PATHOLOGY OF THE REPRODUCTIVE ORGANS IN EXPERIMENTAL HYPOTHYROIDISM IN GOATS

By
NEMALI MOHAN REDDY

THESIS

Submitted in partial fulfilment of the
requirement for the degree

DOCTOR OF PHYLOSOPHY

Faculty of Veterinary and Animal Sciences
Kerala Agricultural University

Department of Pathology
COLLEGE OF VETERINARY AND ANIMAL SCIENCES
Mannuthy - Trichur

1982

D E C L A R A T I O N

I hereby declare that this thesis entitled PATHOLOGY OF THE REPRODUCTIVE ORGANS IN EXPERIMENTAL HYPOTHYROIDISM is a bonafide record of research work done by me during the course of research and that the thesis has not previously formed the basis for the award to me of any degree, diploma, associateship, fellowship or other similar title of any other University or Society.

Signature of the candidate

Name of the candidate:
Nemali Mohan Reddy

Mannuthy,
15-11-1982.

C E R T I F I C A T E

Certified that this thesis, entitled PATHOLOGY OF THE REPRODUCTIVE ORGANS IN EXPERIMENTAL HYPOTHYROIDISM is a record of research work done independently by Nemali Mohan Reddy under my guidance and supervision and that it has not previously formed the basis for the award of any degree, fellowship or associateship to him.

15-11-82

Name of the Guide: Dr.A.Rajan
(Chairman, Advisory Board)

Designation : Professor and Head
Department of Pathology

Mannuthy,
15-11-1982

ACKNOWLEDGEMENTS

I wish to express my esteemed and profound sense of gratitude to Dr.A.Rajan, M.V.Sc., Ph.D., Professor and Head, Department of Pathology and Chairman of the Advisory Committee for suggesting the problem, for his abiding interest, encouragement, meticulous guidance, unstinted criticism, advice and guidance throughout this work.

I express my gratefulness to Dr.M.Krishnan Nair, M.V.Sc., Ph.D., F.R.V.C.S., Dean, College of Veterinary and Animal Sciences and Member of my Advisory Committee for valuable advice, useful suggestion, generous help and co-operation during the course of the study.

I am grateful to Dr.G.Nirmalan, Ph.D., Professor and Head, Department of Physiology and Bio-chemistry, Dr.K.Chandrasekharan, Ph.D., Associate Professor, Department of Parasitology and Dr.M.C.Ramakrishna Pillai, Ph.D., Associate Professor, Department of Physiology and Bio-chemistry, Members of the Advisory Committee for their valuable help and suggestions.

I am extremely thankful to Dr.E.Sivaraman, Professor and Head, Department of Animal Nutrition, Dr.A.Ramakrishnan

Professor and Head, Department of Poultry Science and Dr.K.Prabhakaran Nair, Professor, Department of Animal Reproduction for providing the facilities in the laboratory.

I acknowledge with thanks the valuable help rendered by Dr.T.Sreekumaran, M.V.Sc., Ph.D. and other staff members of the Department of Pathology.

I wish to express my sincere thanks to my colleagues, in the Department of Pathology for the help and co-operation rendered throughout the work.

I am greatly indebted to late Dr.C.K.S.V.Raja, Professor, Department of Animal Reproduction for his precious advice and valuable help rendered to me throughout the present study.

I am thankful to the Andhra Pradesh Agricultural University for granting me the study leave to undergo the course. I am also thankful to the Kerala Agricultural University for providing me the senior fellowship during the course of study.

My thanks are due to Sri.T.K.Prabhakaran, for typing the manuscript neatly.

Lastly but not the least, I appreciate my wife for encouraging me in undertaking and completing the study silently and cheerfully tolerating all troubles.

Nemali Mohan Reddy

DEDICATED TO MY BELOVED PARENTS

TABLE OF CONTENTS

LIST OF TABLES

LIST OF ILLUSTRATIONS

Sl. No.	Graph No.	
15.	21.	Incidence of free loose head in experimental hypothyroidism.
16.	23.	Incidence of middle piece abnormalities in experimental hypothyroidism.
17.	25.	Incidence of tail abnormalities in experimental hypothyroidism.
18.	27.	Incidence of proximal protoplasmic droplets in experimental hypothyroidism.
19.	28.	Relative weight of the thyroid gland in experimental hypothyroidism.
20.	33.	Relative weight of the pituitary in experimental hypothyroidism.
21.	36.	Relative weight of the adrenal gland in experimental hypothyroidism.
22.	39.	Relative weight of the testis and epididymis in experimental hypothyroidism.
23.	51.	Relative weight of the seminal vesicle and the prostate gland in experimental hypothyroidism.
24.	56.	Relative weight of the ovary in experimental hypothyroidism.
25.	63.	Relative weight of the uterus in experimental hypothyroidism.

Photograph No.

26.	2.	Group I - Stunted growth in experiment i hypothyroidism - The buck on the left side is stunted in growth and dull and has matted hair. The matched healthy control animal is on the right side.
27.	3.	Group II - Stunted growth in kids in experimental hypothyroidism. Experimental

Sl. No.	Photograph No.	
		kid is stunted in growth compared to the age matched control animal on the left side.
28.	4.	Group IV - Stunted growth in experimental hypothyroidism. The control animal is on the left side.
29.	5.	Clinical symptoms - Experimental animal showing periorbital oedema and rough matted hair coat.
30.	6.	Clinical symptoms - Experimental animal- The animal is weak, has loose skin and matted rough hair coat.
31.	7.	Clinical symptoms - Experimental animal showing lacrimation and facial oedema.
32.	22.	Abnormal spermatozoa - Free loose heads.
33.	24.	Abnormal spermatozoa - Middle piece abnormality.
34.	26.	Abnormal spermatozoa - Abnormalities in tail.
35.	29.	Thyroid gland - Enlarged thyroid glands of the experimental group. The glands of the control animal are seen on the right side.
36.	30.	Group T1 - Thyroid - Follicles are seen lined by tall columnar epithelial cells. Lumen is devoid of colloid.
37.	31.	Group T2 - Thyroid - Numerous small follicles are seen lined by tall columnar epithelial cells. Follicular lumen is small and devoid of colloid.
38.	32.	Thyroid - Discontinuation of treatment with thiourea - Distended follicles are seen lined with flattened epithelial cells.

Sl. No.	Photograph No.	
39.	34.	Group T2 - Pituitary - Severe degree of hyperplasia of basophils and chromophobes. Vacuolation and formation of cysts are also evident.
40.	35.	Pituitary - Acidophils are seen in normal proportion. Vacuolation in few of the basophils is still evident after discontinuation of treatment with thiourea.
41.	37.	Adrenal-Group T1 - Hyperplasia and hypertrophy of cells of the zona fasiculata. In focal areas the cells were devoid of fat.
42.	38.	Adrenal-Group T2 - Hypertrophy and hyperplasia and scattered fatty cysts in the zona fasciculata.
43.	40.	Testis and Epididymis- Group I - Reduction in the size of the testis and epididymis. The testis and epididymis of the control animals are on the left side.
44.	41.	Testis and Epididymis - Group II - Reduction in the size of the testis and epididymis in experimental animals. Testis and epididymis of the control animals are seen on the left side.
45.	42.	Testis - Group T1 - Seminiferous tubules showing vacuolar and hyaline degeneration. Primary and secondary spermatocytes are also few.
46.	43.	Testis - Group T2 - A few scattered spermatogonial cells close to the basement membrane are seen in the seminiferous tubules. No evidence of spermatogenesis.
47.	44.	Testis - Group II - Severe degree of hyaline degeneration in the seminiferous tubules. Spermatogenesis is absent. A few scattered interstitial cells are seen.
48.	45.	Testis - After discontinuation of treatment

Sl. No.	Photograph No.	
		with thiourea - Seminiferous tubules are active and organised layering of spermatocytes, spermatids and sperms is seen. Slight degeneration is still evident.
49.	46.	Epididymis - Tubules showing absence of sperms. Interstitial tissue is predominant.
50.	47.	Epididymis - Tubules showing degenerative changes. There is deciliation of the epithelial lining. The cells are of low cubical flattened epithelial type. Interstitial tissue is oedematious.
51.	48.	Epididymis - Tubules are intact with mild degeneration. Sperms are seen in the tubules.
52.	49.	Caput epididymis - Deciliation of the epithelial lining of the tubules. The lining cells are low cuboidal to flattened epithelial type.
53.	50.	Corpus epididymis - Tubules showing degenerative changes. Interstitial tissue is oedematous.
54.	52.	Seminal vesicles - Group I1 - The cells lining the glands are small and inactive with no evidence of secretory activity.
55.	53.	Seminal vesicles - Group I2 - The glands are dilated and cells lining the glands are vacuolated and hyalinised. No secretory activity is seen. The interstitial tissue is oedematous.
56.	54.	Prostate - Group I1 - The cells lining the glands show vacuolation.
57.	55.	Prostate - Group I2 - The acini are small and degenerative changes are seen.
58	57.	Ovaries - Inactive ovaries in group III.

Sl. No.	Photograph No.	
		The small ovaries and enlarged thyroids are seen on the side. The organs are from the control animals in the middle.
59.	58.	Ovaries - Smooth and small ovaries in group IV are seen at the bottom along with enlarged thyroid glands. Active ovaries and the thyroid glands from the control group are seen on the top.
60.	59.	Ovary - Cortex - showing loose cellular stroma and few atrophic follicles.
61.	60.	Ovary - Follicles showing degenerative changes in granulosa cell layer.
62.	61.	Ovary - Cortex - loose, odematous cellular stroma. Degenerating follicles are also seen.
63.	62.	Ovary - stromal tissue consisting of closely packed vacuolated elongated spindle shaped cells. Corpus albicans is also seen.
64.	64	Group III - The hypoplastic ovaries and uterus of the experimental animals are seen on the top. The normal ovaries and uterus of age matched control are at the bottom.
65.	65.	Group IV - Hypoplastic uterus from the experimental animal on the top - Uterus from the control animal at the bottom.
66.	66.	Group III. T1 - Uterus - The glands are small in size, inactive and lined by low cuboidal type of epithelium. No evidence of secretory activity is seen. Moderate interstitial oedema is also evident.
67.	67.	Group III. T2 - Uterus - The glands are small in size and the lining cells are cuboidal to flattened epithelial type. Moderate interstitial oedema is also seen.

67.	Photograph No.	
68.	68.	Group IV - Uterus - Uterine glands are small and slit like. Stroma is relatively loose.

CHAPTER I

INTRODUCTION

INTRODUCTION

The thyroid is an important endocrine gland which plays a pivotal role in maintaining the normal growth and reproduction of the animal. The gland is unique as it contains 70 to 80 per cent of the total body iodine in its product thyroxine. It has been well established that iodine and thyroid functions are closely related and the deficiency of this element could cause different thyroid disorders.

The existence of endemic goitre in man and animals due to absolute or relative deficiency of iodine has been recognised since long. Kelly and Snedden (1960) while documenting the geographical distribution of endemic goitre in Asia have demarcated certain regions in India as endemic zones of goitre and this included the coastal area in Kerala. It is known that endemicity of goitre parallels iodine deficient soil zones and it is also known that the iodine content of the soil varies with the geography of the land and climatic conditions. In heavy rain fall regions like Kerala loss of iodine due to leaching of surface soil is bound to happen. Further, the modern practice of adding synthetic nitrogenous

fertilizers containing no iodine is likely to cause iodine deficient soil. Besides this, the role of goitrogenic substances, widely distributed in nature, cannot also be overlooked in precipitating a hypothyroid state in man and animals. Therefore, it is justifiable to surmise that iodine deficiency does exist in Kerala, although no authentic reports have appeared on this subject.

Goitre, as a clinical syndrome in goats, has been recognised in certain parts of India as early as 1935. However, information on thyroid pathology in domestic animals in India is still very meagre and the basic mechanisms involved in the biological effects of hypothyroidism still remain to be elucidated. Sreekumaran (1976) while studying the pathological features of experimentaly induced clinical hypothyroidism in kids indicated that the reproductive organs were also affected in hypothyroid state. However, he did not make an in-depth study on the reproductive pathology in experimental hypothyroidism. A sub-clinical hypothyroid state can perforce cause subtle degenerative changes in the reproductive organs and this could be an important factor leading to subfertility and infertility in livestock.

There has not been any reported investigation in this direction particularly from this country. Therefore, there is need to make a detailed investigation on this problem to delineate the influence of sub-clinical hypothyroid state on growth and reproduction. Recognition of the existence of sub-clinical hypothyroid state in the animal population causing lowered production, increased susceptability to infection and subfertility is of paramount importance for adopting suitable preventive measures to reduce reproductive disorders and losses due to lowered production and mortality in animal industry. Further, ovines have been pointed out as a good experimental model to study human hypothyroid state since the response in this species in hypothyroid state closely parallels the human (Hoy et al. 1964).

Against this background an experimental model of controlled hypothyroid state was induced in goats, using different dose regimes of thiourea with the objective of studying the sequence of physiopathological changes in different stages of hypothyroid state with special reference to its influence on reproduction. The results obtained have been documented and discussed.

CHAPTER II

REVIEW OF LITERATURE

REVIEW OF LITERATURE

2.1. Historical resume

The name "thyroid" for the gland was first suggested by Wharton (1656) though the diseases of the thyroid were recognized by the chinese source as early as the second millenium B.C. King (1836) recognised thyroid as an organ of internal secretion. The relationship between the thyroid and various body functions was studied by experimental thyroidectomy (Cooper, 1836). Kocher (1883) observed development of myxoedema like syndrome following thyroidectomy. Baumann (1896) demonstrated the presence of considerable quantities of the element iodine in the thyroid and its association with thyroid function. The work of Kendall (1915) lead to the isolation, characterisation and finally synthesis of thyroxine from the thyroid gland. Smith and Smith (1922) first demonstrated that the thyroid activity was regulated by the pituitary hormone. Chemical structure of thyroxine was identified by Harington and Berger (1927). Kimbal (1937) noted the association of hypothyroidism and endemic goitre. Crosa and Pitt-Rivers (1952) identified triiodothyroxine (T_3) in plasma. Thyroglobulin was classed as a glycoprotein and most of the

carbohydrate was found accounted by glucosamine and mannose (Gottschalk and Ada, 1954). Hall et al. (1964) discussed the chemical characteristics of purified thyroglobulin.

2.2. Thyroid pituitary relationship

The first observation that pointed out the relationship between the thyroid and pituitary was made by Miepce in 1851. Subsequently, experimental removal of the thyroid showed hypertrophy of the pituitary and atrophy of the thyroid after removal of the pituitary. These studies indicated that the pituitary and the thyroid were related (Ragowitsch, 1889). The activity of the thyroid gland was found to be controlled by the thyroid stimulating hormone (TSH) secreted by the basophil cells of the anterior pituitary (Smelser, 1944 and Adams, 1946). Adams (1958) and McKenzie (1958) discovered thyroid stimulating factor (TSF) in serum. Its action on thyroid was long standing and it disappeared from the blood slower than the thyroid releasing factor (TRF). The iodine level in the blood and thyroid also affected the control mechanism. TSH was not essential for secretion of hormone but was essential as a stimulating factor (Purves, 1964). The

secretion of TSH by the pituitary was dependent upon the level of unbound free iodine in the blood (Wilson et al. 1968). Certain regions in the hypothalamus and pituitary responded to increased thyroxine level by depressing TSH release. Release of TSH was also influenced by TRH produced in the hypothalamus and it stimulated pituitary to release TSH when the level of free thyroxin (T_f) was low (Maneko, 1970). TSH was found to have a number of effects on the thyroid gland. Due to the action of TSH, the gland increased in size, the height of the follicular epithelial cells was increased and there was loss of colloid (Maneko, 1970). TSH hormone stimulated the accumulation of iodide, its organification and release of thyroxine (Jubb and Kennedy, 1970). The response of the thyroid to TSH was also influenced by the level of stable iodine intake. When the level of stable iodine intake was low there was an increase in the number and size of the cells and in the uptake and release of iodine and these changes were attributed to the increased level of TSH in circulation. Iodine was also reported to enhance the hydrolysis of thyroglobulin, liberated from the gland (Jubb and Kennedy, 1970). DeFesi et al. (1979) recorded maximum increase in the pituitary weight after thyroid-

ectomy in rats. Increase in both pituitary DNA content and total pituitary cell number was observed in thyroidectomised rats when compared to euthyroid animals.

2.3. Thyroid hormone synthesis

Iodine entering the gland was rapidly incorporated into the colloid of thyroglobulin which was the initial substrate for the iodination process. The iodinated thyroglobulin within the colloid of thyroid follicle represented the stored hormone. Within the thyroid gland, the active protease cleaved the iodinated amino acid from thyroglobulin (De Robertis, 1941). All tissues though, contained iodine, 25 to 35% of the total ingested iodine was concentrated in the thyroid gland (Riggs, 1952). Pitman and Wodinsky (1955) indicated that the thyroglobulin was formed in the colloid rather than in the epithelial cells of the thyroid. Thyroglobulin was found to have a molecular weight of about 6,60,000 (Edelhoch, 1960). The thyroglobulin was identified to be a polypeptide, made up of about 5650 amino acid residues, of which about 125 were tyrosyl units (Edelhoch and Wall, 1964). Rush (1969) reported that T_3 forms were in greater proportion in the total hormone. The mono and di-iodo-

tyrosine were coupled for the synthesis of tri-iodothyronine (T_3) and thyroxine (T_4). Three major steps were described in the synthesis of thyroxine. The first step was the concentration of iodide by the gland from the blood; this iodide was enzymatically oxidised to iodine. Then it combined with the protein, thyroglobulin and thus synthesis of thyroxine was initiated (Long, 1971).

2.4. Functions of thyroid hormone

The primary function of the thyroid hormone was considered as regulation of cellular oxidation and stimulation of oxygen consumption for normal growth and development (Barker, 1951). The thyroid hormone regulated the rate of energy turnover in vital organs and this helped in maintaining basal metabolic rate of the body. It exerted influence on the development of hair and pigmentation in animals (Berman, 1960). In conjugation with other hormones it exerted a control, by its action at the cellular level, over growth and development of young animal, temperature regulation, intermediary metabolism and reproduction (Bush, 1969). There was a close link between the catecholamines and peripheral effects of thyroid hormone (Jubb and Kennedy, 1970). Thyroxine

exerted maximum growth rate in the presence of growth hormone. In the absence of thyroid hormone the effect of growth hormone was greatly reduced. It was also involved in the production of ribonucleic acid, mitochondrial activity and cytoplasmic protein synthesis. Barker (1971) reported that thyroxine was essential for full translation of genetic message into the ribonucleic acid and ribosomal synthesis of protein. In addition, many metabolic processes were accelerated such as protein break down, carbohydrate and lipid turnover and calcium metabolism. Nervous functions at all levels were influenced by the thyroid. Exchange of water and salts between the cell and body fluids, spontaneous electrical activity, threshold of sensitivity to variety of stimuli, and reflex time motor behaviour were all influenced by the thyroid (Gorbman and Bern, 1974). Anderson and Harness (1975) observed that for every unit increase in body weight there was a 69 unit increase in the thyroid hormone secretion rate. Louvet et al. (1979) pointed out that hypofunction of the thyroid, evidenced by low T_3 and T_4 caused anovulation and sterility.

2.5. Thyroid pathology

The thyroid gland is prone to variety of diseases

other than hypo and hyper activity. For several reasons the gland gets enlarged and show a large spectrum of gross and microscopical patnological changes.

Thyroid diseases capable of producing clinical signs were classified as (Bush, 1969):

1. Goitre
2. Hypothyroidism
3. Hyperthyroidism
4. Thyroiditis, and
5. Thyroid neoplasia

2.5.1. Goitre

Cohrs (1966) described Goitre as a non-inflammatory, non-malignant enlargement of the thyroid gland and classified goitre on the basis of the morphology as :

1. Atoxic goitre : included most sporadic forms
2. Goitre with functional changes

a) Athyroid or hypothyroid goitre

b) Hyperthyroid goitre (Parenchymatous)

Kaneko (1970) recognised two types of goitre on the basis of functional activity.

1. Non toxic goitre : Produced normal amount of

normone (simple goitre) or less than normal amount of hormone (hypothyroid)

2. Toxic goitre : Characterised by excessive production of hormone (hyperthyroid)

On morphological basis goitre was classified into four patterns (Smith et al. 1972).

1. Colloid goitre
2. Hyperplastic goitre (Parenchymatous)
3. Nodular goitre
4. Exophthalmic goitre

2.5.1.1. Endemic goitre

Goitre due to iodine deficiency was reported in Irish (Marine and Lenhart, 1910). The early chemical studies of McCarrison on the iodide content of soil and water provided no evidence to support the iodine deficiency hypothesis (McCarrison et al. 1927). Stott et al. (1930-31) pointed out the association between high goitre rates and the dolomitic lime in India. The work of McCarrison (1933) drew attention not only to the extent of Himalayan endemic goitre but also to the cause and reaction of the thyroid to various noxious influences - nutritional, toxic or infective. McCarrison (1933) introduced the concept of

goitre "Noxa" and thought in terms of "life line" of the thyroid gland.

