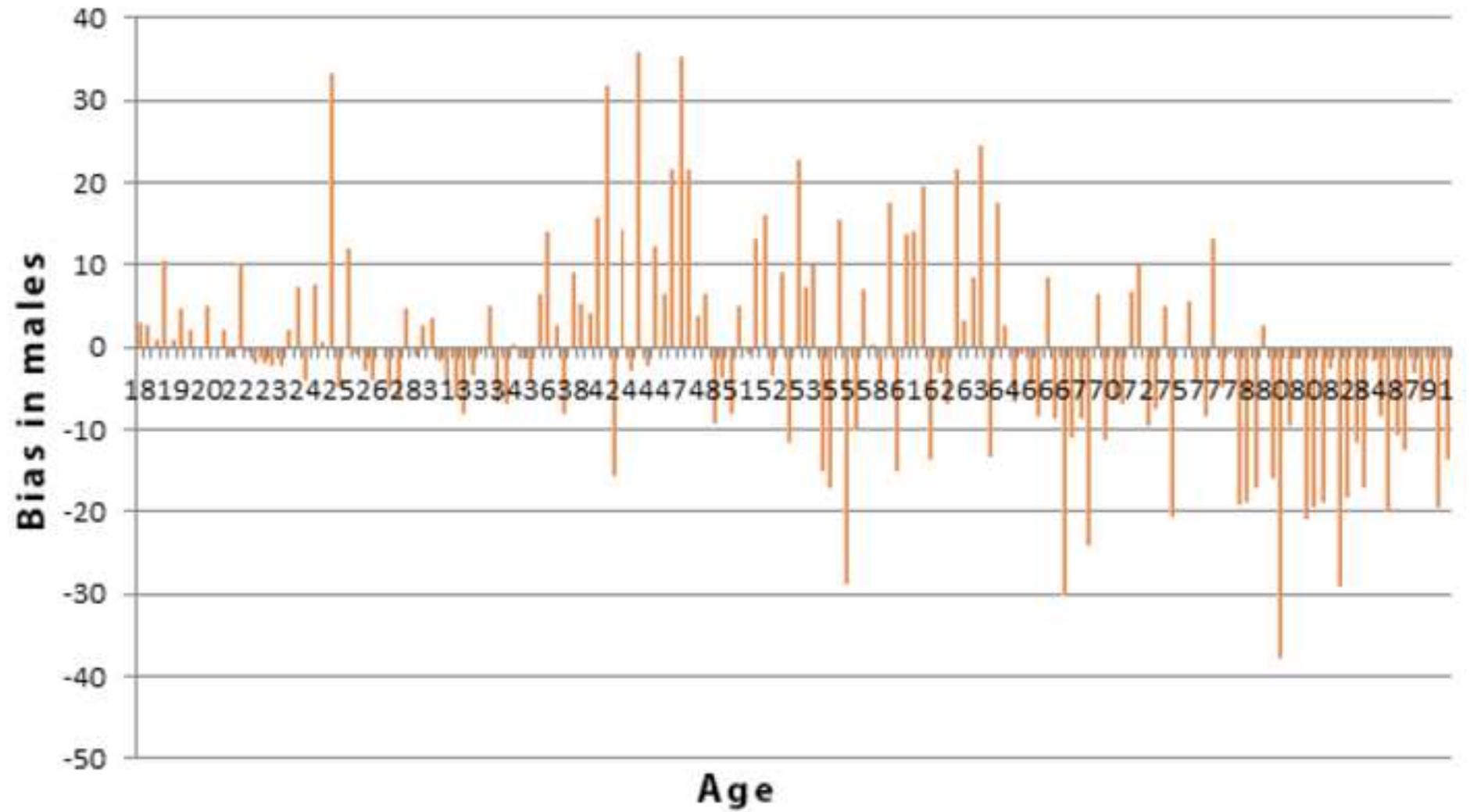
Figure 3. Age bias recoded for each male from the test sample when Rissech's method was performed.

