Quantitative anatomy of the growing clavicle in the human fetus: CT, digital image analysis, and statistical study

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Abstract

Purposes Knowledge of dimensions of fetal long bones is useful in both the assessment of fetal growth and early detection of inherited defects. Measurements of the fetal clavicle may facilitate detection of numerous defects, e.g., cleidocranial dysplasia, Holt–Oram syndrome, Goltz syndrome, and Melnick–Needles syndrome.

Methods Using the methods of CT, digital image analysis, and statistics, the size of the growing clavicle in 42 spontaneously aborted human fetuses (21 males and 21 females) at ages of 18–30 weeks was studied.

Results Without any male–female and right–left significant differences, the best fit growth models for the growing clavicle with relation to age in weeks were as follows: 

- Length: \( y = -54.439 + 24.673 \times \ln(\text{age}) \pm 0.237 \ (R^2 = 0.86) \)
- Width of acromial end: \( y = -12.042 + 4.906 \times \ln(\text{age}) \pm 0.362 \ (R^2 = 0.82) \)
- Width of central part: \( y = -4.210 + 2.028 \times \ln(\text{age}) \pm 0.177 \ (R^2 = 0.77) \)
- Width of sternal end: \( y = -4.687 + 2.364 \times \ln(\text{age}) \pm 0.242 \ (R^2 = 0.70) \)
- Cross-sectional area: \( y = -51.078 + 4.174 \times \ln(\text{age}) \pm 6.943 \ (R^2 = 0.82) \)
- Volume: \( y = -766.948 + 281.774 \times \ln(\text{age}) \pm 19.6 \times 10 \ (R^2 = 0.84) \)

Conclusions With no sex and laterality differences, the clavicle grows logarithmically with respect to its length, width, and volume, and linearly with respect to its projection surface area. The obtained morphometric data of the growing clavicle are considered normative for their respective weeks of gestation and may be of relevance in the diagnosis of congenital defects.

Keywords Clavicle · Size · Growth dynamics · Digital image analysis · Human fetus · Regression analysis

Introduction

Knowledge of dimensions of fetal long bones is useful in both the assessment of fetal growth and early detection of inherited defects. Overall, abnormalities of the fetal clavicle are rare and may exemplify cleidocranial dysplasia, Holt–Oram syndrome, Goltz syndrome, and Melnick–Needles syndrome [1, 2].

The process of clavicle ossification is the earliest among all long bones. It begins at the end of the 6th week of fetal life, which determines the early development of the upper limb, ensuring its adequate mobility [3–6]. The clavicle has two primary ossification centers that fuse in its central part on day 45 [2, 7]. The sternal end of the clavicle develops most slowly, thus undergoing ossification as the very last of all bones. Developmental anatomy and morphometric data provide clinicians of different specialties with relevant information [3]. In archeological studies, the clavicular model is germane to elucidate some mechanisms of evolution, while, in forensic medicine, it serves to determine sex, age, ethnic differences, and body posture [8, 9].

A review of literature concerning morphometric studies of the clavicle revealed that different imaging methods have consecutively been engaged, namely X-rays, ultrasound, and 3D-ultrasound. In this study, based on much more advanced and objective research methods (CT, digital
image analysis), we decided to perform a comprehensive morphometric analysis of the clavicle in the human fetus. The precise purposes of the study were to:

- carry out morphometric analysis of the fetal clavicle with respect to its linear, planar, and spatial parameters to determine their normative values
- assess possible sex differences regarding all analyzed parameters, and
- establish growth dynamics for all analyzed parameters, including mathematical best-matched models for fetal age in weeks.

