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Radiological studies on ossification of iliac crest as an aid in estimation of age

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ABSTRACT

Forensic research and medico legal investigations demand for standardized techniques that help to estimate the chronological age of living individuals. Age estimation methods suggested by various authors have focussed only on the endpoint of epiphyseal developmentand their criteria for each stage of ossification are unclear. This cross-sectional study aims to explore the relationship between the ossification of iliac crest and chronological age in a modern South-Indian population. The pelvic radiographs of 390 subjects (213 males; 177 females) were analysed using a novel scoring method that subdivides the ossification of iliac crest into six stages and each stage of ossification was correlated with chronological age of the individual. The present study revealed that the degree of epiphyseal growth is dependent on the age of the age of the individual, which supports the fundamental basis of age estimation methods.In accordance with previously published studies, significant sex differences with epiphyseal changes in females occurring earlier than males are recorded.The present study is the first to offer a standardized method to undoubtedly identify six stages of iliac crest ossification on radiographs along with their respective age ranges at 95% confidence interval. The standardized criteria of stages eliminates inconsistency between interpretations of same radiographsand the subdivision of the ossification process into six stages enables forensic experts to estimate a more refined age of the individual. On comparison with similar studies, the existence of population variability in the process of ossification was substantiated. Thus, there is need forspecific data on iliac crest ossification from respective geographical areas to ensure more accuracy in age estimation.Further research is required to establish the existence or significance of lurking variables in relation to the ossification process.
Keywords: Determination of age, biological anthropology, epiphysis, iliac crest, forensic medicine, ossification, radiographs, skeletal maturation.

1. INTRODUCTION

While age estimation of unidentified corpses and skeletons for identification purposes has a long tradition in forensic sciences, age estimation of living persons has formed a relatively recent area of forensic research and medico legal importance (A. Schmeling et al., 2007). It is

a prerequisite for personal identification, and is increasingly important in criminal investigations. Forensic age estimation is promptly requested by authorities to ascertain whether the person concerned in a suspected crime has reached the age of criminal responsibility .Such medico legal situationsvary from a child as young as 5 years old, ranging through an adult person agedup to 70 years. The reason for these situations- the former being, according to Section 6 (a) of TheHindu Minority and Guardianship Act (India, 1956), a minor who has not completed the age of 5 years shall ordinarily be in the custody of the mother, andthe latter where in, people allege to be over 70 years old in order to qualify for some State Government's pension scheme for old people. Thus virtually no age is irrelevant in its potential for an aging dispute from a medico legal point of view (J. Payne- James et al. , 2003).An exhaustive list of these ages with its respective medico legal importance wasgiven by Aggarwal (A. Aggrawal, 2000).Among them, Payne-James*et al.*gave a glimpse of the large ranges of ages which a practising medico legal specialist in India, England and Wales may be asked to opine upon along with its respective medico legal importance (A. Schmitt *et al., 2006*).

The demand of forensic age determination in India deals mostly with juvenile or subadults because,themostfrequently encountered legal cases such as eligibility for employment, juvenile delinquencies, rape, kidnapping, attainment of majority, validity ofconsentetc.,fall between the age limits of 14 and 21 years of life. The relevant age thresholds in criminal proceedings are 14, 16, 18 and 21 years of age (A. Schmeling et al, 2004). Therefore our study has focussed the abovementioned age limits and has taken subjects from 13 to 25 years of age for analysis.

The age of an individual can be determined from teeth, ossification of bones and secondary sex characteristics (A. Aggrawal et al, 1991) and an opinion is given only after careful consideration of all the three parameters(J. Payne- James et al. , 2003).However,it is also stated that the study of appearance of ossification centres and epiphyseal union of bones is considered as a reasonably scientific and accepted method for age determination by the courts of law all over the world (K.K Banerjee, 1998). Analysis of ossification of bones provides a very useful method of estimation of age in the living due to the fact that, ossification centres do not appear suddenly and epiphyseal fusion at any particular site does not manifest as a sudden transition from two or more ossified centres to one. Instead it progresses through a series of stages over a period of time that can be seen on gross inspection or by various imaging techniques of bones.

Several studies on age estimation by analysing the appearance ofossification centres and epiphyseal fusion in various bones have been done in India and abroad.Studies (K.K Banerjee, 1998) reveal that the timing of appearance of ossification centres and epiphyseal union varies between individuals from different populations. This may be due to population variability, secular changes (P. Eveleth et al., 1990; M. Lampl et al., 1996) or lack of standardized procedures. Therefore, a need arises for collection of data based on specific population groups. For South Indian population, judicial systems and forensic research rely upon the age-old study provided by Pillai(M.J.S Pillai, 1936).Since medico legal cases involving age estimation demand high levels of accuracy, it is critical to revise these data upon which reliance has been placed(S. Ritz-Timme et al., 2000)

Keeping the above information in mind, an attempt has been made to analyse theepiphysealfusionof iliac crest using digital x-rays in those subjects of South India whose ages has been recorded with reasonable degree of accuracy.

