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NEW METHOD IN THE AGE ESTIMATION BY THE SPHENO-OCCIPITAL SUTURE. 3D CONE-BEAM CT APPLICATION.

Radiology

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ABSTRACT

The aim of the present study was to determine the sequence and timing of closure of the spheno-occipital synchondrosis for a large sample of a modern Italian population to assess if this age marker is a useful tool for age estimation for individuals. The sample consisted of 494 individuals in the age range 0-22 years, who were admitted to the Department of Radiology, Oncology and Anatomo-Pathology of "Sapienza" University of Rome - UOC Head and Neck Radiology - and the Department of imaging, University of L'Aquila, and who had undergone multi-slice CBCT imaging. The average age of the spheno-occipital closure in men is 18.3 years, whilst in women is 16.6. Therefore, it is evident that the closure in female people is faster than 1.7 years. The timing of closure of spheno-occipital suture can be used to understand the age, even if the research has to be implemented.

KEYWORDS

Forensic anthropology population data article; Age estimation; Spheno-occipital synchondrosis; 3D Cone Beam CT, Italian population.

Introduction

Economic globalisation, European integration, and the current armed conflicts have led in recent years to a rise of cross-border migration in Europe, which in turn has led to a steady increase of the foreign population in many European countries. This trend has triggered a growing demand for forensic medicine to assess the age of adolescents and young adults. The examined individuals are unaccompanied minors without valid identification documents who do not know their age or are suspected of not giving their correct age. The date of birth is key information, related to the age of people, because of its several juridical consequences; the aim is to find out whether the adult penal law is applicable to the individual. In most European countries the legally relevant age limit ranges between the 13th and 21st year of life, although in some places individuals can be held accountable for their crimes from the age of 8 (1). Age estimations, done in accordance with good medical practice, are important for criminal proceedings. They ensure equal treatment for offenders; rather they have or not have identification documents.

The Italian judicial system establish that subjects under 14 years of age are never imputable (2), while between 14 and 18 years old imputableness has to be stated in each case by evaluating the subject's capacity (3).

The most suitable procedures currently available are: physical examination with anthropometric measurement, inspection of the signs of sexual maturation, and identification of any developmental disorders that might affect age-appropriate development, radiographic examination of the left hand, examination by a dentist with determination of the dental status and radiographic study of the dentition. An additional radiographic or digital tomographic examination of the collar bones is recommended to check whether an individual has completed his or her 21st year of life.

These methods are more precise in the early phases of development. There are many different biological indicators of age but their reliability is variable with age: for example, below the age of 14–15 years the developing dentition (4-5) and hand/wrist (6-7-8) ossification provide reasonably accurate age estimations, but once these development sites have completed their growth, accurate age estimation becomes far more difficult. Moreover, all the procedures recommended above should be used in combination among them, to increase the diagnostic accuracy and to improve the identification of any relevant developmental disorders. The main difficulties, which have considerable practical implications, regard how the ethnicity and the socioeconomic status (9) affect the developmental systems that are examined for age estimation (10). In fact, further investigations are required to determine the influence of ethnicity on dentition and sexual maturation. A multifactorial method for estimating age was devised based on the development of the 3rd molar tooth, the medial clavicular epiphysis, and the spheno-occipital synchondrosis, using multiple regressions as the means to construct age estimation formulae and CT scanning as the imaging modality (11).

Synchondrosis is defined as the development of a union between two bones by the formation of either hyaline cartilage or fibro-cartilage. A synchondrosis is usually temporary and exists during the growing phase until the intervening cartilage becomes progressively thinner during skeletal maturation and ultimately is obliterated and converted into bone before adult life. In simple terms, a synchondrosis is a cartilaginous joint. Three synchondrosis are present along the midline of the cranial base: the spheno-ethmoidal synchondrosis - between the sphenoid and ethmoidal bones -, the sphenoid synchondrosis - between two parts of the sphenoid bone and the spheno-occipital - synchondrosis between the sphenoid and basioccipital bones (12). The spheno-occipital synchondrosis is the site of union between the occipital and sphenoid bones, situated in the clivus area at the base of the skull, anterior to the foramen magnum and inferior to the pituitary fossa. To date, this synchondrosis has been studied mainly from the point of view of growth of the cranial base (13) and its relationship to dento-alveolar development (14). There are different ideas about its reliability as ethnical and genetic factors seem to have a significant role in determining cranial suture patterns and closure. Many radiological studies about its possible use in forensic pathology for age determination purposes have been performed.