Materials and methods

The study material was 42 human fetuses (21 males and 21 females) aged 18–30 weeks, originating from spontaneous abortions and preterm deliveries. As a prerequisite, with the use of CT and morphological examinations, the sample was built by rejection of malformed fetuses with conspicuous internal or external macroscopic abnormalities, and so could be deliberated normal. The fetuses were acquired before the year 2000 and remained part of the specimen collection of the Department of Normal Anatomy of our university. The study was approved by the Bioethics Committee of the Ludwik Rydygier Collegium Medicum in Bydgoszcz (KB 275/2011). The fetal age was determined based on the crown-rump length. Table 1 lists the characteristics of the study group, including age, number, and sex of the fetuses.

Using the Siemens-Biograph 128 mCT camera, the fetuses were scanned at a step of 0.4 mm, recorded in DICOM formats, and subsequently subjected to morphometric analysis using the Osirix 3.9 software. Of note, Osirix 3.9 allows conducting any type of linear, planar, and three-dimensional reconstructions of the studied objects along with their precise quantitative analysis (Fig. 1).

In all fetuses, quantitative assessment of the following six parameters for either clavicle (Figs. 2, 3) was performed:

1. length of the clavicle (mm), equal to the distance between its acromial and sternal ends in the transverse plane,
2. width of the clavicle acromial end (mm), equal to the distance between its anterior and posterior borderlines in the transverse plane,
3. width of the clavicle central part (mm), equal to the distance between its anterior and posterior borderlines in the transverse plane,
4. width of the clavicle sternal end (mm), equal to the distance between its anterior and posterior borderlines in the transverse plane,
5. projection surface area of the clavicle (mm²), based on the determined contour of the clavicle in the transverse plane,
6. volume of the clavicle (mm³), based on advanced spatial reconstruction of images.

In a continuous effort to minimalize measurement and observer bias, all measurements were achieved by one researcher (M.W.) and verified by the same examiner. Each measurement was done three times under the same settings but at different times, and then averaged. The intra-observer variation between repeated measurements was assessed by

| Gestational age | Crown-rump length (mm) | Number of fetuses | Sex |
|-----------------|-------------------------|-------------------|-----|
| Weeks (Hbd-life) | Mean | SD | Min | Max | ♂ | ♀ |
| 18              | 133.33 | 5.80 | 130.0 | 140.0 | 3 | 1 | 2 |
| 19              | 150.00 | 3.03 | 146.0 | 154.0 | 6 | 2 | 4 |
| 20              | 159.67 | 0.58 | 159.0 | 160.0 | 3 | 2 | 2 |
| 21              | 174.67 | 3.51 | 171.0 | 178.0 | 3 | 2 | 1 |
| 22              | 186.00 | 1.15 | 186.0 | 186.0 | 2 | 0 | 2 |
| 23              | 196.33 | 1.15 | 195.0 | 197.0 | 3 | 1 | 2 |
| 24              | 208.67 | 3.81 | 204.0 | 213.0 | 9 | 5 | 4 |
| 25              | 214.00 | 214.0 | 214.0 | 1 | 0 | 1 |
| 26              | 229.00 | 5.70 | 225.0 | 233.0 | 2 | 1 | 1 |
| 27              | 239.25 | 2.36 | 236.0 | 241.0 | 4 | 4 | 0 |
| 28              | 249.50 | 0.70 | 249.0 | 250.0 | 2 | 0 | 2 |
| 29              | 253.00 | 253.0 | 253.0 | 1 | 0 | 1 |
| 30              | 263.67 | 1.15 | 263.0 | 265.0 | 3 | 3 | 0 |
| Total           |        |     |     |     | 42 | 21 | 21 |
ANOVA and post-hoc RIR Tukey test. The numerical data were statistically analyzed. Distribution of variables was checked using the Shapiro–Wilk (W) test, while homogeneity of variance was checked using Fisher’s test. Since the fetuses studied were inadequately dispersed with fetal age, we tested sex (Student t test for unpaired variables) and laterality (Student t test for paired variables) differences for the whole sample, without considering fetal ages. To compare whether means change with age, Student’s t test for independent variables, and one-way analysis of variance with post-hoc Tukey’s test were used. If no similarity of variance occurred, the non-parametric Kruskal–Wallis test was used. The characterization of growth dynamics for the analyzed parameters was based on linear and curvilinear regression analysis. The match between the estimated curves and fetal age in weeks was evaluated based on the coefficient of determination ($R^2$).