2. MATERIALS AND METHODS

This prospective cross-sectional analysis of the relationship between ossification of iliac crest and chronological age was undertaken using a sample of digital radiographs taken from a population presentedto a private hospital at Chennai in Tamil Nadu, India.
Subjects aged 13-25 years who presented to the hospital from March 2018 to September 2018 through Out-patient or Emergency Departments for some illnesses who require radiographs, were selected by random sampling technique. Only those individuals with documentary evidences of date of birth such as municipal birth certificate, hospital birth certificate, school leaving certificate, driving licence, passports, ration card, voter's card etc. were included in our study. Exclusion criteria were as follows: subjects with known endocrine, metabolic or nutritional disorders, subjects exhibiting trauma or pathology of the bones of the pelvis and hip joint.Based on these criteria, the final number of subjects included in the study was 390 (213 males; 177 females). Sex and age distribution of the sample is shown in tables 1 - 4.
(Antero posterior radiographs of pelvis showing iliac crest) X-rays of the subjects without any trauma or deformities are selected. The chronological age of each individual was calculated using information provided on the date of birth and the registration for x-ray, therefore allowing the calculation of exact age (years) at the time of x-ray.
The sample was sent for radiological assessment. All evaluations were made by one examiner. The investigator was blinded to information on sex and age of the subject being assessed. Based on the criteria of variables given below, the investigators were asked to assign a stage of ossification of iliac crest for each subject.

2.1. Description of variables

The developmental status of ossification of iliac crest was divided into the following six stages.
Stage 0 – Ossification centre not appeared.
Stage 1 – Ossification centre partially appeared.
Stage 2 – Ossification centre fully appeared, epiphyseal cartilage not fused.
Stage 3 – Epiphyseal cartilage partly fused.
Stage 4 – Epiphyseal cartilage fully fused, epiphyseal scar visible.
Stage 5 – Epiphyseal cartilage fully fused, epiphyseal scar no longer visible.

Two age variables were used: AGE1, a 1-year interval grouping, and AGE2, grouping based on age thresholds of medico legal importance such as 14, 16, 18 and 21 years. The firstvariable provides detailed information on the variability of ossification of iliac crest in relation to age while the second oneis in accordance with the demand of forensic age estimation in medico legal proceedings in India.

2.3. Statistics

Results were analysed using SPSS (v.17). Analyses included the use of chi-square, correlation tests and regression analysis.

3. RESULTS

For sake of clarity, results are presented in fourparts referring to: (1) Age and sex distribution of the sample; (2) Data to predict the age of an individual; (3) Sex and age association with degree of ossification of iliac crest; (4) Regression analysis.

3.1. Age and Sex distribution of the sample

Tables 1, 2 and Tables 3, 4 present the distribution of male and female subjects at each stage of ossification of iliac crestby AGE1and AGE2 respectively. For both genders, it is evident that the ossification of iliac crest progresses through a number of definite stages with increasing chronological age.

The age (years) of the youngest and oldest subjects observed at each stage of ossification of iliac crest for males and females are given in Table 5 and Table 6 respectively. Mean age (years) and standard deviation for each stage of ossification are also presented. Although the range in age (years) for each stage of ossification appears to be large, the mean age (years) provides an indication of the typical age at which each stage of ossification occurs, while the age of the youngest and oldest at each stage of ossification demonstrates the population variability. The mean age gradually increases with each stage of ossification and shows variation between male and female subjects.

Figures 7 and 8 present the percentage of degree of ossification of iliac crest by AGE2 for males and females respectively. The maximum percentageofmalesubjects in stages 0, 1& 2, 3&4, 5 fall under the age groups of <14, 16-17.9, 18-20.9, 21-25.9 years respectively. And the maximum percentage of female subjects in stages 0&1, 2, 3, 4, 5 fall under the age groups of <14, 14-15.9, 16-17.9, 18-20.9, 21-25.9 years respectively.This gives a rough idea about the placement of a majority of people under a specific age group (AGE2) of medico legal importance with respect to their ossification status of iliac crest.

3.2.Validations to Predict the Age of an Individual

The age range at 95% confidence interval and mean age for each stage of ossification of iliac crest is graphically represented in figures 9-11. The figures 9 &10could be used as tools to estimate the age of an individual of known sex, from a pelvic radiograph. In cases where sex is not known, figure 11 can be used as an alternative.

3.3.Association of Sex with Degree of Ossification of Iliac Crest

Chi-square test indicates significant sex differences in the degree of ossification of iliac crest for AGE1 (x^2 =12.56;p = 0.028) with female subjects attaining stages 0,1,2,3,4 and 5 on mean values of0.77, 1.56, 2.46, 1.4, 1.21 and 0.6 years respectively, earlier than the male subjects. For AGE2, chi-square test (Table 7) reveals significant sex differences in degree of ossification of iliac crest only for the age groups of 14-15.9 and 16-17.9 years. The remaining age groups show false negatives as their values are less than 5 in more than 20% of the frequencies.