Many authors have focused their attention and research on the spheno-occipital suture. In the next table are reported the scientific results, divided according to the sample used for the study (living/dead person or skeleton rests) and the observation method (Table 1).

The aim of the present study is to determine the sequence and timing of closure of the spheno-occipital synchondrosis on a sample of Italian population, subjected to 3D CBCT.

Given the importance of age-assessment in forensic examinations, the aim of this study is to improve the knowledge in this field and increase the reliability of the gathered data not only for identification, but also for judicial purposes. In order to achieve this goal, unlike similar studies reported in literature, we have analyzed the relationship between age and bone fusion in an alive population sample by using an imaging technique, such as the 3D CBCT scan, which provides a higher definition and lower emission of radiation than conventional X-Rays. We strongly believe that the extent of the studied population together with the technical characteristics of the chosen imaging method guarantee the reliability of the obtained data, providing useful forensic pathologist information for all those cases of judicial interest in which age-determination is crucial.

Material and Methods

The studied sample consisted of cone-beam computerized tomography (3D CBCT) scans of 22years-old Italian living individuals (244 males, 250 females) provided by the Department of Radiology, Oncology and Anatomo-Pathology of "Sapienza" University of Rome – UOC Head and Neck Radiology– and the Department of imaging, University of L'Aquila, CBCT was performed without a contrast medium, by using the NewTom Vg1 Vertical Cone Beam (New-Tom, Verona, Italy). The following technical parameters were used to volumetrically acquire the region to be analyzed: 110kVp, 1-20 mA (pulsated mode), focal spot 0.3 mm, field of view 15x15 cm, and amorphous silicon flat panel. A 20-second acquisition determined an exposure of approximately 3.5 seconds (pulsated), with an estimated dose of about 50μSv. The images were reworked according to the axial, sagittal and coronal planes. CBCT images were viewed on reconstructions according to the axial plane and median sagittal plane MPR (Fig. 1). The exclusion parameters are significant trauma and/or pathology of the skull. The ossification status of the spheno-occipital synchondrosis was assessed using a five-stage system which differs from the one developed by Powell and Brodie (15) and Bassed – Drummer (16). The stages are the following: in stage 1, the synchondrosis is completely open appearing as a hypodense zone; absence of calcification in the joint space (Fig. 2). In stage 2 the superior border has fused whilst the remainder of the fusion site is still open (Fig. 3). In stage 3, half of the length of the synchondrosis is closed (Fig. 4). In stage 4, closure is essentially complete, but the site is still visible through a fusion scar (Fig. 5), and in stage 5 the site has been completely obliterated with the appearance of normal bone throughout (Fig. 6). Stage 4 is a new stage of the growth, added specifically because of the ability to visualize the fusion scar on high resolution CT images, which is not possible with conventional radiography or upon dried skulls. Descriptive and inferential analyses have been realized. In particular for the groups of subject classified into the 5 levels, have been calculated absolute and percentage frequencies towards the qualitative variable: "sex". Furthermore, we have been calculated means, standard deviations, maximum and minimum values for the specific variable: "age". These parameters to identify and to evaluate the values statistically significant differences between age and suture level, an Anova test has been applied (significance limit $\alpha = 0,05$) and, to evaluate in which associated patterns could exists a significant difference, the Tukey ($\alpha \leq 0,05$) post-hoc test. To establish statistical important differences of the spheno-occipital closure between men and women, a Student *t* test have been used; this test compares the mean age of the two groups on the basis of the spheno-occipital closure (significant limit $\alpha = 0,05$). Moreover, the correlation between age and suture level has been found using Spearman's Rho ($\alpha \leq 0.05$). In the end, to quantify the relation between age and suture level, a linear regression model has been used with age as dependent variable and suture level (with values from 1 to 5) as independent variable; the two coefficients of the model, their significance ($\alpha \leq 0.05$), the confidence level (95%) and the R^2, which indicates the percentage of variance of the dependent variable explained by the two independent variables, have been analyzed.