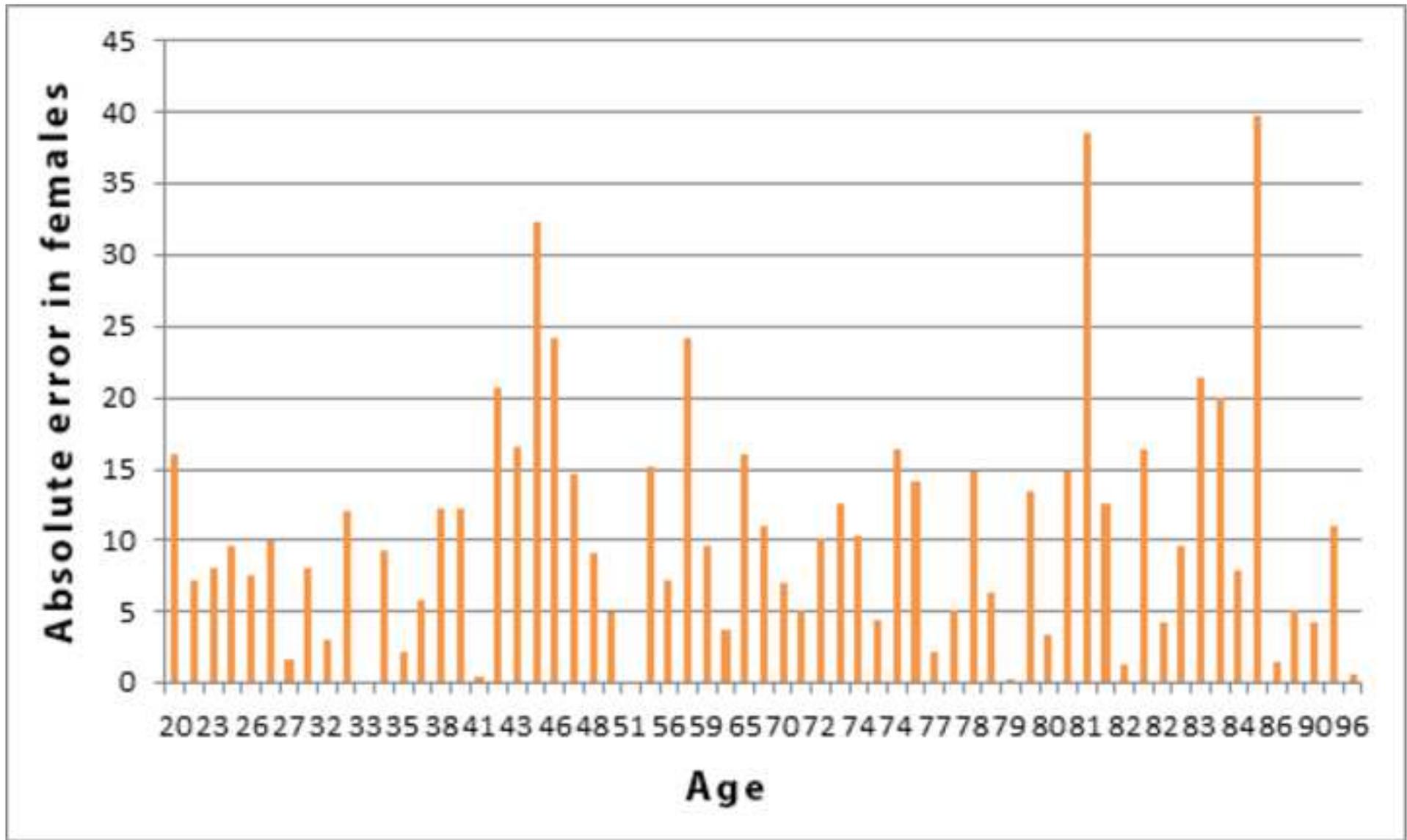
Figure 4. Absolute error for each female from the test sample when Rissech's method was performed.