3.4. Age Association with Degree of Ossification of Iliac Crest

The chi-square test shown in Table 7 indicates a clear association between the degree of ossification of iliac crest and the two age variables AGE1 and AGE2 for both sexes. Correlation tests (table 8) also reveal statistically significant positive correlation between AGE1 and the degree of ossification of iliac crest for both sexes. It indicates that the age increases with the development of iliac crest. Thus we can conclude that the degree of ossification of iliac crest is not independent of age of the individual.

3.5 Regression Analysis

To analyse the percentage of iliac crest development attributed to age, coefficient of determination (R^2) was calculated using linear regression analysis between the degree of ossification of iliac crest and AGE1 (table 9).Regression analysis predicts that the 66.7% and

57.1% of the variation in stages of iliac crest ossification is attributed to its regression on age in males and females respectively.

4. DISCUSSION

In the recent era, medico legal investigations and forensic research demand for techniquesthat help to estimate the chronological age of living individuals. Among such techniques, radiological methods of analysing the epiphyseal changes in bones have been widely used and considered as a reasonably scientific and acceptable method for age estimation by all courts of law (K.K Banerjee, 1998). However,due to population variability suggested by various studies (K.K Banerjee, 1998; M.C Schaefer, 2009), a constant revision of these techniques in specific population groups must be undertaken to maintain accuracy in age estimation. In contrast to techniques of analysing epiphyseal changes in skeletal remains,radiographic analysis of epiphyses in living subjects gives us an opportunity to examine large number of subjects along with the establishment of accurate chronological age. In order to get the most accurate results, a longitudinal study of epiphyseal fusion is ideal. However, such a study involves ethical issues relatingtohealth hazards in subjects due to repetitive x-ray exposures. So cross sectional study involving large number of people in each age group is undertaken to overcome this problem.

The fundamental basis of age estimation methodsisthat the epiphyses show a gradual change over a period of time and their developmentoccursin small increments until the adult state is reached (Mellits ED et al, 1971). However, a majority of studies on ossification of iliac cresthave focussed on the endpoint of epiphyseal development instead of analysing the process of ossification as a whole. Consequently, only two stages of ossification were described: appearance of ossification centre or union of epiphyses. The criteria for these stages have been unclear as the stage of appearance of ossification centre ranges from partial to complete appearance of ossification centre and the stage of union of epiphyses ranges from commencement of fusion of epiphyses to the disappearance of epiphyseal cartilage. Therefore, two or more investigators are likely to have varied interpretation of the same subject. To overcome this variation, we have adopted a six-stage classification representing the major increments in the ossification process, which can be easily identified on radiographs (Figure 1-6).

The distribution of male and female subjects by AGE1 at each stage of ossification (tables 1 &2), clearly indicate that the process of iliac crest ossification slowly progresses through small increments or a series of stages over a period of years until the adult state of complete fusion is obtained. Chi-square test (table 7) and correlation tests (table 8) also revealed a significant association and positive correlation respectively between AGE1 and the stages of iliac crest ossification for both sexes and thus concluding that the degree of epiphyseal growth is dependent on the age of the individual. This is in accordance with the fundamental basis of age estimation methods as described by Mellits et al. 1971

The current study revealed that the females attain stages 0, 1,2,3,4 and 5 of iliac crest ossification at a younger age on mean values of 0.77, 1.56, 2.46, 1.4, 1.21 and 0.6 years respectively, than their male counterparts along with statistical evidence of chi-square test showing significant sex differences in the degree of ossification of iliac crest for AGE1 (x^2 =12.56; p = 0.028). This is in accordance with previous studies such as Flecker, 1932;Galstaun G , 1937, which have revealed that female subjects develop 1-3 years earlier than males. Thus, to ensure more accuracy in age estimation methods, separate age standards are essential for males and females.

Table 10 compares the present study with similar studies on ossification of iliac crest. All these studies have focussed their results only on the endpoint of epiphyseal development and

thereby giving a range for the time of appearance or fusion of iliac crest. In order to compare the present study with similar studies undertaken previously, we have grouped stages 1&2 under the stage of appearance and stages 3, 4&5 under the stage of fusion. Having groupedaccordingly, it is evident that the time of appearance of iliac crest established in the present study is in agreement with the study carried out by Pillai in the same madrassie population in 1936. Whereas, the rest of the studies done in various other geographical populations reveal that the time of appearance or fusion of iliac crest falls within the range described in our study, but in a narrower fashion. This indicates that the process of iliac crest ossification varies betweendifferent geographical populationsand thus substantiating the concept of population variability in the process of ossificationsuggested byB.B.L Agarwaland M.C Schaefer.

Therefore, there is a need for specific data on iliac crest ossification from respective geographical areas to ensure more accuracy in age estimation.

The present study is the first to provide anageinterval (in years)for each of the major increments in the ossification of iliac crest taking place among 95% of the entire population (figures 9-11). Thereby, an investigator can make use of this data to correlate the stage of ossification analysed from a pelvic x-ray and hence predict the age of that individual.Due to significant sex differences, separate data for males and females represented in figures 9 & 10 respectively has to be used in order to obtain more accuracy in age estimation. However, in cases where sex is unknown, figure 11 can be used as an alternative. It is evident that each stage of iliac crest ossification point toshorter periods of life rather than the broad range when entire appearance or fusion is considered. Thus the age of the individual can be estimated precisely with a more refined age range.

