ANATOMICAL STUDIES ON THYROID, PARATHYROID AND ADRENAL GLANDS OF INDIGENOUS ONE-HUMPED CAMEL (*Camelus dromedary*)

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(*Camelus dromedary*)

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AHMADU BELLO UNIVERSITY, ZARIA, NIGERIA

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Declaration

I hereby declare that the work in this dissertation titled "Anatomical Studies on some Endocrine Glands of the Indigenous One-humped Camel" was performed by me in the Department of Human Anatomy, under the supervision of **Dr. A. O. Ibegbu** (B.Sc., M.Sc., Ph.D.), and **Professor S. B. Oladele** (DVM, M.Sc., Ph.D.).

The information derived from the literature has been duly acknowledged in the text and a list of references provided. No part of this work has been presented for another degree or diploma at any institution.

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Certification

This project dissertation titled "**Anatomical Studies on Thyroid, Parathyroid and Adrenal Glands of the Indigenous One-humped Camel"** meets the regulations governing the award of the degree of **Master of Science** of the Ahmadu Bello University, and is approved for its contribution to knowledge and literary presentation.

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Abstract

Information on the anatomical studies of the endocrine glands of the indigenous one-humped camel in Northern Nigeria is scarce, and the fact that these endocrine glands may be involved in the mechanisms of adaptation to hostile environment through which it survives for long periods without food and water in the arid zones. Thirty (30) one-humped camels, comprising fifteen (15) males and fifteen (15) females were used in this study. The animals were divided into 3 groups of 10 animals each. Group A, B and C constitute 10 one-humped camels (5 males and 5 females) aged between 1 to 5 years, 6 to 10 years and above 10 years, respectively. The gross anatomical study of the endocrine glands namely; thyroid, parathyroid and adrenal glands were done to determine their shapes, texture, color, surfaces, and borders, measurements on the weights where obtained using digital weighing balance, and measurements of width, thickness and length were taken using digital vernier caliper. Histological features were studied using routine Haematoxylin and Eosin (H and E) for light microscopy. Histochemical features of the endocrine glands were explored using Periodic Acid Schiff (PAS) special stain; for thyroid and parathyroid glands, Acid Hydrolysis Azure A for thyroid gland, Alcian blue and potassium dichromate for adrenal gland. Hormonal assays of Thyroid Stimulating Hormone (TSH), Triiodothyronine (T3), and Tetraiodothyronine (T4) were studied using three different Enzyme Linked Immuno Servent Assay (ELISA) Kits (obtained from Nazo Medical Diagnostic Services, Lagos) for the T3, T4, and TSH ELISA. The result showed that the thyroid gland was a paired-lobe, and lengthy organ, with blood supply from superior thyroid, middle thyroid and inferior thyroid arteries. Correlation of the weight of the thyroid gland in one-humped camel was directly proportional to the age of the animal, while the correlation of the concentration of the thyroid hormones (T3 and T4) was inversely proportional to the weight and the age of the

thyroid gland in the indigenous one-humped camel. There were higher concentrations of thyroid hormones in the young than in the adult. Histological studies revealed the appearances of numerous vacuolated colloids, and apoptosis of the follicular cells of the adult animals, which highlight the reason for the above correlations. There were follicular and parafollicular cells. The parathyroid glands were two pairs of glands situated around thyroid gland lobes. In addition, accessory parathyroid glands were seen, in this study, situated on the right, while some on the left of the indigenous one-humped camel. The result showed that there was sexual dimorphism in the weight, length, width and thickness in the parathyroid glands in the juvenile age group and the young adult group, especially in the width, which was highly significant ($P<0.01$). The general histological features of the parathyroid gland consisted of densely packed chief cells (light and dark cells), oxyphil cells, and abundance of adipose cells in the adult male and female, but the adipose cells were absent in the young group. The right adrenal gland was positioned superior medial to the right kidney attached to the inferior medial surface of the liver, while the left adrenal gland was superior medially positioned within fat from the left kidney. There was sexual dimorphism in the weight and thickness of the adrenal gland of the female in the juvenile age group. Histologically, the adrenal cortex comprised of three zones generally, but four zones were seen in the young adult groups named; Zona glomerulosa, zona fasciculate, zona intermedia, and zona reticularis. Chromaffin cells of the medulla were identified by potassium dichromate chromaffin reactions. The results showed age and morphometric correlations, sexual dimorphisms and cellular morphology of the endocrine were pertinent to enhance the understanding of how the animal adapts to its environment, as this will provide baseline information for future related researches.

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Abbreviations

H and E = heamatoxylin and eosin

ABU = Ahmadu Bello University

ABUTH= Ahmadu Bello University Teaching Hospital

AKTH = Aminu Kano Teaching Hospital

USB = Universal Serial Bus

TSH = Thyrotropin

T3 = Triiodothyronine

T4 = Tetraiodothyronine

ISO = International Standard Organization

LA= Left Adrenal Gland

RA= Right Adrenal Gland

LK= Left Kidney

RK = Right Kidney

ELISA = Enzyme Linked Immuno Servant Assay

CHAPTER ONE

INTRODUCTION

1.1 Background

Dromedary camel (*Camelus dromedarius)* as a livestock species is gaining economic interest because of its adaptation to harsh climatic conditions and resistance to diseases Bengoumi (1992), Faye (1997), and Faye and Bengoumi (2000). Such adaptation is the basis for the reputation of this specie to be uniquely able, among other large mammals, to survive under harsh desert conditions. One of the dromedary's mechanisms of adaptation to its natural habitat is changes in the activity of its endocrine glands, such as the thyroid gland, parathyroid gland, adrenal gland, pituitary gland and others, which are implicated in growth and development, complex series of metabolic functions, thermoregulatory, specific organ functions, among many, yet numerous morphological variations of these glands are not well explored (Rejeb *et. al.*, 2011).

Dromedary Camel is a specie of animal that is anatomically and physiologically adapted to water and food restrictions and to an excessively hot climate (Ouajd and Kamel, 2009). It can survive in an incredibly harsh environment (up to 70°C), that is lethal to all other species of animal (Ouajd and Kamel, 2009). Camel can store water in all its body compartments (intracellular, extracellular, blood, and especially GIT). Every organ has the extraordinary capacity to store water Karkar (2011). Similarly, it is well known for its feats of endurance, a 750kg camel can carry 450kg luggage and able to travel at 16km/h for up to 18 hours nonstop Snow (1992).

Dromedary camel is described as the 'ship of the desert', as its toes have adapted to walking on sand without sinking, carrying heavy loads and transport them across the deserts Snow

(1992). Even though the traditional usages of camel in most countries have been supplanted by motorized transport, however, its cultural and economic importance have been maintained by camel racing, durbar racing, military mascots (such as the Nigerian Corporal Camel Rakumi in 65 division of 81 Battalion of the Nigerian Army in Lagos ANON (2011), meat and milk provisions, hair and leather textiles, transportation, working animals in farms Yagil (1982) and Snow (1992).

The breeds of the one-humped camel found in Northern Nigeria as indigenous are "Ja" (dark brown), "Fari" (grey white), "Bula" (pied colored), and "Bakin-biri" (Brown Black), identified based on their phenotypic trait of color (Abdussamad *et. al.,* 2011). This needs further investigation because these ecotypes could be of the same breed or of different breeds, the genuineness of which could only be proven by molecular genetic studies, and is yet to be done. The local name of camel in Hausa Language is called "*Rakumi*". A matured female is called "*Taguwa*", while the matured male is called "*Amale*" (Abdussamad *et. al.*, 2011).

1.2 STATEMENT OF RESEARCH PROBLEM:

There is paucity of information on the anatomical studies of the endocrine glands of the indigenous one-humped camel, despite the fact that these endocrine glands are involved in the Dromedary's mechanisms of adaptation to hostile environment through which it survives for long periods without food and water.

1.3 JUSTIFICATION FOR THE STUDY

The studies on the endocrine glands of the indigenous one-humped camel is pertinent to enhance the understanding of how the animal adapts to its environment, as this will provide baseline information on the camel for future related researches.

1.4 SIGNIFICANCE OF THE STUDY

The significance of the present study were:

It could be useful in comparative anatomy and evolutionary studies with other species of the *artiodactyla* order and higher orders like humans. It could also enhance and provide basic and clinical investigative information in endocrinology and related research fields, such as Animal breeding and Biotechnology.

1.5 AIM OF THE STUDY

The aim of the work was to study the anatomical features of thyroid, parathyroid and adrenal glands of the one-humped camel indigenous to Northern Nigeria

1.6 OBJECTIVES OF THE STUDY

The objectives of the research were:

I. To study the gross morphology and morphometry of the thyroid, parathyroid and adrenal glands of the one-humped camel.

II. To study the histology of the thyroid, parathyroid and adrenal glands of the one-humped camel using routine stain (H & E).

III. To study the histochemistry of the thyroid glands using Periodic Acid Schiff (PAS) for demonstration of glycogen and Azure A Acid hydrolysis technique for

demonstration of parafollicular cells; study the histochemistry of parathyroid glands using Periodic Acid Schiff (PAS) for demonstration of glycogen, as well; and to study the adrenal glands of the one-humped camel using Alcian Blue for demonstration of mucin and Potassium dichromate for demonstration of chromaffin cells.

IV. To determine the serum levels of Thyroid Stimulating Hormone (TSH), Triiodothyronine (T3) and Tetraiodothyronine (T4) using ELISA auto-analyzer kits.

V. To determine the sexual dimorphism in the morphometric variables of the thyroid, parathyroid and adrenal glands.

CHAPTER TWO

LITERATURE REVIEW

2.1 Dromedary camel

2.1.1 Etymology of the dromedary camel:

The term "dromedary" is derived from the word *dromos* (Greek word for "road") referring to its first usage in transportation Ouajd and Kamel (2009). However, camel dromedary is used throughout the world to describe the one-humped camel, the Bedouin name of the dromedary Marisa (2011).

The word "camel" was originally from Latin and Greek words *camelus* and *kamēlos,* respectively. Similarly, the Hebrew word *gāmāl* and Arabic word *jamal,* which is linguistically related to Arabic verb *hamala* meaning to bear or carry, related to the primary usage of the camel Herper (2012), and from the same root with the word *jamalun,* which is the Arabic name of camel.

2.1.2 Evolution and Distribution of camel

"Camel" as well, is generally used to describe any of the six camel-like mammals in the family *Camelidae*. It includes the two true camels; the camel dromedary and camel bactrian, and the four South American *camelids*; the *llama* and *alpaca* which are called the "New World camels", and the *guanaco* and *vicuña* called the "South American camels" Bornstein (2010).

Dromedary camel belongs to the Kingdom *Animalia*, Phylum *Chordata*, Subphylum *Vertebrata*, Class *Mammalia*, Order *Artiodactyla* (even-toed ungulates), Suborder *Tylopoda* Family *Camelidae* (camels, llamas, alpacas, guanaco and vicuña), Genus *Camelus* (camels),

and Species *Camelus dromedarius* (one-humped camel) (Gauthier-Pilthers and Dagg, 1981; Nowak, 1991; Wilson and Reader, 2005; Spaulding *et al*., 2009).

The *Tylopoda* (camels, llamas and alpacas) and *chevrotains* have a three-chambered stomach, while the rest of Ruminantia have four-chambered stomachs, hence categorized as Pseudo-ruminant. However, camels do regurgitate ingested forage. In fact, they are more efficient at feed conversion than are ruminants in extracting protein and energy from poor quality forages (Janis and Jarman, 1984; Marisa, 2011).

The *Camelidae* family differs from other true ruminants in not only having horns or antlers, but also their fore-stomachs are different morphologically and physiologically. The even-toed ungulates are generally herbivorous (Janis and Jarman, 1984). Furthermore, the red blood cells (RBCs) of the *Camelidae* are oval in shape, thus unique among all the mammals Bornstein (1990).

Fossils records prove that the early evolution of the *Camelidae* occurred in the North America. The *Camelus* spp appeared during the Pleistocene period, as well as during one of the ice ages when there was a land-bridge between Alaska and Siberia, where the "early camels" spread into Asia. During this migration from the "old world", the camel groups died out in Northern America, however, some *camelids* had migrated to the South America and evolved as the South American Camelids (the guanacos and vicunas) Marisa (2011).

In Asia, two groups estranged to become the two chief types of camel known today the dromedary camel distinct with long legs and a hump, and the Bactrian camel, with two humps and shorter legs. It was thought that the dromedary may have evolved from the Bactrian camel. However, the hump(s) may have been acquired as a result of domestication.

The one-humped camel (*Camelus dromedarius*) is found in the Arabian deserts, while the two-humped camel (*Camelus bactrianus*) is an Asiatic animal (Marisa, 2011).

2.1.3 Lifespan and Reproduction:

The dromedary camel has a lifespan of about 40 to 50 years. Typically, males and females are seasonal breeders. Breeding occurs in winter and overlaps with the rainy season; both vary in respect to the camel's geographic range. The onset of the breeding season is believed to be cured by nutritional status of the camel and the day length. Camels mate in sitting position, with the male mounting from behind. The male usually ejaculates three or four times within a single mating session (Mukasa-Mugerwa, 1981). Copulation time ranges from 7-35 minutes, averaging 11-15 minutes. The pregnancy typically lasts for a period of 15 months, followed by the birth of a single calf. The calf can move freely by the end of the first day. Maternal care generally lasts for 1 to 2 years. Females reach sexual maturity around three years of age and mate around age four or five. Males begin to rut by age three, but do not reach full sexual maturity until six years old (Mukasa-Mugerwa, 1981).

2.1.4 Domestication of camel

Camel dromedary was first domesticated in the Arabian Peninsula about 4000 years ago (Ojoud and Kamel, 2009; Karkar, 2011). Its close connection with Arab history and culture, made it to be called the Arabian camel (Bornstein, 1990). The domestication may have been in southern Arabia, the Bactrian in central Asia around 2,500 BC (Mukasa-Mugerwa, 1981; Bulliet, 1990; Scarre, 1993). To be more specific, the dromedary camel domestication was in the region, now known with the country Yemen, while the Bactrian camel was believed to be domesticated in the historic town of Bakhdi in the Zoroaster's time. It is believed that

before the introduction of dromedary camel in historic country of Bakhtaria, only Bactrian or Bakhdi camel was found (Karkar, 2011).

The dromedary camel is native to the Middle East and the Horn of Africa (dry and hot deserts), while the Bactrian inhabits the Central Asia (cold areas) (Karkar, 2011). Camel came a little late in the record of animal domestication (Karkar, 2011).

2.1.5 Relationship with Humans:

The even-toed ungulates are of more economic and cultural benefit than any other group of mammals (Encyclopedia Britannica, 2008). The camel today is essentially a domestic animal of pastoralists and agropastoralists. However camels are found even in areas of permanent cultivation, i.e; desert oases, along the Nile in Egypt or in Indian and Pakistani villages (Nowak, 1991). Camel is a multipurpose domestic animal that remains central to the subsistence, economic and social livelihoods of pastoralists in the Nigeria-Niger semi-arid zone (Abdussamad *et. al*., 2011).

2.1.6 Population and Geographic range/ Habitat:

The dromedary camels are much more numerous than the Bactrian camels, and they constitute about 90 per cent of the genus *Camelus* in the world. There are about 12 million of One-humped camels in the world, and more than 80 per cent are found in Africa, in more than 18 countries, constituting an important part of the domestic livestock population (Bornstein, 1990). Dromedary camels occupy arid regions of the Middle East through northern India and arid regions in Africa, most notably, the Sahara Desert. They have also been introduced to arid regions of central Australia where some of the only feral populations now persist (Nowak, 1991). The Bactrian inhabits the Central Asia (cold areas). The introduction of the dromedary camel, in most countries, is linked with the Arab Muslim

preachers (Karkar, 2011). The introduction of the camel into northern Nigeria could have occurred due to caravan trade links between the Sahara and Sokoto, Katsina and Kano in northern Nigeria (Blench, 1999). Dromedary camel exists in considerable number in semi-arid and savanna regions of Nigeria. Pastoral groups that originate from the Niger Republic own most of the camels in northern Nigeria (Blench, 1999).

The main breeders of the camel are the pastoral groups from Niger, but are also found in Borno, Yobe, Kano, Jigawa, Katsina, Sokoto, Kebbi, and Zamfara states of northern Nigeria (Abdussamad *et. al.,* 2011). Small-scale camel breeding is, however, undertaken by individual indigenous farmers in northern Nigeria (Blench, 1999). Some local farmers also keep camel largely as a draught animal for land preparation during the rainy season and for transportation of farm produce during harvest period or for transportation of other goods to and from village markets (Abdussamad *et. al.,* 2011).

Working bull camels are being used further south every year. Male camels are being brought further and further south into the centre of the country as portage stock. There seems little doubt that this rather dramatic change is being driven by the changes in the environment. As human population has expanded, land between farms available as a grazing resource has decreased. The cattle population has also grown, putting ever greater pressure on pastures. In many areas of the semi-arid zone, only browse plants remain and some of these are unavailable to any species except camels (Blench, 1999). Camels are specialized in eating browse and can ingest foliage from thorn trees and other armed plants. The environmental degradation of vegetation and the spread of thorns and woody plants give camel, which can digest these plants, a comparative advantage (Abdussamad *et. al.,* 2011).

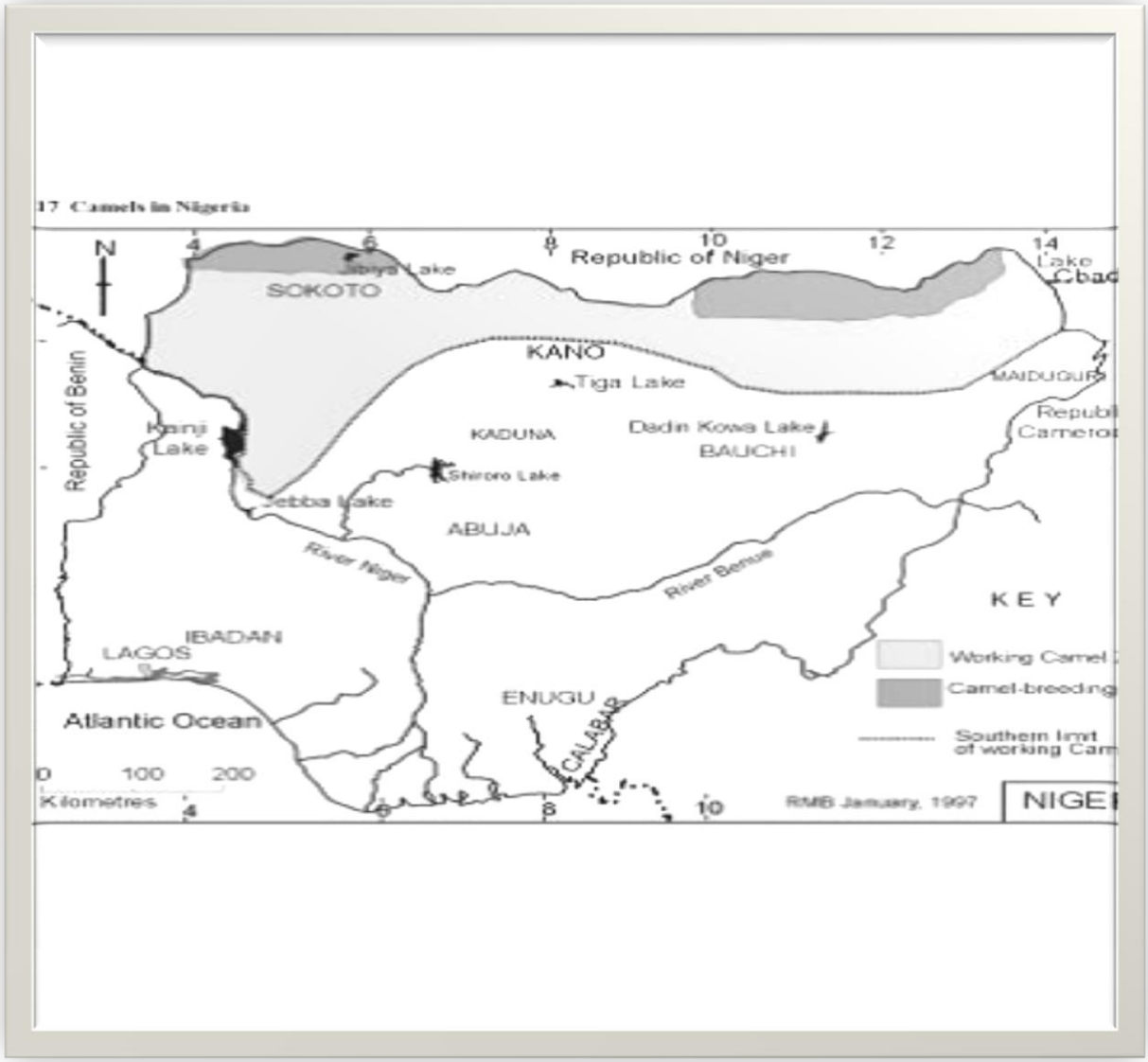


Figure 2.1: DISTRIBUTION OF CAMEL IN NIGERIA (Blench, 1999)

2.1.7 Gross Anatomy and Characteristics of the Dromedary Camel

Camel dromedary is an animal specie that is anatomically and physiologically adapted to water and food restrictions and to an excessively hot climate. It can survive in an incredibly harsh environment (up to 70°C), that is lethal to all other species (animal) (Ouajd and Kamel, 2009). Camel can store water in all its body compartments (intracellular, extracellular, blood, GIT etc). Every organ has the extraordinary capacity to store water (Karkar, 2011).

Dromedary camels are characterized by a single hump, long-curved neck, and deep-narrow chest. The hump is made up of fat bound together by fibrous tissue, as food storage during lack of food. The size of the hump varies with the nutritional status of the camel, becoming smaller and leaning to one side during times of starvation (Kohler-Rollefson, 1991).

Dromedaries are usually light brown or sandy brown in color, however, coloration can range from almost black to nearly white. Hair length is longer on the throat, shoulder, and hump areas (Kohler-Rollefson, 1991).

Male dromedaries, in comparison to females, are about 10% heavier, adult weighing 400-750kg, and are about 10cm taller at shoulder height, measuring 1.8 - 2.15m. Similarly, it is well known for its feats of endurance, a 750kg camel can carry 450kg luggage and is able to travel at 16km/h for up to 18hours nonstop (Snow *et. al.*, 1992). The Dromedary camels have a total of 34 teeth, with a dental formula of 1/3; 1/1; 3/2; 3/3 (Kohler-Rollefson, 1991). The feet of dromedaries are pad-shaped and adapted for traveling on sand. They can be easily injured on sharp stones and are unsuitable for slippery or muddy conditions. The lips of dromedary camels are thickened to allow eating of rough, thorny plants (Kohler-Rollefson, 1991).

Furthermore, male dromedaries have a specialized inflatable diverticulum of the soft palate called the "Dulla". It enlarges on filling with air from the trachea until it hangs out of the mouth of the camel and comes to resemble a pink ball. Copious saliva turns to foam covering the mouth as the male gurgles and makes metallic sounds. It uses it to assert dominance, in courtship to attract females. This occurs in only the one-humped camel (Kohler-Rollefson, 1991; Abu-Zidana, *et. al.,* 2011).

2.2 Endocrine Glands

Despite the differences in their functions the endocrine glands have certain common characteristics: they are ductless glands that secret their secretions directly into the intercellular and perivascular connective tissue spaces from which they reach the circulatory system. The endocrine glands are abundantly enriched with sinusoids; a thin walled vessels intimately associated with parenchymal cells, whose secretory products (hormones) pass directly into the circulatory system (Bacha and Bacha, 2000). Hormones are the chemical substances named because of the word “hormo” means “I arouse to activity” (Ojo, 1987). The circulating hormones regulate the functions of cells in general or that of specific tissues or organs, referred to as “target organs” (Ojo, 1987).

The functions of the endocrine system are intimately linked, coordinated, and sometimes even integrated with nervous system (Dellman, 1993).The endocrine glands include the pituitary gland, thyroid, parathyroids, adrenal glands, hypothalamus, thalamus, islet of Langerhans of the pancreas gland, enterochromaffin cells of the gastrointestinal epithelium, ovarian follicles of the ovary, corpus luteum, pineal gland, thymus, and interstitial cells of testis (Ojo, 1987; Bacha and Bacha, 2000).

2.2.1 Gross organization of mammalian thyroid gland

The gross anatomy of thyroid gland of one-humped camel revealed that the gland is located near the first ring of trachea, and consist of two lobes, with an isthmus connecting the lobes to each other. The colour of the gland is reddish brown (Kausar and Shahid, 2006).

Ahmadpanahi and Yousefi (2012) reported that the thyroid gland of one-humped camel is composed of two lobes, which are positioned each on the lateral surfaces of the trachea. The right lobe is located slightly cranial to the left one lying on the caudal aspect of the larynx, while both lobes are connected together by isthmus on the ventral surface of trachea (Ahmadpanahi and Yousefi, 2012).

In a morphometric study by Kausar and Shahid (2006) and Ahmadpanahi and Yousefi (2012) revealed that the values of the thyroid gland in adult one-humped camel ranges from 45-55kg for weight, 3.5 – 6.5cm for length, 2 – 4cm for width and 0.1 – 1cm for thickness. However, female adult dromedary camel has a little bit higher values of the thyroid gland in the above mentioned variables, especially weight and width, with the exception of thickness which is invariable (Ahmadpanahi and Yousefi, 2012). The statistical analysis revealed non-significant difference in various morphometric parameters between the juvenile and adult age groups. However, the values were relatively higher in adult camels compared with juvenile group (Kausar and Shahid, 2006).

The thyroid gland lobes in cattle, sheep and dog are asymmetric in position (Mc Donald, 2003). In horse the thyroid lobes are connected by a thin isthmus (Dyce *et. al.,* 2002). In rat and mouse an isthmus is located at the caudal end of the lobes (Ingbar, 1985).

The thyroid gland weight varies between domestic and wild animals and it depends on the size and weight of the animal (Little, 1991). The weight of the thyroid gland was found to be relatively higher in female than in male Wistar rats. However, the weight difference was not statistically significant (Capen and Martin, 2003). Both female and male adult Wistar rats contained a similar DNA quantity. There is sexual dimorphism in thyroid gland of the Wistar rats, which depends upon a difference in the mean volume of thyroid follicular cells with the males having larger cells than the females (Capen and Martin, 2003).

In humans, thyroid gland develops as pharyngeal outgrowths that migrate caudally to their final position, in the anterior neck, as embryological development continues (Drake *et. al.*, 2010). It is a large, unpaired gland, positioned in the anterior neck, below and lateral to the thyroid cartilage. The thyroid gland lies deep to the sterno-hyoid and sterno-thyroid muscles located anteriorly in the neck at the level of the C5 to T1 vertebrae (Moore and Dalley, 2006). According to Singh (2006) human thyroid gland consists of two symmetrical lobes united by an isthmus which lies anterior to the second, third and fourth tracheal rings. The lobes lie on either side, or anterolateral, of the larynx and trachea, extending from the oblique line of the thyroid cartilage to the sixth tracheal ring (Moore and Dalley, 2006; Singh, 2006), and weighs about 25g. In addition to its own capsule, the gland is enclosed by an envelope of pretracheal fascia (Singh, 2006).

In the adult human, the entire thyroid gland weighs around 15 to 20 g (Burtis and Ashwood, 2001). Each lobe measures approximately 4.0 cm in length, 2.0 cm in thickness and 2.5 cm in width (Burtis and Ashwood, 2001).

The thyroid gland, highly vascular, is supplied by superior and inferior thyroid arteries. These vessels lie between the fibrous capsule and loose fascia sheath (Moore and Dalley,

2006). Usually the first branches of the external carotid arteries, the superior thyroid arteries descend to the superior poles of the gland pierce the pre tracheal layer of deep cervical fascia and divides into anterior and posterior branches supplying mainly the antero-superior aspect of the gland (Moore and Dalley, 2006). The inferior thyroid arteries, the largest branches of the thyro-cervical trunks arising from the subclavian arteries, run supero-medially, posterior to the carotid sheaths to reach the posterior aspect to the thyroid gland and supply the postero inferior aspect, include the inferior poles of the gland (Moore and Dalley, 2006). The right and left superior and inferior thyroid arteries anastomose extensively within the gland, ensuring its supply, while providing potential collateral circulation between the subclavian and external carotid arteries (Moore and Dalley, 2006). In approximately 10% of people, a small unpaired thyroidea ima artery arises from the brachiocephalic trunk or the aortic arch (Moore and Dalley, 2006).

The gland is drained by three pairs of thyroid veins usually forming a thyroid plexus of veins on the anterior surface of the thyroid gland and interior to the trachea (Moore and Dalley, 2006). The superior thyroid veins accompany the superior thyroid arteries they drain the superior poles of the thyroid gland, the middle thyroid veins do not accompany but run essentially parallel with the inferior thyroid arteries, they drain the middle lobes. The usually independent inferior thyroid veins drain the inferior poles (Moore and Dalley, 2006).

The lymphatic vessels of the thyroid gland run in the interlobular connective tissue, usually near the arteries (Moore and Dalley, 2006). From there, the vessels pass initially to pharyngeal, pre tracheal and para tracheal lymph nodes which drain in turn to the superior and inferior deep cervical nodes (Moore and Dalley, 2006). Laterally, lymphatic vessels located along the superior thyroid vein pass directly to the inferior deep cervical lymph

nodes. Some lymphatic vessels may drain into the brachiocephalic lymph nodes or thoracic ducts (Moore and Dalley., 2006).

The nerves of the thyroid gland are derived from the superior middle and inferior cervical sympathetic ganglia (Moore and Dalley, 2006). They reach the gland through superior and inferior thyroid per arterial plexuses that accompany the thyroid arteries. These fibers are vasomotor, not secretor motor. They cause constriction of blood vessels. Endocrine secretion from the thyroid gland is hormonally regulated by the pituitary gland (Moore and Dalley, 2006).

2.2.2 Histological organization of mammalian thyroid glands

The mammalian thyroid gland is surrounded by a thin capsule of connective tissue and divided into lobules by thin trabeculae (Bacha and Bacha, 2000). The trabeculae are continuous with sparse intralobular connective tissue that contains numerous sinusoids (Bacha and Bacha, 2000). In the pig and cow the connective tissue is abundant. Each lobule consists of numerous follicles of various sizes that are frequently filled with colloid (Bacha and Bacha, 2000). The follicular cells vary in height, depending on the state of activity of the follicle. Their appearance changes from squamous or low cuboidal in the resting stage to cuboidal or columnar in the active stage. In an active follicle the periphery of the colloid adjacent to the apical surface of the follicular cells is vacuolated. In an inactive follicle, the colloid has a smoother peripheral surface and vacuoles are not present (Bacha and Bacha, 2000).

Parafollicular cells or C cells occur among the follicular cells and between them. They are larger and have a paler cytoplasm than the follicular cells. Their nuclei are relatively large

and pale. They occur usually singly, but may also appear in groups. In dogs, parafollicular cells occur particularly in abundance (Bacha and Bacha, 2000).

According to Kausar and Shahid (2006) the histology of the thyroid gland of the native dromedary camel in Pakistan consists of a connective tissue capsule and trabeculae which extend from the capsule into the substance of the gland, dividing it into lobules. Each lobule consisted of two sized follicles in variable numbers, the large and small.

The large follicles were lined by low cuboidal epithelium, while the small follicles were lined by high cuboidal to columnar epithelium (Kausar and Shahid, 2006). The follicular epithelium of the thyroid gland of the one-humped camel consisted of a single type of epithelial cell which varied from flattened to columnar in shape (Atoji *et. al.*, 1999). The follicles had colloid material in their lumen, probably an apocrine secretion from the lining epithelial cells. The parafollicular or C-cells were absent in thyroid glands of camel (Kausar and Shahid, 2006; Rejeb *et. al*., 2011). However, Ahmadpanahi and Yousefi (2012) reported that thyroid gland contained follicles, follicular epithelium and parafollicular (C) cells in microscopic examination; and that the parafollicular cells were about 5% of the cell population.

The size of the thyroid follicles is highly variable in camel dromedary; they are larger for adult one-humped camel, especially during the summer. Follicles on the periphery are larger than central ones (Rejeb *et. al.*, 2011).

According to Rejeb *et. al*., (2011) the age is significant in morphometric measurement of the cells, the largest follicular surface area was recorded for animals of the age above 15 years ($6873\pm670\ \mu m^2$); the smallest value corresponded to animals between the age 3 to 5 years

(about puberty age) (4912±467 μm^2), while dromedary camel less than 3 years (juvenile age) were intermediate with a mean value of 5650±386 μm^2 ($P<0.05$).

2.2.3 Hormones of the Thyroid Gland

The follicular cells produce two main thyroid hormones, thyroxine (T4) and triodothyronine (T3). The functional unit of the thyroid gland is the thyroid secretory follicle (Larsen *et al*., 2008), in which thyroid hormone is synthesized first by iodide organification and then by iodothyronine formation (Taurog, 1986) to form T4 and T3. Although other iodothyronines are formed, their biological functions appear to be limited (Chan and Singer, 1993). The major secretory product of the thyroid gland is T4. Of circulating T3, only approximately 15 – 20% is directly secreted by the thyroid and the remainder is derived from peripheral conversion of T4 to T3 by monodeiodination (1, 2). The process of monodeiodination of T4 can lead to the production of either active T3 or inactive reverse T3 (rT3) (Cheron *et. al*., 1979; Berry *et. al*., 1991).

Thyroid hormones are found in all tissues of the body and exert their effect on growth and development as well as a complex series of metabolic, thermoregulatory, and specific organ functions (Chan and Singer, 1993) through the binding of T3 to a nuclear receptor that regulates and modulates specific gene transcription (Sterling, 1979). The homeostatic control of release of thyroid hormones is controlled through feedback mechanisms by the glycoprotein thyrotropin (thyroid stimulating hormone, TSH) secreted by the anterior pituitary gland. Both T4 and T3 on the pituitary thyrotroph affect the release of TSH. The secretion of TSH by the pituitary is regulated by the thyrotropin releasing hormone (TRH) (Chan and Singer, 1993), which is a tripeptide released by the hypothalamus. TRH stimulates the release of TSH by the pituitary. The thyroid gland also secretes calcitonin hormones by the parafollicular or C cells which lower blood calcium (Ojo, 1987).

The highest concentrations of thyroid hormones were observed for dromedary camel between the ages of 3 to 5 years, while the lowest was observed in older dromedary above 15 years ($P<0.05$). Highest and lowest serum concentrations were 20.41 ± 0.30 (S.E.M.) Pmol/l and 12.64 ± 0.30 (S.E.M.) Pmol/l, 5.70 ± 0.11 (S.E.M.) Pmol/l and 3.80 ± 0.11 (S.E.M.) Pmol/l, and 0.25 ± 0.01 (S.E.M.) miU/l and 0.09 ± 0.01 (S.E.M.) miU/l for T4, T3 and TSH respectively (Rejeb *et. al*., 2011).

2.3 Gross and histological organization of mammalian parathyroid gland

The parathyroid glands are classified as internal and external. Those that are adjacent to or embedded in the thyroid gland are the internal parathyroids. The external parathyroids lie a variable distance away from the thyroid gland. The parathyroid glands are surrounded by a thin capsule of connective tissue, which may be absent where the glands are deeply embedded within the thyroid gland. A stroma of connective tissue is well developed in the pig and cow, but is sparse in other mammals (Bacha and Bacha, 2000).

The parathyroid glands, in one-humped camel, are situated on the dorsal border on the cranial pole of the thyroid gland. It is located as separate parathyroid structure from the architecture of the thyroids (Metwally *et. al.*, 2006). Each is surrounded by a capsule of connective tissue. Its parenchyma is highly vascularized (Metwally *et. al.*, 2006). The parathyroid gland in camel is mainly identical in histological structure to other mammals (Nagpal *et. al.*, 2001). It is identical to histology of parathyroid glands of goat and cow (Roy *et. al.*, 1999; Charles *et. al.*, 2000).

The parenchyma of the parathyroid gland consists primarily of clusters and cords of principal or chief cells (Bacha and Bacha, 2000). In many studies it was reported that mammalian parathyroid gland contains two types of cells; the principal cells (or chief cells)

and the oxyphil cells (Ojo, 1987; Dellmann, 1993; Bacha and Bacha, 2000; Fleischer, *et. al.*, 2004; Singh, 2006). There are two different functional stages of the principal cell. The light principal cell is inactive and has a large, pale nucleus and pale, acidophilic cytoplasm. The dark principal cell is a smaller, active cell with a small, dark nucleus and a deeply acidophilic cytoplasm. In the sheep and goat, light cells tend to be located peripheral to the more central, dark cells. In the other domestic mammals these cells are distributed randomly (Bacha and Bacha, 2000). However, some described three types of principal cells; light, dark and clear cells (inactive (light) and active (dark) forms) (Ojo, 1987; Dellmann, 1993; Fleischer *et. al.*, 2004; Singh, 2006).

The Oxyphil cells are large cells with an acidophilic cytoplasm and pyknotic nucleus (Bacha and Bacha, 2000; Singh, 2006). They have been reported to occur in small quantity in the horse and cow, particularly older animals **(**Bacha and Bacha, 2000**).** Oxyphil cells do not occur in the parathyroid gland of the dog, cat, rat, and man until ten years of age (Ojo, 1987 and Dellmann, 1993). The oxyphil cells are not present in human fetal parathyroid gland (Fleischer *et. al.*, 2004). They first appear in late childhood and increase in number following advancement in age often forming nodules in parathyroid of older person (Fleischer *et. al.*, 2004). Oxyphil cells are absent in chicken and many species of lower animals (Fleischer *et. al.*, 2004).

The parathyroid secretes parathormone which regulates the level of calcium and phosphorous in the plasma (Ojo, 1987). Hence, the glands are essential for life in most animals and human (El-Zoghby, 2004; Bensley, 2007).

In mammals and lower species of animal it have been reported that the oxyphil cells contain glycogen particles and free ribosome, granular endoplasmic reticulum, Golgi apparatus and

secretary granule which are poorly developed, suggesting that oxyphil cells of normal parathyroid do not have an active function in the biosynthesis of parathyroid hormone (Capen and Martin, 2003). In most mammals there is a higher oxidation and hydrolytic enzymes activity in the chief cells than in the oxyphil cells (Nakanishi *et al*., 2004). The functions of the Oxyphil cells are not known (Singh, 2006).

In humans, most people have four parathyroid glands (right and left superior parathyroid glands, and right and left inferior parathyroid glands). Approximately, 50% of people have more, while some have two glands (Moore and Dalley, 2006). The superior parathyroid glands are more constant in position than the inferior ones which are usually at the border of the cricoid cartilage (Moore and Dalley, 2006). The superior parathyroid glands usually lie slightly more than one centimeter superior to the point of entry of the inferior thyroid arteries into the thyroid gland. The inferior parathyroid glands usually lie slightly more than one centimeter inferior to the inferior thyroid artery entry point (Moore and Dalley, 2006).

The parathyroid glands are small flattened oval in shape and usually lie external to the thyroid capsule on the medial half of the posterior surface of each lobe of the thyroid gland, or inside its sheath (Moore and Dalley, 2006).

The glands are supplied by the inferior thyroid artery which provides the primary blood supply to the posterior aspect of the thyroid gland where the parathyroid glands are located (Moore and Dalley, 2006). However they may also be supplied by branches from the superior thyroid arteries; the thyroid ima artery or the laryngeal, tracheal and esophageal arteries. Parathyroid veins drain into the thyroid plexus of veins of the thyroid and trachea (Moore and Dalley, 2006). Lymphatic vessels from the parathyroid glands drain with those from the thyroid gland into deep cervical lymph nodes and Para tracheal lymph nodes

(Moore and Dalley, 2006). The nerves supply of parathyroid gland is derived from thyroid branches of the cervical sympathetic ganglia (Adolffaller and Schünke, 2004). Like the nerves to the thyroid, they are vasomotor rather than secretomotor because these glands are hormonally regulated (Moore and Dalley, 2006).

2.4 Gross and histological organization of mammalian adrenal glands

The Adrenal glands or suprarenal glands are two in number; right and left, found resting on the superior pole of the kidneys (Dellmann, 1993; Ojo, 1987). The paired adrenal glands are situated close to the anterior end of the kidneys (Bacha and Bacha, 2000). They are flattened endocrine organs located in retroperitoneal space along the medial cranial poles of the kidneys in most mammals (Prasad and Sinha., 1981; Rosol *et al*., 2001).

But in one-humped camel they are near the cranial pole not resting on top but towards the medial border of the kidney (Nabipour *et. al.*, 2008). The glands were embedded in fat. The right gland is more cranial than the left. The right gland of dromedary camel is reported to be approximately bean shaped; whereas the left gland is almost triangular (Nabipour *et. al.*, 2008). In other mammals left gland often is larger than the right (Ojo, 1987; Dellmann, 1993).