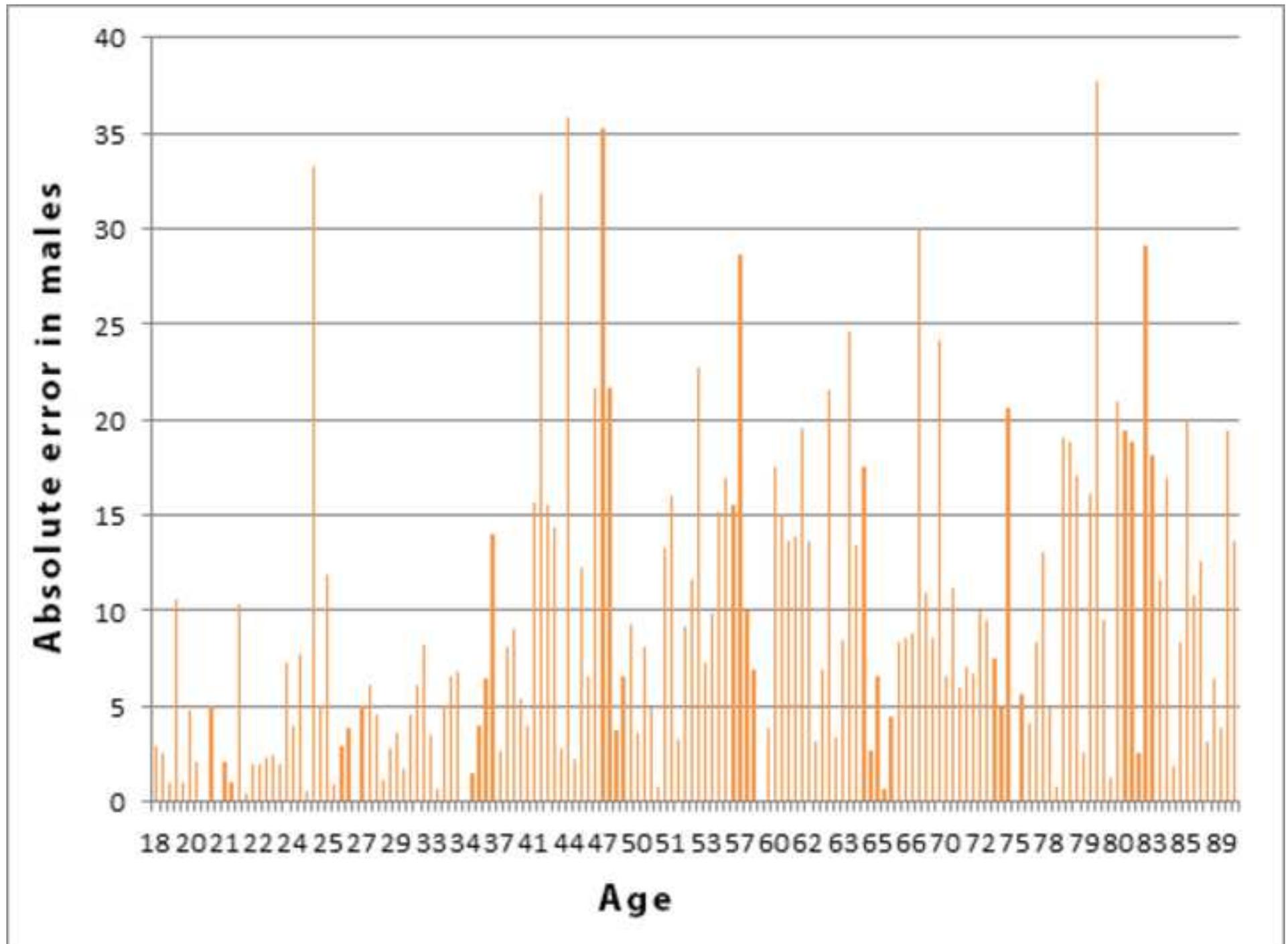
Figure 5. Absolute error for each male from the test sample when Rissech's method was performed.











TABLES

| <i>Age interval</i> | <i>Female</i> | <i>Male</i> |
|-------------------------|---------------|-------------|
| <i>18 - 27</i> | 22 | 76 |
| <i>28 - 37</i> | 21 | 43 |
| <i>38 - 47</i> | 21 | 40 |
| <i>48 - 57</i> | 18 | 51 |
| <i>58 - 67</i> | 15 | 56 |
| <i>68 - 77</i> | 32 | 41 |
| <i>78 - 87</i> | 42 | 57 |
| <i>88 - 97</i> | 13 | 14 |
| <i>Total</i> | 184 | 378 |
| | | |
| <i>Reference</i> | 114 | 210 |
| <i>Test</i> | 70 | 168 |
| <i>Total</i> | 184 | 378 |

Table 1: Distribution of the 562 individuals sampled from the Colombian Human Bone Reference Collection by sex and age group.