Linear regression analysis (Table 9) predicts that, 66.7% (in males) and 57.1% (in females) of iliac crestdevelopment isexplained by age. The remaining 33.3% (in males) 42.9% (in females) can be attributed toinherent variability or unknown,lurking variables which may also have an impact on the pace of ossification.A list such variables described by K.S. Narayan Reddy include health, nutritional, endocrine and environmental factors. The common denominator among most of these factors is socio-economic status of the individual. This is in accordance to the study by Schmeling A.*et al* (Schmeling A.*et al*, 2006) which reports that low socio-economic status delays development and thus their age would be underestimated. The study also commented that,in criminal proceedings, this underestimation of age would not result in a derogatory situation of the individual concerned.However, such a comment is questionable and may not be accepted by all individuals. Hence, further research is required to establish the existence or significance of these variables in relation to the ossification process.

AGE2 variable is considered to give an insight about the distribution of various stages of iliac crest ossification in relation to the age thresholds of medico legal importancesuch as 14,16,18 and 21 years. Tables 3&4 indicate that the process of iliac crest ossification slowly progresses through a number of definite stages as the age group increases. It is also confirmed by chi-square tests (table 7) revealing significant association between the degree of ossification of iliac crest and AGE2. Significant sex differences in development of iliac crest are shown only for the age groups of 14-15.9 and 16-17.9 years (table 6). In other age groups, more samples are required to analyse sex differences using chi-square test. Figures 7 and 8 reveal the placement of majority of people under a specific age group of medico legal importance with respect to their ossification status of iliac crest. This data summarized in table 10 can be used to get a rough idea about the placement of individuals in an age group of medico legal importance when a pelvic x-ray is presented. However, for accurate estimation of age, the data (figure 9-11) with age ranges at 95% confidence interval must be used.

5. CONCLUSION

At present, there is an obvious dearth for standardized procedures to estimate the age of individual from radiographs. Age estimations methods that are used currently focus only on the endpoint of epiphyseal development and their criteria for each stage of ossification is unclear leading to inconsistency between two or more interpretations of the same radiograph. Thus, there is a necessity to standardize the technique of age estimation using radiographs and to remove this inconsistency. The present study is the first to offer a standardized method to undoubtedly identify six stages of iliac crest ossification on radiographsalong with their respective age ranges at 95% confidence interval.The subdivision of the ossification process into six stages enables forensic experts to estimate a more refined age of the individual.

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Figures 1 – 6 show the findings that correspond to each of the stages 0 – **5.**

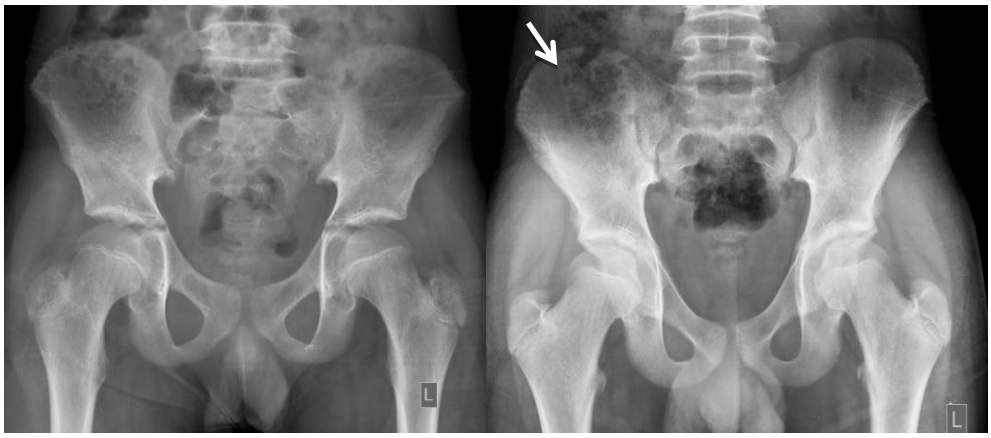


Figure 1: X-ray Pelvis- AP view showing stage 0
Figure 2: X- ray Pelvis – AP view showing stage 1

Figure 3: X- ray Pelvis – AP view showing stage 2
Figure 4: X- ray Pelvis – AP view showing stage 3

Figure 5: X- ray Pelvis – AP view showing stage 4
Figure 6: X- ray Pelvis – AP view showing stage 5

