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Original Article

Fine structure of thyroid gland in wild caught female bat *Taphozous kachhensis* (Dobson) during reproductive cycle

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ABSTRACT

The thyroid gland has long been recognized as an important modulator of reproductive function. Both hyper- and hypo-thyroidism are associated with reproductive dysfunction and infertility. In addition, thyroid hormones appear to play a key role in the expression of events that underlie seasonal reproductive cycles. Despite the considerable information is available on the histology of thyroid gland in some of the bats, literature related to study is very few, and further information available on the ultrastructure is also scanty. The specimen of *Taphozous kachhensis* was collected from Ambai Nimbi. Many collections were made during the breeding season so as to coincide with the time of reproductive cycle and to get an accurate pregnancy record. Thyroid is removed from the bat and cut into 1–2 mm piece and immersed in fresh ice-cold 3% gluteraldehyde solution for 2–4 h and send for further process. The thyroid gland is more active during estrus than the pregnancy and lactation. The cell organelles observed in follicular cell of thyroid gland indicate more synthetic activity. While during pregnancy and lactation the thyroid gland is not synthetically active but the appearance of more lysosomal bodies during pregnancy indicate the utilization of stored colloid and release of thyroid hormone in blood stream. The parafollicular cells are more developed during pregnancy and lactation than estrus as indicated by ultrastructural characteristics. These cells may secrete calcitonin, serotonin, and somatostatin and may be responsible for controlling the seasonal changes in plasma calcium concentration in this bat.

1. Introduction

The thyroid gland has long been recognized as an important modulator of reproductive functions. Both hyper- and hypo-thyroidism are associated with reproductive dysfunction and infertility. Direct effects of thyroid hormones on cellular metabolism and hormone responsiveness have been demonstrated in the gonads and the sex accessory organs. In addition, thyroid hormones appear to play a key role in the expression of events that underline the seasonal reproductive cycles. The thyroid gland is an important modulator for reproductive functions. The metabolic hormone, thyroxin, T4 is the physiological regulator of energy balance as well as in maintaining normal reproductive function in mammal [12,14]. They reported that the T_3 is essential for mammalian reproduction in cattle, the deficiency of which result into female infertility. In 1994, work by researchers in Michigan implicated thyroid hormones as having a role in decreasing LH secretion at the end of the breeding season, resulting in the normal cessation of follicular activity and ovulation. Along with follicle stimulating hormone (FSH), luteinizing

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hormone (LH) and indirectly, gonadotropin-releasing hormone (GnRH) are hormones responsible for follicle growth and ovulation. Electron microscopic structures of follicular cell of thyroid gland in *Hipposideros lankadiva* [13]. Thyroid follicle have been characterized by the presence of dilated rough endoplasmic reticulum, well developed Golgi apparatus, mitochondria with lamellar cristae and apical blebs towards the luminal plasma membrane, suggesting synthesis of thyroid hormone during late pregnancy and lactation. Although significant contribution have been made to thyroid histology and endocrinology of mammals including bats, a very little information is available on cyclic changes in the thyroid gland during different phases of reproductive cycle.

Despite the considerable information is available on the ultrastructure of thyroid gland in some of the bats, literature related to study is very few. Detailed study of the ultrastructural and functional characteristic of the thyroid gland of *Taphozous kachhensis* during different phases of reproductive cycle has been undertaken because earlier no such study has ever been made on this microchiropteran species. And also find out the probable role of endocrine gland in the control of reproduction in this Emballonurid bat.

2. Material and methods

Taphozous kachhensis (Dobson) is an exclusive Indian Emballonurid bat found in caves, tunnels and temples. This species was selected bat selected for present study because of its unique habits. The gestation length of female of this species is about 150 days. The collection of the specimen commenced in February 2006 and the last specimen for the present study was collected in May 2009. The specimen of Taphozous *kachhensis* were collected from Ambai Nimbi, about 45 km from Bramhapuri Taluka, District, Chandrapur, Maharashtra, India. Many collections were made during the breeding season so as to coincide with the time of reproductive cycle and to get an accurate pregnancy record. During the day time, their roosting places were visited and the specimens were netted at random with the help of a butterfly net. These bats are very sluggish in nature after collection they were sexed and only the females were brought to the laboratory. Weight was recorded with sensitive spring balance before they were sacrificed.

2.1. Transmission electron microscopy

Thyroid, adrenal gland, pituitary gland and the ovary of the species from pregnant, non-pregnant and lactating specimen were selected for electron microscopic study.

2.1.1. Fixation

Thyroid, adrenal gland, pituitary gland and ovary were removed from the bat and cut into 1–2 mm piece and immersed in fresh ice-cold 3% gluteraldehyde solution. The fixation was carried out over a period of 1–2 h at 4 °C. A fresh change of cold gluteraldehyde was given at the end of fixation and the tissue were washed in cold 0.1 M sodium cocodylate buffer for half an hour with 3–4 changes to ensure complete removal of excess gluteraldehyde. Post fixation with OSO_4 or osmification with 1% OSO_4 in sodium cocodylate buffer was carried out for 2 h at 4 °C.

2.1.2. Dehydration

Dehydration of tissue was carried out by passing the fixed tissues through a graded series of alcohol of increasing concentration of the dehydrating agent in water ending with absolute alcohol. Most epoxy resins are soluble in ethyl alcohol and acetone but they mix much in propylene oxide. Thus tissues were passed through intermediate solvent, propylene oxide over a period of half hour.

