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Short report

Age estimation from clavicular epiphyseal union sequencing in a Northwest Indian population of the Chandigarh region

Jagmahender Singh MSc, PhD, Anthropologist *, K.H. Chavali MBBS, MD, DNB, PGDHA, Assistant Professor

Department of Forensic Medicine & Toxicology, Govt. Medical College Hospital, Sector 32, Chandigarh 160030, India

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ABSTRACT

Medial clavicular epiphysis is the long bone epiphysis to fuse last and therefore, is useful for estimating age in the post-pubertal period. Age estimation was done from clavicles obtained from 343 cadavers (252 males and 91 females) of known age. The data was subjected to statistical analysis to see whether the difference obtained in the various ages was significant or not. Commencement of fusion was seen as early as 18 years of age in both male and female clavicles. No clavicle showed complete fusion until the age of 22 years. Complete fusion of the medial end of the clavicle was seen latest at 32 years in the male clavicles while the same was observed at 31 years in the female bones, i.e., the medial epiphysis of female clavicles fused one year earlier than their male counterparts. Advanced stages of clavicular epiphyseal union were seen in most of the clavicles after 24 years of age in females and 23 years of age in males. No significant difference was noticed in both sexes for the right and left clavicles as regards to the occurrence of various stages of epiphyseal union.

1. Introduction

Age estimation of unidentified human skeletal remains is one of the many objectives that are required to be achieved not only in forensic practice but also in physical anthropology for the purposes of identification and biological profiling. The changes of developmental processes like epiphyseal–diaphyseal union, dental eruption patterns, etc. as well as those of the degenerative processes like dental attrition, etc. are useful for estimation of age of such skeletal remains. Both these developmental and degenerative processes affect different anatomical structures/regions of the human body at different stages of life in a more or less regular and recognizable pattern depending on the stage of life and the anatomical region/part being observed.[1] Different aging methods are available for different anatomical regions of the human body like dentition, epiphyseal–diaphyseal union, cranial suture closure, morphology of the auricular surface of ilium, fusion of sternal and sacral elements and phase analysis of sternal end of ribs; but due to destructive taphonomic processes or commingling factors, few or none of these age diagnostic indicators/features of bones may be available for analysis in archaeological, palaeontological or forensic materials, thus making it essential to develop reliable aging methods from a variety of long bones including the clavicle.

It is obvious that the problem of identifying a recently dead person is totally different from identifying one having undergone advanced decomposition, skeletonization or purposeful dismemberment aimed at hindering recognition. Personal effects need cautious interpretation since relatives'/friends' identification might be coloured by emotions or ulterior motives. Availability of bones under such circumstances often proves highly useful in approaching identification. Bones can often provide leads in determining age as well as sex (primary parameters for identification) of the deceased. The human clavicle displays the longest period of growth related activity than other long bones, thus retaining its predictive value as a useful and reliable indicator of age in the first three decades of life when other growth related indicators would have become inactive and/or useless for this purpose. Of the long bones, medial clavicular epiphysis is the last to fuse and therefore, its different developmental stages have greatest discriminatory value, being useful for estimating age at death in the post-pubertal period.[2,3]

Both osteological and radiological examination of medial end of the clavicle are prescribed for studying the union of clavicular epiphyses and the two methods produce differing results.[4] As the methods based on radiological sources cannot be directly applied to dry bone material owing to the intrinsic optical and soft tissue complications, examination using bone material is preferred because of its reliability and accuracy.[5–13]

Populations of geographically different regions exhibit different anthropological observations due to various factors. Thus, formulae derived for estimation of age, sex or stature for one population

* Corresponding author. Tel.: +91 172 2665253x1064.
E-mail addresses: jagmindera@hotmail.com (J. Singh), drkhchavali@gmail.com (K.H. Chavali).

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Table 1
State-wise distribution of study population. [Males ($n = 252$ pairs), Females ($n = 91$ pairs)].

