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Histological, Histometrical and Hormonal Assay ComparativeStudy of Thyroid Glands between Suckling and Adult Local Male Cats (*Felis catus)*

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Abstract

The present study was amid to describe on histological, histometric and hormonal assay characteristics of the thyroid glands in suckling and adult local male cats(*Felis catus)*appeared the healthy. The study was conducted on ten suckling (5) and adult (5) local male cats. The histological structural of thyroid gland in suckling and adult cats was similarstatistical all histological measurements of left lobe in suckling and adult cats were higher significantly than those of the right one. The thickness of capsule, diameter of large, medium, small follicle and height of epithelium histological measurements of right and left thyroid lobes in adult cats were higher significantly then those in suckling at p <0.05. these variations in may be due to the differences of studied cats.

Keywords: Histological, Histometrical, Thyroid gland, Thyroid hormones.

Conclusion:

Thyroid gland histological structure was similar insuckling and adult cats, this finding mean that the thyroid gland was function at birth but difference in the histological measurements means that the thyroid gland was continuous in its growth and development with the growth of animal body and body organs.

I. Introduction

The domestic cat (*Felis catus* or *Felis domestica*)used as laboratory animal since ithasa similar physiological characteristic to laboratory rabbits and rodents and it is highly used in biomedical and behavioral researches, especially of neurological conditions (James, 1995).Histologically, the structure of thyroid gland is varies depending to numerous factors. Mostly in species, thyroid is envelope by a capsule that send trabeculainto the glandular parenchyma which dividing it into numerous lobules, which contained of ancombination of variesshapeandsize of follicles and have a single layer of epithelial cells lining them and differ in the quantity of colloid material supported by inter follicular connective tissue rich in capillaries (Louise and Waugh, 2002; Igbokwe and Ezeasor, 2015).Some of them contain a small amount, and few are completely devoid of this substance (Enemali*et al*., 2016; Kadhim, 2022).

II. Materials and Methods:

Animals' ethics

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The Committee on Research Ethics in the Faculty of Veterinary Medicine of the Baghdad, authorized every procedure utilized in this study with the assigned code number PG/ 2335-IN 25/10/2023

Animals of study

The current investigation was conducted out on ten healthy sucking (1-7 weeks) age and adult local male cats (1 years) age. After recording the body weight in sucking was (464.00 ±29.63) gm and for adult was (3320.00±204.02)gmthese animals were used for histological, histochemical and hormonal assay of thyroid gland.

Study design

Fivesuckling and five of adult male's cats used for histological, histochemical studies. After extirpation of thyroid glands, it washed by normal saline solution. Small piece of tissue (1 cm) from the two thyroids right and left lobs was fixed with (10%) formalin solution for (48)hr.washed with running water overnight then dehydrated in a seconding grades alcohol, cleared in xylene then embedded in paraffin wax section of (6-7µm) and stained with hematoxylin and eosin stain for describe the histological structure, Masson's Trichromestain for collagen fibers(Khaleel *et al.*, 2017; Kadhim and Khaleel, 2021;Khaleel, 2022 and Khaleel and Alkhazraji, 2022).Toluidine blue stain for C-cells and PAS stain for glycoprotein and mucopolysaccharides identification(Luna,1968; Bancroft, 2012 andKhaleel *et al.*, 2023).The histological sections were inspected using an Olympus/Japan light microscope at various magnifications (X4, X10, X40, and X100)and the microscopy was captured on camera using a Sony Cyber-shot 14.2 mega pixel digital camera (Khaleel and Salih 2017)

Hormonal analysis

Blood samples were collected from fivesuckling and adult local male cats at morning after anesthesia by heart puncturing into the gel tubes. After centrifugation, the serum used for hormonal assay test of (T3 and T4) by using ELISA kit according to the manufacturer's instructions (Braverman, 1996; Gharib and Mazzaferri, 1998and Batah and Mirhish, 2019)

Data analysis

The t-test was used for statistical analysis of the recommended comparison approaches of microscopic variances of the right and left thyroid glands between sucking and adult cats and the significance level at $P<0.05$ (SAS, 2018).

III. Results and Discussion:

Histological results of thyroid gland in suckling and adult cats:

Generally, the thyroid gland's structure in suckling and adult male cats was similar to the common pattern of histological structure of thyroid in mammals and in domestic animals (Dellmann, 1993).

Thyroid Capsule:

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Thyroid gland at suckling and adult male cats appearedenvelope by a thin connective tissue capsule made of two layers; thin external layer of adipose tissue with clear cellular limits interposes the fine and coarse bundles of collagenous fibers with a few elastic fibers, and an inner dense irregular connective tissue contains fire collagen and elastic fibers with fibroblasts in a spindle shape (Fig.1, 2, 3). Fine connective tissue trabeculae was extended from the capsule and penetrate the glandular parenchyma and divided it into distinct different size of lobules which made up of variable size and shape follicles (Fig. 4, 5).

In suckling cats, the mean capsule thickness of rightand left lobes waslisted in (Table 1).The mean thickness of right lobe was lower significantly ($p < 0.05$) than that of left lobe (Table 1).This result was similar to that reported (Batah, 2019) in Albino rats and guinea pigs, while the capsule thickness of right and left lobes in adult cats were listed in (Table 1). The mean thickness of right lobe was lower significantly ($p < 0.05$) than that of left lobe (Table 1). This finding wasdisagreeing with that by (Batah, 2019) in Albino rats, while similar to in guinea pigs who said the means capsular thickness of left lobe was higher than the right lobe in Albino rat, this difference may be due to different in gland activity, the thickness of capsule in right and left lobes of adult cats were higher than significantly at $p < 0.05$ them those of the suckling cats (Table 2).

Thyroid Follicles

Bothleft and rightthyroid lobes were consisted of anaccumulation of numerous shape and size follicles,there were threechief sizes small, medium large-size follicles were identified, and different shapes of follicles which were oval, irregular,round and elongated shapes (Fig.6,7,8)this was in agree with (Hammodi and Al Aamery, 2023)in *FelisCatus*. Each follicle was surrounded by a basement membrane, thin collagen connective tissue fibers with fibroblast and numerous blood capillaries (Fig. 6,7) This feature was observed previouslyin Weasel *(Herpestesjavanicus*) by (Al-Aamery and Dauod, 2016).Largefollicles were distributed among small and medium-sized follicles were under the capsule and toward the center (Fig.2,4,5,6).

