DOI: 10.7860/IJARS/2023/60683.2916

Original Article

Anatomy Section

Morphometry and Histology of Thyroid Gland in Developing Human Foetuses: A Cross-sectional Study

ROLI JOSHI[1], PANKAJ SINGH[2], SHALINI SRIVASTAVA[3]

ABSTRACT

Introduction: The thyroid gland regulates the basal metabolic rate, somatic growth, and physical growth. The thyroid gland plays a major role in the normal growth of a foetus during the prenatal period. There are few literatures on foetal thyroid gland.

Aim: To study the various morphometric and histological changes during intrauterine life in both gender of human foetuses.

Materials and Methods: This cross-sectional study was carried out in the Department of Obstetrics and Gynaecology, Dr. Sushila Tiwari Hospital Government Medical College, Haldwani (UK) and Saraswati Medical College, Unnao in the period of August 2015-2022. Government Medical College, Haldwani for duration of three years on medically terminated 70 foetus of both sexes of known gestational age ranging from 10 to 40 weeks. These foetuses were divided among three groups as group 1 (10-20 weeks), group 2 (21-30 weeks), group 3 (31-40 week). Midline dissection of neck was done to expose the thyroid gland. Morphometric parameters like length, width, and thickness of each lobe and the isthmus were measured by digital Vernier caliper, weight was also taken by digital weighing machine, then tissue processing was done and paraffin blocks were prepared. All sections were stained with Haematoxylin and Eosin stain (H&E) and studied under the microscope aided camera. By using Analysis of Variance (ANOVA) test and Independent sample test (t-test) comparison among different age groups and gender has been done.

Results: There was a significant increase in foetal thyroid weight, length and widths of the right and left lobes, and length of the isthmus with increase in gestational age, while no significant relation was found between male and female foetuses of measured morphometric parameter. The histological features revealed that development of thyroid gland starts earlier and thin capsule was seen at 12 week and it became thicker and more vascular till 35 weeks. Follicles started to form at 14 week and matured follicle were seen at the age of 24 weeks, at 25th week, the lining epithelium of follicles was low columnar to cuboidal, colloid material was seen almost in every follicle. C cells were found at 22 weeks. At the 36 weeks, the follicles were seen spherical and reached to maturity level.

Conclusion: As the gestational age increases, the morphometric parameters must increase, if not so, it means the growth of gland is hampered. The knowledge of the growth and differentiation of foetal thyroid will be helpful for clinicians, to judge the thyroid structure prenatally.

Keywords: Colloid, Histological differentiation, Thyroid disorder, Thyroid follicle

INTRODUCTION

The thyroid is one of the largest and earliest differentiated endocrine glands in humans, and it is important for brain growth and development during pregnancy and the first few years after birth [1]. The foetal thyroid function is dependent on the embryogenesis, differentiation, and maturation of the thyroid gland. In foetus, Thyroid hormones thyroxine (T4) and Triiodothyronine (T3) are detectable early in gestation and have important developmental, metabolic, and maturational effects and circadian rhythm [2]. Deficiency of thyroid hormones during intrauterine life impairs growth and compromises its adaptation to extrauterine life. Thyroid gland is the first endocrine gland to develop at the 4th week of gestation, by the 8th week thyroid gland has almost acquired its final position and shape with two lateral lobes connected inferiorly through the isthmus. The endodermal tissue derived from the foramen caecum will give rise to the numerous follicles of the thyroid gland. Colloid begins to appear in the 11th week in follicles; thereafter, iodine concentration and the synthesis of thyroid hormones and by 18 weeks, the levels of foetal thyroid-stimulating hormone and thyroxine begin to increase, reaching adult levels by 35 weeks [3].