Age interval (years)	Punjab		Haryana		Himachal Pradesh		Chandigarh		Western U.P.		Total
	M	F	M	F	M	F	M	F	M	F	
17–25	19	19	28	9	14	4	6	4	6	1	110 (32.07%)
26–35	24	8	30	10	11	3	6	1	4	0	98 (28.57%)
36–45	21	2	13	0	8	1	3	0	4	0	52 (15.16%)
46–55	12	8	6	4	4	2	1	0	1	0	38 (11.08%)
56–65	10	4	2	3	4	1	1	0	0	1	26 (7.58%)
66–75	5	2	3	1	1	1	0	1	0	0	14 (4.08%)
>75	2	0	3	0	0	0	0	0	0	0	5 (1.46%)
Total	93	44	85	27	42	12	17	6	15	2	343 (100%)
	137 (39.94%)		112 (32.65%)		54 (15.74%)		23 (6.70%)		17 (4.96%)		

cannot be uniformly applied to all populations in general. Population specific data, hence, are required to be studied and formulae derived for each population for establishment of the biological profile. Again, the formulae so derived need to be examined periodically to observe any modification that may be required to be made as a result of the various changes in the population parameters over time. Only one study was conducted more than three decades earlier by Jit and Kulkarni[8] on a similar composition of Northwest Indian population of Chandigarh region using radiographs. As bone specimens are considered better than radiographs for the purpose of ageing and sexing,[14] the present study was undertaken with the aim of obtaining more reliable results from the epiphyseal union timings of medial clavicular epiphysis of the current population of the same region. Though it needs to be established by further studies, probably, a number of nutritional, environmental and occupational changes that have occurred in the last 2–3 decades and thus affected the socio-economic status of the population of the region in general,[3] could have resulted in a shift in age-thresholds of timings of different stages of epiphyseal union as compared to the earlier study. Majority of the methods of age estimation using dry bones are either from archaeological material of different populations or from modern populations of different regions that cannot be directly compared with the present study population.[15] So the present study was undertaken with another objective of evaluating some standard of age estimation from the clavicle of the population of the Northwest Indian population, using different epiphyseal union stages.

2. Material and methods

The study was a prospective one conducted on 343 pairs of clavicles (males = 252, females = 91) collected from autopsy cases of adult (17–94 years) cadavers of known age, brought for postmortem examination to the Department of Forensic Medicine, Postgraduate Institute of Medical Education and Research (PGIMER), Chandigarh, during the period between March 2008 and September 2008. Prior clearance had been obtained from the institutional ethics committee for this purpose.

The population selected for this purpose consisted of cadavers ethnically belonging to the five Northwest Indian states/Union Territory, i.e., Punjab, Haryana, Himachal Pradesh, Western Uttar Pradesh and Chandigarh. Table 1 shows the state-wise distribution of subjects of the studied population.

Both side clavicles were removed from the cadavers after obtaining consent of the next kin of the deceased. After removal, the clavicles were subjected to boiling in a solution containing caustic soda, sodium chloride, commercially available Henko® washing powder (Henkel Detergents Ltd., Gurgaon) and liquid ammonia as per the procedure prescribed by Fenton et al.[16] The bones showing any grossly visible pathology, fracture or any deformity were excluded from the study sample. The bones were boiled carefully for 10–15 min or until their muscular coverings were removed. While boiling, repeated careful inspection of the bones was done to ensure that the clavicular epiphyses were retained. Muscular attachments, if any remaining after boiling, were removed manually by scraping carefully with blunt scalpel. The wet bones were then wiped with a piece of gauze and dried using a hair-dryer to remove any residual moisture on the surface. The bones were examined for various stages of epiphyseal union of the inner (medial) end of the clavicles. No prolonged defatting or drying process was performed on the bones as they were to be replaced into the body before its handing over to the relatives after conduction of the postmortem examination.

A score was assigned to each clavicle based on the stage of epiphyseal union of its medial end, according to criterion adopted by Schaefer and Black.[11] The scores used were: 0 = no fusion, 1 = fusion commencing or less than one-third of the epiphysis showing fusion, 2 = active fusion or approximately one-half of epiphysis showing union, 3 = recent fusion or more than three-fourths of the epiphysis showing union, 4 = complete union.(Figs. 1–5)

3. Results

Table 2 shows the number and percentages of right and left clavicles at different stages of epiphyseal union. No statistically significant difference was noticed in the observations between the right and left clavicles as regards to incidence/occurrence of different stages of epiphyseal union. Commencement of fusion was seen as early as 18 years of age in both male and female clavicles of either side. No clavicle showed complete fusion until the age of 22

Fig. 1. No fusion.

Fig. 2. Fusion commencing.