The medium and small size follicle were dominatedlyin center of gland and observed as spherical or oval shape (Fig. 1,2, 4),similar results were reported by (Igbokwe and Ezeasor, 2015) in adult male African giant rats, while in other species such as rats the larger follicles tend to be only peripherally present (Hartoft-Nielsen *et al.*, 2005). The thyroid follicles appeared not homogenous or in different shapes such as oval and rounded follicles which were predominate.These finding were similar to that of (Enemali*et al*., 2016) in adult male African giant rats, (Igbokwe, 2010) in grass cutter, (Seraphim, 2014) in *Hipposideroslankadiva* and (Sekulic *et al*., 2007) in pigs.

In the right and left lobes of thyroid gland, the mean follicles diameters of large, medium and small-sized follicles at suckling werelisted in(Table 1). Statistically, there were significant differences in these parameters between left and right lobes at $p< 0.05$ (Table 2).Statistically, revealed the means diameter of follicles in left lobe was higher significantly at $p < 0.05$than those of right lobe (Table 1).In adult cats mean follicles diameters of large, medium and small -sized follicles werein adult for the right and left lobes were listed in (Table 1). The thyroid follicular lining epithelial cells was mostly

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simple cuboidal epithelium contains spherical nuclei and in some follicles were simple squamous epithelium (Fig.8,9)**.**

The mean height of follicular epithelial cellsat suckling and adult cats for the right and left were listed in (Table 1).Statistical no significant difference between right and leftlobes at (p< 0**.**05) (Table 2)**.**Similar result was reported by (Batah andMirhish, 2019) in Albino rats and guinea pigs.

Parafollicular cells or Clear cells (C-cells)

Generally, few parafollicular cells were present in comparison with follicular cells, it appeared asrounded to oval large cells, exhibit lighter-stained cytoplasm than of the follicular cells with densely stained nuclei**,**found as a single cell or in cluster of two-three cells located between the follicular cellsand rested with the follicular cells on the basement membrane but notreaches to the follicular lumen.Some other cells located within interfollicular tissue (Fig**.**7,8,9).These finding correspond with that of (Chen *et al*., 2000) in hamsters and Albino rats and (Igbokwe, 2010) in African grasscutter. These cells were spreadun equally in the gland and tend to be concentrated in the lobule center (Kaufman *et al.,* 2010; Chavhana and Dhaman, 2015 and Batah and Mirhish, 2019).

Colloid substance

Many of thyroid follicles were contains a different amount of eosinophilic colloid material within the follicular lumen. There were uniformity and nonuniformly stained of colloid substance in which many or few peripheral visible empty vacuoles showed as clear areas in a stored colloid. Some follicles identified without colloid material (Fig. 5,8,10). Colloid material showed appeared as variable degree of density inside the follicles, some of them were contain densely stained eosinophilic colloid substancethat react positive towards PAS stain and showed magenta color, while follicles contained faintly stained colloid with PAS stain (Fig. 11). These results coincide with (Parchami and Dehkordi, 2012) in rabbits, in sheep and gazelles(Salih, 2015; Iman and Ali, 2017), (Enemali*et al.,* 2016) in African giant pouched rats and indicate that the colloid substance which was thyroglobulin that was iodinated glycoprotein substance (Kierszenbaum and Tres, 2012; Santos *et al.,* 2013)

Table 1. Comparative histological measurements of left andright lobes of thyroid gland in suckling andadult local male cats.

Type of animal	Histological measurements(µm)	Right Lobe	Left Lobe	T- test
		Mean ± SE	Mean ± SE	
Suckling	Thickness of capsule	245.00 ±2.23	265.00 ±4.47	1.49 *
	Diameter of large Follicle	175.40 ±0.81	185.00 ±2.23	11.53 *

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	Diameter of Medium Follicle	84.00 ±1.04	96.00 ±1.87	5.48 *
	Diameter of small Follicle	36.40 ±2.11	45.00 ±2.23	4.94 *
	Height of epithelium	6.60 ±0.60	9.60 ±0.24	7.09 *
Adult	Thickness of capsule	303.00±4.89	320.00 ±8.36	22.35 *
	Diameter of large Follicle	195.80±0.96	213.40±6.20	14.49 *
	Diameter of Medium Follicle	110.60±1.63	127.70±2.97	7.62 *
	Diameter of small Follicle	53.10 ±2.62	72.00 ±4.96	12.93 *
	Height of epithelium	12.80 ±0.63	18.00 ±0.52	3.72 *
* (P<0.05) Significant, NS: Non-Significant.				

Table 2. Histological measurements of right and left lobes of thyroid gland in suckling and adult localmale cats.

Parts of gland	Histological measurements(µm)	Suckling	Adult	T- test
		Mean ± SE	Mean ± SE	
Right Lobe	Thickness of capsule	245.00 ±2.23	303.00±4.89	25.09 *
	Diameter of large Follicle	175.40 ±0.81	195.80±0.96	11.66 *
	Diameter of medium Follicle	84.00 ±1.04	110.60±1.63	8.73 *
	Diameter of small Follicle	36.40 ±2.11	53.10 ±2.62	10.08 *
	Height of epithelium	245.00 ±2.23	12.80 ±0.63	3.21 *
Left	Thickness of capsule	265.00 ±4.47	320.00 ±8.36	18.93 *
	Diameter of large Follicle	185.00 ±2.23	213.40±6.20	3.02 *
	Diameter of medium Follicle	96.00 ±1.87	127.70±2.97	4.39 *

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Lobe	Diameter of small Follicle	45.00 ±2.23	72.00 ±4.96	6.71 *
	Height of epithelium	9.60 ±0.24	18.00 ±0.52	1.58 *
* (P<0.05) Significant, NS: Non-Significant.				

Table 3. Comparison between suckling and Adultlocal male cats in level of thyroid hormones.

Hormones	Suckling	Adult	T-Test
	Mean ± SE	Mean ± SE	
T4 (µg/dl)	2.89 ± 0.19	4.24 ± 0.27	1.02 *
T3 (ng/ml)	1.65 ± 0.07	3.47 ±0.20	0.996 *
* (P<0.05) Significant.			