The study of histological differentiation might help us to understand the morphological changes during the development of the human thyroid. Functional and anatomical abnormalities of the thyroid are common among the diseases of endocrine glands, affecting approximately one in 2000-4000 newborns [2]. Limited work has been done on the morphometry and histology of this peculiar gland [3]. The present study was undertaken to analyse the thyroid gland in male and female foetuses of various gestational ages, to observe the sexual dimorphism and progressive morphometric and histomorphic changes.

MATERIALS AND METHODS

The cross-sectional study was conducted on 70 human foetuses (37 males and 33 females) of different gestational age, in the Department of Anatomy, Government Medical College, Haldwani Uttarakhand, India. The foetuses were obtained from the Department of Obstetrics and Gynaecology, Dr. Sushila Tiwari Hospital Government Medical College, Haldwani (UK) and Saraswati Medical College, Unnao in the period of 2015-2022. After Ethical Committee approval (through letter no. 134/EC/R-12-12-2013) and permission from the concerned authorities of the Institute, the human foetuses aged between 10th to 40th gestational weeks were collected. Written consent from foetal parents were taken after informing that burial with honors will be done after foetal dissection for research purposes.

Inclusion criteria: Only spontaneous aborted and stillborn foetuses of gestational ages between 10 to 40 weeks were included.

Exclusion criteria: Cases with twin or congenital anomalies were excluded.

Study Procedure

These foetuses were preserved in 10% formalin. The peritoneal and orbital cavities were also injected with 10% formalin, and after 7 days dissection was done so that the original data can be availed, as shrinkage of tissue was minimal or none till this period. The gestational age of foetuses was calculated from the crown-rump length and ultrasonography. The foetuses were divided into three groups according to their gestational age as group 1 (10-20 weeks), group 2 (21-30 weeks), group 3 (> 31weeks). The midline dissection of the anterior aspect of neck was done to expose the thyroid gland. The infrahyoid muscles along with surrounding connective tissue were separated and the bilobed thyroid gland was removed. Measurements of the following parameters of in-situ gland were taken through digital Vernier caliper [Table/Fig-1].

[Table/Fig-1]: Photograph showing measurement of lobe of right and left thyroid gland: a) length; b) width showing with black arrow.

1. **Length of right and left lobes-** between the upper and lower poles
2. **Length of isthmus-** between superior and inferior border
3. **Width of right and left lobes-** between lateral margin to a perpendicular line drawn at the meeting point of isthmus and lobes
4. **Width of isthmus-** between perpendicular lines drawn at the meeting point of isthmus and lobes
5. **Thickness of right, left lobes and isthmus-** at maximum anteroposterior dimension.

Once the gland was removed from its anatomical position and extra tissues were removed carefully, the outer surface of foetal thyroids was dried with blotting paper and then weighed by a digital weighing machine.

The thyroid tissue was subjected to routine processing by dehydration in graded alcohols, clearing in xylol, and embedded in paraffin. Sections of 5 μ thickness were cut with a rotatory microtome and stained with H&E stain. The histological features were observed by using a compound light microscope and photography was done with a digital camera. The histological study was done for recording the developmental changes in the gland microanatomically.

STATISTICAL ANALYSIS

Statistical analysis was carried out using Statistical Package for Social Sciences (Chicago, IL, version 22.0 for windows). All quantitative variables were estimated using measures of central location (mean, median) and measures of dispersion (standard error and standard deviation). Descriptive statistics is used to define all parameters. Morphometric parameters by one-way ANOVA test within and between the groups. Independent sample test (t-test) was used to note sexual dimorphism.

RESULTS

Number of foetuses in each group were as group 1 (n=22), group 2 (n=23), group 3 (n=25) and there were 37 males and 33 females in this study.

Length, weight, width and thickness of right and left lobes of thyroid and isthmus were statistically significant among all three groups of different gestational age [Table/Fig-2].

There were no statistically significant differences in length, breadth, thickness of both the lobes of thyroid, weight of thyroid, and length, breadth and thickness of isthmus between male and female fetuses [Table/Fig-3].