Fig. 4. Recent fusion.

years. Complete fusion was seen for the first time at the age of 22 years in both sexes, however, a high percentage of clavicles showed active process of fusion at that age. Advanced stages of clavicular epiphyseal union were observed after the age of 24 years in females and 23 years in males. It was noticed that by 31 years of age, all female cases showed complete fusion. Furthermore, the medial epiphysis of female clavicles fused one year earlier than their male counterparts.

As seen from Table 3, no significant difference was observed in frequency distribution of different stages of epiphyseal union in clavicles of either side between the two sexes. Remarkably significant age differences in the degree of epiphyseal union were observed in right as well as left clavicles of both sexes, as no incomplete fusion was noticed in females beyond 31 years of age and only one pair of clavicle of a male showed recent fusion in the 30+ age-group. On the other hand, in the age group of 17–30 years, complete fusion in female clavicles was observed to be about 10% less than that in male clavicles (Table 4). The differences in stages of epiphyseal union between two age-groups in each sex were found to be statistically highly significant. But, within the same age group, no significant sex differences were found (Table 5).

4. Discussion

The clavicle has been studied extensively in both forensic and archaeological anthropology because it survives burial/decomposition/putrefaction rather successfully (owing to a relatively higher proportion of compact bone) thus, helping in estimation of age and sex of young adults.[17–19] It displays a uniquely prolonged period of growth related activity and so is arguably one of the most valuable specimens in the estimation of age at death.[2] A clavicle with no evidence of a fused or fusing epiphysis is most likely to have come from an individual less than 18 years of age. Complete fusion is unlikely before 22 years and is nearly complete by around 30 years of age.[2,5,9,20] Age estimation based on extent of epiphyseal union or ossification patterns of medial clavicle has been studied by many workers using bone specimens,[7,9,15] radiographs,[21] computed tomographs,[22] magnetic resonance imaging,[23] ultrasounds,[24] etc., and it has been found to be a useful indicator of age in the early years of life.[7–10,15,22,25,26] McKern and Stewart[7] found that epiphyseal union of medial clavicle in young American males starts at the age of the 23 years and at 30, clavicles in some individuals are still seen to be in a state of active fusion. Webb and Suchey[9] found that epiphyseal union of medial clavicle in a modern American sample starts earlier in females than in males and complete union occurs at 21 years or older in both the males and females. Female standards can

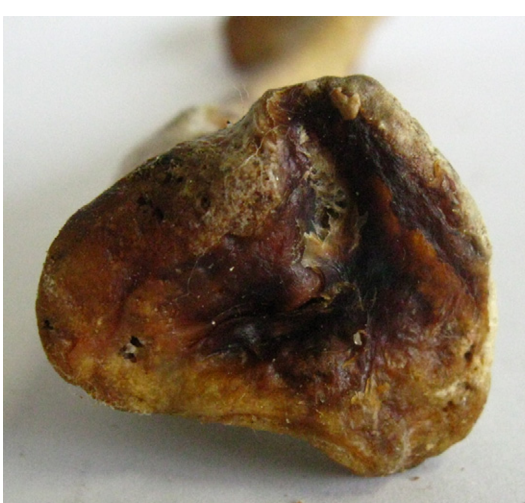

Fig. 3. Active fusion.

Fig. 5. Complete fusion.

Table 2
Frequency distribution of different stages of epiphyseal union in right and left clavicles.