In humans, the adrenal glands are located between the superomedial aspect of the kidneys and the diaphragm, where they are surrounded by connective tissue containing considerable amount of fat (Moore and Dalley, 2006). Despite their name, they are separated from the kidneys by a thin septum. The shape and relations of the glands differs on both sides (Moore and Dalley, 2006). The pyramidal right gland is more apical situated over the superior pole) relative to the right kidney. The crescent shaped left gland is medial to the superior half of

the kidney and is related to the spleen, stomach, pancreas and lift cross of the diaphragm (Moore and Dalley, 2006).

The glands are covered by a capsule of dense irregular connective tissue that sometimes contains smooth muscle. Clusters of epithelioid cortical cells occur in the capsule. Thin trabeculae project partially into the parenchyma (Bacha and Bacha, 2000)

Each adrenal gland is organized into two parts the peripheral cortex and a central medulla (Bacha and Bacha, 2000; Moore and Dalley, 2006). These parts have different embryological origin and different functions (Moore and Dalley, 2006). The gland, in most animals, is surrounded by a connective tissue capsule (Kikuta and Murakami, 2003). A macroscopic cross section of the gland revealed that it consists of distinct outer part or cortex and an inner portion called the medulla (Prasad *et. al.,* 1981; Ojo, 1987; Dellmann, 1993; Rosol *et. al*., 2001). In ruminants and swine, the cortex is flesh colored and cream or light yellow. In equines, dogs and cats, the medulla of the gland is reddish brown due to the presence of blood flow in the medullary veins (Gelberg *et. al.*, 2000). The adrenal cortex consists of 3 zones: Zona glomerulosa, Zona fasciculata, and Zona reticularis (Ojo, 1987; Dellmann, 1993). The adrenal cortex is divided into four zones; zonas glomerulosa, intermedia, fasciculate, and reticularis (Bacha and Bacha, 2000). In gunea pig, the cortex is divided into three zones (without intermedia zone) (Colby *et. al.,* 1992), as well as in rats (Moog *et al*., 1998), and in buffalo and in some other mammals (Banks, 1993).

The zona glomerulosa or zona multiformis is the outermost zone. In the carnivore, horse, and pig the parenchymal cells of this region are columnar and arranged into arcs. In the horse the columnar cells are especially tall. In ruminants the zona glomerulosa contains polyhedral cells that form irregular clusters or cords (Bacha and Bacha, 2000).

The zona intermedia are seen more often in the horse and carnivore than in other domestic mammals. It lies between the zona glomerulosa and the zona fasciculata. It consists of small, closely packed cells (Bacha and Bacha, 2000).

The zona fasciculata is the widest zone of the cortex (Bacha and Wood, 1990; Junqueira *et. al.*, 1995; Bacha and Bacha, 2000). Zona fasciculata forms 60% of the cortex (Hussein, 2003). It is formed by radially arranged cords of cuboidal or polyhedral cells. The cords are one or two cells thick and separated by sinusoids (Bacha and Bacha, 2000). The cytoplasm of the cells in this zone frequently appears foamy because of the presence of numerous lipid vacuoles (Bacha and Wood, 1990; Junqueira *et. al.*,, 1995; Bacha and Bacha, 2000).

The zona reticularis is the innermost zone of the adrenal cortex. It is arranged as an irregular zone of the adrenal cortex. It is arranged as an irregular network of anastomosing cords of cells surrounded by sinusoids. (Bacha and Bacha, 2000)

The human histology of the adrenal cortex is made up of the cells arranged in cords. Sinusoids intervene between the cords. On the basis of the arrangement of cells the cortex is divided into: The outer most layers called the Zona glomerulosa. Here cells are arranged as inverted U-shaped formation. This zone consists of one fifth of the cortex (Singh, 2006). The next layer is Zona fasciculata. Here the cells are arranged in straight columns, two cells thick. Sinusoids intervene between the columns. This layer forms the middle three fifth of the cortex (Singh, 2006). Zona fasciculata in humans is 65% of the gland (Junquiera *et. al.*,, 1995). The innermost layer of the cortex that is the inner one fifth is called the Zona reticularis It is so called because it is made up of cords that branch and anastomose with each other to form a kind of reticulum (Singh, 2006).

With the light microscope the cells of the Zona glomerulosa are seen to be small, polyhedral or columnar with basophilic cytoplasm and deeply staining nuclei (Singh, 2006). The cells of the zona fasciculata are large polyhedral with basophilic cytoplasm and vesicular nuclei. The cells of the zona fasciculata are very rich in lipids. These cells also contain considerable amount of vitamin C. (Singh, 2006). The cells of the zona reticularis are similar to those of the zona fasciculata but the lipid content is less. The cytoplasm is eosinophilic. The cells often contain brown pigment.

The adrenal medulla is composed mostly of columnar or polyhedral chromaffin cells, which form clusters and anastomosing cords separated by sinusoids. In domestic mammals, an outer and inner zone of the medulla can often be distinguished. The former consists of larger, more darkly stained cells, while the latter contains smaller, more lightly stained cells. Ganglion cells, either individually or in clusters, are scattered through the medulla. Projections of the zona reticularis may appear within the medulla, because the cortex and medulla interdigitate at their junction (Bacha and Bacha, 2000).

In the human adrenal gland, the medulla is made up of groups of cells which are separated by wide sinusoids. These cells are columnar and have basophilic cytoplasm (Singh, 2006). They are considered to be modified postganglionic sympathetic neurons (Singh, 2006).

According to Moore and Dalley (2006), human embryological development shows that the adrenal cortex is derived from mesoderm and secretes corticosteroids and androgens. These hormones cause the kidneys to retain sodium and water in response to stress, increasing the blood volume and blood pressure. They also affect muscles and organs such as the heart and lungs (Moore and Dalley, 2006). The cells of the zona glomerulosa produce the mineralocorticoid aldosterone and deoxcoticosterone. These hormones affect the electrolyte

balance in the body. The cells of the zona fasciculata produce glucocorticoids cortisone and cortisol (Singh,2006) These hormones have wide effects including carbohydrate metabolism and protein metabolism. Cells of this zone also produce small amount of androgen (Singh, 2006). Cells of the zona reticularis also produces some glucocorticoids and sex hormones both oestrogen and androgen (Singh, 2006).

The adrenal medulla is derived from neural crest cells associated with sympathetic nervous system. The chromaffin cells of the medulla are related to sympathetic ganglion cells in both derivation (neural crest cells) and function (Moore and Dalley, 2006). These cells secrete epinephrine into the blood stream in response to signals from presynaptic nerves (Moore and Dalley, 2006). The powerful medullary hormones epinephrine and norepinephrine activate the body to a fight or flight status in response to traumatic stress. They also increase heart rate and blood pressure, dilate the bronchioles and change blood flow patterns, preparing for physical exertion (Moore and Dalley, 2006). The secrete noradrenalin and adrenalin into the blood (Singh, 2006). This secretion takes place mainly at stress (fear, anger) and results in widespread effects similar to those of the stimulation of the sympathetic nervous system(e.g., increase in heart rate and blood pressure) (Singh, 2006).

The hormones of adrenal cortex are among the most potent substances available to the animal and are necessary for the maintenance of life (Ojo, 1987; Dellmann, 1993). They enable the animals to function in a constantly changing environment (Bacha and Wood, 1990). Total adrenalectomy led to death within few weeks (Ojo, 1987; Dellmann, 1993). Adrenalectomized animals are unable to withstand stressful conditions such as exposure to cold, toxins or trauma and changes in the environment that may have no great effect in normal circumstances may be fatal to the animal (Ojo, 1987; Dellmann, 1993).

The hormones secreted by the adrenal cortex are: mineralocorticoids (aldosterone, and deoxycorticosterone), which are secreted by the zona glomerulosa, and affect the salt and water balance. Glucocorticoids (cortisone and hydrocortisone) are secreted by zona fasciculata and affect carbohydrate metabolism. The Zona reticularis secretes. The medulla secretes adrenalin (the same as sympathetic nerve synapses) and noradrenalin (Ojo, 1987).

The maximum response of the animal to stressful conditions in the environment is possible by activation of these glands and the mechanism of their hormones (Ojo, 1987; Singh, 2006).

In humans the blood supply to these glands is profusely and from three sources: The aorta (one or more middle suprarenal arteries), the inferior phrenic artery (6-8 superior suprarenal arteries), and the renal artery (one or more inferior suprarenal arteries) (Moore and Dalley, 2006).

The venous drainage of the adrenal glands is large suprarenal vein which drains into the inferior vena cava, whereas the longer left suprarenal vein, often joined by the inferior phrenic vein empties into the left renal vein (Moore and Dalley, 2006).

The suprarenal lymphatic vessels arise from the plexus deep to the capsule of the gland and from one in its medulla drain the glands of lymph (Moore and Dalley, 2006). These glands have rich innervations from the adjacent celiac plexus and the greater thoracic splanchnic nerves. These preganglionic sympathetic fibers pass through the hilum and synapse on the cells in the medulla. The suprarenal cortex receives only a vasomotor nerve supply (Moore and Dalley, 2006).

CHAPTER THREE

Materials and Methods

3.1 Experimental Animals

Thirty (30) one-humped camels, comprising fifteen (15) males and fifteen (15) females, between the ages 1-5, 6-10 and above 10 years, representing juvenile, adult, and old ages, respectively were used for the study. The camel gland's tissues to be studied were obtained from the Kano Abattoir.

An introduction letter was issued from the Department of Human Anatomy to the Veterinary Department of Kano State Ministry of Agriculture and Mineral Resources for permission to carry out the procurements of the tissues. The permission was granted and we received maximum cooperation and assistance throughout the procurements.

Physical examinations were carried out on each camel, 10 minutes prior to slaughter, by Veterinarians attached to Veterinary Department of Kano State Ministry of Agriculture. Only apparently healthy camels were used for this study (Plate 3.1).

The age of the animals was determined according to their dental formulae, as described by *Faye* (1997).



Plate 3.1: How the One-humped Camels were inspected when going to Slaughter House

3.2 STUDY DESIGN

The animals were divided into 3 groups of 10 animals in each group for the study, as follows:

Group A constitutes 10 one-humped camels (5 males and 5 females) aged between 1 to 5 years old.

Group B constitutes 10 one-humped camels (5 males and 5 females), aged between 6 to 10 years old.

Group C constitutes 10 one-humped camels (5 males and 5 females), aged above 10.

3.3 HORMONAL ASSAYS

Blood samples were collected at the point of slaughter via the jugular vein at the neck (Plate 3.2) into bottles containing ethylene diamine tetra acetic and centrifuged at 10,000 rmp for 10 min, the resulting blood serum were used for hormonal assays for the quantitative determination of Thyrotropin (TSH), Triodothyronine (T3), and Tetraiodothyronine (T4) concentrations in One-humped camel serum by a MicroPlate Immunoenzymetric assay.

The hormones were assayed using three different ELISA Kits; the T3 ELISA kit, T4 ELISA kit as prescribed by INTECO Diagnostic, UK, ISO 13485 accredited company (purchased from NAZO Medical Diagnostic Services No. 13 Winfunke Olowe Crescent, Off Lagos/ Abeokuta Express Way, Lagos state) as follows: For Thyrotropin (TSH) ELISA the reagents were prepared by washing the buffer concentrate to 1000ml with distilled or deionized water in a suitable container and stored at room temperature (20 – 27^{o}C). The Working substrates were prepared by preparing 1 + 1 mixture of Substrate A and Substrate B, and were used

immediately after preparation. Before the test, all the reagents serum references were brought to room temperature (20 – 27^{o}C)

The test procedures were:

- The microPlates' wells were formatted for each serum reference, and camel specimen to be assayed in duplicate. All unused microwell strips were replaced back into the aluminum bag and store at 2-8^{o}C.
- 0.050ml (50µl) of the appropriate serum reference or specimen into the assigned well
- 0.100ml (100µl) of the TSH Enzyme Reagent was added to each well. NB: It is very important to dispense all reagents close to the bottom of the coated well.
- The microplate was swirled gently for 20-30 seconds to mix and cover
- Then it was incubated for 60 minutes at room temperature
- The contents of the microplate were discarded by decantation. The plate was taped and blot dry with absorbent paper
- 300 µl of wash buffer was added and decanted (taped and blotted). This was repeated twice, for a total of three (3) washes.
- 0.100 ml (100 µl) of working substrate solution was added to all wells. The reagents were added in the same order always to minimize reaction time differences between wells.
- It was incubated at room temperature for fifteen (15) minutes.
- Stop solution of 0.050 ml (50 µl) was added to each well and mixed gently for 15-20 seconds.
- The absorbance was read in each well at 450 nm (using a reference wavelength at 620-630 nm to minimize well imperfections) in a microPlate reader.

The result was read within thirty (30) minutes of adding the stop solution.

For Thyroxine (T4) and Triodothyronine (T3) quantitative concentration determination

The reagents were prepared as follows:

- The working reagents A= T4-Enzyme conjugate solution and T3-Enzyme conjugate solution were diluted 1:11 with total (T3/T4) conjugate buffer in a suiTable container. And was used immediately
- Wash buffer: The content of wash concentrate was diluted to 1000ml with distilled water.

Working substrate solution was prepared 1 + 1 mixture of substrate A and substrate B. Use immediately after preparation. Enough quantities were prepared. Before proceeding with the assay, all the reagents, serum concentrations were brought to room temperature.

The complete test procedures were:

- The microplates' wells were formatted for each serum reference (T4 and T3), and the camel specimen to be assayed in duplicate. All unused microwell strips were replaced back into the aluminum bag and store at 2-8^{o}C.
- 0.025ml (25µl) of the appropriate serum reference or specimen were pipeted into the assigned well, for T4. While for T3 is 0.050ml (50 µl)
- 0.100ml (100µl) of working Reagent A, T4/ T3-enzyme conjugate solution were added to each well.
- The microPlate was swirl gently for 20-30 seconds to mix and cover
- Then it was incubated for 60 minutes at room temperature
- The contents of the microPlate were discarded by decantation. The Plate was tap and blot dry with absorbent paper
- 300µl of wash buffer was added and decanted (taped and blotted). This was repeated twice, for a total of three (3) washes.

- 0.100ml (100 µl) of working substrate solution was added to all wells. The reagents were added in the same order always to minimize reaction time differences between wells.
- It was incubated at room temperature for fifteen (15) minutes.
- To stop solution 0.050ml (50 µl) was added to each well and mixed gently for 15-20 seconds.
- The absorbance was read in each well at 450nm (using a reference wavelength at 620-630nm to minimize well imperfections) in a microplate reader.

The result was read immediately, within thirty (30) minutes of adding the stop solution.

The hormonal assays were carried out in the Chemical Pathology Department of the Ahmadu Bello University Teaching Hospital, Shika, Zaria, Kaduna State.

3.4 DISSECTION OF THE ENDOCRINE GLANDS:

After the camels were slaughtered (Plate 3.2), the skin of the animal was reflected. Then, the neck detached (Plates 3.3 and 3.4), the upper part of the respiratory track as well as the associated organs were separated and removed intact from the heavy muscles of the posterior neck (Plate 3.5). Incision was made sagitally, through the muscles of the anterior neck, specifically sterno-thyroid muscles. This exposed the thyroid cartilage, cricoid cartilage, tracheal rings and the thyroid gland. The pictures of the thyroid glands, with the related structures, were taken in situ. The thyroid glands were removed, and the morphological and morphometric studies were done, using sensitive digital weighing balance, digital vernier caliper, and immediately fixed in 10% neutral buffered formalin.

The parathyroid glands were located around the thyroid gland, hence were removed along with the thyroid, the morphological and morphometric studies were done using sensitive

digital weighing balance, digital vernier caliper and immediately fixed in 10% neutral buffered formalin.

The camel's abdominal region was dissected while the camel was in sitting position through a lateral incision in between the border between the thoracic and abdominal regions and accessed the viscera (Plate 3.6). Pictures were taken to view the adrenal glands in-situ (Plate 4.29). Other pictures were taken after removing the viscera out of the camel, and fats were removed. The right adrenal gland was located between the right kidney and the liver. The right adrenal glands was removed, the morphological and morphometric studies were done using sensitive digital weighing balance, digital vernier caliper, and fixed immediately in 10% neutral buffered formalin, while some in potassium dichromate (for chromaffin special reaction study). The left adrenal gland was located between the left kidney and the spleen. It was removed, weighed using sensitive digital weighing balance, and fixed immediately in 10% neutral buffered formalin, while some in potassium dichromate.



Plate3.2: How camel is being slaughtered in Kano Abbatoire (A young camel being slaughtered)



Plate 3.3: How the skin was dissected



Plate 3.4: The Head and Neck of the One-humped camel were cut from the body

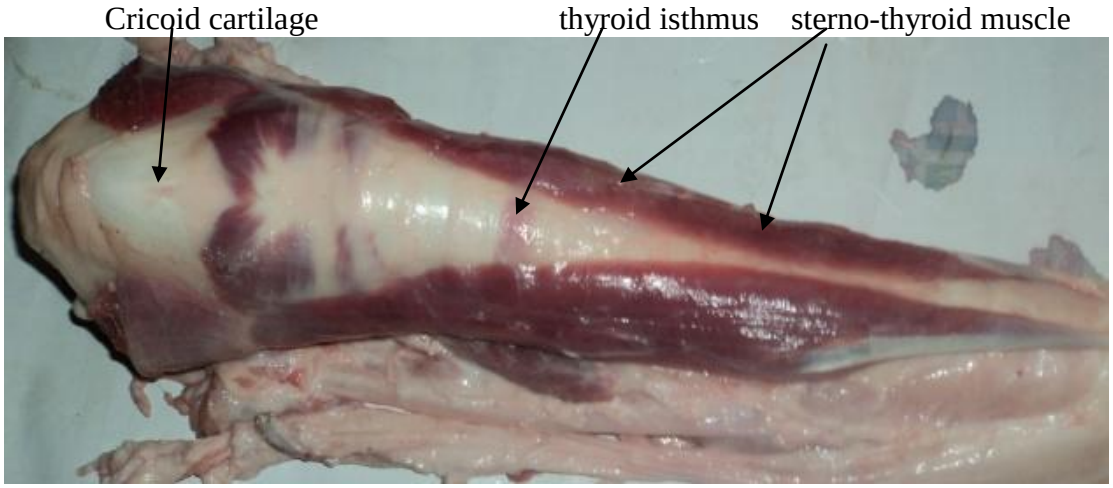


Plate 3.5: Sagital incision at the anterior neck exposed the neck muscles and reflected the ventral structures (trachea, the esophagus and the related muscles were seen).

Boarder between Thoracic and Abdominal regions



Plate 3.6: To access the glands, a cut on the border between the thoracic and abdominal region was made to remove the viscera and the glands

3.5 GROSS ANATOMICAL STUDIES:

Morphological studies: The glands were observed grossly to determine the shape, color, surfaces, borders and angles. The glands were then photographed in situ (Plate 4.1).

Morphometric Studies: The weights of the glands were obtained immediately after dissection, and linear measurement of their width, thickness and length were taken as shown in Plate 3.7 using digital vernier caliper.



Plate 3.7: Taking the length measurement of one of the thyroid lobes using digital vernier caliper

3.6 HISTOLOGICAL STUDIES

The glands were prepared for standard histological study using haematoxylin and eosin (H and E) for light microscopy, according to Bancroft and Gamble (2008).

The tissues were taken to grossing room of the Department of Histopathology, Aminu Kano Teaching Hospital (AKTH). Then the tissues were cut and mounted on blocks, and processed for H and E techniques through the following procedures:

Samples from the glands were dehydrated in two changes of graded series of ethanol (70%, 80%, 90%, 95% and 100%). The gradual process was employed to prevent the tissues from sudden shrinkages of the tissue which could distort the cells and consequently alter the results.

The tissues were transferred into two changes of Xylene for two hours for clearing process.

The tissue were then infiltrated with molten paraffin in an embedding mold and an oven set at 60^{o}c

Tissues were allowed to solidify to obtain a homogenous mass containing embedded tissues. The embedded tissues were then mounted on the blocks.

The mounted blocks were then affixed into a rotator microtome and trimmed into suitable sizes (5µ) before sectioning. The tissues were then transferred into a water bath of warm water.

Clean glass tubes coated with albumen were used to pick floating tissues from the water bath and kept in a clean and dry place. The slides were then passed through three changes of Xylene for complete removal of wax. They were then dehydrated by passing them through two changes of alcohol, starting from absolute alcohol, followed by graded alcohol (70%,

80%, 90%, 95% and 100%) for two hours. The tissues were then stained with haematoxylin and eosin stain for 25 minutes. The stained tissues were placed under gently running water to wash off. Following proper staining the tissues were again dehydrated by passing them through two changes of graded alcohol (70%, 80%, 90%, 95% and 100%) for two hours. The tissues were then cleared in Xylene. DPX was used as a mountant to mount the tissue on the slides. The mounted tissues were covered with cover slip and allowed to dry. The stained slides were examined using binocular light microscope at different magnification (X40, X100, X250 and X400). Photomicrographs of the slides were taken using Amscope digital camera attached to computer system via USB.

3.7 HISTOCHEMICAL STUDIES

For histochemical studies, the tissue blocks fixed using 10% neutral buffered formalin and prepared for H and E slides were used. In case of thyroid gland and parathyroid glands, tissue blocks were sectioned and stained with Periodic Acid Schiff (PAS) special stain to demonstrate glycogen and glycoprotein. The thyroid and adrenalin tissue blocks were also sectioned and stained with Acid Hydrolysis Azure A technique special stains specifically to demonstrate C-cells in thyroid. The hydrochloric acid in the solution hydrolysis precedes staining with Azure A and suppresses basophilia due to nucleic acids and any acid mucins present. Adrenalin secretin cells of the adrenal medulla are also stain to a varying degree (Cook, 1974). The adrenal gland tissue blocks were also sectioned and stained with Alcian blue special stains to show mucin in the adrenal cortex (Bancroft and Gamble, 2008).

Some adrenal gland tissues were fixed with potassium dichromate fixative for the demonstration of the chromaffin cells (Bancroft and Gamble, 2008). The fixation lasted for 24 hours after which tissues were processed. Tissues were washed in water and oxidized in 5% chromic acid for one hour after which they were washed in running water for one to two

minutes. Tissues were then treated with 2% sodium bisulphate to remove the chromic acid. Tissues were then washed in water for 5 minutes after which they were placed in methylamine silver for about three hours at 60^{o}C. Tissues were then toned in 0.2% gold chlorine solution for about two minutes then rinsed in distilled water. After which tissues were placed in 3% sodium thiosulphate for two minutes and were washed in running water for two to three minutes, sections of tissues were obtained and were counter stained with 0.2% acetic acid. Finally, sections were dehydrated, cleared and mounted in DPX.

Slides were studied on the binocular light microscope Olympus at different magnifications (X40, X100, X250 and X400). Photomicrographs of slides were taken using a microscope armscope digital camera attached to a computer system via USB.

3.8 STATISTICAL ANALYSIS

Data were expressed as mean ± standard error of the mean. Independent sample t-test were used to analyse the differences in morphometric variables between right and left lobes of the thyroid and right and left adrenal glands, as well as between the superior and inferior lobes in the right and left sides of parathyroids within each of the groups. Similarly, the differences between male and female were determined within groups A, B, and C. Comparison were also done between all males and females regardless of their ages.

Pearson's correlation analysis was carried out between morphometric measurements of weight, length and width of the glands with the concentration of the hormones. The level of significance was set as $P< 0.05$. The statistical analysis was done using student package for IBM SPSS version 20.0

CHAPTER FOUR

RESULTS

4.1 THYROID GLAND

4.1.1 Gross Anatomical studies

Gross studies of the thyroid gland of the one-humped camel indigenous to northern Nigeria showed that it was a paired-lobe organ located on the lateral surface of the trachea (Plate 4.1). The two lobes were joined together at inferior pole by an isthmus which passes on the ventral surface of the third (3^{rd}) to fifth (5^{th}) tracheal rings. It has lengthy lobes that extend from lower boarder of the cricoid cartilage (CC) and up to the fifth (5^{th}) tracheal ring (Plate 4.1).

The glands were seen as reddish brown organs (Plate 4.1). The thyroid gland lobes appeared triangular, and like a long shield in shape (Plate 4.2). The isthmus is thin and elongated (Plate 4.1). The blood supply is from superior, middle and inferior thyroid arteries as shown in Plate 4.3, which are direct branches from the external carotid or common carotid artery.

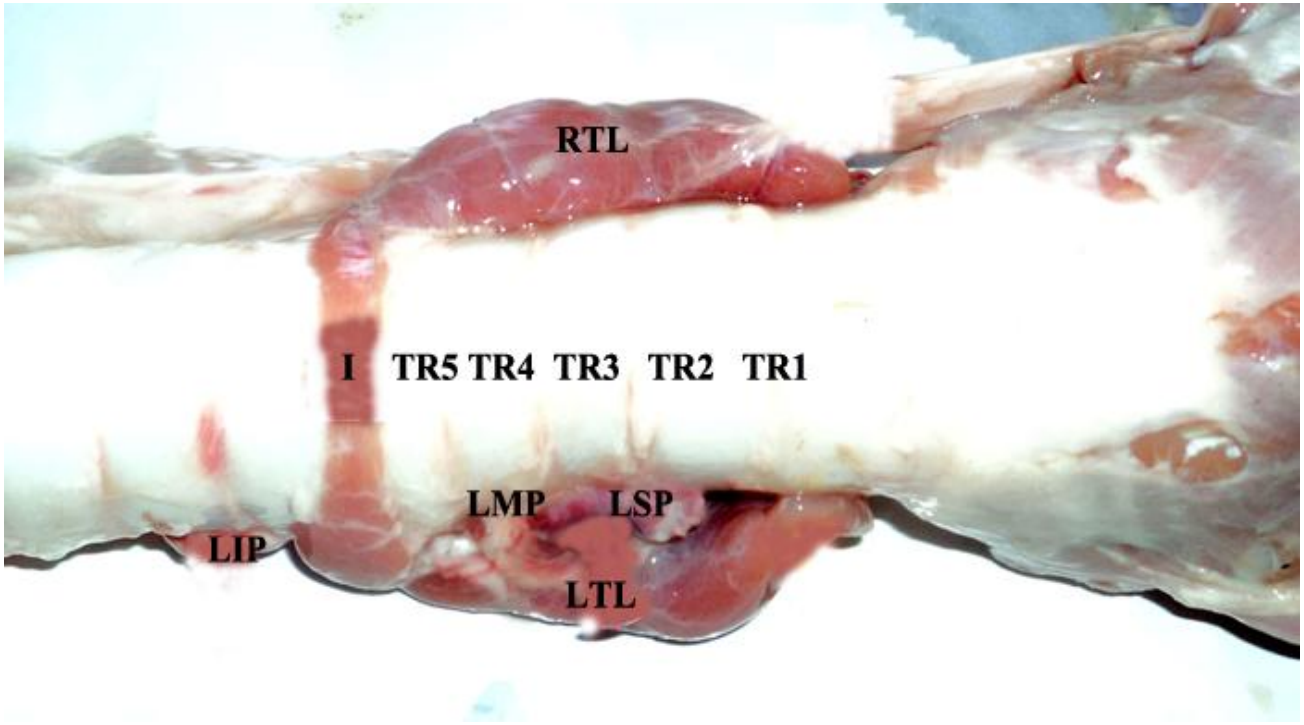
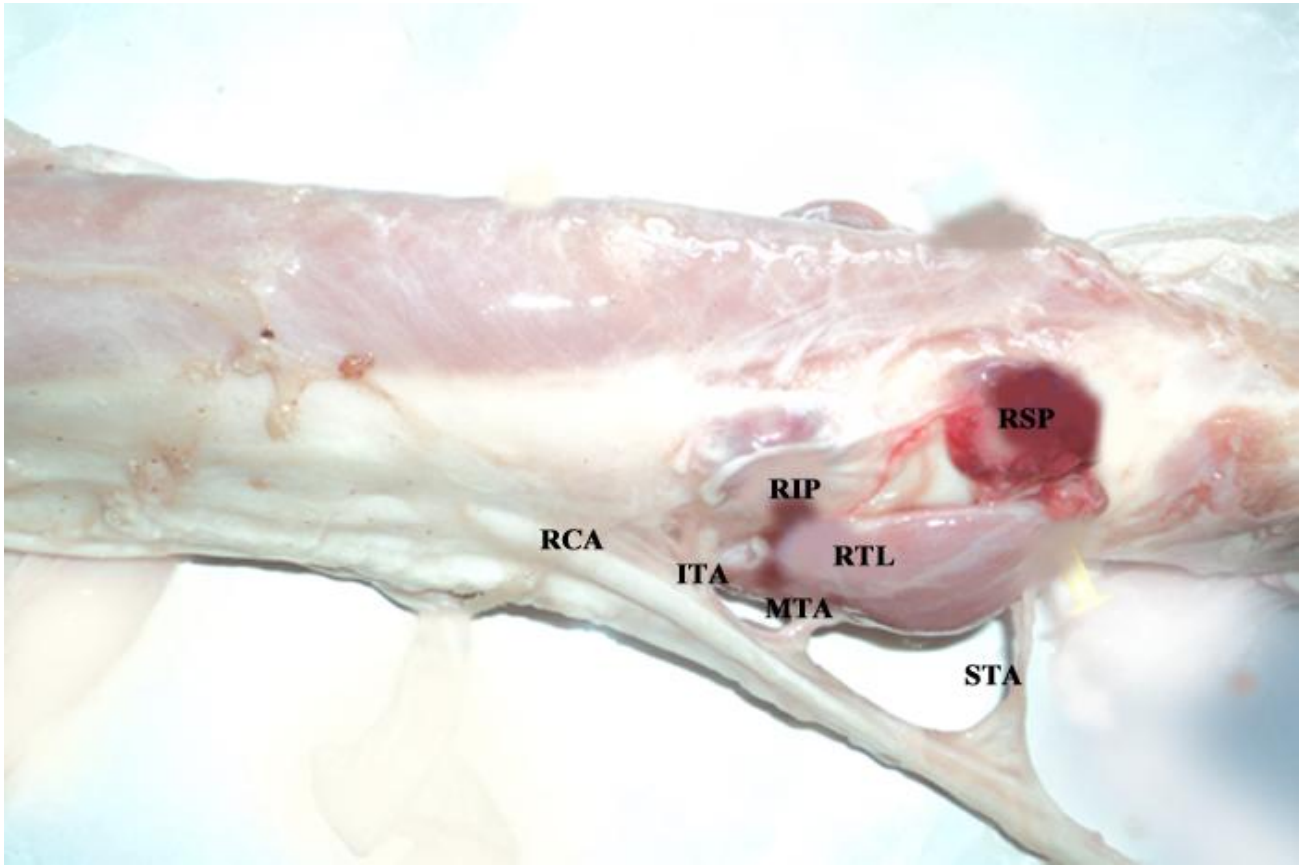


Plate 4.1: The thyroid gland and parathyroid glands of indigenous Camel dromedary. Note the location of the Isthmus (**I**), the right thyroid lobe (**RTL**), left thyroid lobe (**LTL**). Note also the left superio parathyroid (**LSP**) gland, left inferior parathyroid (**LIP**), and left middle parathyroid (**LMP**). **TR1** denoted the first tracheal ring, **TR2** second tracheal ring, **TR3** third tracheal ring, **TR4** fourth tracheal ring, and **TR5** fifth tracheal ring



Plate 4.2: The shape of the thyroid lobe (left) of indigenous one-humped camel is triangular



Pl ate 4.3: Blood supply to the thyroid and parathyroid glands; showing branches of right common carotid artery (**RCA**); inferior thyroid artery (**ITA**), middle thyroid artery (**MTA**), and superior thyroid artery (**STA**) in indigenous one-humped camel. N**ote** Right Superior Parathyroid (**RSP)**, Right Inferior Parathyroid (**RIP**) and Superior Thyroid Artery (**STA**).

4.1.2 Morphometric Studies of the Thyroid gland

The study showed that the morphometric variables expressed as the mean plus /or minus standard error of mean of the weight, length, width and thickness in the female on e-humped camel, among group A, between the age 1 – 4 years (juvenile), were: 34.48 ±3.64 g, 77.76± 2.99 mm, 29.03 ± 1.05 mm, and 11.93 ± .51mm, respectively (Table 4.1). The mean of the weight, length, width, and thickness of the male one-humped camel, among group A (juvenile), were also presented in Table 4.1. The mean of the weight, length, width, and thickness in male, between the ages 5 to 10 years (young adult or group B) were: 45.64±5.01mm, 93.10±7.93mm, 41.30±6.12mm, and 21.10±5.05mm, respectively (Table 4.1). Those of the female adult of the same group B were also presented (Table 4.1). The adult female one-humped camel, above the age of 10 years, among the group C, in the study, showed that the mean of the weight, length, width, and thickness were: 44.44 ± 3.91 mm, 86.64 ± 5.28 mm, 29.80 ± 1.24 mm, and 11.40 ± 0.40mm, respectively (Table 4.1), while those of males in the same group are shown also in Table 4.1.

The differences in the morphometric values of weight, length, width, and thickness of the thyroid gland in one-humped camel, between male and female within group A was determined and the result showed that there was sexual dimorphism in the length of thyroid gland of the one-humped camel significantly longer in female than the male (juvenile age) (Table 4.1). In the adult, the result showed there was differences in the morphometric values of weight, length, width, and thickness between the males and females which was not significant. There was sexual dimorphism in the thickness of the thyroid gland between males and females in group C, constituting one-humped camel above ten (10) years of age, with that of the male thicker than that of the female (Table 4.1).

The differences in the morphometric variables were compared between all the male and female in all the groups, regardless of their age, the result showed that there was no statistical significant difference (Table 4.2).

The thyroid glands have two lobes and the difference in morphometric variables between the lobes in each group was determined and the result showed that the differences were not significant.

Correlation between the thyroid gland morphometric variables (weight, length, width, and thickness) and the concentration of Thyroid Stimulating Hormone (TSH), Tetraiodothyronine (T4), and Triodothyronine (T3) hormones were also determined and showed that the correlation was highly significant (at $P<0.01$), between weight and length, weight and width, length and width, width and thickness, weight and Triodothyronine (T3) hormone, and between the Tetraiodothyronine (T4) and Triodothyronine (T3) (Table 4.4, Figure 4.1, 4.3 and 4.4). The correlation was also significant at $P<0.05$ between the thickness of the thyroid gland and tetraiodothyronine (T4) hormone (Table 4.4).

4.1.3 Hormonal Assay:

The concentrations of the Thyroid Stimulating Hormone (TSH), Tetraiodothyronine (T4), Triodothyronine (T3) hormones were in group A female were found to be 9.69 $\pm$8.43 µlu/ml, 7.11 $\pm$ 0.36 µg/dl, and 1.92 $\pm$ 0.17 ng/ml, respectively (Table 4.5), and that of male in the same group A is shown (Table 4.5). In group B male the TSH, T4, and T3 concentrations were: 7.62 $\pm$ 4.45 µlu/ml, 8.21$\pm$0.57 µg/dl, and 1.76$\pm$0.24 ng/ml, respectively (Table 4.5), while that of the female, in the same group B, is shown also in Table 4.5. The hormonal concentration of TSH, T4, and T3 in group C female are 15.40 $\pm$8.12 µlu/ml,

6.25±0.74 µg/dl, and 1.22±0.14 ng/ml respectively (Table 4.5). The male hormonal concentrations in the group C are shown in the same Table 4.5.

The differences of the concentrations of the TSH, T4, and T3 between the males and females within each of the groups (A, B, and C) were determined and found not to be statistically different.

However, correlation analysis have shown strong correlation ($P<0.01$), of T4 and T3 hormones between all the groups (A, B, and C), regardless of their sex (Table 4.4, Figures 4.3 and 4.4)

Table 4.1 Morphometric Mean values (± SEM) of thyroid gland of the one-humped camel within the groups A, B, and C.

	GROUP A		GROUP B		GROUP C	
	Mean ± SEM		Mean ± SEM		Mean ± SEM	
VARIABLES	**Female (n=5)**	**Male (n=5)**	**Female (n=5)**	**Male (n=5)**	**Female (n=5)**	**Male (n=5)**
Weight (g)	34.48 ± 3.64	27.04 ±4.39	49.09 ± 6.97	45.64 ± 5.01	44.44 ±3.91	53.30±7.02
Length (mm)	77.76 ± 2.99**	65.63 ± 4.51	89.55±3.50	93.10±7.93	86.64±5.28	86.90±6.26
Width (mm)	29.03 ± 1.05	25.06 ± 2.62	37.71 ± 1.66	41.30±6.12	29.80±1.24*	35.20±1.39*
Thickness (mm)	11.93 ± 0.51	11.14 ± 0.95	14.27 ± 1.14	21.10 ±5.05	11.40 ±0.40	10.80 ± 0.97

Analysis by Students T-test between Female and Male One-humped Camel among the age groups. Data was expressed in mean ± standard error of mean (SEM) of values collected.

*=P< 0.05 (difference is significant between male and female)

**= P < 0.01(difference significant between Male and Female)

Table 4.2 Morphometric mean values (±SEM) of mean of thyroid gland of the one-humped camel differences between male and female in all the groups (A, B, and C) regardless of their age.

	Mean ± SEM	
Variables	**Female (n=15)**	**Male (n=15)**
Weight (g)	42.67±3.16	41.99±4.20
Length (mm)	84.65±2.54	81.89±4.64
Width (mm)	32.18±1.27	33.85±2.76
Thickness (mm)	12.54±.52	14.35±2.06

Analysis by Students T-test between Female and Male One-humped Camel in all the age groups. Data is expressed in mean ± standard error of mean (SEM) of values collected.

Table 4.3 Comparison between right and left lobes of thyroid gland in indigenous one-humped camel within each of the groups (A, B, and C)

	Mean ± SEM					
	Group A		**Group B**		**Group C**	
Variables	**Right Lobe (n=10)**	**Left Lobe (n= 10)**	**Right Lobe (n=10)**	**Left Lobe (n= 10)**	**Right Lobe (n=10)**	**Left Lobe (n= 10)**
Weight (g)	14.43±1.41	15.37±1.54	24.06 ± 2.91	22.99 ± 1.57	24.17 ± 1.96	24.66 ± 2.28
Length (mm)	70.82±3.26	72.58±3.76	92.59 ± 4.85	90.08 ±3.95	85.30 ± 3.74	88.10 ± 4.67
Width (mm)	28.66±1.95	25.42±1.43	33.56 ± 1.51	45.21 ±6.01	33.10 ± 1.54	32.40 ± 1.485
Thickness (mm)	11.88±0.68	11.20±0.53	13.94 ± 0.96	21.49 ± 5.60	11.00 ± 0.683	11.00 ± 0.730

Analysis by students T-test between right and left lobes of thyroid gland within each age group. Data is expressed in mean ± standard Error of mean (SEM) of values collected.

Table 4.4 Correlation of thyroid gland morphometric variables and hormone concentrations of TSH, T4, and T3

Variables	Length (mm)	Width (mm)	Thickness (mm)	TSH (µlu/ml)	T4 (µg/dl)	T3 (ng/ml)
Weight (g)	0.655**	0.528**	0.211	-0.353	-0.193	**-0.494****
Length (mm)		0.812**	0.665**	-0.295	0.085	**-0.251**
Width (mm)			0.847**	-0.289	0.212	**-0.142**
Thickness(mm)				-0.211	0.378*	**0.022**
TSH (µlu/ml)					-0.291	**0.130**
T4 (µg/dl)						**0.558****
T3 (ng/ml)						

Pearson Correlation

*= Correlation is significant at the P<0.05 level (2-tailed).

** =Correlation is significant at the P<0.01 level (2-tailed).

TSH = Thyroid Stimulating Hormone

T4= Tetraiodothyronine

T3= Triiodothyronine

Table 4.5 Mean values (± SEM) of hormonal concentrations of thyroid hormones (thyroid function test) of the one-humped camel within the Groups A, B, and C to determine sexual dimorphism.