**Results**

No statistically significant differences ($p > 0.05$) in assessing intra-observer reproducibility of clavicle measures were found. The morphometric values obtained were characterized by normality of distribution and homogeneity of variance. As a consequence, numerical variables have been expressed as mean ± standard deviation. The lengths and three widths of the fetal clavicles have separately been presented for males (Table 2) and females (Table 3), and their projection surface areas and volumes have separately been displayed for males (Table 4) and females (Table 5). The statistical analysis for the whole sample revealed neither sex nor bilateral differences ($p > 0.05$) regarding all analyzed parameters. Therefore, we aggregated investigated the growth dynamics of the established parameters, as functions of fetal age in weeks, without considering sex or side. Thus, all individual data—not only their means—irrespective of sex and side were involved in the six regression formulae. An increase in length and width
of the fetal clavicle at varying ages in weeks followed natural logarithmic functions. Between weeks 18 and 30, the mean length of the clavicle increased from $16.97 \pm 0.49$ to $28.70 \pm 1.54$ mm on the right and from $18.94 \pm 0.59$ to $29.39 \pm 0.21$ mm on the left, following the function $y = -54.439 + 24.673 \times \ln(\text{age}) \pm 0.237$ ($R^2 = 0.86$)\textup{(Fig. 4a).} At the same period, the mean width of the clavicle acromial end increased from $2.34 \pm 0.12$ to $4.20 \pm 0.54$ mm on the right, and from $2.46 \pm 0.09$ to $4.48 \pm 0.43$ mm on the left, following the function: $y = -12.042 + 4.906 \times \ln(\text{age}) \pm 0$. 

\begin{table}
\centering
\caption{Length and widths of the fetal clavicle in males} 
\begin{tabular}{llllllllll}
\hline
Gestational & \multicolumn{2}{c}{Mean} & \multicolumn{2}{c}{SD} & \multicolumn{2}{c}{Mean} & \multicolumn{2}{c}{SD} & \multicolumn{2}{c}{Mean} & \multicolumn{2}{c}{SD} & \multicolumn{2}{c}{Mean} & \multicolumn{2}{c}{SD} \\
\hline
age (weeks) & & & & & & & & & & & & & & & & \\
\hline
18 & 1 & 16.90 & – & 18.59 & – & 2.23 & – & 2.44 & – & 1.69 & – & 1.73 & – & 2.24 & – & 2.27 & – \\
19 & 2 & 17.03 & 0.37 & 17.54 & 0.67 & 2.07 & 0.15 & 2.38 & 0.10 & 1.78 & 0.02 & 1.76 & 0.16 & 1.91 & 0.00 & 2.30 & 0.02 \\
20 & 2 & 18.86 & 0.41 & 18.51 & 0.82 & 2.47 & 0.95 & 2.69 & 0.42 & 1.92 & 0.33 & 1.78 & 0.21 & 2.00 & 0.06 & 2.42 & 0.02 \\
21 & 2 & 22.08 & 0.52 & 22.40 & 0.21 & 2.83 & 0.08 & 2.94 & 0.19 & 1.99 & 0.21 & 1.84 & 0.01 & 2.81 & 0.04 & 2.66 & 0.25 \\
22 & 0 & – & – & – & – & – & – & – & – & – & – & – & – & – & – & – & – & – \\
23 & 1 & 23.41 & – & 21.36 & – & 3.34 & – & 3.01 & – & 2.07 & – & 2.14 & – & 3.13 & – & 2.54 & – \\
24 & 5 & 23.87 & 1.00 & 22.64 & 1.18 & 3.53 & 0.53 & 3.55 & 0.48 & 2.25 & 0.20 & 2.15 & 0.15 & 2.77 & 0.12 & 2.86 & 0.20 \\
25 & 0 & – & – & – & – & – & – & – & – & – & – & – & – & – & – & – & – & – \\
26 & 1 & 25.61 & – & 27.21 & – & 4.12 & – & 4.07 & – & 2.72 & – & 2.03 & – & 3.35 & – & 2.99 & – \\
27 & 4 & 26.39 & 2.90 & 27.31 & 2.24 & 4.41 & 0.11 & 4.41 & 0.17 & 2.64 & 0.16 & 2.26 & 0.11 & 3.44 & 0.13 & 3.08 & 0.12 \\
28 & 0 & – & – & – & – & – & – & – & – & – & – & – & – & – & – & – & – & – \\
29 & 0 & – & – & – & – & – & – & – & – & – & – & – & – & – & – & – & – & – \\
30 & 3 & 28.70 & 1.54 & 29.39 & 1.04 & 4.20 & 0.54 & 4.48 & 0.43 & 2.69 & 0.17 & 2.66 & 0.17 & 3.23 & 0.29 & 3.09 & 0.17 \\
\hline
\end{tabular}
\end{table}

