Forensic Anthropology Population Data

Assessment of second (I₂M) and third (I₃M) molar indices for establishing 14 and 16 legal ages and validation of the Cameriere's I₃M cut-off for 18 years old in Chilean population

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A B S T R A C T
This retrospective cross-sectional study has two-fold aims: the first is to assess new cut-offs at the legal age thresholds (LATs) of 14 and 16 years old and the second is to validate the cut-off of third molar index I₃M = 0.08 for 18 years of age in Chilean people. Orthopantomographs from 822 Chilean children aged from 11 to 22 (472 girls and 350 boys) were analysed. For LAT of 14 years, cut-offs were found using the ROC curves singly for boys and girls. The cut-offs for boys were I₂M = 0.16 and I₃M = 0.73 while for girls we obtained I₂M = 0.10 and I₃M = 0.77. For LAT of 16 years we obtained the same cut-offs regardless of gender, which were 0.06 and 0.36 for I₂M and I₃M respectively. Concerning the validity of I₃M cut-off for 18 years old in Chilean population, the proportion of correctly classified individuals was 83% and estimated post-test probability, PPV, was 93.2%, with a 95% confidence interval equals to 91.3%, 94.6%. Hence, the probability that a subject positive on the test was 18 years of age or older was 93.2%, confirming the validation of the I₃M cut-off for Chilean population.

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1. Introduction

Rights and duties of a person in society are granted according to their age as stated in the Convention of the Rights of the Child (CRC). A child is “every person below the age of 18 years unless under the law applicable to the child, majority is attained earlier” which leaves the possibility open that in certain circumstances a child can attain the status of adult before reaching 18 years of age [1]. Several countries have different age limits to assign a person certain rights such as the right to vote, to give sexual consent, to work, to drink, to drive and so on. The minimum age of criminal responsibility (MACR) around the world goes from 6 to 18 years, being a minor or an adult in the eyes of the law will make a difference from how the subject will be prosecuted and convicted to the type of facility that will be assigned in case of incarceration [2]. One of the most challenging situations in the judicial system is to establish the real age of a person in circumstances where there are no reliable documents. The high rate of illegal immigration in Europe has made this problem more apparent [3], however this issue also exists in regions of South America and Asia for different reasons. In countries like Colombia and Chile, regional situations like paramilitary presence and the history of dictatorships has created problems registering both living and dead people. In recent years, registration has improved in countries like Brazil but continue to be deficient [4,5]. Chilean laws are complex when it comes to determining the specific age of responsibility since the implementation of the Law of Adolescent’s Criminal Responsibility, or LRPA by its Spanish acronym. In 2011, the National Congress of Chile ratified a new law and fixed two-age thresholds for 14,16 and
18 years old [6]. A subject 14–16 years can only receive a maximum penalty of 5 years. Someone younger than 18 but older than 16 can receive a maximum penalty of 10 years, while full criminal responsibility is attained at 18. In other matters, marriage can be granted at 16 with the consent of a legal guardian, a person cannot work before 18 but some can work at 15 depending on the type of work, consensual sex can be legal for heterosexual couples aged 14, but homosexual couples cannot give consent until 18 [7]. Chile has 100% birth registration, but has problems with illegal immigration, particularly from Peru, with a certain percentage through fake documents. To confront this situation, we should use a method to establish age through biological markers that are associated with those ages relevant to the law in each country. For age estimation in a subject with unknown date of birth, the usual procedure followed in the forensic field is to assess social parameters and physical developmental charts. Since the first of these has not been scientifically proven, and the last one has been demonstrated to be largely influenced by external environment, the use of teeth has become more reliable when estimating age [8]. Several researchers have developed scoring systems through the observation of dental X-rays, using dental calcification stages of permanent teeth [9,10]. The most known method and also used in Chile for estimating age in the living is Demirjian method [10,11], however the use of stages involves a high subjectivity in the appreciation and assigning of the tooth developmental stage. Our research group has been proving several methods for age estimation using orthopanoramic dental radiographs that have been tested in different populations with positive results. They have proposed the possibility of using a statistical cut-off to estimate 18 years old by studying and measuring the relationship between the open apex and the length of the third molar, diminishing the risk of subjectivity inherent to the stages methods [12]. The resulting cut-off was 0.08 and, since the initial research, the accuracy has been tested in numerous samples from different countries. This step is essential to being able to prove the application of the cut-off in several different populations [13–15]. The present study has two-fold aims: the first is to assess new cut-offs at the age thresholds of 14 years old and 16 years old and the second is to validate the cut-off of \[ L_{M} = 0.08 \] for 18 years of age in Chilean people.