2.1.3. Infiltration and embedding

Complete and uniform penetration of tissue by the embedding medium is accomplished through infiltration and embedding. Infiltration involved the gradual replacement of dehydrating agent with embedding medium while embedding consist of complete impregnation of the interstices of a tissue specimen with the medium. This was done as follows:

i) Propylene oxide araldite 'A' solution 1:1 for one hour at room temperature.
ii) Fresh araldite 'A' solution – kept at room temperature in desiccator overnight.
iii) Araldite 'B' solution – for 1 h at room temperature.

Embedding of tissue was done in plastic BEEM capsule with fresh araldite 'B' solution and the capsule was kept in an oven maintained at 60° C for 24–48 h to ensure polymerization. Blocks were freed from the sample by cutting away the plastic, then trimmed with safety razor blade under a stereo-microscope, to a flat surface cone, to remove the excess embedding material ultrathin section of 1–2 μm in thickness were cut on an LKB ultratome V, with glass knife maker. These sections were dried on hot plate (60 °C) and consequently stained with 1% toludine blue (20–30 s) and observed on light microscope. The selected areas for ultrathin section were marked out. The blocks were further trimmed and ultrathin section were marked out the block were further trimmed and ultrathin section or thin sections' 600–900 Å thick corresponding the pale gold colour of section were cut section were collected on 300 mesh copper grids. To enhance the contrast double staining technique was employed. The grid was subjected to 10% alcoholic uranyl acetate for half an hour followed by lead citrate for 10 min. All grids were observed on a JEOL-100S electron microscope at 80 kV accelerating voltage. Micrographs were taken of the desired sample at different planes.

3. Result

3.1. Electron microscopic study of thyroid gland

3.1.1. Thyroid gland during estrus

Bats collected in the month of November are in estrus. The thyroid gland during estrus shows the following ultrastructural characteristics.

The small size follicles are lined by high cuboidal epithelium with spherical nuclei. The chromatin material is

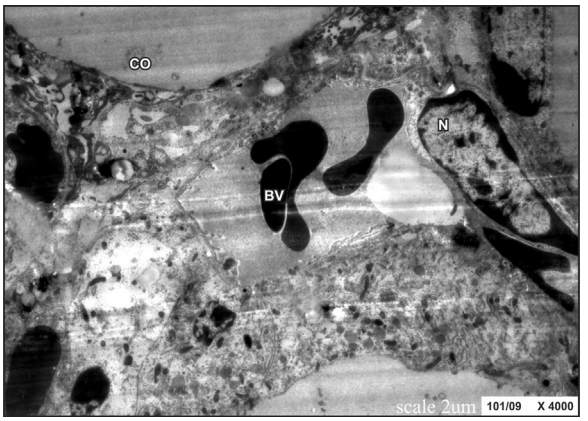

Fig. 1. Electron micrograph of the thyroid gland of female bat during estrus showing small active follicle lined by cuboidal to low columnar epithelium with elongated nucleus [N] and lumina of the follicle filled with colloid material [CO]. 4000×.

attached to the inner surface of the nuclear membrane. Two or three nucleoli were observed within the granular nucleoplasm (Fig. 1). Nuclear pores are frequently observed. Elongated tubules of rough endoplasmic reticulum are frequently observed in the cytoplasm of the follicular cell and are heavily dotted with ribosomes. Some dilated profiles of rough endoplasmic reticulum are also observed in the cytoplasm of the follicular cell with close association with mitochondria. The lumen of small follicle is filled with homogenous colloid material and microvilli are numerous. Golgi complex is inconspicuous. Free ribosomes are also scattered throughout the cytoplasm. Mitochondria are abundant and are evenly distributed throughout the cytoplasm. Mitochondria are mostly spherical but some are elongated in shape with lamellar cristae, having electron dense mitochondrial matrix. Some dense, spherical lysosomal bodies of variable size are seen in the cytoplasm (Figs. 2 and 3). Large size follicles are lined by squamous epithelium with oval nuclei. The number of mitochondria and rough endoplasmic reticulum are reduced.

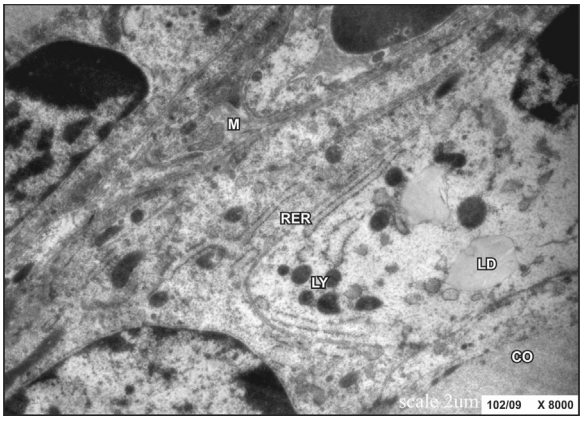

Fig. 2. Electron micrograph of follicular cell of the thyroid gland during estrus. Note the presence of dilated profile of rough endoplasmic reticulum [RER] appeared as elongated cisternae dotted with ribosomes. Note the presence of lysosomes [LY] and few mitochondria [M]. 8000×.