Group 1 (10-20 weeks): The capsule with trabeculae of the gland was thin and become thicker later, and septa was seen into the parenchyma of the gland with large number of blood vessels, lined with endothelium indicating high vascularisation of the gland. Follicular organisation is incomplete [Table/Fig-4]. Maximum number of follicles has a circular and oval shape these are devoid of colloid, which indicates the active state of the thyroid gland. These are also lined by the cuboidal epithelium [Table/Fig-5]. Some follicles have clear small vacuoles in the colloid material. Peripheral follicles are larger as compared to the central ones with more vascularity. Central gland has many clusters of epithelial cells.

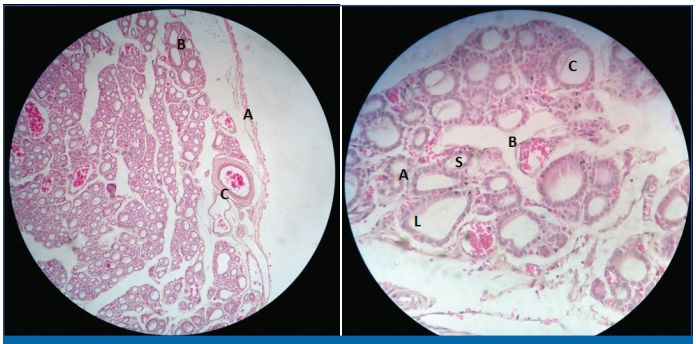
Group 2 (21-30 weeks): At 22 weeks, connective tissue in capsule was thick with large number of blood vessels. In 26 weeks, vascularity was increased and septas [Table/Fig-6] dividing the gland into the different lobules (trabeculae) to the parenchyma of the gland was noticed. Blood vessels were seen invading the gland, but typical lobular pattern was not seen due to the incomplete structure of septa. Peripheral follicles were larger as compared to central follicles and matured in 28th week. In larger follicles lining epithelium was low columnar, and cuboidal in smaller follicles with prominent apical nuclei. Mature thyroid follicles can be seen. Colloid material was seen in almost all follicles but prominent in the peripheral follicles with intrafollicular vacuoles in few. C-cells were also noticed at this stage of development.

Parameters	Group 1 (n=22)		Group 2 (n=23)		Group 3 (n= 25)		p-value
	Range	Mean±SD	Range	Mean±SD	Range	Mean±SD	
Weight (gms)	0.04-0.177	0.111±0.52	0.147-0.484	0.266±0.99	0.347-0.648	0.487±0.99	0.001
Right lobe length (cm)	0.3-1.0	0.631±0.209	0.7-1.8	1.0142±0.320	1.0-2	1.470±0.352	0.001
Left lobe length (cm)	0.2	0.631±0.221	0.6-1.8	1.100±0.393	0.8-1.9	1.374±0.351	0.001
Right lobe breadth (cm)	0.2-0.4	0.308±0.095	0.2-0.8	0.533±0.161	0.4-0.9	0.674±0.145	0.001
Left lobe breadth (cm)	0.2-0.4	0.308±0.760	0.3-0.7	0.492±0.124	0.5-0.9	0.652±0.120	0.001
Thickness of right lobe (cm)	0.2-0.4	0.246±0.066	0.2-0.6	0.367±0.130	0.3-0.8	0.443±0.112	0.001
Thickness of left lobe (cm)	0.1-0.4	0.246±0.077	0.2-0.6	0.375±0.128	0.3-0.7	0.426±0.105	0.001
Length of isthmus (cm)	0.2-0.5	0.300±0.1291	0.2-0.6	0.358±0.1165	0.4-0.613	0.613±0.1517	0.001
Breadth of isthmus (cm)	0.1-0.4	0.277±0.093	0.2-0.342	0.342±0.124	0.3-0.9	0.167±0.065	0.001
Thickness of isthmus (cm)	0.1-0.3	0.162±0.065	0.1-0.3	0.217±0.174	0.1-0.4	0.326±0.185	0.001

