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Histochemical studies on thyroid and parathyroid glands of pig *(Sus scrofa)*[#]

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Abstract

The present histochemical study on thyroid and parathyroid glands was conducted on six Large White Yorkshire pigs. Histochemically, the lining epithelium of follicles of the thyroid gland, along with the basement membrane and thyrocolloid, showed intense positive reaction for PAS, while the capsule showed mild reaction for PAS-AB. The follicular cells and parafollicular cells showed mild positive activity,and the thyrocolloid showed intense positive activity for protein. The calcium was deposited in the cytoplasm of parafollicular cells, thyrocolloid and interfollicular connective tissue. Intense acid phosphatase activity was noticed in the blood vessels and interstitial connective tissue of the capsule and basement membrane of the follicular lining epithelium. Intense alkaline phosphatase activity was noticed in blood vessels of interstitial connective tissue and the basement membrane of the follicular lining epithelium. The fat deposition was noticed in the capsule, trabeculae and interlobular connective tissue. In the parathyroid gland, the colloid substance of mixed follicles showed an intense positive reaction for PAS. The secretory granules of chief cells were positive for PAS and PAS-AB proteins.

Keywords: *Thyroid gland, parathyroid gland, follicular cells, PAS, PAS-AB, domestic pig.*

The pig is considered an excellent biomodel for humans due to its similarity in physiology, organ development, and disease progression to humans (Lunney, 2007). The thyroid gland plays an important role in normal reproduction and productive performance, such as growth, milk and hair fibre production of domestic animals (Tondini, 2007). The thyroid gland is the largest and first recognisable endocrine gland in the development of vertebrates (Hossam *et al*., 2012). Microscopically, follicles are the fundamental units of the thyroid gland,and the follicular lining cells synthesise and secrete the triiodothyronine (T3) and thyroxine (tetraiodothyronine, T4) hormones, which are essential in normal metabolism and homeostasis (Huszenica*et al.,* 2002;Capen and Martin, 2003). In domestic animals, triiodothyronine and thyroxin hormones exert apermissive effect upon the anabolic and metabolic effects on the growth hormone and increases synthesis in pituitary gland (Medrano and Hua,2016).

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Pig is a suitable model to study the holistic gene expression and physiological aspects of the parathyroid gland due to its nutrient requirements, pathophysiology and the functional genomics, which are similar to those of human (Oster *et al.,* 2018). The chief cells of parathyroid gland secrete the parathyroid hormone. The main role of the hormone is to maintain the normal levels of calcium and phosphorus in the blood by increasing the absorption of calcium from the intestine and decreasing the loss of calcium from the urine and resorption of calcium from the bone (Hussain and Al – Taay, 2009).The literature available on the structural details of thyroid and parathyroid glands especially in pig is very scanty. Hence the present study is undertaken to elucidate the histochemical features of thyroid and parathyroid glands of pig.

Materials and methods

For the histochemical studies, the specimens of thyroid and parathyroid glands were collected after slaughter from six Large White Yorkhire pigs in AICRP on pigs. College of Veterinary Science, Tirupati. Fresh tissue samples of 3-5mm thickness were collected from two lateral lobes and pyramidal lobe of thyroid and parathyroid glands of adult pigs and fixed in 10% neutral buffered formalin (NBF). These tissue samples were subjected to routine tissue processing techniques and paraffin sections of about 5-6µm thickness were obtained with the help of Leica Semi-automatic microtome and these sections were subjected to periodic acid -Schiff (PAS) method for mucopolysaccharides, periodic acid-Schiff-alcian (PAS-AB) blue method for acid mucopolysaccharides, Von Kossa's method for calcium (Singh and Sulochana, 1997) Mercury - Bromophenol blue method for proteins (Luna, 1968), Special stain for thyrocolloid and iodine (Desmaris and Laham, 1962). The stained slides were examined under microscope to study the normal histochemical characters of the thyroid and parathyroid glands.

Fresh tissue samples of porcine thyroid gland were used for frozen sections. Sections of 10 µm thickness were obtained using a cryostat and subjected to Gomori's methods for alkaline and acid phosphatase activity and the Oil red 'O' in propylene glycol method for demonstrating fats (Singh and Sulochana, 1997). The stained slides were observed under the light microscope to study and compare the histochemical observations in thyroid gland of pigs.