Figure 7:
Percentage of degree of ossification of iliac crest for males

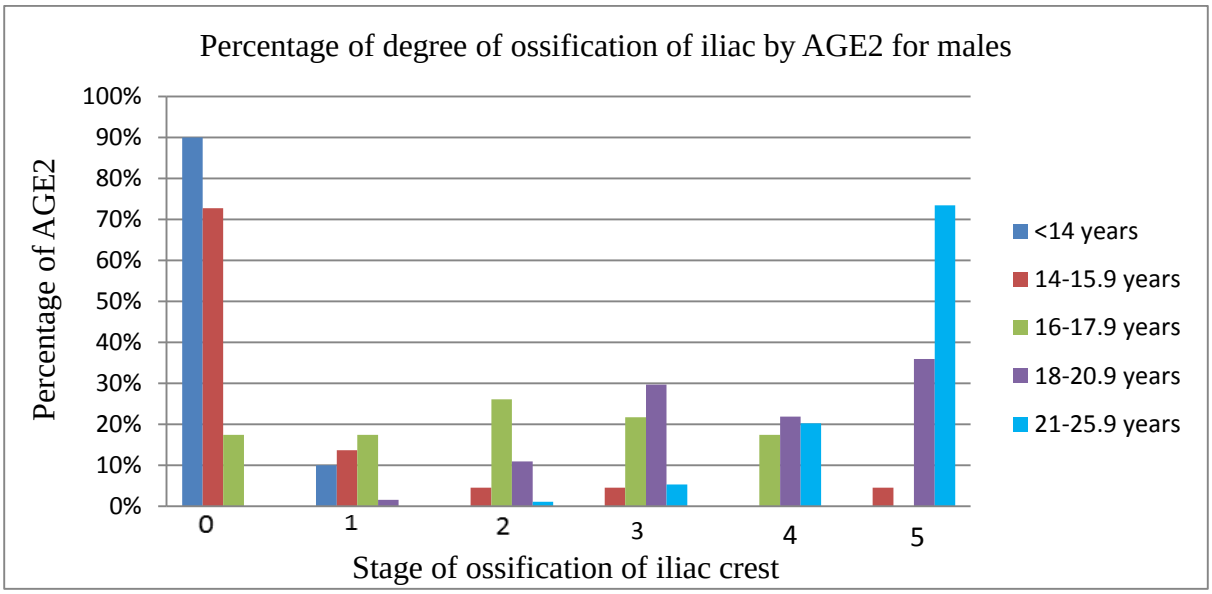


Figure 8:
Percentage of degree of ossification of iliac crest for females