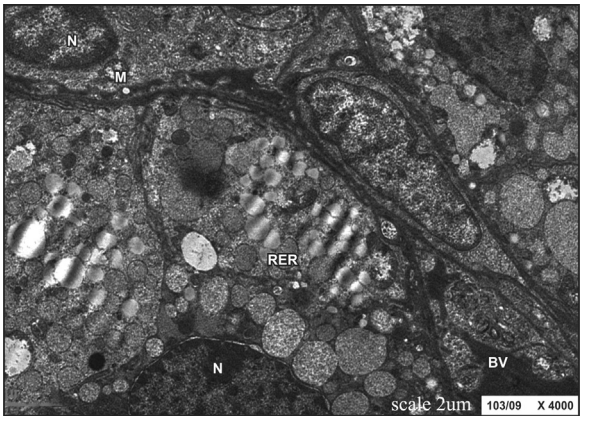

Fig. 3. Electron micrograph of follicular cell of the thyroid gland during estrus showing oval nucleus [N] and clumps of chromatin material is present at periphery. Note the presence of several dilated round profile of rough endoplasmic reticulum [RER]. Several dense bodies [DB] and lipid droplets [LD] are seen in the apical region of the cell. Mitochondria [M] are seen with collapsed cristae. Note the presence of blood vessel [BV]. 4000×.

3.1.2. *Parafollicular cells*

The parafollicular cells are observed at the base of the follicular epithelium between the follicular cells and are filled with electron dense secretory granules. These are elongated to oval in shape and are electron lucent and larger than follicular cell. The nucleus is large and irregular in shape with indented margin and occupies the large portion of the cytoplasm. Nucleolus is clearly visible. Chromatin clumps are seen along with the nuclear membrane (Fig. 4). Rough endoplasmic reticulum occurs as short tubules. Mitochondria are scattered throughout the cytoplasm.

3.1.3. *Thyroid gland during early pregnancy*

Ovulation, insemination and fertilization take place in early December in this species of female bat. Implantation of the blastocyst is observed during the last week of December. By the end of December the entire female

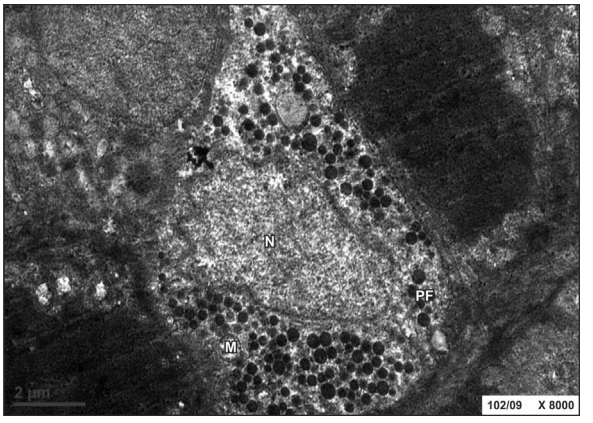

Fig. 4. Electron micrograph of parafollicular cell [PF] during estrus showing large indented nucleus [N]. Secretory granules [SG] are distributed in the cytoplasm. Mitochondria [M] are few with collapsed cristae. Other cell organelles are indistinct. 8000×.

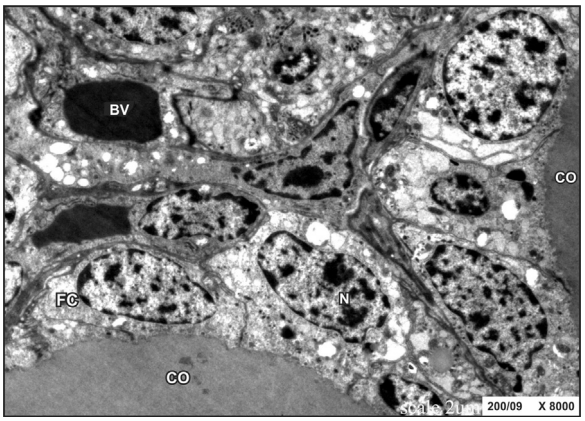

Fig. 5. Electron micrograph of the thyroid gland of female bat during early pregnancy showing active follicle lined by low columnar cells. Lumina of follicle are filled with electron dense colloid material [CO]. 8000×.

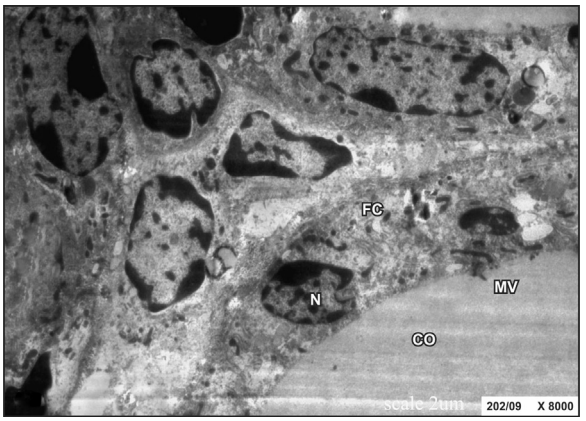

Fig. 7. Electron micrograph of the thyroid gland of female bat during early pregnancy showing Follicular cell [FC] with oval shaped nucleus [N]. 8000×.