Results and discussion

Histochemical observations of thyroid gland

Carbohydrate

In the present study, the capsule and interlobular connective tissue of thyroid gland showed mild positive reaction for Periodic acid-Schiff (PAS) (Fig.1), which concurs with the findings of Rajalakshmi *et al.* (2019) in sheep. The PAS positivity is indicative of the presence of neutral mucopolysaccharides i.e. carbohydrates and glycogen. The follicular lining epithelial cells showed intense positive reaction for PAS (Fig.2), which is in agreement with the findings in sheep reported by Rajalakshmi *et al.* (2019) and Iraqian goat by Nadhim (2017). But Royand Saigal (1987) reported that in sheep,follicular epithelial cells showed no reaction for PAS. Thyrocolloid in the lumen of the follicles showed intense positive reaction for PAS (Fig.2), as noted in humans(Ham and Comarck, 1972; Bloom and Fawcett, 1978 and Harach, 1987), in buffalo by Roy and Yadava (1977), in domestic animals by Dellmann and Brown(1987), in sheep and gazelle by Khaleel and Mohammad Salih (2017) and Iraqian goat by Nadhim (2017). The parafollicular cells and basement membrane of thyroid follicles in the present study showed intense positive reaction for PAS (Fig.2).

In the present study, the follicular lining epithelial cells showed moderate positive reaction for Periodic

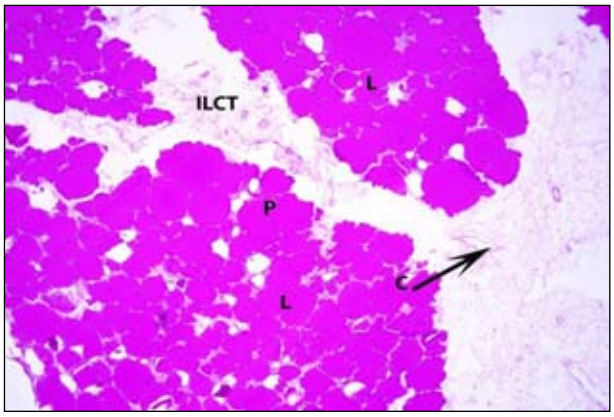

Fig. 1. Photomicrograph showing mild positive reaction for PAS in capsule (C) and interlobular connective tissue (ILCT) of thyroid gland in pig. -P- Parenchyma, L- Lobules.
Periodic Acid Schiff method X 40

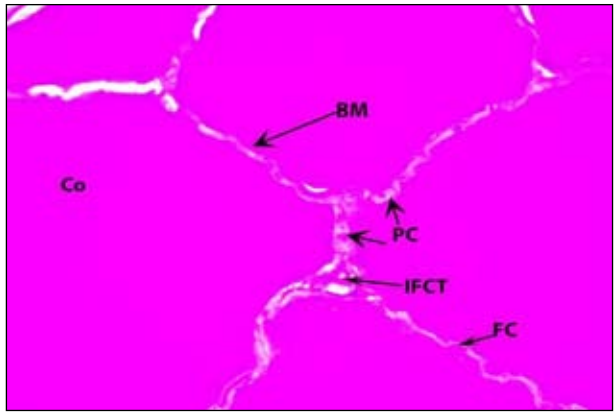

Fig. 2. Photomicrograph showing intense positive reaction for PAS in the follicular cells (FC), basement membrane (BM) and parafollicular cells (PC) of the thyroid gland in pig. Co- Colloid, IFCT-Interfollicular connective tissue.
Periodic Acid Schiff method X 400

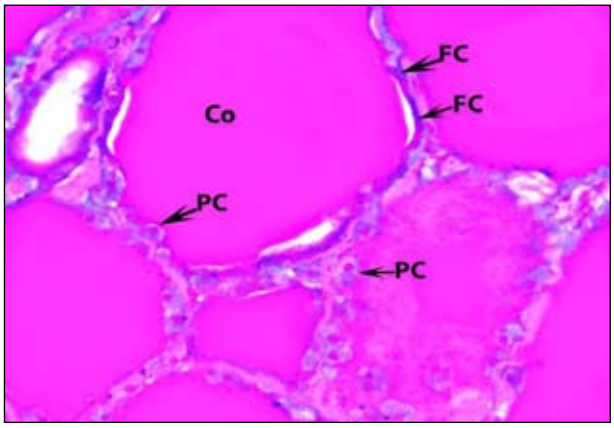

Fig. 3. Photomicrograph showing moderate positive reaction for PAS-AB in follicular cells (FC) and parafollicular cells (PC) of the thyroid gland in pig. Co - Colloid