[Table/Fig-2]: Morphometery of thyroid gland in all groups.
ANOVA was used

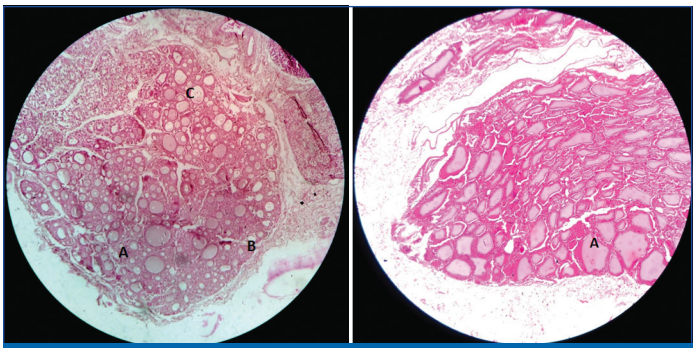
Parameters	Male (n=37)		Female (n=33)		
	Mean	SD	Mean	SD	p-value
Weight (gm)	0.364	0.197	0.293	0.165	0.185
Rt. lobe length (cm)	1.236	0.514	1.078	0.405	0.246
Lt. lobe length (cm)	1.184	0.492	1.017	0.395	0.205
Rt. lobe width (cm)	0.544	0.222	0.535	0.189	0.878
Lt. lobe width (cm)	0.536	0.193	0.500	0.171	0.499
Thickness of rt. lobe (cm)	0.364	0.115	0.378	0.154	0.716
Thickness of lt. lobe (cm)	0.356	0.119	0.374	0.142	0.638
Length of isthmus (cm)	0.464	0.214	0.465	0.185	0.983
Width of isthmus (cm)	0.448	0.204	0.374	0.145	0.158
Thickness of isthmus (cm)	0.192	0.064	0.187	0.694	0.795

[Table/Fig-3]: Morphometery of thyroid gland in all male and female group. Independent t-test



[Table/Fig-4]: Photomicrograph of 13 week foetus at X100 with H&E staining showing connective tissue capsule (A) and trabeculae giving it lobular pattern but of incomplete type (B) and blood vessel in parenchyma of the gland (C). **[Table/Fig-5]:** Photomicrograph of 17-week foetus at X400 with H&E staining showing cuboidal epithelium (A), blood vessel lined by endothelium (B) of the capsule of the gland. showing larger peripheral follicles (L) and smaller central follicles (S). Clear vacuoles and scalloped margin of the colloid. Thickness of the capsule has increased. clear vacuole in colloid material of follicles and having clear vacuole (C) in some follicles, lobular pattern of parenchyma. (Images from left to right)

Group 3 (31-40 weeks): At 35 weeks follicular organisation and cellular differentiation are complete with highly vascularised parenchyma. Follicles are spherical in shape, identical to the adult thyroid follicles [Table/Fig-7]. Follicles present in centre were smaller in size as compare to peripheral follicles. Almost all follicles are filled with colloid material, indicating the complete maturation of the thyroid gland.



[Table/Fig-6]: Photomicrograph of 22 weeks foetus at X100 with H& E staining showing capsule of the gland. follicle (A) filled with colloid, trabeculae (B), clear vacuole in follicle (C). **[Table/Fig-7]:** Photomicrograph of 35 weeks foetus at X100 with H&E staining showing follicles filled with colloid (A). (Images from left to right)