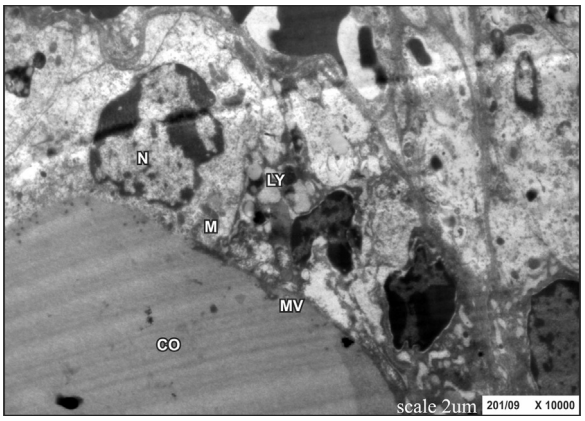

Fig. 6. Electron micrograph of the thyroid gland of female bat during early pregnancy showing the nucleus [N] with chromatin clumps and electron dense colloid material in the lumen of follicle. Note the presence of lysosome [LY] and oval to elongated mitochondria [M]. Note the presence of microvilli [MV] protrude in to the colloid. 10,000×.

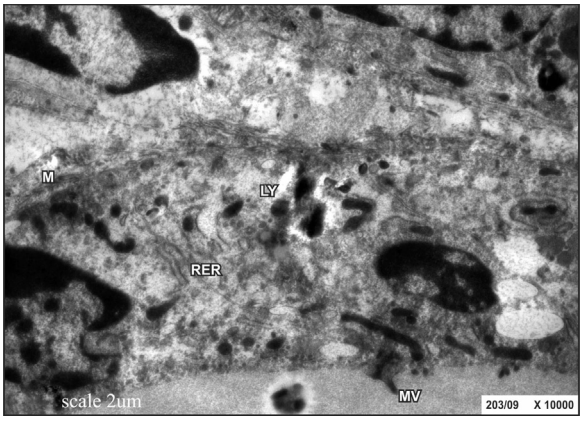

Fig. 8. Magnified view of the follicular cell [FC]. Note the presence of several dilated cisternae of rough endoplasmic reticulum [RER], mitochondria [M] with lamellar cristae, dense bodies [DB] and lipid droplet [LD]. 10,000×.

specimens collected were pregnant. The thyroid gland shows medium to large size follicles but the medium size follicles are more than large follicles.

The medium size follicles are lined by cuboidal epithelium and lumina is filled with homogenous colloid material. Nucleus is spherical with slight indentation (Fig. 5). Rough endoplasmic reticulums are in the form of tubules. Mitochondria are elongated in shape. Lysosomal bodies are seen in the cytoplasm engulfing the lipid droplets (Fig. 6). Microvilli are seen towards the lumen. The nucleus is oval to irregular in shape with chromatin material at periphery. The microvilli are small and few in number (Fig. 7). The Golgi body is inconspicuous. The rough endoplasmic reticulums are much reduced. Numerous free ribosomes are seen in the cytoplasm (Fig. 7). Few rod shape mitochondria are observed in the cytoplasm. The lysosomes are increase and associated with colloid droplets (Fig. 8).

3.1.4. Parafollicular cells

The parafollicular cell of the pregnant bat shows the very specialized structure. They are seen in a group of two to three cells (Fig. 9). The nucleus is large with heterochromatin material. Golgi body is seen juxta nuclear in position. The mitochondria are spherical with lamellar cristae. Large, spherical, electron dense secretary granules of varying diameter are scattered throughout the cytoplasm. Other cell organelles are inconspicuous (Fig. 10).

3.1.5. Thyroid gland during late pregnancy

Bats collected in the month of May shows late pregnancy. Nucleus is seen with well developed nucleoli and chromatin flakes are less as compare to early pregnancy. The lumen of the follicle is filled with homogenous colloid material. Mitochondria are less in number. Few colloid droplets are found throughout the cytoplasm (Fig. 11). Lysosomal bodies are seen near the colloid droplets. The follicular cells of the large follicle are synthetically inactive during late pregnancy.

3.1.6. Parafollicular cells

Parafollicular cell at this stage of pregnancy shows very specialized structure. The nucleus is large irregular with light chromatin material. The rough endoplasmic

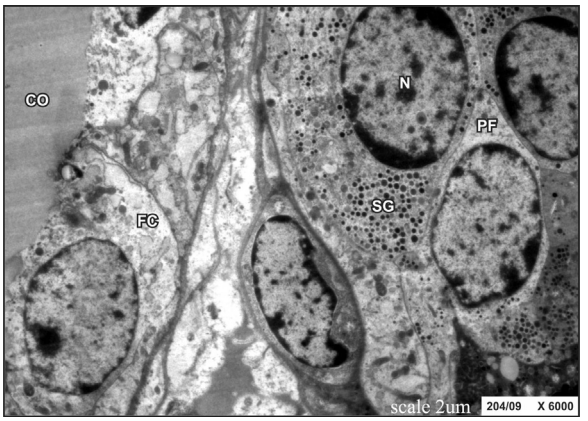

Fig. 9. Electron micrograph of the parafollicular cell of the thyroid gland during early pregnancy. Note the group of parafollicular cells filled with secretory granules. 6000×.

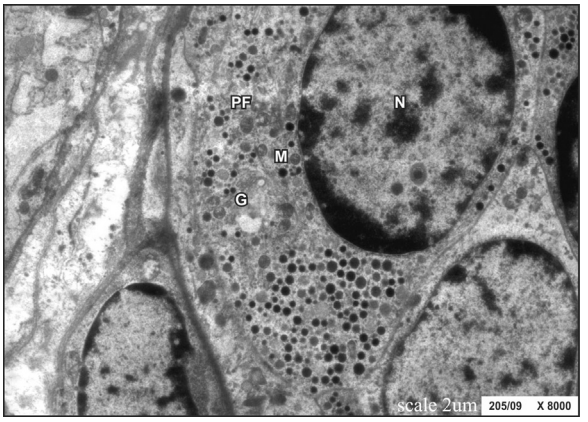

Fig. 10. Electron micrograph of the parafollicular cell of the thyroid gland during early pregnancy. Note the presence of spherical nucleus [N] with well developed nucleoli [NO], spherical mitochondria [M] with lamellar cristae and Golgi apparatus [G]. 8000×.