Since the fetuses did not represent adequate samples for statistical analysis between particular 1-week intervals, we tested sex and laterality differences for the whole sample, without considering fetal ages.
The mean width of the clavicle central part increased from $1.77 \pm 0.07$ to $2.69 \pm 0.17$ mm on the right and from $1.75 \pm 0.11$ to $2.66 \pm 0.17$ mm on the left, following the function $y = -4.210 + 2.028 \times \ln(\text{age}) + 0.177$ ($R^2 = 0.77$)—(Fig. 4c). The mean width of the clavicle sternal end increased from $2.23 \pm 0.04$ to $3.23 \pm 0.29$ mm on the right and from $2.32 \pm 0.06$ to $3.09 \pm 0.17$ mm on the left, following the function $y = -4.687 + 2.364 \times \ln(\text{age}) + 0.242$ ($R^2 = 0.70$)—(Fig. 4d).

Since the fetuses did not represent adequate samples for statistical analysis between particular 1-week intervals, we tested sex and laterality differences for the whole sample, without considering fetal ages.
68.47 ± 6.21 mm$^2$ on the left, following the function $y = -51.078 + 4.174 \times \ln(\text{age}) ± 6.943$ ($R^2 = 0.82$)—(Fig. 4e), with fetal ages expressed in weeks.

On the right and left sides, the mean volume of the clavicle increased from 70.24 ± 8.00 to 183.87 ± 34.96 mm$^3$ and from 76.06 ± 2.81 to 191.71 ± 17.40 mm$^3$, respectively, following the function $y = -766.948 + 281.774 \times \ln(\text{age}) ± 19.610$ ($R^2 = 0.84$)—(Fig. 4f), where age was expressed in weeks.

### Discussion

As reported in the professional literature, to date, assessment of the development of the clavicle has involved different research methods: anatomical dissection, anthropometric measurements, X-rays, ultrasound, and 3D-ultrasound, but a CT examination has not been used yet. Therefore, we decided to perform a precise quantitative assessment of the clavicle in human fetuses based on objective research techniques, i.e., computed tomography and digital image analysis. Of note, our findings have been presented as if describing a developmental sequence in one fetus, even though the numerical data have truly been cross-sectional, resulting from 42 autopsied fetuses.

