

Histogenesis of thyroid gland in buffalo

RAJESH RANJAN¹, ANIL SHARMA², OPINDER SINGH³ and NEELAM BANSAL⁴

Guru Angad Dev Veterinary and Animal Sciences University, Ludhiana, Punjab 141 004, India

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The thyroid gland is the first endocrine gland of the body to develop as a ventral-midline-endodermal diverticulum from the floor of the fore-gut at a level between the first and second pharyngeal arches. Rapid advances have been made in knowledge and understanding of thyroid in human (Fagman and Nilsson 2010) due to its functional importance. However, scanty information is available on development of thyroid in domestic animals especially buffalo, hence the present study was undertaken.

The present study was conducted on buffalo fetuses (11) ranging from 5.2 to 65.0 cm curved crown rump length (CVRL) collected from abattoir. The fetal body was measured as a curved line in centimeter with a calibrated inelastic thread along the vertebral column between the most anterior part of frontal bone to the rump at ischiatic tuberosity and designated as curved crown rump length (CVRL) (Edward 1965). The approximate age of the fetuses was calculated by using following formula as derived by Soliman (1975).

$$Y = 28.66 + 4.496 X \text{ (CVRL} < 20 \text{ cm)}$$

$$Y = 73.544 + 2.256 X \text{ (CVRL} \geq 20 \text{ cm)}$$

where Y is the age in days and X is the CVRL in centimeter.

The thyroid gland was dissected out and fixed in 10% neutral buffered formalin (10% NBF). The samples were processed for paraffin blocks preparation by acetone-benzene schedule and the sections of 5–7 μ m were obtained on glass slides. These paraffin sections were stained with Haematoxylin and eosin for routine morphology (Luna 1968), Masson's Trichrome for connective tissue (Luna 1968), Verhoeff's stain for elastic fibres (Sheehan and Hrapchak 1973), Periodic acid Schiff (PAS) for neutral mucopolysaccharides and Alcian blue at pH 2.5 for acid mucopolysaccharides (Luna 1968).

The thyroid primordium was observed at 5.2 cm CVRL

Present address: ¹Assistant Professor (e mail: rajesh.ranjan837@gmail.com), BSCVM, Jhunjhunu Rajasthan.

²Assistant Professor, Apollo Veterinary College, Jaipur.

³Associate Professor (e mail: singhopinder68@gmail.com),

⁴Associate Professor-cum-Head (e mail: bansal.neelam@rediffmail.com), Department of Veterinary Anatomy, GADVASU, Ludhiana.