Results

The sample of 494 living patients aged 0-22 is made up by 49.4% (n=244) of males and 50.6% (n=250) of females. Observing the age distribution, the most part of the subjects was 14-years-old (39 people); then, we had 38 young people being 12-years-old and 38 were 13-years-old; the 23.3% have an age between 12 and 14. The less represented age is the maximum age, 22, which is presented by only one subject. The 24.3% (n=120) of the subjects is classified into the first suture level, the 17.6% (n=87) into the second, the 13.4% (n=66) into the third, the 21.1% (n=104) into the fourth and the 23.7% (n=117) into the fifth (Table 2). Within every group, patients are distributed as follows: on the first suture level 44.2% (n=53) are males, the 55.8% (n=67) are females; into the second suture level the 58.6% (n=51) are males and the 41.4% (n=36) are females; into the third level the 51.5% (n=34) are males and the 48.5% (n=32) are females, into the fourth level the 43.3% (n=45) are males and the 56.7% (n=59) are females, into the fifth level the 52.1% (n=61) are males and the 47.9% (n=56) are females. The mean age of patients with open suture (first level) is 7.7 years old (SD=3.1), with minimum age as 0.1 years old and maximum age as 15.5 years old; the mean age classified into second suture level is 10.6 years old (SD=3.2), with minimum age 0.9 and maximum age 15.8; into third level mean age is 12.6 (SD=3.1), with minimum age 0.2 and maximum age 18.4; into the fourth level mean age is 16.4 (SD= 2.8), with minimum 6.8 and maximum 21.7; finally, the mean age when the suture is completed (fifth level) is 17.5 (SD=2.6), with minimum 6.6 and maximum age 22.1. Anova test shows significant statistical differences among the five age-groups ($p<0.001$); specifically, the Tukey post-hoc test, which compares every pair, shows a significant difference for every age-couple (the p value is, for every comparison, always less than 0.001) as well as some significant values very near to the limit level for the couple IV/V suturing level (p=0,054), which indicates a similitude of the age for the groups belonging to the IV/V suturing levels. Examining the mean age of every gender and in each and every suturing level, the attention is drawn by the data according which female people reach before male companions the suturing in each level. If we analyze exclusively the V grade of suturing – the one presenting the complete spheno-occipital closure – we can see that 117 people are included in this level (in the amount of 23.7% out the total sample), whose 52.1% (n=61) are male and the residual 47.9% (n=56) are female. The average age of the spheno-occipital closure in men is 18.3 years (DS=2), whilst in women is 16.6 (DS=2.9). Therefore, it is evident that the closure in female people is faster than 1.7 years; this difference is statistically important for the t test of Student ($p<0.001$) (Table 3) Correlation between suture levels and age is high and positive, with a Spearman's Rho equal to 0.80 ($p<0.01$ – two-tailed), to indicate that a strong direct correlation between the two variables exists: the suture level grows as age grows. The linear regression model, with age as dependent variable and suture level as independent variable, shows that as the suture level increases also the age increases of 2.5 years (CI 95% 2.4-2.7). The model interpolates well the observations, with an R- Square of 0.625, indicating that the 62.5% of the dependent variable variance is explained by the independent variables.