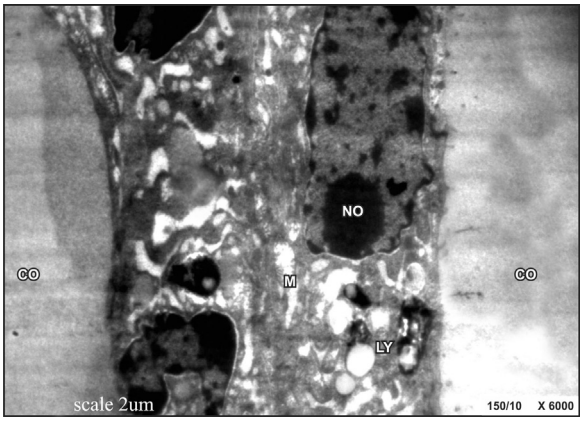

Fig. 11. Electron micrograph of the thyroid gland during late pregnancy showing follicle lined by squamous to cuboidal epithelium. Follicular lumina is filled with homogenous colloid material [CO]. Note the presence of elongated nucleus [N] with irregular outline and well developed nucleoli [NO]. Note the presences of lysosomal body [LY], mitochondria [M] with collapsed cristae. 6000×.

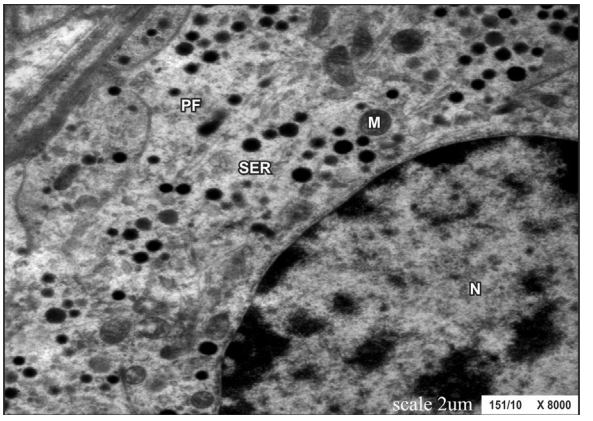

Fig. 12. Electron micrograph of the parafollicular cell [PF] of the thyroid gland during late pregnancy. Note the presence of large number of secretory granules [SG]. Mitochondria [M] are rod to circular shaped with lamellar cristae. 8000×.

reticulum is in the form of small dilated cisternae. Mitochondria are seen scattered in the cytoplasm with lamellar cristae while some mitochondria are with collapsed cristae. The large secretary granules are seen scattered in the cytoplasm (Fig. 12).

3.1.7. Thyroid gland during lactation

Bats collected in the month of June shows lactation. The thyroid gland consists of many small size follicles. The active thyroid follicles are lined with low cuboidal epithelium (Fig. 13). Nucleus is large, spherical with few chromatin material and nucleolus is well developed. Lumen is filled with colloid material. Blood vessels are seen. Rough endoplasmic reticulums with dilated vesicle are seen throughout the cytoplasm. Numerous free ribosomes are scattered throughout the cytoplasm. Round to elongate mitochondria are seen in the cytoplasm of the cell. Microvilli are observed directed towards the lumen. Lysosomal body is observed in the cytoplasm and more in number (Fig. 14). The Golgi body is present in the apical part of cytoplasm of the cell.

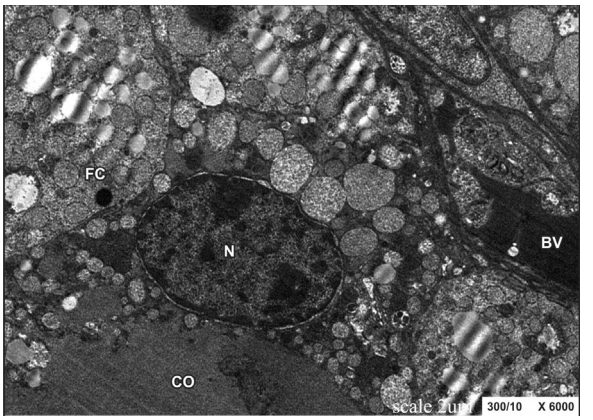

Fig. 13. Electron micrograph of the thyroid follicle during lactation. The active follicles are lined by low columnar epithelium cell with electron dense colloid material [CO]. Note the blood vessel [BV] in the interfollicular area. 6000×.