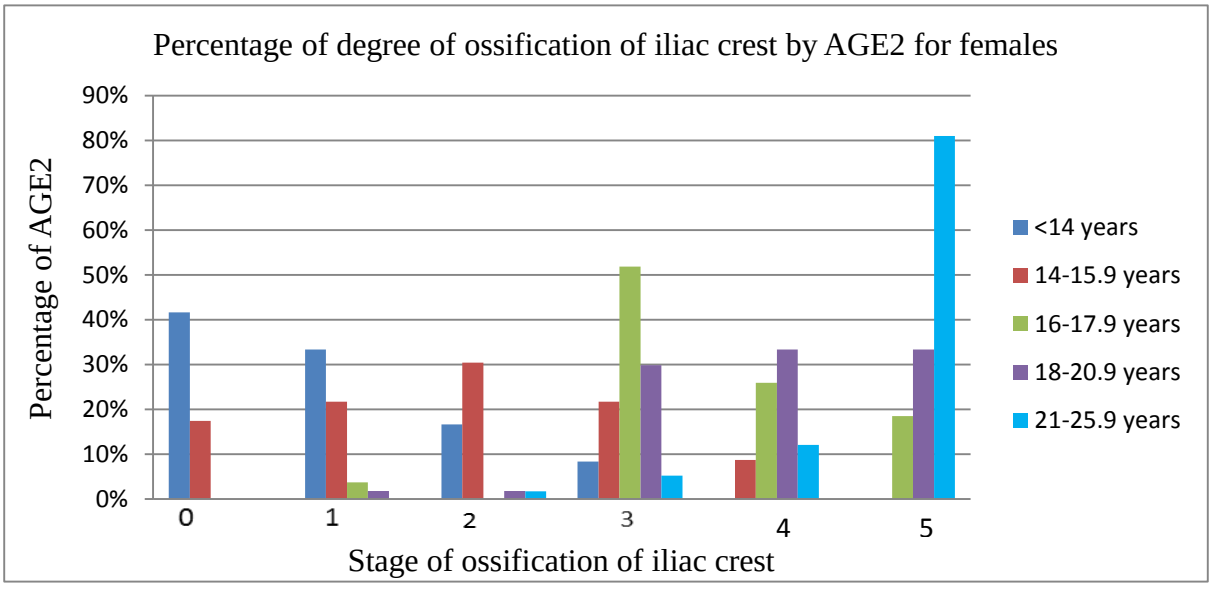


Figure 9
Mean age for ossification of iliac crest by stages in males

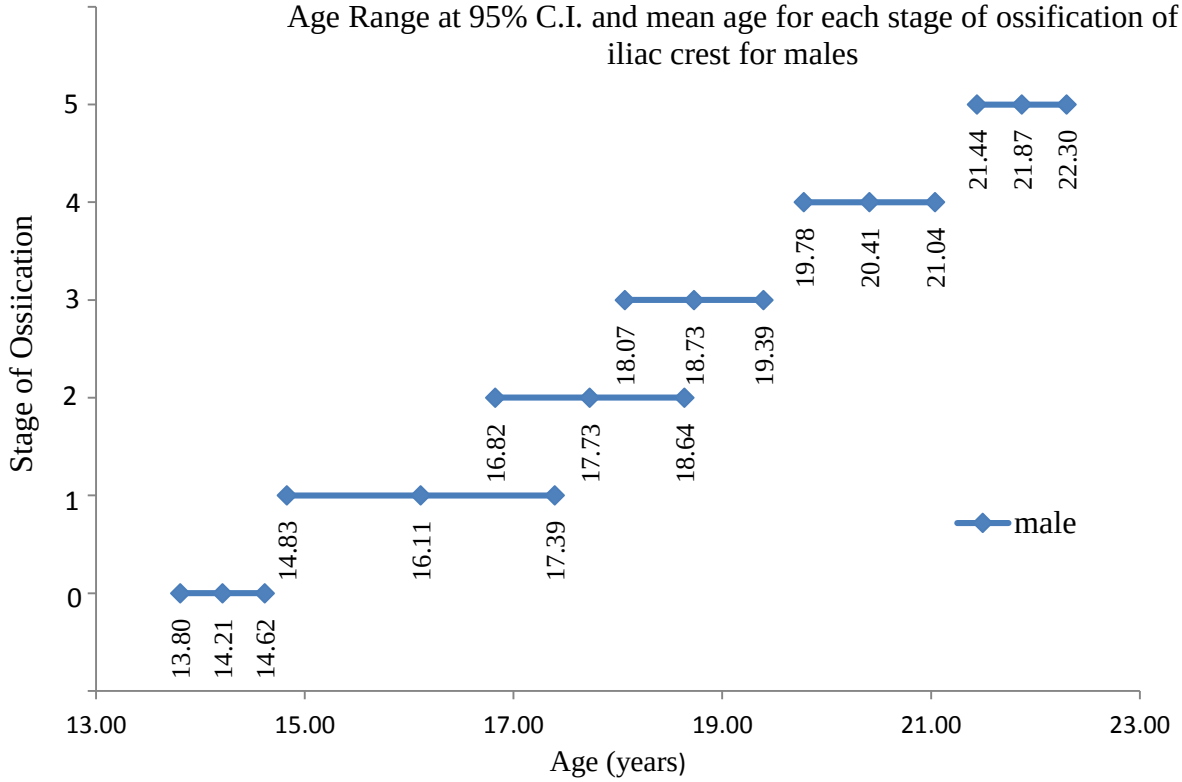
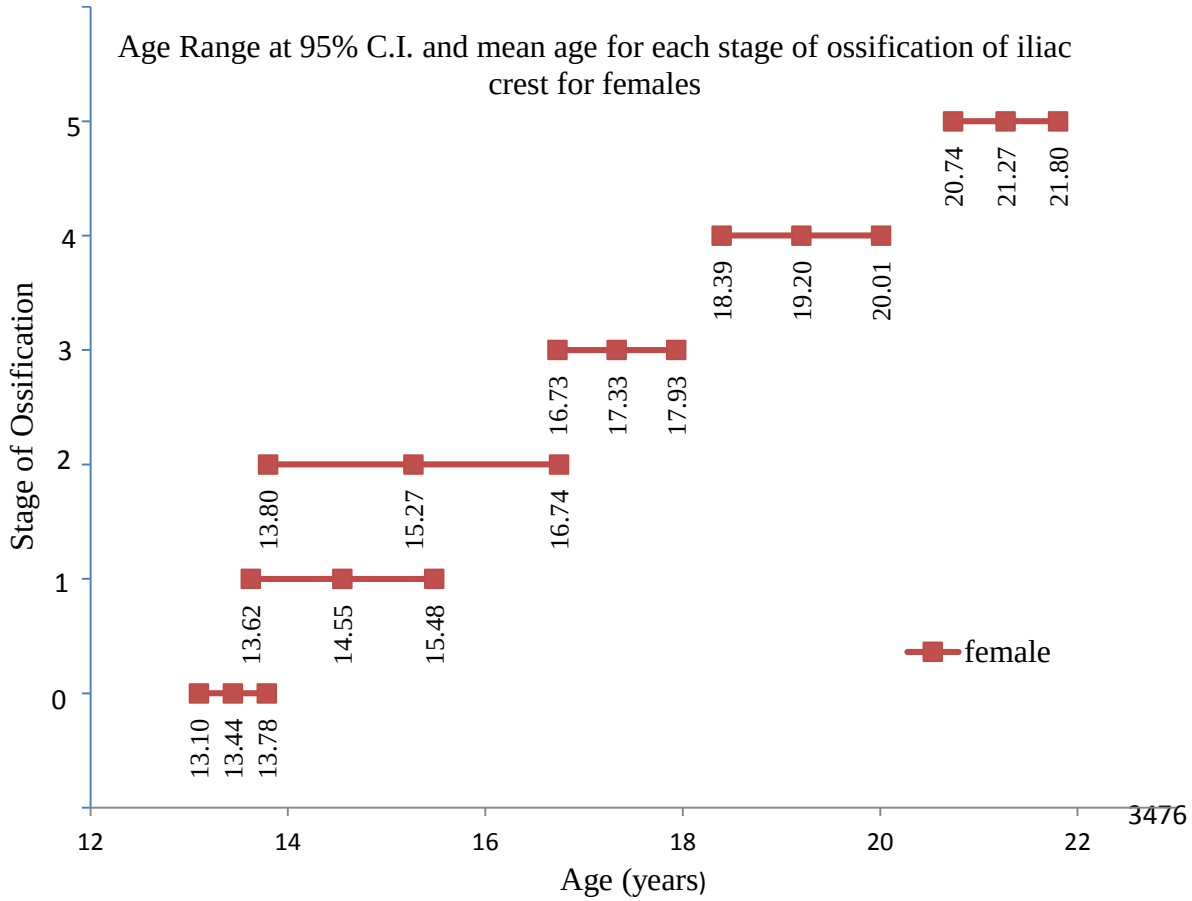