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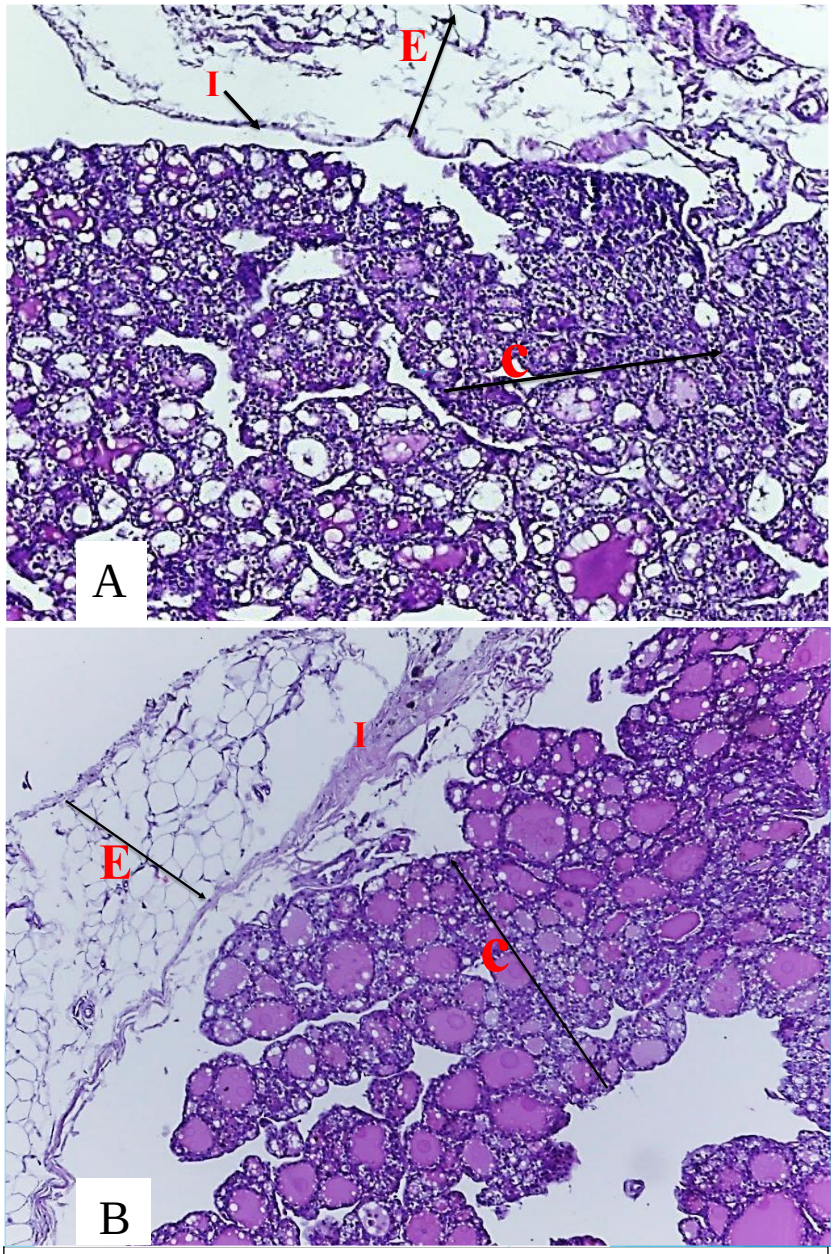

(Fig.1). Histological section of thyroid gland in male cats shows: E- External layer (adipose tissue), I- Internal layer (collagenous connective tissue), C- Thyroid lobule
A- Suckling cats B- Adult cats (H&E stain, X100).

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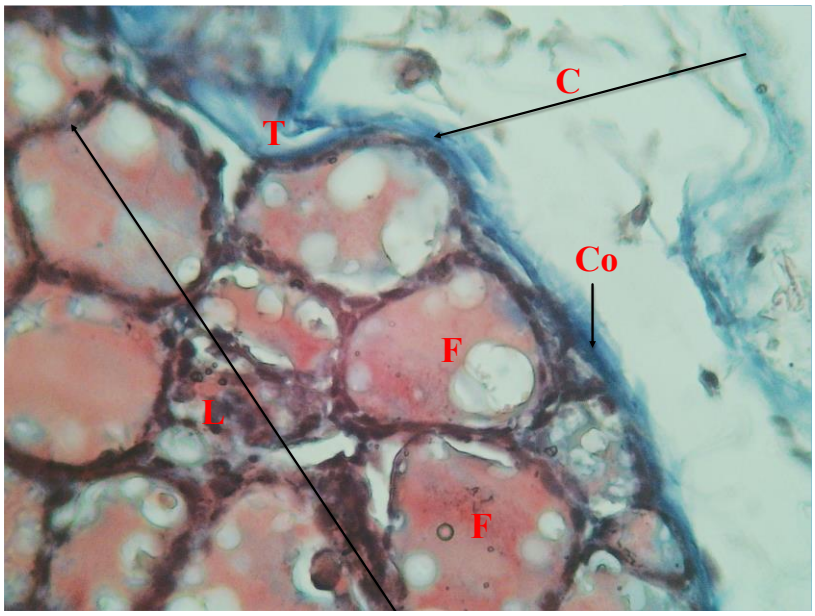