Periodic Acid Schiff – Alcian blue method X 400

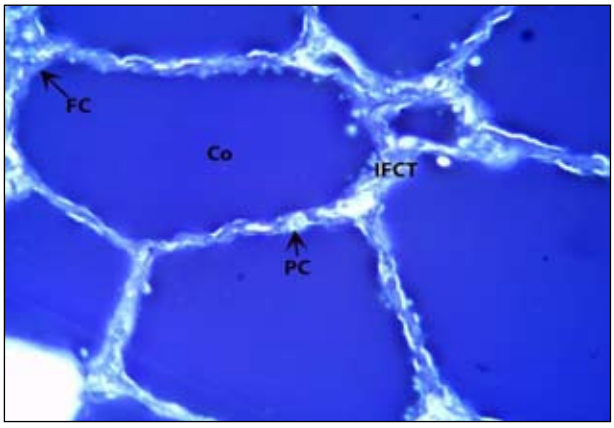

Fig. 4. Photomicrograph showing mild positive activity for proteins in follicular cells (FC), parafollicular cells (PC) and interfollicular connective tissue (IFCT) and intense positive reaction for proteins in thyrocolloid of the thyroid gland in pig. Co – Colloid.

Mercury Bromophenol Blue method X 400

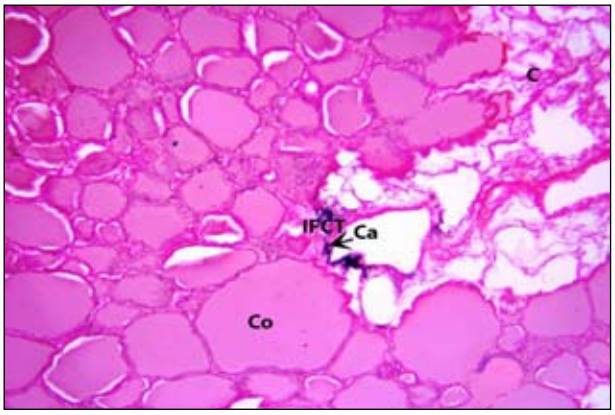

Fig. 5. Photomicrograph showing presence of calcium deposit (Ca) in the capsule (C) of the thyroid gland in pig. Co-colloid, IFCT-Interfollicular connective tissue.

Vankossa's method X 100

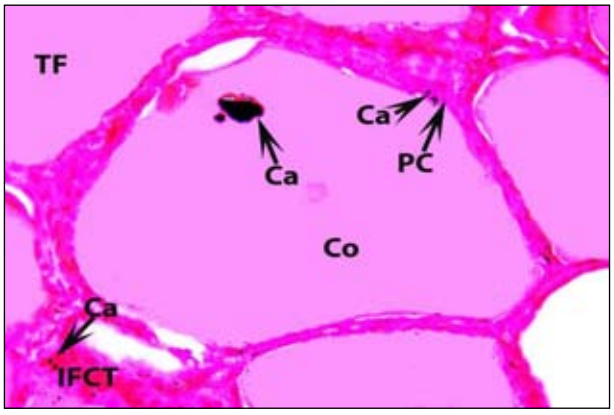

Fig. 6. Photomicrograph showing calcium deposits (Ca) in parafollicular cells (PC), colloid substance (Co) and interfollicular connective tissue (IFCT) of the thyroid gland in pig. TF- Thyroid follicle.

Vankossa's method X 400

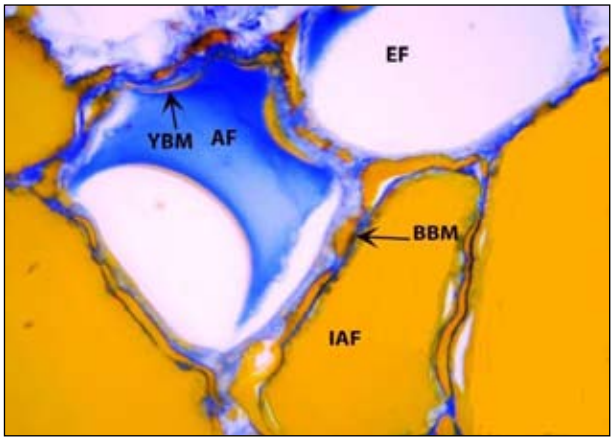

Fig. 7. Photomicrograph showing inactive follicles (IAF) and active follicles (AF) of the thyroid gland in pig. YBM- Yellow basement membrane, BBM- Blue basement membrane, EF- Empty Follicle.