The co-existence of endemic goitre and cretinism was recognised in the mid 16th century and a concrete description of cretinism in Switzerland was made in the early 17th century. An absolute or relative deficiency of iodine was found mainly responsible for endemic goitre. Stott et al. (1931) concluded that drinking of hard water containing excessive amount of calcium was of etiological importance in Himalayan endemic zone. This supported the belief that calcium may be a goitrogenic factor. Levine et al. (1933) reported that the element iodine and inorganic iodine themselves, in large doses have goitrogenic properties. Wilson (1941) suggested that excessive intake of flurine might be a causative factor in Punjab, where he found a close association between the incidence of endemic goitre and flurosis. Murray et al. (1945) indicated that there is a relationship between the distribution of goitre and calcium concentration of drinking water in England. Several antithyroid substances have been isolated from plants and fodders responsible for endemic goitre. Greer (1950) made a comprehensive review of substances which have been reported goitrogenic to experimental animals

and discussed their importance.

Stanbury et al. (1954) studied on the metabolism of iodine in endemic goitre cases in Western Argentina, employing radioactive iodine and indicated that lack of iodine in the diet was the most probable cause. Further, the work of Stanbury et al. (1954) threw light on the mechanism of adaptation of man to iodine deficiency. The mountain slopes of Himalayas, Alps, Pyrenees, and Andes have been the world's most notorious foci for endemic goitre (Kelly and Snedden, 1960). In the Indian subcontinent, the northern frontiers extending from Kashmir in the west to Bengal and Assam in the East formed the extensive Himalayan goitre belt (Ramalingaswami, et al. 1961).

In addition to these factors genetic constitution was suggested to play a role especially in regions where endemic goitre existed for generations (Roche and Michitzky, 1960). Cold climate was reported to influence the prevalence of endemic goitre in regions of borderline iodine supply as a result of increased demands for thyroid hormone (Scrimshaw, 1964). Suzuki et al. (1965) reported endemic goitre in Japan due to excessive iodine intake. The geographical distribution of this condition has been well studied. On the basis of the nature of occurrence

of the disease, the terms endemic and sporadic atoxic goitre have been used. An endemic goitre area was defined as one in which more than 10% of the population showed clinical signs of thyroid enlargement (Koutras, 1971). Halik and Zavadsky (1978) recorded mass outbreak of goitre in breeding rams in an endemic goitre area. The thyroid glands were enlarged and diffuse colloid goitre was evident histologically. The goitre regressed after the administration of iodine preparations. Walton and Humphrey (1979) diagnosed congenital goitre as a cause for premature and stillbirth in ewes in an area of high altitude and high rainfall in New Guinea.

2.5.1.2. Colloid goitre

Colloid goitre was considered as an involutionary phase of hyperplastic goitre (Hollis, 1959). The gland was unable to return to its normal size, when the demand of tissue for thyroxine was met or increased in size due to the accumulation of colloid in quantities commensurate with the increase in thyroid epithelium. Follicles were packed with colloid, and there was greater variation in their size and some times they coalesced to form cysts. All follicles were filled with deeply staining colloid

(Jubb and Kennedy, 1970). The follicular epithelium remained taller than normal and much of the increased vascularity persisted (Wilson, 1975).

2.5.1.3. Parenchymatous goitre

According to Jubb and Kennedy (1970) the morphological indication of increased stimulation of the thyroid gland by the thyroid stimulating hormone was hyperplasia of the thyroid epithelium. In this condition, the vascularity of the gland was greatly increased as also the total volume of the gland which sometime became enormous. The follicular lumina were smaller and many disappeared and the colloid was reduced in amount with variable staining affinity or was completely absent. An early and characteristic sign of stimulation was the appearance of vacuoles around the periphery of the colloid or peripheral scalooping (Wilson, 1975).

2.5.1.4. Nodular goitre

According to Smith et al. (1972) this type was frequent in old animals. They described well defined nodules in one or both thyroid lobes. These nodules were clearly demarcated from the rest of the thyroid tissue and the histological appearance of the nodular goitre

usually varied from nodule to nodule. Many follicles were greatly distended with colloid. While others were small and devoid of colloid. In the simplest form, the epithelial cells were inactive and colloid was deeply stained. In some follicles the hyperplastic lining cells were thrown into small papillary folds. Retrogressive changes were reported as common in nodular goitre (Smith et al. 1972).

2.6. Hypothyroidism

Ruminant hypothyroidism mainly occurs in areas of endemic goitre region (Roger, 1931). Jubb and Kennedy (1970) reported that in domestic animals hypothyroidism was generally caused by congenital goitre and was encountered in the new born. Goats were considered particularly susceptible to both development of congenital goitre and associated effects of hypothyroidism (Mason and Wilkinson, 1973). They classified hypothyroidism into the following categories. The last five categories were associated with thyroid gland enlargement.

1. Primary hypothyroidism due to lack of functioning thyroid.
2. Secondary hypothyroidism due to pituitary insufficiency.

3. Hypothyroidism due to extreme degree of iodine deficiency.
4. Hypothyroidism due to goitrogen
5. Hypothyroidism due to dyshormonogenesis
6. Hypothyroidism due to autoimmune thyroiditis
7. Hypothyroidism due to neoplasia.

Hypothyroidism was more severe in young growing animals and it caused interference with overall development than in mature adults (Ferguson et al. 1956). Calderbank (1958) reported cases of infertility associated with iodine deficiency. A close association between the thyroid and gonadal functions and the loss of libido in males and suboestrum in females was reported (Calderbank, 1963). Wallach (1965) pointed out that hypothyroidism was generally characterised by lowered body temperature, increased sensitivity to low environmental temperature and retardation of growth. In hypothyroid domestic animals the gestation was significantly prolonged. New born goats showed myxoedema, alopecia and high mortality rate (Jubb and Kennedy, 1970). Underwood (1971) indicated that reproductive failure was a common feature of iodine deficiency. The essential feature of hypothyroidism was an abnormally low basal metabolic rate. Retardation of growth with regard to stature and sexual development was

a common finding in the young. Elevated cholesterol level in the blood serum was usual (Smith et al. 1972). Wilson (1975) summarised the effects of hypothyroidism in ruminants. They were

1. Retention of placenta
2. Infertility
3. Lowered milk production
4. Lowered resistance to infection
5. Increased susceptibility to ketosis and
6. Late abortion, still birth and weak off springs

In goats Sreekumaran (1976) observed increased serum cholesterol and protein levels and decreased protein bound iodine in experimental hypothyroidism in goats.

2.6.1. Hypothyroidism due to iodine deficiency

The main cause of simple hypothyroidism was iodine deficiency in the environment (Southcott, 1945). Goldschmidt (1954) surveyed the factors influencing the iodine content of the soil. The iodine requirements were also influenced by the composition of diet as a whole (Scott et al. 1961). Calderbank (1963) suggested that although soil is the source for iodine for both water and crops, there may be little or no correlation between

the iodine content of the soil and pasture growing on it. Soil in the neighbourhood of the sea are not always richer in iodine than those in lands but was dependent upon the prevailing wind, the amount of precipitation and the nature and reaction of the soil. The geographic and climatic history of the land determined the distribution of iodine in soil and water supplied to some extent. The effect of heavy rain was also a determining factor (Serimshaw, 1964). Blokhina (1970) stated that some micro-elements may also influence the availability of iodine. High protein intake was found to interfere with utilization of iodine (Wilson, 1975). Walton and Humphrey (1979) reported that the iodine deficiency in soil in the high lands of Papua, New Guinea was due to high annual rainfall leaching the iodine from the soil.

2.6.2. Hypothyroidism due to goitrogens

Presence of goitrogens in the feed stuffs was described as another important cause for hypothyroidism in animals. Many plants and forages have been known to contain goitrogens. Calderbank (1963) described two main types of goitrogens. A thiocynate type which inhibited thyroidal uptake of iodine and this blocking effect was overcome by

simulataneous administration of iodine. A thiouracil type which interfered with the organic binding of the iodine in the thyroid and this effect was reversed by the administration of thyroxine. Thiocyanate was about 25 times more potent than nitrate in inhibiting thyroid function (Greer et al. 1961). The goitrogens like the thiocyanate were described as anionic goitrogens and those like thiouracil were designated as organic goitrogens (Catt, 1970).

2.6.2.1. Natural goitrogens

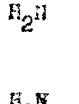
Greaney et al. (1928) observed development of goitre in rabbits fed on cabbage diet. They detected the presence of goitrogen in the cabbage. Sharpless et al. (1939) demonstrated the goitrogenic action of soyabean flour meal in rats by producing enlarged thyroid by feeding soyabeen flour. Kennedy and Purves (1941) produced goitre in rats fed Brassica seeds. The weights of the glands were found to be increased by 300 times and histologically they observed hyperplasia of the thyroid glands. Examination of pituitary showed a rapid increase in the basophil cells and this was associated with hyalinization and vacuolation and formation of 'signet ring' cells. There was also simultaneous loss of acidophil cells in the rats treated with

Brassica seed diet (Griesbach, *et al.* 1941). Rapid proliferative changes occurred in the second and third week after treatment. All the halogens, if present in excess were capable of displacing iodine and caused iodine deficiency. Fluoride was reported as one of the factors responsible for goitre in Punjab (Wilson, 1941). Sharplese and Metzger (1941) showed that arsenic at 0.02 level in the diet in rams caused decrease in growth, increase in thyroid weight and decrease in iodine concentration of the thyroid gland. Astwood *et al.* (1945) found in the seeds of cochlearia and *Conringia orientalis* presence of 5,5-dimethyl-2-thio-oxazolidone, a goitrogenic substance. Astwood *et al.* (1949) isolated a goitrogen L-5-vinayl-2-thiooxazolidone (goitrin) from rutabagas and turnips. A marked decrease in follicular colloid and decrease of follicular epithelium were observed in sheep and goats fed cauliflower leaves (Spisni and Gravaglia, 1954). Butler *et al.* (1957) observed a decrease in total iodine content of thyroid and also inhibition of the conversion of inorganic iodine to organically bound iodine in sheep fed white clover. Sinclair and Andrews (1958) noted that a heavy diet of kale to pregnant ewes caused high incidence of goitre and hypothyroidism in new born lambs. Setchell *et al.* (1960)

reported the goitrogenic action of perennial grass. Greer and Whalion (1961) derived 5-phenyl-2-thiooxazolidon from the seeds of Barbarea vulgaris and from the seeds, leaves, stems and roots of Resedea luteola by enzymic hydrolysis. This compound and the 3-phenyl and 4-phenyl-2 analogous were demonstrated to have antithyroid activity. Greer (1962) reported that goitrin was present as an inactive thioglycoside, progoitrin and it was released by the action of enzyme myrosinase contained in the plants. A high incidence of goitre was observed in new born lambs of ewes and sheep grazing on pasture dominated by white-clover (George et al. 1966). Thyroid glands were heavier than normal and showed severe hyperplasia of the lining epithelial cells of the follicles and complete absence of colloid in lambs which had grazed rape (Russel, 1967). Simple goitre and hypothyroidism were observed in ruminants when fed Brassica seeds and Brussels sprints (Wood and Henderson, 1968). Akiba and Matsumoto (1977) observed 2-5 times enlargement of the thyroids in chicks fed rapeseed meal compared to the controls and they demonstrated that goitrin in rape-seed inhibited biosynthesis and release of thyroid hormone from the gland.

2.6.2.2. Chemical goitrogens

Goitrogenic chemicals like thiourea and allied compounds have been frequently employed as experimental goitrogens in animals to supress thyroid activity.



C = S Thiourea

A discrete chemical that produced goitre was first reported by Barker et al. (1941) who found that treatment of hypertension with thiocyanate some times produced goitre and hypothyroidism in man. Mac Kenzie and Mac Kenzie (1941) found that goitre in the rat resulted from sulphaguanidine administration. Kennedy (1942) observed enlarged thyroids in rats treated with thiourea. The glands were three times heavier than normal and almost completely devoid of colloid. Pituitary showed an increase in basophil cells and loss of acidophil cells. A decrease in adrenal size among rats fed thiouracil was reported (Baumann and Marine, 1945). Jones et al. (1946) observed resorption of foetus in rats fed thiouracil. They indicated that thyroid hormone was necessary for full utilization of oestrogen or progesterone. Jones (1946) observed

hypertrophy of the thyroid gland, congestion of vessels and depletion of colloid in rats treated with repeated doses of 1000 mg/kg body weight of thiourea. Involution of the adrenal cortex occurred in rats treated with thiouracil (Zarrow and Money, 1949). The involution of the adrenal cortex after thiouracil treatment was both morphological and physiological in nature. Sellers and Ferguson (1949) observed exopthalmos in rats treated with thiouracil.

D'Angelo et al. (1951) reported thyroid hyperplasia in guinea pigs treated with propylthiouracil. Histological changes were uniformly evident after 15 to 18 days treatment and were characterised by increased vascularity and increase in the height of the acinar epithelial cells. Colloid resorption was a constant feature. The microscopic changes in prolonged dosing with propylthiouracil were colloid resorption and high vascularity. The follicular epithelial cells assumed cord like formation. Hall (1952) reported reduction in serum calcium level in rams dosed with thiouracil. Swanson and Boutman (1953) observed symptoms of hypothyroidism in two young and one old dairy bulls after treatment with thiouracil. The weight of the thyroid gland in the treated animals were

twice the weight of the normal. Histologically the follicles were filled with colloid and lined by low cuboidal epithelial cells. Prolonged administration of thiouracil to rats resulted in macroscopical and microscopical changes simulating the late fibro-lymphoid stage of struma fibrosa (Clausen, 1954). Harkness et al. (1954) recorded the effects of oral administration of thiouracil on the collagen content of thyroid of rats. Increased weight and collagen content of the thyroid gland were noticed during the treatment.

Goldberg et al. (1957) observed in propylthiouracil treated rats enlarged thyroid glands with tall columnar cells, numerous mitotic figures, scanty colloid, papillary infoldings and increased vascularity. In the pituitary hyperplasia and hypertrophy of beta cells with a characteristic granularity and vacuolation and complete absence of granulated alpha cells were observed. Lascelles and Setchell (1959) administered methylthiouracil at the dose rate of 0.5, 1.5 and 4.5 g daily to six Merino sheep after conception. The offsprings had goitre and retardation of ossification centres. A reduction in protein bound iodine and increase in cholesterol value were reported. In hamster, colloid goitre was produced

by thiouracil administration (Follis, 1959). Extensive thyroid hyperplasia and loss of colloid accompanied by an increase in vascularity were noticed in the first week after thiouracil administration. When thiourea was removed from the diet, the follicles got filled up with colloid and the epithelial cells became flattened, but some had residual epithelial springs projecting into the colloid when compared to the normal. Most of the follicles were large in size. McCarthy et al. (1959) reported adrenal atrophy among rats fed goitrogens, thiouracil and tapzole. In addition to the changes in the adrenal among thiouracil fed rats, Lazo-Wasem (1960) observed thyroid and pituitary hypertrophy with a concomitant reduction in the adrenocorticotrophic hormone (ACTH) level.

Mayberry and Astwood (1961) described the mode of action of thiourea and related compounds to inhibit the formation of iodothyroxine and their coupling to form iodothyronine. They were found to diminish the inorganic iodide content of thyroid and had a slight inhibitory effect on iodide pump (Danowaka, 1962).

Nangia et al. (1975) observed high blood levels of protein and cholesterol in methimazole treated birds.

Nangia and Gulati (1976) recorded retarded growth in methimazole induced hypothyroid birds. Sreekumaran (1976) induced hypothyroidism in kids using thiourea. Histological picture was characterised by hypertrophy of the epithelium in the thyroid gland, hypertrophy and hyperplasia of the basophilic cells in the pituitary and depletion of fat and focal areas of haemorrhage in the adrenal gland.

Prasad and Singh (1979) reported decreased plasma protein bound iodine in methimazole treated hypothyroid birds. Replacement therapy with thyroxine counteracted the effects of methimazole. Burstein et al. (1979) recorded considerable loss of weight in hypothyroidism in rats induced by propylthiouracil. A significant drop in the growth hormone level was described as the cause for the weight loss. Davidson et al. (1979) demonstrated that doses of thiourea in rats inhibited in vivo protein bound iodine formation.

2.6.3. Dyshormonogenesis

Falconer (1966) reported the occurrence of congenital goitre due to a inherited defect in the biosynthesis of thyroid hormone leading to increased production of thyroid

stimulating hormone in sheep. This caused hyperplasia of the thyroid gland. High serum protein bound iodine concentration and low hormonal iodine were reported from these goitrous animals than the controls. The concentration of thyroid stimulating hormone was also significantly high in goitrous sheep. Mac et al. (1968) in their study on congenital goitre in Merino sheep observed enlargement of the thyroid glands in affected animals. The enlargement was 25 times more than the control. There was significant increase in the cholesterol level in affected sheep.

2.6.4. Surgical thyroidectomy

Marston and Peirce (1932) recorded reduction in growth rate and metabolic rate in thyroidectomised Merino sheep. Degranulation of acidophil cells after thyroidectomy was observed in rats (Zeckwer et al. 1935). Silberg and Silberg (1940) indicated a delay in endochondral ossification in thyroidectomised immature guinea pigs. In rats, Contopoulos et al. (1958) reported that after thyroidectomy in rats there was atrophy of the pituitary and the plasma contained only decreased amount of somatotrophic hormone and the anterior pituitary contained only decreased amounts of thyroid stimulating hormone, interstitial cell

stimulating hormone and growth hormone. After thyroidectomy the acidophil cells became more granulated and there was concomitant increase in the number of cells accompanied by enlargement and vacuolation of the cell cytoplasm. A significant decrease in the number of acidophil cells was observed in thyroidectomised rats (Solomon and Greep, 1959). Yatvin et al. (1964) noticed a decrease in protein : deoxyribonucleic acid ratio in thyrodectomised rats. Belonje (1967) reported an increase in plasma globulin and sedimentation of red cells in thyro-parathyroidectomised Merino rams. In thyroideotomised goats there was reduction in phosphorus excretion into long bones and endogenous excretion of phosphorus. This was accompanied by hypophosphatemia (Symonds, 1969, 1970). Mc Intosh et al. (1979) observed somatic changes like delayed osseous development in the limbs and increase in the pituitary weight after foetal thyroidectomy. The thyroidectomised lambs failed to survive for more than few hours after birth.

2.6.5. Radio thyroidectomy

Goldberg and Chaikoff (1950, 1951) produced an early state of hypothyroidism in rats by injecting various doses of I^{131} in rats. They observed hypertrophy and

hyperplasia of the basophil cells accompanied by degranulation of the acidophils. A drop in protein bound iodine from 6.7 to 0.8 ug % was observed in a Jersey bull after subcutaneous injection of carrier free I^{131} (Lewis, 1956). Thyroid adenoma, fibroma, and fibrosarcoma were produced in sheep following daily administration of I^{131} at different levels (Bustad et al. 1957). Interfollicular fibrosis, oedema and arterial damage were also reported (Marks et al. 1957). Potter et al. (1960) indicated papillary and follicular carcinoma in rats by single injection of I^{131}. Ayoub (1968) reported damage of the thyroid gland and reduction in the rate of radio active iodine uptake by the thyroid and its release into blood plasma in goats on administration of radio iodine. Cone et al. (1975) recorded high plasma thyroid stimulating hormone level in radio thyroidectomized rats.

2.7. Thyroid status and male reproduction

McKenzie and Berliner (1937) observed that in ram summer sterility was influenced by the thyroid. Turner and Cupps (1940) noted reduced anterior pituitary gonadotrophin content in thyroidectomised male rats. Similarly

a reduction in the gonadotrophic potency of the pituitary following thyroideotomy was reported amongst young male goats (Meineke et al. 1941). Schultze and Davis (1946) observed increased conception rate, high sperm motility and greater resistance of spermatozoa in bulls fed iodinated casein.

Kumaran and Turner (1949) induced mild hypothyroidism in birds by feeding 0.6% thiouracil. There was progressive depression of the secretion of the interstitial cell stimulating hormone (ICSH), without any effect on the secretion of the pituitary gonadotrophin and follicular stimulating hormone (FSH). Mild hyperthyroidism on the other hand had slight stimulatory effect on the secretion of FSH and more pronounced effect on ICSH.

Maqsood (1952) observed that the thyroid gland had an important role in the maintenance of male fertility. In his study on the effect of thyroxine supplementation on the reproductive performance of mammals recorded precocious sexual development, increased sex libido and improvement in semen picture of a bull treated with thyroxine. Mignett (1952) noted a decline in libido and deterioration of semen quality in iodine deficient bulls.

Mukherjee et al. (1953) observed that the effect of high ambient temperature on the semen quality of bucks could not be eliminated completely by feeding gamma thyroprotein daily. The apparent increase in the concentration of sperms was offset by decrease in the volume. Jovanovic et al. (1953) reported decreased libido and high percentage of sterility in males associated with enzootic goitre.

Lewis (1956) recorded a significant drop in the plasma protein bound iodine (PBI) level from 6.7 ug % to 0.8 ug % in the blood serum of bulls within 33 days following inactivation of the thyroid gland by subcutaneous injection of carrier free I^{131}. Lehon and Mixner (1958) observed better reproductive performance in Holstein cattle associated with high PBI values.

