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TECHNICAL NOTE

ODONTOLOGY

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Estimation of Gestational Age from Tooth Germs: Biometric Study of DentaScan Images

ABSTRACT: The few available studies on fetal age estimation concern very small samples, and statistical analysis is sometimes inadequate. In this survey, we used germs of deciduous teeth to estimate fetal age. Forty-nine fetuses and 40 mandibles were scanned, and observations and measurements were made on DentaScan images. After checking their repeatability and reproducibility (analysis of variance), we defined thresholds using Fisher's linear discriminant analysis to calculate the probability that a fetus was over or below a predefined age threshold. The forensic threshold which is of particular interest in France is 22 weeks amenorrhea. Relationships between fetal age and deciduous germ measurements were then sought by multiple linear regression. The thresholds gave very good results: 91.84% of good probability for the threshold of 22 weeks amenorrhea with no chance of error. The most precise age evaluation obtained nevertheless gave a range of ±4.6 weeks amenorrhea, so greater accuracy is still needed.

KEYWORDS: forensic science, fetus, age determination, deciduous teeth, gestational age, DentaScan

Estimation of fetal age is of considerable interest in medico-legal anthropology, as well as in other fields, such as biologic anthropology, anatomy, fetal pathology, or obstetric gynecology. The aim of our study was to develop a simple, standardized, reliable, and reproducible technique for estimating fetal age. Charts are usually based on direct or indirect measurements of long bones (1) or the mandible (2,3). Dental organs have long been used for age determination in both immature and adult subjects (4–11). Although they are more rarely applied in the fetus, a variety of techniques are employed:

- Anatomic (12), using sequential tables of centers of calcification,
- Histologic (13–15), based on tooth ring analysis,
- Radiologic (16), but this method cannot be used at <36 weeks amenorrhea (WA),
- Ultrasound (17), which is not always applicable as tooth germs can only be detected after 29 WA.

The main drawback of these fetal studies is that fetal age is estimated from references, such as crown-root length (18,19). In addition, these charts have been established in nonrepresentative populations (12–15). There is little information on the origin of the samples. As far as we are aware, this is the first time that computed tomography (CT) has been used for the observation of tooth germs.

Materials and Methods

Materials

Forty-nine fetuses and 40 mandibles underwent CT scan. The fetuses were aged between 19 and 41 WA, and their sex (Fig. 1), weight, and crown-root length were known. They were obtained from therapeutic or spontaneous abortions or maternal salvage had died *in utero* or were stillborns. Normal fetuses were selected from a collection of 700 fetuses according to several radiologic, fetopathologic, and histologic criteria. Their karyotype was normal, without trisomy or external, internal, or bony abnormalities. Fetuses with congenital defects or growth retardation were not included. The study sample was selected at the legal medicine laboratory of the Medical Faculty, Marseille, France.

Methods

Measurement Systems and Apparatus—The observations and measurements were carried out on two-dimensional reconstructed images obtained by DentaScan (GE Healthcare, La Penne S/Huveaune, France). At the present time, this is the most accurate method of maxillary investigation (20). A General Electric HiSpeed CT system (General Electric, Milwaukee, WI) was used. Images were processed with DentaScan V2.0.8a software. This displays numbered sections, and the number of each section corresponds to its position in millimeters in relation to the beginning of a previously drawn line ([21]; Fig. 2).