2. Material and methods

2.1. Sample

This work is a retrospective cross-sectional study of radiographs. Orthopantomographs were collected from 822 Chilean children aged from 11 to 22. The OPGs of patients with bilaterally missed lower third or second molars were excluded from the study since it was impossible to obtain complete data from them. Additional exclusion criteria were systemic diseases, congenital anomalies, endodontic treatments of the lower second and third molars, large carious lesions involving the dental pulp and poor-quality X-rays (Table 1).

2.2. Measurements

Following Cameriere et al. [12,16], we evaluated dental maturity of the lower third and second left molars, \( L_{M} \) and \( L_{m} \) respectively, dividing the sum of the distance between the inner side of the two open apices of each tooth by its length. X-ray images were processed by computer-aided drafting program (Image J 1.49).

2.3. Statistical analysis

To assess the new legal age thresholds (LAT) at the age of 14 and 16 years, individual age was used as a dichotomous variable according to whether the person in question was older or younger than the considered LAT value. Using the second and third molars we identified a procedure which allowed to assign an individual to populations younger, if the test was negative, or older, if the test was positive, to LAT. With individual age as a dichotomous response variable (A = 1 if an individual is at least LAT, A = 0 otherwise), and gender, the second (\( I_{2M} \)) and third (\( I_{3M} \)) molar maturity indexes as predictor variables, we derived a logistic model to predict whether an individual is older (A = 1) or younger (A = 0) than LAT. The estimates of the model parameters were obtained by the maximum likelihood method. The predictive accuracy of the model was assessed by the determination of the characteristic receiver operating curve (ROC curve). All significant variables were used to test the medico-legal question as to whether an individual is older or younger than LAT. The test was performed to identify thresholds (cut-offs) that could be used to assign an individual to the population of those younger or older than LAT. It was established that the test is positive if at same time, for a given pair, \( c_1 \) and \( c_2 \), of the cut-offs, we have \( I_{3M} < c_1 \) and \( I_{2M} < c_2 \).

Cut-offs were estimated so that the Sensitivity, \( S_e \), of the test (i.e., the proportion of individuals equal to or older than LAT whose test is positive) and specificity, \( S_p \) (i.e., the proportion of individuals younger than LAT whose test is negative) minimized the distance from the ideal pair of values \( S_e = 1 \) and \( S_p = 1 \).

To not increase artificially the sensitivity of the test for LAT at the age of 14 and 16 years, we restricted the sample to subjects younger than 20 years.

As regard the second aim of this study, Cameriere et al. [12] established a cut-off value \( c^* = 0.08 \) for LAT = 18 years old using \( I_{3M} \) values, and we validated this cut-off on the Chilean population. To validate \( c^* \) on this population, we compare the post-test probability, PPV, obtained analysing the Chilean sample with the post-test probability, PPV*, obtained using \( c^* \).

To not increase artificially the specificity of the test for eighteen we restricted the sample to subjects older than 13 years. The significance level was set at 5%.

Statistical analysis of data and related graphs was carried out by the R statistical program version 3.4.0 [17].

3. Results

When we consider the LAT at the age of 14 years, we observed that only 8.5% of boys and 3.5% of girls, aged less than 14 years, have the third molar completely mature. However, 14.6% of boys and 16.2% of girls, aged less than 14 years, have the second molar completely mature. The mean value of \( I_{2M} \) in boys, 0.092 (standard error 0.009), was significantly higher than in girls, 0.068 (standard error 0.006). To examine the effect of including one of the three

<table>
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<tr>
<th>Age</th>
<th>Male</th>
<th>Female</th>
<th>Total</th>
</tr>
</thead>
<tbody>
<tr>
<td>11</td>
<td>13</td>
<td>12</td>
<td>25</td>
</tr>
<tr>
<td>12</td>
<td>31</td>
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<td>28</td>
<td>37</td>
</tr>
<tr>
<td>22</td>
<td>11</td>
<td>15</td>
<td>26</td>
</tr>
<tr>
<td>Total</td>
<td>350</td>
<td>472</td>
<td>822</td>
</tr>
</tbody>
</table>
factors, gender, I2M, and I3M, in or excluding it from the model, we considered the difference in deviance among different models with or without gender, I2M, and I2M (Table 2). The deviance analysis points out that both gender and molar maturity indexes are significantly related to the LAT at the age of 14 years. The maximum likelihood estimates of parameters of the logistic model used to evaluate the probability that an individual was 14 years of age or older are listed in Table 3. When the second and third molars were used to assign an individual to the population of those younger than 14 years, two cut-offs were found using the ROC curve to assess their discrimination capacity. Fig. 1 shows the resulting ROC curves for boys and girls. The area under the ROC curve (AUCR) for girls, 0.868 (95% confidence interval 0.834–0.902), is significantly lower than the AUCR for boys 0.94 (95% confidence interval 0.915–0.964). Measures of the test accuracy, reported in Table 4, point out the better discriminative property of the test in the boys rather than in the girls. Both sensitivity and specificity result higher in boys than in girls and, consequently, the positive predictive value results significantly higher in boys than in girls.