Desmaris and Laham method X 400

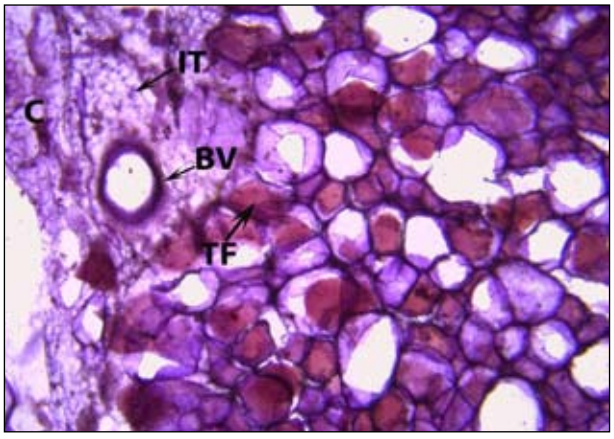

Fig. 8. Photomicrograph showing intense positive activity in blood vessel (BV) and interstitial connective tissue (IT) of capsule (C) for acid phosphatase of the thyroid gland in pig. TF- Thyroid Follicle.

Gomori's method for acid phosphatase X 40

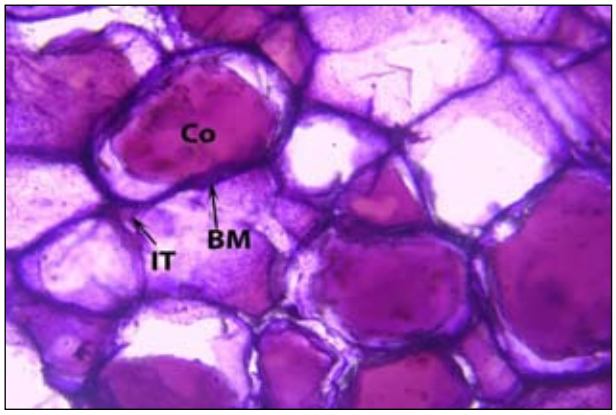

Fig. 9. Photomicrograph showing intense positive activity in basement membrane (BM) of follicular lining epithelium, moderate positive activity in colloid substance (Co) and interstitial connective tissue (IT) for acid phosphatase of the thyroid gland in pig.

Gomori's method for acid phosphatase X 400

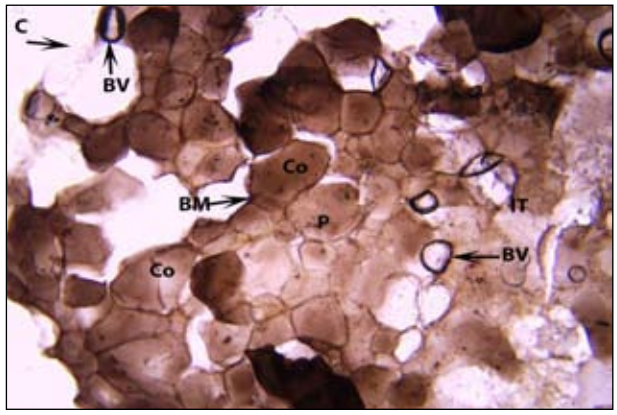

Fig.10. Photomicrograph showing intense positive activity in the blood vessels (BV) of capsule (C) and parenchyma (P), mild to moderate positive activity in colloid (Co) and basement membrane (BM) of follicular lining epithelium for alkaline phosphatase of the thyroid gland in pig.

Gomori's method for alkaline phosphatase X 40

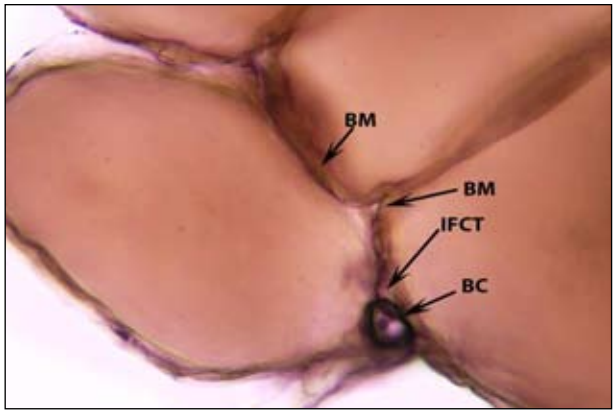

Fig.11. Photomicrograph showing intense positive activity for blood capillaries (BC) in interfollicular connective tissue (IFCT) and mild to moderate activity in basement membrane (BM) of follicular lining epithelium for alkaline phosphatase of the thyroid gland in pig.