| Sex | Age | e ≤ 5 years | | | | e ≤ 10 years | | | | e ≤ 20 years | | | | N | Mean bias | Mean absolute error | Not estimated |
|---------|-------|---------------|---------------|---------------|-------------|----------------|---------------|---------------|--------------|----------------|---------------|---------------|--------------|-----|-----------|---------------------|---------------|
| | | n | - | + | 0 | n | - | + | 0 | n | - | + | 0 | | | | |
| Females | <20 | - | - | - | - | - | - | - | - | - | - | - | - | - | - | - | - |
| | 20-29 | 1 (12.5%) | 1 (100%) | 0 (0%) | 0 (0%) | 7 (19.4%) | 1 (14.3%) | 6 (85.7%) | 0 (0%) | 8 (100%) | 1 (0%) | 7 (100%) | 0 (0%) | 8 | 8.09 | 8.51 | 4 |
| | 30-39 | 3 (42.9%) | 1 (33.3%) | 2 (66.7%) | 0 (0%) | 5 (71.4%) | 3 (60%) | 2 (40%) | 0 (0%) | 7 (100%) | 4 (57.1%) | 3 (42.9%) | 0 (0%) | 7 | -2.29 | 6.37 | 0 |
| | 40-49 | 2 (22.2%) | 0 (0%) | 2 (100%) | 0 (0%) | 3 (33.3%) | 0 (0%) | 3 (100%) | 0 (0%) | 7 (77.8%) | 0 (0%) | 7 (100%) | 0 (0%) | 9 | 15.00 | 15.00 | 0 |
| | 50-59 | 2 (33.3%) | 0 (0%) | 1 (50%) | 1 (50%) | 4 (66.7%) | 0 (0%) | 3 (75%) | 1 (25%) | 5 (83.3%) | 0 (0%) | 4 (80%) | 1 (20%) | 6 | 10.00 | 10.00 | 0 |
| | 60-69 | 0 (0%) | 0 (0%) | 0 (0%) | 0 (0%) | 0 (0%) | 0 (0%) | 0 (0%) | 0 (0%) | 2 (100%) | 0 (0%) | 2 (100%) | 0 (0%) | 2 | 13.50 | 13.50 | 0 |
| | 70-79 | 3 (21.4%) | 0 (0%) | 3 (100%) | 0 (0%) | 7 (50%) | 1 (14.3%) | 6 (85.7%) | 0 (0%) | 14 (100%) | 3 (21.4%) | 11 (78.6%) | 0 (0%) | 14 | 3.88 | 8.77 | 3 |
| | 80-89 | 4 (28.6%) | 2 (50%) | 2 (50%) | 0 (0%) | 7 (50%) | 4 (57.1%) | 3 (42.9%) | 0 (0%) | 10 (71.4%) | 6 (60%) | 4 (40%) | 0 (0%) | 14 | -10.73 | 14.06 | 0 |
| | ≥90 | 2 (66.7%) | 1 (50%) | 1 (50%) | 0 (0%) | 3 (100%) | 2 (66.7%) | 0 (0%) | 1 (33.3%) | 3 (100%) | 2 (66.7%) | 0 (0%) | 1 (33.3%) | 3 | -5.33 | 5.33 | 0 |
| | Total | 17 (27%) | 5 (29.4%) | 11 (64.7%) | 1 (5.9%) | 36 (53.1%) | 11 (30.6%) | 23 (63.9%) | 2 (5.6%) | 56 (88.9%) | 16 (28.6%) | 38 (67.9%) | 2 (3.5%) | 63 | 2.52 | 10.63 | 7 |
| Males | <20 | 4 (80%) | 0 (0%) | 4 (100%) | 0 (0%) | 4 (80%) | 0 (0%) | 4 (100%) | 0 (0%) | 5 (100%) | 0 (0%) | 5 (100%) | 0 (0%) | 5 | 3.62 | 3.62 | 0 |
| | 20-29 | 24 (37.4%) | 13 (54.2%) | 9 (37.5%) | 2 (8.3%) | 28 (25.8%) | 15 (53.6%) | 11 (39.3%) | 2 (7.1%) | 30 (100%) | 15 (50%) | 13 (43.3%) | 2 (6.7%) | 31 | 1.90 | 4.43 | 4 |
| | 30-39 | 8 (44.4%) | 6 (75%) | 2 (25%) | 0 (0%) | 17 (94.4%) | 11 (64.7%) | 6 (35.3%) | 0 (0%) | 18 (100%) | 11 (61.1%) | 6 (33.3%) | 1 (5.6%) | 18 | -0.50 | 5.26 | 0 |
| | 40-49 | 4 (26.7%) | 2 (50%) | 2 (50%) | 0 (0%) | 6 (40%) | 2 (33.3%) | 4 (66.7%) | 0 (0%) | 10 (66.7%) | 3 (30%) | 7 (70%) | 0 (0%) | 15 | 12.59 | 15.34 | 1 |
| | 50-59 | 6 (28.6%) | 4 (66.7%) | 2 (33.3%) | 0 (0%) | 13 (61.9%) | 7 (53.8%) | 6 (46.2%) | 0 (0%) | 20 (95.2%) | 11 (55%) | 9 (45%) | 0 (0%) | 21 | -0.26 | 10.36 | 2 |
| | 60-69 | 5 (20%) | 3 (60%) | 2 (40%) | 0 (0%) | 12 (48%) | 8 (66.7%) | 4 (33.3%) | 0 (0%) | 22 (88%) | 13 (59.1%) | 9 (40.9%) | 0 (0%) | 25 | -0.13 | 12.27 | 1 |
| | 70-79 | 6 (28.6%) | 4 (66.7%) | 2 (33.3%) | 0 (0%) | 15 (71.4%) | 9 (60%) | 6 (40%) | 0 (0%) | 20 (95.2%) | 13 (61.9%) | 7 (100%) | 0 (38.1%) | 21 | -4.07 | 8.79 | 2 |
| | 80-89 | 5 (25%) | 5 (100%) | 0 (0%) | 0 (0%) | 8 (40%) | 8 (100%) | 0 (0%) | 0 (0%) | 17 (100%) | 17 (100%) | 0 (0%) | 0 (0%) | 20 | -13.47 | 13.47 | 0 |
| | ≥90 | 0 (0%) | 0 (0%) | 0 (0%) | 0 (0%) | 2 (100%) | 2 (100%) | 0 (0%) | 0 (0%) | 2 (100%) | 2 (100%) | 0 (0%) | 0 (0%) | 2 | -16.16 | 16.16 | 0 |
| | Total | 62 (39.2%) | 37 (59.7%) | 23 (37.1%) | 2 (3.2%) | 105 (66.5%) | 62 (59.1%) | 41 (39%) | 2 (1.9%) | 144 (91.1%) | 85 (59%) | 56 (38.9%) | 3 (2.1%) | 158 | -0.89 | 9.44 | 10 |