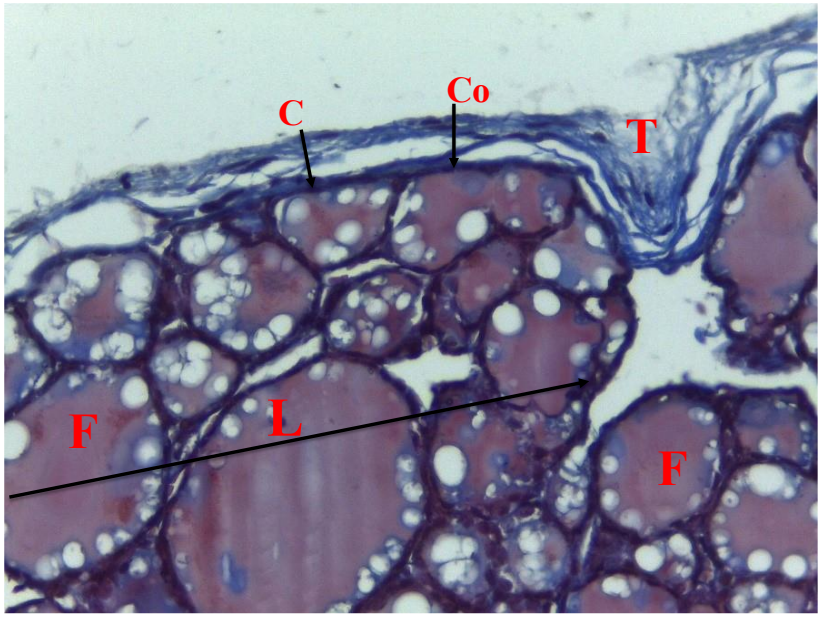

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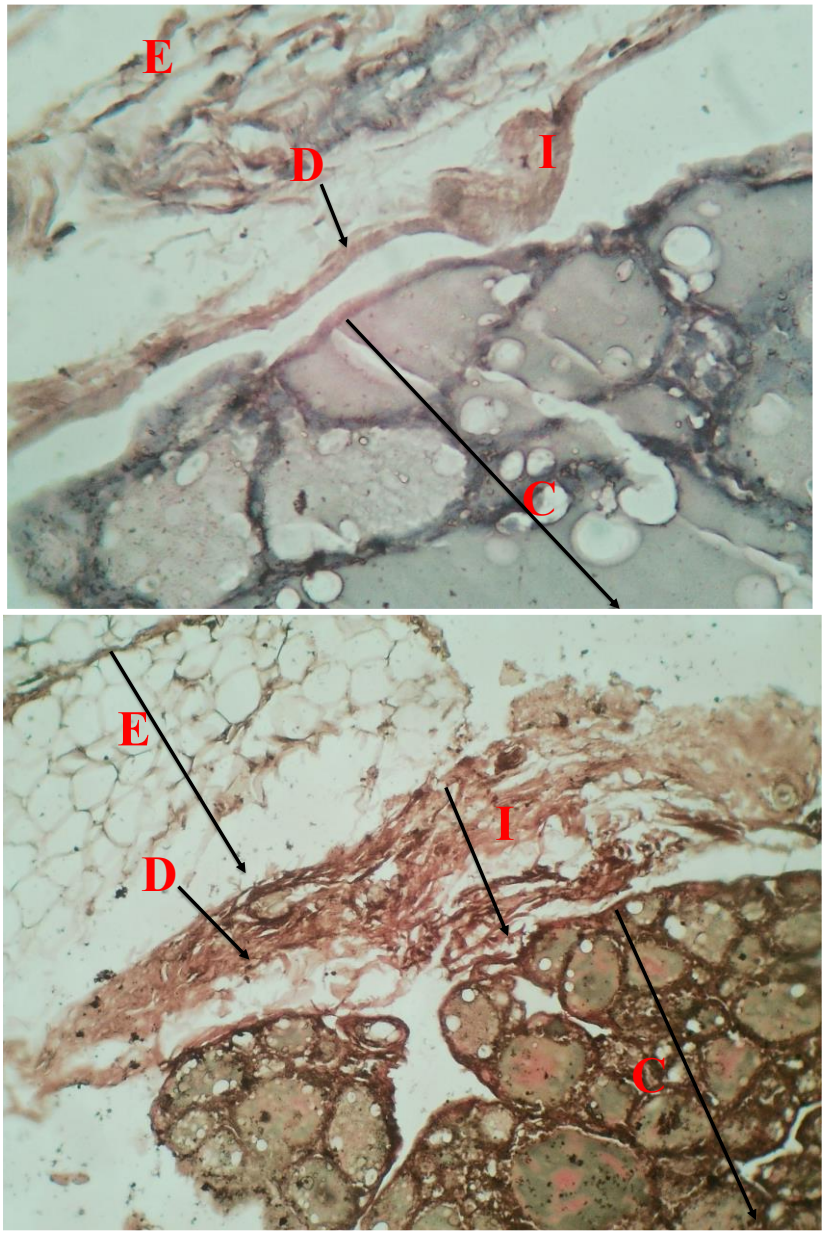

(Fig.3). Histological section of thyroid gland in male cats shows: E- External layer (adipose tissue), I- Internal layer (collagenous connective tissue), C- Thyroid lobule, D- elastic fibers.
A- Suckling cats B- Adult cats (Verhoffs stain, X100).

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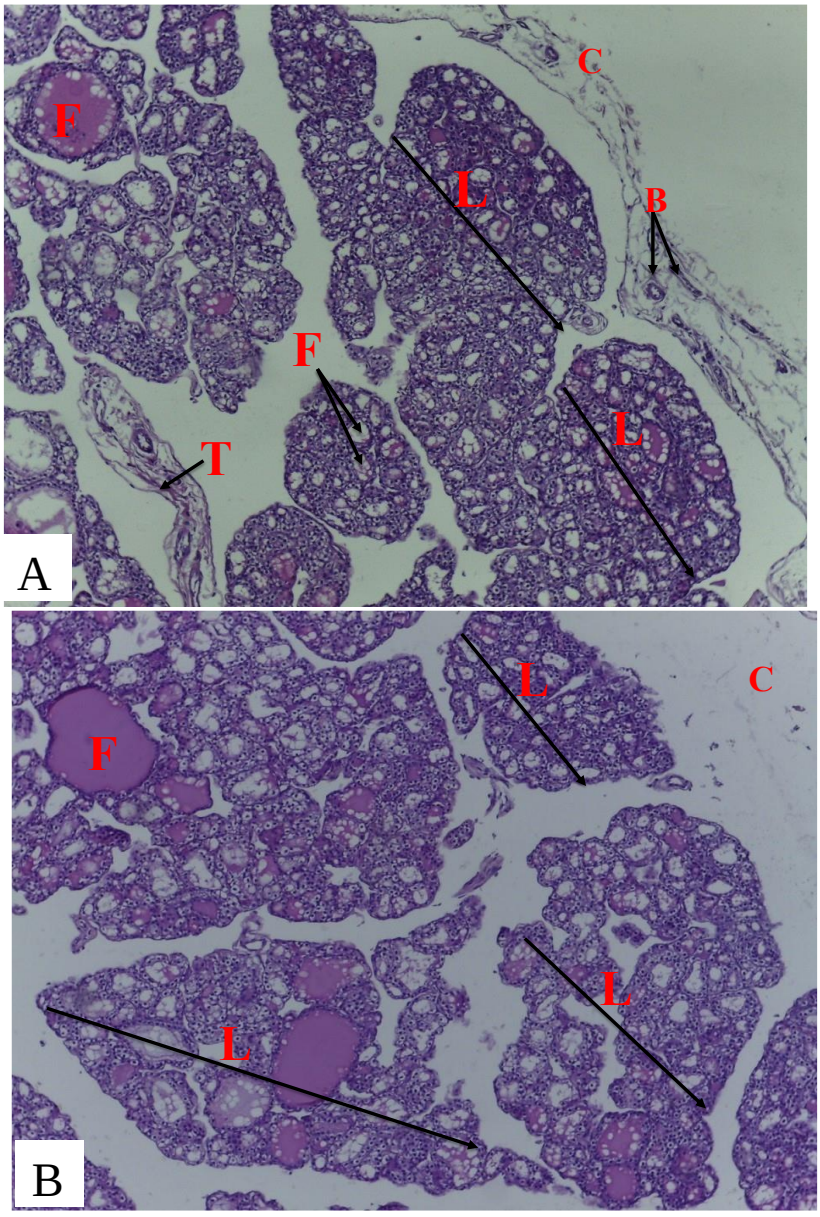