Discussion

The techniques used to study the timing of closure of spheno-occipital suture are many and different: direct observation (17-18), histological exams and x-ray imaging (19). The last technique – X-ray imaging – especially the CT, allows to highlight middle stages of closure that can not be seen through other techniques. The values obtained as result depend also from the examined sample: dead bodies and bone rests or living people. We report the results of studies carried out over dead bodies. Ford (20), in his study about dry skulls, indicated as the age of closure of spheno-occipital synchondrosis ranges between 17 and 25 years; according Irwin (21), the complete fusion happens at 18 years. Powell and Brodie (15), through X-ray, concluded that the complete fusion happens between 13 and 16 years in men and between 11 and 14 years in women, at least in American people. Melsen (22) reported the age range 12 - 17 years, linking it with the eruption of II and III molar tooth (23). Ingervall and Thilander (18), observing directly Swedish skulls, reported the age of 13 years for female and 16 years for males. Mann *et al*. (24), examining an American people sample, stated that the spheno-occipital synchondrosis completes its closure at 16 years in women and 18 years in men. El-Sheikh e Ramadan (25), using CT over a sample of Arabian people, observed that the complete closure of spheno-occipital synchondrosis happens at 18 years in male and at 16 years in female. Coqueugniot et al (26) highlighted, through the direct observation of a Portuguese dry skull that the closure of the suture happens at 14 years in women and at 19 in men. Akhlaghi et al (17), through direct observation of an Iranian sample, has decreeded that the closure of spheno-occipital suture ends in male at 21 years and at 19 years in female. Bassed et al (16), through CT, studying an Australian sample, has observed that the closure of synchondrosis is complete at 17 years, both in men and women. Shirley e Jantz (27), through direct observation of an American sample, has ascertained that the complete fusion of spheno-occipital synchondrosis happens at 23 years in males and 20 years in females. Lingawii (28), through CT, has reported that, in Arabian people, the fusion of suture can be observed from 15-years-old, for both genders, whilst Lottering et al (29) indicated 16-years-old for males and 14 years for females, examining an Australian sample. The researches carried out over living people use always the CT method. Madeline and Elster (30), examining an American sample, indicate how the closure of spheno-occipital closure happens at 16-years-old in women and 18-years-old in men. Okamoto et al (31), studying Japanese people, both male and female, reports the age of 13. Usama et al (32) indicates an age older than 15 years for women and older than 16 years for men. Franklin and Flavel (33) have reported the closure of the suture, in an Australian sample, at 10 years in girls and at 13 years in boys. Only one article has a mixed sample (living people and dead bodies): Sahni et al (34), over an Indian sample, set the complete fusion of spheno-occipital suture between 13 and 17 years in females and between 15 and 19 years in males. The results of this study are different because of the kind of the sample, the methodology and

the statistical analysis: first of all, we have to consider the quantity and the homogeneous allocation of the sample, parameters that strongly affect the efficiency of the results. For example, the sample used by Kahana et al (35) was formed by 21 subjects, exclusively female. The absence of just one and only classification of suturing status as well as of a statistical analysis are the reasons of the missing standards for the results. These differences depend also by the ethnic group and their anthropologic-social elements. In this study, the sample is wide (494 subjects), but selected according geographical area (Italy), balanced between women and men (49.4% male; 50.6% female) and divided into the five classification groups according the gender and age (reference in tables and graphics in Results Section). The technical method here used, CBCT, is innovative and has never been used to these aims. This technique provides high quality images using very few radiations, MPR reconstructions of the picture on all planes of the area and with lower costs than traditional CT. The statistical analysis has confirmed the correlation between suture levels and age exist and show value high and positive, with a Spearman's Rho equal to 0.80 ($p<0.01$ – two-tailed), to indicate that a strong direct correlation between the two variables exists: the suture level grows as age grows. The linear regression model, with age as dependent variable and suture levels as independent variables, shows that increasing the suture level also the age increases of 2.5 years. Talking about the mean age when the complete fusion of spheno-occipital suture happens, the examined sample reports the full occlusion at 18.3-years-old in men and at 16.6-years-old in women. On the other hand, the fusion of the suture starts (II grade) at 9.9-years-old in female and 11-years-old in males. It is confirmed that the bone development ends about 1.7 year sooner in women than in men. Only the articles of Irwin (21), Okamoto (31), Bassed (16) and Lingawii (28) don't show any differences between sexes. Our results match with those by Madeline, which used a pentaphasic classification system, a sample formed by living people, but coming from USA. However, our data are not so different from the ones reported into the literature, most of all if we consider that the several studies analyze different geographical groups and that no other studies carried out over Italian population are available. Okamoto (31), analyzing a Japanese sample, reports as age of fusion 13-years-old both for men and women; Usama et al (32), over a Yemeni sample, indicates as fusion age 15-years-old in female subjects and 16-years-old in male; according to Franklin et al (33), in an Australian study, the complete occlusion is set at 10-years-old in girls and 13-years-old in boys. If we compare our results with those obtained by dead bodies samples, they will match with El-Sheikh et al (25) ones but will be very different than others. That depends by the research method (macroscopic observation, Rx or CT) and the status of the dead bodies or their skeletal rests, beyond their geographical belonging. These results are promising and suggestive that scientific community has seen the big picture, considering the spheno-occipital suture an age indicator.