When we consider the LAT at the age of 16 years, we observed that 23% of boys and 27% of girls, aged less than 16 years, have the second molar completely mature. On the contrary, only 6% of boys and 4% of girls have the third molar completely mature. In Table 5, we summarized the comparison of the nested models obtained including one of the three factors in or excluding it from the model. The deviance analysis points out that molar maturity indexes are significantly related to the LAT at the age of 16 years, but gender is not. The maximum likelihood estimates of parameters of the logistic model used to evaluate the probability that an individual was 16 years of age or older are listed in Table 6.

When the second and third molars were used to assign an individual to the population of those younger than 16 years, two cut-offs were found using the ROC curve to assess their discrimination capacity. The resulting ROC curve (Fig. 2) has an AUCR of 0.890 (95% confidence interval 0.863–0.916). To test the legal question of whether an individual is older or younger than 16 years of age, we determined two cut-offs which were 0.06 and 0.36 for I2M and I3M respectively. This means that the test is positive, i.e. the individual is older than LAT = 16 years of age, if at the same time, we have I3M < 0.36 and I2M < 0.06. The estimated sensitivity of this test was 0.79 and its specificity was 0.81 (Table 7). As far as the second aim of this study, we measure the performance of the cut-off reported in [12] evaluating the validity of the I3M method on Chilean sample. The results were summarized in a 2 × 2 contingency table (Table 8) which displays the numbers of individuals who have I3M ≥ 0.08 and younger than 18, I3M ≥ 0.08 and older than 18, I3M < 0.08 and younger than 18, and I3M < 0.08 and older than 18. Table 8 displays the strong association between adult age and positivity of the test (i.e. I3M < 0.08). In fact, 588 on a total of 709 individuals were correctly classified. Using the results reported in Table 8 we assessed that the sensitivity of the test was 70.5% with a 95% confidence interval equal to (64%, 76.5%), and its specificity was 88.4% with a 95% confidence interval equal to (85.3%, 91.1%). The proportion of correctly classified individuals was 83%. Estimated post-test probability, PPV, was 93.2%, with a 95% confidence interval equals to (91.3%, 94.6%). Hence, the probability that a subject positive on the test (i.e. I3M < 0.08) was 18 years of age or older was PPV" = 93.2%. The cut-off value obtained using the Chilean sample was c1 = 0.11 and the estimated PPV was 92.5% with 95% confidence interval (91.0%–94.0%). Since this PPV is not significantly different from PPV*, obtained using the cut-off c* = 0.08, this result confirms the validity of c* on a Chilean sample.

### Table 2

<table>
<thead>
<tr>
<th>Model</th>
<th>df</th>
<th>Deviance</th>
<th>Residual df</th>
<th>Residual deviance</th>
<th>p-Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Null</td>
<td>0</td>
<td>283.03</td>
<td>719</td>
<td>895</td>
<td></td>
</tr>
<tr>
<td>I2M</td>
<td>1</td>
<td>73.99</td>
<td>718</td>
<td>538.09</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>I2M + I3M</td>
<td>1</td>
<td>5.98</td>
<td>717</td>
<td>532.12</td>
<td>0.01</td>
</tr>
</tbody>
</table>

![ROC for 14 years of age - boys](image1)

![ROC for 14 years of age - girls](image2)

**Fig. 1.** Receiver operating characteristic curve for “14 years of age or older” status for boys (left) and girls (right).