Age (years)	Sex	Total no. of cases	No fusion [N, (%)]		Fusion commencing [N, (%)]		Active fusion [N, (%)]		Recent fusion [N, (%)]		Complete fusion [N, (%)]	
			Right	Left	Right	Left	Right	Left	Right	Left	Right	Left
17	M	4	4 (100)	4(100)	–	–	–	–	–	–	–	–
	F	2	2 (100)	2(100)	–		–	–	–	–	–	–
18	M	5	3(60)	2(40)	1(20)	2(40)	1(20)	1(20)	–	–	–	–
	F	5	3(60)	1(20)	2(40)	4(80)	–	–	–	–	–	–
19	M	8	2(25)	1(12.5)	4(50)	5(62.5)	1(12.5)	1(12.5)	1(12.5)	1(12.5)	–	–
	F	2	1(50)	–	–	1(50)	–	–	1(50)	1(50)	–	–
20	M	13	1 (17.7)	1(7.69)	5(38.46)	5(38.46)	4(30.77)	4(30.77)	3(23.08)	3(23.08))	–	–
	F	5	–	1(20)	3(60)	2(40)	1(20)	–	1(20)	2(40)	–	–
21	M	5	1(20)	–	–	1(20)	2(40)	2(40)	2(40)	2(40)	–	–
	F	2	1(50)	–	–	1(50)	–	1(50)	1(50)	–	–	–
22	M	7	–	–	1 (14.29)	1(14.29)	3 (42.86)	2(28.57)	1 (14.29)	2(28.57)	2 (28.57)	2(28.57)
	F	8	–	–	2 (25)	1(12.5)	3 (37.5)	2(25)	1 (12.5)	3(37.5)	2 (25)	2(25)
23	M	7	–	–	1(14.29)	1(14.29)	–	–	4(57.14)	4(57.14)	2(28.57)	2(28.57)
	F	5	–	–	–	–	1(20)	1(20)	3(60)	2(40)	1(20)	2(40)
24	M	9	–	–	–	–	2(22.22)	2(22.22)	1(11.11)	1(11.11)	6(66.67)	6(66.67)
	F	5	–	–		–	2(20)	2(22.22)	3(60)	2(66.67)	–	1(33.33)
25	M	15	–	–	–	–	3(20)	4(26.67)	7(46.67)	4(26.67)	3(33.33)	7(46.67)
	F	3	–	–	–	–	–	1(50)	3(100)	1(25)	–	1(25)
26	M	16	–	–	–	–	–	1(6.25)	4(25)	2(12.5)	12(75)	13(81.25)
	F	5	–	–	–	–	1(20)	1(20)	1(20)	2(40)	3(60)	2(40)
27	M	5	–	–	–	–	–	–	5(100)	5(100)	–	–
	F	1	–	–	–	–	–	1(50)	1(100)	–	–	1(50)
28	M	7	–	–	–	–	–	–	4(57.14)	3(42.86)	3(42.86)	4(57.14)
	F	4	–	–	–	–	–	–	–	1(25)	4(100)	3(75)
29	M	3	–	–	–	–	–	–	1(33.33)	1(33.33)	2(66.67)	2(66.67)
	F	2	–	–	–	–	–	–	1(50)	–	1(50)	2(100)
30	M	14	–	–	–	–	–	–	8(57.14)	5(35.71)	6(42.86)	9(64.29)
	F	1	–	–	–	–	–	–	1(100)	1(100)	–	–
31	M	3	–	–	–	–	–	–	1(33.33)	1(33.33)	2(66.67)	2(66.67)
	F	1	–	–	–	–	–	–	–	–	1(100)	1(100)
32	M	6	–	–	–	–	–		1(16.67)	–	5(83.33)	6(100)
	F	2	–	–	–	–	–	–	–	–	2(100)	2(100)

vary 1–2 years from those of males but, in general, the overall timing of epiphyseal fusion in both sexes is similar. Ji et al[10] found that epiphyseal union proceeds faster in females than in males and comparatively more faster in Japanese males than in American males. Krietner et al[15] tomographically studied the medial extremity of clavicle and concluded that ossification of clavicular epiphysis and its union lasts for several years and may be a useful adjunct in forensic age estimation in living individuals. Schaefer and Black[12] proposed that appropriate population specific standards of epiphyseal union of medial clavicle should be devised as Bosnian clavicles studied by them started and attained complete union 1–3 years earlier than the Americans. Schulze et al[22] proposed that computed tomography of medial clavicle epiphysis will only be suitable for age estimation around the age of 21 years in living persons.

It is established that in the long bones, the epiphyseal ossification centers appear and fuse with the metaphyses earlier in females than males.[8,9,25,27] But earlier, Todd and D'Errico[6] and Flecker[28] had categorically denied any such sexual differences in appearance and fusion of these epiphyses of the sternal (medial) end of clavicle. Galstaun[27] further opined that the earliest complete fusion took place one year later in the females than the males. Jit and Kulkarni[8] reported that in both sexes the earliest partial fusion of medial epiphysis occurs at the age of 18 years and latest by 23 years whereas earliest complete fusion occurred at the age of 22 years in males and at 23 years in females. They further opined that the age of a person showing partial fusion can be estimated to be above 18 years but below 24 years and a person with a complete fusion must be above 22 years of age. The age at which epiphyseal ossification commences, fusion begins and total fusion occurs, shows a considerable degree of variability which, in turn, can be explained in terms of individual maturation timings that are influenced by a variety of predisposing hereditary and environmental factors.[2]