	Mean ± SEM					
	GROUP A		**GROUP B**		**GROUP C**	
VARIABLES	**Female (n=5)**	**Male (n=5)**	**Female (n=5)**	**Male (n=5)**	**Female (n=5)**	**Male (n=5)**
TSH (µlu/ml)	9.69 ±8.43	9.07 ±6.86	-1.45 ± 0.25	7.62 ± 4.45	15.40 ±8.12	-1.00± 0.69
T4 (µg/dl)	7.11 ± 0.36	7.71 ± 0.57	7.80±0.63	8.21±0.57	6.25±0.74	6.41±0.53
T3 (µg/dl)	1.92 ± 0.17	8.21 ± 0.57	2.34 ± 0.59	1.76±0.24	1.22±0.14	1.04±0.09

Analysis by t-test to determine sexual dimorphism in One-humped Camel among the age groups (A, B, and C). Data is expressed in mean ± standard Error of mean (SEM) of values collected.
*****= $P < 0.05$(Significant between Male and Female)

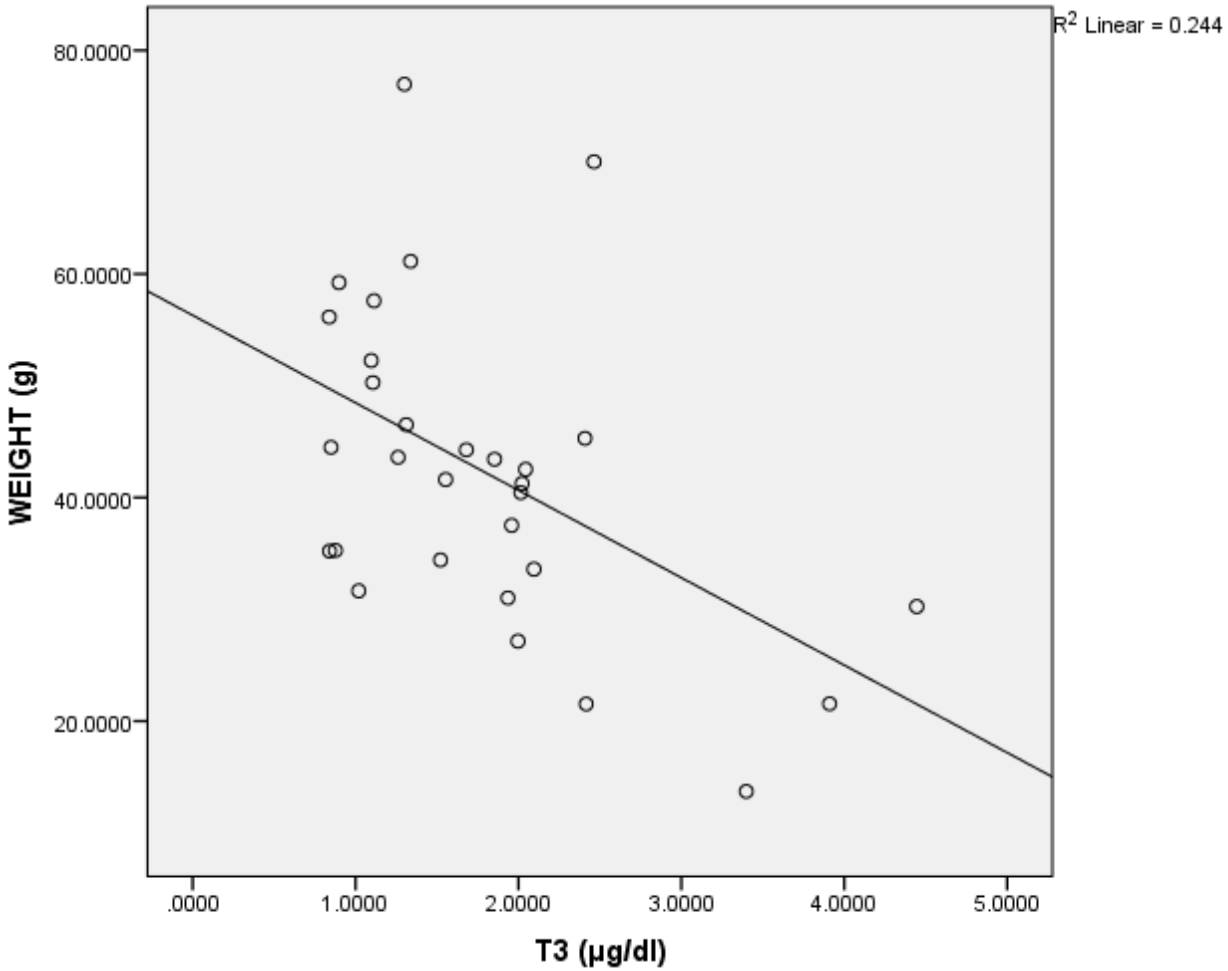


Figure 4.1 Scattergram graph showing strong correlation between the Weight of thyroid gland against the concentration of Triodothyronine (T3). Note the relationship is inverse.

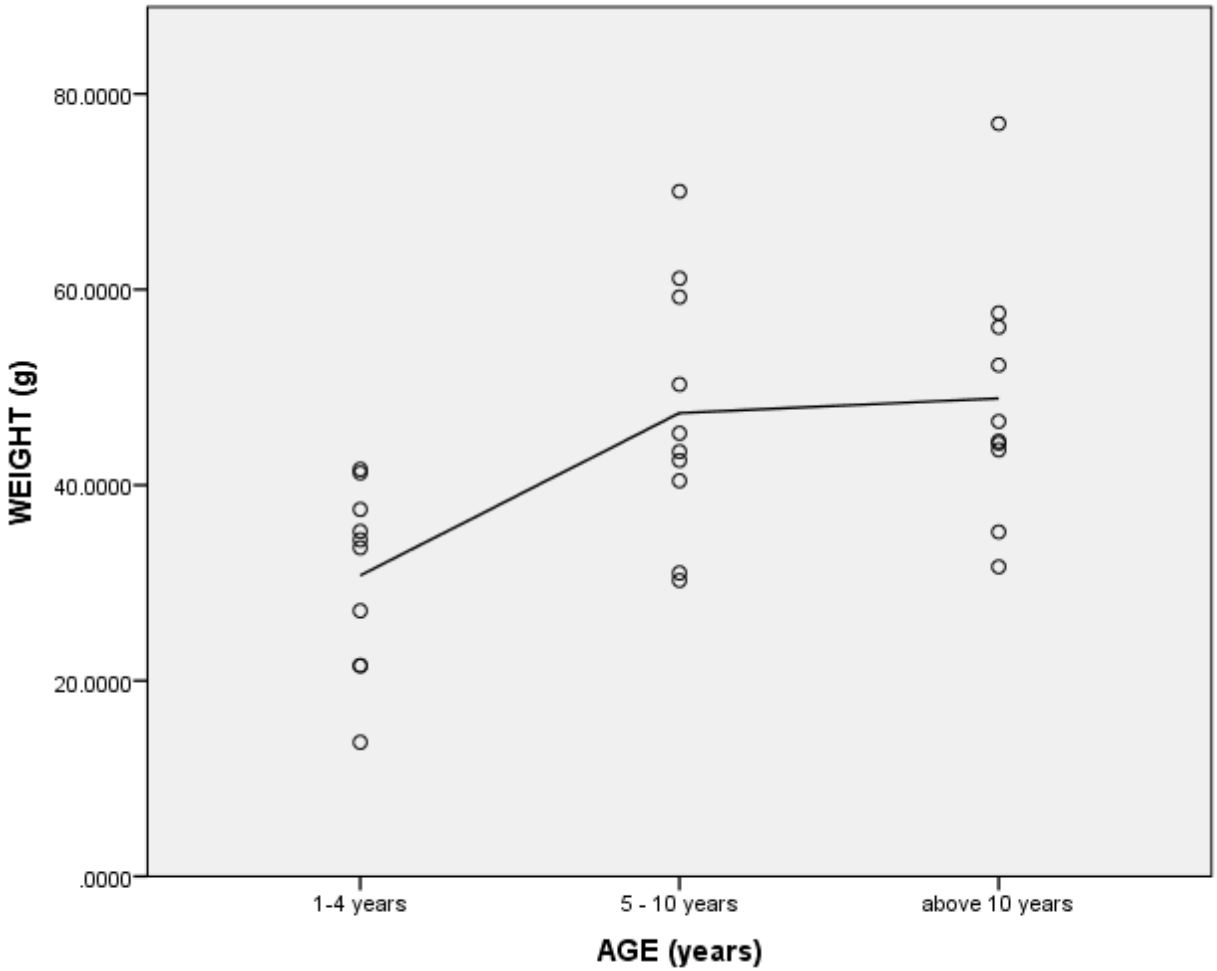


Figure 4.2 Scatter gram graph showing strong correlation between the weights of thyroid gland against the respective age in groups of the animals. **Note** the line is straight upward (directly proportional) then it becomes constant

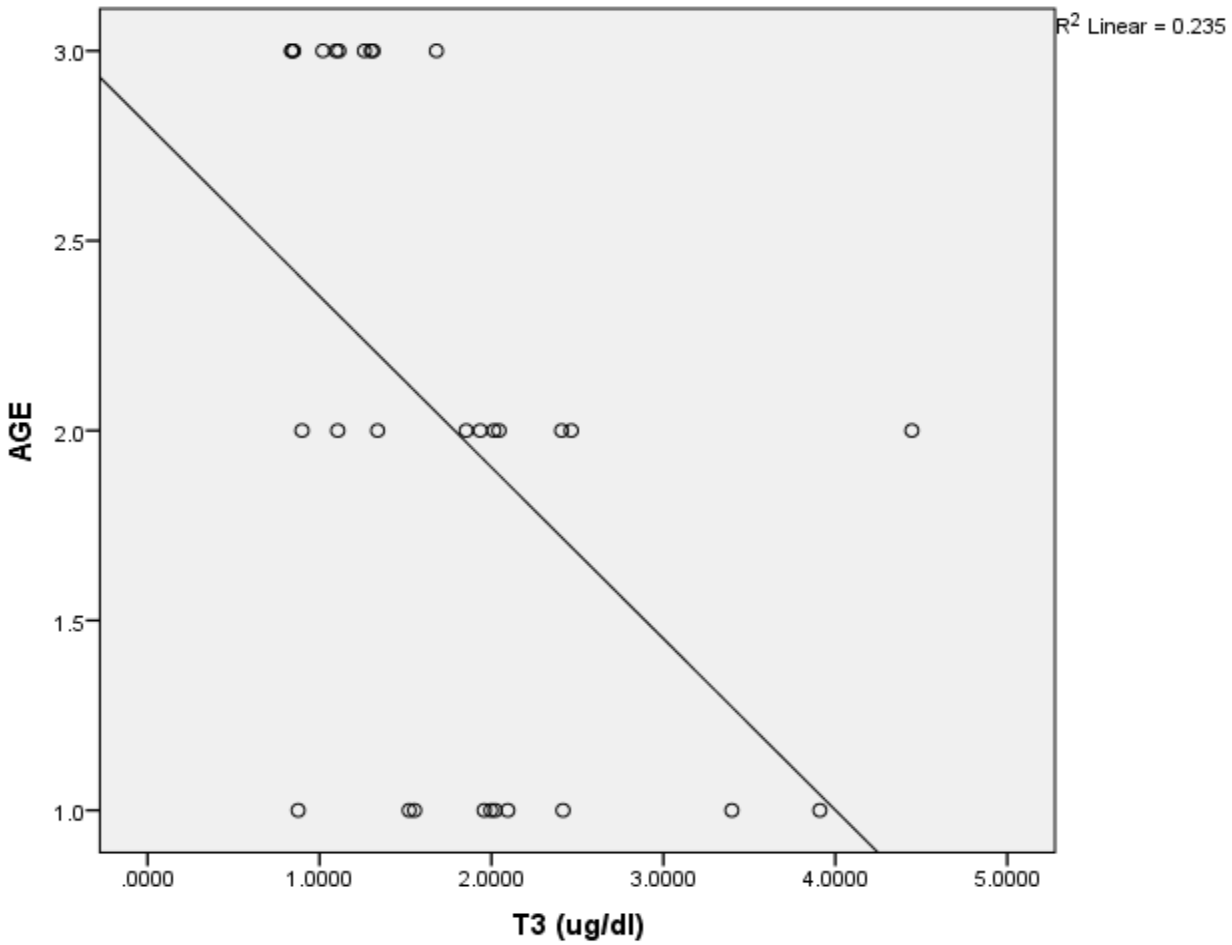


Figure 4.3: Scatter plot graph showing correlation of age with T3 concentrations.
Note the correlation between the **Triiodothyronine (T3)** and the Age are inversely proportional

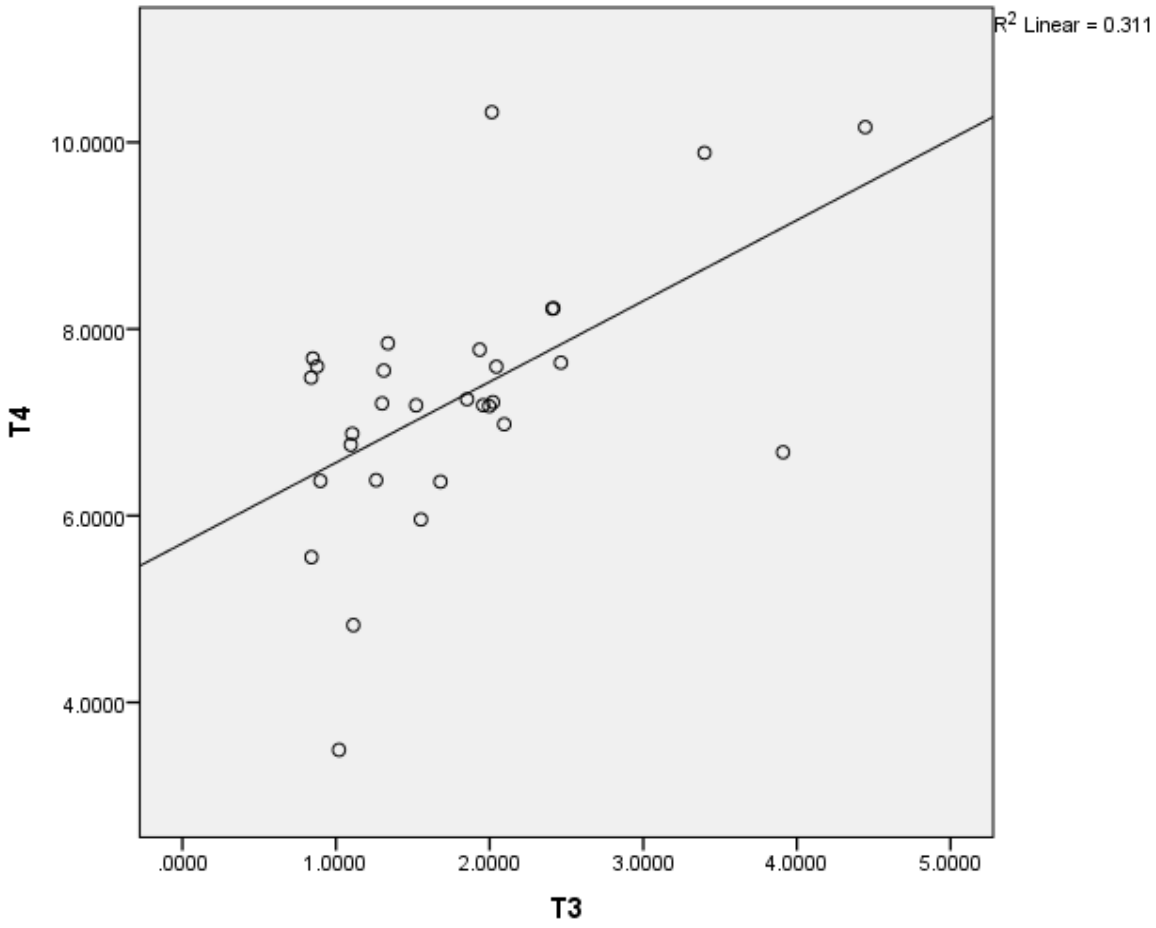


Figure 4.4: Scatter plot graph showing correlation of T4 concentration to T3 concentration. Note the correlation between the **Tetraiodothyronine (T4),** and **Triiodothyronine (T3)** is directly proportional.

4.1.4 Histological and histochemical studies of the thyroid gland

Group A of the study comprised of ten (10) indigenous one-humped camels (with equal number of female and male) between the age of 1- 4 years (juvenile age).

The results of the histology and histochemical studies were as follows:

The result of H&E stained thyroid gland tissue slides revealed that the thyroid gland tissue is surrounded by a thin capsule of connective tissue, which invade the parenchyma and provide a passage, by thin trabeculae**,** for the rich capillary plexus/ sinusoids (Plate 4.5), and divided it into lobules (Plates 4.4, 4.5, and 4.6)**.** The sinusoids were seen more prominent and numerous in female of group A than in male of the same group (campare Plates 4.5 and 4.6). This intralobular connective tissue consists of numerous follicles of various sizes (Plate 4.4) that filled with colloid (Plates 4.4, 4.5, and 4.6), with bigger follicles situated towards the periphery more in female of group A than in the male (Plate 4.4). The shapes of the follicles were round, oval and irregular (Plate 4.4).

Each follicle is made up of simple follicular epithelial cells that encircled it (Plates 4.4, 4.5, and 4.6), and consist of parafollicular cells as well (Plate 2). The follicular and parafollicular cells rest on a thin basal lamina**,** but are separated from the thin trabeculae that surrounds the follicles/ lobules (Plate 4.5)**.** Follicular cells of neighboring follicles may come into contact with each other and disrupt the continuity of the basal lamina and seen within the follicles/ lobules, and more prominent in male than in female of group A (compare Plates 4.4 and 4.5)

The follicles are mostly inactive and filled with colloids and had smoother peripheral surface of the follicles in both male and female of group A (Plates 4.4, 4.5, and 4.6). The follicular

cells are low cuboidal in shape (Plates 4.5 and 4.6), and with very few vacuolated follicles seen only at the periphery in female (Plate 4.4).

Parafollicular cells or C cells occurred among the follicular cells and between them (Plates 4.5 and 4.6). They were larger and have a paler cytoplasm than the follicular cells, and occur singly, and in groups (Plates 4.5 and 4.6). This is virtually the same in both female and male.

Special histochemical studies revealed that the colloid in the follicles were negative to Periodic Acid Schiff (PAS)**.** The PAS slides did not stained different from the H&E (Plate 4.8). No difference between the sexes of the same group.

A special histochemical technique was used in order to differenciate parafollicular cells from the follicular with some degree of certainty, and Azure **A Acid Hydrolysis** was utilized which deeply stained the parafollicular cells purple blue (Plate 4.8)

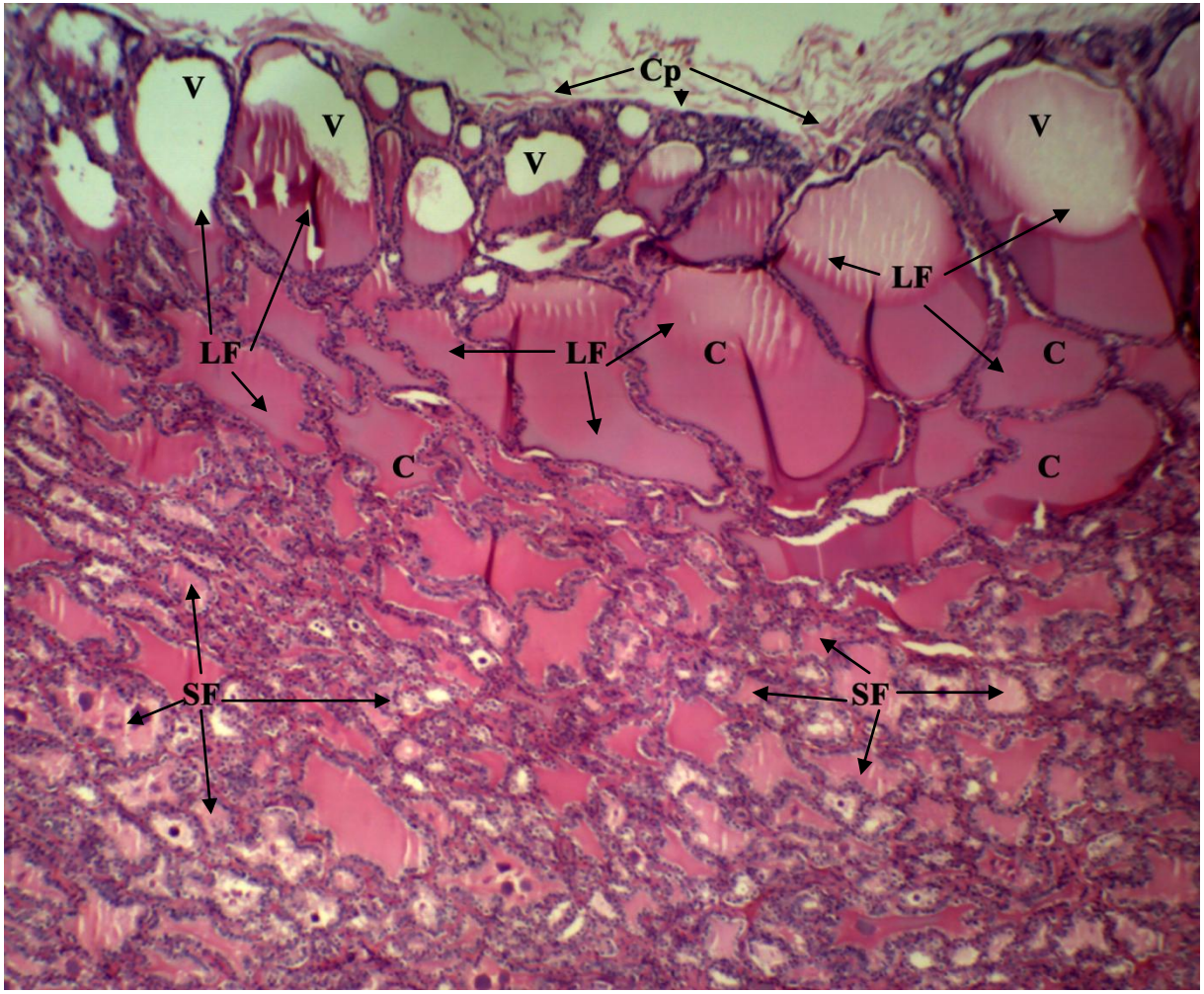


Plate 4.4: Photomicrograph of the thyroid gland tissue **(Transverse Section)** of female indigenous one-humped camel of **group A,** showing the basic structures of the thyroid tissues. Note the Capsule (**Cp**). Follicles of different sizes, Large follicles (**LF**) at the periphery, and small follicles (**SF**) surrounding the Colloids (**C**). The vacuoles (**V**) in some colloids, vacuolization of the colloids (H&E × 40).

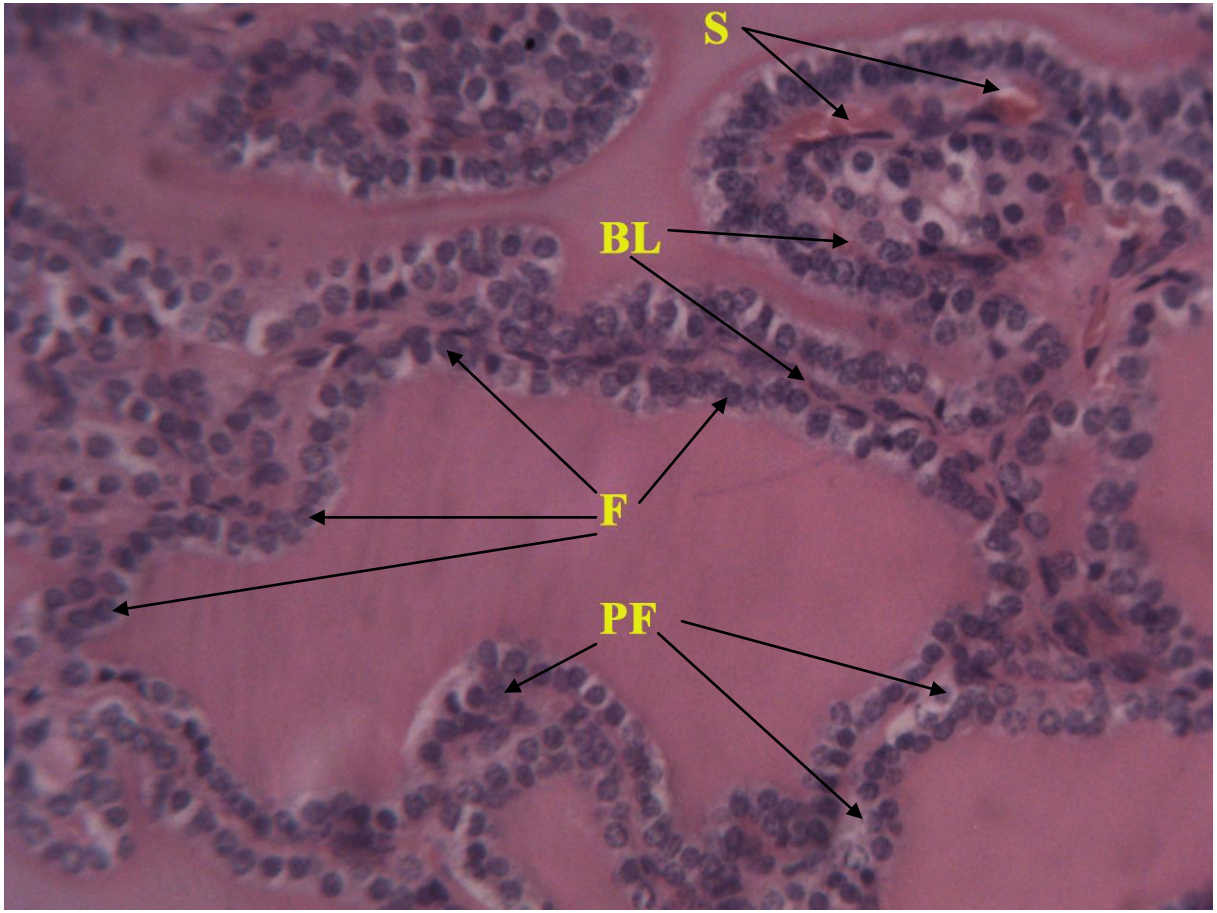


Plate 4.5: Photomicrograph of thyroid gland tissue **(Transverse Section)** of female indigenous one-humped camel of group A. Note the Follicular cells (**F**) laying on basal lamina (**BL**), and the Parafollicular cells (**PF**). There are lots of sinusoids (**S**) around the basal lamina (**BL**) (H&E × 250).

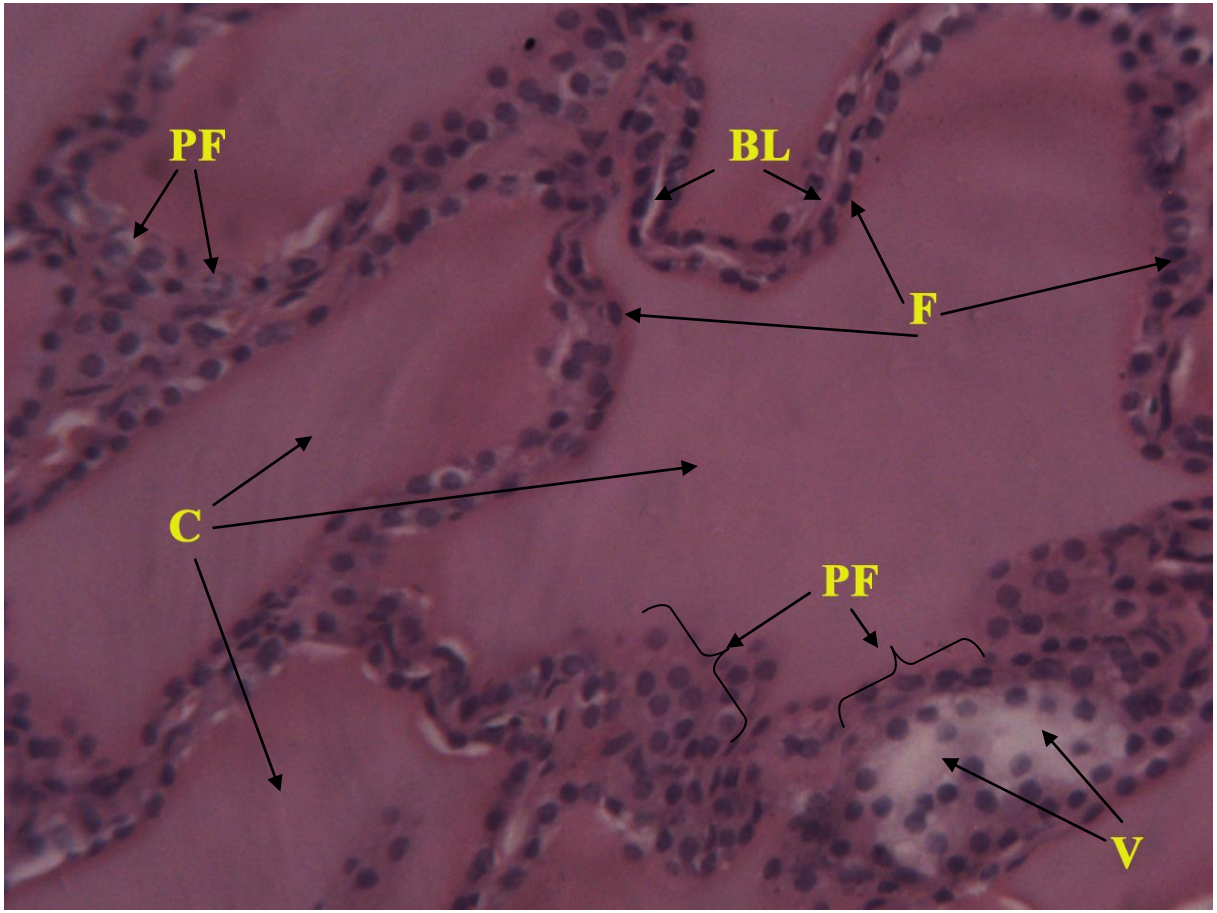


Plate 4.6: Photomicrograph of thyroid gland tissue **(Transverse Section)** of **male** indigenous one-humped camel in group A. Note the follicles surrounded by follicular cells (**F**), filled with colloids (**C**). The parafollicular cells (**PF**) here appeared in groups and sometimes in between the follicular cells. The vacuoles were little. The basal lamina (**BL**) upon which the follicular cells rest also separated the follicles (H&E × 250).

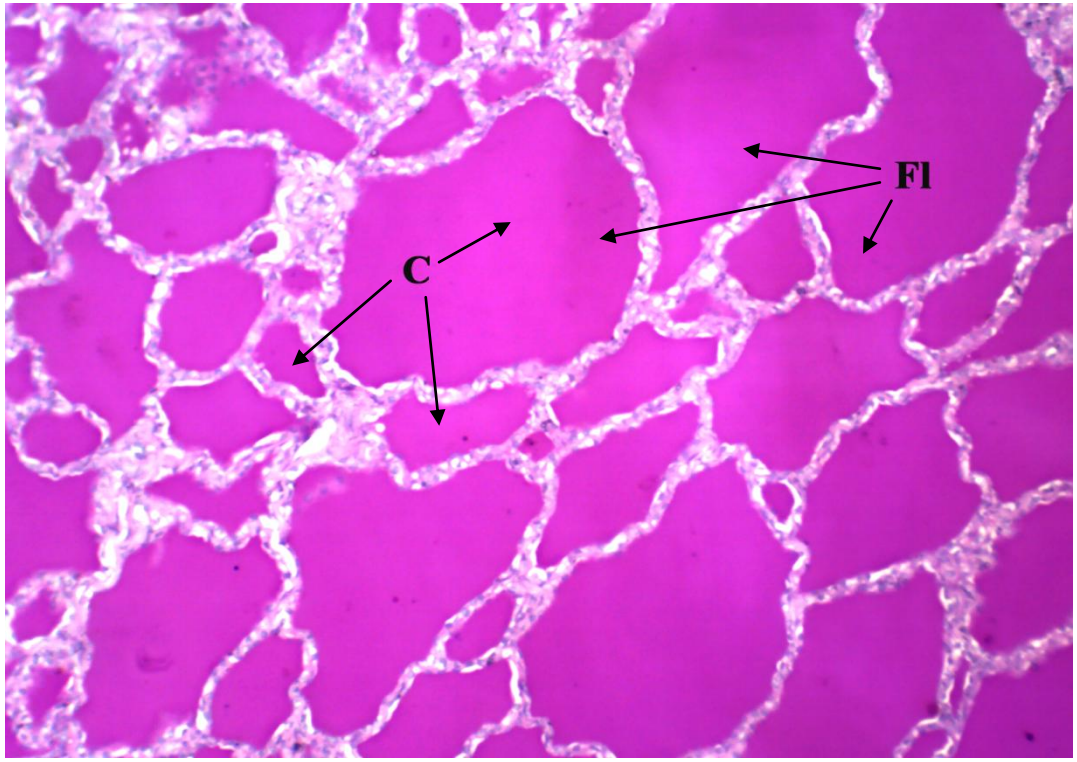


Plate 4.7: Photomicrograph of thyroid gland tissue **(Longitudinal Section)** of **male** indigenous one-humped camel in group A. Note the colloid (**C**) within the follicles (**Fl**) reacted negative to the Periodic Acid Schiff (PAS) reagent (PAS × 100).

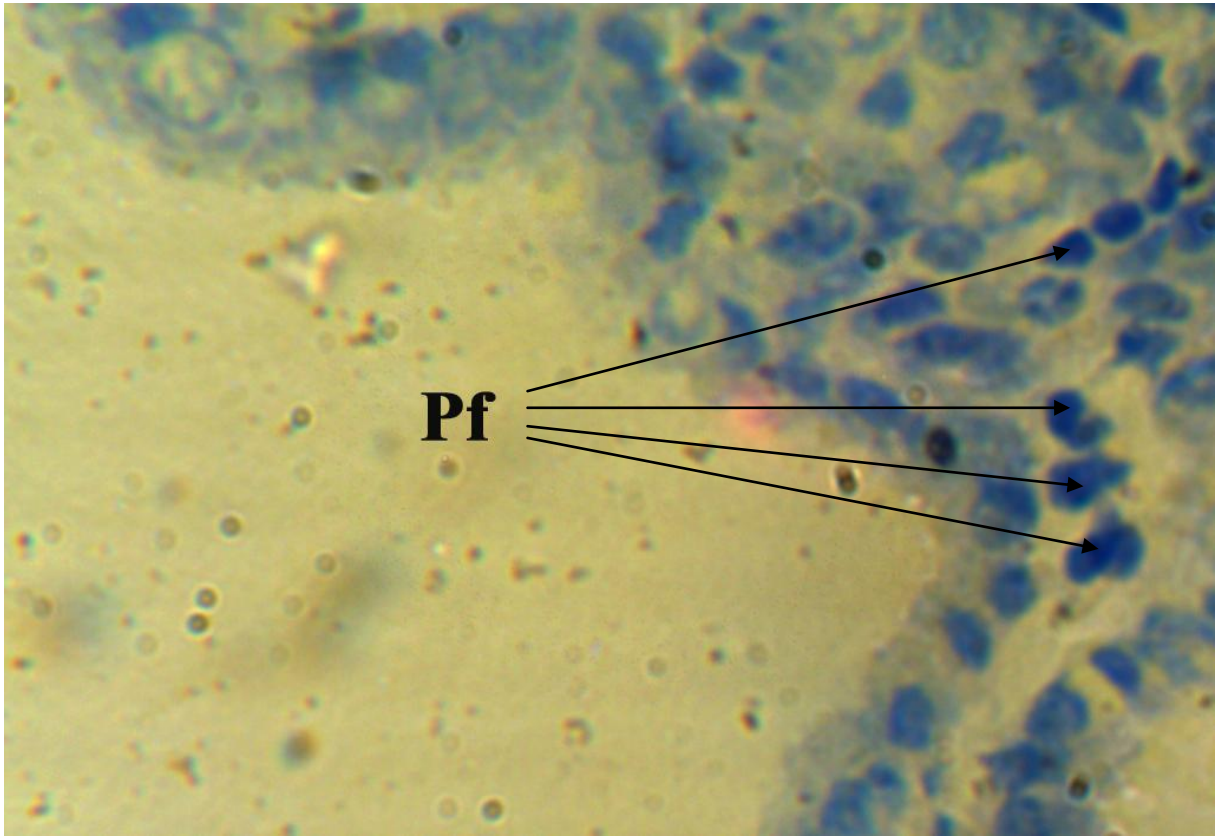


Plate 4.8: Photomicrograph of thyroid gland tissue **(Longitudinal Section)** of **male** indigenous one-humped camel in group A, demonstrating the parafollicular cells (**Pf**) that stained purple blue well with Azure A Acid Hydrolysis technique (Azure × 450).

Group B of the study consists of ten (10) equal number of female and male indigenous one-humped camels between the age of 5- 10 years (young adult).

The result of the histology study is as follows:

The result of the thyroid gland tissue slides stained with Haematoxylin &Eosin (H&E) revealed that the tissue is surrounded by a thin **capsule** of connective tissue, which invade the parenchyma and provide a passage, by thin **trabeculae,** for the rich capillary plexus/ sinusoids (Plate 4.9), and divided it into **lobules**, which contain follicles (Plates 4.9 and 4.10)**.** The sinusoids were seen more prominent and numerous in female of group B, but it was also present in males of the same group (Plates 4.9 and 4.10). There are **follicles** of various sizes (Plate 4.9) with some filled with **colloid,** while some **vacuolated** (Plate 4.9). The follicles with vacuoles were, mostly, bigger occupied more than half of the tissue from the periphery in male of group B than in the female (Plates 4.9 and 4.10). There are vacuoles in the follicles scattered more in female than in male. Most of the bigger follicles at the periphery are empty or vacuolated in male of the group B (Plate 4.9). The shapes of the follicles were round, oval and irregular (Plate 4.9).

Each follicle is surrounded by simple **follicular** epithelial cells (Plate 4.9); which consist of **parafollicular** cells as well (Plates 4.9, 4.10 and 4.12). The cells rest on a thin **basal lamina,** but are separated from the thin trabeculae that surround the follicles/ lobules (Plate 4.9)**.** The follicular cells of the neighboring follicles may come into contact with each other and disrupt the continuity of the basal lamina and may be seen within the follicles/ lobules, and are more prominent in females than in males of group B (Plate 4.9).

The follicular cells are more columnar in shape in females while those of males are more cuboidal in shape (Plate 4.9 and 4.10). Some of the cells show some level of apoptosis and is more prominent in female than in male (Plate 4.10).

Parafollicular cells or C cells occured among the follicular cells and between them (Plates 4.10 and 4.12). They were **larger** and have a **paler cytoplasm** than the follicular cells and occur singly, as well as in groups (Plate 4.9 and 4.12).

Histochemical studies revealed that the colloid in the follicles were negative to **Periodic Acid Schiff (PAS)** (Plate 4.11)**.** The PAS slides did not stained different from the H&E. No difference was seen between the sexes of the same group.

A special histochemical technique was used also, in order to differenciate parafollicular cells from the follicular cells, to achieve certainty. Azure **A Acid Hydrolysis** was utilized which deeply stained the parafollicular cells purple (Plate 4.12)

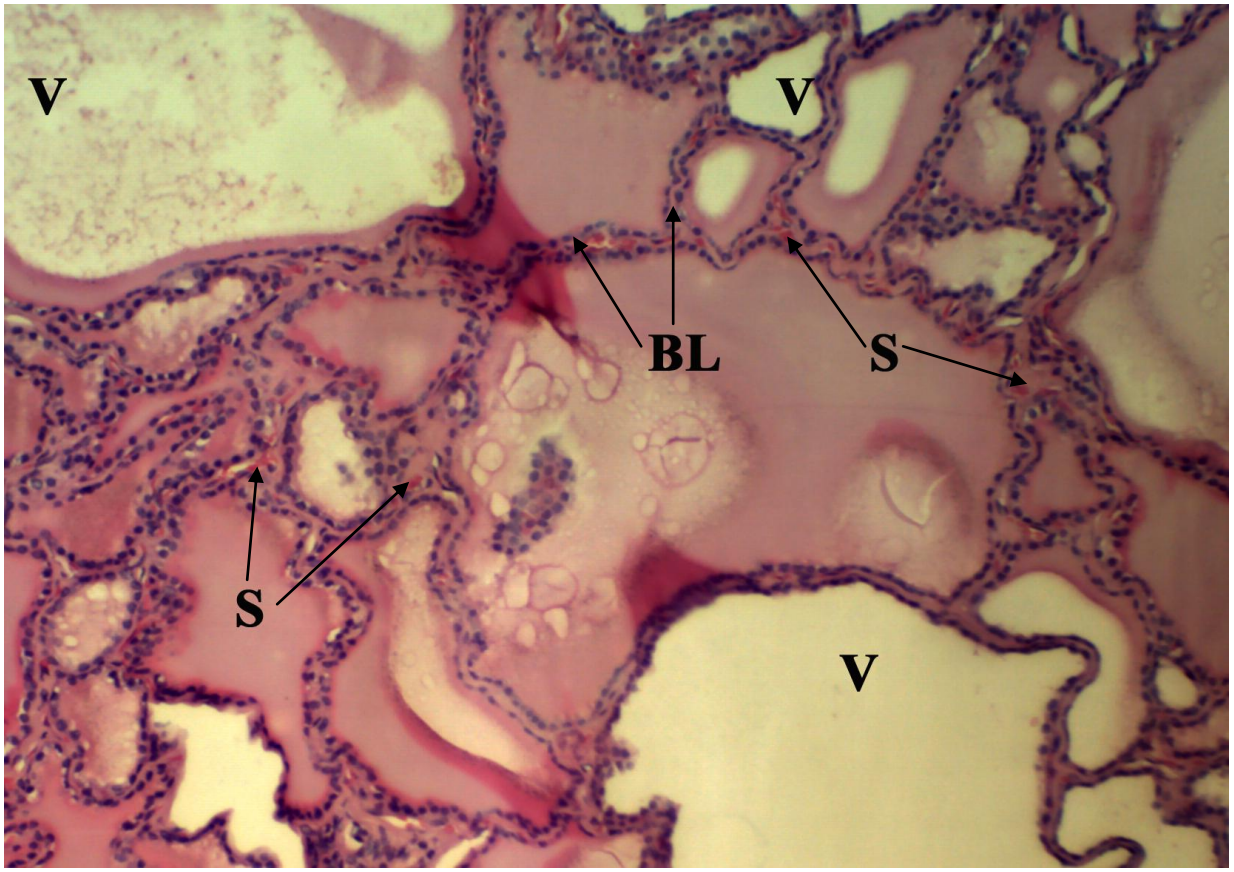


Plate 4.9: Photomicrograph of thyroid gland tissue **(Longitudinal section)** of **male** indigenous one-humped camel in group B. Note almost half of the tissue was vacuolated, the vacuoles (**V**) within the follicles, the basal lamina (**BL**) upon which the cells rested, sinusoid (**S**) of blood capillaries (H&E × 100).