Gomori's method for alkaline phosphatase X 400

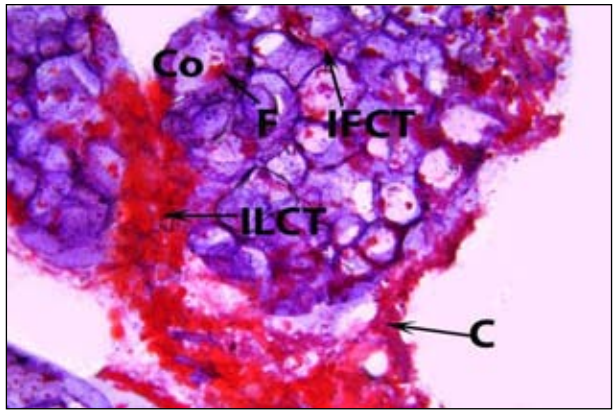

Fig.12. Photomicrograph showing fat deposition (F) in capsule (C), interlobular connective tissue (ILCT), interfollicular connective tissue (IFCT) and colloid substance (Co) of the thyroid gland in pig.

Oil Red 'O' X 40

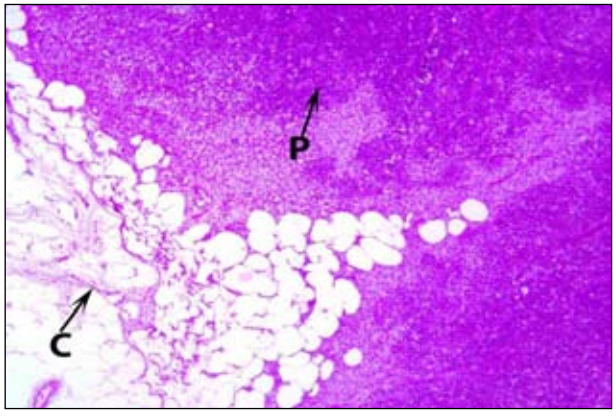

Fig.13. Photomicrograph showing mild positive reaction for PAS in the capsule (C) of the parathyroid gland in pig. P- Parenchyma.

Periodic Acid Schiff method X 40

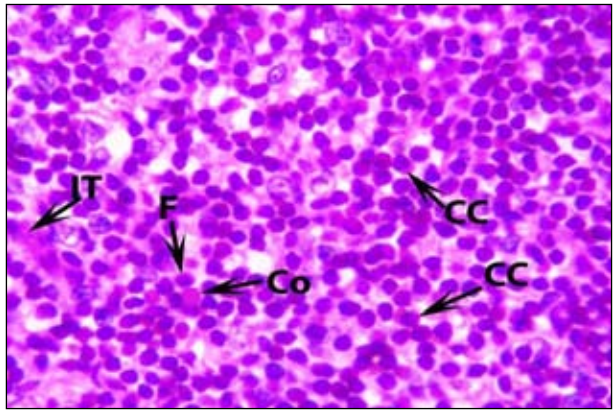

Fig.14. Photomicrograph showing positive reaction for PAS in chief cells (CC), colloid (Co) of follicle (F) and interstitial connective tissue (IT) of the parathyroid gland in pig.

Periodic Acid Schiff method X 1000

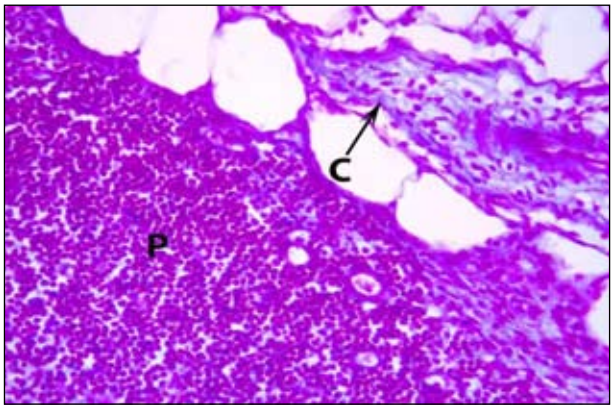

Fig.15. Photomicrograph showing mild positive reaction for PAS-AB in capsule (C) of the parathyroid gland in pig. P- Parenchyma.

Periodic Acid Schiff – Alcian blue method X 40

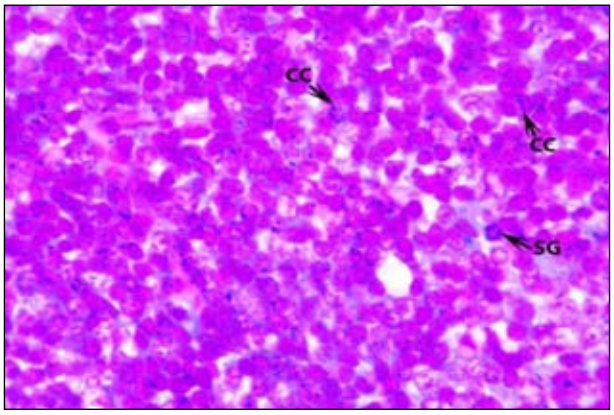

Fig.16. Photomicrograph showing positive for PAS-AB in secretory granules (SG) of chief cells (CC) of the parathyroid gland in pig.