Brooks and Ross (1962) observed that exogenous administration of L-thyroxine in feed at 0.2, 0.3 and 0.4 ug % concentration failed to have any significant effect on the adverse influence of high ambient temperature on the semen quality in rams.

Balcionus et al. (1963) reported that administration of iodinated salts containing 0.87 mg/kg in feed in rams at the rate of 0.5 mg/kg day for 14 days, favourably

increased the ejaculate volume, sperm concentration, motility and resistance with a significant decrease in the percentage of abnormal spermatozoa. Baryaan (1966) correlated the increase in iodine (I^{131}) uptake directly with the sperm concentration and volume in bulls and indirectly with the sperm resistance.

Mahtiev (1966) recorded an improvement in fertility and semen picture following iodine supplementation in infertile rams maintained in iodine deficient areas. Sperm concentration, motility and fertilizing ability increased and the number of pathological sperms decreased in treated iodine deficient animals.

Prasad and Singh (1971) observed the effects of propylthiouracil and thyroxine on testis. A four fold increase in the weight of the testis was noted when compared with normal chicks at 8 weeks of age. Testis of propylthiouracil treated birds showed a compact arrangement of seminiferous tubules, against loose arrangement of such tubules in normal birds.

Kapoor et al. (1974) correlated the PBI level with different semen characteristics. Significant positive correlation was observed between the PBI level and the

live sperm percentage and total fructose, and negative correlation between the PBI level and the calcium in semen. However, no relationship between the PBI and other attributes of semen such as volume, mass activity, motility and sperm concentration was stated.

Sharma and Singh (1975) recorded more coiled seminiferous tubules lined with two or more layered germinal epithelium in hypothyroid birds as against single layer of epithelium in the control group. The leydig cells were more aggregated in clumps in the intertubular space and capillaries were more wide opened in methemazole treated birds.

Sreekumaran (1976) observed that the seminiferous tubules contained only few primary and secondary spermatocytes in experimentally induced hypothyroid state in male kids. There was complete absence of spermatozoa and germinal layer in some tubules. The lumen of the tubules contained only a net work of fibres and scattered round cells. There was also moderate interstitial oedema.

Pezely et al. (1979) reported that thyroidectomy or treatment with thyroxine did not affect the basal concentration of testosterone in quails. There was, however, a strong inhibition of the growth of the testis.

2.8. Thyroid status and female reproduction

2.8.1. Thyroid and oestrus cycle

Reproductive performance in animals and human beings is dependent on the functional state of the thyroid gland. Hunt (1944) observed that in young female rats the mitotic activity of the thyroid increased somewhat in early oestrus, was maximal during metestrus and was minimal during the dioestrus interval.

Soliman and Reineke (1950) in their study on the uptake of radioactive iodine by the thyroid in rats recorded maximum iodine concentration in the thyroid during oestrum with a gradual decline through metoestrum and dioestrum. Oestrogen in small doses consistantly increased the level of thyroid iodine and progesterone had the opposite effect. It was postulated that rhythmic alterations in the thyroid during the oestrus cycle were probably controlled by changes in the levels of gonadal hormones.

Soliman and Dadawi (1956) reported that the thyroid activity in rats increased during the oestrus phase when compared to the other three stages of the cycle and that the level of thyrotropic hormone in the blood closely paralleled the state of thyroid.

Robertson and Falconer (1961) recorded a cyclic variation in the degree of the thyroid activity, iodine uptake and concentration of PBI in different stages of the oestrus cycle in ewes. All these were maximum during oestrum. In cows the levels of thyroid hormone were significantly higher during oestrus as compared to that of dioestrus (Soliman et al. 1964).

Dokumow et al. (1971) correlated the changes in PBI concentration with changes in the vaginal exfoliative cytology in women and reported the maximum concentration of PBI at the ovulation time and the lowest level with the formation of corpus leuteum.

Soliman et al. (1973) recorded a progressive decrease in the fresh and dry weight of the thyroid in buffaloes during different stages of oestrus cycle. The respective weights were 25.17 and 7.12 g. Kelly et al. (1974) failed to record any significant influence on the serum thyroid hormone levels on the state of oestrus cycle in mares. The decrease in serum thyroxine following ovulation was not significant.

Sharma and Sharma (1976) recorded the PBI level in the serum in oestrum as 5.91 ug/100 ml. Vadodaria et al. (1977)

in their studies on thyroid activity in relation to reproductive performance of Surti buffalo heifers found a mean serum PBI level of 8.88 ug %. The lowest PBI level (5.46 ug %) was observed during the medium breeding period ie. July to October. The highest serum PBI level was observed just before ovulation (10.6 ug %) whereas the lowest level was observed immediately after ovulation (8.17 ug %). The serum PBI level was high at the follicular phase (9.48 ug %) than at the luteal phase (8.3 ug %). Further Vadodara et al. (1980) observed increased activity of the thyroid follicles during ovulation than during luteal phase.

2.8.2. Thyroid and pregnancy

Annison and Lewis (1959) observed limited changes in the thyroid metabolism of pregnant ewes. The rate of turnover of thyroxine was slightly increased, but was not accompanied by increased levels of plasma PBI or thyroxine binding protein.

Soliman et al. (1963) in their studies on the levels of thyroid and thyrotrophic hormones in the blood of Friesian cows during pregnancy, observed in increase in thyroid hormones during the ninth month and abundance of thyrotrophic

hormone during the first four months of pregnancy. After parturition the thyroid hormone level was low while the level of thyrotrophic hormone was increased.

Gaiton (1968) observed increased thyroid activity as evidenced by the rate of uptake of iodine, in late pregnancy in rats. Significant increase in the thyrotrophic hormone level during the first and second trimester of pregnancy which returned to normal at term was recorded in rats (Malkastan et al. 1969).

Elwishy et al. (1973) observed that the total gonadotrophin and thyrotrophin levels in the blood of buffaloes were low after calving. The level gradually increased by the third week. On the other hand, cows had higher levels with a similar pattern, the peak being during the first post-parturient week. Vzaimosuyaz (1973) recorded the PBI level as 18.1 ug/100 ml, 12.1 ug/100 ml and 8.0 ug/100 ml at the sixth, seventh and eighth month of pregnancy in cows. After parturition, the serum PBI was 7-8 ug/100 ml and within 30 to 60 days from calving the PBI level rose and the cows conceived. Hassan and Akkam (1980) observed linear increase in PBI level upto the fourth month of pregnancy in cross-bred cows. It remained constant upto the seventh month and there after increased up to the end of pregnancy.

Milk yield decreased linearly throughout pregnancy and this indicated a negative correlation between PMI and lactation.

2.8.3. Thyroid and infertility

Chu and You (1944) reported that feeding small doses of desiccated thyroid to thyroidectomised rabbits prevented the hypertrophy of ovarian follicles, while large doses had inhibitory action. Similar treatment in normal rabbits had no effect on the ovary; and large doses caused incomplete inhibition of ovarian activity.

In dairy animals there was significant reduction in the symptoms manifested by the cows in oestrus. However, the production of ovum was normal (Spielman et al. 1946). Aranow et al. (1946) observed menstrual irregularity with frequent occurrence of amenorrhoea in thyroidectomised and thiouracil treated rhesus monkey.

Krohn (1947) found that daily subcutaneous administration of propylthiouracil disturbed the oestrus rhythm of adult albino mice, causing lengthening, irregularity or complete disappearance of cycles. Krohn and White (1950) observed longer and variable oestrus cycles in hypothyroid rats. Conception was normal but with a smaller litter size

and high foetal resorption.

Thiouracil fed mice showed continuous oestrus as judged by vaginal smears, while slightly hyporthyroid mice exhibited regular oestrus cycles. Sexual maturation was accelerated at all levels of thyroprotein feeding as evidenced by changes in genitalia. The thiouracil fed groups had ovaries packed with follicles, but no corpora lutea (Soliman and Reineke, 1952).

Brownstand and Fowler (1959) reported that the ovulation rate tended to be lower among sows maintained on 0.15% thiouracil. Moberg (1959) described retention of placenta in bovines associated with sub-optimal iodine intake. The incidence of retention was reduced by 47% in herds which were supplemented by the inhalation of vapour from iodine crystals placed in front of each animal. Kovalskii et al. (1970) treated anestrous associated with iodine deficiency in dairy cows by supplementing potassium iodide. Oestrum with normal ovulation and fertilisation was induced within an average period of 148 days, when treated with 1.75 mg of potassium iodide per kg.

Barakat et al. (1971) recorded no deviation in total thyroid iodine content in normal female buffaloes and in

buffaloes with cystic ovary, inactive ovary, hydrosalpinx mucometra, perimetritis with salpingitis, metritis, perimetritis with salpingitis and oophoritis.

Srinivas (1979) reported very low concentration of PBI amongst buffaloes with cystic ovarian degeneration. The mean value was 1.68 ± 0.04 ug %. The highest concentration of serum PBI was seen in animals with uterine infections. The average values were 10.45 ± 0.6 ug % and 9.40 ± 0.3 ug % for endometritis and metritis respectively.

2.9. Incidence of goitre in sheep and goats

The incidence of goitre and hypothyroidism has been reported by several workers in sheep and goats. Love (1942) studied changes in the thyroid gland of four new born kids. He noticed parenchymatous goitre in all the four kids and alopecia in two still born kids. Mointosh (1943) reported that in hypothyroidism the new born lambs were weak and the wool growth was poor with focal areas of denudation.

Southcott (1945) recorded congenital goitre in lambs. The lambs were weak or born dead. Histologically follicles were depleted of colloid and were filled with finger like processes of the lining epithelium. The epithelial cells

were low cuboidal or columnar type.

Bauman (1948) described goitre in new born kids. In six cases myxodema and complete hairlessness were noticed. Histologically two were parenchymatous type and four mixed type. Andrews et al. (1948) noticed hyperplasia of the thyroid epithelium and depletion of colloid in the thyroid gland of new born lambs from the dams which were fed a diet free of iodised salt.

In Serbia, Jovanovic et al. (1953) investigated on the incidence of goitre in goats, sheep, cattle, pig and horses. The incidence of goitre in animals and human was parallel. The number of animals affected with goitre was large.

Otomatus (1954) made a study on the incidence of goitre in sheep in Japan. He recorded congenital goitre in 25% of new born lambs. In adults he documented an incidence of 33%. The gestation period of goitrous sheep was much longer than that was observed for normal sheep. The new born lambs were weak and died within few hours. The thyroid glands exhibited various forms of enlargement. Typical strona parenchymatosa was noted histologically.

Jovanovic (1955) investigated on the nature and incidence of goitre in domestic animals. The diffuse colloid

type of goitre was the commonest in sheep and goats. The parenchymatous goitre was observed only in goats.

Setchell et al. (1960) reported neonatal mortality associated with thyroid enlargement in lambs. He observed that the affected lambs were weak and lethargic bearing coarse coat. The glands showed varying degree of colloid change histologically.

Crowth (1962) recorded hypothyroidism in sheep. He observed adverse effect on wool growth and increased incidence of alopecia and still born lambs and kids. Watson et al. (1962) observed in congenital goitrous Doorset Horn lambs an increased size and weight of the thyroid glands and decrease in the iodine content of the glands. There was pronounced hyperplasia of the thyroid epithelial cells in these animals.

Wallach (1965) reported goitrogenic hypothyroidism in feeder lambs. In the lambs he observed low basal metabolic rate, retardation of growth rate and increased sensitivity to low environmental temperature. There was significant enlargement of the glands and they were palpable. Tall columnar epithelial cells were seen lining the follicles.

George et al. (1966) studied the thyroid glands of dead lambs histopathologically and observed parenchymatous and transitional parenchymatous goitre.

2.10. Incidence of goitre and hypothyroidism in sheep and goats in India

So far the endocrine system has not been a subject of detailed pathological investigation in this country. Only very few reports have appeared on the incidence of endocrine disorders in domestic animals from India.

Lall (1952) reported congenital goitre in three kids with enlarged thyroid. The thyroid glands histologically showed hyperplasia of the lining cells of the follicles. The acidophils of the pituitary were few in number and there was increase in the number of basophil cells. In the testis the seminiferous tubules were found immature.

Dutt and Kehar (1959) studied 1000 thyroid glands collected from sheep and goats from Bareily slaughter house. The incidence of goitre was common in goats (10%), particularly in female goats. Not a single case of goitre was observed in sheep.

Dutt and Vasudeva (1963) described a case of hypothyroidism in a ram. There was loss of weight, irregular appetite and intermittent diarrhoea. At autopsy the thyroid glands were found to be cystic and slightly enlarged. On histological examination the follicles were found

to be atrophic and lined with low cuboidal epithelium. Seminiferous tubules in the testis were atrophic and some of the tubules showed multinucleated giant cells. Pituitary showed short cords of hypertrophied epithelial cells projecting as finger like processes in some places and also hyperplasia of the basophils.

Roy et al. (1964) made a comparative study of the thyroid glands of 25 human and 50 goats, collected from a severely endemic area in the Himalayan belt. The thyroid glands of goats were large, pale and hyperplastic with intense lobular hyperplasia. In human thyroids grossly visible, well circumscribed, greyish white multiple nodules were noticed. There was extreme reduction of organic iodine content of thyroids in man and animals from endemic area. Microscopically the epithelial cells were tall columnar type and they were thrown into papillary folds. Follicles contained little or no colloid. Human thyroid showed intense epithelial and stromal hyperplasia.

Rajkumar (1970) observed enlargement of the thyroid glands in 16 kids out of 29 Barbari kids in a Government Farm in Uttar Pradesh. Taking gross enlargement of the thyroid gland as the criterion for the diagnosis of goitre

in village flock, he recorded an incidence of 0.54 % in local goats, 7.02 % in Barbari X Local and 16.67 % Alpine X Local goats.

Sreekumaran (1976) described the clinico-pathological features of experimental hypothyroidism in kids. Histologically the thyroid gland showed formation of colloid depleted microfollicles and the pituitary was hypertrophied with hyperplasia of basophil cells. Both male and female gonads showed degenerative changes.

CHAPTER III

MATERIALS AND METHODS

MATERIALS AND METHODS

3.1. Survey studies

Blood samples (5 ml) from the jugular vein were collected at random from goats (21) maintained at the All India Co-ordinated Research Project on Goats for Milk Production, Mannuthy with normal reproductive performance and from all goats (29) that were diagnosed to have reproductive disorders. The protein bound iodine (PBI) of all these animals was estimated to assess the thyroid status. The animals studied were categorised into the following groups.

1. Normal males (7)
2. Normal females in oestrum (7)
3. Pregnant (7)
4. Animals showing postpartum anoestrum (8)
5. Repeat breeders (15)
6. Animals which showed delayed puberty (6)

3.2. Experimental studies

3.2.1. Design of the experimental study

Clinically healthy, cross-bred kids of the age group between two to five months and cross-bred adults between 12 to 24 months comprising of both sexes were randomly

selected from the AICRP on goats, Kerala Agricultural University, Mannuthy and the Government goat farm, Kommeri for the study. All the animals were housed in pens under hygienic conditions, separately, in groups according to the age and sex. The animals in all the groups were sub-divided into three different groups.

Grouping:

Group I : Adult male goats (12)
1. C - Control group (4)
2. T1- Treatment group I.(4)
3. T2- Treatment group II (4)

Group II : Male kids (11)
1. C - Control group (3)
2. T1 - Treatment groupI(4)
3. T2 - Treatment group II (4)

Group III : Adult female goats (11)
1. C - Control group (3)
2. T1 - Treatment group I (4)
3. T2 - Treatment group II (4)

Group IV : Female kids (11)
1. C - Control group (3)
2. T1 - Treatment group I (4)
3. T2 - Treatment group II (4)

Except the control group of animals all the animals in other two groups T1 and T2 were daily administered

thiourea orally ($H_2NCS\ NH_2$-Sarabhai -M-Chemicals) at different dose levels. Group T1 was given 50 mg/kg and group T2 100 mg/kg body weight respectively. Throughout the experiment male animals were not allowed to have access with the females and were maintained on concentrates and green Jack tree leaves. Water was given ad libitum. On the 90th day of the experimental period both the control and experimental animals were sacrificed leaving at least one animal in each sub-group. They were maintained without treatment with thiourea for a further period of 90 days and sacrificed.

Body weight, haemogram (Total RBC, WBC, Differential leukocyte count, Packed Cell Volume, Haemoglobin content) serum cholesterol, protein bound iodine and total serum protein values of all the animals were recorded before commencement of the experiment and subsequently at fortnightly intervals. The goats were observed daily and clinical symptoms, if any, manifested were observed and recorded. The female animals were specially watched for symptoms of heat and the males for libido.

The study covered the following aspects:

1. Clinical symptoms and reproductive behaviour

2. Growth rate
3. Determination of protein bound iodine
4. Estimation of total serum protein
5. Estimation of serum cholesterol
6. Haemogram - Total RBC, WBC, Differential leucocyte count, packed cell volume, Haemoglobin content.
7. Evaluation of semen
8. Biometrics and Gross pathology of endocrines and reproductive organs
9. Histopathological and histochemical examination of tissues.

3.3. Techniques

3.3.1. Clinical symptoms and reproductive behaviour

All the animals were daily observed closely for clinical symptoms, if any. The adult female goats were specially watched twice daily for oestrum using a teaser buck. Adult males were observed for libido at the time of semen collection.

3.3.2. Growth rate

All the experimental and control group of animals were weighed at the commencement of the experiment and thereafter

at fortnightly intervals. The live weight was taken as the criterion to study the growth rate of the animals.

3.3.3. Blood chemistry

Blood samples (5 ml) for analysis were collected in a sterile test tube without adding any anticoagulants. The blood was allowed to clot and then serum was separated by centrifugation for blood chemistry.

3.3.3.1. Protein bound iodine

The protein bound iodine in the serum was estimated employing the method of Faulkner et al. (1961).

3.3.3.2. Total serum protein

The Biuret assay method of Inchiosa (1964) was adopted for the estimation of total protein content in blood serum.

3.3.3.3. Serum cholesterol

Serum cholesterol was estimated employing the method of Zak (1957).

3.3.4. Haematology

Five millilitres of blood were collected separately from the jugular vein using reagent grade Ethylene diamine tetra acetic acid (disodium salt) (EDTA) as the anticoagulant at the rate of 5 mg for every 5 ml of blood

for haematological studies.

3.3.4.1. Erythrocyte count

Erythrocyte counts were made following the techniques of Schalm (1965).

3.3.4.2. Haemoglobin

The method of Miale (1967) for the determination of haemoglobin was modified in this estimation. The cyanmethemoglobin was prepared as detailed by Miale (1967). But the readings were taken in Erma Haemophotometer as against Spectronic 20.

3.3.4.3. Packed cell volume

The method described by Wintrobe (1961) was adopted.

3.3.4.4. Leucocyte count

Leucocytes were enumerated by the method described by Schalm (1965).

3.3.4.5. Differential leucocyte count

The technique of Schalm (1965) was adopted.

3.3.5. Semen characteristics

The semen samples were collected using the artificial vagina for sheep and goat from six bucks (two from each sub group). On the 90th day of the experiment half the

number of the animals were sacrificed and from the rest semen samples were collected till the end of the experiment. Semen characteristics were recorded before the commencement of the experiment and subsequently at fortnightly intervals.

The ejaculated volume was noted directly from the graduated collection vial. The initial motility of the sperms was assessed by examining a small drop of undiluted semen, immediately after collection under high power of the microscope at 37°C. The motility was expressed in percentage. Sperm concentration was measured using the haemocytometer. The percentage of live sperms was estimated by differential staining method using Magrosin-Eosin stain as described by Blom (1950). The smears used for differential counts of live and dead spermatozoa were also made use of for assessing the sperm abnormalities. For detecting the protoplasmic droplets semen samples fixed in buffered formal saline (Hancock, 1957) were used.

3.5.6. Postmortem examination

The control and the experimental groups of animals were sacrificed by exsanguination at the end of the experiment. The carcass was weighed. A detailed autopsy was conducted following the autopsy procedure advocated by

FAO/SIDA (1968).

Immediately after slaughter the endocrine glands (thyroid, pituitary and adrenals) were dissected out and weighed after removing the loose fat and fascia. Testis were separated from the epididymis and their weights were recorded. Ovaries and tubular genitalia excluding the vagina were separated from the broad ligament and other surrounding tissues and weighed independently.

3.3.7. Histopathology

The endocrine glands and the reproductive organs were incised and examined for gross lesions. Appropriate samples of tissue were collected in 10% buffered neutral formalin for histopathological examination. Tissues were processed by routine paraffin embedding technique (Armed forces Institute of Pathology, 1968). Paraffin sections cut at 5 to 6 microns thickness were stained routinely with haematoxylin and eosin method of Harris as described by Disbery and Mack (1970). Reticulin stain (James, 1967) and periodic acid schiff stain as detailed by Armed forces Institute of Pathology (1968) were employed wherever necessary.

CHAPTER IV

RESULTS

RESULTS

4.1. Survey of thyroid status in goats

The data on the level of protein bound iodine (PBI) in goats with normal and impaired reproductive performance are presented in figure 1.

The PBI level in pregnant animals was higher than in females in oestrum and fertile males respectively. The level of PBI in animals with reproductive disorders was significantly low (Table 1). Animals with postpartum anoestrum had the lowest level of PBI. In repeat breeder animals the level of PBI was lower than in animals with a history of delayed puberty (Fig. 1).