(Fig.4). Histological section of thyroid gland in male cats shows: C-Capsule, B-blood vessels, L-Lobules,F-Follicles, T-Trabeculae. A- Suckling cats B- Adult cats(H &E stain X100).

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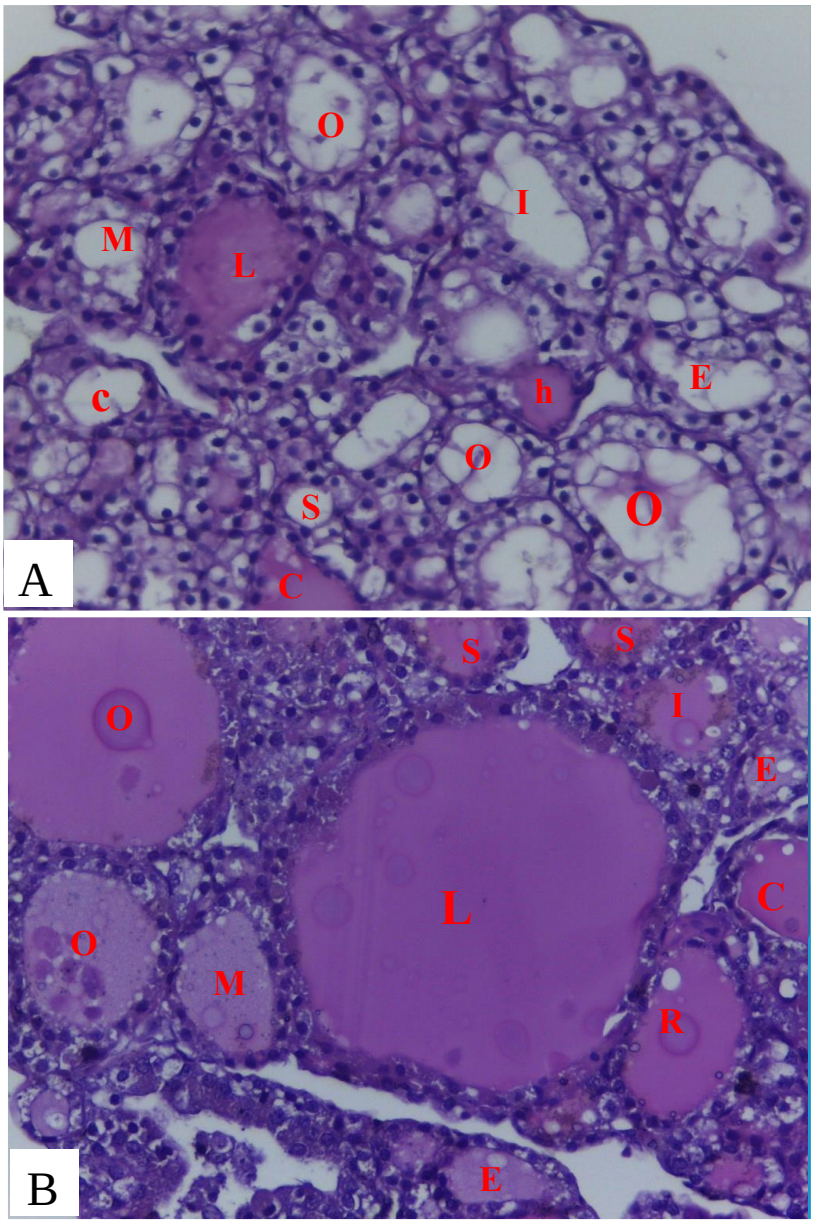

(Fig.5). Histological section of thyroid gland in male cats show Follicles: S- Small size follicle, M-Medium size follicle, L- Large size follicle, O- Oval shape, R- Round shape, I- Irregular shape, E- Elongated shape, C- Colloid material.
A- Suckling cats B- Adult cats(H &E stain X400).