DISCUSSION

Numerous previous studies dealing with the growth and development of thyroid gland in human foetuses with respect to the gestational weeks have been limited to short periods of gestation [2,3,4]. In the current study, the morphometric parameters of the thyroid gland increased in proportion to estimated gestational age, with the mean weight of the gland being more in male foetuses. The lengths of both lobes and isthmus increased considerably with gestational age, while this was consistent in study of Cicekcibasi AE et al., who did his study on 60 aborted foetuses [5]. The results of the present study showed that all the parameters as length, thickness, width weight of both lobes and isthmus was increasing with gestational age and significantly related with it, however, Jalaja Y and Anbarasi CP told that all the parameters of both lobes are significantly related while parameters of isthmus had non significant relation with gestational age, clear reason is not defined for it as it is the matter of further studies on isthmus parameters [4]. In the present study, all the mean values are slightly higher than those of Cicekcibasi AE et al., [5]. Non significant relation of thyroid lobes parameters between males and females foetuses are countered by Jalaja Y and Anbarasi CP which are similar to current study [4]. Gregory JK et al., did not notice any significant differences in right and left lobe dimensions in his ultrasound investigation of 100 male and female neonatal thyroids [6].

Very limited studies are there on this important topic and minor differences observed in them compared to previous studies can be attributed to anthropometric differences between races, maternal dietary habits, antenatal care, and population socio-economic status [6].

This study helps us to associate the morphological changes with histological changes during the development of foetal thyroid gland. According to Mukhiya R et al., the staging of histological differentiation helps us to understand the morphological and functional disorders of the thyroid glands which was also stated by Mukhia R and Lokanadham S and Devi SV [7,8]. According to Ham AW and Carmack DH and Kotian SR et al., at the 12th gestational week, the capsule was very thin, with less number of blood vessels but as the gestational age advanced, the capsule became thicker and the thyroid gland becomes more vascular, similar to the findings of the present study [9,10]. In the present study, the thyroid follicles differentiation started from the periphery of the gland and extended centrally, as the periphery of the gland was more vascular than the centre throughout the gestational age of foetus. In the present study, thin rim of colloid appeared in the developing thyroid follicles of 15-17 gestational weeks' foetuses and increased as the gestational age increases, also observed in the study of Anupriya A and Kalpna R [11]. In present study, from 28th week onward, the colloid appeared irregularly and eroded at periphery along the apices of follicular cells of the thyroid gland in accordance with the finding with Junqueira LC and Carneivo J [12]. In the foetuses of 37th to 40th gestational week the thyroid follicles showed full maturation and were somehow similar to that of an adult, also stated by Mukhiya R et al., and matured thyroid follicle filled colloid and increased vascularity was seen in group 3 cases as noticed by Vahini P [7,13]. Tanriover O et al., found that the number and size of follicles increased as gestational age increased, our study also shows the follicular size is more at the periphery as compare to centre which was also measured by Bande AN et al., at periphery of the gland, the diameter of the follicles was 110-250 µm and that of central follicle was 60-120 µm [14,15].

This study helps us to determine the histological changes during the development of foetal thyroid gland. The present study would help the clinicians to understand the development of thyroid gland and correlate with histopathological changes in certain thyroid gland disorders.

Limitation(s)

The limitation of this study was the constricted range of gestational age of foetuses from 10th to 40th weeks. Hence, it cannot be commented how the early developmental changes must have appeared in the foetuses before 10 weeks.

CONCLUSION(S)

The knowledge of morphometric changes and developmental anomalies of the thyroid gland with increasing foetal age is of excessive

importance to radiologists, gynecologists, and pediatricians to avert both maternal and foetal difficulties in foetal and neonatal period. The study gives the knowledge on thyroid growth and differentiation in different gestational ages.

Acknowledgment

The authors sincerely thank those who donated their bodies for anatomical research. Results from such research can potentially increase humankind's overall knowledge that can then improve patient care. We would like to thank our Department of Anatomy, Obstetrics and Gynecology for their support.