The study revealed neither sex nor bilateral differences regarding the examined morphometric parameters of the clavicle. The lack of differences between the right and left fetal clavicles was also confirmed by Mohsin et al. [3]. On the contrary, both sex and bilateral differences were found in adults, since numerous authors [10–13] reported clavicle length to be greater in men than in women. Albeit preponderance of researchers [4, 8, 10, 11, 13–16] reported the left clavicle to be consequently longer than the right one, only Mays et al. [16] and Auerbach and Raxter [14] proved statistically significant right–left differences. We share the view of Mohsin et al. [3] that in the fetus, the right and left clavicles develop symmetrically. The subsequent bilateral differences observed in adults are related with the development of the dominant upper limb, type of work, and everyday life conditions. However, the literature lacks quantitative data on clavicle length in fetuses of different ethnic origin [2, 3, 9, 17].

In this study, based on autopsied human fetuses aged 18–30 weeks, the mean lengths of the right and left clavicles increased from 16.97 to 28.70 mm, and from 18.94 to 29.39 mm, respectively. According to Mohsin et al. [3], in their autopsy material consisting of 15 human fetuses aged 14–33 weeks, the mean lengths of the right and left clavicles increased from 14.12 to 34.58, and from 14.43 to 34.71 mm, correspondingly. Black and Scheuer [9] evaluated clavicle length in in utero fetuses using ultrasound, and on autopsy specimens using anthropometric techniques. The mean clavicle length in the former group increased from 17 mm at 16 weeks to 41 mm at 40 weeks, while in the latter group increased from 8.2 mm at 12 weeks to 44.1 mm at 40 weeks. In the material under examination, an increase in clavicle length has followed the function $y = -54.439 + 24.673 \times \ln(\text{age})$ that clearly corresponds with the findings by Sherer et al. [2].

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| Gestational age (weeks) | N | Morphometric parameters of the female clavicle |
|-------------------------|---|---------------------------------------------|
|                         |   | Projection surface area (mm$^2$) | Volume (mm$^3$) |
|                         |   | Right | Mean | SD | Left | Mean | SD | Right | Mean | SD | Left | Mean | SD |
| 18                      | 2  | 32.81 | 1.40 | | 33.91 | 0.72 | | 74.65 | 3.42 | | 77.49 | 1.89 |
| 19                      | 4  | 24.45 | 0.30 | | 25.01 | 1.01 | | 55.39 | 1.39 | | 56.61 | 2.98 |
| 20                      | 1  | 27.10 | – | | 29.90 | – | | 63.14 | – | | 72.06 | – |
| 21                      | 1  | 33.50 | – | | 29.90 | – | | 81.07 | – | | 70.03 | – |
| 22                      | 2  | 45.30 | 0.14 | | 39.80 | 0.14 | | 111.89 | 0.99 | | 94.73 | 0.90 |
| 23                      | 2  | 41.75 | 8.56 | | 41.25 | 7.42 | | 101.21 | 20.45 | | 99.54 | 17.06 |
| 24                      | 4  | 47.65 | 10.05 | | 46.15 | 8.08 | | 125.82 | 27.51 | | 121.17 | 20.55 |
| 25                      | 1  | 40.70 | – | | 40.10 | – | | 102.56 | – | | 98.25 | – |
| 26                      | 1  | 71.20 | – | | 65.40 | – | | 180.14 | – | | 164.81 | – |
| 27                      | 0  | – | – | – | – | – | – | – | – | – |
| 28                      | 2  | 75.70 | 3.54 | | 71.90 | 3.68 | | 199.41 | 6.64 | | 183.40 | 11.41 |
| 29                      | 1  | 65.50 | – | | 54.60 | – | | 168.34 | – | | 151.79 | – |
| 30                      | 0  | – | – | – | – | – | – | – | – | – |

Since the fetuses did not represent adequate samples for statistical analysis between particular 1-week intervals, we tested sex and laterality differences for the whole sample, without considering fetal ages.