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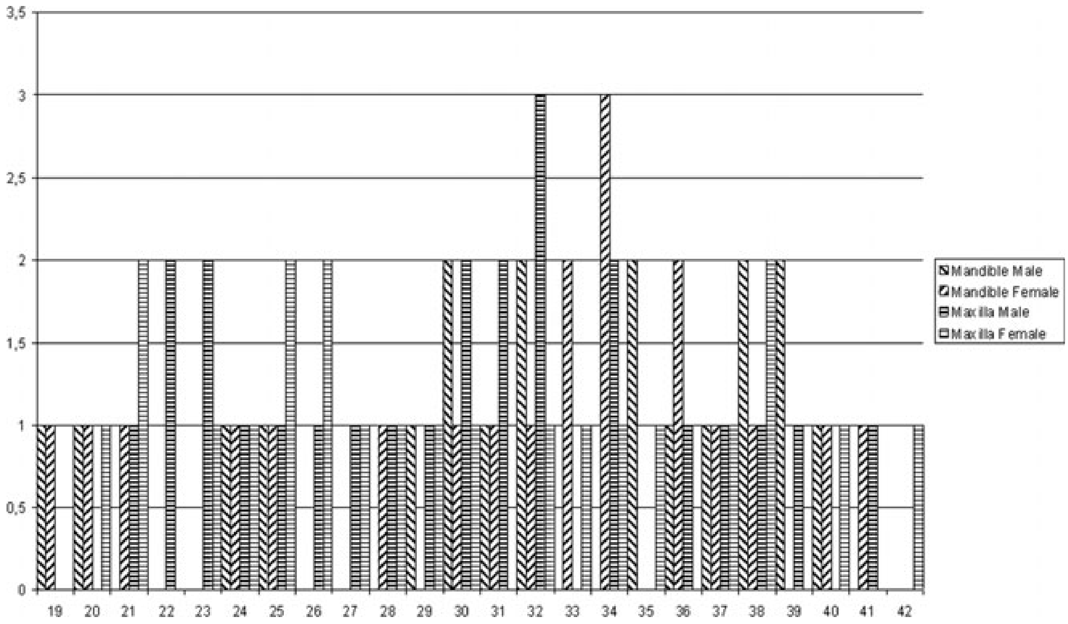

FIG. 1—*Maxillas and mandibles: distribution of fetuses by age and sex.*

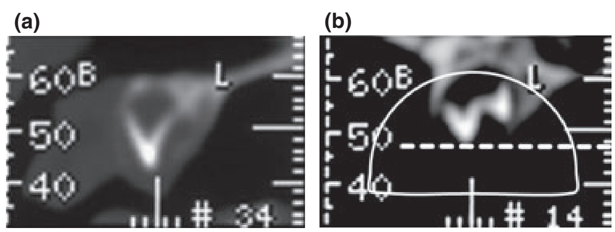

FIG. 2—*DentaScan sections. The curve passes through the center of the tooth germs. (a) Oblique coronal section showing image 34 through image 61. (b) Oblique coronal section showing image 14 through image 54.*

Data Collection

Qualitative Data—Two types of qualitative observations were made:

The *presence or absence* of all tooth germs was recorded, from sectors 5 to 8: *51, 52, 53, 54, 55, 61, 62, 63, 64, 65, 71, 72, 73, 74, 75, 81, 82, 83, 84, 85.* A score of 1 was attributed if the germ was present and 0 if it was absent. The germs were also scored according to their degree of maturation:

Mineralization of the cusp tips only (Fig. 3*a*),

Coalescence of the cusp tips (Fig. 3*b*),

Complete mineralization of the crown (Fig. 3*c*).

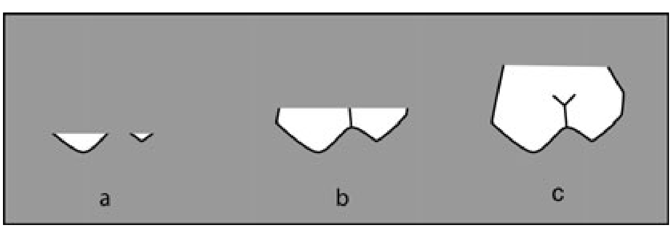

FIG. 3—*Index of maturation of tooth germs. (a) Mineralization of cusp tips only, (b) coalescence of cusp tips, and (c) complete mineralization of the crown.*

Quantitative Data—Measurements were made in pixels between the points of mineralization: cusps, fissures, and necks, with three measurements being made for cuspid teeth: lingual (L), cervical (C), and buccal (V), and six measurements for multicuspid teeth as shown in the diagrams (Fig. 4): occlusal (O), lingual (L), cervical (C), buccal (V), buccal aspect of the occlusal surface (VA), and lingual aspect (LA).

All qualitative data were tested. Measurements were made by two independent observers. Thirty germs were selected at random and two observers each carried out two series of measurements between the points of mineralization of the cusps, fissures, and necks. All cases were tested for error because of intra-observer and inter-observer bias using analysis of variance (ANOVA).