REFERENCES

[1] Moore KL. The Developing Human's Clinically Oriented Embryology. 7th ed. USA: Saunders; 2016. Pp. 215-17.
[2] Kratzsch J, Pulzer F. Thyroid gland development and defects. Best Pract Res Clin Endocrinol Metab. 2008;22(1):57-75.
[3] Patel J, Landers K, Li H, Mortimer RH, Richard K. Thyroid hormones and foetal neurological development. Journal of Endocrinology. 2011;209(1):01-08.
[4] Jalaja Y, Anbarasi CP. Morphology and histogenesis of human fetal thyroid gland. Natl J Clin Anat. 2022;11(3):148-153. DOI: 10.4103/NJCA.NJCA_93_22
[5] Cicekcibasi AE, Salbacak A, Seker M, Ziylan T, Tuncer I, Buyukmumcu M. Developmental variations and clinical importance of the fetal thyroid gland. A morphometric study. Saudi Med J. 2007;28(4):524-28.
[6] Gregory JK, Guse DM. Unique variant of levator glandulae thyroideae muscle. Clin Anat. 2007;20(8):966-67.
[7] Mukhia R, Mansur D, Pandey M, Taneja BK. Histological studies on the foetal thyroid gland of developing human foetuses. JCMS Nepal. 2021;17(1):75-80.
[8] Lokanadham S, Devi SV. Gestational age related developmental anatomy and histogenesis of human fetal thyroid gland. World J Med Sci. 2011;6(4):173-77.
[9] Ham AW, Carmack DH. Histology, 8th edition. J B Lippincott Company. Philadelphia and Toronto. 1979;801-02.
[10] Kotian SR, Salva MN, Pandey AK, Kalthur SG. Human fetal thyroid gland: a morphological and histological study. J Morphol Sci. 2021;38:186-89.
[11] Anupriya A, Kalpna R. Morphological and histological features of human foetal thyroid gland. Int J Sci Stud. 2016;3(10):136-40.
[12] Junqueira LC, Carneivo J. Basic Histology, Text and Atlas, 11th edition. McGraw Hill Company. 2005;411-15.
[13] Vahini P. Morphology and histogenesis of human fetal and adult thyroid gland. J Res Med Dent Sci. 2021;9(9):127-28.
[14] Tanriover O, Comunoglu N, Eren B, Comunoglu C, Turkmen N, Bilgen S, et al. Morphometric features of the thyroid gland: A cadaveric study of Turkish people. Folia Morphol (Warsz). 2011;70(2):103-08.
[15] Bande AN, Doshi MA, Vaidya RB, Dhobale MR, Mudiraj NR. Histogenesis of thyroid gland in human fetuses of different gestational age. JCDR. 2018;12(10):05-08.

PARTICULARS OF CONTRIBUTORS:
1. Associate Professor, Department of Anatomy, NC Medical College, Israna Panipat, Haryana, India.
2. Assistant Professor, Department of Anatomy, Saraswati Medical College, Unnao, Uttar Pradesh, India.
3. Professor, Department of Pathology, Saraswati Medical College, Unnao, Uttar Pradesh, India.

NAME, ADDRESS, E-MAIL ID OF THE CORRESPONDING AUTHOR:
Dr Roli Joshi,
1718, Valencia Mahagun Mezzaria, Noida, Uttar Pradesh, India.
E-mail: roli_joshi2005@yahoo.com

PLAGIARISM CHECKING METHODS: [Jain H et al.]
- Plagiarism X-checker: Oct 12, 2022
- Manual Googling: Mar 28, 2023
- iThenticate Software: Mar 31, 2023 (11%)

ETYMOLOGY: Author Origin

EMENDATIONS: 6

AUTHOR DECLARATION:
- Financial or Other Competing Interests: None
- Was Ethics Committee Approval obtained for this study? Yes
- Was informed consent obtained from the subjects involved in the study? Yes
- For any images presented appropriate consent has been obtained from the subjects. Yes

Date of Submission: **Oct 11, 2022**
Date of Peer Review: **Dec 01, 2022**
Date of Acceptance: **Apr 03, 2023**
Date of Publishing: **Sep 01, 2023**