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**Table 5** Projection surface area and volume of the fetal clavicle in females.
authors performed an ultrasound cross-sectional study of 623 consecutive patients between 14 and 42 weeks' gestation, and proved an increase in clavicle length in accordance with the function
\[ y = -75.30 + 32.70 \times \ln(\text{age}) \pm (-0.41 + 0.08328 \times \text{age}) \]. Nevertheless, 20 years earlier, Yarkoni et al. [17] had ultrasonically evaluated clavicle length in 85 human fetuses aged 15–40 weeks and reported a commensurate increase in its length to follow the function:
\[ y = 1.118303 + 0.9788639 \times \text{age} \]. As it turned down, the clavicle length expressed in millimeters

Fig. 4  Regression lines for length (a), width of the acromial end (b), width of the central part (c), width of the sternal end (d), projection surface area (e), and volume (f) of the clavicle.
Conclusions

In terms of morphometric parameters, the fetal clavicle shows neither sex nor bilateral differences. We found the width of the sternal end of the right and left clavicles increased from $1.08 \pm 0.27$ to $2.65 \pm 0.28$ mm on the right, and from $1.03 \pm 0.30$ to $2.06 \pm 0.416$ mm, respectively, for the sternal end. As reported by Mohsin et al. [3], the widths of the clavicle increased logarithmically. Interestingly enough, in contrast to our study, Mohsin et al. [3] also focused on the vertical dimensions of the right and left clavicles.

Vertical dimensions of the right and left clavicles were consistently substantially larger than the mean width of the right and left clavicle acromial end increased from $4.47 \pm 0.53$ mm, and from $2.00 \pm 0.38$ to $4.55 \pm 0.64$ mm, respectively. Of note, in this study, the mean width of the clavicle central part increased from $1.77$ to $2.69$ mm on the right, and from $1.75$ to $2.66$ mm on the left.

We found the width of the sternal end of the right and left clavicles increased from $2.34$ to $4.20$ mm on the right, and from $2.00$ to $3.09$ mm, respectively. Of note, in this study, the mean width of the left clavicle central part increased from $1.08 \pm 0.27$ to $2.65 \pm 0.28$ mm on the right, and from $1.03 \pm 0.30$ to $2.06 \pm 0.416$ mm, respectively, for the sternal end. As reported by Mohsin et al. [3], the widths of the clavicle increased logarithmically.

To date, computed tomography has not been used for the assessment of the fetal clavicle development in the human fetus obtained in this study may be of potential relevance. Obviously, the width of the shoulder girdle is greater than the dimensions of the head, impeding or even preventing a natural delivery. Nevertheless, according to Sherer et al. [2], the width of the clavicle length in all fetuses, but the overall presence and configuration of this important anterior component of the shoulder girdle should be noted. If possible, to our opinion, ultrasound morphometric evaluation of clavicle length may help in diagnosing many congenital defects, such as clavicular hypoplasia, often observed in Caffey syndrome [11], Meckel-Gruber syndrome, as well as in other syndromes characterized, among other things, by agenesia or hypoplasia in chondrodysplasia punctata or Caffey syndrome (trisomy 18), and symmetrically, in the shoulder girdle.

The numerical data describing the clavicle growth in the human fetus obtained in this study may be of potential relevance. Obviously, the width of the shoulder girdle is greater than the dimensions of the head, impeding or even preventing a natural delivery. Nevertheless, according to Sherer et al. [2], the width of the clavicle length in all fetuses, but the overall presence and configuration of this important anterior component of the shoulder girdle should be noted. If possible, to our opinion, ultrasound morphometric evaluation of clavicle length may help in diagnosing many congenital defects, such as clavicular hypoplasia, often observed in Caffey syndrome [11], Meckel-Gruber syndrome, as well as in other syndromes characterized, among other things, by agenesia or hypoplasia in chondrodysplasia punctata or Caffey syndrome (trisomy 18), and symmetrically, in the shoulder girdle.
2. The fetal clavicle increases logarithmically with respect to its length, width, and volume, and linearly with respect to its projection surface area.
3. The obtained morphometric data of the growing clavicle are considered normative for their respective weeks of gestation and may be of relevance in the diagnosis of congenital defects.

Compliance with ethical standards

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

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