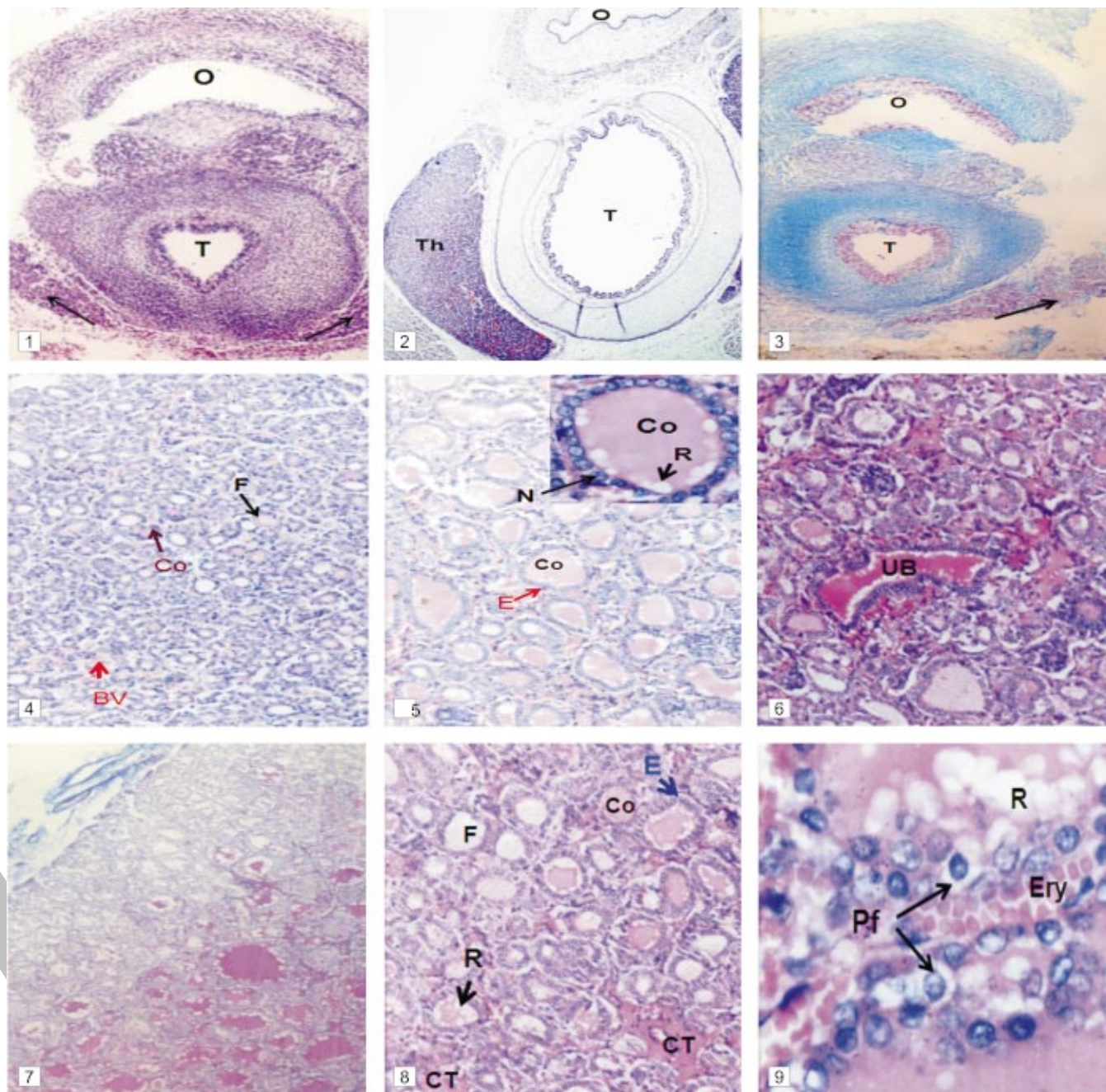
(52 days) of gestation on the ventro-lateral aspect of trachea (Fig.1) and comprised undifferentiated mesenchymal cells and few blood vessels up to 13.7 cm CVRL (90 days) (Fig.2). The moderately PAS positive parenchyma was surrounded by developing capsule (Fig.3). Kay *et al.* (2007) also reported the location of thyroid immediately anterior to the trachea in human beings.

At 18.5 cm CVRL (112 days) compact epithelial strands separated by loose connective tissue with few blood vessels were observed. Similar observations were made by El Sheikh *et al.* (1966) in camel. In the present study, the follicular arrangement began to appear in the developing thyroid at 20 cm CVRL (119 days) of gestation (Fig.4). The strongly PAS positive colloid also appeared in some of the follicles at this stage. Alwan (2009) reported the follicular development with colloid in sheep at 20 cm CVRL (75 days) while it appeared at 52 days of gestation in camel (El Sheikh *et al.* 1966). The collagen and reticular fibers in the capsule and h-stroma increased with advancing gestation.

The developing follicles were usually lined by simple cuboidal epithelium with large and centrally placed nuclei (Fig.5). Titlbach *et al.* (1987) reported cuboidal to columnar epithelium in developing cat thyroid. Similar findings were observed by El sheikh *et al.* (1966) in camel and Alwan (2009) in sheep. However in the present study, no change was observed in the follicular lining epithelium during the entire period.