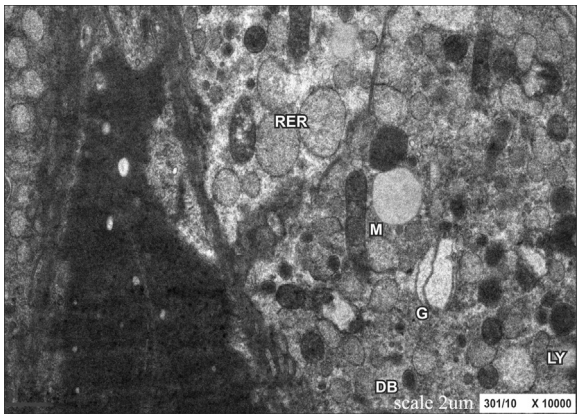

Fig. 14. Electron micrograph of the follicular cell during lactation. Note the presence of several ovoid to elongated mitochondria [M] with lamellar cristae, lysosomes [LY] and dense bodies [DB]. Note the presence of hypertrophied Golgi body [G], dilated round profile of rough endoplasmic reticulum [RER]. Blood vessel [BV] is seen in the intrafollicular space. 10,000×.

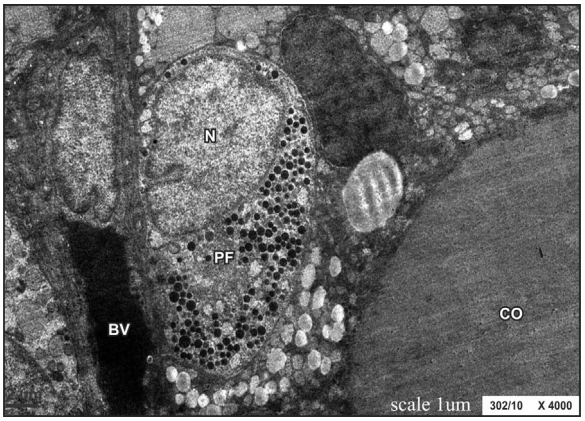

Fig. 15. Electron micrograph of the parafollicular cell [PF] during lactation showing indented nucleus [N]. Blood vessel is seen near theparafollicular cell. 4000×.

3.1.8. *Parafollicular cells*

The parafollicular cell is well developed during lactation as compared to the pregnancy. The secretary granules are seen scattered towards one pole of the cytoplasm and are uniform in diameter. The secretory granules are moderately dense (Fig. 15). The nucleus is irregular with slight indentation. Intercellular junction between parafollicular and follicular cell are prominent. Cisternae of rough endoplasmic reticulum are observed in the cytoplasm. Free ribosomes are distributed throughout the cytoplasm. Mitochondria are seen with collapsed cristae. Other cell organelles are not conspicuous (Fig. 16).

4. Discussion

The thyroid gland has long been recognized as an important modulator of reproductive function. Both hyper- and hypo-thyroidism are associated with reproductive dysfunction and infertility. Direct effects of thyroid hormones on cellular metabolism and hormone responsiveness have been demonstrated in the gonads and the sex accessory organs. In addition, thyroid hormones appear to play a key role in the expression of events that underlie seasonal reproductive cycle.

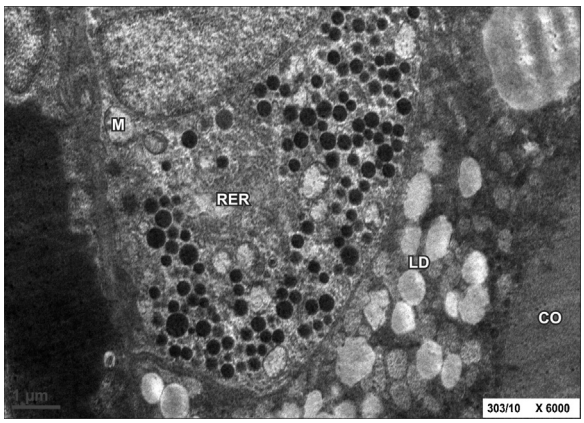

Fig. 16. Electron micrograph of the parafollicular cell during lactation showing large secretory granules [SG], dilated cisternae of rough endoplasmic reticulum [RER] and mitochondria [M] with collapsed cristae. 6000×.

Thyroid is an endocrine gland that secretes hormones including thyroglobulin, triiodothyronine and thyroxin. Thyroxin hormone secreted by this gland plays an important role in metabolism of the body [18]. In *Taphozous kachhensis*, the location of thyroid gland in the body is similar to other bats like *Hipposideros lankadiva* [13], *Taphozous longimanus* [7], i.e., with the first ring of trachea and consisted of two lobes on both side and an isthmus connecting these lobes. In *Grass cutter* the thyroid gland shows that it is a bi-lobed organ located on the lateral surface of the trachea between the first and third tracheal rings. The two lobes were totally separated, without an isthmus [5].

In *Taphozous kachhensis*, the gland appeared reddish brown in colour which is in concordance with the findings of *Megaderma lyra lyra* [17]. In *Taphozous kachhensis*, the thyroid follicles, colloid and the epithelial cell height show significant variation during the reproductive cycle. During estrus thyroid gland shows heterogeneous population of follicles. It is mainly composed of medium size follicles and few large and small follicles.

The medium size follicles are lined by cuboidal epithelium cells and are synthetically active follicles. The small size follicles are lined by low columnar epithelium and synthetically most active follicles. The large size follicles are lined by low cuboidal to squamous epithelium and are synthetically inactive follicles.

In *Hipposideros lankadiva* [13], thyroid gland is active during estrus similar to that are reported in *Taphozous kachhensis*. During early pregnancy mainly large follicles are seen indicating hypothyroid condition. Similar hypothyroid condition is also observed in *Macrotus californicus* [2] and *Taphozous longimanus* [7,16]. In present study the thyroid gland is active during early part of pregnancy. In *Hipposideros lankadiva*, after arousal from torpor, thyroid gland become active and remains till late pregnancy [13]. In *Taphozous kachhensis*, thyroid activity decreases at the

end of pregnancy as indicated by large follicle in the thyroid gland of the pregnant bat.