All the studies conducted earlier were done either on dry bones or using radiographs, ultrasounds or computerized tomographic scans (Table 6). The present study was conducted on fresh bones obtained from the bodies of cadavers brought for autopsy and hence some difference in observations between the previous studies and the current study was assumed. In the present study, the earliest age at which the process of union was observed was 18 years which is in agreement with the observations of most of the previous studies except for the observation of Kreitner et al.[15] where earliest fusion was observed at 16 years. The difference could be probably due to the difficulty in evaluating the CT scans which were used for the study. There is however, some disagreement on the latest age when partial union was observed. The current study shows the latest age showing partial union as 31 years which is higher than the latest age

Table 3
Sex-wise frequency distribution of different stages of epiphyseal union in clavicles of either side.

Side	Sex		No fusion	Fusion commencing	Active fusion	Recent fusion	Complete fusion	Chi-square
Right	Male ($N = 252$)	n (%)	11 (4.4)	12 (4.8)	16 (6.3)	42 (16.7)	171 (67.9)	4.355[a]
	Female ($N = 91$)	n (%)	7 (7.7)	7 (7.7)	8 (8.8)	17 (18.7)	52 (57.1)	
Left	Male ($N = 252$)	n (%)	8 (3.2)	15 (6.0)	17 (6.7)	34 (13.5)	178 (70.6)	3.592[a]
	Female ($N = 91$)	n (%)	4 (4.4)	9 (9.9)	8 (8.8)	15 (16.5)	55 (60.4)	

[a] $p > 0.05$, not significant.

Table 4
Comparative frequency distribution of different stages of epiphyseal union between two age-groups of same sex in left and right clavicles.

Side	Sex	Age-group (years)		No fusion	Fusion commencing	Active fusion	Recent fusion	Complete fusion	Chi-square
Right	Male ($N = 252$)	17–30	n	11	12	16	41	38	129.379[a]
		($N = 118$)	%	9.3	10.2	13.6	34.7	32.2	
		30+	n	–	–	–	1	133	
		($N = 134$)	%				0.7	99.3	
	Female ($N = 91$)	17–30	n	7	7	8	17	11	55.965[a]
		($N = 50$)	%	14	14	16	34	22	
		30+	n	–	–	–	–	41	
		($N = 41$)	%	–	–	–	–	100	
Left	Male ($N = 252$)	17–30	n	8	15	17	33	45	113.063[a]
		($N = 118$)	%	6.8	12.7	14.4	28	38.1	
		30+	n	–	–	–	1	133	
		($N = 134$)	%	–	–	–	0.70%	99.30%	
	Female ($N = 91$)	17–30	n	4	9	8	15	14	48.842[a]
		($N = 50$)	%	8	18	16	30	28	
		30+	n	–	–	–	–	41	
		($N = 41$)	%	–	–	–	–	100	

[a] Significant ($p < 0.01$).

Table 5
Comparative frequency distribution of different stages of epiphyseal union between the two sexes of same age-group in left and right clavicles.

Side	Age-group (years)		Sex	No fusion	Fusion commencing	Active fusion	Recent fusion	Complete fusion	Chi-square value
Right	17–30	Male	n	11	12	16	41	38	2.579[a]
		($N = 118$)	%	9.3	10.2	13.6	34.7	32.2	
		Female	n	7	7	8	17	11	
		($N = 50$)	%	14	14	16	34	22	
	30+	Male	n	–	–	–	1	133	0.308[a]
		($N = 134$)	%	–	–	–	0.7	99.3	
		Female	n	–	–	–	–	41	
		($N = 41$)	%	–	–	–	–	100	
Left	17–30	Male	n	8	15	17	33	45	1.899[a]
		($N = 118$)	%	6.8	12.7	14.4	28	38.1	
		Female	n	4	9	8	15	14	
		($N = 50$)	%	8	18	16	30	28	
	30+	Male	n	–	–	–	1	133	0.308[a]
		($N = 134$)	%	–	–	–	0.7	99.3	
		Female	n	–	–	–	–	41	
		($N = 41$)	%	–	–	–	–	100	