Periodic Acid Schiff – Alcian blue method X 1000

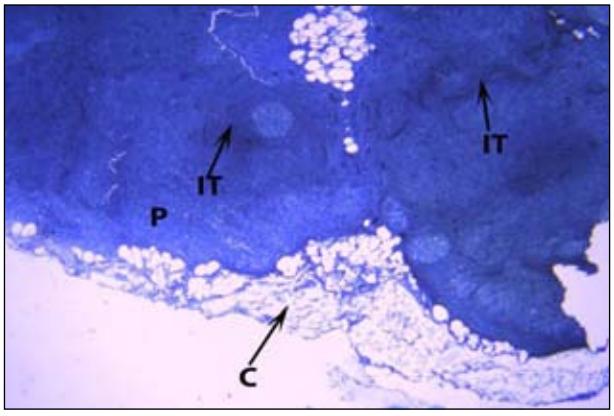

Fig.17. Photomicrograph showing mild positive activity for proteins in the capsule (C) and intense activity in interstitial connective tissue (IT) and parenchyma (P) of the parathyroid gland in pig. P- Parenchyma.

Mercury Bromophenol Blue method X 40

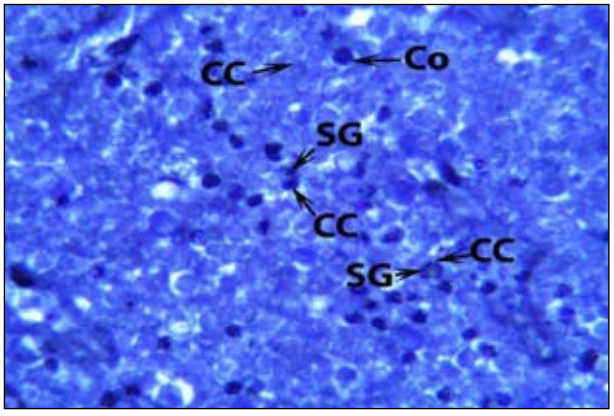

Fig.18. Photomicrograph showing positive activity for proteins in secretory granules (SG) of chief cells (CC) of the parathyroid gland in pig. Co- Colloid.

Mercury Bromophenol Blue method X 1000

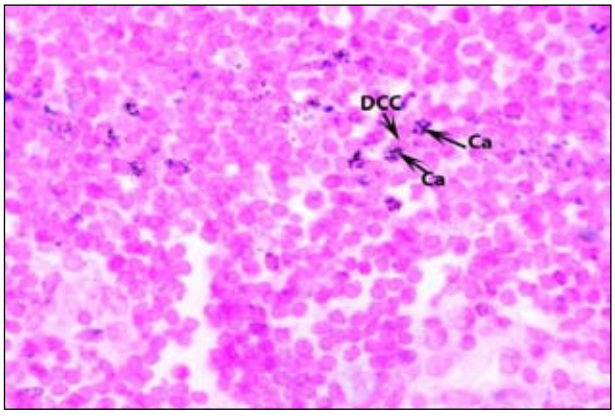

Fig.19. Photomicrograph showing calcium deposits (Ca) in the dark chief cells (DCC) of the parathyroid gland in pig.

Vankossa's method X 1000

acid-Schiff-alcian blue(PAS-AB), which concurs with the findings of Roy and Saigal (1987) in sheep. The PAS-AB activity is indicative of the presence of acid mucopolysaccharides. The parafollicular cells showed a moderate positive reaction for PAS-AB. Contrary to these findings, Roy and Saigal (1987) reported that there is no reaction in parafollicular cells for PAS-AB in sheep. The thyrocolloid showed no reaction for PAS-AB in this study (Fig.3).

Proteins

In the present study, the capsule showed mild positive activity for proteins. The follicular lining cells, parafollicular cells and interfollicular connective tissue showed mild positive and thyrocolloid showed intense positive activity for proteins (Fig.4). The presence of proteins is indicative of the presence of glycoprotein, which was similar to the findings of Roy (1971) in buffalo.

Calcium

In the present study, the calcium deposits were present in the capsule (Fig.4), cytoplasm of parafollicular cells, thyrocolloid substance and interfollicular connective tissue (Figs.5,6), which is agreed partly with the findings reported by Sanjeev and Rakesh (2016) in goat.