The follicular epithelium is low cuboidal to squamous during sexually quiescence period indicating low synthetic activity while, the follicular epithelium is columnar during sexually active period indicating increased synthetic activity [6,7,9,10], supporting the present observation.

The histological sections also revealed that the shape and size of the follicles are not homogenous; since large follicles were usually found in the peripheral and smaller ones were located centrally. This is in agreement with the report of Hartoft-Nielsen et al. [4]. The observation in the *Taphozous kachhensis* is in contrast to the arrangement reported by Sanap et al. [11] in cattle and Baishya et al. [1] in Assam barbari goats.

The thyroid gland during estrus shows the small size follicles which are lined by high cuboidal epithelium with spherical nuclei. The chromatin material is attached to the inner surface of the nuclear membrane. Two or three nucleoli were observed within the granular nucleoplasm. Nuclear pores are observed. Elongated tubules of rough endoplasmic reticulum are frequently observed in the cytoplasm of the follicular cell and are heavily dotted with ribosomes. Some dilated tubules of rough endoplasmic reticulum are also observed in the cytoplasm having close association with mitochondria. The lumen of follicle is filled with homogenous colloid material and microvilli are numerous. Golgi complex is inconspicuous. Free ribosomes are also scattered throughout the cytoplasm.

Mitochondria are abundant and are evenly distributed in the cytoplasm. Mitochondria are mostly spherical but few are elongated in shape with lamellar cristae and having electron dense mitochondrial matrix. Some dense spherical lysosomal bodies of variable size are seen in the cytoplasm. Large size follicles are lined by squamous epithelium with oval nucleus. The number of mitochondria and rough endoplasmic reticulum are reduced.

The thyroid gland during pregnancy shows medium to large size follicles but the medium size follicles are more than large follicles. The medium size follicles are lined by cuboidal epithelium and lumina is filled with homogenous colloid material. Nucleus is spherical with slight indentation. Rough endoplasmic reticulums are tubular and are dotted with ribosomes. Lumen of the follicle contains homogenous colloid material. Mitochondria are elongated in shape. Lysosomal bodies are seen in the cytoplasm. Microvilli are seen towards the lumen.

Large follicles are lined by squamous epithelium. The nucleus is oval to irregular in shape with some chromatin material at periphery. The microvilli are small and few in number. The Golgi body is inconspicuous. The rough endoplasmic reticulums are much reduced. Numerous free ribosomes and few rod shaped mitochondria are observed in the cytoplasm.

During late pregnancy, the thyroid follicles are lined by squamous epithelium cell. Nucleus is seen with well developed nucleoli and chromatin flakes are less as compare to early pregnancy. The lumen of the follicle is filled with homogenous colloid material.

Mitochondria are less in number. Few colloid droplets are found throughout the cytoplasm. Lysosomal bodies are seen near the colloid droplets. The follicular cells of the large follicles are synthetically inactive during late pregnancy.

During lactation the thyroid gland consists of many small size follicles. The active thyroid follicles are lined with low cuboidal epithelium. Nucleus is large spherical with few chromatin material and nucleolus is well developed. Lumen is filled with colloid material. Blood vessels are seen [3].

Rough endoplasmic reticulum is in the form of small to medium size polymorphic and dilated vesicles seen throughout the cytoplasm. Numerous free ribosomes are scattered throughout the cytoplasm. Round to elongated mitochondria are seen in the cytoplasm of the cell. Microvilli are observed and they are directed towards the lumen. Lysosomes are observed in the cytoplasm and more in number. The Golgi is present in the apical part of the cytoplasm. The Golgi complex is greater in complexity and size. The ultrastructural features of follicular cells indicate that these cells actively elaborate the thyroid hormone during lactation. Similar observation was recorded in *Taphozous longimanus* [16], *Hipposideros lankadiva* [13].

In *Taphozous longimanus* [7], the thyroid follicle are lined by cuboidal epithelium during estrus and filled with colloid. Mitochondria are seen with lamellar cristae. They are closely opposed to the element of the rough endoplasmic reticulum. The cells show many pleomorphic profiles of rough endoplasmic reticulum. During pregnancy thyroid composed of small follicles lined by low columnar epithelium. Golgi body is well developed. Mitochondria are oval to rod shaped with lamellar cristae. Rough endoplasmic reticulum are dilated A large lipid droplets are seen.

In *Megaderma lyra lyra* [17], the follicles in the thyroid gland are of variable size and shapes during different phases of reproductive periods. During estrus small follicles were numerous with cuboidal epithelium. Medium size follicles were less in number with low cuboidal epithelium. During pregnancy the large follicles are increase in number and the lumina is filled with little amount of colloid. During lactation small follicles are increases in number with low columnar cell. The ultrastructural changes in the follicular cell of *Megaderma lyra lyra* are similar to present observation in the thyroid follicular cell of *Taphozous kachhensis*.

In thyroid gland of dolphin [15], the rough endoplasmic reticulum was seen adjacently to mitochondria at the basal and lateral regions of follicular cells. Golgi complex, multivesicular bodies, granules with various size and electron density are observed in the apical regions of cells. Microvilli are poorly developed at the apical surface of the cells.