### Table 3

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Value</th>
<th>Std. error</th>
<th>t Value</th>
<th>p-Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Intercept</td>
<td>3.873</td>
<td>0.301</td>
<td>12.849</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Gender</td>
<td>-0.556</td>
<td>0.230</td>
<td>-2.417</td>
<td>0.016</td>
</tr>
<tr>
<td>I2M</td>
<td>-3.176</td>
<td>0.317</td>
<td>-10.028</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>I3M</td>
<td>-20.508</td>
<td>2.837</td>
<td>-7.229</td>
<td>&lt;0.001</td>
</tr>
</tbody>
</table>

### 4. Discussion

The issue of people with unreliable documents or any document at all is increasing worldwide as situations like armed
conflicts, violence and poor work opportunities in their original countries, make people to take the risk to travel in search of better conditions. This population tend to have a lack of reliable documents since they either loose them or even destroy them out of fear, some others alter them to seem older to have the opportunity to work and get state benefits or instead to appear younger before the law and get a softer penalty in a criminal case. Chronological age in criminal law is determinant for a judge to decide which set of rules will be used to approach a case and it is impossible to do so without a method that can estimate as accurately as possible an individual’s age [18–20].

18 years old is the most common age for full criminal responsibility in the world. Instead, there are many different ages for other LATs. In Chile, 14 and 16 years are important for several law issues.

Regarding 14 years cut-off just few articles are published [18,19,21,22], and only in the first two articles, the authors, using teeth, pointed out different cut-offs according to gender.

As far as cut-offs at LAT of 16 years is concerned, at present, no scientific article has yet been published. Our results underlined that both $I_{2M}$ and $I_{3M}$ are significantly related to the LAT at the age of 16 years but gender is not.

In the last decade, several articles have been published about age estimation using maturity index, $I_{3M}$. Some of these concerned the South American population specifically Colombia, Peru and Brazil [5,14,23]. In Colombia, for girls, the sensitivity test was 95.1% (95% CI 87.1%–95%) and specificity was 93.8% (95% CI 87.1%–98.8%). The proportion of correctly classified individuals was 95.1%. For boys, the sensitivity test was 91.7% (95% CI 85.1%–96.8%) and specificity was 90.6% (95% CI 82.1%–97.8%). The proportion of correctly classified individuals was 89.7%. In Peru, the accurate classification, sensitivity and specificity were 0.96, 0.96, 0.96 and 0.90, 0.84 and 0.95 in males and females, respectively. In Brazil, the results show that the sensitivity of the test was 77.4% and its specificity was 86.2%, with a 95% confidence interval. The proportion of correctly classified individuals was 87.4%. Concerning these results, it is important to highlight two issues: first, in both Chilean and Brazilian samples the sensitivity’s values are slightly lower than in the other ones but the specificity’s values are very high in all the studied samples. The specificity of $I_{3M}$ is the ability of the test to identify correctly those who are not yet 18 years old (i.e. all individuals younger than 18 years of age must be correctly classified). In fact, in a forensic scenario, offenders who are not classed as children, but instead deemed to be legally adult, accordingly enter the adult justice system exposing them to additional mental and physical risks, and receiving also significant breaches of his/her human rights. Second, and according to the studies already carried out in some neighbour countries such as Colombia, Brazil and Peru, the obtained results showed that the cut-off (0.08) is not population specific and can be also applied with the same reliability in those Latin American contexts where the estimation of the legal age of criminal responsibility (18 years) is a part of the identification process in forensic practice.

To the best of our knowledge, studies that assess age using dental calcification are becoming increasingly important but there are few researches regarding tooth development in the Chilean population, and none in the forensic field. According to the
literature, only two studies have been carried out in Chile. In Pérez Flores et al. [24] the dental age was estimated, in a sample of 159 children aged between 3 and 14 years, by the Demirjian method and it was determined that the range between chronological and dental age was similar, showing a degree of correlation nearly perfect between both ages. Cadenas et al. [25] studied the dental development of a sample of 363 Chilean children aged between 5 and 15 years and highlighted a general overestimation in both girls and boys.

The present research, following the previous studies of Cameriere et al. [12], tests the applicability of using the relationship between apex and tooth for second and third molar, to determine the LAT in Chilean population.

Some other researchers have tested the use of dental development to establish an ideal cut-off for the different LAT [5,13–16,26–28]. When testing the I3M accuracy of the cut-off value (0.08) in the Chilean, we also look for the cut-off obtained from the specific Chilean sample to avoid a biased result. Our 0.08 cut-off did not show a significant difference in the estimated PPV compared with the one estimated from the Chilean sample. This result highlights the validity of the 0.08 cut-off for identifying the adults in Chilean population.

This is the first study verifying the reliability of the I3M in discriminating between minors and adults in a Chilean sample of children. Furthermore, for the first time, I3M is evaluated in combination with I3m to find cut-offs for fourteen and sixteen years. Next steps will be to test I3M in different populations as cut-off for eighteen years, and the combination of I3M and I3m for LATs in different countries.

References