4.2. Experimental studies

4.2.1. Clinical signs and reproductive behaviour

The experimental animals in all the groups, dosed with thiourea showed similar symptoms with slight variation in the degree of manifestation of symptoms. The animals showed progressive weakness and stunted growth from the second fortnight onwards till the 6th fortnight (Fig. 2, 3 and 4). Subcutaneous oedema of moderate degree was evident on the face, periorbital space, eyelids and lower portion of the limbs (Fig. 5). The animals were not active and

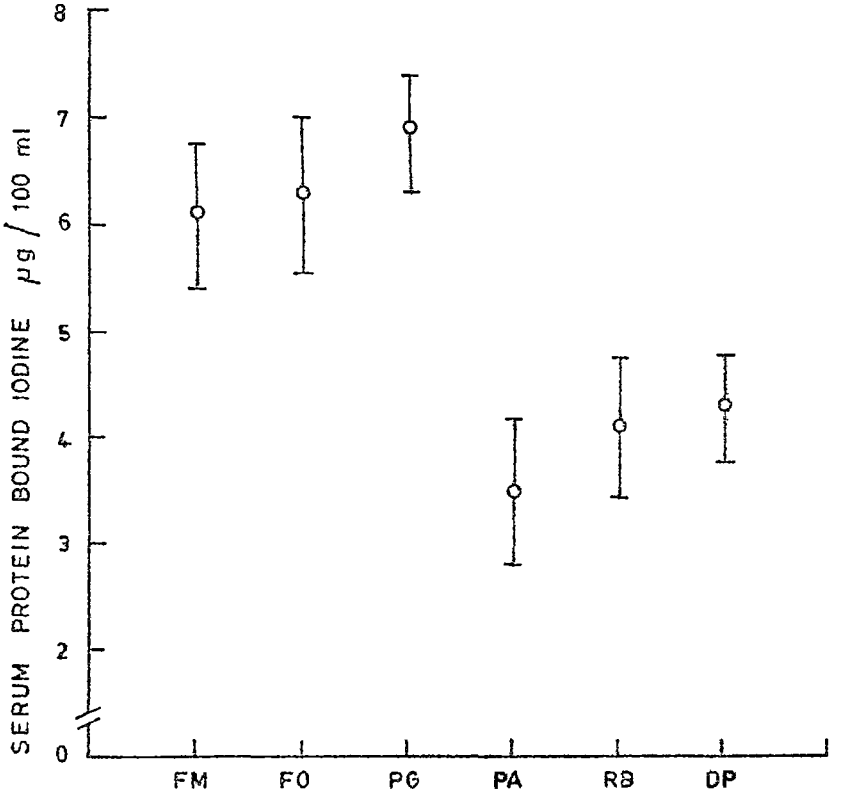


FIG. 1. THE LEVEL OF PROTEIN BOUND IODINE IN GOATS WITH NORMAL AND IMPAIRED REPRODUCTIVE PERFORMANCE

Table 1. Analysis of variance : The level of protein bound iodine in goats with normal and impaired reproductive performance

Source	df	SS	MSS	F
Treatment	4	65.5356	16.3840	42.329*
Error	38	14.7084	0.3871	
Total	42	80.244		

* Significance at 5% level
All treatments are significantly different

Mean values

1. Females : 6.29
2. Pregnant : 6.91
3. Post partum anoestrus : 3.51
4. Repeat breeder : 4.17
5. Delayed puberty : 4.03
6. Males : 6.09

C D

1 and 2	: 0.577	1 and 3	: 0.652
1 and 4	: 0.702	1 and 5	: 0.674
2 and 3	: 0.552	2 and 4	: 0.609
2 and 5	: 0.577	3 and 4	: 0.661
3 and 5	: 0.652	4 and 5	: 0.702

consumption of feed was considerably reduced. The hair coat was rough (Fig. 6) and there was slight to moderate watery discharge from the eyes (Fig. 7). Symptoms shown by the animals in groups II and IV were slightly more severe than those shown by the animals in groups I and III. The kids were lethargic and carried their head in drooping position. They remained isolated from others and did not show any tendency to move about like the control animals. The gait was unsteady.

There was loss of libido in bucks by the 3rd and 6th fortnight following the administration of thiourea. One animal each in group T1 and T2 of group I refused to ejaculate the semen even in the presence of a female in oestrum. In T1 animals there was only slight protrusion of the penis from the orifice with weak erectile movements, while in T2 animals no erectile movements and penial protrusion were noticed. Animals in group III became anoestrus from the 3rd fortnight onwards. On discontinuation of treatment with thiourea, the clinical symptoms manifested gradually disappeared. Animals in all the experimental groups appeared almost normal by the 12th fortnight. Group I animals showed normal libido by the 8th fortnight and group III animals showed signs of heat

by the 9th fortnight.

4.2.2. Growth rate

The data on growth rate are presented in figure 8 and 9.

Group I (Adult males)

There was sharp reduction in weight during the 1st fortnight in T1 animals and on the otherhand T2 animals showed a slight increase. Thereafter, the rate of reduction of weight in both T1 and T2 groups was progressive till the 6th fortnight. On discontinuation of treatment with thiourea in both the groups there was slight increase in the weight till the 12th fortnight (Fig. 8).

Group II (Male kids)

The T1 animals showed moderate increase in weight during the 1st fortnight and subsequently the weight remained almost constant. Thereafter a slight reduction in weight till the 6th fortnight was observed. The T2 animals did not gain any body weight and the initial weight remained constant till the 6th fortnight. On discontinuation of oral administration of thiourea there was steady weight gain in both T1 and T2 animals till the 12th fortnight (Fig. 8).

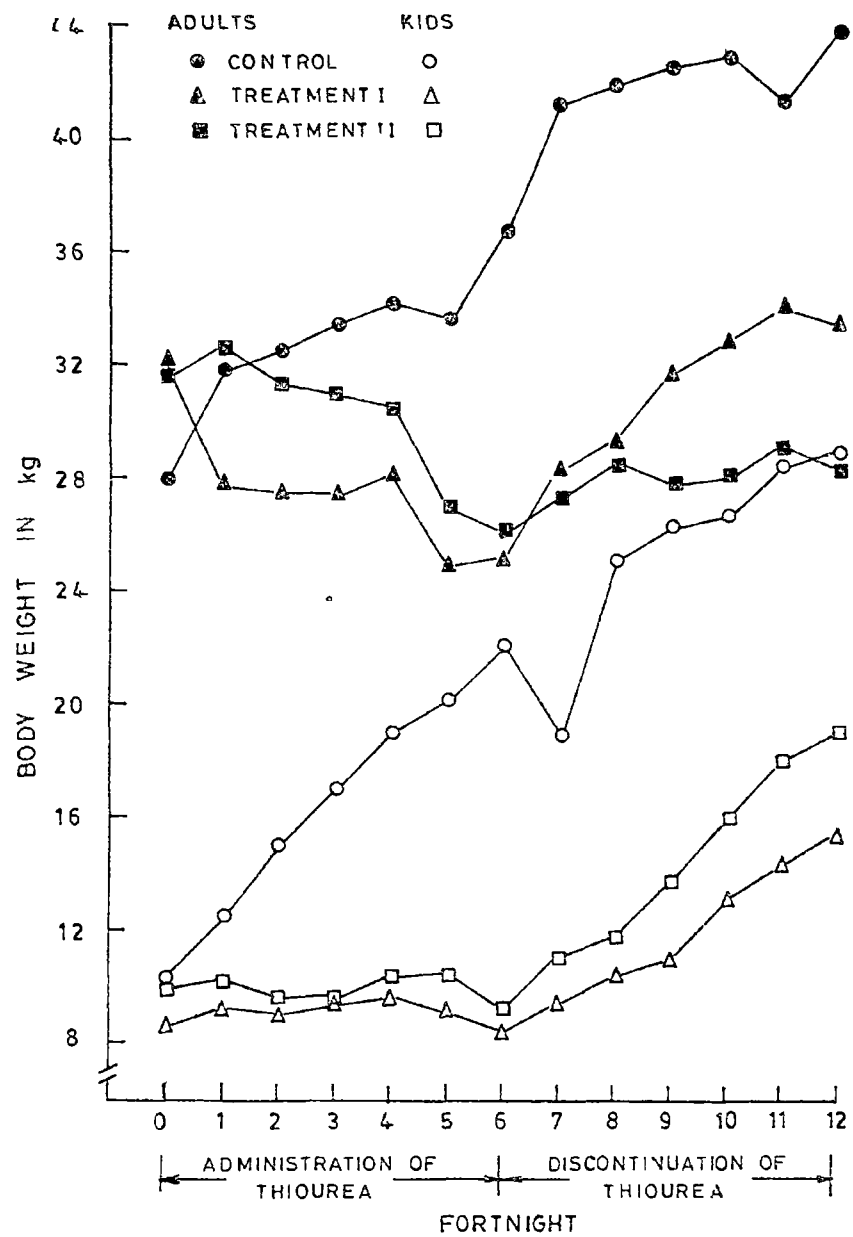


FIG.8. GROWTH RATE OF MALE GOATS IN EXPERIMENTAL HYPOTHYROIDISM

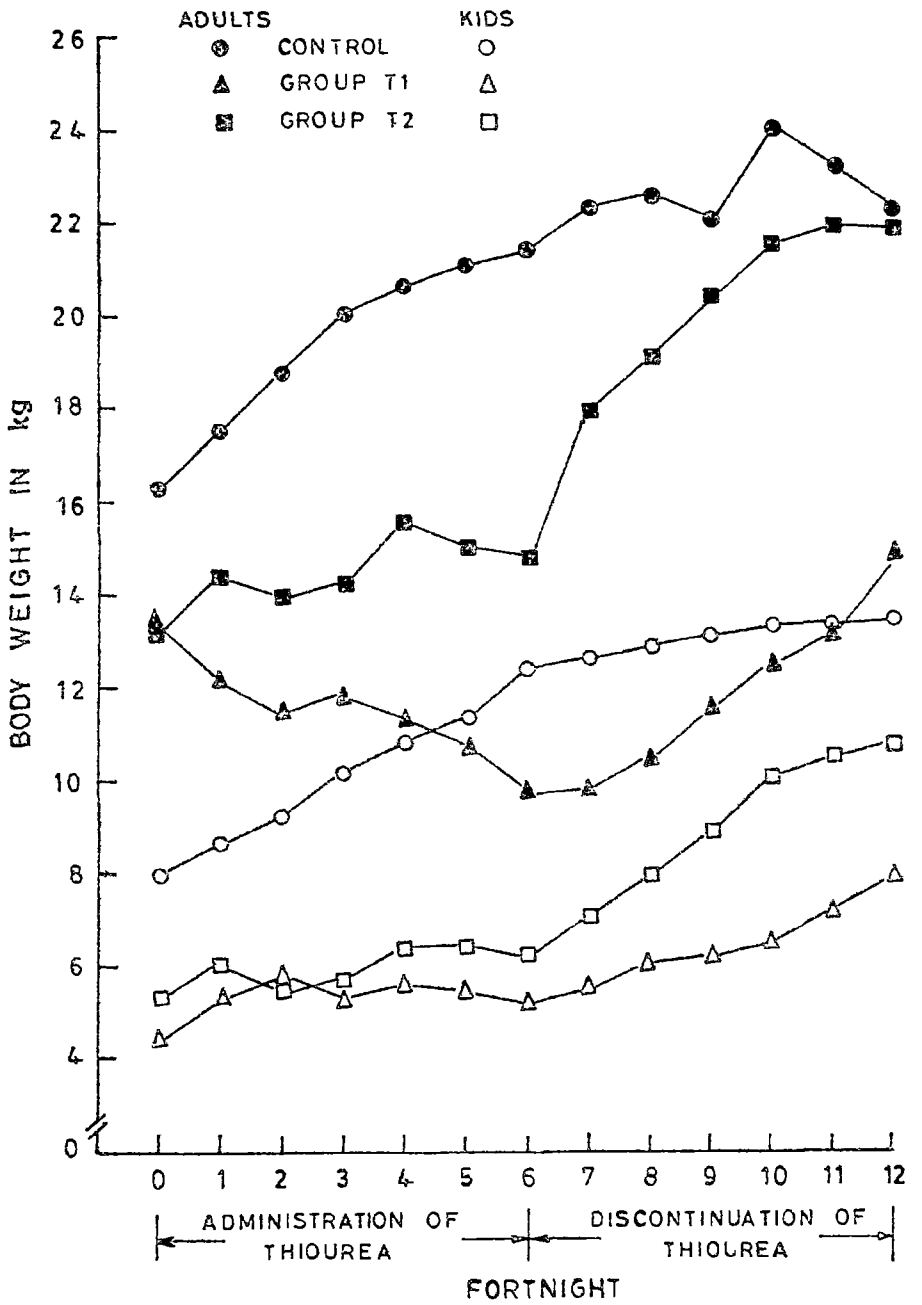


FIG. 9. GROWTH RATE OF FEMALE GOATS IN EXPERIMENTAL HYPOTHYROIDISM

Group III (Adult females)

In T1 animals there was sharp decrease in weight during the 1st fortnight but in the T2 animals there was increase in body weight. Thereafter, a steady decrease in weight was recorded in T1 animals till the 6th fortnight while in T2 animals the weight remained almost constant till the 6th fortnight. On discontinuation of treatment with thiourea there was progressive gain in weight in animals in both the groups till the 12th fortnight (Fig. 9).

Group IV (Female kids)

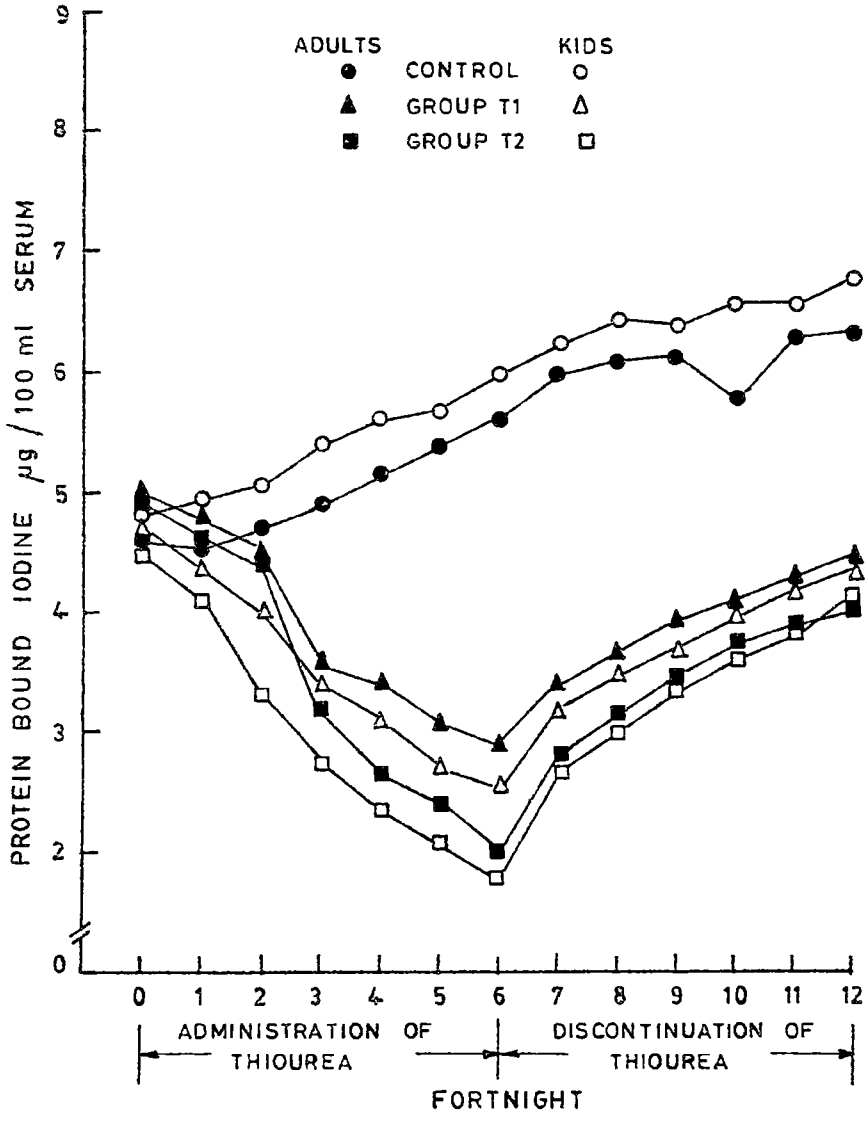
There was slight increase in weight during the 1st and 2nd fortnight in T1 animals and in T2 animals there was increase till the 4th fortnight and thereafter the weight remained constant till the 6th fortnight. On discontinuation of treatment with thiourea a steady gain in weight was observed till the 12th fortnight (Fig. 9).

4.2.3. Blood chemistry

4.2.3.1. Protein bound iodine (PBI)

The data on serum PBI level are presented in figure 10 and 11.

There was a marked decrease in the PBI level in all the animals dosed with thiourea. A steep fall in the PBI



FIG·10. THE LEVEL OF PROTEIN BOUND IODINE IN MALE GOATS IN EXPERIMENTAL HYPOTHYROIDISM

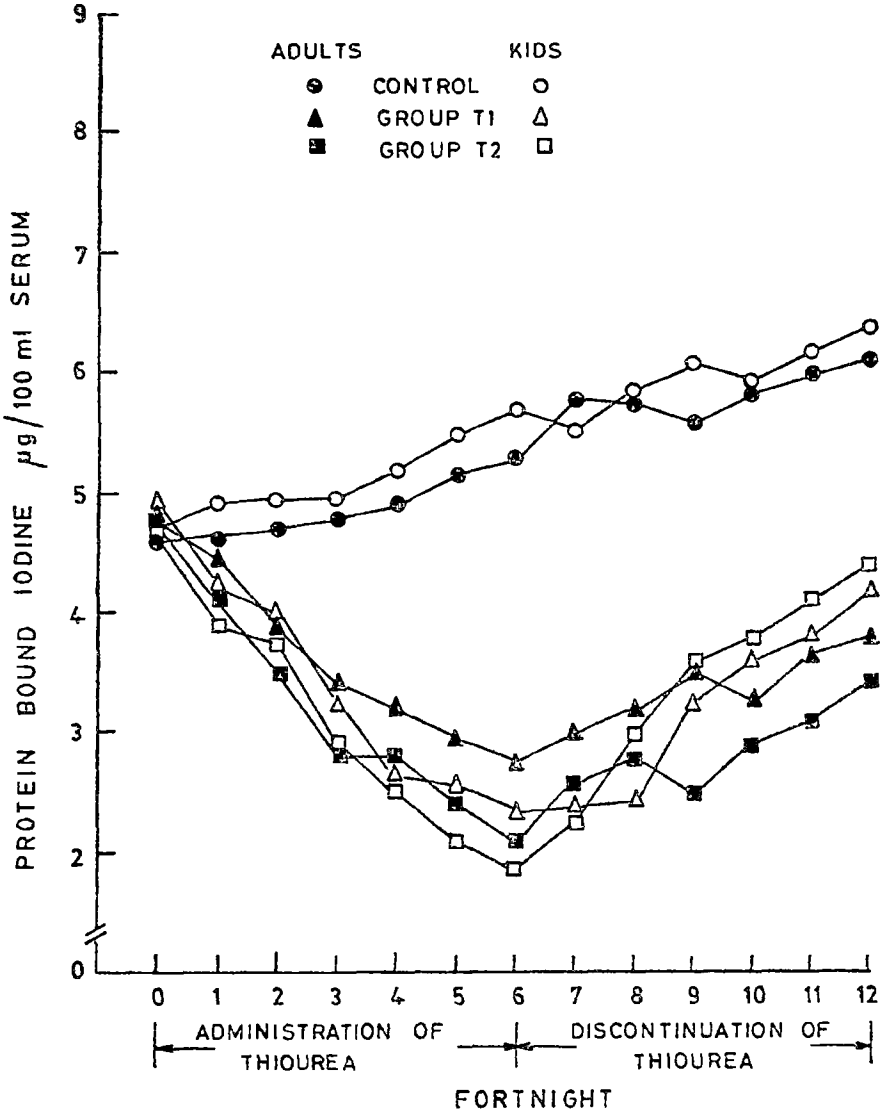


FIG.11. THE LEVEL OF PROTEIN BOUND IODINE IN FEMALE GOATS IN EXPERIMENTAL HYPOTHYROIDISM

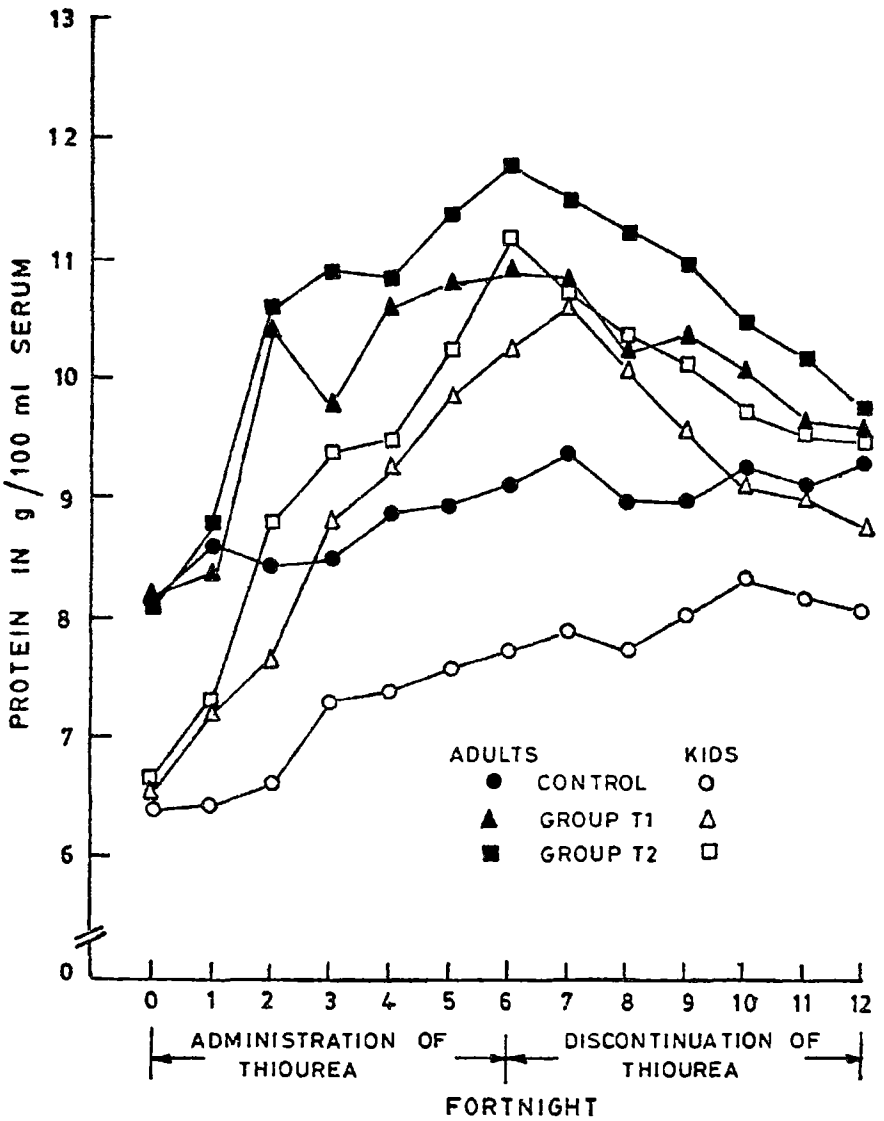


FIG. 12. TOTAL SERUM PROTEIN LEVEL IN MALE GOATS IN EXPERIMENTAL HYPOTHYROIDISM

level was observed in T2 animals. However, reduction of PBI was gradual in animals in T1 group. Lower PBI values were noted in groups II and IV. On discontinuation of treatment with thiourea there was gradual but progressive increase in the PBI level and it reached almost the normal level by the 12th fortnight. In the group T1, the PBI level returned to almost normal level when compared to the T2 group. In animals in the T2 group the PBI level was slightly lower than in the T1 group by the 12th fortnight.