Table 2. Validation of Rissech's method on Colombian test sample. The table is divided into absolute error less or equal than 5 years ($|e| \leq 5$ years), 10 years ($|e| \leq 10$ years) and 20 years ($|e| \leq 20$ years). The latter ($|e| \leq 10$ years and $|e| \leq 20$ years) include $|e| \leq 5$ years and the $|e| \leq 10$ years results, respectively. "n" indicates the number of individuals infraestimated, overestimated or perfectly estimated of each age interval (-, +, and

0, respectively). "N" indicates the total number of individuals estimated in each age interval. "Not estimated" corresponds to the number of individuals in which was not possible estimate the age in each age interval. Perfectly estimate means that the estimated age coincides with the chronological age of the individual. Mean bias and mean absolute error include all the estimated individuals (individuals with absolute error higher than 10 years are also included).

| Age | Females | | | Males | | | t | p |
|-------|---------|-----------|-------|-------|-----------|-------|--------|--------|
| | n | Mean bias | SD | n | Mean bias | SD | | |
| 18-39 | 15 | 3.25 | 8.28 | 54 | 1.26 | 6.68 | -0.953 | 0.340 |
| 40-64 | 15 | 13.00 | 9.40 | 51 | 5.38 | 14.45 | -1.921 | 0.059 |
| ≥ 65 | 33 | -2.57 | 14.25 | 53 | -9.10 | 10.60 | -2.430 | 0.017* |
| Total | 63 | 2.52 | 13.44 | 158 | -0.89 | 12.51 | -1.790 | 0.075 |

Table 3. Sex differences in bias obtained in the Colombian test sample using as reference the Colombian reference sample. Student T test was applied in the total sample and each age interval. SD means standard deviation.

| Age | Females | | | Males | | | t | p |
|-------|---------|---------------------|------|-------|---------------------|------|--------|-------|
| | n | Mean absolute error | SD | n | Mean absolute error | SD | | |
| 18-39 | 15 | 7.51 | 4.40 | 54 | 4.63 | 5.12 | -1.982 | 0.052 |
| 40-64 | 15 | 13.00 | 9.40 | 51 | 12.61 | 8.74 | -0.151 | 0.880 |
| ≥ 65 | 33 | 10.99 | 9.24 | 53 | 11.29 | 8.17 | 0.161 | 0.873 |
| Total | 63 | 10.63 | 8.50 | 158 | 9.44 | 8.23 | -0.969 | 0.333 |

Table 4. Sex differences in absolute error obtained in the Colombian test sample using as reference the Colombian reference sample. Student T test was applied in the total sample and each age interval. SD means standard deviation.