The ultrastructure of follicular cell of estrus bat, *Hipposideros lankadiva* [13] showed the presence of dilated rough endoplasmic reticulum, well developed Golgi apparatus and mitochondria with lamellar cristae and apical blebs towards the luminal plasma membrane. During embryonic diapause, thyroid follicles are lined by flattened epithelium. Cytoplasm is vacuolated. Cell organelles are not well developed, suggesting a reserve or storage state of the cells and maternal hypothyroidism. A bat arouse from torpor thyroid gland shows medium size follicle and few large follicle. The lumina of follicle are filled with colloid material. The cell organelles are well developed

suggesting a synthesis of thyroid hormone during late pregnancy and lactation. The ultrastructural changes in the follicle of bat *Hipposideros lankadiva* [13] are similar to those observed in the thyroid follicular cell in this species of bat during reproductive cycle except during embryonic diapause.

4.1. Parafollicular cells

In *Taphozous kachhensis*, the parafollicular cells are observed at the base of the follicular epithelium between the follicular cells and are filled with electron dense Secretory granules. These are elongated to oval in shape and are electron lucent and larger than follicular cell. The nucleus is large and is irregular in shape with indented margin and occupies the large portion of the cytoplasm. Nucleolus is clearly visible. Chromatin clumps are seen along with the nuclear membrane. Rough endoplasmic reticulum is occurs in short tubular forms. Mitochondria are scattered throughout the cytoplasm.

In *Taphozous longimanus* [8], the parafollicular cells are present at the base of the follicular epithelium between the follicular cells. The cells are elongated, irregular or oval and are abetting to the inter follicular area. Similar observations were noted in the bats, *Myotis* and *Nyctula* [9].

In *Taphozous kachhensis*, the parafollicular cell indicates features of high activity during the active phase. They have several dilated profiles of rough endoplasmic reticulum dotted with ribosomes. Mitochondria possess lamellar cristae. Several dense cytoplasmic granules of varying shapes and sizes are observed in the cytoplasm. All these features suggest that the cell is in active synthetic state during the different phases of the reproductive cycle. This is differing from the observation in *Taphozous longimanus* [8], in which the morphological characteristic of parafollicular cells during estrus, pregnancy and lactation is suggestive of low synthetic activity and storage of granules during the reproductive cycle.

In *Hipposideros lankadiva* [13], the parafollicular cell indicates feature of high activity during the active phase. They have several dilated profiles of rough endoplasmic reticulum dotted with ribosomes. Mitochondria possess lamellar cristae. Several dense cytoplasmic granules of varying shapes and sizes are observed in the cytoplasm. All these features suggest that the cell is in active synthetic state during the different phases of the reproductive cycle.

The thyroid gland in active bats, *Myotis* and *Nyctula* [9], have large number of granule-containing parafollicular cells which undergo alterations in relation to seasonal changes in the physiological state of the bat. Some of the parafollicular cells of bats caught in late summer contain small dense secretory granules packed in the Golgi apparatus, while the large granules are found enclosed within a ribosome studded membrane which may increase by direct secretion of newly synthesized material at their surface [9]. During early hibernation, the large dense secretory granules are absent, while cells containing small granules are either partly or totally degranulated. Similar observations have been reported for parafollicular cells of estrus and pregnant bats of *Taphozous longimanus*. Nunez [9,10] suggest that the two types of granules present in bat thyroid parafollicular cells have different fates during the annual cycle of physiological activity.

In *Taphozous longimanus* [8], parafollicular cells during estrus are elongated or oval. They are electron-lucent and larger than the follicular cells. The nucleus is large and irregular in shape with indented margin and occupies a large portion of the cytoplasm. The nucleus contains finely condensed chromatin material, and chromatin clumps are seen along the nuclear envelope. Nuclear pores are seen. Rough surfaced endoplasmic reticulum occurs as short tubular segments. Golgi apparatus is inconspicuous. Mitochondria are sparse and scattered throughout the cytoplasm. Some membrane-enclosed areas show a light flocculent material. They are of the same size as the dense granules or larger. Some dense granules are partly or totally degranulated. Small desmosomes and intercellular junctions are observed between the parafollicular cells and follicular cells.

In *Megaderma lyra lyra* [17], it is reported that the parafollicular cells during pregnancy shows abundant secretory granules near nucleus and also in cytoplasm. The endoplasmic reticulum is dilated and other cell organelles are scanty. During lactation parafollicular cell were more developed than pregnancy. The rough endoplasmic reticulum is tubular and dotted with ribosomes, Golgi complex is inconspicuous and mitochondria with lamellar cristae. Secretory granules are abundant of high electron density. The present observations are in agreement with these findings.

5. Conclusion

The thyroid gland is more active during estrus than the pregnancy and lactation. The cell organelles observed in follicular cell of thyroid gland indicate more synthetic activity to secrete more thyroglobulin which is stored in the lumina of follicle. While during pregnancy and lactation the thyroid gland is not synthetically active but the appearance of more lysosomal bodies during pregnancy indicate the utilization of stored colloid and release of thyroid hormone in blood stream. During pregnancy and lactation T3 level is high and this hormone is responsible for growth of embryo during pregnancy. The parafollicular cells are more developed during pregnancy and lactation than estrus as indicated by ultrastructural characteristics. These cells may be secrete calcitonin, serotonin, and somatostatin and may responsible for controlling the seasonal changes in plasma calcium concentration in this bat.

Conflict of interest

The authors declare that there is no conflict of interest that could be perceived as prejudicing the impartiality of the review.

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