Figure 10
Mean age for ossification of iliac crest by stages in females

Figure 11: Mean age for ossification of iliac crest by stages in both sexes



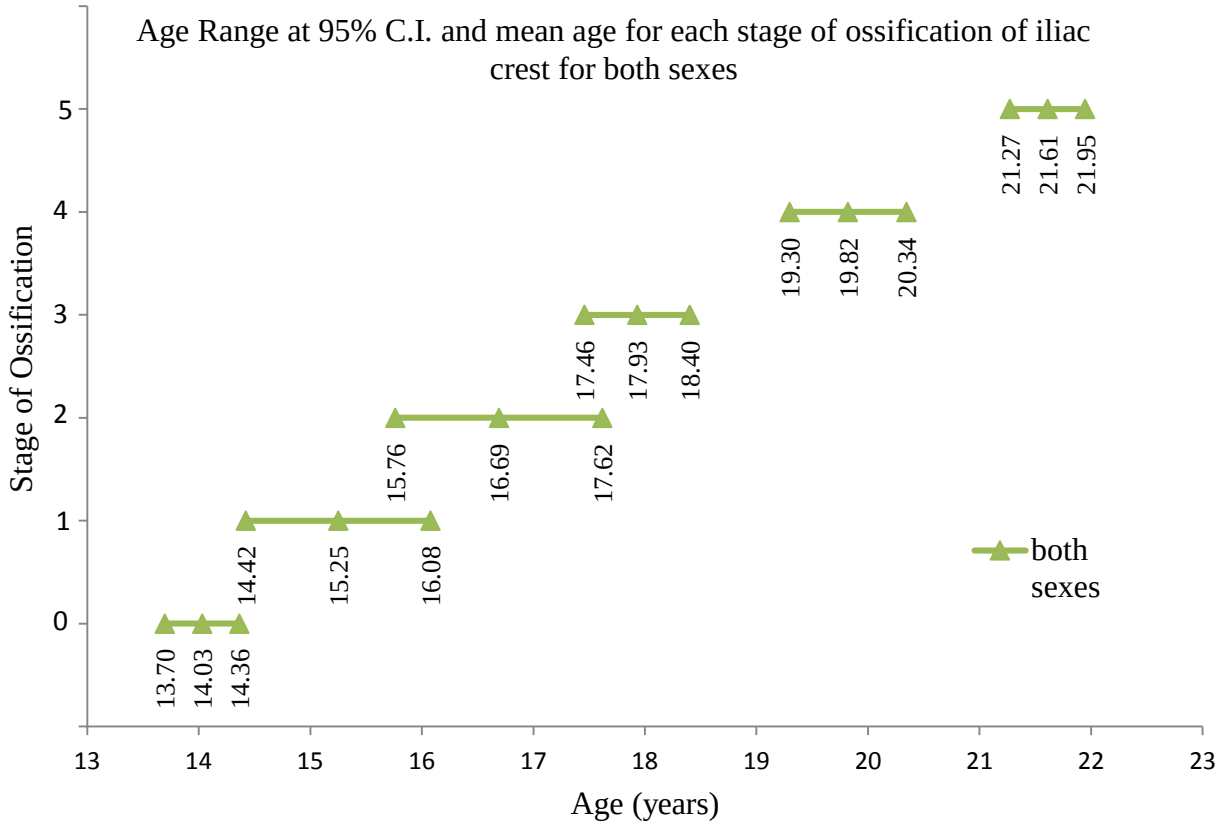


Table 1:
Distribution of male subjects at each stage of ossification of iliac crest by AGE1