4.2.3.2. Total serum protein (TSP)

The data on total serum protein level are given in figure 12 and 13.

A progressive increase in the TSP level was observed in all the experimental animals dosed with thiourea. The increase in the TSP level was slightly higher in group T2 than in T1. Group I animals recorded increased TSP level over group III. On discontinuation of treatment thiourea there was gradual fall in the TSP level till the 12th fortnight. During the 12th fortnight the TSP levels of T1 group was comparable with that of the control group. However, in the T2 group, the recorded values were slightly higher than the initial values.

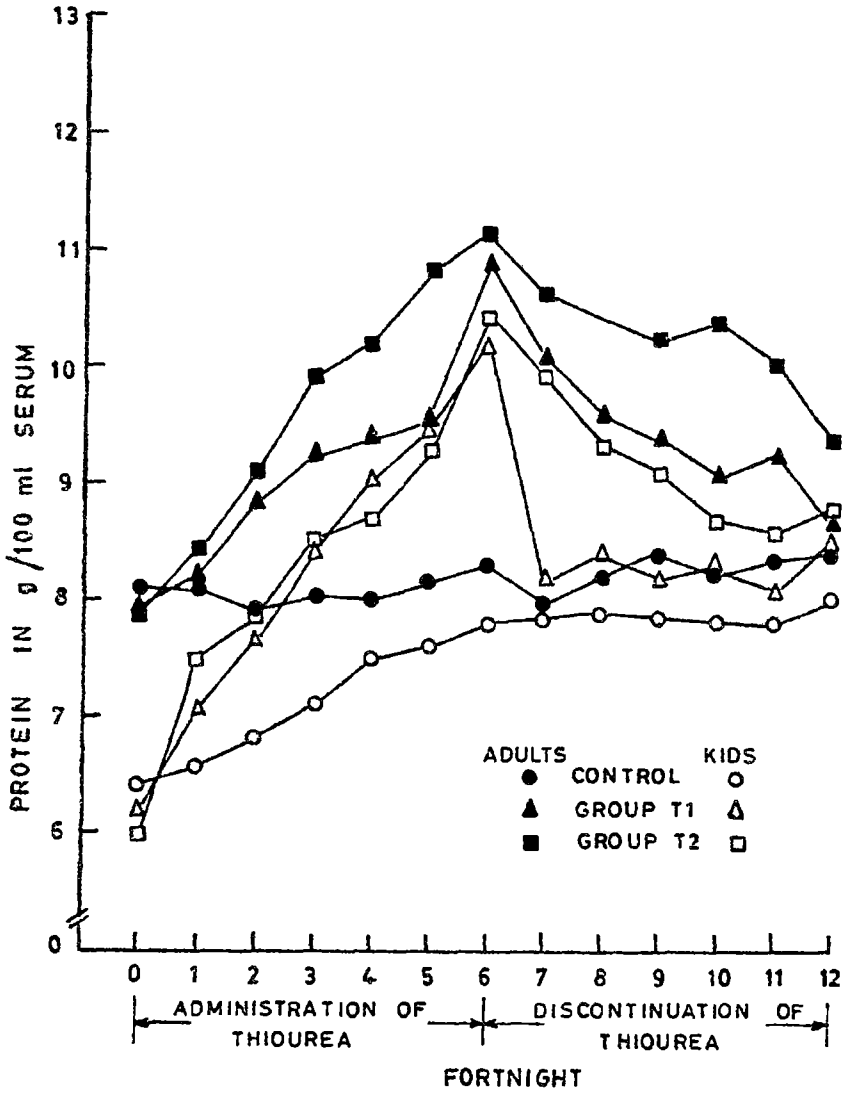


FIG. 13. SERUM PROTEIN LEVEL IN FEMALE GOATS IN EXPERIMENTAL HYPOTHYROIDISM

4.2.3.3. Serum cholesterol (SC)

The data on serum cholesterol level during the experimental period are shown in figure 14 and 15.

Animals in all the groups dosed with thiourea showed marked progressive increase in the SC level till the 6th fortnight. The SC was high in T2 animals of group I and III from the 1st fortnight onwards and it reached the highest level and above the level recorded in group II during the 6th fortnight. On discontinuation of treatment with thiourea the SC level came down gradually. The reduction was marked in T2 animals. However, the SC level remained higher than in the control animals by the 12th fortnight.

4.2.4. Haematology

4.2.4.1. Erythrocyte count

The data on erythrocyte count are shown in table 2.

There was slight reduction in the erytarocyte count by the 6th fortnight in all the animals dosed with thiourea. On discontinuation of thiourea the erythrocyte count increased progressively and was comparable with the erythrocyte count of control animals by the 12th fortnight.

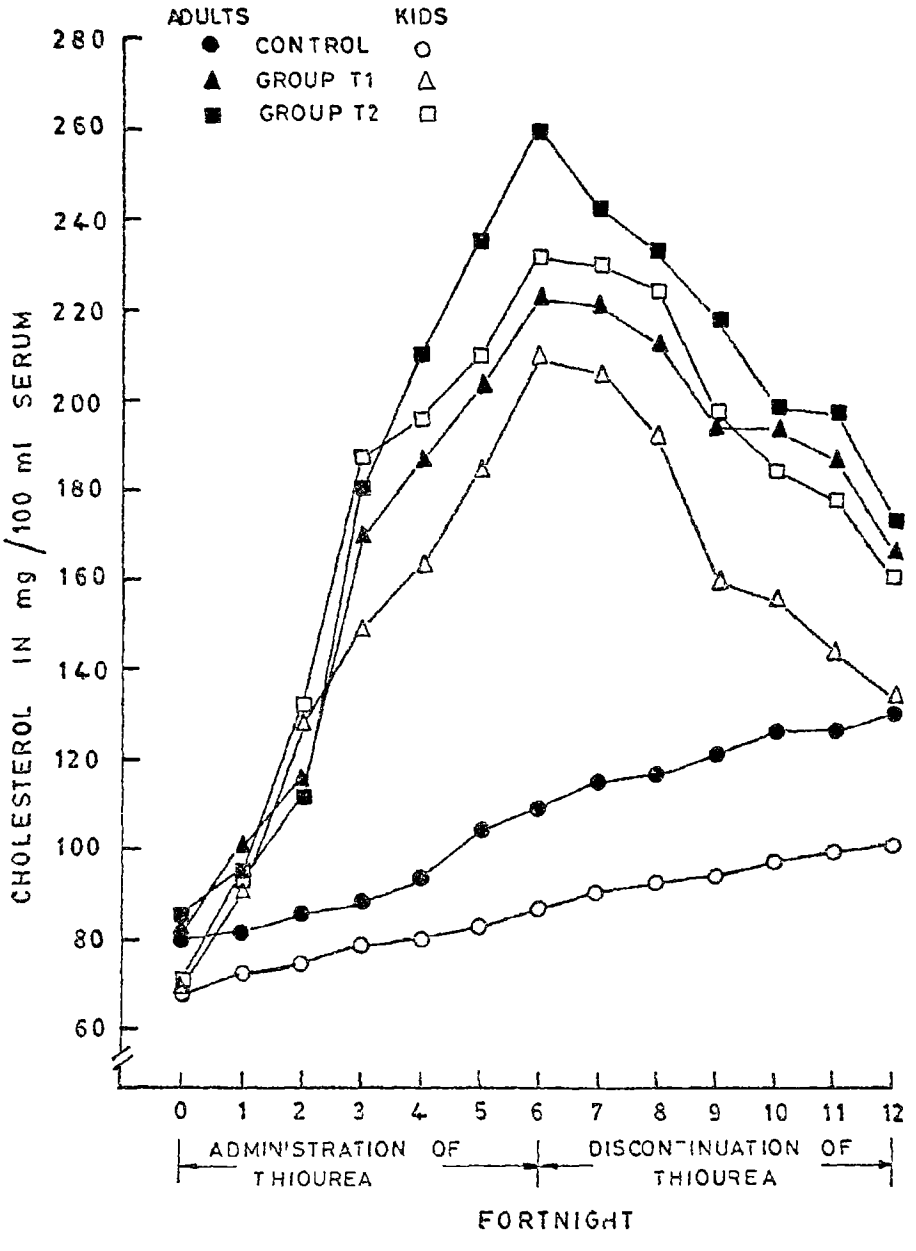


FIG.14. SERUM CHOLESTEROL LEVEL IN MALE GOATS IN EXPERIMENTAL HYPOTHYROIDISM

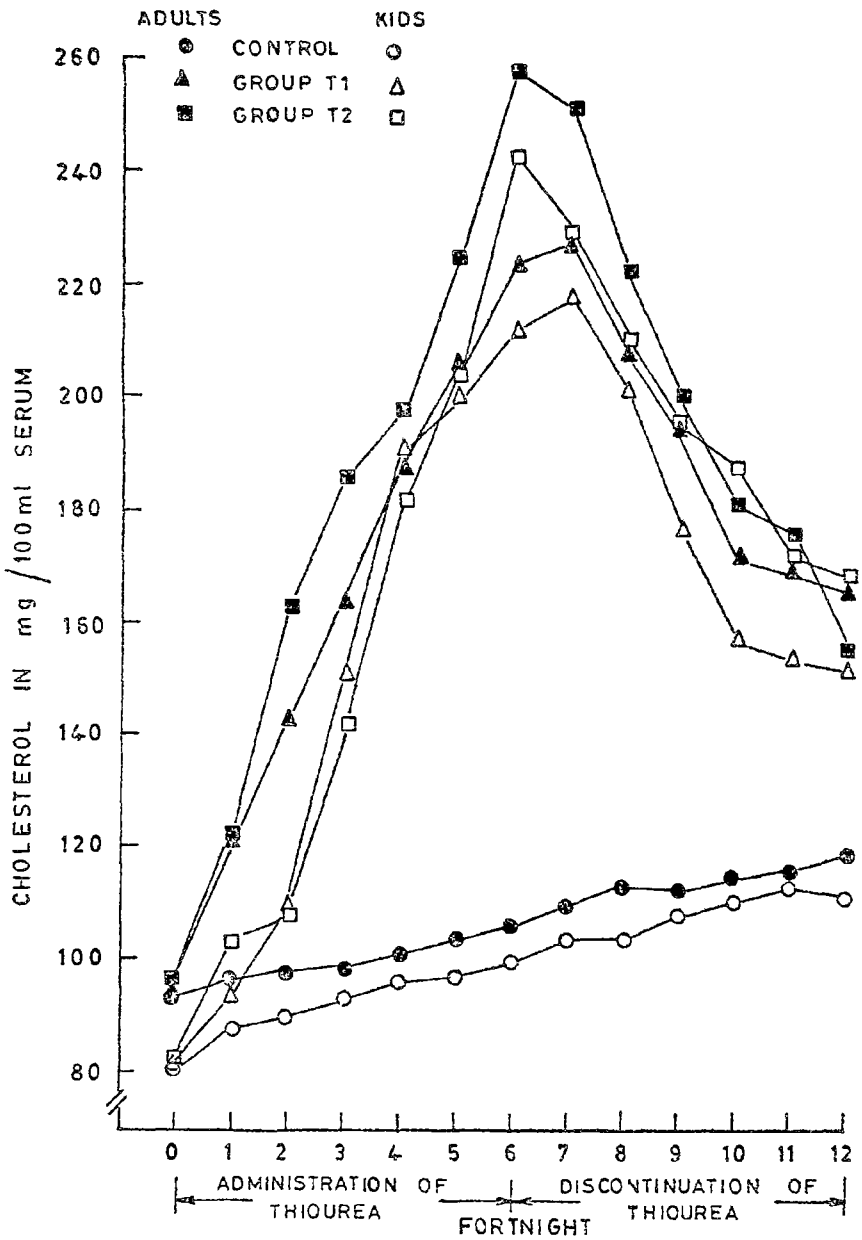


FIG. 15. SERUM CHOLESTEROL LEVEL IN FEMALE COATS IN EXPERIMENTAL HYPOTHYROIDISM

Table 2. Total erythrocyte count during the experimental period (10^6/cmm)

Sl. No.	Group	Before experiment	Fortnight: Administration of thiourea						Fortnight: Discontinuation of thiourea					
			1	2	3	4	5	6	7	8	9	10	11	12
Group I - Adult males														
1.	C	15.58	15.38	15.50	14.24	15.62	15.22	15.70	16.00	15.64	16.65	16.19	15.80	16.07
2.	T1	15.40	14.44	14.34	12.67	12.18	11.46	11.23	12.81	12.92	15.38	15.65	15.42	15.66
3.	T2	15.13	14.37	13.72	13.66	11.89	10.67	10.87	11.08	11.34	14.42	13.24	14.09	14.97
Group II - Male kids														
1.	C	15.52	14.78	15.00	14.83	15.20	15.16	15.42	16.00	16.42	16.25	15.25	10.20	12.20
2.	T1	15.33	16.01	15.78	14.26	12.44	12.62	10.79	11.89	14.83	15.03	13.38	9.60	10.40
3.	T2	15.96	15.79	15.72	11.87	11.52	12.15	9.65	11.65	13.50	16.00	14.83	9.00	9.60
Group III - Adult females														
1.	C	10.98	13.04	13.12	12.80	13.29	14.40	14.95	15.25	15.34	15.19	15.02	14.52	15.10
2.	T1	11.32	10.79	10.81	8.85	10.17	11.25	11.92	14.37	14.38	15.80	14.85	14.88	13.67
3.	T2	10.65	11.83	10.06	8.17	11.92	13.86	10.13	15.83	15.67	14.67	15.17	15.67	15.68
Group IV - Female kids														
1.	C	15.11	15.60	15.25	15.19	14.83	14.44	15.33	14.84	14.01	15.60	15.24	15.08	16.17
2.	T1	14.77	15.50	14.57	14.36	12.58	11.17	11.00	13.50	15.70	14.60	15.67	15.37	16.28
3.	T2	14.66	15.10	13.27	12.97	14.18	10.00	9.77	14.00	13.37	14.60	15.83	15.67	16.00

C : Control T1 : Treatment with thiourea (50 mg/kg) T2 : Treatment with thiourea (100 mg/kg)

The data are mean values

4.2.4.2. Haemoglobin

The data on haemoglobin level during the experimental period are presented in table 3.

There was progressive reduction in the haemoglobin level in all the animals dosed with thiourea till the 6th fortnight. The lowest value was observed in group IV. On withdrawal of thiourea, the haemoglobin level returned to almost initial level by the 12th fortnight.

4.2.4.3. Packed cell volume (PCV)

The data on PCV are presented in table 4.

There was gradual reduction in PCV till the 6th fortnight in all the groups of animals dosed with thiourea. Lowest values for PCV were recorded in group II. All the animals in T2 groups had low PCV values. On discontinuation of tratment with thiourea the PCV values came back to the normal level by the 12th fortnight in both the T1 and T2 animals.

4.2.4.4. Total leucocyte count

The data on leucocyte count are given in table 5.

There was no significant difference in the leucocyte count of control animals and animals dosed with thiourea

Table 3. The haemoglobin value during the experimental period (g/100 ml)

Sl. No.	Group	Before experiment	Fortnight: Administration of thiourea 1	2	3	4	5	6	Discontinuation of thiourea 7	8	9	10	11	12
Group I - Adult males														
1	C	9.95	9.80	10.30	10.50	9.25	9.25	9.20	9.40	9.00	10.50	10.30	10.60	10.30
2.	T1	10.00	8.60	8.75	8.65	6.80	6.00	5.65	6.10	5.90	8.90	9.40	9.40	9.60
3.	T2	9.80	9.35	8.95	7.75	6.35	6.05	5.35	5.70	5.90	7.30	6.90	7.80	8.60
Group II - Male kids														
1.	C	11.27	10.33	9.87	10.13	12.33	8.80	8.80	10.20	10.40	11.40	10.70	10.20	12.20
2.	T1	10.30	9.45	10.10	7.40	7.47	5.47	5.60	6.20	6.40	10.50	9.50	9.60	10.40
3.	T2	12.30	9.25	10.05	6.50	6.20	6.00	4.80	6.20	7.20	8.20	8.20	9.00	9.80
Group III - Adult females														
1.	C	8.20	8.00	7.47	7.67	7.27	8.00	8.47	8.60	8.60	8.70	8.60	8.60	9.00
2.	T1	7.70	4.25	4.90	4.45	4.30	5.10	5.70	6.60	6.80	8.00	8.40	9.00	10.60
3.	T2	8.55	5.20	4.75	4.33	5.00	6.65	5.20	6.00	8.20	10.20	9.00	9.00	10.00
Group IV - Female kids														
1.	C	9.13	8.93	9.53	9.37	8.80	8.13	8.40	8.00	7.80	9.40	9.40	9.00	9.70
2.	T1	8.80	7.55	9.00	8.60	6.85	6.40	4.70	5.40	7.20	10.00	9.20	8.60	10.40
3.	T2	8.75	7.00	8.07	7.80	7.40	4.80	4.80	6.00	6.60	9.00	10.20	9.20	9.40

C : Control T1 : Treatment with thiourea (50 mg/kg) T2 : Treatment with thiourea (100 mg/kg)

The data are mean values

Table 4. The packed cell volume during the experimental period (%)

Sl. No.	Group	Before experiment	Fortnight: Administration of thiourea						Discontinuation of thiourea					
			1	2	3	4	5	6	7	8	9	10	11	12
Group I - Adult males														
1.	C	32.25	30.75	31.00	30.00	33.25	31.25	34.50	34.50	33.50	39.00	37.50	34.00	35.00
2.	T1	33.00	28.50	28.25	25.25	24.25	22.25	21.75	23.00	23.50	32.50	33.00	32.50	34.00
3.	T2	31.00	28.75	29.25	26.50	22.00	21.50	21.00	21.50	22.50	26.50	26.00	28.00	29.50
Group II - Male kids														
1.	C	33.00	30.67	30.00	29.00	31.33	31.00	32.00	36.00	38.50	37.50	31.50	33.50	37.00
2.	T1	33.50	35.50	33.50	27.75	24.67	17.33	17.33	22.00	29.00	35.00	26.00	33.00	33.00
3.	T2	35.25	34.25	24.50	21.00	19.67	20.00	16.67	22.00	27.00	30.00	29.00	30.00	32.00
Group III - Adult females														
1.	C	24.67	26.00	27.00	27.00	30.33	28.67	29.67	31.50	32.00	31.50	29.50	32.00	30.50
2.	T1	24.50	20.50	21.25	15.50	20.50	31.00	23.50	25.00	26.00	32.00	29.00	32.00	34.00
3.	T2	21.00	23.75	20.00	13.00	23.50	21.50	18.50	22.00	24.00	28.00	31.00	34.00	34.00
Group IV - Female kids														
1.	C	30.67	32.33	31.67	31.33	30.33	28.67	32.00	29.00	30.00	33.00	31.50	30.50	25.50
2.	T1	29.50	32.50	29.50	29.50	24.50	20.67	21.00	27.00	30.00	30.00	35.00	34.00	36.00
3.	T2	29.50	31.25	26.33	25.00	28.00	18.50	17.50	24.00	26.00	30.00	34.00	34.00	36.00

C : Control T1 : Treatment with thiourea (50 mg/kg) T2 : Treatment with thiourea (100 mg/kg)

The data are mean values

Table 5. Total leucocyte count during the experimental period (10^3/cmm)

Gl. No.	Group	Before experiment	Fortnight: Administration of thiourea						Discontinuation of thiourea					
			1	2	3	4	5	6	7	8	9	10	11	12
Group I - Adult males														
1.	C	13.75	12.83	13.20	12.73	13.71	12.19	13.50	13.50	12.95	13.70	12.75	14.98	13.80
2.	T1	12.08	11.38	12.01	12.41	11.59	9.71	10.27	12.40	9.93	11.58	14.60	9.38	12.38
3.	T2	12.79	12.76	12.44	11.73	12.29	10.98	9.81	11.69	12.55	7.23	10.10	12.58	11.63
Group II - Male kids														
1.	C	11.93	11.03	11.67	12.32	10.25	11.68	9.77	12.65	14.23	14.70	11.69	13.28	12.15
2.	T1	13.25	14.31	15.25	12.44	15.25	15.03	13.25	12.45	13.15	13.40	11.90	13.10	13.75
3.	T2	11.84	11.56	10.41	12.93	12.78	13.13	9.62	10.55	11.25	15.25	9.40	14.60	8.40
Group III - Adult females														
1.	C	12.82	10.58	13.07	12.98	11.40	11.03	9.42	12.55	11.98	10.58	13.68	14.15	12.95
2.	T1	12.46	10.43	15.75	14.40	9.75	13.90	14.50	11.60	12.25	10.60	13.10	12.15	11.15
3.	T2	13.93	12.24	14.64	14.95	11.43	13.53	15.10	15.50	15.95	13.70	11.35	16.25	16.70
Group IV - Female kids														
1.	C	10.63	9.97	10.55	12.85	11.33	13.15	13.52	12.43	8.38	13.55	13.20	12.48	11.22
2.	T1	11.21	14.00	16.23	16.28	14.66	12.08	14.38	10.30	9.00	10.85	14.25	15.66	11.35
3.	T2	11.72	12.75	14.98	15.58	11.88	14.20	14.68	10.55	11.80	11.20	13.55	13.10	13.95

C : Control T1 : Treatment with thiourea (50 mg/kg) T2 : Treatment with thiourea (100 mg/kg)

The data are mean values

during the experimental period. However, groups III and IV showed slightly higher leucocyte count than the initial values.