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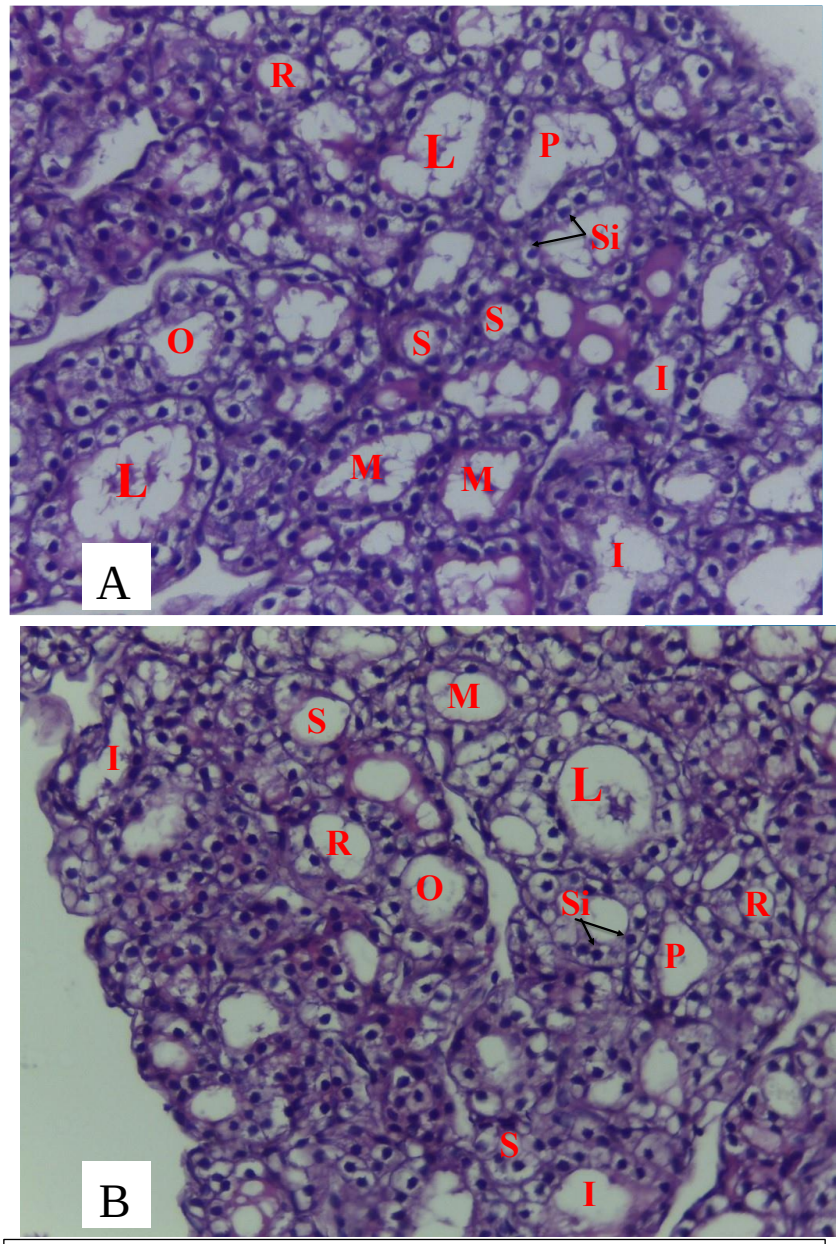

(Fig.6). Histological section of thyroid gland in male cats shows: S- Small follicle, M-Medium follicle, L- Large follicle, R- Rounded follicle, O- Oval follicle, P- Polygonal follicle, I- Irregular follicle A- Suckling cats B- Adult cats(H &E stain X400).

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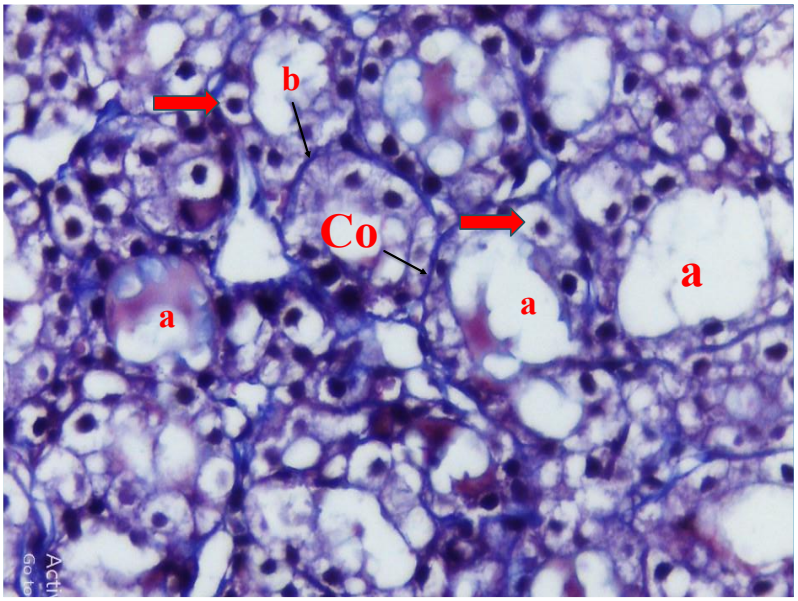

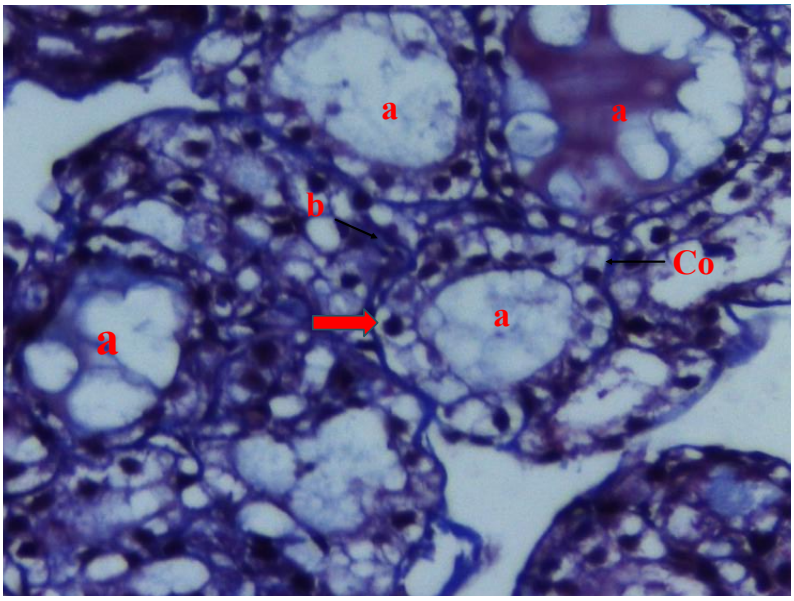

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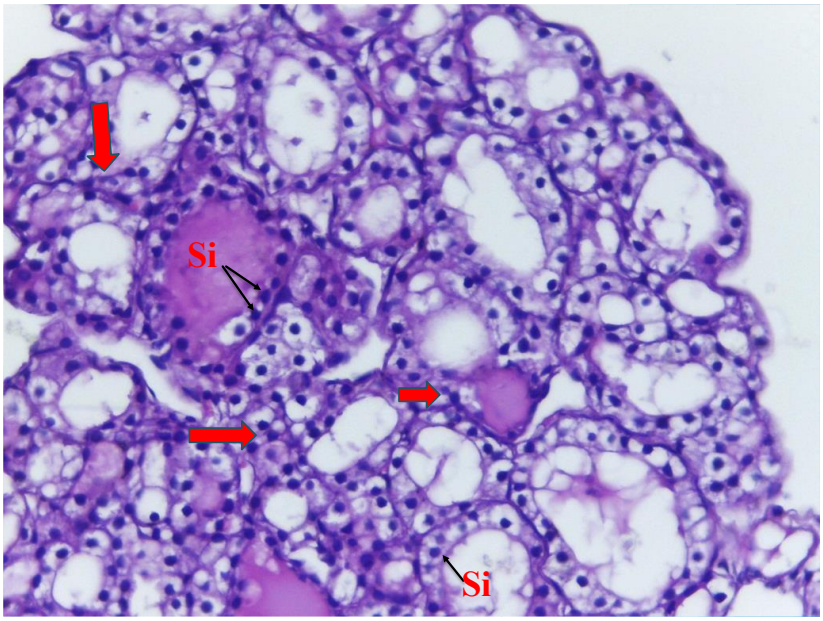