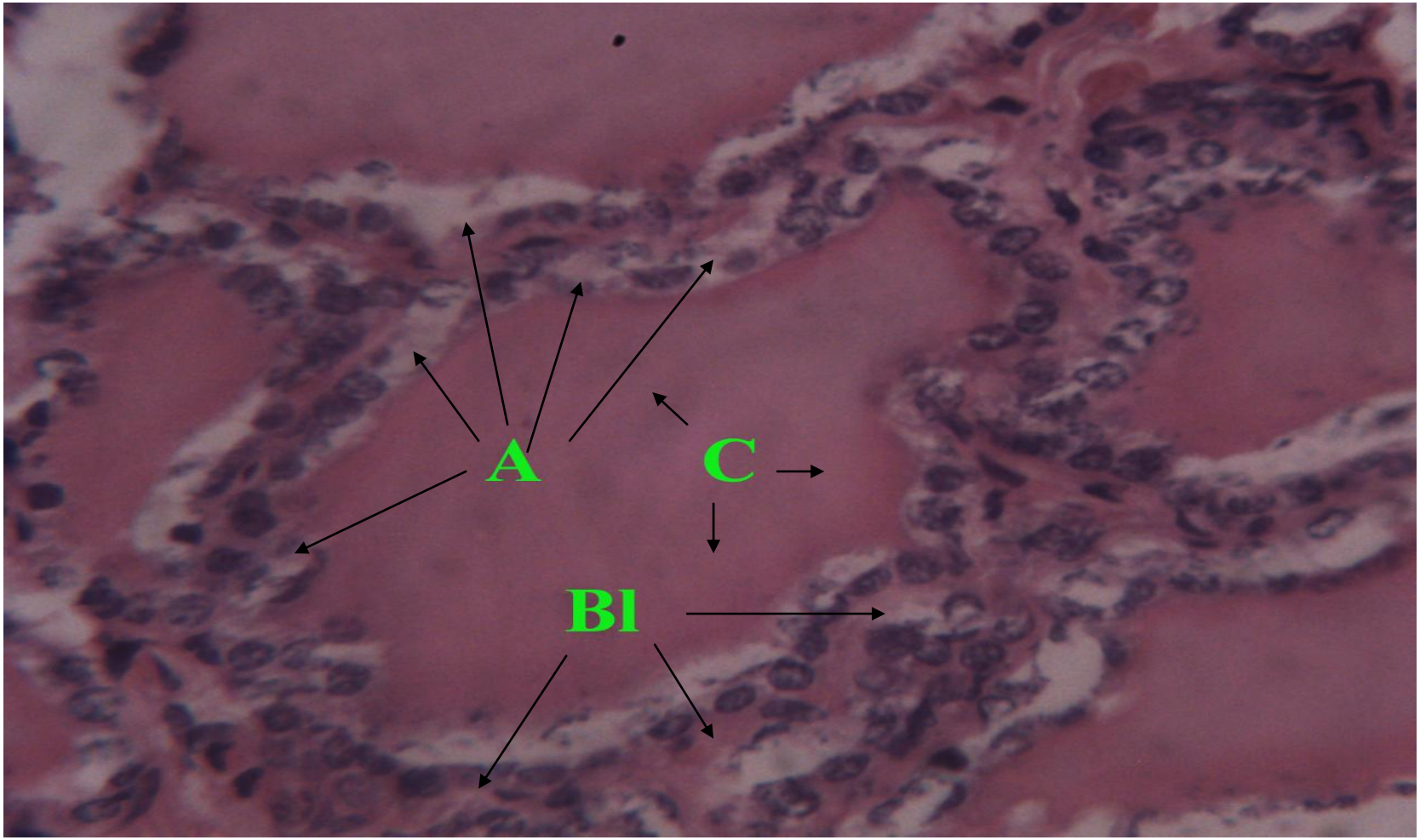


Plate 4.10: Photomicrograph of thyroid gland tissue **(Transverse section)** of **female** indigenous one-humped camel in group B. The thyroid cells are showing some level of **apoptosis (A)**. Some of the follicles were filled with colloids while some where vacuolated. **BL** is the basal lamina upon which the follicular cells lied.
(H&E × 400)

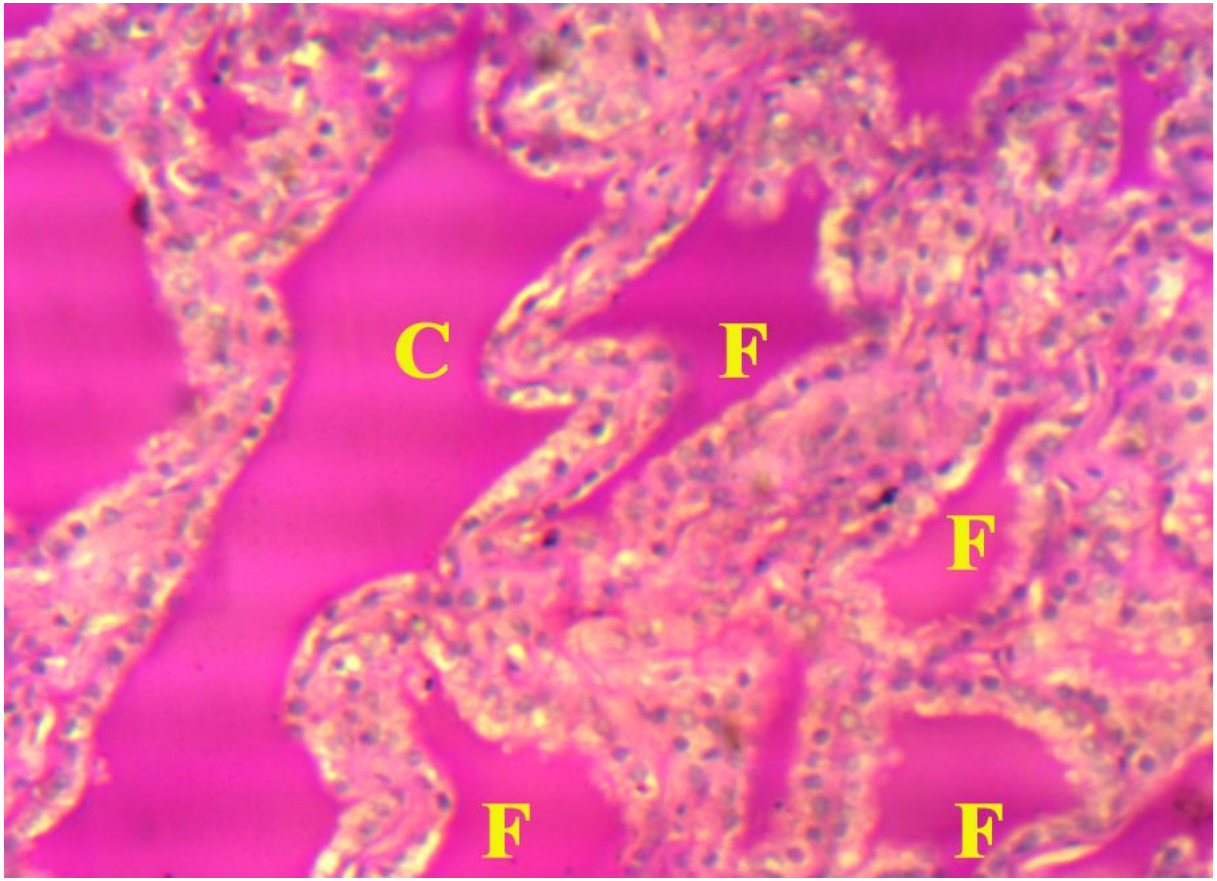


Plate 4.11: Photomicrograph of thyroid gland tissue **(Longitudinal section)** of **male** indigenous one-humped camel in group B. Colloid (**C**) contained within the follicles (**F**) reacted negative to the Periodic Acid Schiff reagent (PAS × 150).

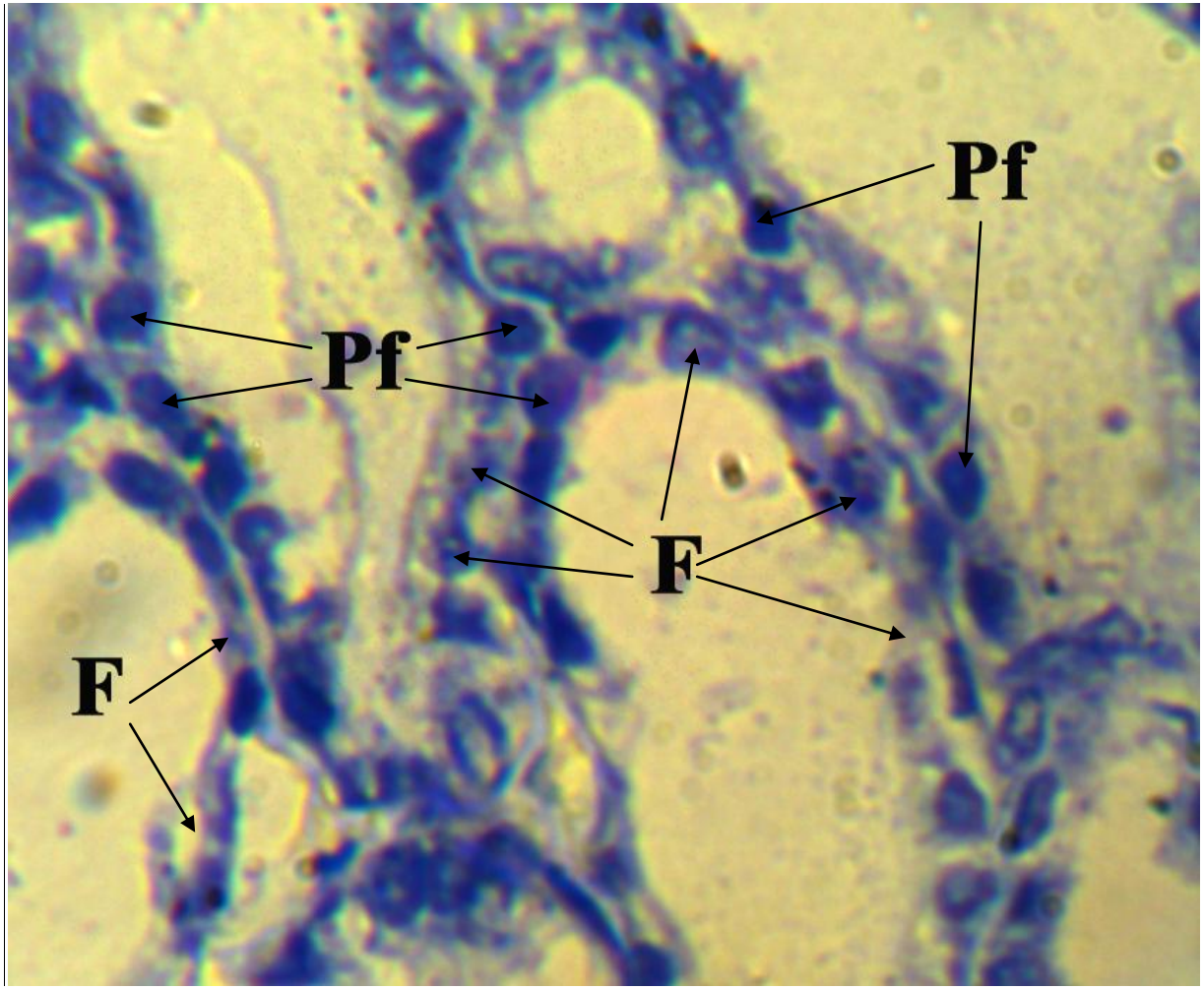


Plate 4.12: Photomicrograph of thyroid gland tissue **(Longitudinal section)** of **Male** indigenous one-humped camel in group B. Note how the parafollicular cells **(Pf)** nuclei were clearly and deeply stained purple blue by the Azure A acid hydrolysis technique, while the follicular cells were irregular and poorly stained (nucleus not clear) (Azure A ×450).

Group C of the study consists of ten (10) equal number of female and male indigenous one-humped camel above the age of 10 years (old adult).

The result of the histological study is as follows:

The result of the thyroid gland tissue slides stained with Heamatoxylin and Eosin (H&E) revealed that the tissue was surrounded by a thin **capsule** of connective tissue, which invade the parenchyma and provide a passage, by thin **trabeculae,** for the rich capillary plexus/ sinusoids (Plate 4.13), and divided it into **lobules** (Plates 4.13, 4.14, 4.15, and 4.16)**.** Blood vessels, specifically arteries, were seen more prominent and numerous in male of group C (Plate 4.13). They have **follicles** of various sizes scattered (Plates 4.13 and 4.14). Most of the follicles were **vacuolated** (Plates 4.13, 4.14, 4.15, and 4.16).The vacuoles are relatively more in male (compare Plates 4.13 and 4.14). The shapes of the follicles were round, oval and irregular (Plates 4.13 and 4.14), they are more irregular in male (Plates 4.13 and 4.14).

Each follicle is surrounded by simple **follicular** epithelial cells (Plates 4.13, 4.14, 4.15 and 4.16) and consist of **parafollicular** cells as well (Plates 4.14 and 4.16). The follicular and parafollicular cells rest on a thin **basal lamina,** but are separated from the thin trabeculae that surrounds the follicles/ lobules (Plates 4.13 and 4.14)**.** The follicular cells of the neighboring follicles may come into contact with each other and disrupt the continuity of the basal lamina and seen within the follicles/ lobules (Plates 4.13 and 4.14).

Some of the follicular cells look columnar while some look cuboidal in shape both in female and male (Plates 4.14 and 4.16). Some of the cells are undergoing **apoptosis** in both male and female (Plates 4.14 and 4.16).

Parafollicular cells or C cells occur among the follicular cells and between them (Plates 4.14 and 4.16). They were **larger** and have a **paler cytoplasm** than the follicular cells, and occur singly, as well as in groups (Plates 4.14 and 4.16).

Histochemical studies revealed that the colloid in the follicles reacted negative to Periodic Acid Schiff reagent **(**Plate 4.15**).** The PAS slides did not stained different from the H&E. No difference was seen between the sexes of the same group.

A special histochemical technique was used also, in order to differentiate parafollicular cells from the follicular cells, to achieve certainty. Azure **A Acid Hydrolysis** was utilized which deeply stained the parafollicular cells purple (Plate 4.16).

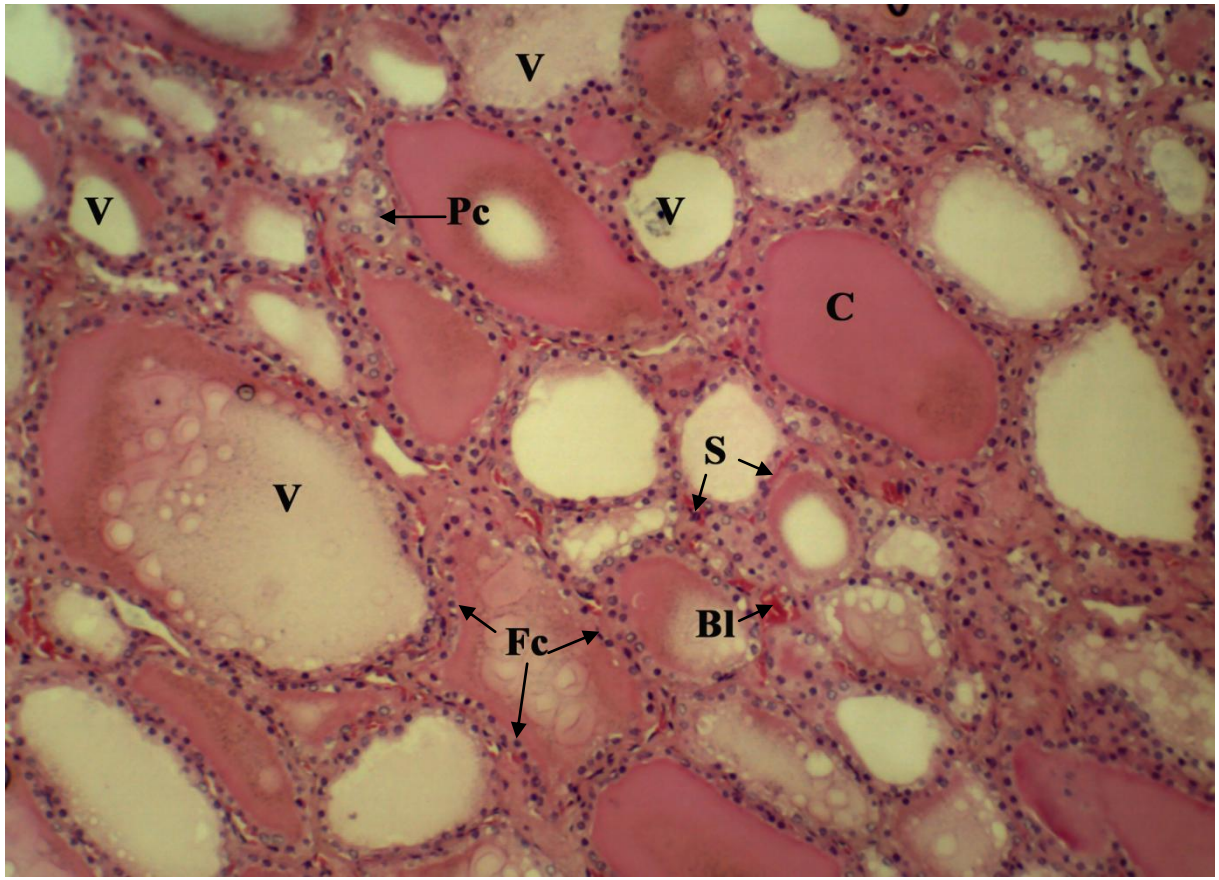


Plate 4.13: Photomicrograph of thyroid gland tissue **(Transverse section)** of **male** indigenous one-humped camel in group C. Note the spread of **vacuoles (V)** all over the tissue in the follicles. The follicles were formed by follicular cells (**Fc**), and parafollicular cells (**Pc**), and contain colloids (**C**), mostly vacuolated (**V**). The follicular cells (**Fc**) were based on basal lamina (**BL**) in this tissue were mostly bathed with sinusoids (**S**) or blood capillaries all over the tissue in these group (H&E × 100).

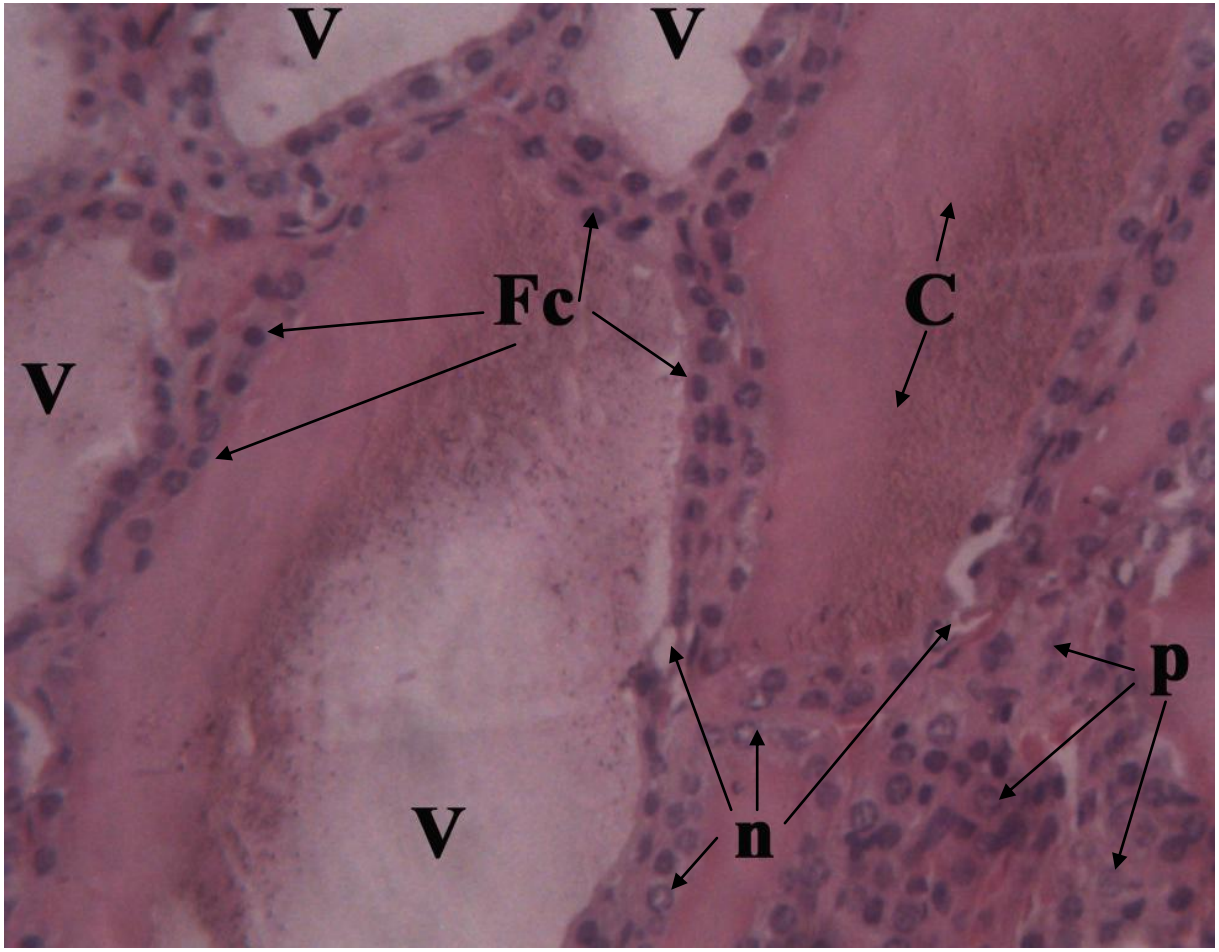


Plate 4.14: Photomicrograph of thyroid gland tissue **(Transverse section)** of **female** indigenous one-humped camel in group C. Note the follicles, the follicular cells (**Fc**) showing a little of apoptosis (**n**), the parafollicular cells (**P**). The follicles have vacuoles (**V**) (H&E × 250).

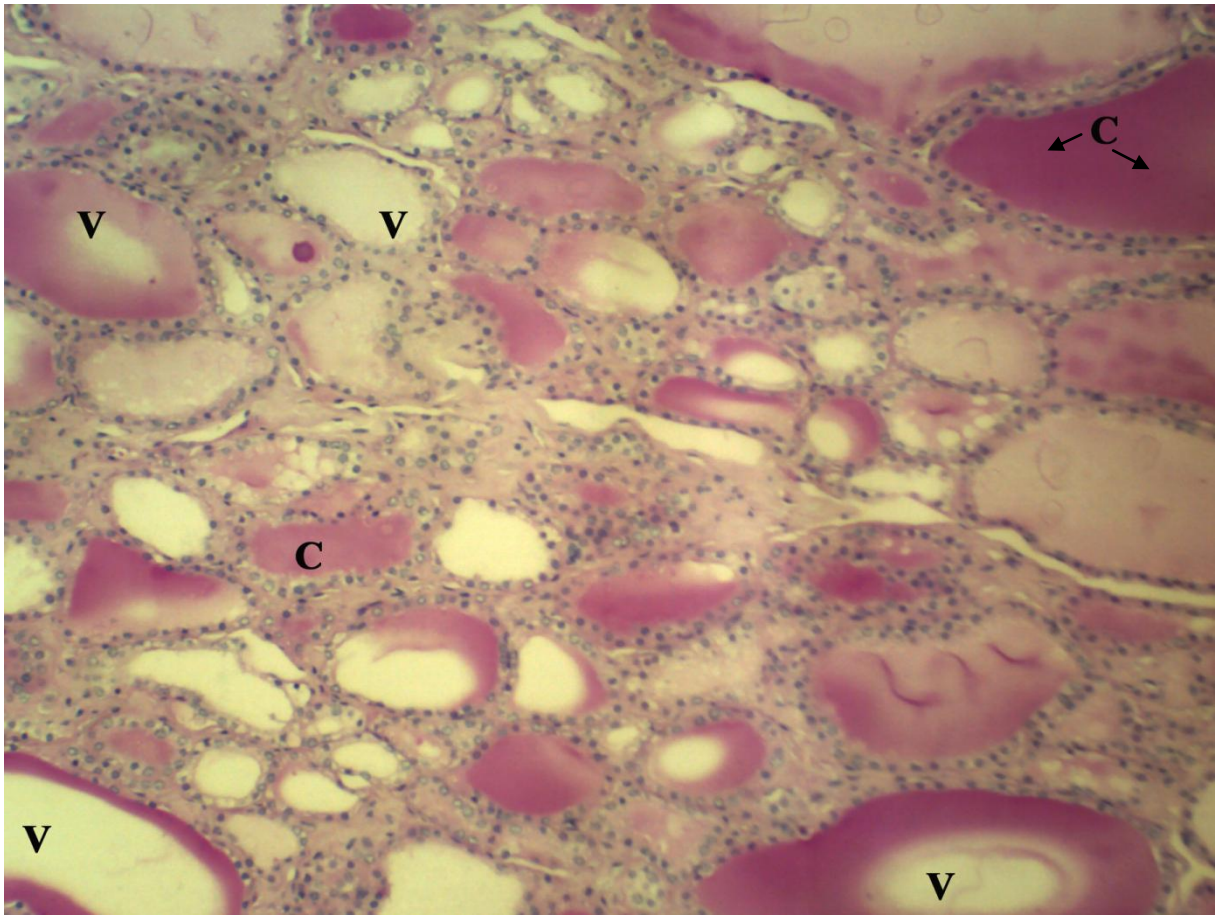


Plate 4.15: Photomicrograph of thyroid gland tissue **(Transverse section)** of **male** indigenous one-humped camel in group C showing vacuolated (**V**) follicles. The colloids (**C**) were negatively stained to Periodic Acid Schiif (PAS × 100).

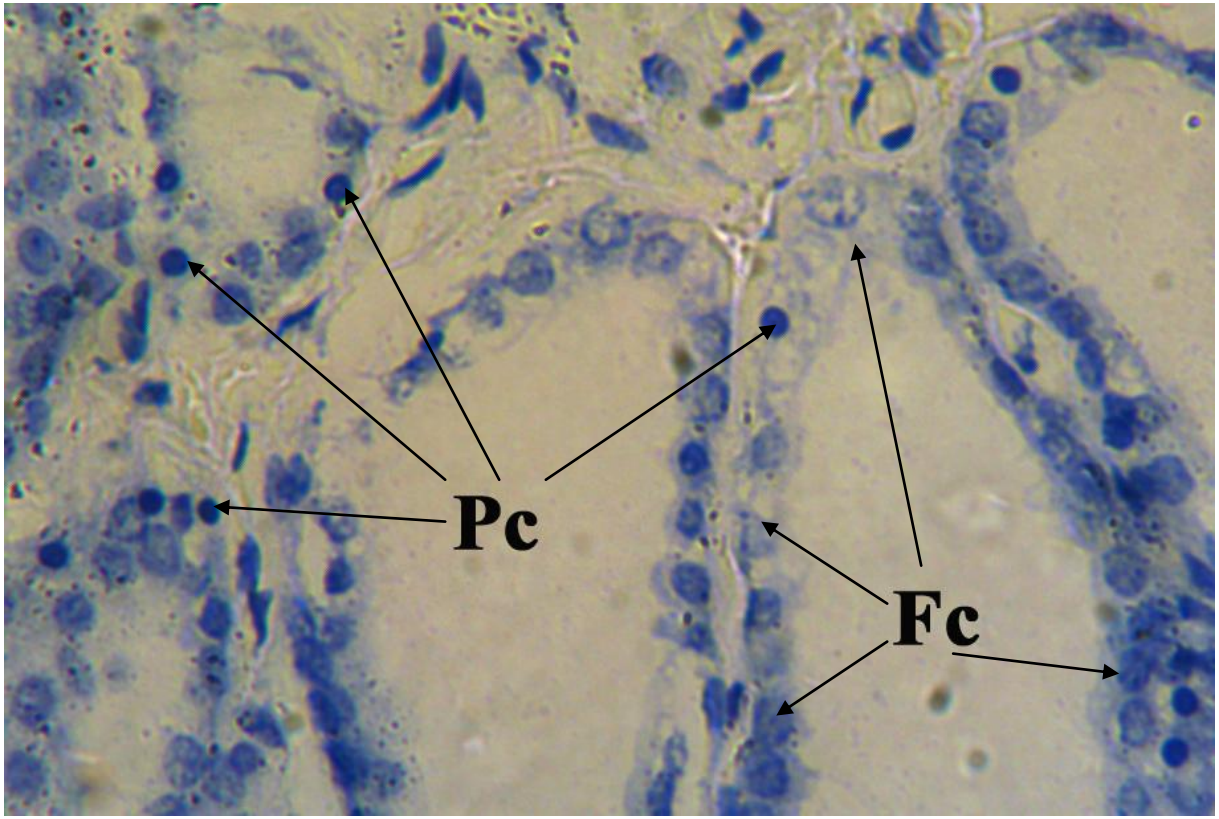


Plate 4.16: Photomicrograph of thyroid gland tissue **(Transverse section)** of **male** indigenous one-humped camel in group C. Azure A Acid hydrolysis stained the parafollicular cells **(Pc)** deep purple blue. Note the nuclei of the parafollicular cells (**Pc**) how they stand out clearly from the follicular cells **(Fc)** nuclei (Azure A × 400).

4.2.0 Parathyroid Gland

4.2.1 Gross Anatomy and morphological studies

The parathyroid glands were found usually to be two pairs of glands situated around both the right and left thyroid gland lobes, namely right superior parathyroid, left superior parathyroid, right inferior parathyroid, and left inferior parathyroid (Plate 4.17). However, right middle parathyroid glands and left middle parathyroid glands were seen in some of the specimen animals interchangeably, embedded into the thyroid gland, internally positioned (Plate 4.17). Most of the superior and inferior parathyroids are external glands positioned some little distance from the thyroid gland (Plate 4.17).

The glands were seen as light/ pale brown organs (Plate 4.18). They appeared like a little thyroid gland, triangular in shape, roughly (Plate 4.18).

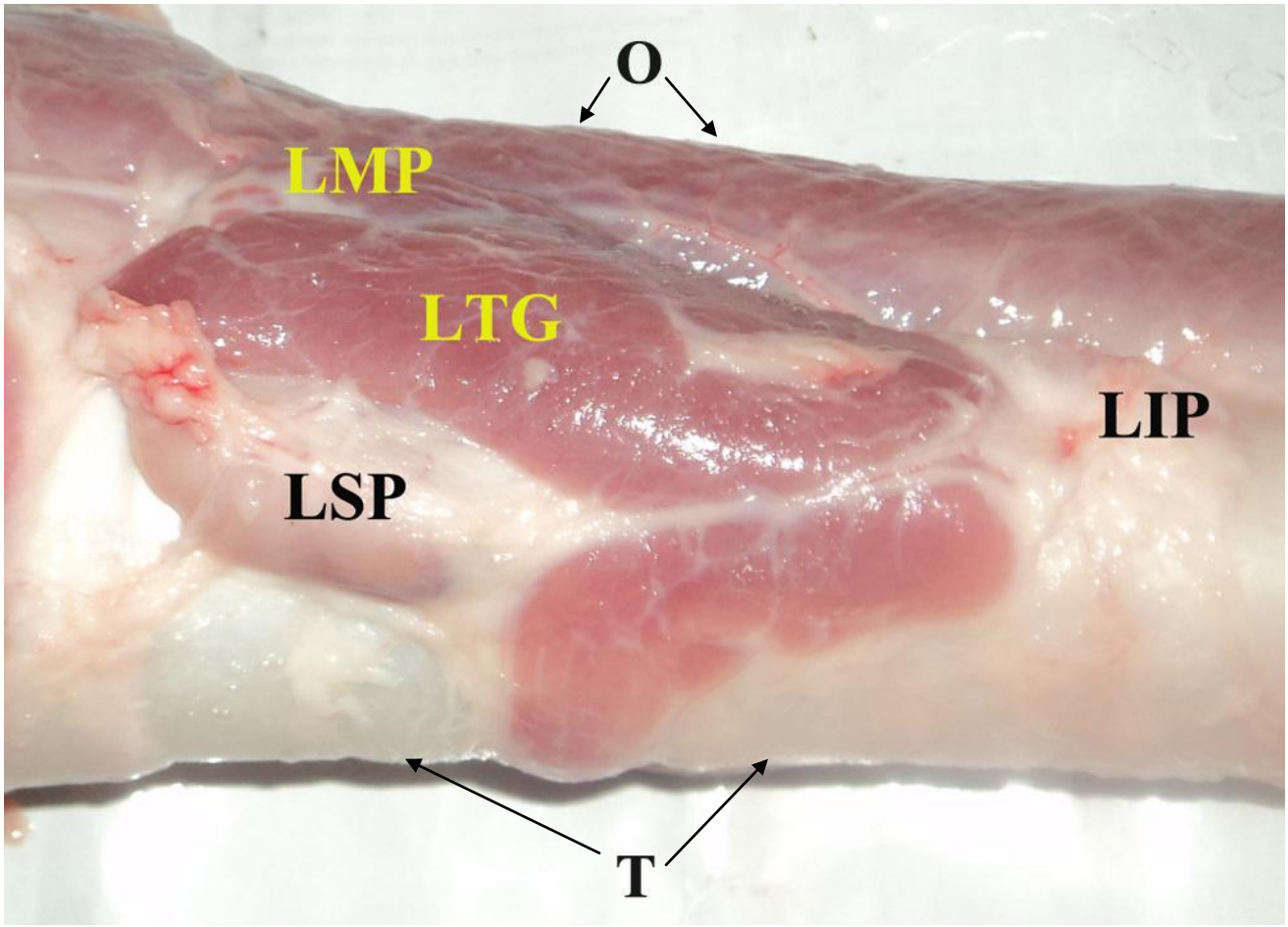


Plate 4.17: The left superior parathyroid (**LSP**), left middle parathyroid (**LMP**) and left inferior parathyroid (**LIP**) glands situated around the left thyroid gland (**LTG**). Note the left superior parathyroid gland (**LSP**) here was positioned anteriorly, the trachea (**T**) and the esophagus (**O**).



Plate 4.18: The picture of the parathyroid thyroid gland of indigenous one-humped camel is roughly triangular in shape.

4.2.2 Morphometric Studies of the Parathyroid

The study showed that the morphometric variables expressed as the mean plus /or minus standard error of mean of the weight, length, width and thickness of the right superior parathyroid glands in the female one-humped camels, among group A, between the age 1 – 4 years (juvenile), were: 0.41 ±0.08 g, 18.57±0.35 mm, 6.69 ± 0.67 mm and 3.59 ± 0.25 mm, respectively (Table 4.6). The mean of the weight, length, width, and thickness of the right superior parathyroid, right inferior parathyroid, left superior parathyroid, left inferior parathyroid in the male and female one-humped camels, between the ages 1-4 years (juvenile age), otherwise called group A, were also presented in Table 4.6. The mean of the weight, length, width, and thickness of the right superior parathyroid glands, in the males, between the ages 5 to 10 years (adult), otherwise called group B, were: 2.07 ± 0.31 g, 62.51±15.58 mm, 52.21±7.08 mm, and 17.82±11.33 mm, respectively (Table 4.6). Those of the female adults of the same group C were also presented, and the right inferior parathyroid, left superior parathyroid, left inferior parathyroid both in the males and females (Table 4.6). The adult female one-humped camels, above the age of 10 years, among the group C, in the studies, showed that the mean of the weight, length, width, and thickness of the right superior parathyroid were: 0.31 ± 0.07 g, 7.36 ± 1.33 mm, 5.16 ± 1.56 mm, and 0.82 ± 0.29 mm, respectively (Table 4.6), while those of males in the same group, and the right inferior parathyroid, left superior parathyroid, left inferior parathyroid both in both the sexes is presented in the Table 4.6.

The differences in the morphometric values of weight, length, width, and thickness of the thyroid gland in indigenous one-humped camel, between males and females, within group A were determined and found that sexual dimorphism in the weight of the right inferior parathyroid gland in both the males and the females were significant at the $P<0.05$ (Table

4.6). It was also significant at the P<0.05 on the weight of the left inferior parathyroid, in both males and females (Table 4.6). The differences in the length of the right superior parathyroid was significant at P<0.05 in both males and females (Table 4.6). The length of the right inferior parathyroid in the females, show significant differences at P<0.05, as well (Table 4.6). The width of the right inferior parathyroid differences were significant in both the male and female at P<0.05 (Table 4.6). Similarly, the width differences on the left inferior parathyroid in both females and males were significant at P<0.05 (Table 4.6). The thickness of the right superior and inferior parathyroid, in both the males and females, differences were significant at P<0.05 (Table 4.6).

In group B, it is only in the width that sexual dimorphism showed significance, being highly even at P<0.01 in the female right superior parathyroids, while in the males were at P<0.05 (Table 4.6). The left inferior parathyroids also showed significance differences in the width at P<0.05 in both the females and the males (Table 4.6).

In the group C all the morphometric variables showed no significant differences between the sexes within the groups (Table 4.6).

The differences in the morphometric variables were compared between all the males and females in all the groups, regardless of their age, and it showed that it was only the width of the right superior parathyroid that was significant difference at P<0.05, in both the males and females (Table 4.7).