4.2.4.5. Differential leucocyte count

The data on differential count are set out in table 6.

There was no significant variation in the differential leucocyte count of animals in the experimental and control group. However, a slight neutrophilia was observed in animals dosed with thiourea.

4.2.5. Semen characteristics

4.2.5.1. Ejaculate volume

The data on ejaculate volume are documented in figure 16.

There was decrease in the semen volume from the 4th fortnight (0.15 ml) onwards till the 6th fortnight (0.1 ml) in all the animals dosed with thiourea. On discontinuation of the treatment with thiourea semen volume in T1 animals returned to the initial level by the 8th fortnight and there after remained within the normal level while in the T2 animals the volume returned to

Table 6. Differential leucocyte count during the experimental period (%)

Sl. No.	Group and Parameter	Before experiment	Administration of thiourea — Fortnight 1	2	3	4	5	6	Discontinuation of thiourea — Fortnight 7	8	9	10	11	12
Group I - Adult males														
1.	Control													
	N	39.50	41.75	44.75	48.75	40.25	40.75	29.00	43.50	45.50	42.00	57.50	46.50	48.00
	L	55.50	49.50	51.75	48.25	56.25	56.50	56.00	53.50	47.50	43.50	40.50	51.50	49.00
	M	..	0.75	0.75	..	..	0.50	..	0.50	..	..	..	..	0.50
	E	5.00	8.00	2.75	3.00	3.50	2.25	5.00	2.50	6.50	7.50	2.00	2.00	2.50
2.	Treatment 1 (T1)													
	N	45.25	50.75	40.75	36.25	41.00	32.50	56.00	55.50	62.50	54.00	45.50	43.00	54.00
	L	52.25	44.00	53.70	61.75	57.25	67.25	43.25	43.50	36.50	43.50	51.50	50.50	37.50
	M	0.50	2.25	2.50	0.25	0.50	0.25	0.25	1.00	..	..	3.00	..	..
	E	2.00	4.03	4.00	2.00	1.25	2.50	0.50	..	1.00	2.50	..	5.50	8.50
3.	Treatment 2 (T2)													
	N	47.75	52.25	50.00	44.25	46.75	46.00	59.50	51.50	55.00	47.50	48.50	49.50	59.00
	L	56.00	44.50	46.78	53.25	51.75	52.00	40.50	48.50	43.00	50.00	50.50	50.50	40.00
	M	0.25	..	0.25	0.25	0.25	..	0.25	..	0.50	0.50	0.50	..	..
	E	1.00	2.25	3.00	2.25	1.25	2.00	1.00	..	1.50	2.00	..	..	1.00

T1 : Treatment with thiourea (50 mg/kg) T2 : Treatment with thiourea (100 mg/kg)
N : Neutrophil L : Lymphocyte M : Monocyte E : Eosinophil
The data are mean values

Table 6. Continued

Sl. No.	Group and Parameter	Before experiment	Fortnight: Administration of thiourea 1	2	3	4	5	6	Fortnight: Discontinuation of thiourea 7	8	9	10	11	12
Group II - Male kids														
1. Control														
	N	28.67	39.33	45.33	50.33	37.33	29.33	30.67	38.00	42.00	57.50	50.50	36.00	42.00
	L	71.00	58.00	51.33	47.00	61.00	69.33	68.00	60.50	50.00	41.50	46.50	63.00	55.00
	M	0.33	0.67	0.33	0.67	0.67	1.33	0.33	..	..	..	0.50	0.50	1.00
	E	0.33	1.00	3.00	3.00	1.00	..	1.00	0.50	..	1.00	3.50	0.50	2.00
2. Treatment 1 (T1)														
	N	30.00	41.00	39.75	37.00	40.67	41.00	38.00	42.00	47.00	47.00	54.00	48.00	38.00
	L	67.75	58.00	60.25	61.25	58.67	57.33	60.50	55.00	50.00	53.00	44.00	52.00	60.00
	M	1.75	0.67	..	0.50	0.33	0.67	0.50	2.00	3.00	..	1.00	..	..
	E	0.50	1.00	..	1.25	0.33	1.00	1.00	1.00	..	..	1.00	..	2.00
3. Treatment 2 (T2)														
	N	31.25	31.25	27.50	37.50	37.33	42.67	42.00	39.00	52.00	54.00	47.00	43.00	33.00
	L	66.25	67.25	71.75	60.50	61.67	55.33	55.00	59.00	43.00	44.00	42.00	57.00	66.00
	M	1.75	0.50	..	1.50	0.67	1.67	1.00	1.00	1.00	1.00	..	..	..
	E	0.50	1.00	0.75	1.50	0.33	0.33	2.00	1.00	1.00	1.00	1.00	..	1.00

T1 : Treatment with thiourea (50 mg/kg) T2 : Treatment with thiourea (100 mg/kg)
N : Neutrophil L : Lymphocyte M : Monocyte E : Eosinophil

The data are mean values

Table 6. Continued

Sl. No.	Group and parameter	Before experiment	Fortnight: Administration of thiourea 1	2	3	4	5	6	Fortnight: Discontinuation of thiourea 7	8	9	10	11	12
Group III - Adult females														
1.	Control													
	N	35.67	38.33	45.33	44.33	52.33	37.67	48.00	45.50	41.50	46.00	50.00	42.00	50.00
	L	63.33	55.67	53.00	48.67	46.00	60.00	50.33	52.00	55.00	49.50	46.50	54.50	41.50
	M	0.67	..	0.67	0.67	..	0.33	0.33	1.00	..	1.50	1.50	0.50	..
	E	0.33	6.00	1.00	6.33	1.67	2.00	1.33	1.50	3.50	3.00	2.00	3.00	0.50
2.	Treatment 1 (T1)													
	N	35.75	38.50	32.25	26.25	37.50	38.15	42.00	52.10	63.00	72.00	45.00	51.00	52.00
	L	65.25	57.00	67.00	70.00	60.50	59.00	45.50	46.00	36.00	27.00	49.00	44.00	47.00
	M	0.75	..	0.25	0.75	0.50	1.50	..	1.00	..	..	..	1.00	..
	E	0.75	4.00	0.50	0.75	1.50	1.00	12.00	1.00	1.00	1.00	6.00	4.00	1.00
3.	Treatment 2 (T2)													
	N	33.50	34.50	36.25	33.33	43.00	40.50	45.50	45.00	43.00	70.00	49.00	38.00	42.10
	L	61.50	63.25	61.50	65.33	54.00	57.50	52.00	51.00	55.00	30.00	48.00	54.00	57.00
	M	1.00	..	0.50	..	0.50	..	..	3.00	..	..	1.00	..	..
	E	2.00	2.25	1.75	1.33	2.50	2.00	2.50	2.00	2.00	..	2.00	6.00	1.00

T1 : Treatment with thiourea (50 mg/kg) T2 : Treatment with thiourea (100 mg/kg)

N : Neutrophil E : Eosinophil M : Monocyte E : Eosinophil

The data are mean values

Table 6. Continued

Sl. No.	Group and Parameter	Before experiment	Fortnight: Administration of thiourea					Fortnight: Discontinuation of thiourea					
			1	2	3	4	5	6	7	8	9	10	11	12
Group IV - Female kids														
1.	Control													
	N	33.67	45.67	29.67	26.33	28.67	36.33	42.00	42.00	40.50	43.50	51.00	46.50	39.00
	L	63.67	66.33	69.00	66.67	64.33	60.00	55.00	57.00	57.50	53.50	44.50	50.00	59.00
	M	1.00	..	0.67	1.67	3.33	1.00	..	1.00	1.00	..	1.00	1.00	1.00
	E	1.33	..	1.00	2.00	4.33	2.67	3.00	..	1.00	3.00	3.50	2.50	1.00
2.	Treatment 1 (T1)													
	N	37.75	38.50	27.50	26.50	31.75	40.00	47.00	42.00	46.00	47.00	46.00	40.00	40.00
	L	60.50	56.25	71.00	70.75	66.50	57.67	52.00	55.00	53.00	53.00	53.00	59.00	60.00
	M	1.00	0.25	2.50	3.40	0.25	1.00	0.50	2.00	1.00	..	1.00	1.00	..
	E	0.75	..	0.50	1.75	1.25	1.67	0.50	1.00	..	..	..	..	..
3.	Treatment 2 (T2)													
	N	42.00	50.50	31.67	30.00	31.50	39.00	20.00	36.00	35.00	46.00	44.00	55.00	46.00
	L	57.00	47.00	66.00	66.33	66.50	59.00	79.00	62.00	61.00	54.00	55.00	40.00	48.00
	M	1.00	..	1.67	1.00	..	1.00	..	..	2.00	..	1.00	1.00	1.00
	E	..	..	0.67	0.67	2.00	1.00	1.00	2.00	2.00	..	..	4.00	5.00

T1 : Treatment with thiourea (50 mg/kg) T2 : Treatment with thiourea (100 mg/kg)
N : Neutrophil L : Lymphocyte M : Monocyte E : Eosinophil

The data are mean values

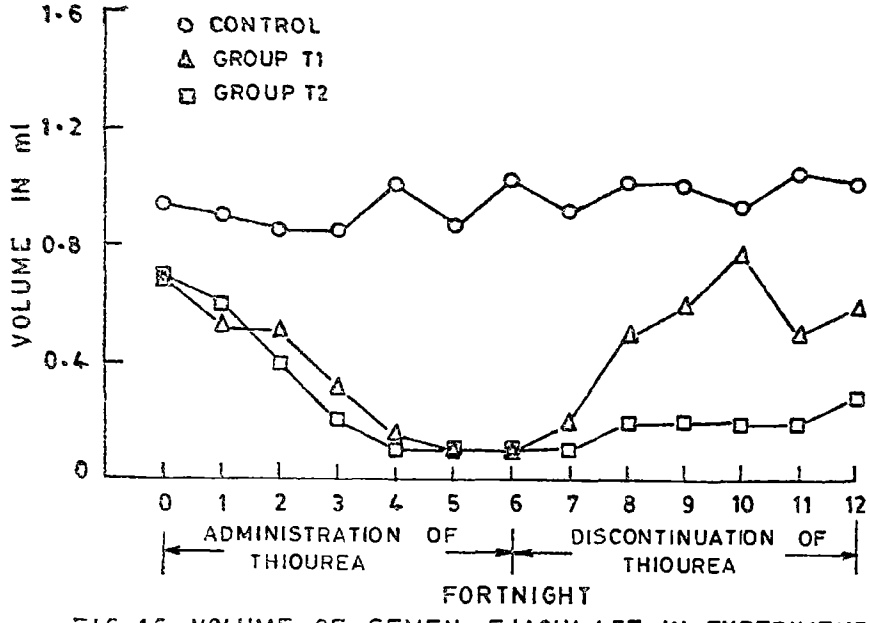


FIG.16 VOLUME OF SEMEN EJACULATE IN EXPERIMENTAL HYPOTHYROIDISM

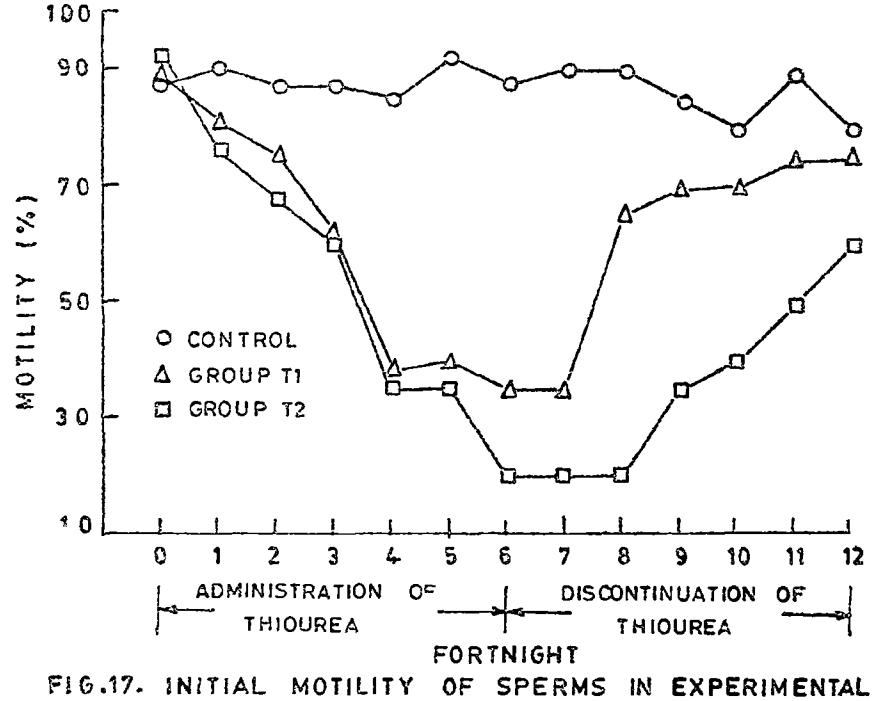


FIG.17. INITIAL MOTILITY OF SPERMS IN EXPERIMENTAL HYPOTHYROIDISM

slightly lower normal level (0.3 ml) during the 12th fortnight.

4.2.5.2. Initial motility

The data on the percentage of motile spermatozoa are given in figure 17.

The percentage of motile sperms in T1 and T2 animals was low by the 4th fortnight. The lowest percentage of motile sperms was recorded in T2 animals during the 5th and 6th fortnight. On discontinuation of thiourea administration the percentage of motile sperms increased gradually and reached the normal level by the 8th and 11th fortnights in T1 and T2 animals respectively.

4.2.5.3. Sperm concentration

The data on sperm concentration during the experimental period are shown in figure 18.

The sperm concentration decreased gradually and the lowest values were recorded between the 4th and 6th fortnight in animals dosed with thiourea. The sperm concentration was only 1.14 billions per ml during the 6th fortnight in T2 animals. On discontinuation of thiourea treatment, the sperm concentration increased and reached

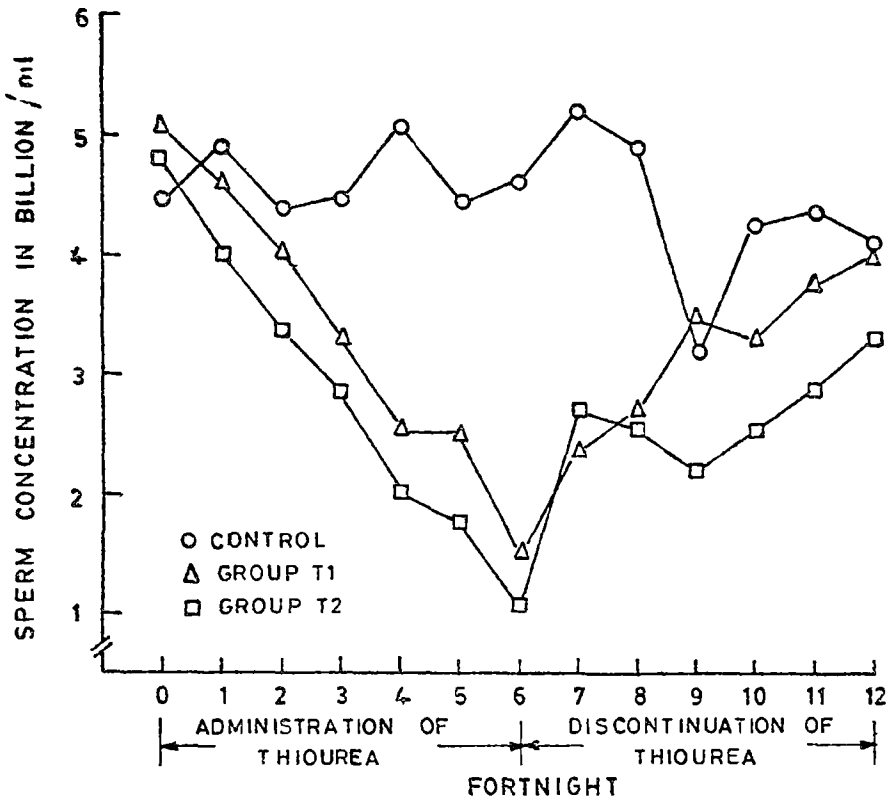


FIG.18. SPERM CONCENTRATION IN EXPERIMENTAL HYPOTHYROIDISM

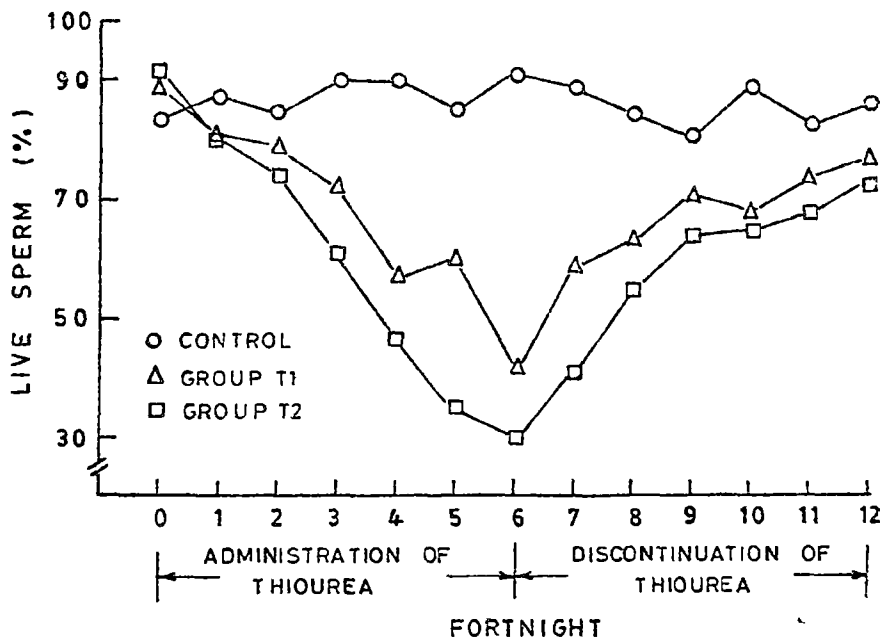


FIG.19. PERCENTAGE OF VIABLE SPERMS IN EXPERIMENTAL HYPOTHYROIDISM

the initial level by the 9th and 12th fortnights in T1 and T2 groups respectively.