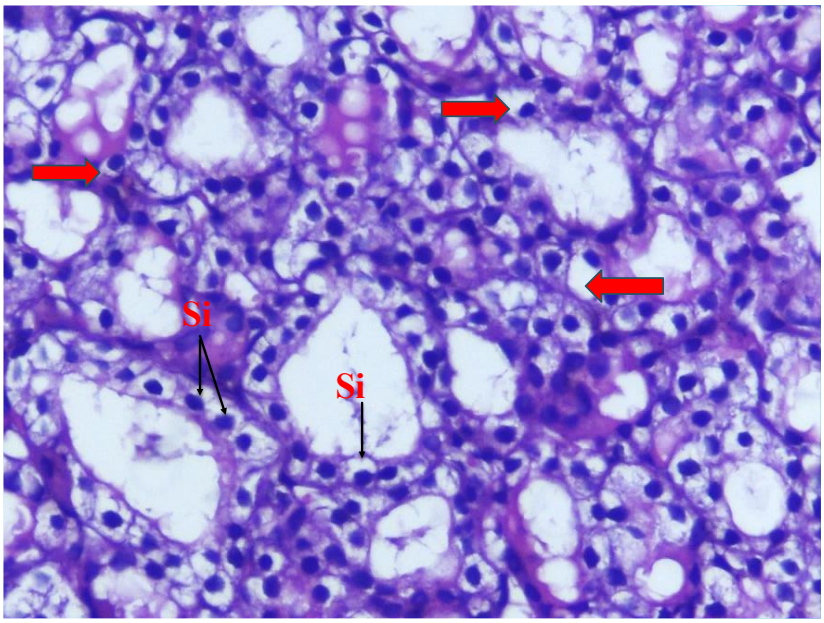

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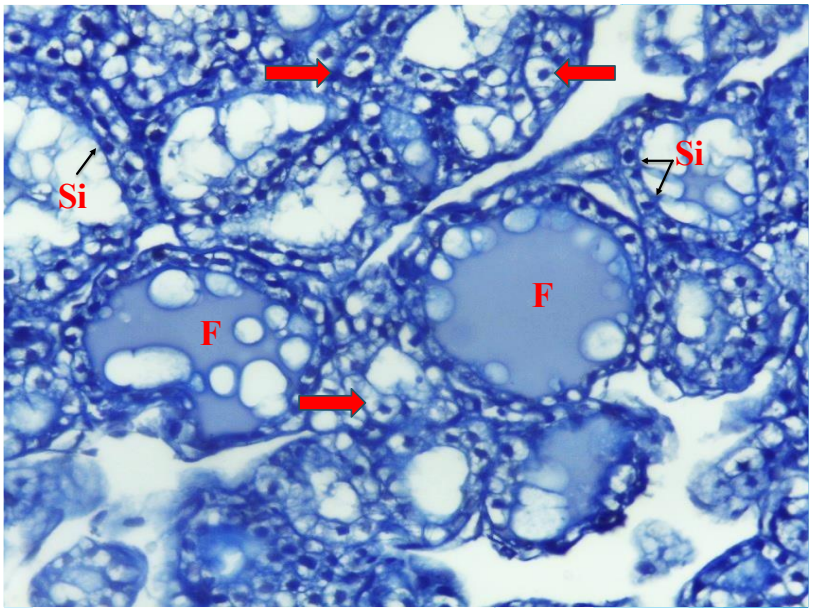