The differences of morphometric variables between all the right parathyroid glands and all the left parathyroid glands were determined and the study showed that the differences are significant only at the weight of the superior parathyroid glands at p<0.05 in both the right and left of group A (Table 4.8), and weight of inferior parathyroid glands in both the right and left of group B (Table 4.8). The length differences is also significant in the inferior

parathyroid of both the right and left in group A, being highly significant with $p<0.01$ (Table 4.8). Lastly is the thickness which the difference was significant in the inferior parathyroid on both the right and the left of group C at $p<0.05$ (Table 4.8)

Table 4.6: Morphometric values and sexual dimorphism within each group of the indigenous one-humped camel's parathyroid gland

	Mean ± Std. Error Mean					
	Group A		**Group B**		Group C	
Variables	**Female (n=5)**	**Male (n=5)**	**Female (n=5)**	**Male (n=5)**	**Female (n=5)**	Male (n=5)
Weight RSP (g)	0.41 ± 0.08	0.95 ± 0.03	1.89 ± 0.54	2.07 ± 0.31	0.31 ± 0.07	0.98 ± 0.37
Weight RIP (g)	1.36 ± 0.04*	0.49±0.26*	0.87 ± 0.30	0.40 ± 0.21	0.51 ± 0.17	2.19 ± 0.83
Weight LSP (g)	1.17 ± 0.40	1.36±0.31	2.38 ± 0.93	1.20 ± 0.18	0.65 ± 0.28	0.67 ± 0.18
Weight LIP (g)	1.11 ± 0.08*	0.63 ± 0.16*	1.18 ± 0.43	1.54 ± 0.41	1.50 ± 0.57	1.08 ± 0.59
Length RSP (mm)	18.57 ± 0.35*	25.71 ± 2.35*	34.32 ± 2.8	62.51 ± 15.58	7.36 ± 1.33	15.20 ± 4.65
Length RIP (mm)	27.04 ± 0.87*	16.96 ± 4.10	16.66 ± .89	39.82 ± 12.04	17.34 ± 3.94	28.42 ± 5.39
Lenght LSP (mm)	22.44 ± 2.28	26.98 ± 0.64	42.9 ± 8.07	30.08 ± 2.95	13.87 ± 5.26	15.22 ± 2.37
Length LIP (mm)	27.47 ± 0.32	21.48 ± 4.12	24.83±3.58	23.56 ± 3.33	19.58 ± 5.56	17.44 ± 5.59
Width RSP (mm)	6.69 ± 0.67	10.08 ± 1.59	10.75 ±.48**	52.21 ± 7.08*	5.16 ± 1.56	6.40 ± 2.46
Width RIP (mm)	11.60±1.11*	6.02±0.82*	8.80 ± 0.79	27.06 ± 10.51	7.60 ± 1.23	11.16 ± 1.36
Width LSP (mm)	10.55 ± 1.67	10.58 ± 1.02	11.7± 1.05*	7.79 ± 0.82*	5.93 ± 1.70	9.06 ± 1.95
Width LIP (mm)	10.51 ± 0.93*	7.78 ± 0.57*	9.13 ± 0.93	9.62 ± 1.08	6.88 ± 1.65	4.92 ± 2.24
Thickness RSP (mm)	3.59 ± 0.25*	6.18 ± 0.45*	4.95 ± 0.30	17.82 ± 11.33	0.82 ± 0.29	1.62 ± 0.76
Thickness RIP (mm)	6.43 ± 0.26*	3.60 ± 0.60*	4.27 ± 0.41	5.85 ± 1.94	1.66 ± 0.37	3.18 ± 0.82
Thickness LSP (mm)	5.91 ± 0.85	5.92 ± 0.60	7.21 ± 1.09	11.57 ± 1.79	1.46 ± 0.91	1.40 ± 0.40
Thickness LIP (mm)	6.31 ± 0.37	5.07 ± 0.89	4.44 ± 0.47	6.62 ± 1.17	1.54 ± 0.44	1.45 ± 1.02

Student's t-test analysis for sexual dimorphism between the sex within the groups
***= P<0.05**
****** = P<0.01**
RSP= right superior parathyroid, RIP= right inferior parathyroid, LSP = left superior parathyroid, LIP = Left Inferior parathyroid.

Table 4.7 Sexual dimorphism between male and female parathyroid glands in all the one-humped camel studied

Variables	**Mean ± Std. Error Mean**	
	Female (n=15)	**Male (n=15)**
Weight RSP (g)	0.88 ± 0.26	**1.33 ± 0.23**
Weight RIP (g)	0.91 ± 0.14	**1.03 ± 0.35**
Weight LSP (g)	1.40 ± 0.38	**1.08 ± 0.15**
Weight LIP (g)	1.26 ± 0.23	**1.082 ± 0.25**
Length RSP (mm)	20.08 ±3.11	**34.47 ± 7.42**
Length RIP (mm)	20.35 ±1.80	**28.40 ± 4.94**
Length LSP (mm)	26.41 ±4.46	**24.09 ± 2.08**
Length LIP (mm)	23.96 ±2.22	**20.83 ± 2.47**
Width RSP (mm)	7.54 ± 0.83*	**22.90 ± 6.04***
Width RIP (mm)	9.33 ± 0.72	**14.75 ± 4.06**
Width LSP (mm)	9.39 ± 1.04	**9.15 ± 0.79**
Width LIP (mm)	8.84 ± 0.77	**7.44 ± 0.94**
Thickness RSP (mm)	3.12 ± 0.48	**8.54± 3.95**
Thickness RIP (mm)	4.12 ± 0.56	**4.21 ± 0.75**
Thickness LSP (mm)	4.86 ± 0.83	**6.30 ± 1.26**
Thickness LIP (mm)	4.10 ± 0.57	**4.38 ± 0.80**

***=P<0.05**

Table 4.8 Differences between right and left parathyroid glands

	Mean ± Std. Error Mean					
	Group A		**Group B**		**Group C**	
Variables	**Right Gland (n=10)**	**Left Gland (n=10)**	**Right (n=10)**	**Left (n=10)**	**Right (n=10)**	**Left (n=10)**
Weight SP (g)	0.68 ± 0.18*	1.26 ± 0.24*	1.98 ± 0.30	1.79± 0.49	0.64 ± 0.21	0.66 ± 0.16
Weight IP (g)	0.93 ± 0.19	0.87 ± 0.11	0.64± 0.19*	1.36± 0.29*	1.35 ± 0.49	1.29 ± 0.39
Length SP (mm)	22.14 ± 1.63	24.71 ± 1.35	48.41±8.82	36.49±4.58	11.28 ± 2.63	14.55 ± 2.73
Length IP (mm)	22.00 ± 2.60**	24.47 ± 2.19**	28.24±6.88	24.19±2.31	22.88 ± 3.65	18.51 ± 3.73
Width SP (mm)	8.39 ± 0.99	10.57 ± 0.92	31.48±7.68*	9.74 ±0.90*	1.39 ± 0.78	7.49 ± 1.33
Width IP (mm)	8.81 ± 1.14	9.15 ± 0.69	17.93±5.83	9.37 ±0.68	9.38 ± 1.05	5.90 ± 1.35
Thickness SP (mm)	4.88 ± 0.49	5.92 ± 0.49	11.38±5.76	9.39 ±1.23	1.22 ± 0.41	1.43 ± 0.47
Thickness IP (mm)	5.02 ± 0.57	5.69 ± 0.50	5.06 ± 0.97	5.53 ±0.70	2.42 ± 0.49*	1.49 ± 0.52*

***= P<0.05**
****=P<0.01**

4.2.3 Histological and Histochemical Studies of the Parathyroid:

Group A of the study consists of ten (10) equal number of female and male indigenous one-humped camels between the ages of 1- 4 years (juvenile age).

The result of the histological study is as follows:

Haemotoxylin and Eosin (H&E): The parathyroid glands are surrounded by a thin capsule of connective tissue. It has abundant stroma of connective tissue, with big smooth muscles all over (Plate 4.19).

The tissue of the parathyroid gland consists of clusters of principal or chief cells (Plates 4.19, 4.20 and 4.21).

There are two different functional stages of the principal cell. The **light principal cell** has a large, pale nucleus and pale cytoplasm (described as inactive) (Plates 4.20 and 4.21). The **dark principal cell** is a smaller cell, with a small, dark nucleus and a dark cytoplasm (active cell) (Plates 4.20 and 4.21). The dark cells tend to be located peripheral to the more central, light cells (Plates 4.20 and 4.21). The presence of Oxyphil cells was observed (Plates 4.20 and 4.21).

The result of histochemical studies showed that the tissue of the parathyroid gland in indigenous one-humped camel, reacted negatively to the periodic acid schiff (PAS) (Plate 4.22)**.**

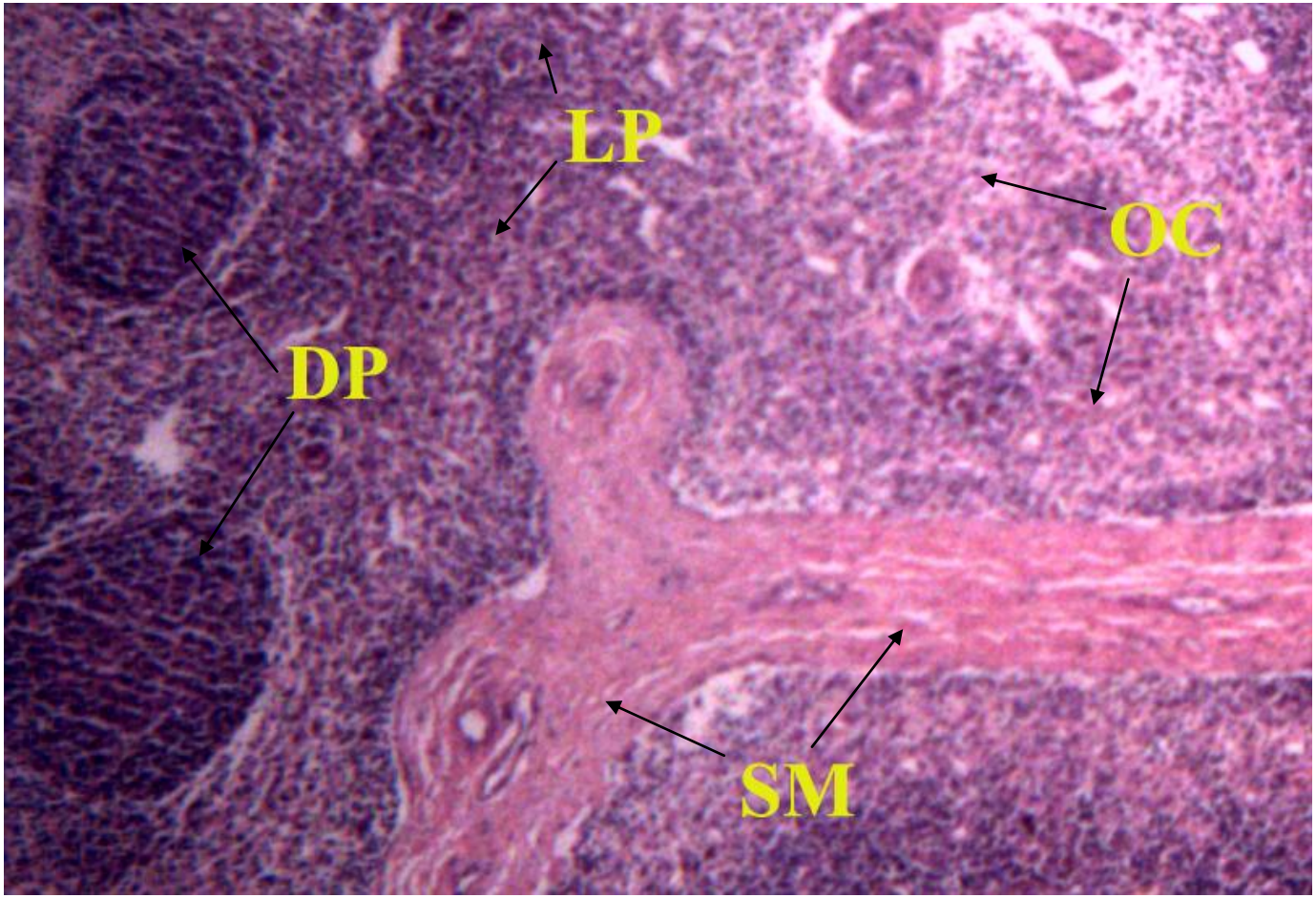


Plate 4.19: Photomicrograph of Parathyroid gland tissue section (longitudinal) among the female of group A. It showed clusters of dark principal cells **(DP),** the light principal (**LP**) cells and the oxyphil cells (**OC**) and smooth muscle **(SM)** (H&E × 40)**.**

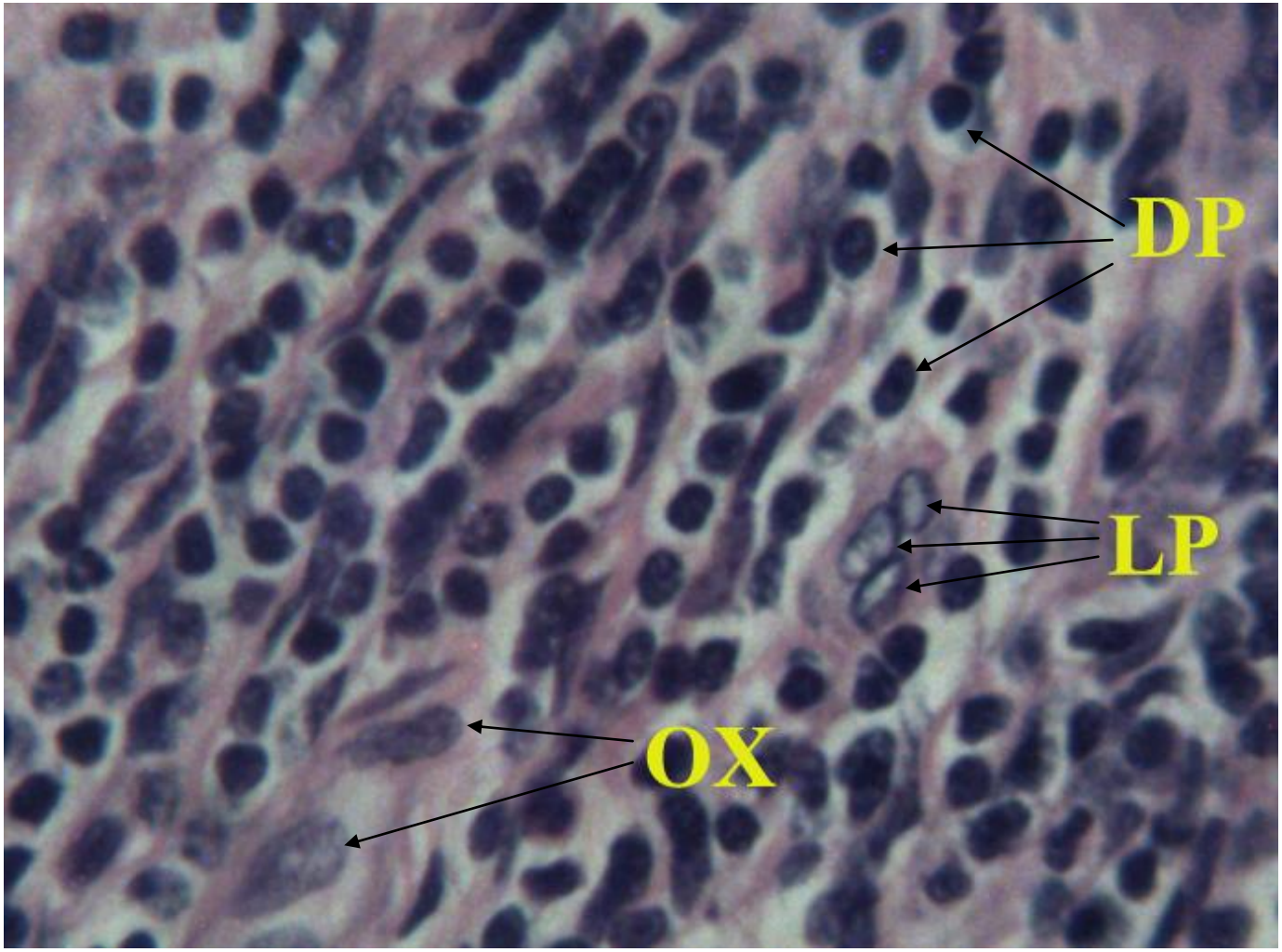


Plate 4.20: Photomicrograph of Parathyroid gland tissue section (longitudinal) among the female of group A. Note the **Dark Principal (DP)**, and the **Light Principal (LP),** and the Oxyphil cells **(OX)** with paller and bigger cytoplasm and Nucleus (H&E × 250).

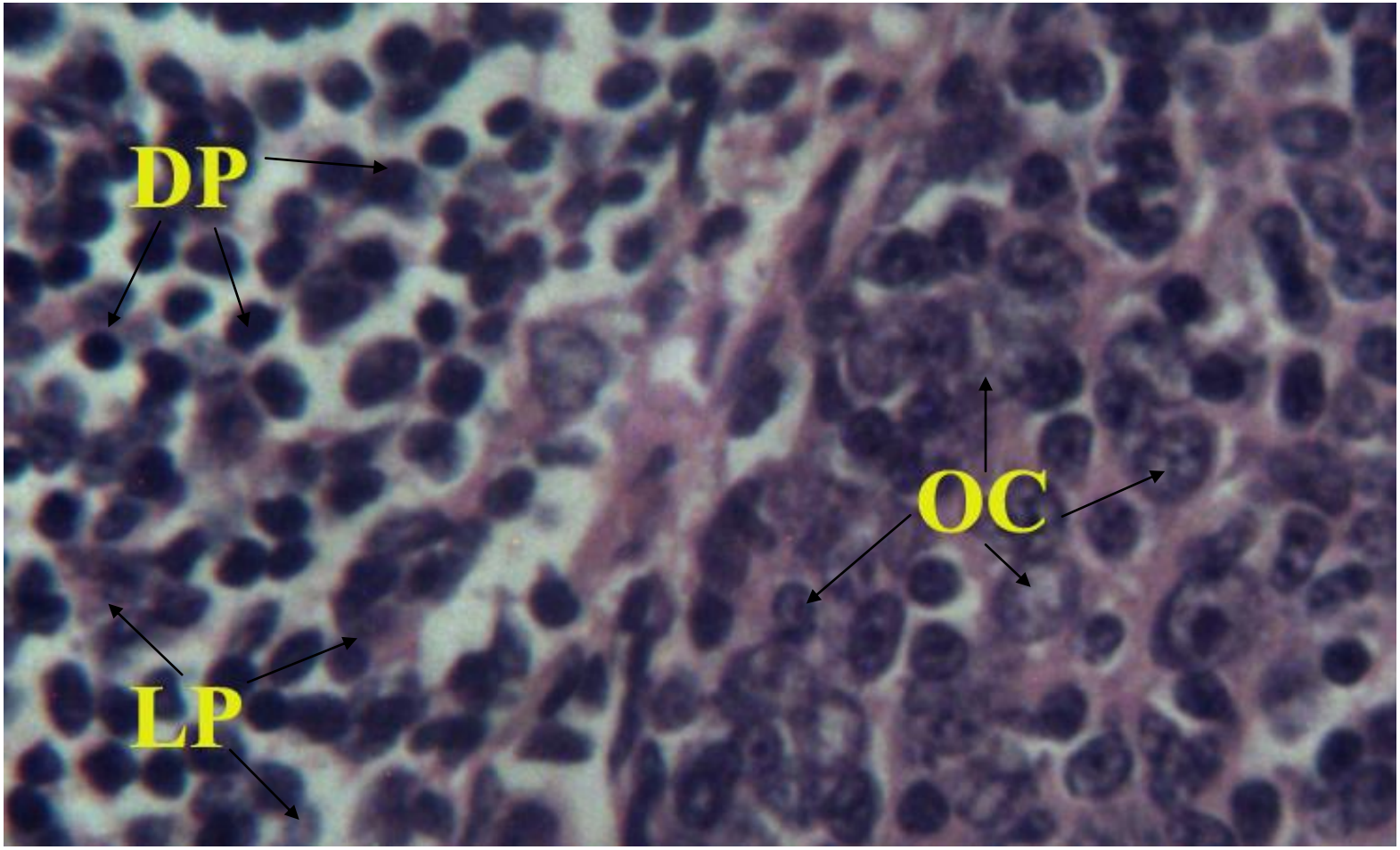


Plate 4.21: Photomicrograph of Parathyroid gland tissue section (longitudinal) among the male of group A note the principal cells; the **dark principal (DP)**, and **light principal (LP)** packed cords, while some were scattered**.** The **Oxyphil cells (OC)** are showned with paller and bigger cytoplasm and nucleus(H&E × 450)

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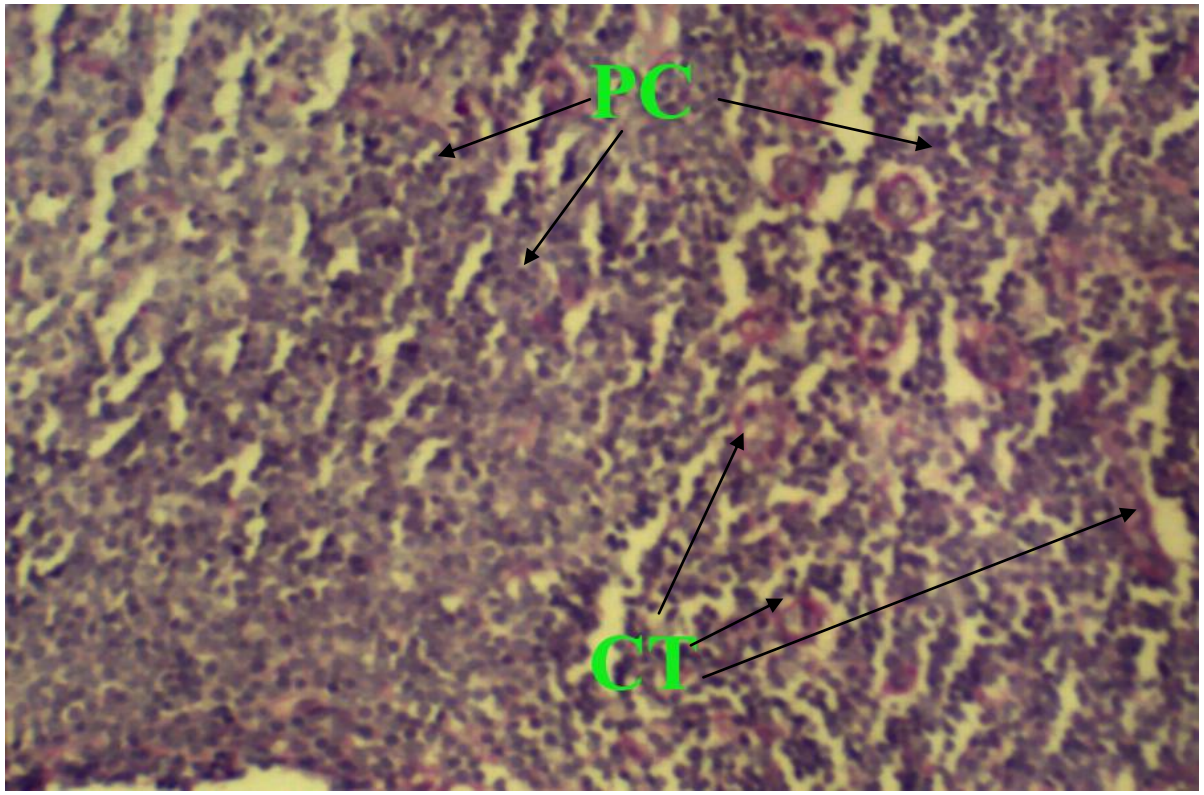


Plate 4.22: Photomicrograph of Parathyroid gland tissue section (Longitudinal) among the male of group A, showing the principal cells (**PC**) and connective tissues (**CT**), but the reaction to PAS is negative (PAS × 100)

Group B of the study consists of ten (10) equal number of female and male indigenous one-humped camels between the ages of 5 - 10 years (young adult).

The results of the histology study are as follows:

Haemotoxylin and Eosin (H&E) stain

It has abundant stroma of connective tissue, with big smooth muscles all over (Plates 4.23 and 4.24).

The tissue of the parathyroid gland consists of clusters and cords of principal or chief cells (Plates 4.23 and 4.24).

There are two different functional stages of the principal cell. The **light principal cell** has a large, pale nucleus and pale cytoplasm (described as inactive) (Plates 4.23 and 4.24). The **dark principal cell** is a smaller cell, with a small, dark nucleus and a dark cytoplasm (active cell) (Plates 4.23 and 4.24). The dark cells tend to be located peripheral to the more central, light cells (Plates 4.23 and 4.24). It also has abundant Oxyphil cells, which are large cells with an acidophilic cytoplasm and **pyknotic** nucleus (Plates 4.23 and 4.24). There are abundance of Adipose cells especially in the male (compare Plates 4.23 and 4.24).

The result of histochemical studies showed that the tissue of the parathyroid gland in indigenous one-humped camel, reacted negatively to the **Periodic Acid Schiff (PAS)** (Plate 4.25)**.** The PAS slides did not stained different from the H&E.

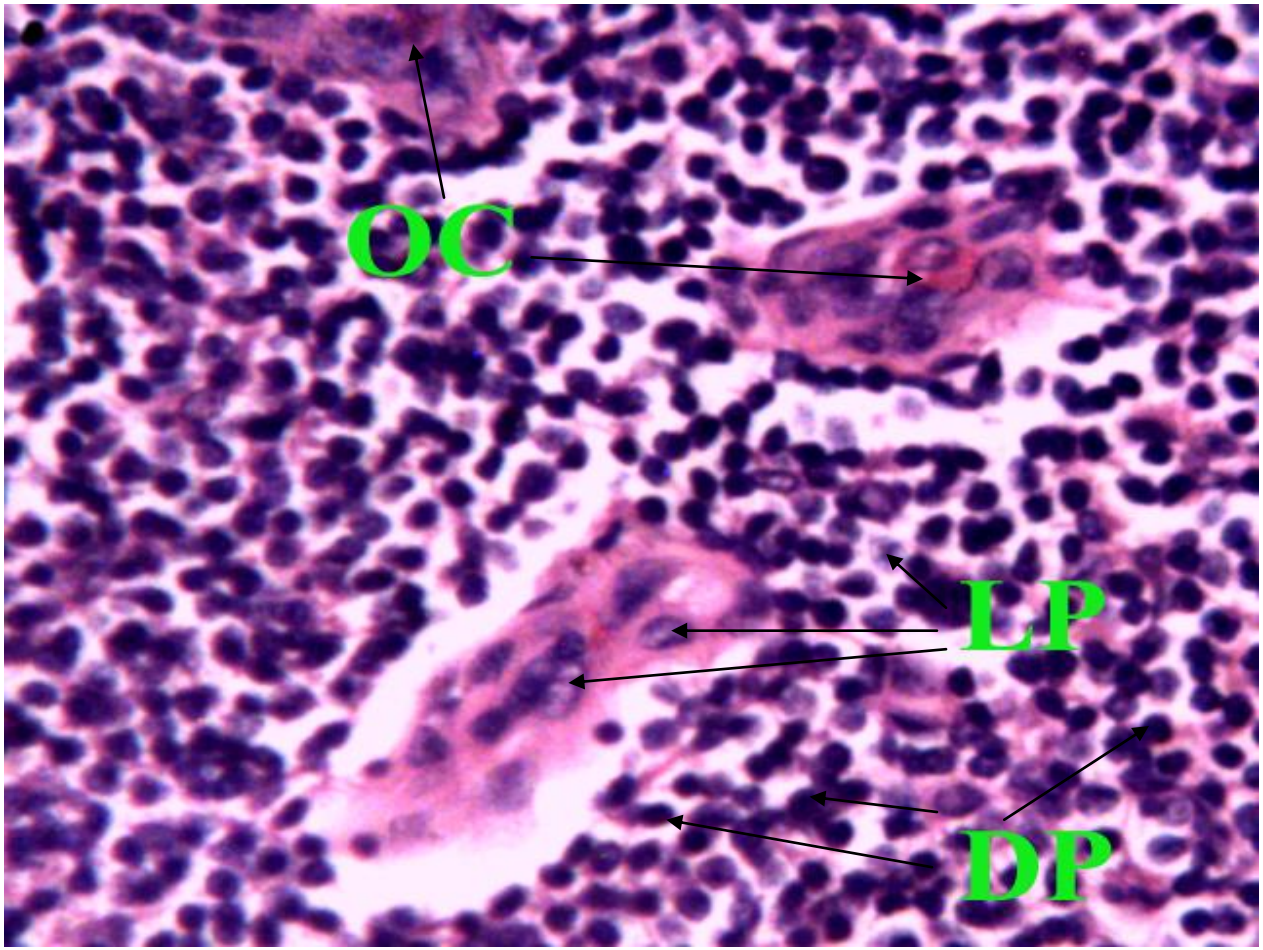


Plate 4.23: Photomicrograph of parathyroid gland tissue section (longitudinal) among the female of group B showing clusters and cords of **light principal cell (LP)** and **dark principal cell** (**DP**) and abundant oxyphil cells (**OC**) and stroma of vascular connective tissue (H&E × 250)

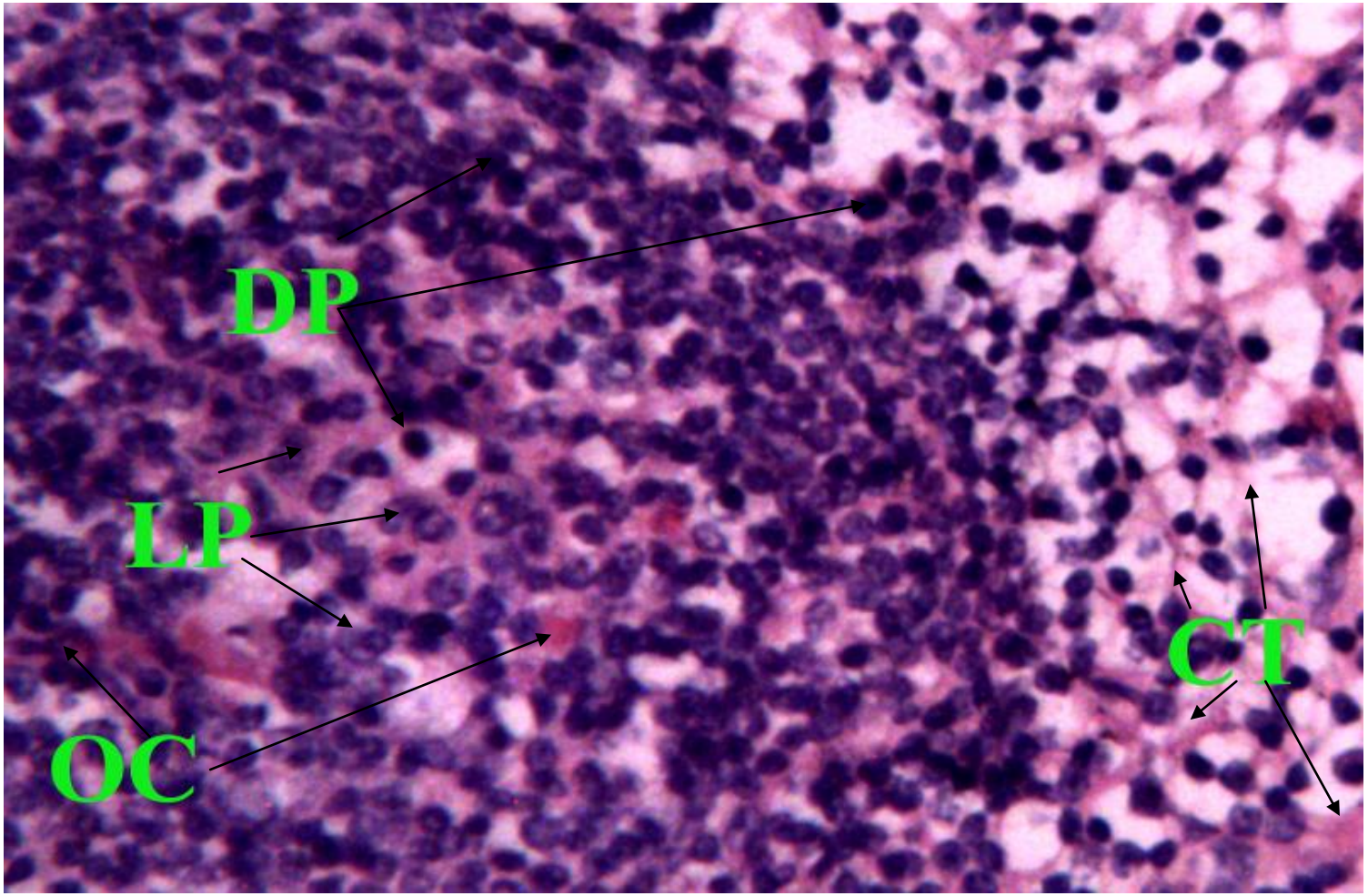


Plate 4.24: Photomicrograph of Parathyroid gland tissue section (transverse) among the male of group B showing clusters and cords of principal cells; **light principal cell (LP)** and **dark Principal cell** (**DP**), abundant oxyphil cells (**OC**), and mesh of vascular connective tissue (**CT**) (H&E × 250).

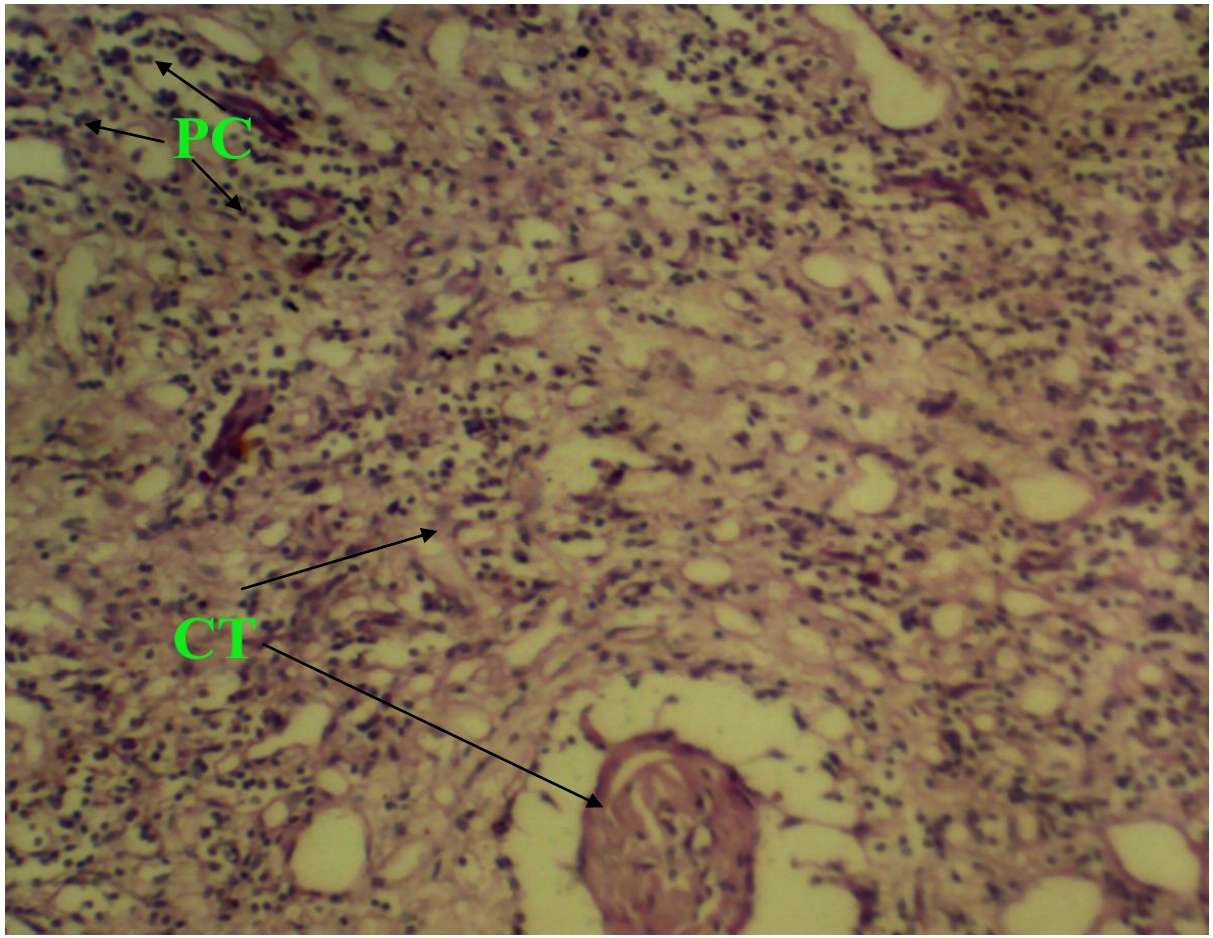


Plate 4.25: Photomicrograph of Parathyroid gland tissue section (transverse) among the male of Group B showing the principal cells (**PC**), and connective tissue (**CT**), which the reaction is negative to PAS (PAS × 100).

Group C of the study consists of ten (10) equal number of female and male indigenous one-humped camels above the age of 10 years (old adult).

The result of the histological study is as follows:

Haemotoxylin and Eosin (H&E) stain:

It has abundant stroma of connective tissue, with big smooth muscles all over (Plates 4.26 and 4.27).

The tissue of the parathyroid gland consists of clusters and cords of principal or chief cells (Plates 4.26 and 4.27).

There are two different functional stages of the principal cell. The light principal cell has a large, pale nucleus and pale cytoplasm (described as inactive) (Plates 4.26 and 4.27). The **dark principal cell** is a smaller cell, with a small, dark nucleus and a dark cytoplasm (active cell) (Plates 4.26 and 4.27). The dark cells tend to be located peripheral to the more central, light cells (Plates 4.26 and 4.27). It also has abundant Oxyphil cells, which are large cells with an acidophilic cytoplasm and **pyknotic** nucleus (Plates 4.26 and 4.27). There are abundance of Adipose cells especially in the female (compare Plates 4.26 and 4.27).

The result of histochemical studies showed that the tissue of the parathyroid gland in indigenous one-humped camel, reacted negatively to the **Periodic Acid Schiff (PAS)** (Plate 4.28)**.** The PAS slides did not stained different from the H&E. No difference was observed between the sexes of the same group.

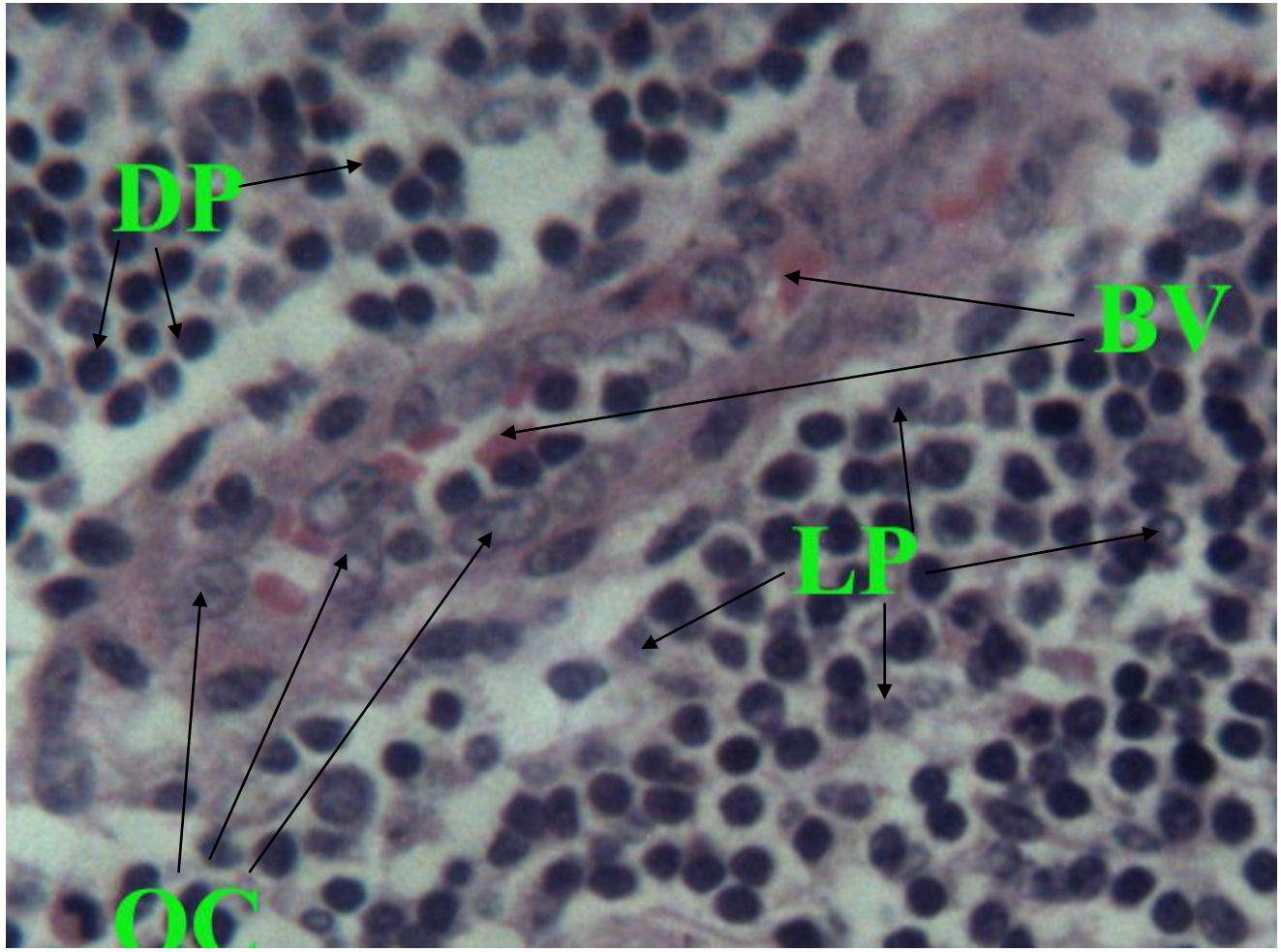


Plate 4.26: Photomicrograph of Parathyroid gland tissue section (transverse) among the female of group C, showing the clusters of dark principal cells (**DP**) and light principal cells (**LP**). Note the oxyphil cells (O**C**) with larger and paler nucleus, and blood vessels (**BV**) (H&E × 400).