Relation Between Age and Measurements Recorded

Maturation scores were analyzed using Fisher's linear discriminant formula. This method yields the probabilities of belonging to an age group below or above a given threshold by two statistical methods: stepwise analysis and simultaneous data entry. So that the predictive function should not be calculated as a function of the individual tested, we used the N−1 technique.

Age Estimation. Ascending and descending linear regression was applied in N−1 in all fetuses with cuspid teeth (central and lateral incisors), 25 maxillary and 21 mandibular teeth, to measurements which had previously been shown to be repeatable and reproducible.

Results

Intra- and Inter-observer Error

Qualitative Data—

α- Presence or absence of germs

No statistical difference at a significance threshold of 5% was found for the two qualitative observations in either maxillary or mandibular teeth.

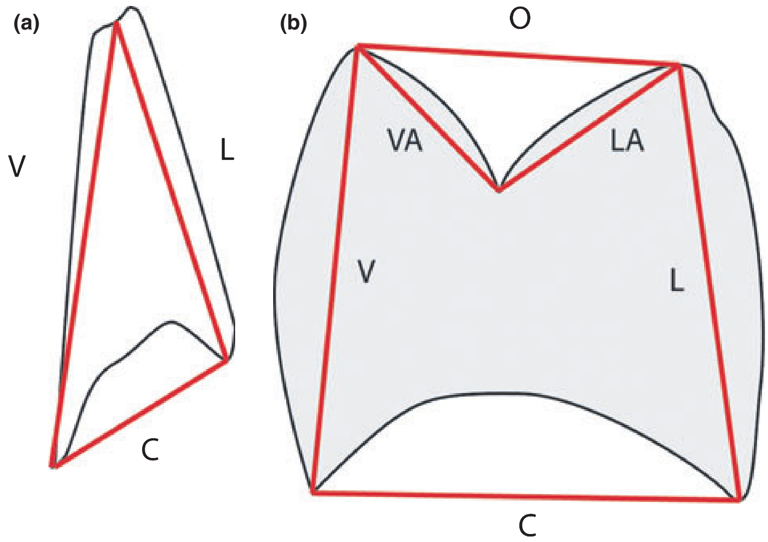

FIG. 4—*Measurements made on cuspid teeth (a): lingual (L), cervical (C), and buccal (V), and on multicuspid teeth (b): occlusal (O), lingual (L), cervical (C), buccal (V), buccal aspect of the occlusal surface (VA), and the lingual aspect (LA).*

β – Index of tooth germ maturation
Using ANOVA, once again no statistical difference was found at a significance threshold of 5%.

Quantitative Data—Only one metric measurement was excluded: buccal measurement of cuspid germs (measurement 1), s = 0.012 in intra-observer evaluation. This was repeatable but not reproducible (s = 0.906).

Correlation with Age

Discriminant Analysis—Fisher's linear discriminant analysis yielded a set of well-classified individuals whose ages could be estimate. Reliability was good at all thresholds from 19 to 40 WA. Only one threshold, 25 WA, presented an error (wrongly classified individual), which was nevertheless below 5%. For the 28 to 30 WA thresholds, the maximum probability of correct classification was 100%.

Linear Regressions—We carried out eight multiple linear regressions: maxillary and mandibular ascending and descending multiple linear regression, for two confidence intervals: 95% and 99%. These regressions were only carried out on the incisors because these were the most numerous germs: 25 maxillary and 21 mandibular (Table 1). So that the predictive function should not be calculated as a function of the individual tested, we used the N−1 technique. Figure 5 shows residual dispersion between −10 and +10 WA with a mean ± 4.6 WA. These observations were made in individuals aged 27–40 WA.

TABLE 1—*Maxillary and mandibular ascending and descending multiple linear regression for two confidence intervals: 95% and 99%. Maxillary multiple linear regression is more accurate and with no chance of error.*

	95% (WA)—	Error %	99% (WA)—	Error %
Maxillary ascending (25)	10.60	3.41	14.40	0
Maxillary descending	9.23	0	12.59	0
Mandibular ascending (21)	11.29	3.45	15.47	0
Mandibular descending	10.90	0	15.02	0

$$Y = 1.61 \times 52\,L - 0.773 \times 52\,C + 0.588 \times 51\,C + 20.315 \pm 4.61 \text{ with } 95\%\text{ CI}$$

Discussion

We propose an original and innovative method of estimating fetal age by study of tooth germs. Its reproducibility and repeatability were validated by statistical tests.