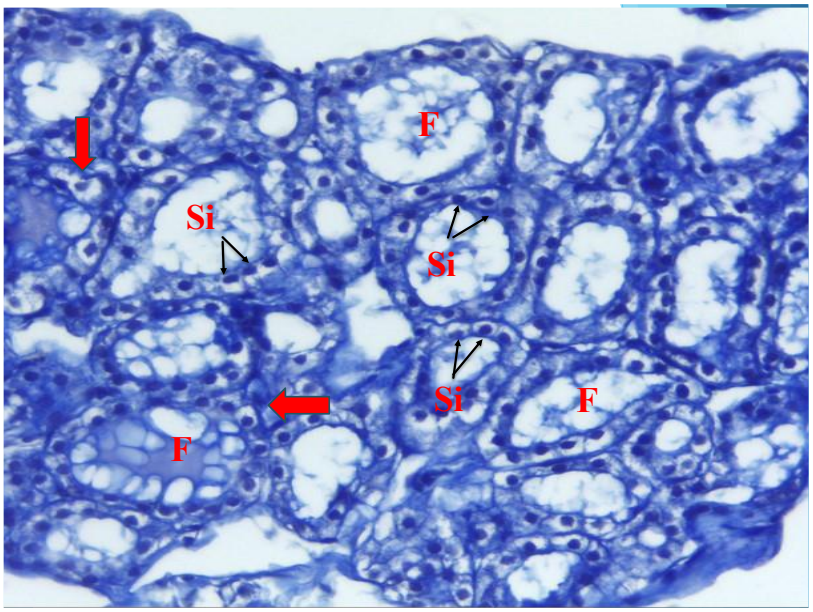

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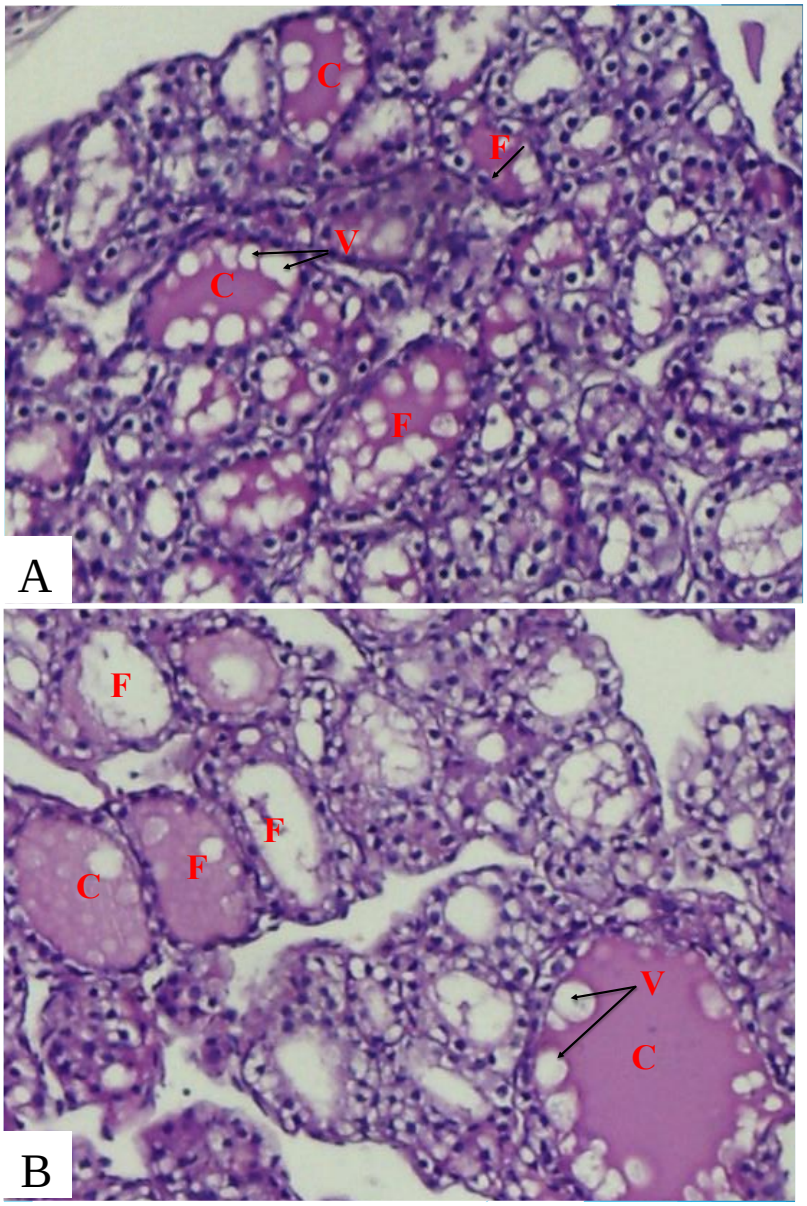

(Fig.10). Histological section of thyroid gland in male cats shows: F- Follicle, C- Colloid material, V- Vacuole.
A- Suckling cats B- Adult cats(H &E stain X400).

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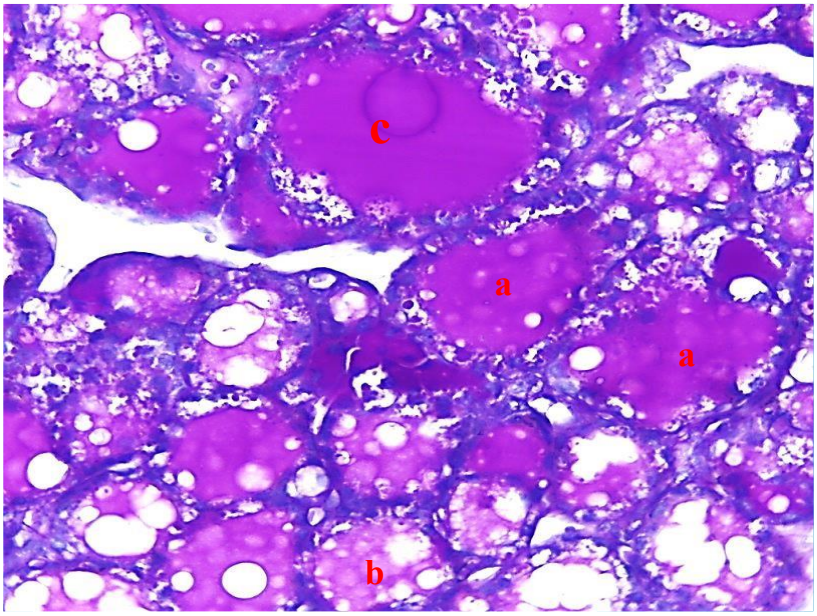

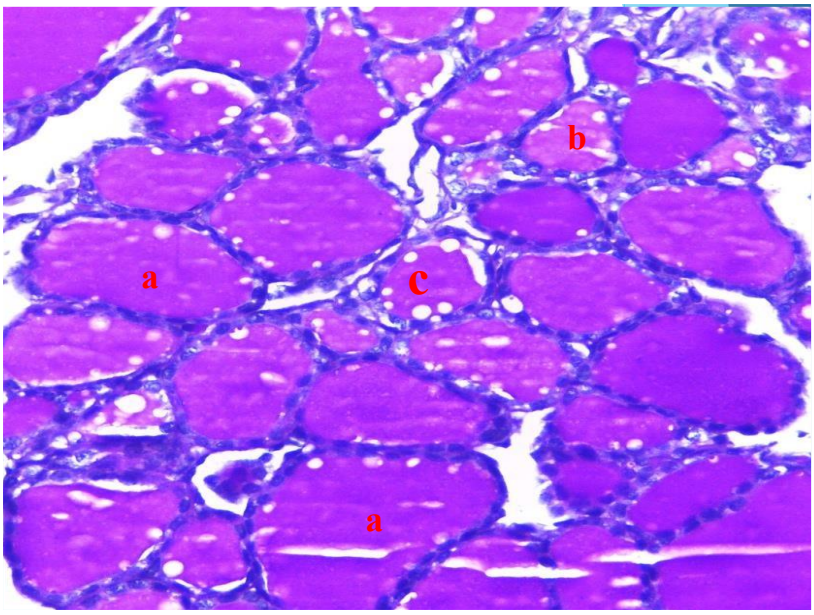

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