Male		Iliac crest Stages of ossification					
AGE1 (years)	N	Stage 0	Stage 1	Stage 2	Stage 3	Stage 4	Stage 5
13-13.9	10	9	1	0	0	0	0
14-14.9	10	10	0	0	0	0	0
15-15.9	12	6	3	1	1	0	1
16-16.9	12	3	1	4	3	1	0
17-17.9	11	1	3	2	2	3	0
18-18.9	20	0	0	3	10	2	5
19-19.9	14	0	0	2	3	3	6
20-20.9	30	0	1	2	6	9	12
21-21.9	27	0	0	1	2	11	13
22-22.9	27	0	0	0	3	3	21
23-23.9	16	0	0	0	0	2	14
24-24.9	10	0	0	0	0	3	7
25-25.9	14	0	0	0	0	0	14
Total	213	29	9	15	30	37	93

Table 2:
Distribution of female subjects at each stage of ossification ofiliac crest by AGE1

Female		Iliac crest Stages of ossification					
AGE1 (years)	N	Stage 0	Stage 1	Stage 2	Stage 3	Stage 4	Stage 5
13-13.9	12	5	4	2	1	0	0
14-14.9	10	4	1	3	2	0	0
15-15.9	13	0	4	4	3	2	0
16-16.9	10	0	1	0	5	3	1
17-17.9	17	0	0	0	9	4	4
18-18.9	25	0	1	0	15	4	5
19-19.9	12	0	0	1	2	4	5
20-20.9	20	0	0	0	0	11	9
21-21.9	20	0	0	1	1	4	14
22-22.9	12	0	0	0	1	0	11
23-23.9	11	0	0	0	1	0	10
24-24.9	6	0	0	0	0	1	5
25-25.9	9	0	0	0	0	2	7
Total	177	9	11	11	40	35	71

Table 3:
Distribution of male subjects at each stage of ossification of iliac crest by AGE2

Male		Iliac crest Stages of ossification					
AGE2 (years)	N	Stage 0	Stage 1	Stage 2	Stage 3	Stage 4	Stage 5
<14	10	9	1	0	0	0	0
14-15.9	22	16	3	1	1	0	1
16-17.9	23	4	4	6	5	4	0
18-20.9	64	0	1	7	19	14	23
21-25.9	94	0	0	1	5	19	69
Total	213	29	9	15	30	37	93

Table 4:
Distribution of female subjects at each stage of ossification of iliac crest by AGE2

Female		Iliac crest Stages of ossification					
AGE2 (years)	N	Stage 0	Stage 1	Stage 2	Stage 3	Stage 4	Stage 5
<14	12	5	4	2	1	0	0
14-15.9	23	4	5	7	5	2	0
16-17.9	27	0	1	0	14	7	5
18-20.9	57	0	1	1	17	19	19
21-25.9	58	0	0	1	3	7	47
Total	177	9	11	11	40	35	71

Table 5:
Mean, SD and range in age (years) for each stage of ossification of iliac crest for males**.**

Male					
Stage of ossification	N	Age (years) of the youngest subject	Age (years) of the oldest subject	Mean age (years)	SD
Stage 0	29	13	17	14.21	1.114
stage 1	9	13	20	16.11	1.965
stage 2	15	15	21	17.73	1.792
Stage 3	30	15	22	18.73	1.856
stage 4	37	16	24	20.41	1.95
Stage 5	93	15	25	21.87	2.112

Table 6:
Mean, SD and range in age (years) for each stage of ossification of iliac crest for females.

Female					
Stage of ossification	N	Age (years) of the youngest subject	Age (years) of the oldest subject	Mean age (years)	SD
Stage 0	9	13	14	13.44	0.527
Stage 1	11	13	18	14.55	1.572
stage 2	11	13	21	15.27	2.494
stage 3	40	13	23	17.33	1.94
stage 4	35	15	25	19.2	2.435
stage 5	71	16	25	21.27	2.286

Table 7:
Chi-square test for sex differences in degree of ossification of iliac crest by AGE2 ($p = 0.05$)

Chi-square	AGE2				
	<14 years	14-15.9 years	16-17.9 years	18-20.9 years	21-25.9 years
x^2	5.81	17.85	21.7	5.36	1.78
P	0.121	0.003	0.001	0.252	0.618

Table 8:
Chi-square test for the association between degree of ossification of iliac crest and AGE1 and AGE2 by sex

Chi-square	Degree of ossification of iliac crest	
	Male	Female
AGE1 x^2 *p*	 291.99 0.000	 237.14 0.000
AGE2 x^2 *P*	 228.6 0.000	 177.51 0.000

Table 9:
Correlation test between AGE1 and degree of ossification of iliac crest

Sex	Pearson's correlation coefficient (r)**
Male	.817
Female	.756
Both	.774

**Correlation is significant at the 0.01 level (2-tailed)

Table 10
Maximum percentage of age group(AGE2) in each stage of ossification of iliac crest

Stage of iliac crest ossification	Maximum percentage of AGE2 (in years)	
	Male	Female
Stage 0	<14	<14
Stage 1	16-17.9	<14
Stage 2	16-17.9	14-15.9
Stage 3	18-20.9	16-17.9
Stage 4	18-20.9	18-20.9
Stage 5	21-25.9	21-25.9