CT imaging yields very precise observations and measurements. The germs can be visualized in the very early stages of crown mineralization. This technique has the advantage of being independent of fetal position, unlike the ultrasonic or radiographic techniques previously mentioned.

Probabilistic approach: using Fisher's linear discriminant analysis to determine thresholds of fetal age, we obtained in our sample a very good percentage of correctly classified fetuses at almost all thresholds, with a single error which did not exceed 5% at the 25 WA threshold. According to the law of January 8, 1993, the medico-legal threshold which concerns us in France is 22 WA (and/or fetal weight greater than 500 g). This is the fetal viability threshold and as soon as it is exceeded official registration is mandatory. If fetal age is below this threshold, the fetus is declared dead and nonviable. At this threshold, maturation scores showed a

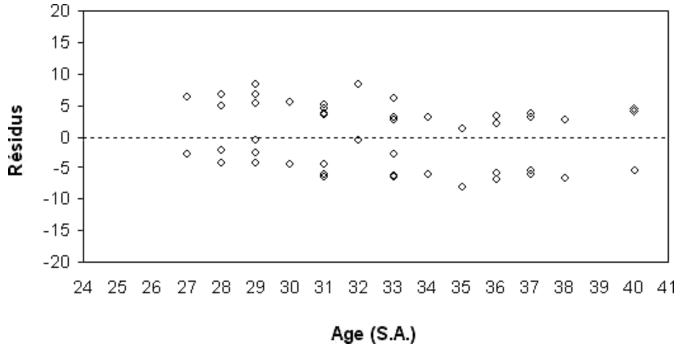

FIG. 5—*Distribution of residues of maxillary multiple linear regression according to fetal age (27–40 WA). 95% confidence interval with no chance of error.*

mean efficacy of 91.84% of correctly classified and 8.16% of undetermined individuals with 100% reliability. For international interest, we can use this probabilistic approach using a range of different WA values; for example in Portugal and Finland 22 weeks, the United Kingdom 24 weeks, in Australia and the United States 20 weeks (federal guidelines) (22).

Estimation of fetal age: using ascending and descending maxillary and mandibular linear regression (95% and 99% confidence intervals), the most precise estimate obtained was ±4.6 WA and the least precise was ±7.7 WA. This result is less accurate than those obtained by measurement of long bones (1,23), which was precise within a range of ±2 to 3 WA.

Our results are therefore less satisfactory, but as we are working on dental material our method enables an estimation of age when only the skull is available, in an archeological context for example. Age estimation is possible even if the degree of accuracy is less.

Conclusion

New medical imaging techniques, CT in particular, allow precise, simple, and rapid investigation while preserving the structures in their anatomic relationship. Fetuses therefore remain intact for any future study using other lines of research. Computerization makes it simple and repeatable at will, with no damage to the original material, and little training is required. The images are also easily and permanently preserved using inexpensive and readily obtainable material.

A requisite in forensic medicine is reliability of results. When an autopsy is demanded by the judicial authorities, we need to be able to make an accurate and reliable statement at various legal thresholds, which will determine the subsequent legal consequences. Where fetal age estimation is concerned, Fisher's linear discriminant function gives us this possibility for the 22 WA legal threshold. We observed great reliability and a mean efficiency of nearly 92% for this threshold. Multiple linear regression does not give good results compared with methods of estimation using other indicators, such as the long bones (1,22). Nevertheless, we obtain a dental and not bony regression formula which enables us to estimate age even if only deciduous dental organs are present. This study could be further developed using a larger sample, which would increase the number of individuals in each age group. It would also be very interesting to consider in this sample the maxillary and mandibular teeth of the same individual, to increase the predictive power of our analysis. With a larger sample, it would be possible to determine more closely the variability of fetal odontogenesis in each age group and to develop more robust and accurate models for age estimation.

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