Iodine

In the present study, the inactive thyroid follicles showed yellow coloured colloidal substance and blue coloured basement membrane, whereas the active thyroid follicles showed blue coloured colloidal substance and yellow coloured basement membrane (Fig.7). These findings were similar to the observations reported in buffalo by Roy (1971). Blue colour indicates positive reaction for iodine and yellow colour indicates negative reaction for iodine.

Acid phosphatase activity

In the present study, intense positive activity for acid phosphatase was noticed in the basement membrane of the lining epithelium of thyroid follicles, whereas thyrocolloid of thyroid follicles showed mild to moderate activity for acid phosphatase enzyme (Figs.8,9). In contrast to these findings, Roy and Saigal (1986) noted absence of acid phosphatase enzyme activity in thyrocolloid of thyroid gland of sheep.

Alkaline phosphatase activity

The blood vessels present in the interstitial connective tissue showed intense positive activity for alkaline phosphatase, which concurs with the findings of Roy and Saigal (1987) in sheep and (Greep, 1966) in human. In the present study mild to moderate activity was recorded in thyrocolloid and basement membrane of follicular lining epithelium. Mild activity was also noticed in the capsule (Fig.10). However, there was an intense activity of alkaline phosphatase in the blood capillaries of interfollicular connective tissue (Fig.11).

Fat

In the present study, large amount of fat deposition was observed in the capsule and interlobular connective tissue of thyroid gland, thyrocolloid of the follicles and interfollicular connective tissue in between the follicles (Fig.12). Similarly, Rajalakshmi *et al.* (2019), also noticed the accumulation of fat in capsule, interstitial connective tissue, thyrocolloid and interfollicular connective tissue of thyroid gland in sheep.

Histochemistry of parathyroid gland

Carbohydrates

In the present study, the capsule of the parathyroid gland showed a mild positive reaction for PAS (Fig.13), which is similar to the findings of Rajalakshmi *et al.* (2018) in sheep. The PAS positivity indicated the presence of neutral mucopolysaccharides in the porcine parathyroidcapsule. The chief cells also showed positive reaction for PAS (Fig.14). Similar findings in goats were also reported by Roy *et al.* (1984). The colloid substance in the follicles showed intense positive reaction for PAS (Fig.14).

In the present study, the capsule of the parathyroid gland showed a mild positive reaction for PAS-AB (Fig.15), indicating the presence of acid mucopolysaccharides. Contrary to these findings, Rajalakshmi *et al.* (2018) stated that the capsule showed no reaction for PAS-AB in sheep. The secretory granules of the chief cells were positive for acid mucopolysaccharides (Fig.16).

Proteins

In the present study, the capsule showed mild positive activity for proteins (Fig.17). The secretory granules of chief cells showed intense positive activity for proteins (Figs.17,18), which is similar to the findingsin rabbit(Hara *et al.*,1969).

Calcium

In the present study, calcium deposits were present in the dark chief cells of the gland (Fig.19) as observed in Mongolian gerbils by Boquist and Lundgren(1975).

Conclusion

In thyroid gland, the lining epithelium, the basement membrane and thyrocolloid of thyroid follicles, while in parathyroid gland the colloid substance present in the mixed follicles and secretory granules in the chief cells showed intense positive reaction for PAS indicating the presence of neutral mucopolysaccharides. The capsule of thyroid gland and the secretory granules present in the chief cells of the parathyroid gland showed positive reaction for PAS-ABindicating the presence of acid mucopolysaccharides.

Conflict of interest

The authors declare that there is no conflict of interest in this study.

References

Bloom, W. and Fawcett, D.W. 1978.*Textbook of Histology*. (10th Ed.).W.B. Saunders Publishing Company, Philadelphia, pp. 500-513.

Boquist, L. and Lundgren, E. 1975. Effects of variations in calcium concentration on parathyroid morphology *in vitro*. *Laboratory Investigation; Journal of Technical Methods and Pathology*. **33(6)**: 638-647.

Capen, C.C. and Martin, S. 2003. The thyroid gland. In: McDonalds *Veterinary Endocrinology and Reproduction*.(5th Ed.). M. H. Pineda & M. P. Dooley, Iowa State Press, Ames, pp: 35–69.

Dellmann, H.D. and Brown, E.M. 1987.*Textbook of*

Veterinary Histology.(3rd Ed.). Lea &Febiger, Philadelphia, pp. 374-375.

Desmaris, A. and Laham, Q.N. 1962. The relation between the staining properties of the thyroidal colloid and its iodine content. *Canadian J.Biochem.Physiol.***40**: 227-236.

Greep, O.R. 1966.*Histology*.(2nd Ed.). Mcgraw-hill book Company, pp. 781-794.