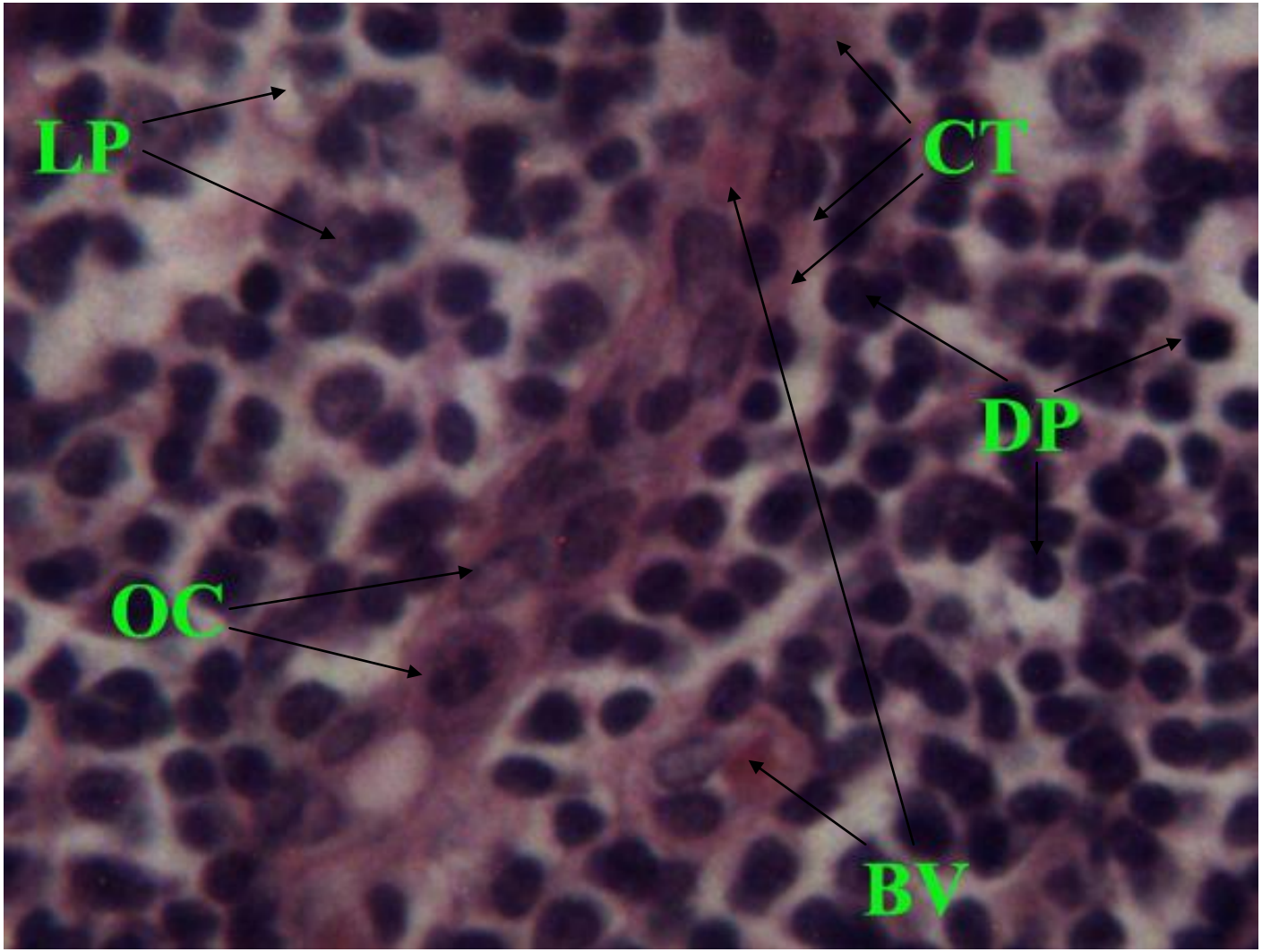


Plate 4.27: Photomicrograph of parathyroid gland tissue section (transverse) among the male of Group C, showing the clusters of dark principal cells (**DP**) and light principal cells (**LP**). Note the oxyphil cells (O**C**) with larger and paler nucleus, and blood vessels (**BV**) (H&E × 400)

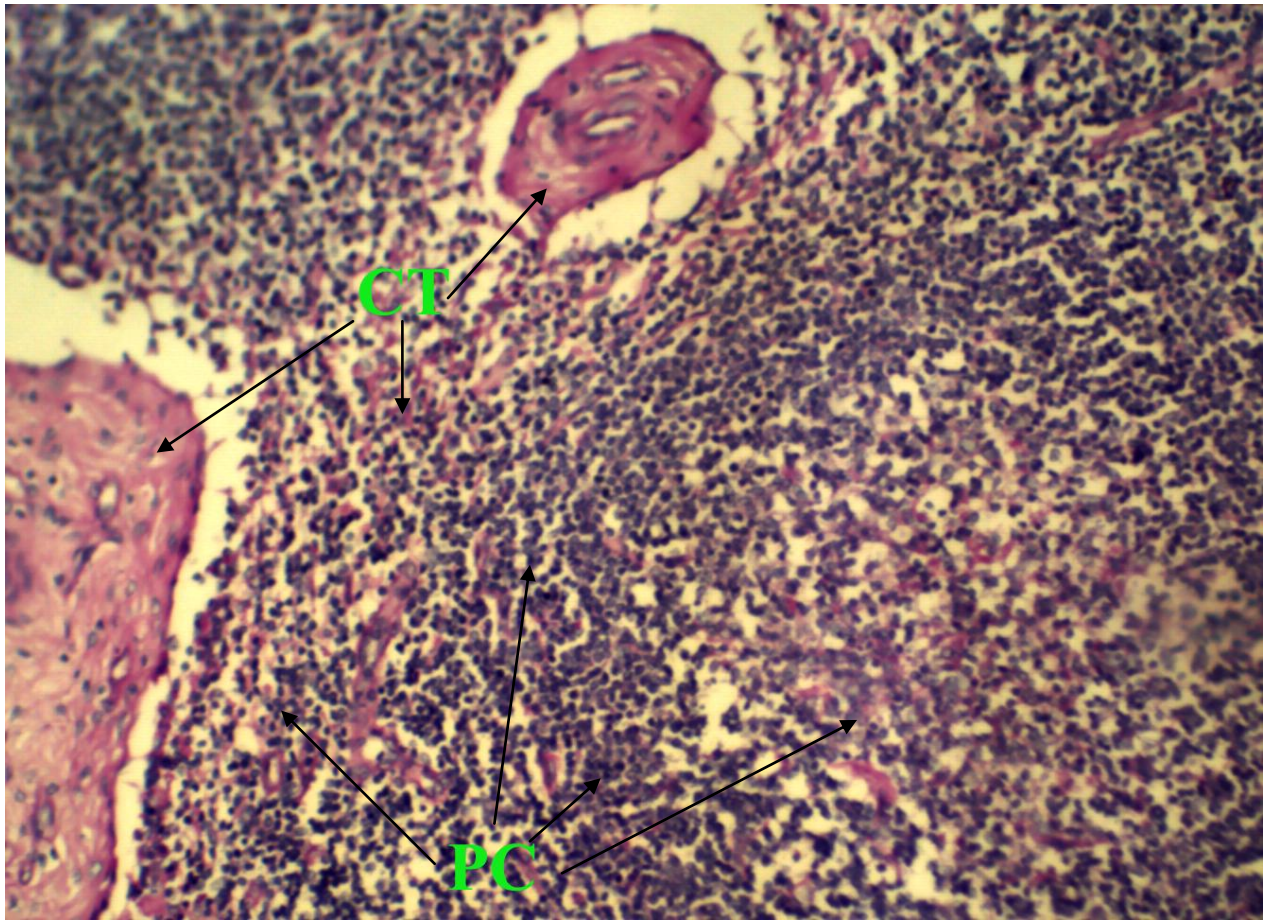


Plate 4.28: Photomicrograph of Parathyroid gland tissue section (transverse) among the male of group C, showing the principal cells (**PC**), and connective tissue (**CT**), the reaction is negative to PAS (PAS × 100)

4.3.0 Adrenal Glands:

4.3.1 Gross anatomy and morphological studies:

This study revealed that the right adrenal was found superior medial to the right kidney, not attached to it (Plate 4.29), but it was found to be attached to the inferior medial surface of the liver (Plate 4.29). The left adrenal gland was found to be superior medially away from the left kidney (Plate 4.29). The right adrenal gland was triangular in shape while the left adrenal gland was irregularly oval.

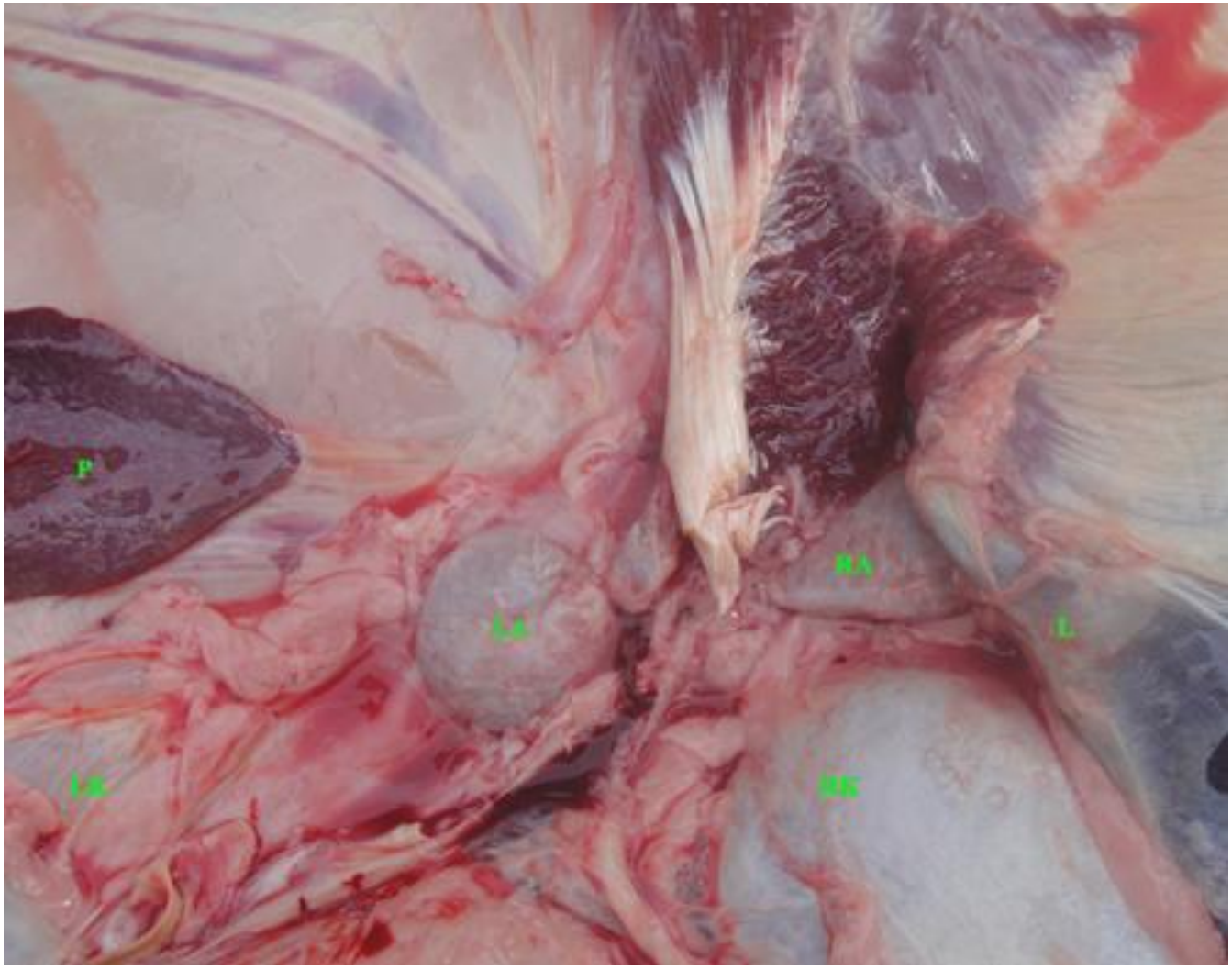


Plate 4.29: Some of the internal organs showing left adrenal gland (**LA**) far above medial to the left kidney (**LK**), the right adrenal gland (**RA**) above the right kidney (**RK**) but below the inferior boarder of the liver (**L**), far left is the pancreas (**P**) reflected aside

4.3.2 Morphometry of the Adrenal glands

The study showed that, in female of group A, the morphometric variables expressed as the mean of the weight, length, width and thickness of the right adrenal gland were: 16.49±1.55 g, 47.80±1.03 mm, 37.81±1.86 mm, and 20.22 ±1.47 mm, respectively (Table 4.11). The mean of the weight, length, width and thickness of the left adrenal gland in the indigenous one-humped camel were also presented in Table 4.11. The mean of the weight, length, width and thickness of the right and left adrenal glands of the indigenous one-humped camel were also presented in Table 4.11. Those of the groups B and C were also presented in Table 4.11. The differences in the morphometric values between the sexes were significant at $p<0.05$ in the weight of the right and left adrenal glands, length of right adrenal and thickness of left adrenal gland, in both the male and the female (Table 4.11).

Table 4.9 Value of adrenal gland used to determine sexual dimorphism among the groups

Mean ± Std. Error Mean						
	Group A		**Group B**		**Group C**	
Variables	**Female (n=5)**	**Male (n=5)**	**Female (n=5)**	**Male (n=5)**	**Female (n=5)**	**Male (n=5)**
Weight RG (g)	16.49±1.55*	10.54 ± 1.63*	23.65 ± 1.52	22.28 ± 1.26	23.73 ± 1.23	**23.44 ± 1.96**
Weight LG (g)	16.90±1.69*	7.24 ± 1.4.31*	22.23 ± 2.16	21.71 ± 1.02	20.92 ± 1.41	**19.76 ± 1.34**
Lenght RG (g)	47.80±1.03*	54.71±2.43*	55.14 ± 1.07	57.31 ± 1.87	60.83 ± 4.94	**62.70 ± 4.82**
Length LG (mm)	42.64 ± 1.94	37.88 ± 3.18	49.32 ± 0.65	48.95 ± 6.57	45.68 ± 1.28	**42.46 ± 2.16**
Widht RG (mm)	37.81±1.86	35.33 ± 4.55	39.72 ± 2.16	33.40 ± 2.53	37.51 ± 1.40	**29.79 ± 6.95**
Widht LG (mm)	35.63±2.31	33.65 ± 2.20	41.71 ± 1.59	37.08 ± 3.21	37.24 ± 1.25	**37.04 ± 1.64**
Thickness RG (mm)	20.22 ±1.47	15.15 ± 1.79	17.52 ± 0.42	18.64 ± 0.86	13.64 ± 1.35	**12.90 ± 0.76**
Thickness LG (mm)	18.43±0.79*	15.05 ± 0.33*	17.64 ±1.20	18.67 ± 0.55	15.44 ± 0.75	**13.04 ± 0.85**

*=P<0.05

RG = Right Gland of Adrenal

LG = Left Gland of Adrenal

4.3.3 Histological and Histochemical Studies of the Adrenal glands:

Group A:

The glands are covered by a capsule of dense irregular connective tissue (Plates 4.30 and 4.32). Clusters of cortical cells occur in the capsule (Plates 4.32). Thin trabeculae of the connective tissue capsule project into the parenchyma of the cortex reaching far deep into the zona fasciculata, more in the female than the male (Plates 4.30, 4.32, and 4.33). In male, the trabeculae coils to form a cluster like arrangement (Plates 4.32 and 4.33). The adrenal gland tissue is organized into two parts; the peripheral cortex and a central medulla (Plates 4.30 and 4.31). The adrenal cortex is further subdivided microscopically into Zona glomerulosa (Plates 4.30, 4.32, and 4.33), Zona fasciculata (Plates 4.30, 4.32, 4.33 and 4.34), and Zona reticularis (Plates 4.30, 4.31, and 4.35).

The zona glomerulosa or zone multiformis is the outermost zone (Plates 4.30, 4.32 and 4.33). The parenchymal cells of this region are columnar and polyhedrally arranged into arcs or clusters or cords as an inverted U-shaped formation by the trabeculae (Plates 4.32 and 4.33).

The zona fasciculata is formed by arranged cords in straight columns of cuboidal or polyhedral cells separated by sinusoids (Plates 4.30, 4.32 4.33 and 4.34). The cytoplasm of the cells in this zone frequently appears foamy because of the presence of numerous lipid vacuoles (Plate 4.34).

The zona reticularis is the innermost zone of the adrenal cortex, and it is the widest zone (Plates 4.30 and 4.31). It is arranged as an irregular network of anastomosing cords of cells surrounded by sinusoids. The cells are polyhedral in shape (Plate 4.35).

The adrenal medulla is composed mostly of columnar or polyhedral chromaffin cells, which form clusters and anastomosing cords separated by sinusoids, and have basophilic cytoplasm (Plates 4.31 and 4.36).

The medulla may appear within the zona reticularis, because the cortex and medulla interdigitate at their junction (Plate 4.31).

Histochemical staining of the adrenal tissue with Alcian Blue revealed how the connective tissue trabeculae and the sinusoids reacted with alcian blue reagent and stained blue (Plate 4.37).

When the gland was fixed in potassium dichromate fixative, the stored catecholamine granules of adrenal medullary cells (chromaffin cells) were oxidised to brown colour (Plate 4.38)

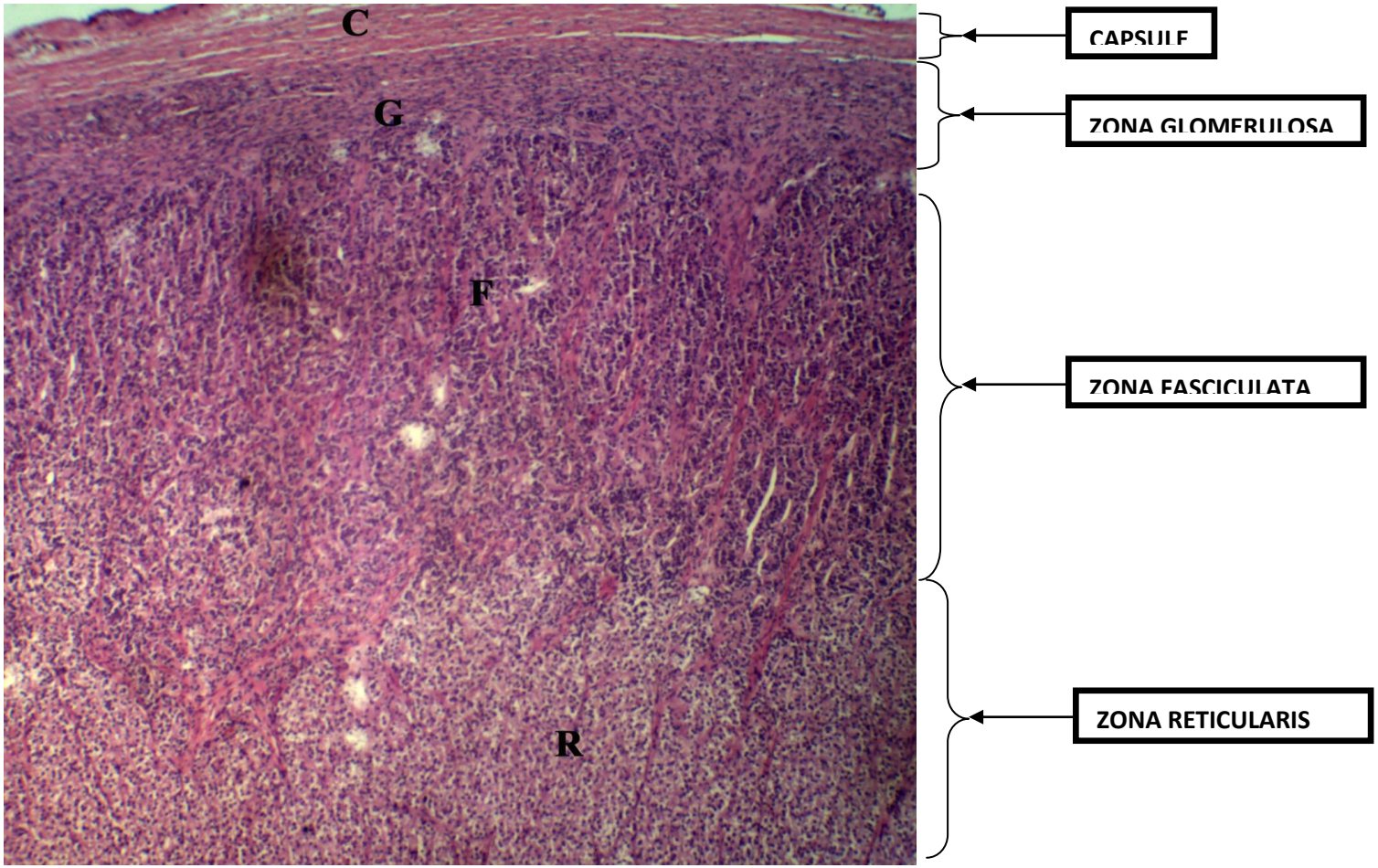


Plate 4.30: Photomicrograph of Adrenal gland tissue section (transverse) among the female of group A. Note Connective tissue capsule (**C**) and its trabeculae penetrating the zones in different arrangements up to the zona reticularis. The three zones of the adrenal cortex where distinct above; cells of the zona glomerulosa (**G**) were lying immediately behind the capsule (**C**) and composed of cells arranged in irregular ovoid clusters, the zona fasciculata (**F**) is the middle of the three cortical zones. It consists of cells in an irregular network of branching cords. The zona reticularis (**R**) is the innermost layer of the adrenal cortex (H&E × 40).

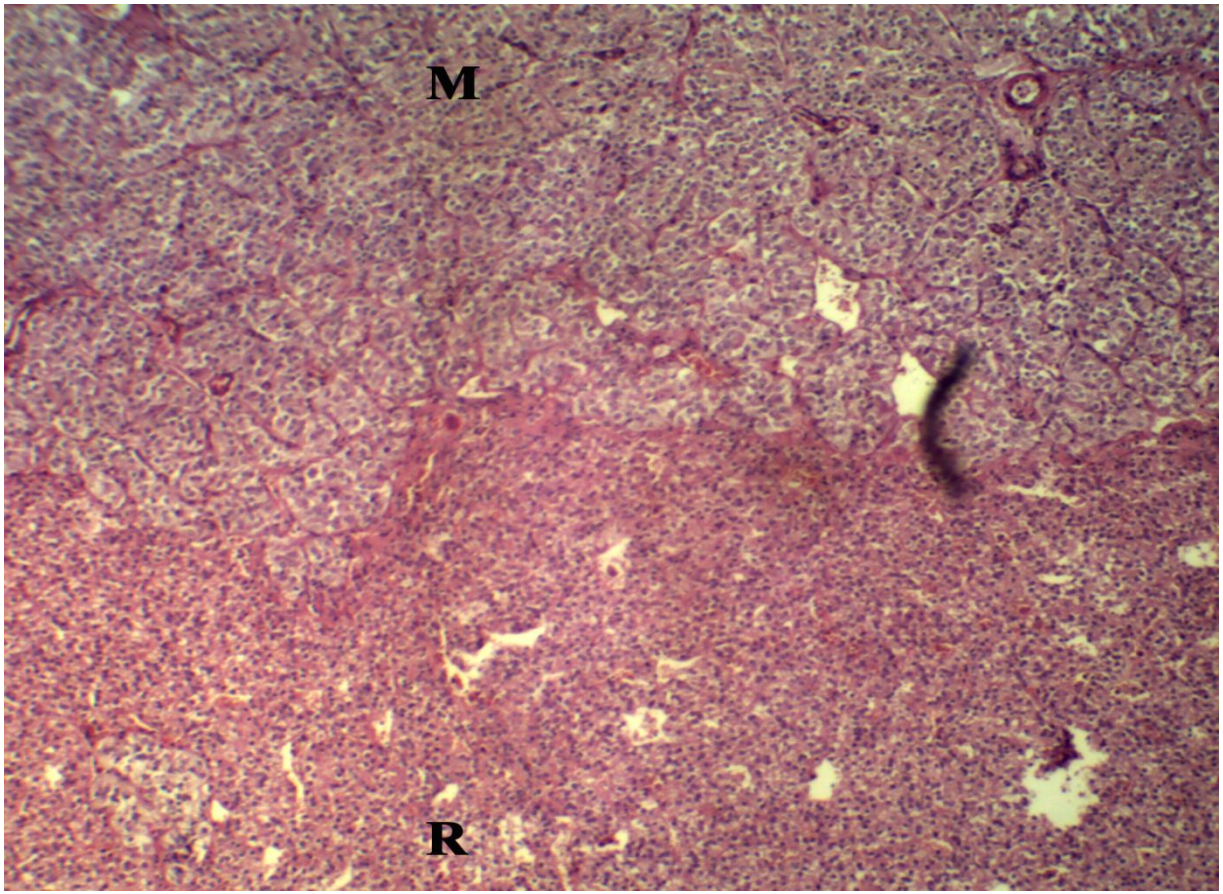


Plate 4.31: Photomicrograph of Adrenal gland tissue section (longitudinal) among the female of Group A. The continuation of the zona reticularis (**R**) which lies next to the adrenal medulla having irregular network of anastomosing cords of cells surrounded by sinusoids; the medulla (**M**) consists of mostly of columnar or polyhedral chromaffin cells, which form clusters and anastomosing cords separated by sinusoids (H&E × 40)

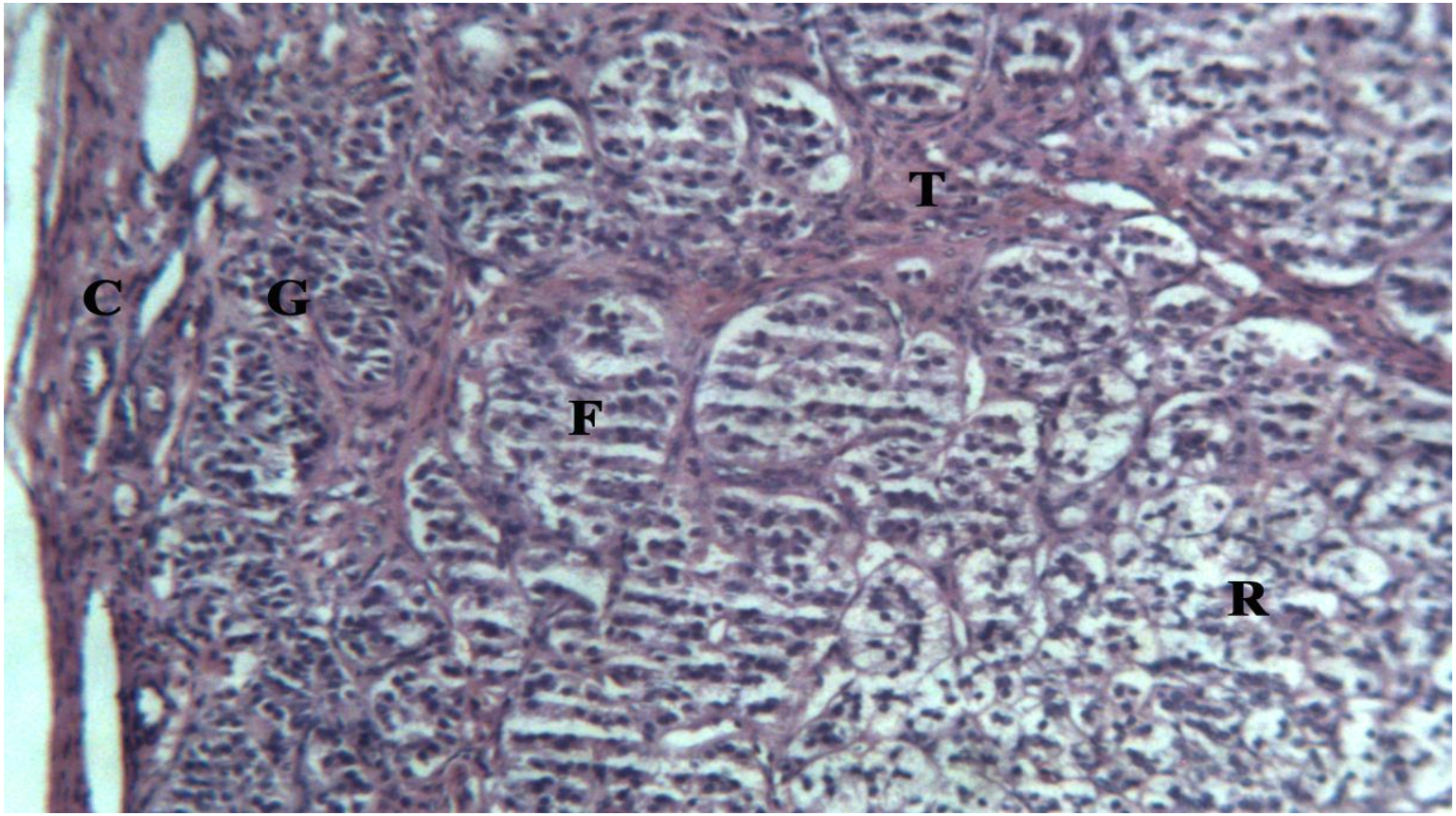


Plate 4.32: Photomicrograph of Adrenal gland tissue section (transverse) among the male of Group A. Note Capsule (**C**) and it trabeculae (**T**) and it fenestration into the cortex. The zones in the adrenal cortex, Glomerulosa (**G**) which is the outer zone lying immediately behind the capsule (**C**) and composed of cells arranged in irregular and tight ovoid clusters. The zona fasciculata (**F**) is the middle of the three cortical zones; consist of cells in irregular circular cords inside straight columns. The zona reticularis (**R**) is the innermost layer of the adrenal cortex have irregular network of anastomosing cords of cells surrounded by sinusoids (H&E × 100).

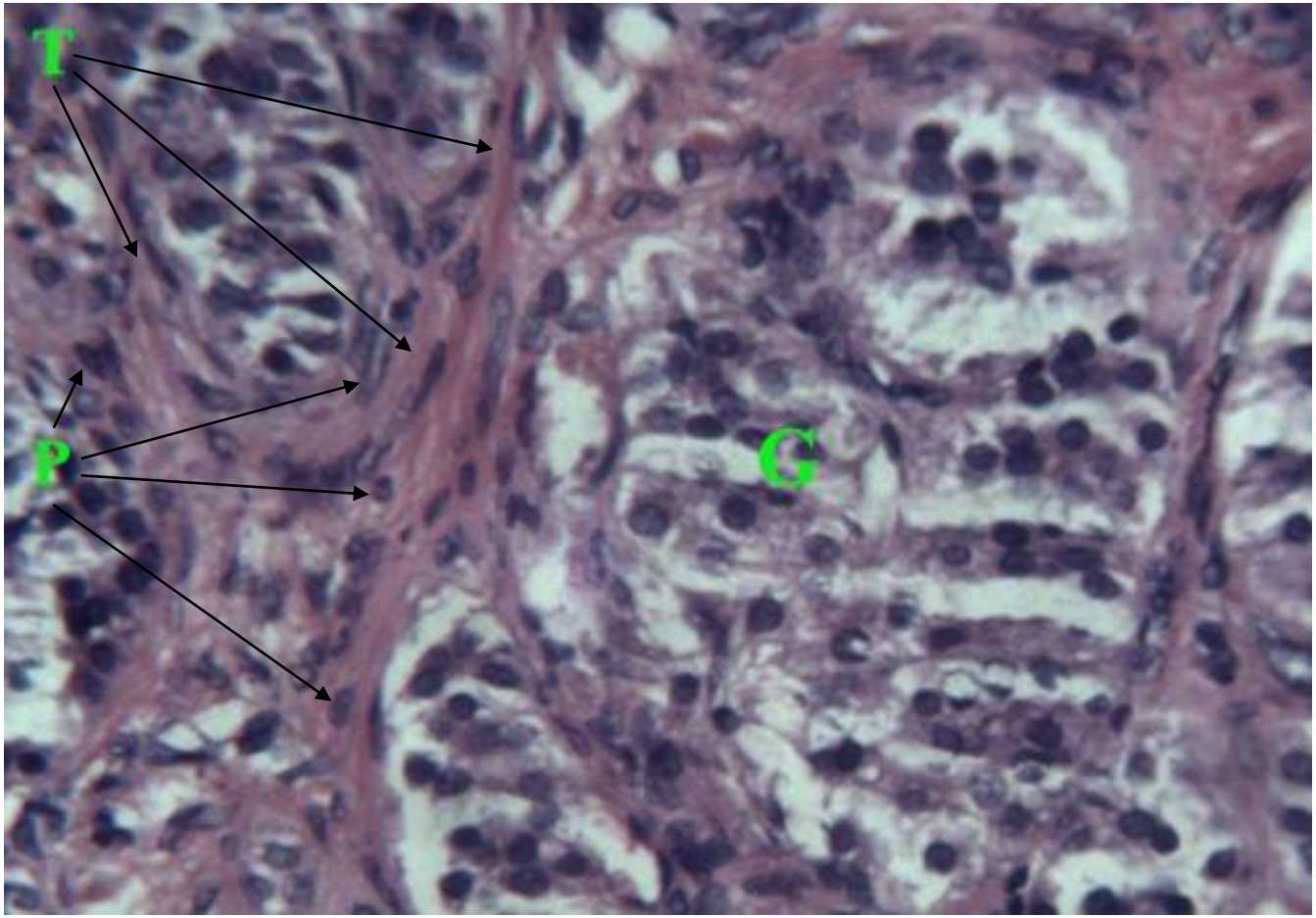


Plate 4.33: Photomicrograph of Adrenal gland tissue section (transverse) among the male of Group A showing a higher magnification of the Zona glomerulosa (**G**), the polyhydral shape of the parenchymal cells (**P**), and the arrangement of the trabeculae (**T**) (H&E × 250).

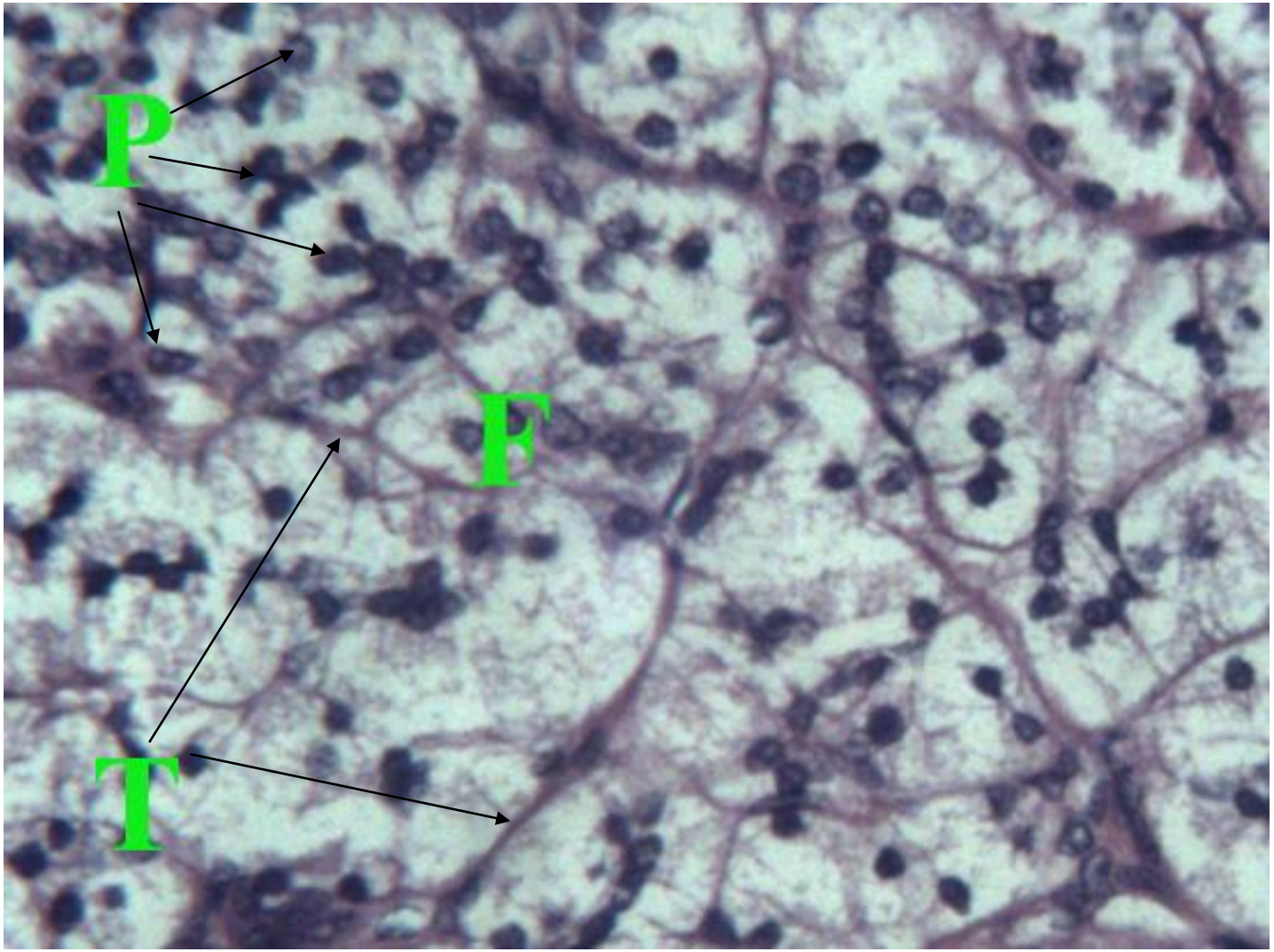


Plate 4.34: Photomicrograph of Adrenal gland tissue section (transverse) among the male of Group A, showing higher magnification of the Zona fasciculata (**F**), noting the thin trabecular (**T**) network, and the polyhedral shape of the parenchymal (**P**) cells and their sparse arrangement (H&E × 250).

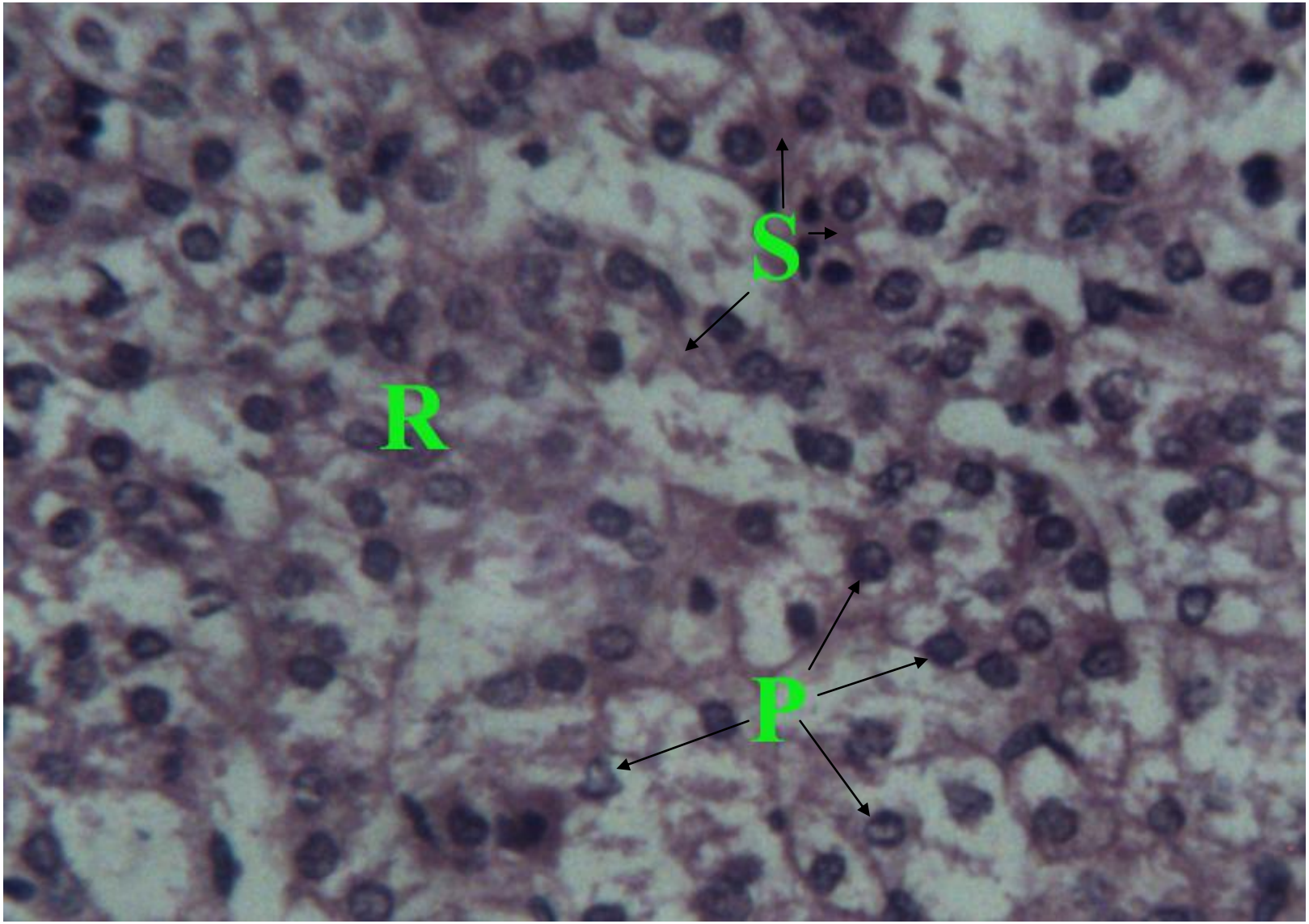


Plate 4.35: Photomicrograph of Adrenal gland tissue section (transverse) among the male of Group A, showing magnification of Zona reticularis (**R**), irregular network of anastomosing cords of parenchymal cells (**P**) polyhedral in shape surrounded by sinusoids (**S**) (H&E × 250).