[a] $p > 0.05$, not significant.

observed in almost all the earlier studies except the observations of Webb and Suchey[9] on female clavicles, where it was observed to be 33 years. The lowest observation of 24 years was reported by Galstaun[27] and Jit and Kulkarni,[8] both studies having been conducted using radiographs on different Indian populations. The other study by Flecker[29] using radiographs was conducted on an Australian population and also gives almost a similar observation i.e., 25 years as the latest age demonstrating partial fusion. However, conventional radiographs of medial clavicular epiphyses used for the purpose of aging the living individuals have been found to be often suboptimal because of overlapping ribs, vertebrae and mediastinal shadows[29–31] which can prevent the determination of the exact

Table 6
Comparative analysis of epiphyseal union timings of medial clavicle (age in years).

Study	Year	Ethnicity & sample/method	Sample size	Age range of partial union	Latest age of non union	Age at earliest complete union	Age at latest incomplete union	Age at complete union in all cases
Todd and D'errico[6]	1928	American (Dry bones)	166 (♂–130 ♀–36)	19–27	–	23	27	28,[a] 29[b]
McKern and Stewart[7]	1957	American (Dry bones)	374 (♂)	18–30+	18	23	30+	31
Webb and Suchey[9]	1985	American (Dry bones)	859 (♂–605, ♀–254)	17–30♂, 16–33♀	24♂, 23♀	21 (both sexes)	30♂, 33♂	31♂, 34♂
Schaefer and Black[12]	2005	Bosnian (Dry bones)	114 (♂)	?–28	22	21	28	29
Schaefer and Black[13,14]	2007	Bosnian (Dry bones)	258 (♂)	17–29	23	21	29	30
Flecker[29]	1933	Australian (Radiographs)	437	?–25	–	–	25	26
Galstaun[27]	1937	Bengali (Radiographs)	654 (♂–445, ♀–209)	18–24	–	19♂, 20♀	24♂, 23♀	25♂, 24♀
Jit and Kulkarni[8]	1976	North Indian (Radiographs)	684 (♂–391, ♀–293)	18–24	21 (both sexes)	22♂, 23♀	24♂, 23♀	25♂, 24♀
Kreitner *et al.*[15]	1998	German (CT scan)	380 (♂–229m, ♀–151)	16–26	16	22	26	27
Present study		North-West Indian (Fresh bones)	360 (♂–254m, ♀–106)	18–31	21 (both sexes)	22 (both sexes)	31♂, 30♀	32♂, 31♀

♂ = Male.
♀ = Female.
[a] = Whites.
[b] = Blacks.

stage of medial epiphyseal development.[27,32] Complete union in all cases was seen at 32 years of age, which is higher than the observations of all the previous studies.

Wherever, the sex difference in fusion has been studied, it has been observed that complete union occurs earlier in the females as compared to males by around one year. The present study findings in this aspect are in agreement with those of other studies.

Only one other study[10] gives the latest age of complete union to be earlier in males than in females. The studied population however, was an American one. Recent studies on American populations have yielded different results. The differences in age for different stages of epiphyseal union of the clavicle as given by different authors including the present study could, perhaps, be attributed to different methodologies of data collection and/or scoring techniques. So, it may be concluded that the radiographs and bone specimens give different estimates of age on the basis of study of different stages of epiphyseal union of medial end of the clavicle. Also, factors like the enhanced socio-economic status of an individual[3] might be responsible for the differences in the age estimates between the present study and the earlier study conducted on the same population by previous workers more than three decades earlier.

5. Conclusions

In the present study, all left sided clavicles showed complete union one year earlier than the right clavicles, which however, was not statistically significant. The medial epiphysis of female clavicles fused one year earlier than their male counterparts. Further, it was observed that both male and female clavicles equally showed latest incomplete fusion at 31 and 30 years of age respectively. On the other hand, 39% of male clavicles showed complete fusion at 32 years' age as compared to 29% cases among females at 31 years' age. A male or female clavicle showing complete fusion or no fusion can probably be more than 22 years and less than 21 years, respectively. It was noticed that by 31 years of age, all female cases showed complete fusion. No definite comment could be made about the timings of earliest commencement of epiphyseal union as the individuals younger than the age of 17 were not considered in the present study.

Conflict of interest statement

There is no conflict of interest of any author.

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Ethical approval

None.

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