Ham, D.C. and Cormack, D. 1972.*Histology*.(8th Ed.). J. B. Lippincott Publishing Company, Philadelphia, pp. 824-844.

Hara, J., Yamada, K. and Iwatsutsumi Y. 1969. Some aspects of proteins histochemistry in the parathyroid gland of the rabbit. *Nagoya J.Med.Sci.*.**32**: 159-168.

Harach, R.H. 1987. Mixed follicles in the human thyroid gland. Acta Anatomica.**129**: 27-30.

Hossam, H. Authman, A.M.M., Zoki, A.M.S. and Mohammad, G.F. 2012. Effect of seasonal temperature changes on thyroid structure and hormones secretion of white grouper (*Epinephelus aeneus*) in Suez Gulf. *J. Egypt Life Sci.*.**9(2)**:700-705.

Hussain, A.M. and Al– Taay, M.M. 2009. Histological study of the thyroid and parathyroid glands in Iraqi buffalo "*Bubalus bubalis*" with referring to the seasonal changes. *Basrah J.Res.***8(1)**: 26-38.

Huszenica, G.Y., Kuleszar, M. and Rudas, P. 2002. Clinical endocrinology of thyroid gland functions in ruminants.*Vet. Med. – Czech*, **47**: 199–210.

Khaleel, I.M and Mohammad Salih, A.A. 2017. Comparative histomorphological and histochemical study of thyroid gland in adult indigenous gazelle and sheep. *J.Entomol.Zool.Stud.*.**5(6)**: 1236-121.

Luna, L.G. 1968.*Manual of histological staining methods of armed force institute of pathology*.(3rd Ed.). McGraw Hill Book Co., New York, 87p.

Lunney, J. 2007. Advances in swine biomedical model genomics. *Int. J. Biol. Sci.* **3(3)**: 179-184.

Medrano, R.F. and Hua, H.J. 2016. Advances in Thyroid gland Hormones Function relate to Animal Nutrition*Annals Thyroid Res.***2(1)**: 45-52.

Nadhim, A.S. 2017. Histological and histological analysis of thyroid gland in slaughter male local Iraqian goats (*Capra aegagrus*). *Int. J.Agric.Sci.Vet. Med.***5(2)**: 59-66.

Oster, M., Keiler, J., Schulze, M., Reyer, H., Wree, A. and Wimmers, K. 2018 Fast and reliable dissection of porcine parathyroid glands - A protocol for molecular and histological analyses.*Annals Anat.***219**: 76-81.

Rajalakshmi, K., Ramesh, G., Kumari, U., Sivakumar, M., Uma, S. and Lakkawar, A.W. 2018. Microanatomy of the parathyroid glands in sheep (*Oviesaries*). *J. Entomol. Zool. Stud.***6(3)**: 988-993.

Rajalakshmi, K., Ramesh, G., Kumari, U., Kannan, Siva Kumar, M., Sridevi, P. and Lakkawar, A.W. 2019. Microanatomy of the thyroid gland in sheep (*Ovis aries*). *Int.J. Chem Stud.***7(2)**: 404-415.

Roy, K.S. 1971. Histological and histochemical studies on the thyroid glands of Indian buffaloes (*Bos bubalis*). *M.V.Sc thesis* submitted to Rajendra Agricultural University, Patna, and Bihar, 76p.

Roy, K.S. and Saigal, R.P. 1986. Histochemical and enzymatic observations on the ultimobranchial follicles of the sheep thyroid. *Indian J. Anim. Sci.***56**: 1013-1016.

Roy, K.S. and Saigal R.P. 1987. Histochemical study on thyroid gland of sheep during early pregnancy. *Indian J. Anim. Sci.*.**57**: 26-28.

Roy, K.S., Saigal, R.P. and Nanda, B.S. 1984. Histomorphological and histochemical studies of internal parathyroid gland of the goat. *Indian J. Anim. Sci*. **54**: 465-468.

Roy, K.S. and Yadava, R.C.P. 1977. Histological and certain histochemical studies on the thyroid gland of the Indian buffalo (*Bubalus bubalis*). *Indian J. Anim. Sci.***45**: 201-208.

Sanjeev, J. and Rakesh, M. 2016. Light microscopic studies on the thyroid gland of goats. *Rumin. Sci.***5(1)**: 25-28.

Singh, U.B. and Sulochana, S. 1997.*Handbook of histological and histochemical techniques*. Premier publishing house, Hyderabad, pp: 39-62, 69.

Tondini, T. 2007. Thyroid hormones in small ruminants: Effects of endogenous, environmental and nutritional factors.*Animal*.**1**: 997–1008. ■