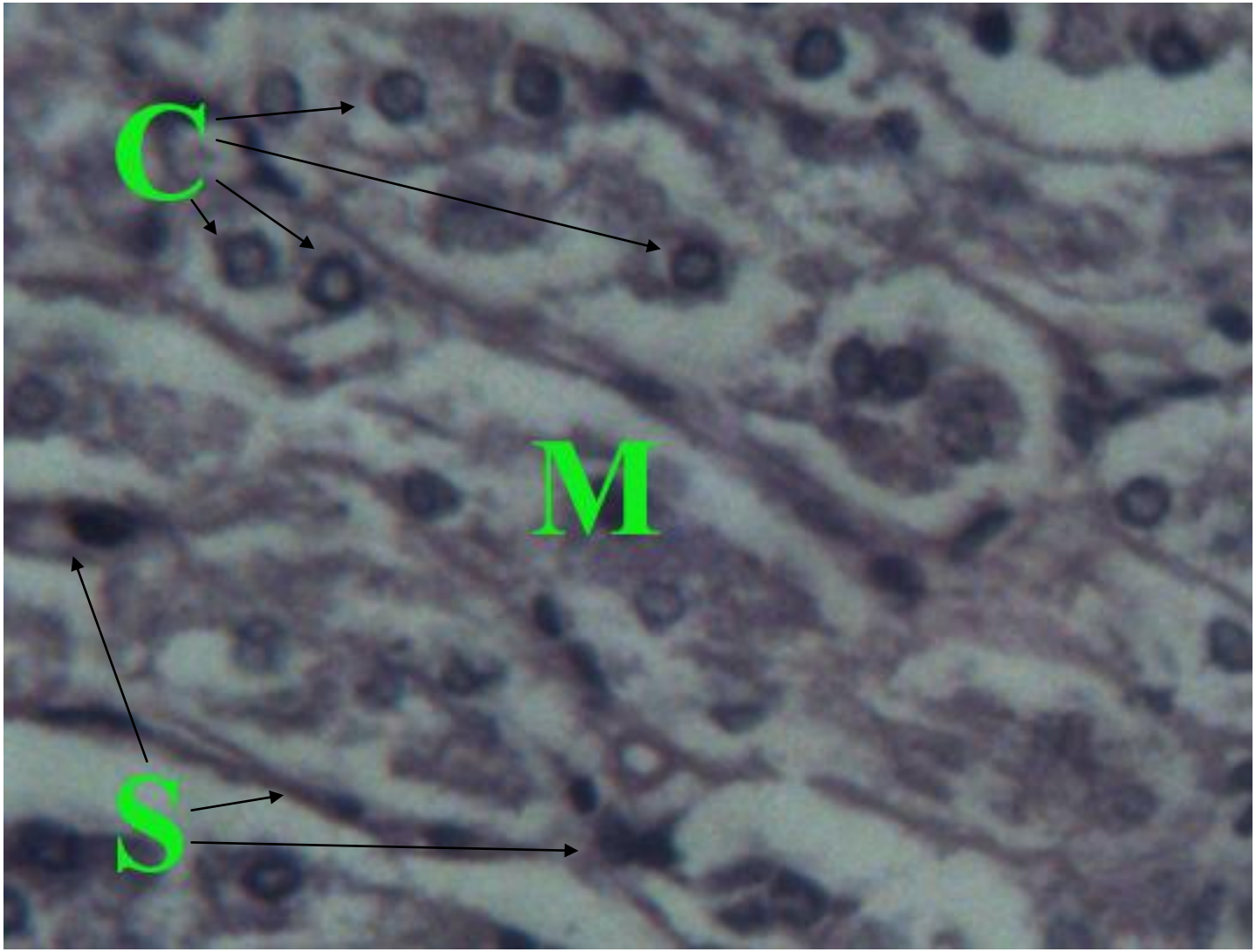


Plate 4.36: Photomicrograph of Adrenal gland tissue section (transverse) among the male of group A, showing the adrenal medulla (**M**) with polyhedral shaped chrommafin cells (**C**) surrounded by sinusoids (**S**) (H&E × 250).

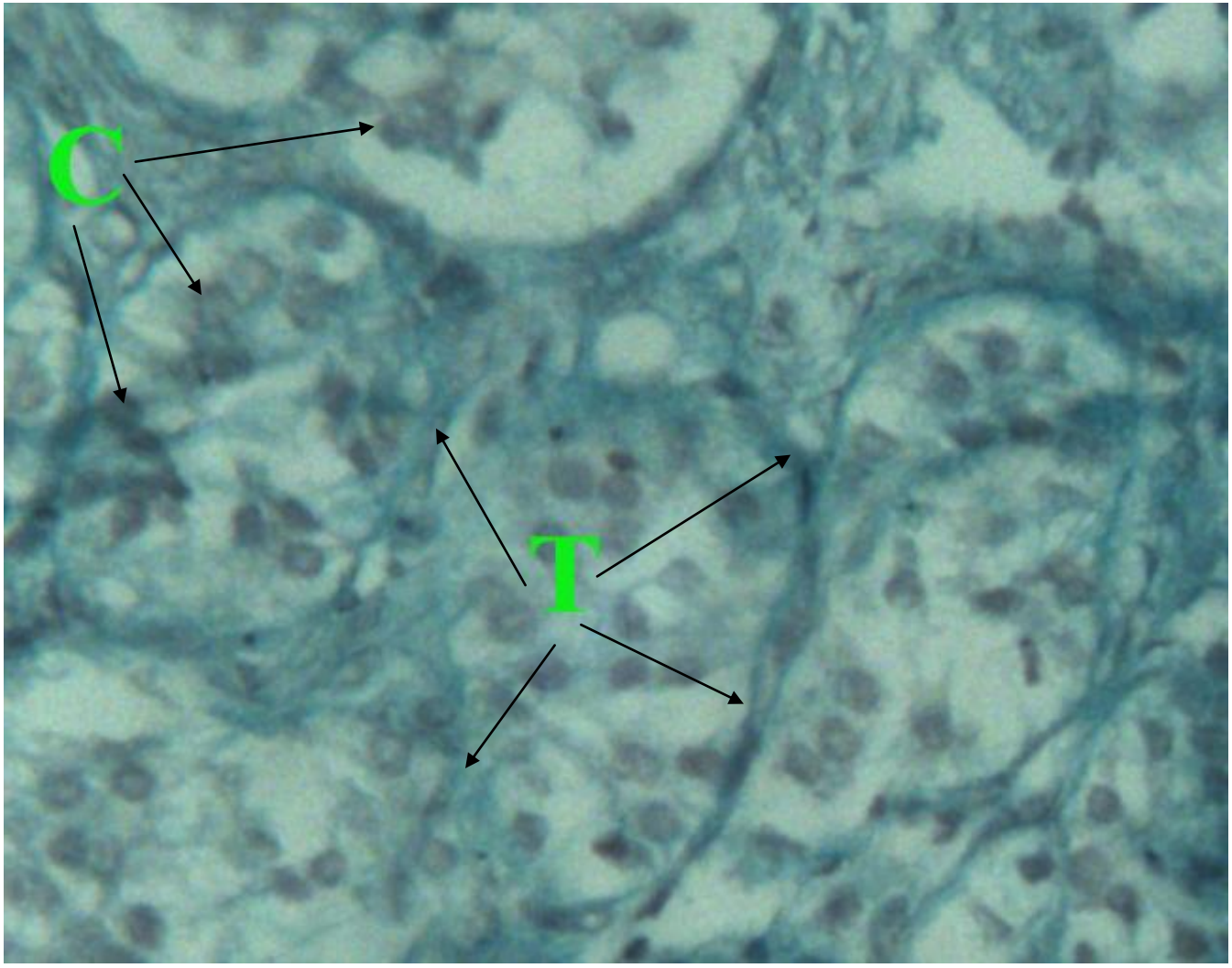


Plate 4.37: Photomicrograph of Adrenal gland tissue section (longitudinal) among the group A (male). Note the connective tissue trabecular (**T**) stained blue, the nucleus of the cells (**C**) (Alcian Blue × 250)

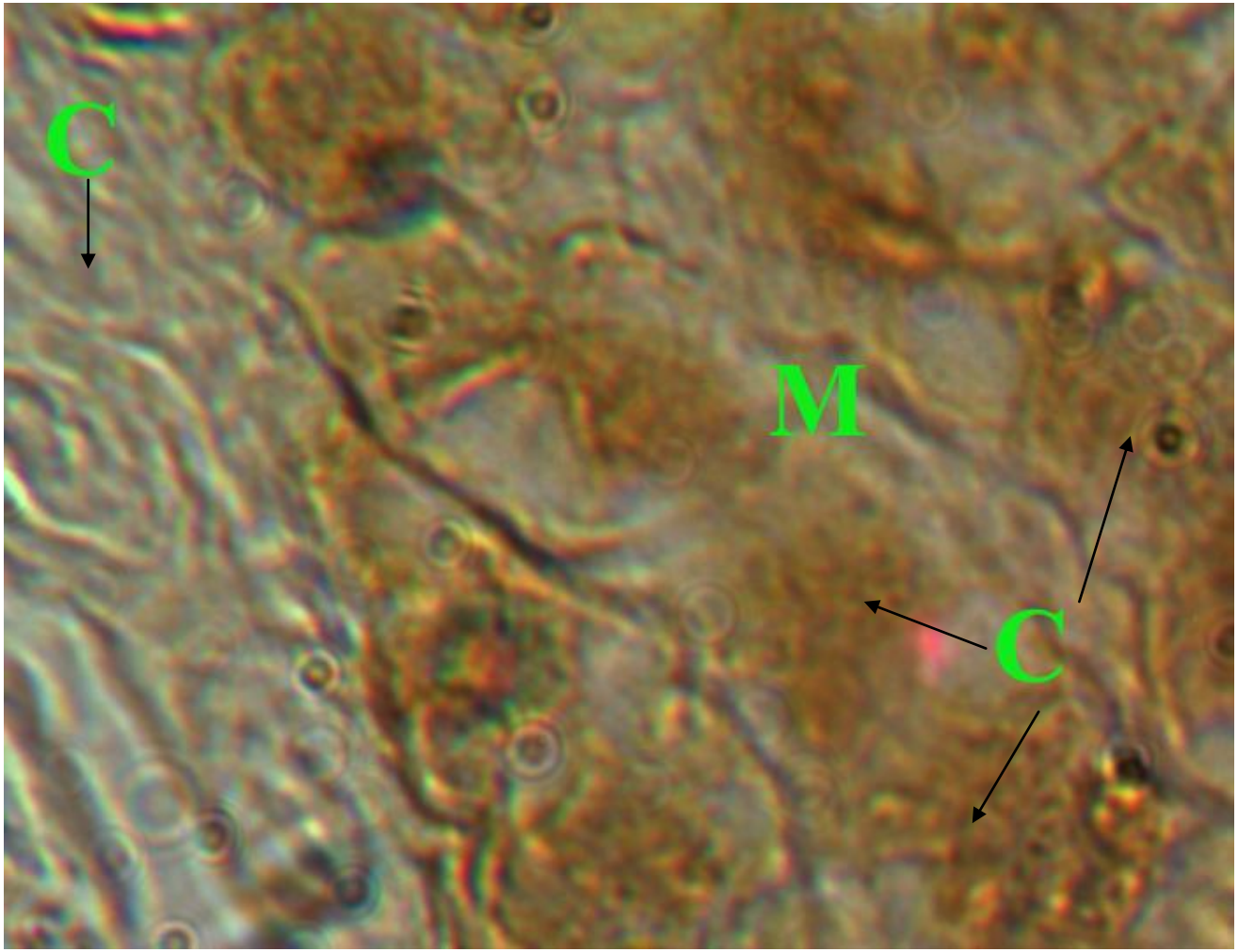


Plate 4.38: Photomicrograph of Adrenal gland tissue section (transverse) among the Group A (male), showing chromaffin cells (**C**) in the medulla (**M**) been oxidized to brown color by the potassium dichromate. Note the cortex (**C**) (potassium dichromate × 450).

Group B:

This group (young adults) shares the same features with the previous group having thick capsule of dense irregular connective tissue as shown in Plates 4.39, 4.40 and 4.41. Thin trabeculae of the connective tissue capsule projecting into the parenchyma of the cortex reaching far deep into the zona fasciculata, more in the males than the females in this group (Plates 4.39, and 4.41). Some part of the trabeculae coils a little bit to form an inverted u-shaped arrangement (Plates 4.39 and 4.41).

The adrenal gland tissue is organized into two parts; the peripheral cortex and a central medulla (Plates 4.39 to 4.42).

The adrenal cortex consists of Zona glomerulosa or zone multiformis which is the outermost zone (Plates 4.39, 4.40, and 4.41). The parenchymal cells of this region are columnar and polyhedrally arranged into arcs or clusters or cords as an inverted U-shaped formation by the trabeculae (Plates 4.39, 4.41).

The zona fasciculata is the next layer, formed by arranged cords in straight columns of cuboidal or polyhedral cells separated by sinusoids. The cytoplasm of the cells in this zone frequently appears foamy because of the presence of numerous lipid vacuoles (Plates 4.39, 4.40 and 4.41).

The zona reticularis is the innermost zone of the adrenal cortex (Plates 4.40 and 4.42). It is arranged as an irregular network of anastomosing cords of cells surrounded by sinusoids. The cells are polyhedral in shape (Plates 4.40 and 4.42).

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There is appearance of zona intermedia in the female of this group, which is situated between the zona fasciculate and zona reticularis (Plate 4.40).

The adrenal medulla is composed mostly of columnar or polyhedral chromaffin cells, which form clusters and anastomosing cords separated by sinusoids, and have basophilic cytoplasm (Plates 4.40 and 4.42).

The medulla may appear within the zona reticularis, because the cortex and medulla interdigitate at their junction (Plates 4.40 and 4.42).

When the gland was fixed in potassium dichromate fixative, the stored catecholamine granules of adrenal medullary cells (chromaffin cells) were oxidised to a brown colour (Plate 4.44).

Outer and inner zone of the medulla can often be distinguished. The former consists of larger, more darkly stained cells, while the latter contains smaller, more lightly stained cells (Plate 4.44).

Alcian Blue special histochemical stain was positive with the trabeculae of the connective tissue capsule, and the sinusoids within the adrenal parenchyma (Plate 4.43).

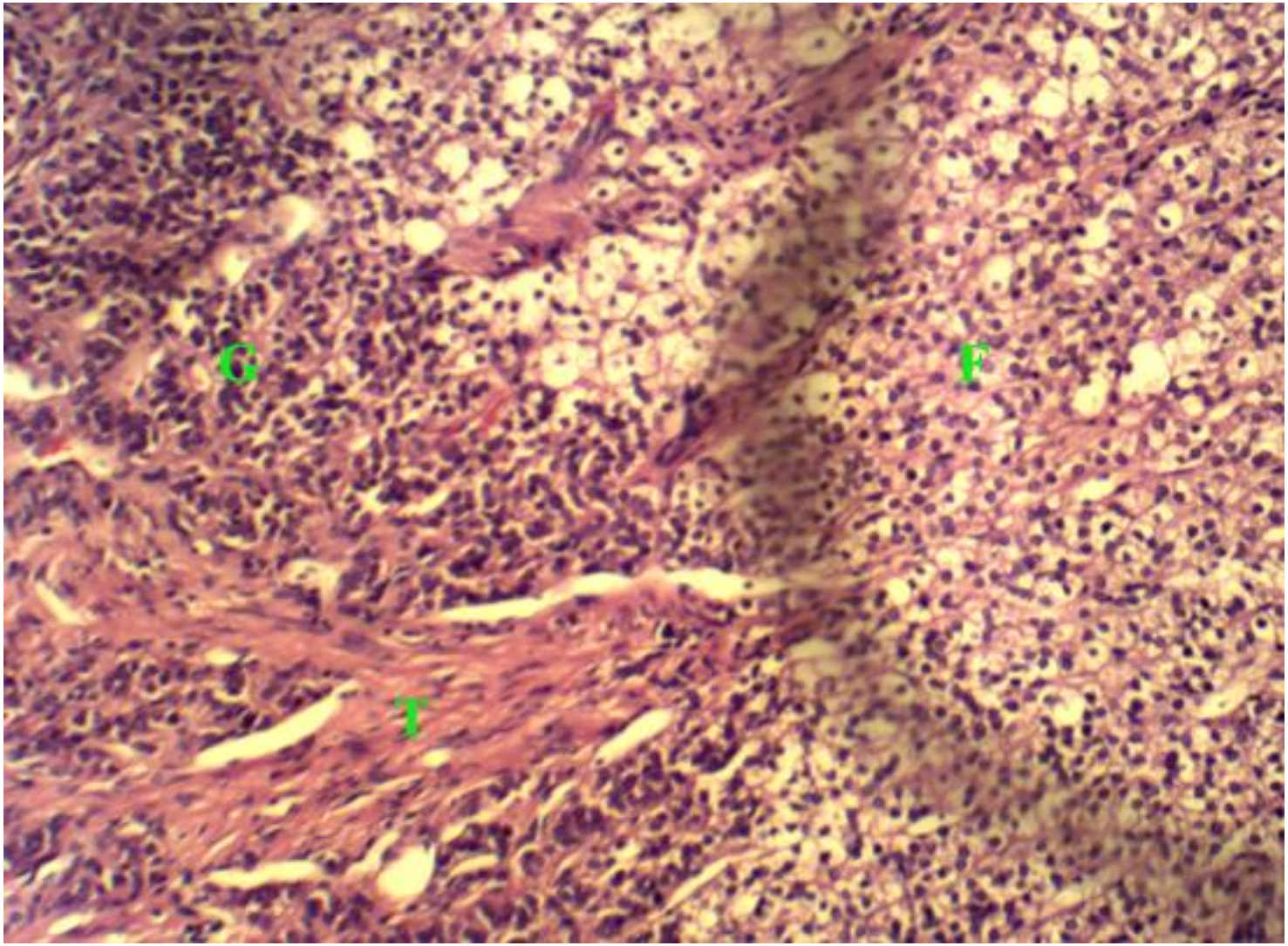


Plate 4.39: Photomicrograph of adrenal gland tissue section (longitudinal) among the female of Group B showing the connective tissue trabeculae (**T**) extension from the capsule, the zona glomerulosa (**G**), and zona fasciculata (**F**) (H&E × 100).

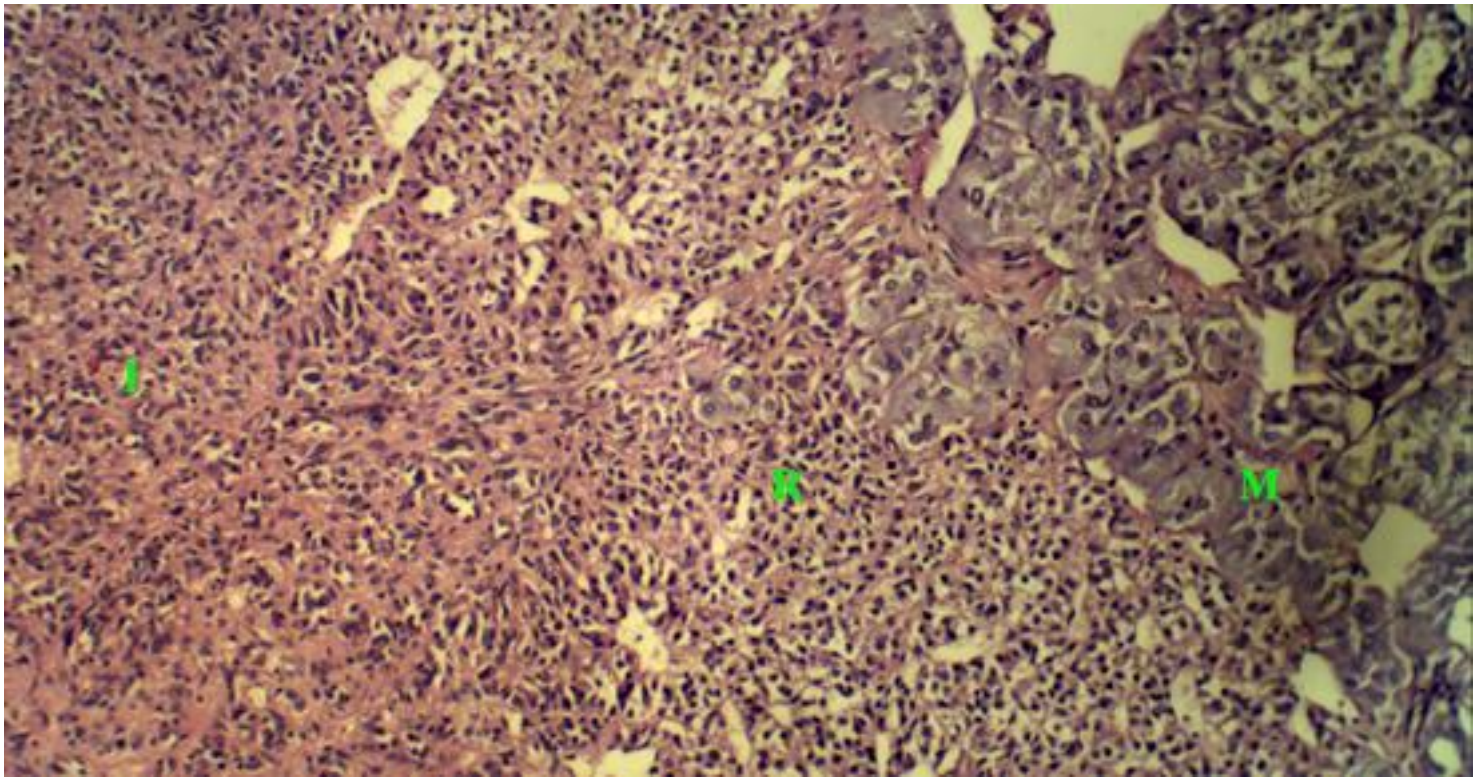


Plate 4.40: Photomicrograph of adrenal gland tissue section (longitudinal) among the females of group B, showing the presence of zona intermedia (**I**), zona reticularis (**R**), and the adrenal medulla (**M**) (H&E × 100).

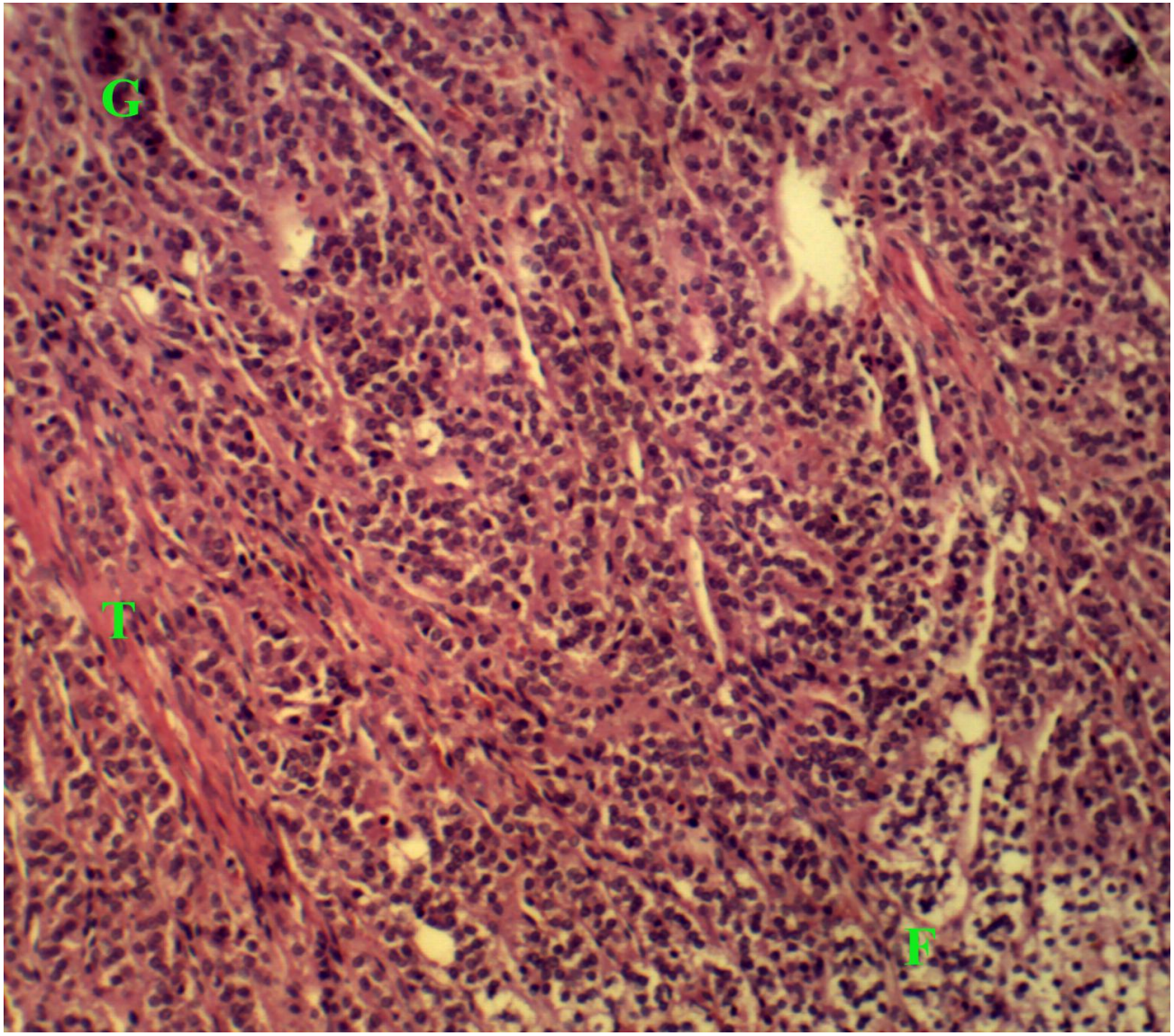


Plate 4.41: Photomicrograph of adrenal gland tissue section (longitudinal) among the males of Group B, showing the connective tissue trabeculae extending into the zona gromerulosa (**G**) and zona fasciculata (**F**) (H&E × 100)

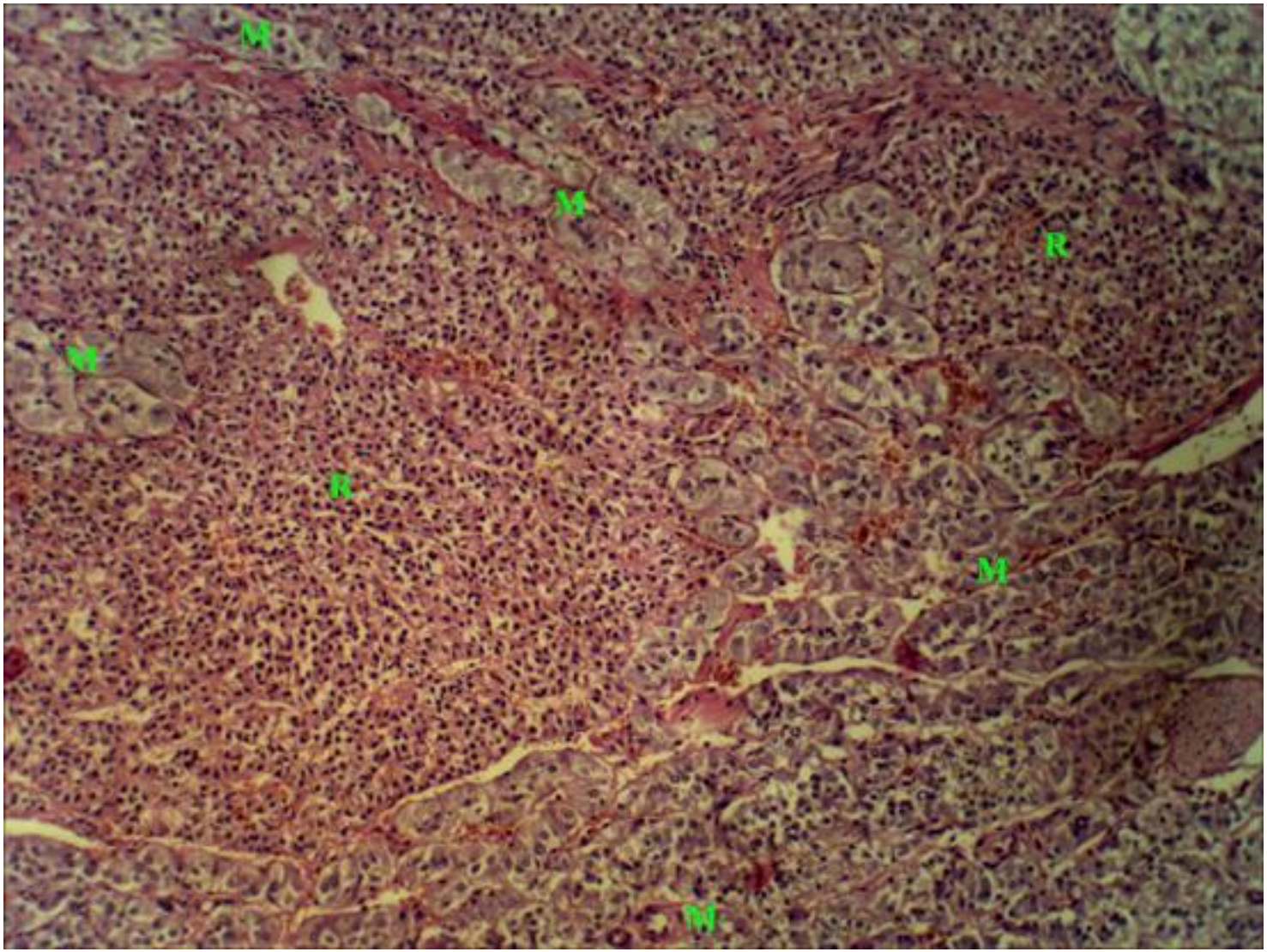


Plate 4.42: Photomicrograph of adrenal gland tissue section (longitudinal) among the male of Group B, showing that the adrenal medulla (**M**) tissue interdigitate with zona reticularis (**R**) of adrenal cortex. Note the medulla tissue (**M**) in the middle of zona reticularis (H&E × 100).

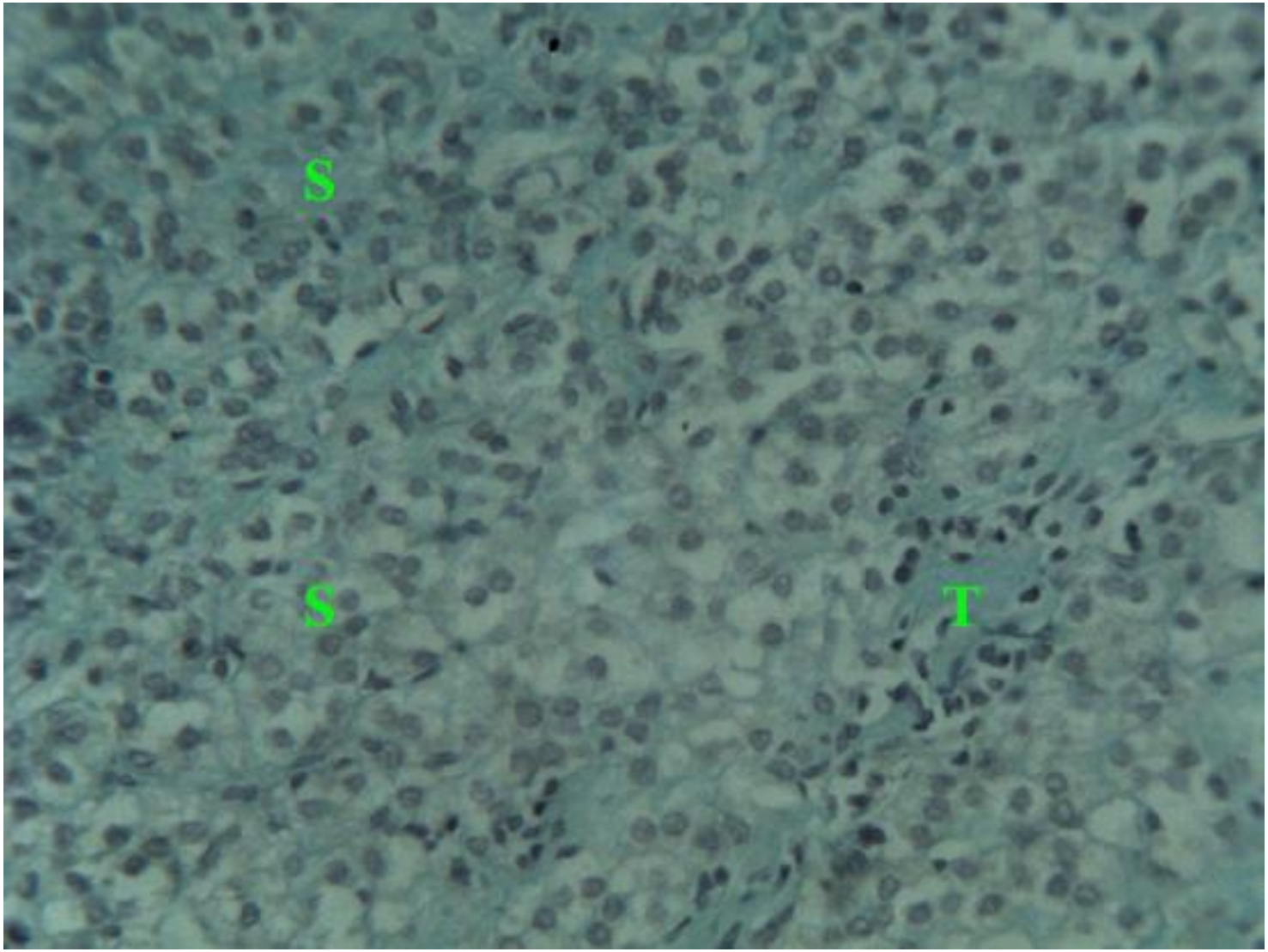


Plate 4.43: Photomicrograph of adrenal gland tissue section (transverse) among the Group B (female), showing the reaction of the sinusoids (**S**) and the connective tissue trabeculae (**T**) with the Alcian blue reagent, the color turned light blue color (Alcian Blue × 250)

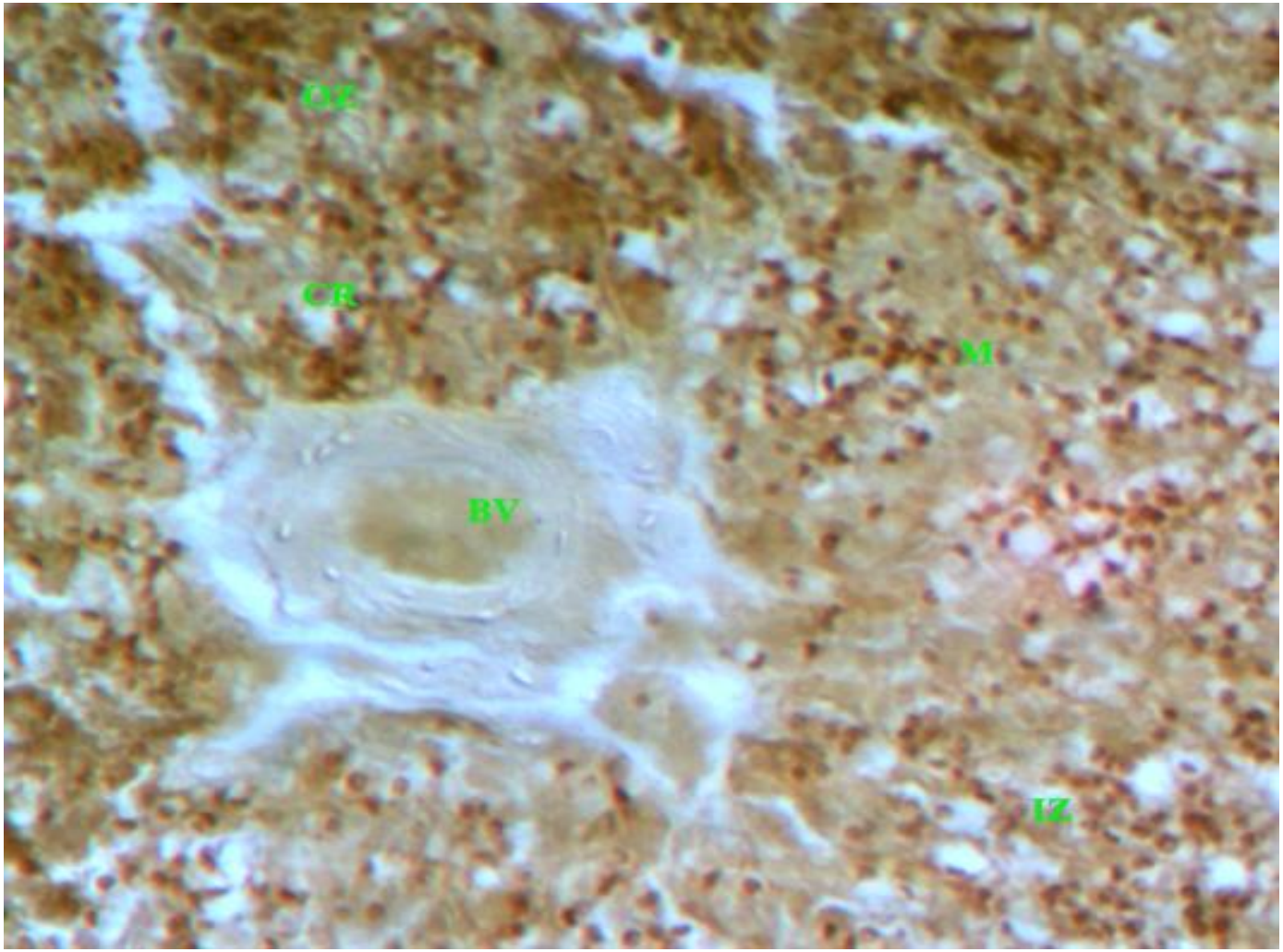


Plate 4.44: Photomicrograph of adrenal medulla tissue section (longitudinal) among the Group B (female), showing chromaffin cells reaction (**CR**) with potassium dichromate, and **inner** zone (**IZ**) and **outer** zone (**OZ**) of the adrenal medulla (**M**), and blood vessels **(BV)** (Potassium Dichromate × 150).

Group C:

This group (old adults) shares similar features with the previous group having thick capsule of dense irregular connective tissue as shown in Plates 4.45, and 4.46. Thick trabeculae of the connective tissue capsule project into the parenchyma of the cortex reaching far deep into the zona fasciculata, more in the male than the female in this group (Plates 4.45 and 4.46). Some part of the trabeculae coils a little bit to form an inverted u-shaped arrangement (Plates 4.45 and 4.46).

The adrenal gland tissue is organized into two parts; the peripheral cortex and a central medulla. The adrenal cortex consists of Zona glomerulosa or zone multiformis is the outermost zone (Plates 4.45 and 4.46). The parenchymal cells of this region are columnar and polyhedral arranged into arcs or clusters or cords as an inverted U-shaped formation by the trabeculae (Plates 4.45 and 4.46).

The zona fasciculata is the next layer, formed by arranged cords in straight columns of cuboidal or polyhedral cells separated by sinusoids. The cytoplasm of the cells in this zone frequently appears foamy because of the presence of numerous lipid vacuoles (Plates 4.45 and 4.46).

The zona reticularis is the innermost zone of the adrenal cortex. It is arranged as an irregular network of anastomosing cords of cells surrounded by sinusoids. The cells are polyhedral in shape (Plate 4.47).