4.2.5.4. Livability of spermatozoa

The data on the percentage of live sperms are presented in figure 19.

There was gradual reduction in the percentage of live sperms in animals in group II dosed with thiourea. It reached the lowest level by the 6th fortnight in T1 animals and during the 4th fortnight in T2 animals. The lowest number of live sperms were observed during the 6th fortnight in T2 animals. On discontinuation of administration of thiourea the percentage of live sperms increased and almost reached the normal level by the 7th and 9th fortnights respectively in T1 and T2 groups.

4.2.5.5. Abnormal spermatozoa

4.2.5.5.1. Abnormal heads

The data on abnormal sperm heads are given in figure 20.

Slight increase in the number of abnormal heads was observed in T1 and T2 animals. Highest number of abnormal heads were noted during the 6th fortnight in T2 group.

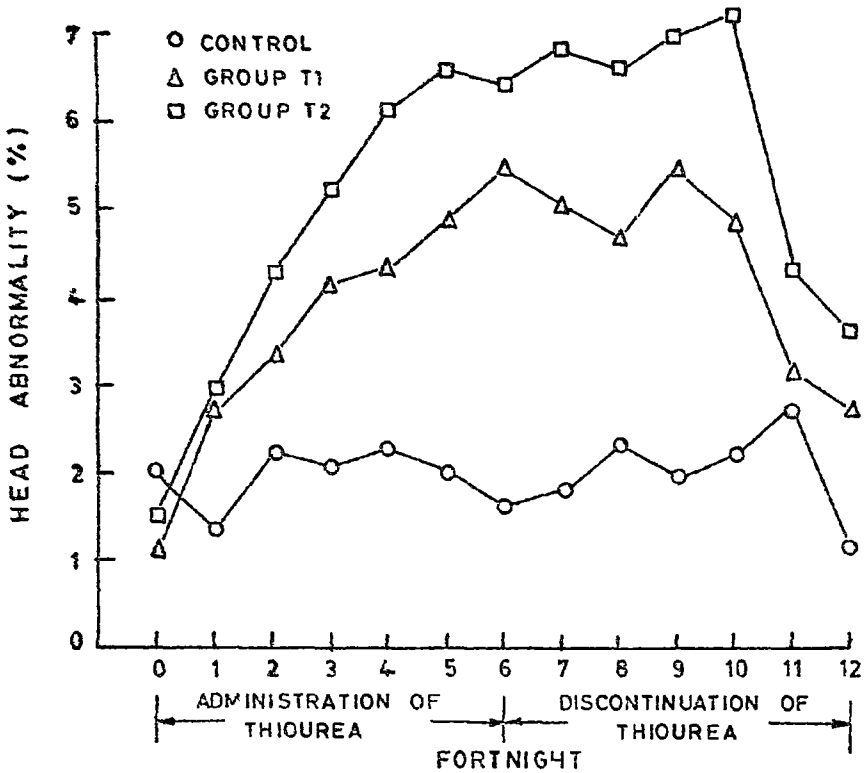


FIG.20. INCIDENCE OF SPERM HEAD ABNORMALITIES IN EXPERIMENTAL HYPOTHYROIDISM

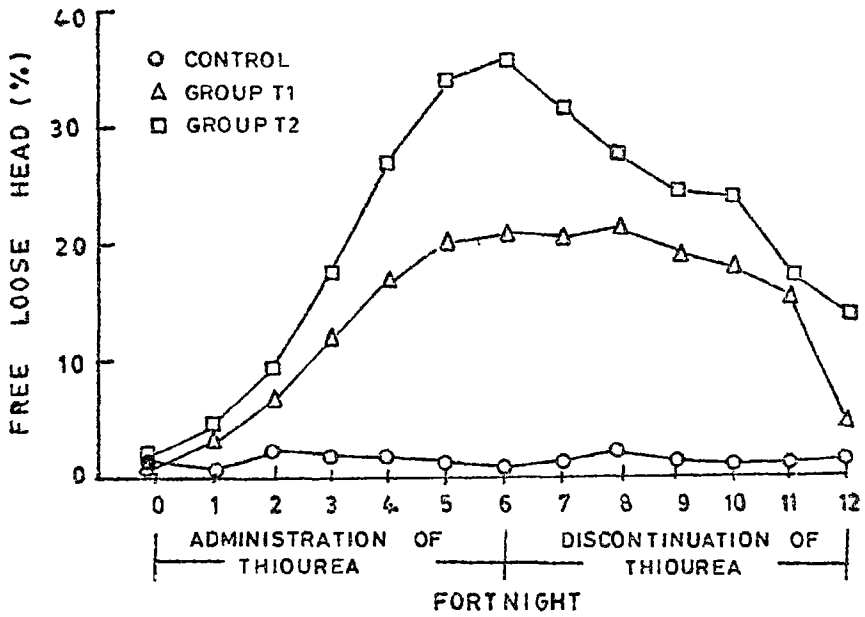


FIG.21. INCIDENCE OF FREE LOOSE HEAD IN EXPERIMENTAL HYPOTHYROIDISM

On discontinuation of treatment with thiourea the percentage of abnormal heads gradually came down and was almost absent by the 10th fortnight in T1 animals and during the 11th fortnight in T2 animals.

4.2.5.5.2. Free loose heads

The data on tailless free heads during the experimental period are presented in figure 21.

A high incidence of free loose heads in T1 and T2 animals of group I was noted during the 2nd fortnight. Thereafter, the increase was gradual till the 6th fortnight. Highest number of free loose heads was recorded in animals in the T2 group (Fig. 22). After discontinuation of oral feeding of thiourea the presence of free loose heads became low by the 12th fortnight in both T1 (4.2%) and T2 (8.4%) groups.

4.2.5.5.3. Middle piece abnormalities

The data on the percentage of abnormal sperms with middle piece abnormalities during the experiment are given in figure 23.

Abnormalities in the middle piece was encountered during the 3rd fortnight. Thereafter, a gradual increase

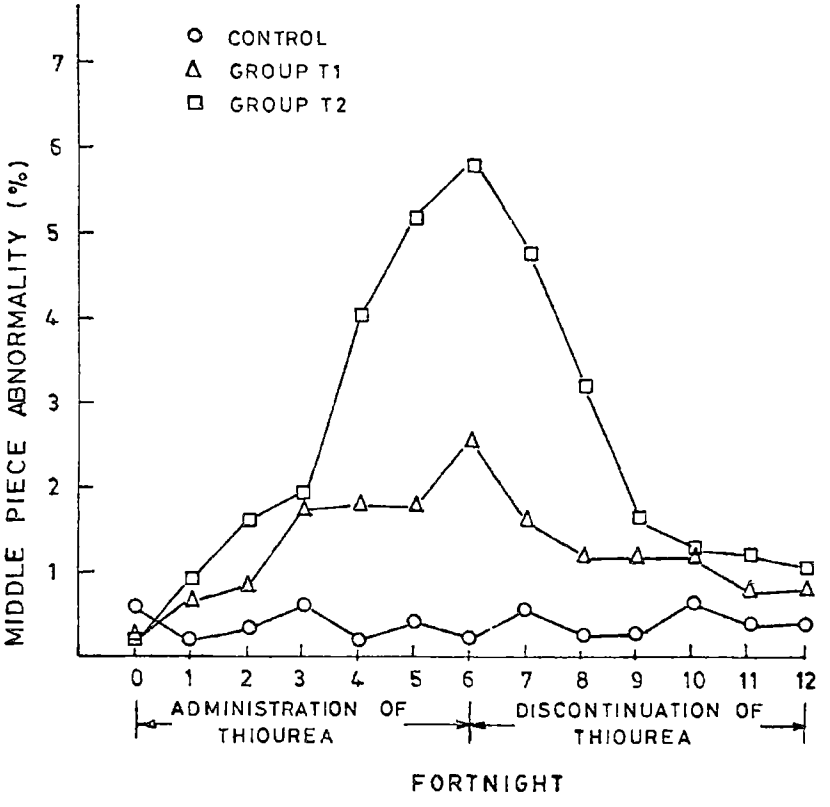


FIG. 23. INCIDENCE OF MIDDLE PIECE ABNORMALITIES IN EXPERIMENTAL HYPOTHYROIDISM

in the percentage of sperms with middle piece abnormalities was observed till the 6th fortnight. Highest number of abnormal middle piece was noted in T2 animals (Fig. 24). On withdrawal of thiourea, the incidence of abnormal middle piece gradually decreased and returned to the normal level.

4.2.5.5.4. Tail abnormalities

The data on the incidence of tail abnormalities are given in figure 25.

In group I animals dosed with thiourea there was progressive increase in the percentage of tail abnormalities during the 1st and 2nd fortnights respectively in T1 and T2 animals and thereafter remained high till the 6th fortnight (Fig. 26). Animals in the T2 group showed highest number of tail abnormalities. On discontinuation of thiourea administration the percentage of abnormalities gradually decreased and the percentage of sperms with abnormal tail were comparable to the normal level by the 12th fortnight.

4.2.5.5.5. Proximal protoplasmic droplets

The data on sperms with proximal protoplasmic droplets are given in figure 27.

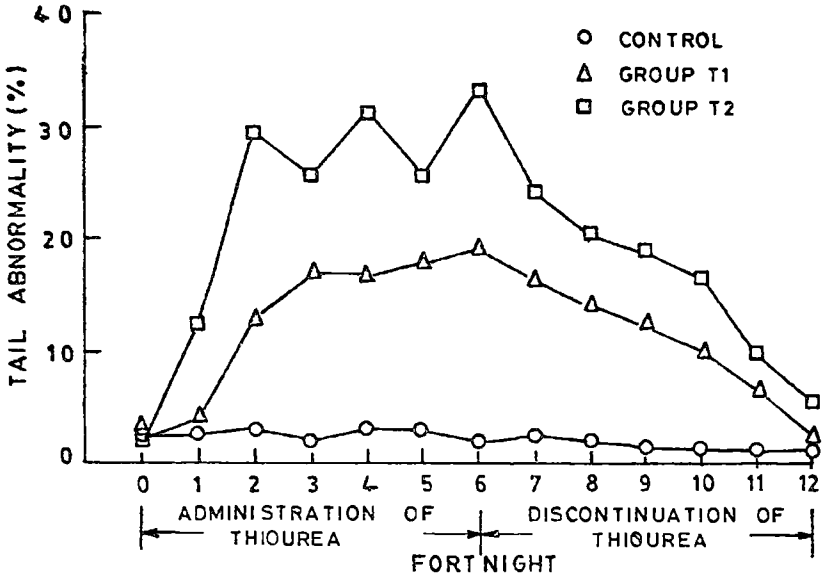


FIG.25. INCIDENCE OF TAIL ABNORMALITIES IN EXPERIMENTAL HYPOTHYROIDISM

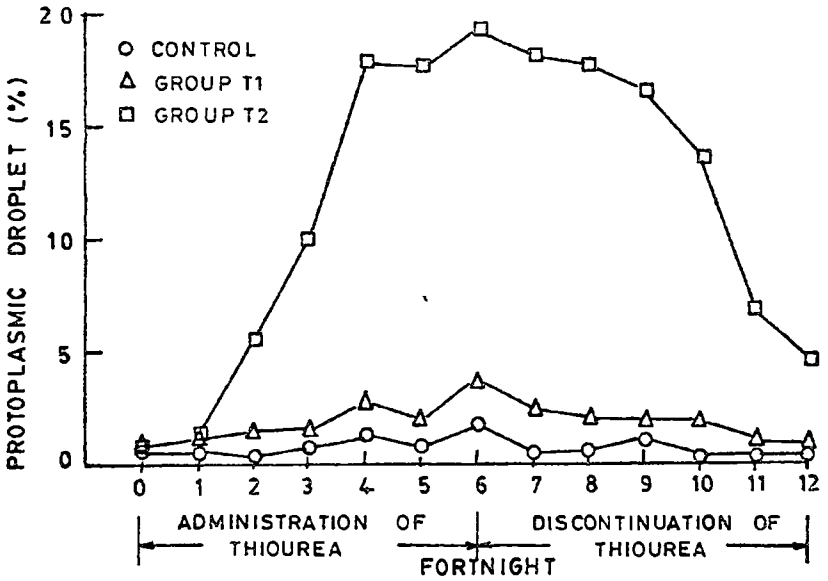


FIG.27. INCIDENCE OF PROXIMAL PROTOPLASMIC DROPLETS IN EXPERIMENTAL HYPOTHYROIDISM

Sperms with protoplasmic droplets were high during the 4th to 6th fortnight in T1 animals and during the 3rd to 11th fortnight in T2 animals. On discontinuation of treatment with thiourea the percentage of sperms with proximal protoplasmic droplets gradually came down and reached the normal level by the 11th fortnight in T1 animals. However, in T2 animals the level was slightly higher even at the end of the 12th fortnight.

4.2.6. Autopsy findings

General

The carcases of all the experimental animals dosed with thiourea were very much emaciated. Subcutaneous oedema of the facial region was evident. There was gelatinisation of the subcutaneous fat in the regions of the neck, hind and fore limbs. Hydropericardium of moderate degree was observed. There was slight hypertrophy and dialatation of the left ventricles. Gelatinisation of the coronary fat was also evident. The carcases of animals which were sacrificed after discontinuation of administration of thiourea did not show any gross lesions.

4.2.6.1. The thyroid gland

The data on the relative weights of the thyroid

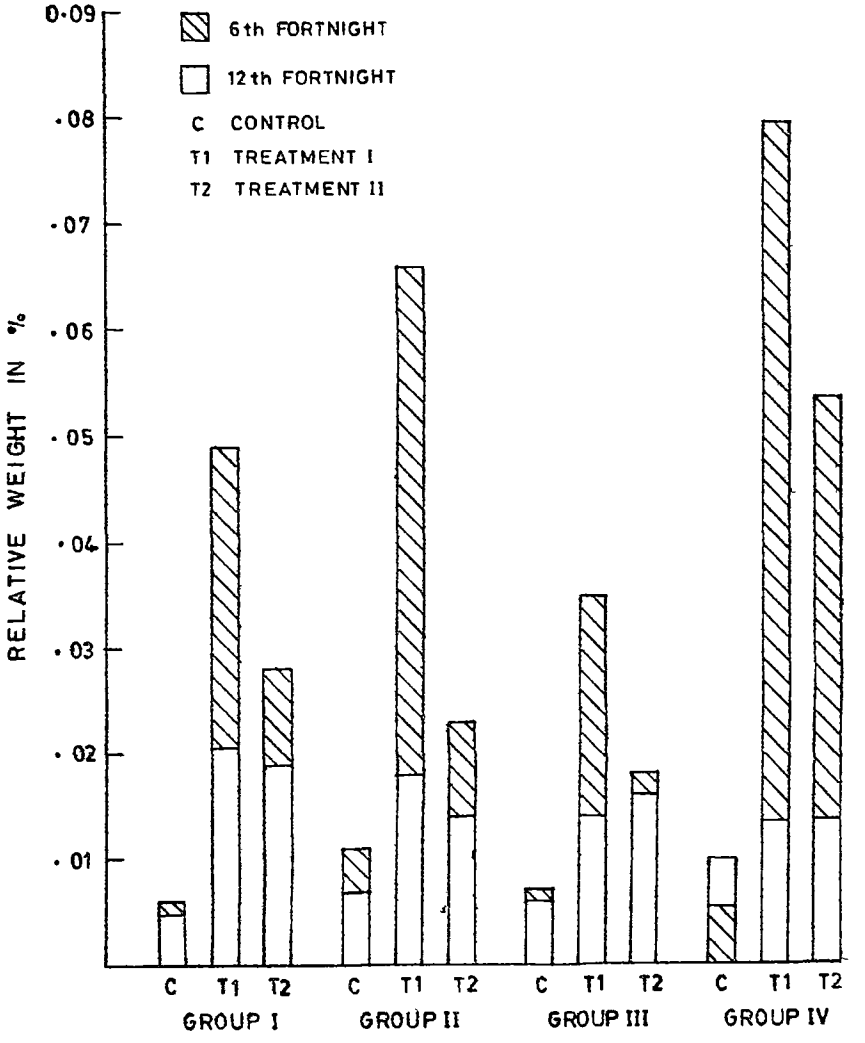


FIG. 28. RELATIVE WEIGHT OF THE THYROID GLAND IN EXPERIMENTAL HYPOTHYROIDISM

gland are presented in figure 28.

There was increase in the relative weight of the thyroid gland in all the animals dosed with thiourea. The increase was more marked in groups II and IV. The increase in the weight of the gland in T2 animals was 5 to 8 times more than the weights of the thyroid gland of age matched control animals. The thyroid gland weighed much less in animals maintained after discontinuation of thiourea when compared with animals dosed with thiourea. However, the weight was slightly above the relative weight of the thyroid gland of control animals. Maximum weight was observed in the thyroid glands of T1 and T2 animals of the group IV.

Gross appearance

The thyroid glands in animals dosed with thiourea were elongated and markedly enlarged and were pale to dark brown in colour (Fig. 29).

Histopathology

The histological picture was characterised by the presence of numerous microfollicles of varying size. Most of the follicles were devoid of colloid and a few contained pale staining vacuolated colloid. The follicles

were lined by tall columnar epithelial cells (Fig. 30). These cells were hypertrophic and had abundant pale acidophilic cytoplasm and a large hyperchromatic basal nucleus. Some of the cells showed vacuolar degeneration of the cytoplasm. In a few of the follicles the lining cells had the tendency to form more than one layer and occasionally the proliferated epithelial cells projected into the lumen of the follicles. Rarely it filled the lumen of the follicles. In some of the follicles there was peripheral scalooping of the colloid. Few of the follicles contained masses of degenerated desquamated epithelial cells. The content of the follicles gave negative reaction with PAS. In focal areas follicles were absent and only groups of dense dark staining nuclear material amidst a mass of homogenous slightly basophilic granular material were seen. This was more pronounced in animals in T2 group. The stroma was scanty. Vessels were moderately engorged. Many of the follicles contained thin pale colloid in T1 animals in groups I and III while most of the follicles were devoid of colloid in T1 animals of groups II and III. In all the animals in the T2 group numerous microfollicles without any colloid were evident (Fig. 31). The hyperplastic changes in the thyroid were

more severe in kids than in the adult animals and in the groups given higher dose of thiourea. The thyroid gland in animals after discontinuation of treatment with thiourea was characterised by well defined follicles with uniform pale staining colloid. The follicles were lined with cuboidal to low columnar epithelial cells. Some of the follicles were distended and were lined with flattened epithelial cells (Fig. 32). The cells were mostly vacuolated. In T2 animals the size of the follicles were relatively large, and few of them contained desquamated epithelial cells.

4.2.6.2. The pituitary

The data on the relative weights of the pituitary gland are presented in figure 35.

There was increase in the relative weight of the pituitary gland of all animals dosed with thiourea. The animals in groups II and III showed pronounced increase in the relative weight of the pituitary. The maximum increase was observed in T2 animals of group II. On discontinuation of thiourea there was decrease in the relative weight of the pituitary and the weight of the pituitary in these animals was comparable with the weight

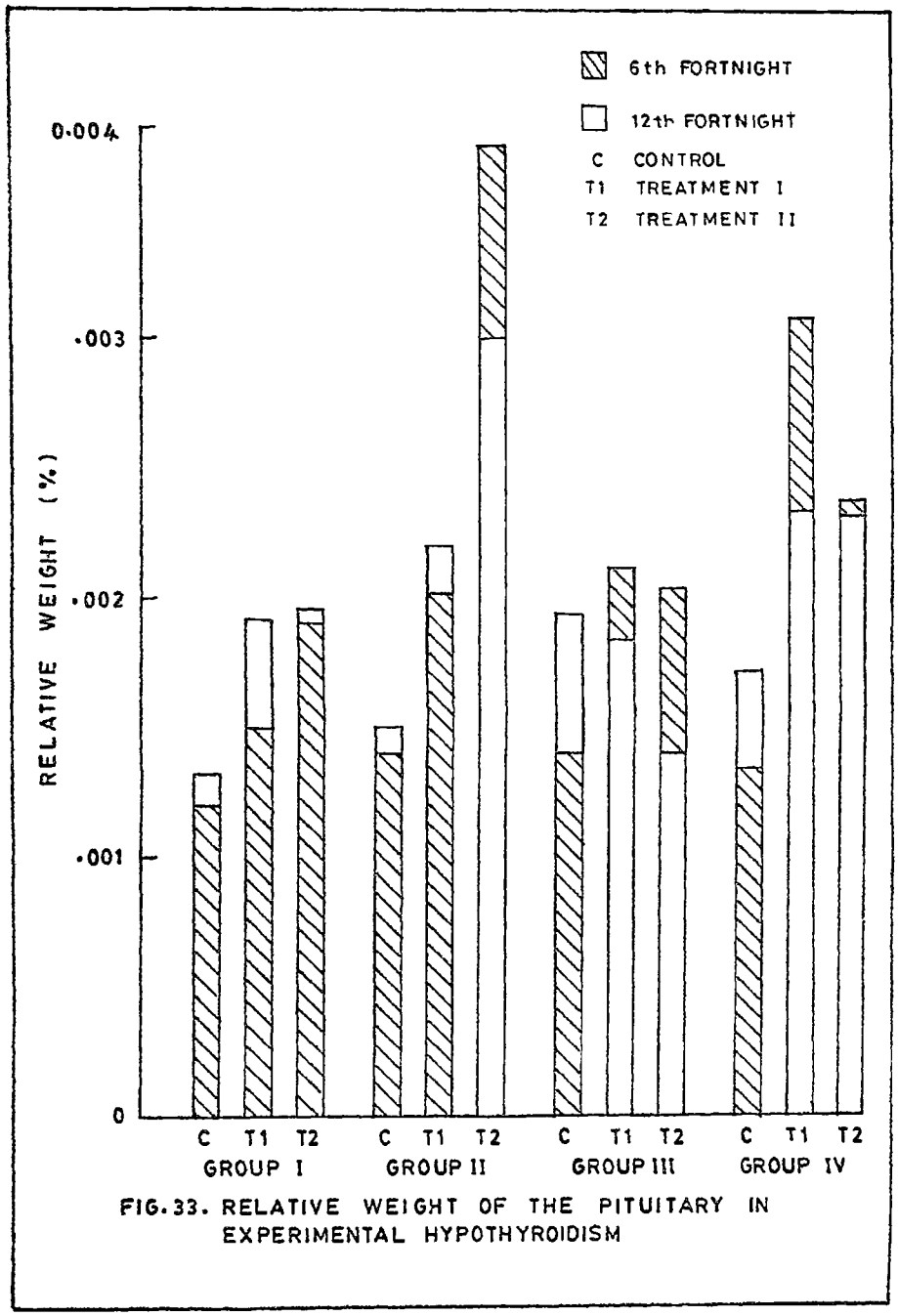


FIG. 33. RELATIVE WEIGHT OF THE PITUITARY IN EXPERIMENTAL HYPOTHYROIDISM

of the pituitary of the control animals.