The large irregular colloid filled follicles lined with stratified epithelium that appeared to be ultimobronchial follicles were observed in the developing thyroid at 25 to 28 cm CVRL (130–137 days) (Fig.6). Sayed *et al.* also (2005) observed remnants of ultimo-bronchial in the thyroid gland of buffalo.

Throughout the period of study, follicles were observed to be large in centre and small towards the sub capsular area. The capsule was moderate to strongly positive for acid mucopolysaccharides (Fig.7). The follicular epithelium showed mixed reaction for mucopolysaccharides. Similar findings were observed by El sheikh *et al.* (1966) in camel.



Figs 1-9. 1. Section from neck region of a 5.2 cm CVRL buffalo foetus (52 days) showing developing trachea (T), oesophagus (O) and thyroid primordium (arrow) at the ventro-lateral of trachea. H and E $\times$ 100; 2. Section from neck region of a 13.7cm CVRL buffalo foetus (90 days) showing developing trachea (T), oesophagus (O) and thyroid primordium (Th) at the ventro-lateral of trachea. H and E $\times$ 40. 3. Section from neck region of a 5.2 cm CVRL buffalo foetus (52 days) showing developing trachea (T), oesophagus (O) and thyroid primordium (arrow) having PAS positive paranchyma. PAS-AB $\times$ 100. 4. Photomicrograph of thyroid gland from 20 cm CVRL buffalo foetus (119 days) showing initiation of follicular arrangement (F), colloid (Co) and blood vessel (BV). H and E $\times$ 200. 5. Photomicrograph of thyroid gland from 41 cm CVRL buffalo foetus (166 days) showing thyroid follicles of different sizes lined with simple cuboidal epithelium (E). H and E $\times$ 200. [Inset- Spherical nuclei (N), resorption vacuoles (R) H and E $\times$ 1000]. 6. Photomicrograph of thyroid gland from 28 cm CVRL buffalo foetus (137 days) showing presence of Ultimobronchial follicle (UB) lined with stratified epithelium. H and E $\times$ 200. 7. Photomicrograph of thyroid gland from 65 cm CVRL buffalo foetus (220 days) showing large follicles in the centre and small at the periphery. PAS-AB $\times$ 100. 8. Photomicrograph of thyroid gland from 28 cm CVRL buffalo foetus (137 days) showing presence of resorption vacuoles (R) in the colloid material of thyroid follicles, simple cuboidal epithelium (E) and developing connective tissue (CT). H and E $\times$ 200. 9. Photomicrograph of thyroid gland from 65 cm CVRL buffalo foetus (220 days) showing parafollicular cells (Pf), resorption vacuoles (R) and erythrocytes (Ery). H and E $\times$ 1000.

The resorption vacuoles in the colloid material were first observed at 28 cm CVRL (137 days) (Fig.8). El Sheikh *et al.* (1966) observed clear vacuoles in the intra follicular colloids by 70 days of gestation in camel and Alwan (2009) reported its presence at 30 cm CVRL (100 days) in sheep. The presence of colloid and resorption vacuoles is indicative of secretory activity of gland (El Sheikh *et al.* 1966). The C-cells or parafollicular cells were observed at 65 cm CVRL (220 days) of gestation in between the epithelial cells of developing thyroid follicles (Fig.9). Titlbach *et al.* (1987) reported the presence of C-cells in cat foetal thyroid at 38–39th day of gestation.

SUMMARY

The present study was conducted to study histogenesis of thyroid gland in buffalo fetuses (n=11) ranging from 5.2 cm (52 days) to 65.0 cm (220 days) curved crown rump length (CVRL). The parenchyma of thyroid gland comprised randomly arranged undifferentiated mesenchymal cells with large number of blood vessels up to 13.7 cm CVRL (90 days). Initiation of follicular arrangement was observed in some of the developing follicles at 20 cm CVRL (119 days). The resorption vacuoles appeared in the colloid at 28.0 cm CVRL (137 days) and invasion of connective tissue in the parenchyma of gland was observed. Ultimo-bronchial follicles were observed at 25 to 28 cm CVRL (130–137 days) and C-cells at 65 cm CVRL (220 days).

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