The adrenal medulla is composed mostly of columnar or polyhedral chromaffin cells, which form clusters and anastomosing cords separated by sinusoids, and have basophilic cytoplasm (Plate 4.47).

When the gland was fixed in potassium dichromate fixative, the stored catecholamine granules of adrenal medullary cells (chromaffin cells) are oxidised to a brown colour (Plate 4.49).

Outer and inner zone of the medulla can often be distinguished. The former consists of larger, more darkly stained cells, while the latter contains smaller, more lightly stained cells (Plate 4.49).

Alcian Blue special histochemical stain was positive with the trabeculae of the connective tissue capsule, and the sinusoids within the adrenal parenchyma (Plate 4.48).

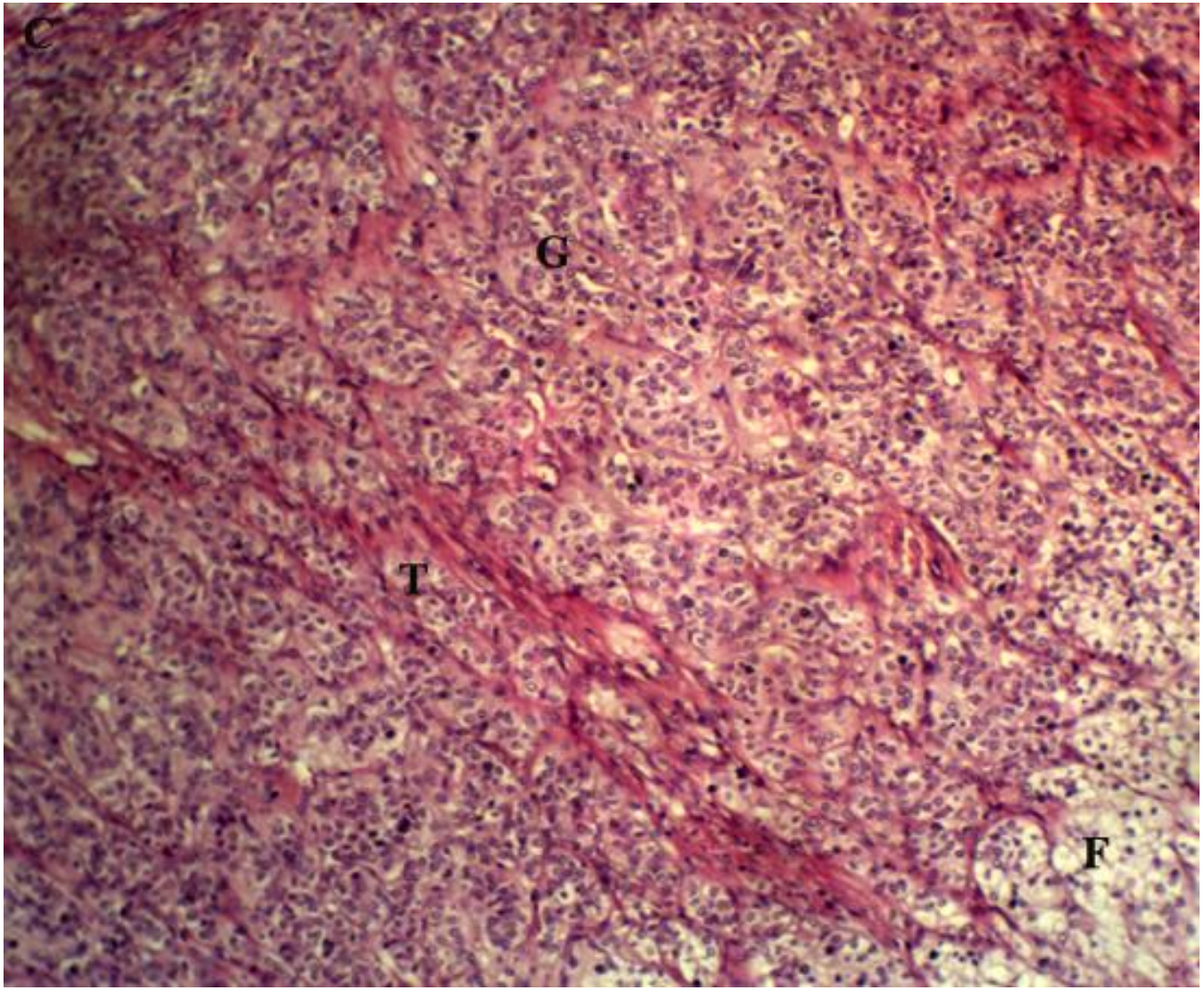


Plate 4.45: Photomicrograph of adrenal gland tissue section (longitudinal) among the female of group C, showing the connective tissue capsule (**C**) and trabeculae (**T**), the zona glomerulosa (**G**) and the zona fasciculata (**F**) (H&E × 100)

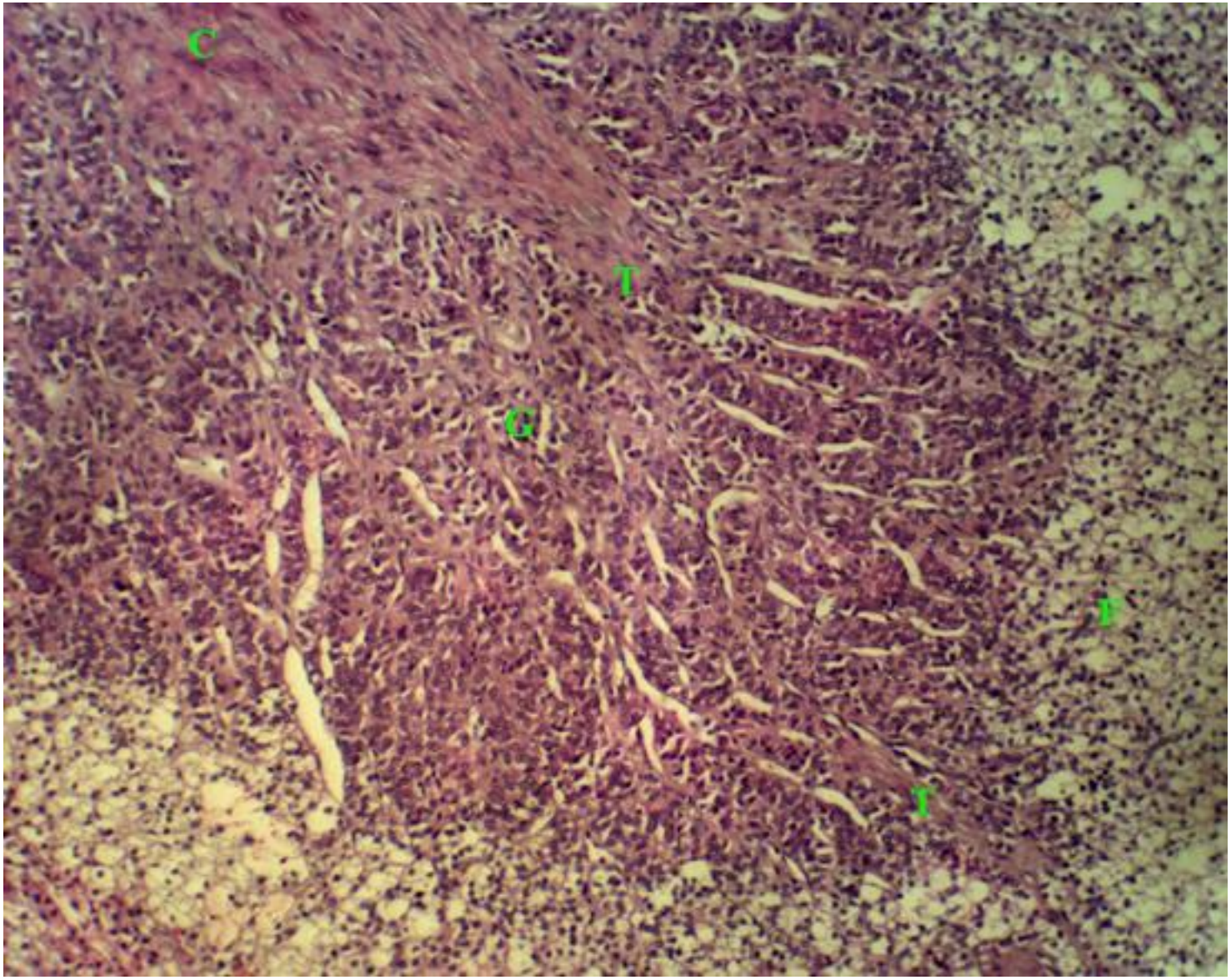


Plate 4.46: Photomicrograph of adrenal gland tissue section (transverse) among the male of group C, showing the connective tissue capsule (**C**) and trabeculae (**T**), the zona glomerulosa (**G**) and the zona fasciculata (**F**) (H&E × 100)

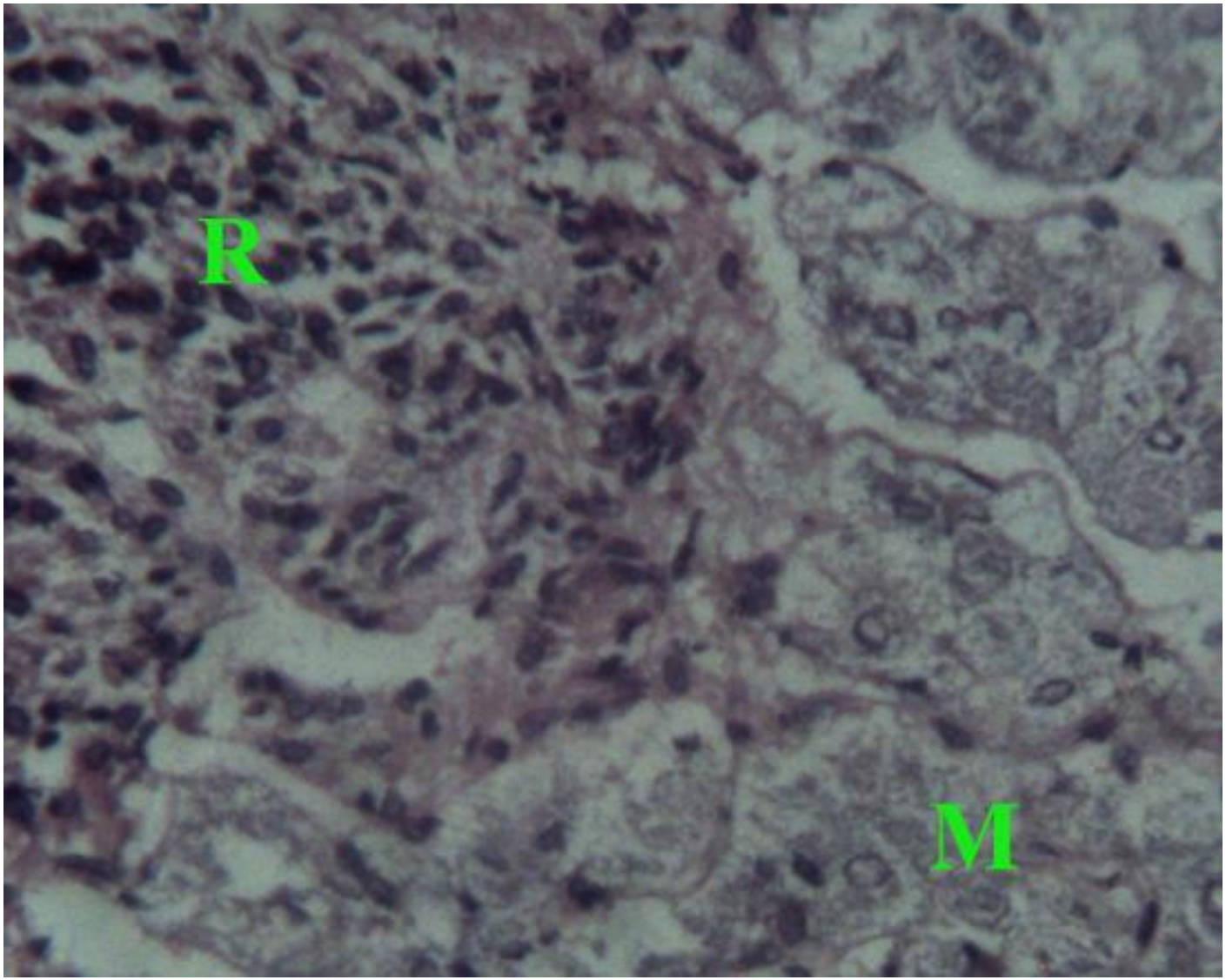


Plate 4.47: Photomicrograph of adrenal gland tissue section (transverse) among the male of group C, showing the zona reticularis (**R**), and the adrenal medulla (**M**) (H&E × 250).

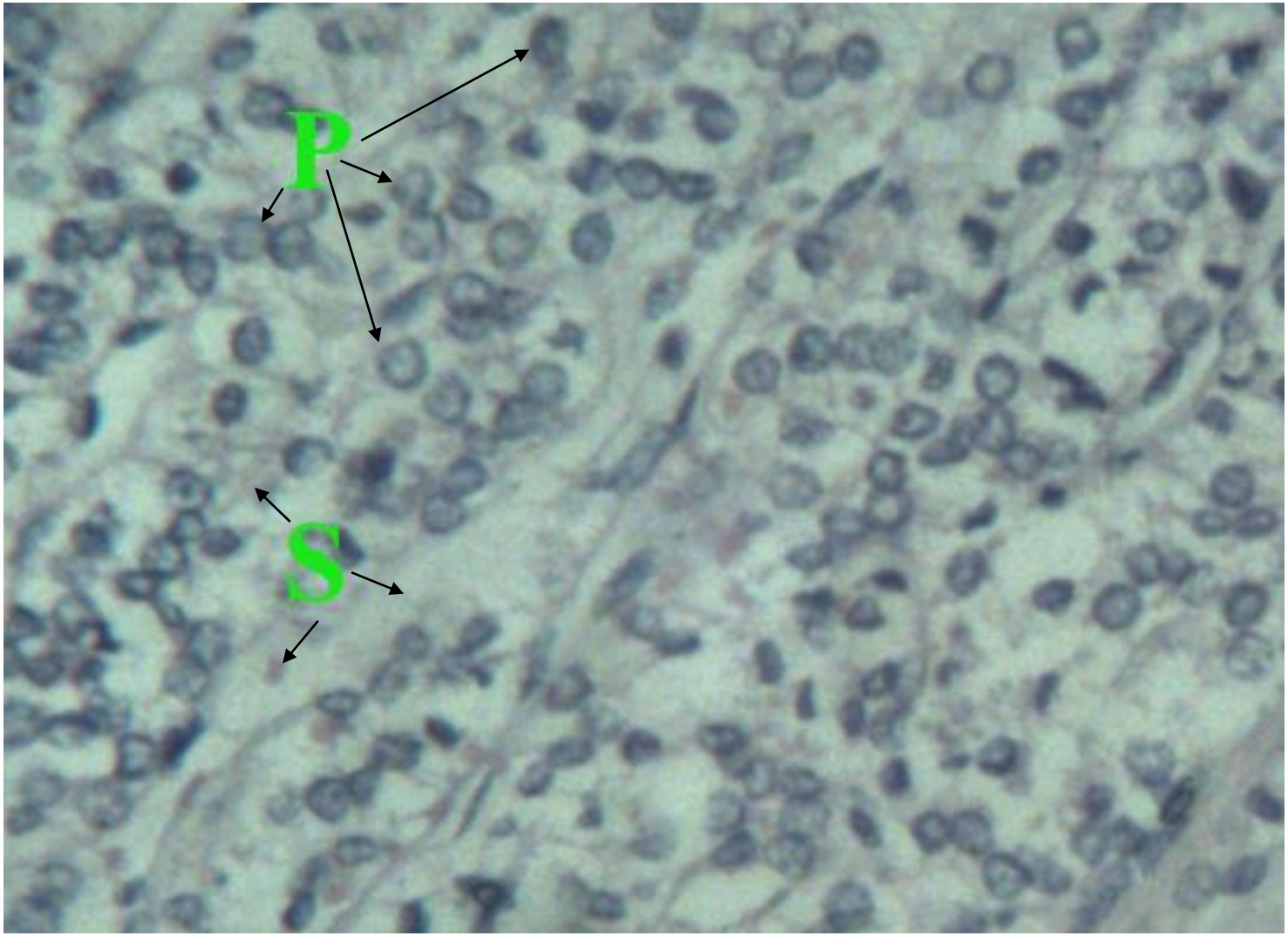


Plate 4.48: Photomicrograph of adrenal gland tissue section (transverse) among the male of Group C, showing the parenchymal cells (**P**), and sinusoids (**S**) which colored light blue to colorless after staining with Alcian blue reagents (Alcian Blue × 100).

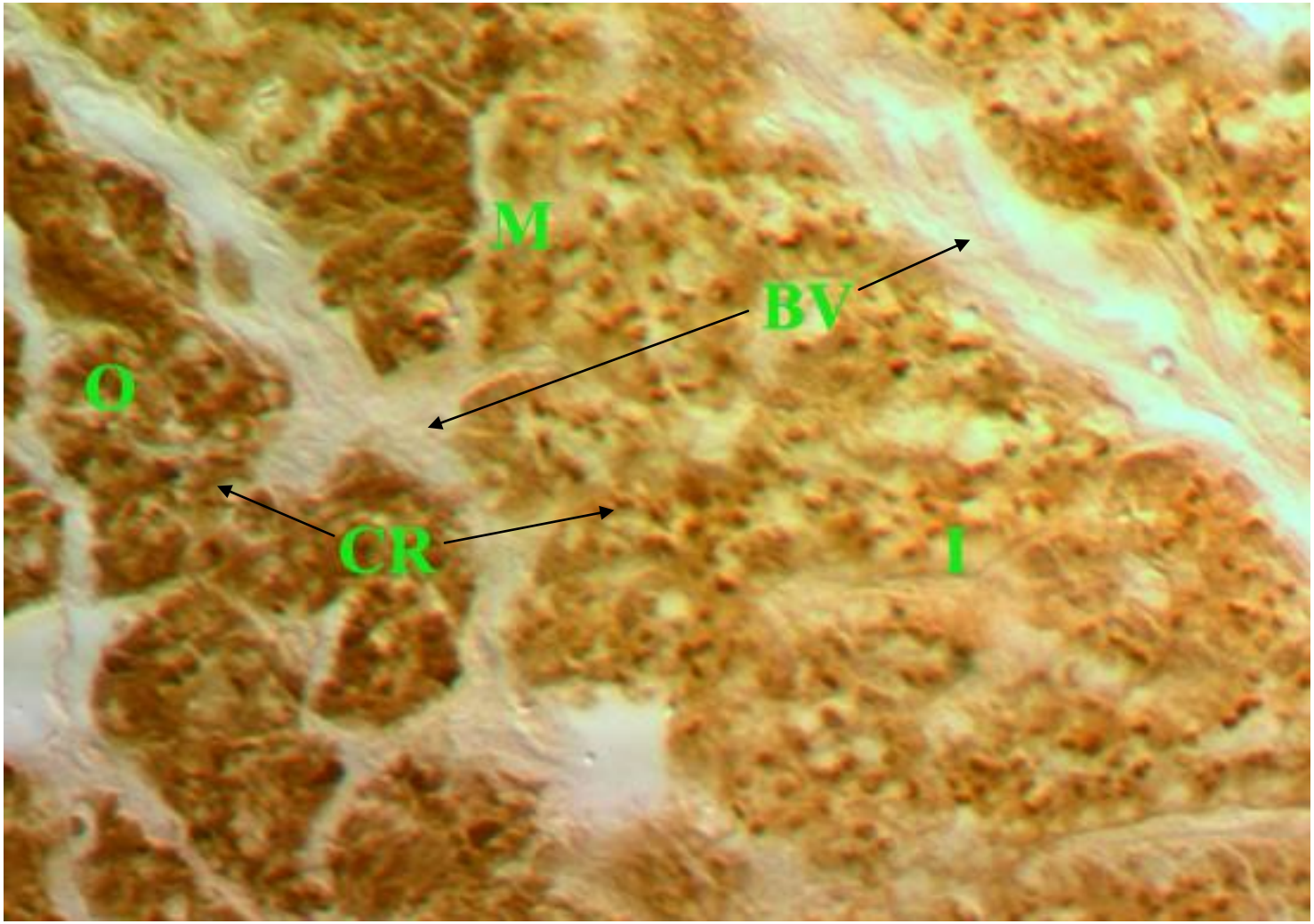


Plate 4.49: Photomicrograph of adrenal medulla tissue section (transverse) among the male of Group C, showing chromaffin cells reaction (**CR**) with the potassium dichromate fixative, the **outer zone** (**O**) is distinct from the **inner zone** (**I**) within the *adrenal* medulla (**M**) (potassium dichromate × 400).

CHAPTER FIVE

DISCUSSION

The gross anatomical study of the thyroid gland of indigenous one-humped camel showed that it was a paired-lobe organ located on the lateral surface of the trachea, joined together at inferior pole ventrally by an isthmus which passes on the ventral surface of the third (3^{rd}) to fifth (5^{th}) tracheal rings. The thyroid gland has lengthy lobes that extend from lower boarder of the cricoid cartilage and up to the fifth (5^{th}) tracheal ring. This conforms with what was found in the one-humped camel in Pakistan by Kausar and Shahid (2006) and in Iran reported by Ahmadpanahi and Yousefi (2012). The thyroid gland lobes in cattle, sheep and dog are asymmetric in position (McDonald, 2003). The thin isthmus that connects the lobes of the thyroid gland in camel in this study is similar to what was found in the horse (Dyce *et. al.*, 2002). The human thyroid gland is similar to the bilobes and isthmus arrangements present in the indigenous one-humped camel but the lobes, in humans, is positioned anterior to the second, third and fourth tracheal rings (Singh, 2006; Moore and Dalley., 2006). In rats, mice and African giant rats (AGR), the isthmus is located at the caudal end of the lobes (Ingbar, 1985) different from the presentation found in the present study.

The present study revealed that the thyroid glands were seen as reddish brown organs, which agreed with the work on one-humped camel in Iran (Ahmadpanahi and Yousefi, 2012), and in Pakistan (Kausar and Shahid, 2006).

The present work revealed that the arterial blood supply to the thyroid gland in one-humped camel indigenous to Northern Nigeria are from superior thyroid arteries, middle thyroid arteries and inferior thyroid arteries, but this is different from what is found in humans which are supplied, usually by two arteries; superior and inferior thyroid arteries, and also thyroidea ima artery in 10% of humans (Moore and Dalley., 2006; Singh, 2006).

Morphometric findings in the present work revealed that the weight, length, width and thickness of the thyroid gland in adult one-humped camel indigenous to Northern Nigeria agreed with the previous works done on one-humped camel in Pakistan, Egypt, Sudan and Iran. The ranges of the morphometric values of the thyroid gland of one-humped camel previously studied were: weight ranges 45 – 55 g, length ranges 3.5 – 6.5 cm, width ranges 2 – 4 cm and thickness ranges 0.1 – 1 cm (Kausar and Shahid, 2006; Ahmadpanahi and Yousefi, 2012), which were higher than those of the humans, horses, rats, mice, AGRs, and most of the other domestic ruminants (Ingbar, 1985; Dyce *et. al*.,, 2002; Hajorska, 2002; Capen and Martin, 2003; Moore and Dalley., 2006). The result from the present study showed that there was sexual dimorphism in length of the thyroid gland among the juvenile age group, with the females having significantly higher values than the males. The width in the group C showed significant differences with the males having higher values than in the females.

The morphometric values (weight, length, width and thickness) of the thyroid gland in the indigenous one-humped camel, regardless of the age groups, were found to be slightly higher in females than in males in weight and length, while males have slightly higher values than the females in width and thickness of the thyroid gland. These findings were similar to the findings of Ahmadphanahi and Yousefi (2012) on sexual dimorphism in the width of the thyroid gland of one-humped camel and the invariability in the thickness, but this present work differed with his work in the weight of the thyroid gland of the dromedary camel, which the author found that sexual dimorphism was significant. The result of this study corresponds with the report on Wistar rats by Capen and Martin (2003) that the weight of the thyroid gland was greater in females than in males but not statistically significant.

The result of comparison of the morphometric variables (weight, length, width, and thickness) between the two lobes (right and left) of the thyroid gland within each of the groups revealed that there were no significant differences between them, hence they are symmetrical, this conforms with that of humans (Singh, 2006; Moore and Dalley., 2006). However, thyroid gland lobes in cattle, sheep and dog have been found to be asymmetric in position (McDonald, 2003).

The present study revealed that the weight of the thyroid gland in one-humped camel was directly proportional to the age of the animal, especially in the juvenile age to the young adult age, which showed sharp increase, however, it was not so from the young adult to old adult.

Amazingly, the findings in the present study on the concentration of the thyroid hormones (T3 and T4) synthesized and secreted by the thyroid gland showed that the concentrations of the thyroid hormones were inversely proportional to the weight and the age of the thyroid gland in the indigenous one-humped camel. Correspondingly, the histological studies revealed the appearances of numerous vacuolated colloids within the follicles in the young adult group and the old adult group, and apoptosis of the cells of the thyroid cells in the young adult, and old adult. However, in the juvenile age group the vacuoles were less or absent, and have intact thyroid cells without any sign of death of cells. This signifies that due to lack of the need for growth and development in the adult indigenous one-humped camel, the follicular cells that are responsible for the synthesis and secretion of one of the hormone responsible for those roles die, while some follicles develop vacuoles to reduce the quantity of the hormones. These coincide with the findings in the humans where weight, length of thyroid gland decreased in approximately proportional level with age (Abdullah *et.*

al., 2010). The number of thyroid follicles and parafollicular cells were decreased with progressive increase of age (Abdullah *et al*., 2010).

Parafollicular cells were demonstrated using the Azure A Acid Hydrolysis special stains which distinctly differentiated the parafollicular cells from the follicular cells resolving the controversy of the existence or non-existence of the parafollicular cells in the thyroid gland of the one-humped camel (Kausar and Shahid, 2006; Rejeb *et al*., 2011). Although, Ahmadpanahi and Yousefi (2012) had reported that thyroid gland of the one-humped camel contained parafollicular (C) cells, they were about 5% of the cell population.

The general histological features of the thyroid glands of dromedary camel were similar to those of other mammals (Abdel-Magied *et. al.*,, 2000). In this study the sinusoids were seen more prominent and numerous in the females of juvenile age, and young adult than in male of the same groups, while the males of old adult have more prominent blood vessels than the females in the thyroid gland tissue. This may have linked with the higher values in morphometric variables of weight, length in the females over the males of group A and B, as well as male over the female in the old age group.

Follicles of various sizes appeared across all the groups (A, B and C). This is in conformity with what was found in most mammals (Bacha and Bacha, 2000). Vacuoles appeared in the follicles of group B and group C. The vacuoles were larger in group B than in group C. The vacuoles occupied more than half of the tissue in males than in the females of group B. The vacuoles are relatively more in males of group C. This may explain the morphometric studies why females thyroid gland had greater weight than the males, because the males follicles were more vacuolated, hence less colloid content, which may contribute to the overall weight of the thyroid gland of one-humped camel.

The shape of the thyroid gland follicles in the indigenous one-humped camel in all the groups of this study were round, oval and irregular, but it was more irregular in males of group C.

Follicular epithelial cells encircled the follicles**,** but the follicular cells of neighboring follicles frequently, come into contact with each other and disrupt the continuity of the basal lamina as seen within the follicles/ lobules, and are more prominent in males than in females of group A, but more prominent in females than in males of groups B and C.

The follicular cells are mostly resting/ inactive, and low cuboidal in shape, the follicles were filled with colloids and had smoother peripheral surface of the follicles with little or no vacuoles, in both males and females of group A. While the groups B and C follicular cells were seen in an active secretive stage, more columnar in shape in females than the males who were more cuboidal in shape. Some of the cells showed some level of apoptosis and high occurrence of vacuoles in the follicles, which were more prominent in females than in males. These variations in histology between the juvenile age group and other two adult groups were indeed related to the hormonal secretions of the thyroid cells, where it was established in our study that the thyroid hormones had higher concentrations of T3 and T4 in the juvenile age than in adult (group B and C), as it has been established that thyroid hormones play vital role on growth and development (Chan and Singer, 1993) which were more in juvenile age than adult age.

The slightly higher morphometric values and histological variations in the females than the males in all the groups showed that the females may have higher metabolic activities than the male especially in group A (juvenile) and group B (young adult). This may be related to

the reason why female dromedary camels reach sexual maturity around three years of age and mate around age four or five. Males begin to rut by age three, but do not reach full sexual maturity until six years old (Mukasa-Mugerwa, 1981).

Parafollicular cells or C cells indeed occur among the follicular cells and between them in all the age groups with some degree of certainty, using Azure A Acid Hydrolysis special stains which deeply stained the parafollicular cells purple blue. This have solve the controversy about the existence of parafollicular or C-cells in the dromedary camel, where Kausar and Shahid (2006) and Rejeb *et. al.,* (2011) argued that parafollicular cells were absent in thyroid glands of camel. However, Ahmadpanahi and Yousefi (2012) had reported the presence of parafollicular cells, which the present study confirmed.

In the present study, the parathyroid glands were found to be two pairs of glands around both the right and left thyroid glands. This corresponds with what McDonald (2003) had reported that most animal species consisted of two pairs of parathyroid glands in the anterior cervical region. However, in this study accessory parathyroid glands do occur, by description, as right middle parathyroid glands and left middle parathyroid glands in some of the animals interchangeably, embedded into the thyroid gland, internally positioned. Most of the superior and inferior parathyroids are external glands positioned some little distance from the thyroid gland. Similar result was found in other mammals, especially in the Pig and Cow (Bacha and Bacha, 2000).

Sexual dimorphism in the weights of the right and left inferior parathyroid glands in both the males and the females were higher in group A (juvenile age). It was significant in the length of the right superior and inferior parathyroid glands. The differences in the width of the right and left inferior parathyroids were significant in both the males and females. There was

sexual dimorphism in the thicknesses of the right superior and inferior parathyroids. In the group B (young adult), it was only in the width that sexual dimorphism was highly significant in the female right superior parathyroid. The left inferior parathyroid was also significant. Group C (old adult) shows no significant differences between the sexes in all the morphometric variables within the groups.

The general histological structures found in the present study of the parathyroid gland in the one-humped camel indigenous to Northern Nigeria was identical generally to the other mammals. Specifically it was identical to other camel (Nagpal *et. al.*,, 2001), to cow (Charles *et. al.*, 2000) in goat (Roy *et al*., 1999) and in humans (Singh, 2006). The parathyroid glands are surrounded by a thin capsule of connective tissue. It has abundant stroma of connective tissue, with big smooth muscles all over, no difference among all the age groups. This conforms to what were found in the pig and cow especially the well developed stroma of connective tissue, but is sparse in other mammals (Bacha and Bacha, 2000).

In many studies it was reported that mammal parathyroid gland contains two types of cells; the principal cells (or chief cells) and the oxyphil cells (Dellmann, 1993; Ojo, 1987; Bacha and Bacha, 2000; Fleischer, *et. al.*, 2004; Singh, 2006). The same was found in the present study.

There were abundance of adipose cells especially in the males of the young adult (group B), while in the group C (old adult) was abundant in females. It was not seen in the younger groups (group A).

The result of histochemical studies in all the age groups (A, B and C) showed that the tissue of the parathyroid gland, in indigenous one-humped camel, reacted negatively to the

Periodic Acid Schiff (PAS)**.** This was because the parathyroid gland tissues were normal, as PAS is used to detect glycogen in tissues that have abnormal excess glycogen or they specifically store glycogen.

The adrenal glands or suprarenal glands were paired flattened endocrine organs found resting on the superior pole of the kidneys (Dellmann, 1976; Ojo, 1987), situated close to the anterior end of the kidneys (Bacha and Bacha, 2000), and located in retroperitoneal space along the medial cranial poles of the kidneys in most mammals (Prasad and Sinha, 1981; Rosol *et. al*., 2001).

However, this study revealed that the right adrenal was found superior medial to the right kidney, a little bit far away from the superior pole of the kidney and found to be attached to the inferior medial surface of the liver, while the left adrenal gland was found to be superior medially away from the left kidney. The right adrenal gland was triangular in shape, while the left adrenal gland was irregularly oval in this study, which agrees with the previous studies on the suprarenal gland of the dromedary camel (Ahmadphanahi and Yousefi, 2012). The right gland of dromedary camel according to Nabipour *et. al.,* (2008) was approximately bean shaped, whereas the left gland is almost triangular. This also corresponds, to the shape found in most mammals as reported by Bacha and Bacha (2000).

Sexual dimorphism was significant in the group A (juvenile age) with the weight of the right and left adrenal glands significantly higher in the females than in the males, so it was in the thickness of the left adrenal gland. It was significant also in the length of the right adrenal gland of the same group, but with the males higher than the females. It means that young female dromedary camels had heavier and thicker adrenal glands than their male counterparts. And these glands are important in producing corticosteroids and androgenes hormones by it cortex which helps in sodium and water retention in response to stress, it also

affects muscles. The zona glomerulosa produces the mineralocorticoids, aldosterone and deoxcoticosterone which affect the electrolyte balance in the body (Singh, 2006). The zona fasciculata produces glucocorticoids, cortisone, cortisol, and small amount of androgen (Singh, 2006), which have wide effects including carbohydrate metabolism and protein metabolism (Singh, 2006). The cells of the zona reticularis also produce some glucocorticoids and sex hormones (both oestrogen and androgen) (Singh, 2006; Moore and Dalley, 2006). With the above mentioned higher morphometric values in females more than in males and explained the corresponding functions of the adrenal tissues, it may explain the reason why the female dromedary camels get matured quicker and earlier than the males, have more endurance and more active than their male counterparts.

Histologically, the adrenal glands of mammals are covered by a capsule of dense irregular connective tissue that sometimes contains smooth muscle (Bacha and Bacha, 2000), which was found to be true in this study of the indigenous one-humped camels. Clusters of epithelioid cortical cells occur in the capsule in most mammals (Bacha and Bacha, 2000), indigenous one-humped camel with this study inclusive. Thin trabeculae projected partially into the parenchyma as reported by Bacha and Bacha (2000). This study observed that the trabeculae reach far deep into the zona fasciculata, more in the females than the males. In males, the trabeculae coil to form a cluster like arrangement.

Each adrenal gland is organized into two parts the cortex and a central medulla (Bacha and Bacha, 2000; Moore and Dalley, 2006). The same was found in the one-humped camels in this study. The adrenal cortex consists of Zona glomerulosa, Zona fasciculata, and Zona reticularis. Dellmann (1993) and Ojo (1987) had reported the same thing in most mammals including humans. In gunea pigs three zones were reported (Colby *et. al.,* 1992), as well as

in rats (Moog *et. al*., 1998), and in Buffalo and in some other mammals (Banks, 1993). However, Bacha and Bacha (2006) had reported additional zona intermedia in some animals. This work conforms with the above works; with zona intermedia found in the females of group B (young adult), which is situated between the zona fasciculate and zona reticularis. The arrangements and nature of the cells found in these zones and the medulla were similar to what was found in most mammals as reported by Bacha and Bacha (2000).

Histochemical staining of the adrenal tissue with alcian blue revealed how the connective tissue trabeculae and the sinusoids reacted with alcian blue reagent and stained it purple blue. Similarly, potassium dichromate fixative demonstrated the chrommafin cells of the adrenal medulla as it reacted with the stored catecholamine granules of the cells (chromaffin cells) and oxidised them to brown colour.

CHAPTER SIX

SUMMARY, CONCLUSION AND RECOMMENDATION

6.1 Summary

The gross anatomical study of the thyroid gland of indigenous one-humped camel showed that it was a paired-lobe, long that extended from lower boarder of the cricoid cartilage to the fifth (5^{th}) tracheal ring. The work revealed that the blood supplies to the thyroid gland in one-humped camel indigenous to Northern Nigeria are from superior thyroid arteries, middle thyroid arteries and inferior thyroid arteries. Sexual dimorphisms were not significant in all the morphometric values of weight, length, width and thickness of the thyroid gland, regardless of the age groups in the indigenous dromedary camel, however the females had slightly higher values than the males in weight and length, while males had slightly higher values than the females in width and thickness of the thyroid gland. The correlation of the weight of the thyroid gland in one-humped camel was directly proportional to the age of the animal. While the correlation of the concentration of the thyroid hormones (T3 and T4) were inversely proportional to the weight and the age of the thyroid gland in the indigenous one-humped camel, meaning that thyroid hormones concentrations were higher in the young than in the adult. Histological studies revealed the appearances of numerous vacuolated colloids, and apoptosis on the follicular cells of the adult, which further support the above correlation. The thyroid gland in indigenous one-humped camel had follicular cells and parafollicular cells. The parafollicular cells were demonstrated using the Azure A Acid Hydrolysis special histochemical stains which distinctly differentiate the parafollicular cells from the follicular cells to settle the controversy of the existence or non-existence of the parafollicular cells in the thyroid gland

of the one-humped camel. Periodic Acid Schiff (PAS) special stains reacted negatively to the colloid in the thyroid gland follicles in all the groups.

The parathyroid glands were usually two pairs of glands situated around thyroid gland lobes. However, accessory parathyroid glands may be situated, either on the right or on the left, described as right middle parathyroid gland and left middle parathyroid gland. Sexual dimorphism was significant in the weight, length, width and thickness in some of the parathyroid glands in juvenile age group. In the adult groups sexual dimorphism were not significant in the morphometric variables, it was only among the young adult group that sexual dimorphism was significant only in the width of the female right superior parathyroid. The general histological structures of the parathyroid gland found in this study of the one-humped camel indigenous to Northern Nigeria was identical generally to other mammals. The parathyroid gland consisted of densely packed chief cells (light and dark cells), oxyphil cells, and abundance of adipose cells in the adult males and females, but absent in the younger group. The cells reacted negatively to PAS. The right of the paired adrenal gland was positioned superior medial to the right kidney attached to the inferior medial surface of the liver, while the left adrenal gland was superior medially positioned held within fat from the left kidney. The right adrenal gland was triangular in shape, while the left adrenal gland was irregularly oval. Sexual dimorphism was statistically significant in the weight and thickness of adrenal gland of the females in the juvenile age group, inferring that the young females dromedary camel had heavier and thicker adrenal glands than the males. Histological studies showed that adrenal glands have cortex and medulla. The adrenal cortex comprised of three zones, and younger and older camel, but four zones in young adult groups; Zona glomerulosa, Zona fasciculate, Zona intermedia, and Zona reticularis. Chromaffin cells of the medulla were identified.

6.2 Conclusion

The study established that:

- ✓ That the arterial blood supply to the thyroid gland of one-humped camel were from superior, middle and inferior thyroid arteries
- ✓ There was sexual dimorphism in the morphometric parameters of weight, length, thickness, and width of the thyroid gland with female indigenous one-humped camel having higher values, with the exception of width which was higher in male old adult indigenous one-humped camel.
- ✓ The thyroid hormone had higher concentrations of T4 (8.21±0.57µg/dl) and T3 (8.21±0.57µg/dl) in young indigenous one-humped camel than in adult.
- ✓ Thyroid gland of indigenous one-humped camel had parafollicular cells as demonstrated by Azure A Acid Hydrolysis special stains
- ✓ The indigenous one-humped camel have accessory parathyroid glands positioned between the superior and inferior parathyroid glands
- ✓ There was sexual dimorphism in the morphometric variables of width, length and weight of the parathyroid glands of indigenous one-humped camel with the females having higher values than their male counterparts
- ✓ There was abundance of adipose cells in the parathyroid glands of the adult indigenous one-humped camel, but not seen in the juvenile camels.
- ✓ The adrenal glands of indigenous one-humped camel was found to be not resting on top of the superior pole of the kidney, as it were on other animals, but the right adrenal gland was found to be attached to attached to the inferior medial surface of the liver, while the left adrenal gland embedded in fat superior medially away from the kidney

- ✓ Sexual dimorphism was significant on the weight and thickness of the adrenal glands of the indigenous one-humped camel with female higher than male.

Generally, the gross, morphometric, histological, histochemical and hormonal studies of the thyroid, parathyroid and the adrenal glands of the indigenous one-humped camel showed age, hormonal and morphometric correlations, sexual dimorphisms and cellular morphology which were pertinent to enhance the understanding of how the animal adapts to its environment, as well, will provide baseline information for future related researches.

6.3 Recommendation

Further studies on seasonal effects, nutritional correlations, detail hormonal and immunohistochemical studies, expression of genes and neurochemical characteristics should be carried out to give further insight to understanding these endocrine glands, in order to unravel the mechanism of adaptation of indigenous one-humped camel to unfavorable weather conditions, as well as to provide basic clinical investigative information in endocrinology.

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