Conclusion

The timing of closure of spheno-occipital suture can be used to understand the age, even if the research has to be implemented. The next studies will have to use samples that have a statistical value, selected according the ethnic groups or the geographical areas, favoring the age of adolescence. To corroborate the chosen technique, we reaffirm that CBCT method provides with accurate imaging using very few radiations, so that it can be used also with living people.

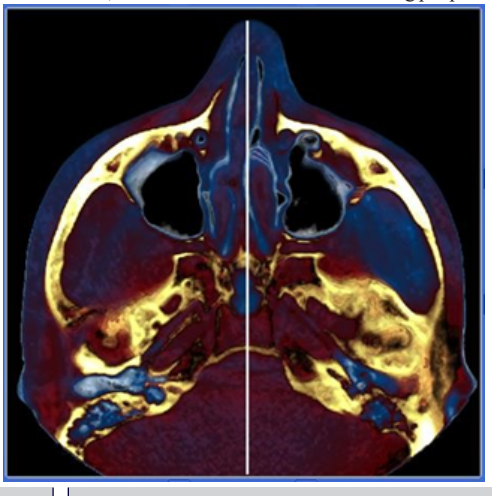

FIG. 1–3D cone-beam image, the processing emphasizes the acquisition plan used for our study.

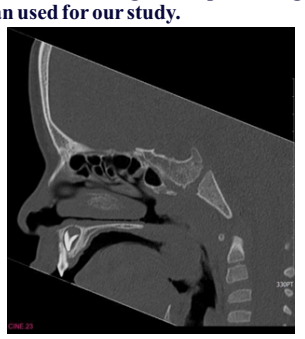

FIG. 2–3Dcone-beam image, sagittal reconstruction. Grade I. synchondrosis fully open.

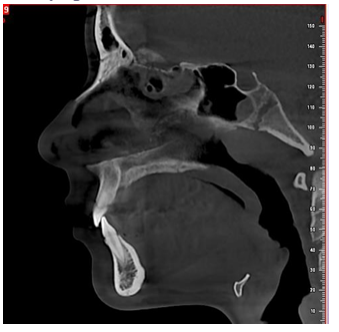

FIG. 3–3Dcone-beam image, sagittal reconstruction. Grade II upper edge of the suture partially melted.

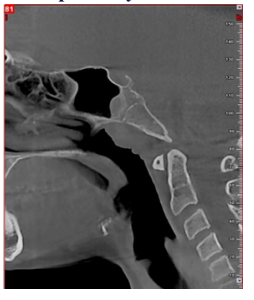

FIG. 4–3Dcone-beam image, sagittal reconstruction. Grade III, evident rhyme suture ossified but not closed.

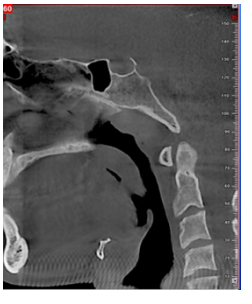

FIG. 5–3Dcone-beam image, sagittal reconstruction. Grade IV suture fused which is still appreciable rhyme conjugation.

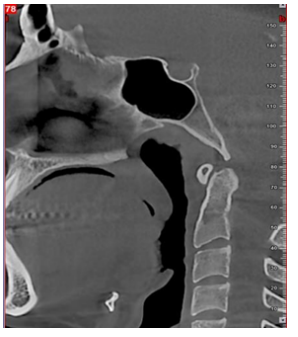

FIG. 6–3Dcone-beam image, sagittal reconstruction. Grade V, synchondrosis completely melted.

AUTHOR	METHODIC	POPULATION	FUSION AGE	
			M	F
Madeline et al	CT	UnitedStates	18	16
Okamoto et al.	CT	Japanese	13	13
Shani et al.	CT	Indian	15-19	13-17
Mann et al.	CT	UnitedStates	18	16
Usama et al.	CT	Yemeni	16	15
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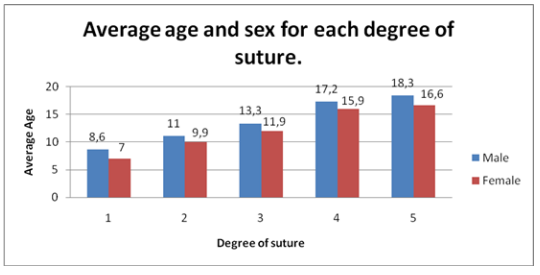
TABLE 1- Other authors on suture spheno-occipital in living population. Modificated from summary by Lottering N, MacGregor DM, Alston CL, Gregory LS. Ontogeny of the sphenooccipital synchondrosis in modern Queensland, Australian population using computed tomography.Am J PhysAnthropol. 2015 May;157(1):42-57.

Closure suture degree	N	%
1	120	24,3
2	87	17,6
3	66	13,4
4	104	21,1
5	117	23,7
Tot	494	100

TABLE 2- Table showing the distribution by degree of suture sample study. Absolute data and percentages



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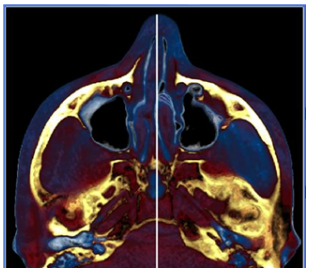
Figure(s) and caption(s)



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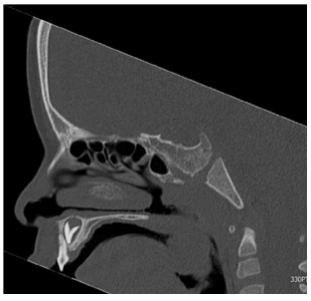

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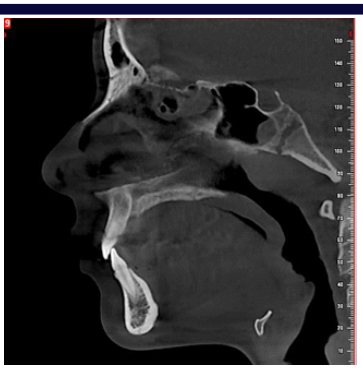

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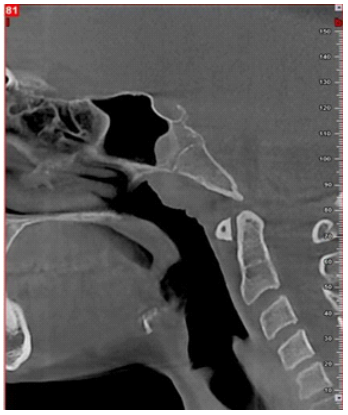

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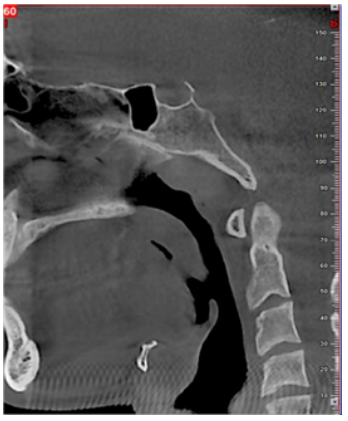

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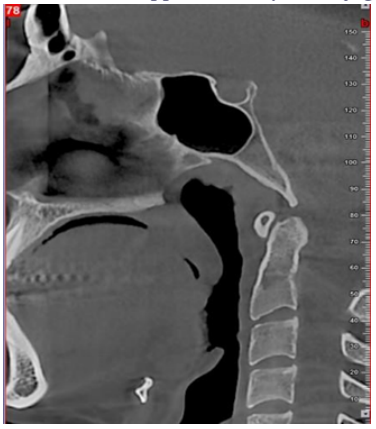

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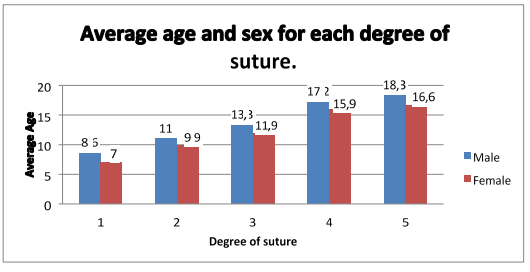

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