Gross appearance

The pituitary glands were slightly enlarged in all the animals treated with thiourea.

Histopathology

Histologically there was diffuse hyperplasia and hypertrophy of the basophils and chromophobes in the pars distalis. Acidophils were proportionately much less. There was degranulation and vacuolation of most of the basophils. The hypertrophic basophils were flattened epithelial cells with deeply staining basophilic granular cytoplasm and hyperchromatic centrally placed nucleus. The PAS staining revealed deep purple cytoplasm. In T2 animals more severe degree of hyperplasia of the basophil cells and chromophobes characterised by vacuolation and formation of cysts in focal areas was evident (Fig. 34). Nodular hyperplasia of the chromophobes and basophils was also evident in all the animals in the T2 group. Vessels were moderately engorged. Animals in all the groups showed similar histological changes in the pituitary. After discontinuation of treatment with thiourea the basophils as well as the chromophobes were less in

number and the acidophils appeared in normal proportion. The acidophils were hypertrophic and hyperplastic and had dense acidophilic cytoplasm. Vacuolation in a few basophils was still evident in some areas (Fig. 35). However, the histological picture was almost normal in all the groups of animals.

4.2.6.3. The adrenal gland

The data on the relative weights of the adrenal gland are presented in figure 36.

Animals in all the groups dosed with thiourea showed an increase in the relative weight of the adrenal gland. The increase was marked in groups III and IV. The highest increase in the relative weight of the adrenal was observed in T1 animals of group IV. There was slight increase in the relative weight of the adrenal in T2 animals over T1 animals of groups I and II. In animals after discontinuation of treatment with thiourea, the relative weight of the adrenal gland was almost the same as the weight of adrenal glands in the control animals.

Gross appearance

The adrenal glands were moderately enlarged in all the animals dosed with thiourea.

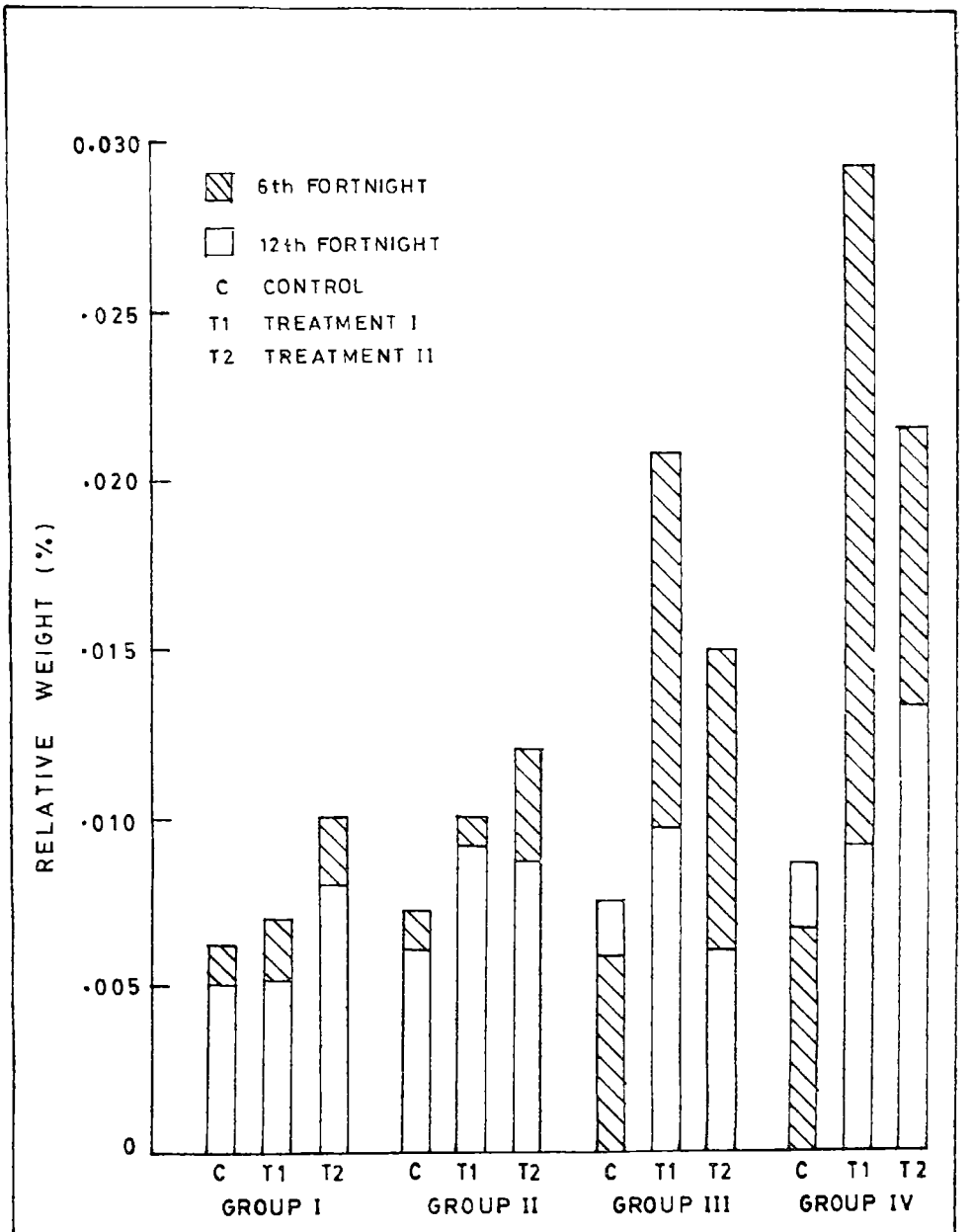


FIG.36 RELATIVE WEIGHT OF THE ADRENAL GLAND IN EXPERIMENTAL HYPOTHYROIDISM

Histopathology

There was moderate diffuse hyperplasia and hypertrophy of the cells of the zona fasciculata. In focal areas the cells were depleted of fat and in certain other areas there were many fatty vacuoles in the cytoplasm and occasionally there were scattered fatty cysts in the zona fasciculata (Fig. 37). Focal areas of hyaline degeneration and necrosis were evident in the cortico-medullary junction. Few accessory cortical nodules characterised by groups of zona fasciculata cells encapsulated by fibrous tissue were evident. The adrenal glands in animals after discontinuation of treatment with thiourea revealed reduction in the width of the zona fasciculata. There was no evidence of any degenerative changes. The capsule showed a few accessory cortical nodules. There was no difference in the histological picture of adrenal glands of animals in different groups.

4.2.6.4. The testis

The data on the relative weights of the testis are shown in figure 39.

All the animals in groups I and II dosed with thiourea showed decrease in the relative weight of the testis. The

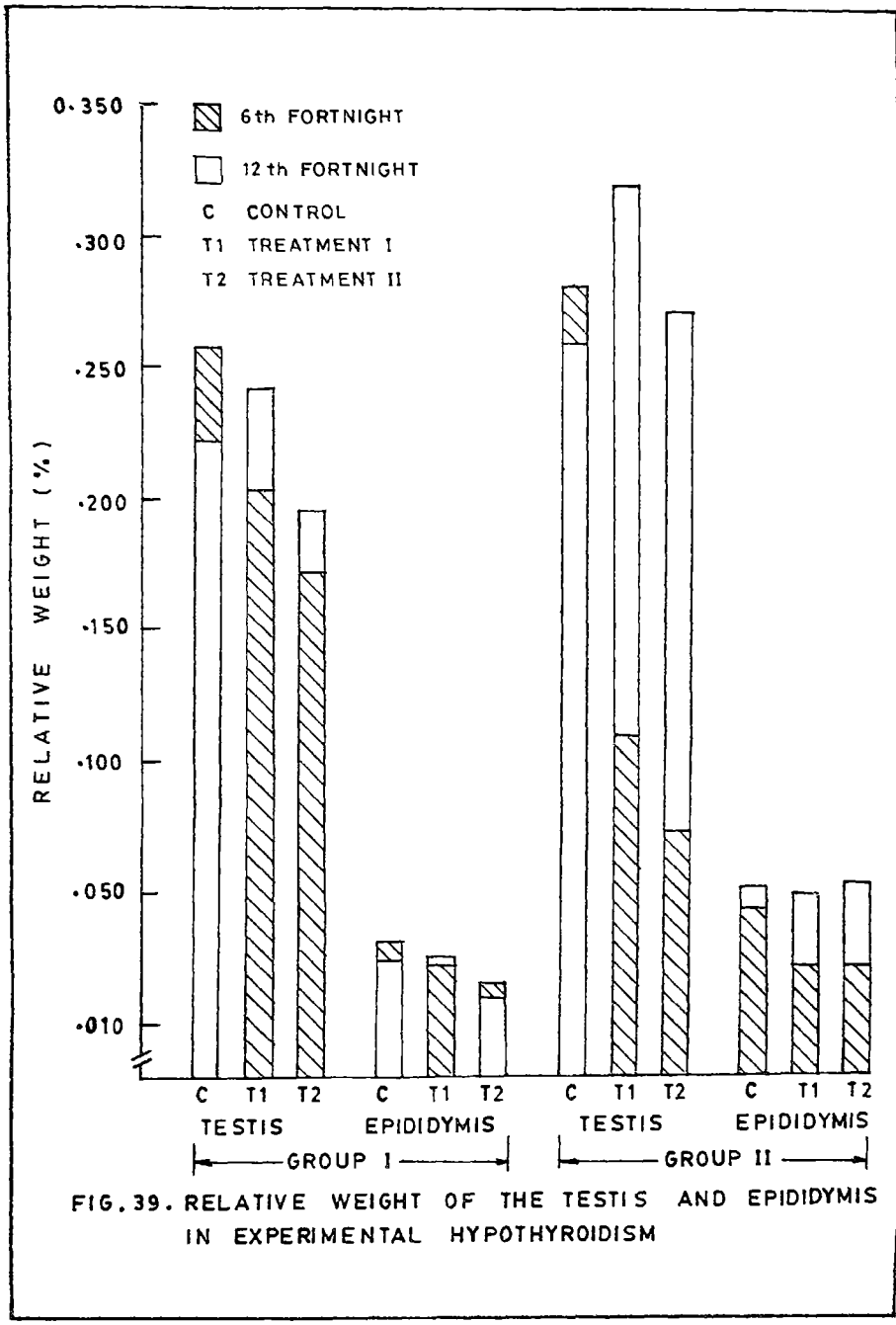


FIG. 39. RELATIVE WEIGHT OF THE TESTIS AND EPIDIDYMIS IN EXPERIMENTAL HYPOTHYROIDISM

decrease was more pronounced in group II when compared to group I. The reduction in the weight of the testis was more in T2 animals than in T1 animals. After discontinuation of treatment with thiourea there was increase in the relative weight of the testis and it reached almost the same weight as that of the age matched control animals.

Gross appearance

The testis of animals in groups I and II was small in size, soft in consistancy and was pale in colour (Fig. 40 and 41).

Histopathology

The seminiferous tubules were small and were lined by single layer of spermatogonial cells when compared to the normal tubules. The tubules contained only a few inactive primary and secondary spermatocytes (Fig. 42). Spermatogenesis was completely absent. Some of the tubules contained degenerated and desquamated cells without any evidence of sperms (Fig. 43). Many tubules contained pink stained granular material. Only a few of the tubules contained sperms and most of the tubules were devoid of any sperms. There was moderate degree of interstitial

oedema. The interstitial cells were scattered and few in number. There was no evidence of sertoli cells in most of the tubules. However, a few of them contained scattered sertoli cells. Only a few scattered spermatogonial cells close to the basement membrane were evident in the testis of animals in group II (Fig. 44) when compared to group I animals. The testis of animals in the T2 group showed severe degree of degeneration than in T1 animals of group I. After discontinuation of treatment with thiourea the spermatogonial cells in the seminiferous tubules were active and the mitotic activity in the cells was evident. The tubules were well formed with organised layering of spermatocytes and spermatids (Fig. 45). Many of the tubules contained sperms. In T2 animals some of the tubules still showed degeneration and desquamation of epithelial cells. The lining cells of the seminiferous tubules were active in group II.

4.2.6.5. The epididymis

The data on the relative weights of the epididymis are shown in figure 39.

There was decrease in the relative weight of the epididymis in animals dosed with thiourea. The reduction

in the weight of the epididymis was marked in group II. In T2 animals of group I, the relative weight of the epididymis was slightly low after discontinuation of treatment with thiourea.

Gross appearance

The epididymis in animals dosed with thiourea was smaller in size (Fig. 40 and 41) when compared to the control animals.

Histopathology

The tubules were smaller in size and some of the tubules did not contain any sperms (Fig. 46). But most of the tubules contained sperms but were only few in number. There was decilation of the epithelial lining of the tubules and the epithelial cells had desquamated in focal areas. Some of the tubules contained scattered desquamated degenerated cells. The lining cells were low cuboidal to flattened epithelial cells and the interstetial tissue was predominant (Fig. 47). The number of tubules were less. There was slight interstitial oedema. In group II animals there was no evidence of sperms at all. On discontinuation of treatment with thiourea, the

epithelial lining of the tubules was found to be intact and the tubules contained sperms (Fig. 48). There was no evidence of degenerative changes. However, a few tubules in T2 animals contained desquamated epithelial cells. There was slight interstitial oedema. The changes in the caput, corpus and cauda of the epididymis were similar (Fig. 49 and 50).

4.2.6.6. The seminal vesicles

The data on the relative weights of the seminal vesicle are presented in figure 51.

In animals in groups I and II dosed with thiourea, there was decrease in the relative weight of the seminal vesicles. The reduction in the weight was marked in T2 animals of group II. After discontinuation of treatment with thiourea the relative weight of the seminal vesicle increased and was almost the same as that of the seminal vesicles of the control animals.

Gross appearance

The seminal vesicles were slightly smaller in size in animals treated with thiourea.

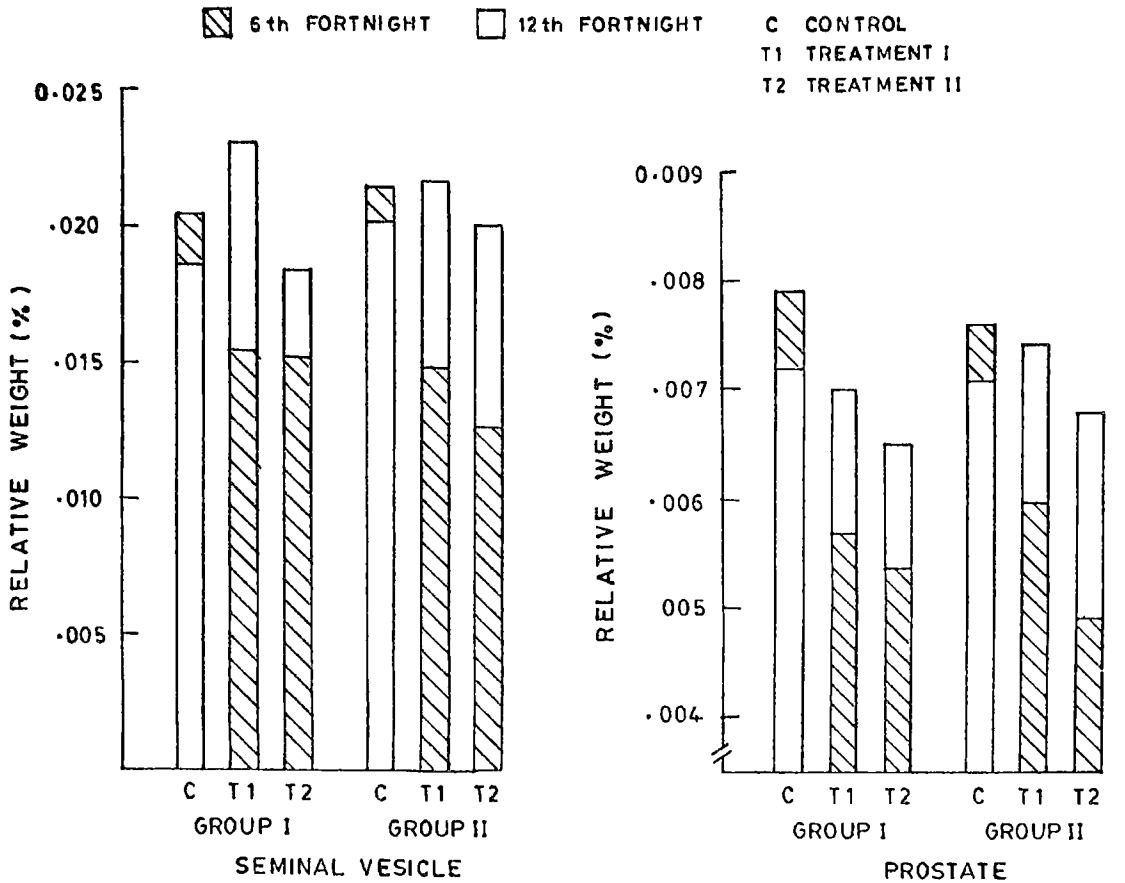


FIG 51 RELATIVE WEIGHT OF THE SEMINAL VESICLE AND THE PROSTATE GLAND IN EXPERIMENTAL HYPOTHYROIDISM

Histopathology

The glands were very small and lined by low columnar, cuboidal or flattened epithelial cells. The cells lining the glands were small and inactive. The secretory activity was not evident in the lumen of the acini (Fig. 52). The cells were vacuolated and hyalinised (Fig. 53). Some of them contained corpora amylacea. In T2 animals of group I the stroma was abundant and the glandular tissue was only less when compared to the animals in T1 group. In group I animals the acini were small in size. The seminal vesicles in animals after discontinuation of thiourea revealed active glands lined by tall columnar epithelial cells. Acini contained secretory material. Stroma was less. In T2 animals only few acini contained secretory material.

4.2.6.7. The prostate gland

The data in the relative weights of the prostate gland are presented in figure 51.

There was marked reduction in the relative weight of the prostate gland in animals dosed with thiourea. The reduction in weight was maximum in T2 animals in the group II when compared to the animals in group I. After

discontinuation of treatment with thiourea the relative weight of the prostate was almost the same as the weight of the prostate gland in the control animals of both the groups. However, the prostate gland of T2 animals in group I weighed only slightly less than that of the control animals.

Gross appearance

There was slight reduction in the size of the prostate in all the animals dosed with thiourea.

Histopathology

The cells lining the glands showed vacuolation and the secretory material was only little in the lumen of the acini. Majority of the acini were small and showed degenerative changes and desquamated cells filled the lumen (Fig. 54). There was hyalinisation of the epithelial lining cells. There was no evidence of any secretory activity particularly in animals in the T2 group (Fig. 55). After discontinuation of treatment with thiourea the prostate histologically showed well formed acini, lined by tall columnar epithelial cells. Most of the acini in T1 animals were more well formed and the cells were intact than in T2 animals.

4.2.6.5. The ovary

The data on the relative weights of the ovary are given in figure 56.

There was decrease in the relative weight of the ovaries in animals in groups III and IV dosed with thiourea. Maximum reduction in weight was observed in T2 animals of group III when compared to T1 and T2 animals of group IV. The relative weight of the ovaries in animals maintained after discontinuation of treatment with thiourea increased and they weighed almost the same as that of the control animals.

Gross appearance

The ovaries in animals treated with thiourea were small, pale, smooth and inactive (Fig. 57 and 58).

Histopathology

The ovaries were inactive. The germinal layer was poorly developed and the cells were of inactive low cuboidal type (Fig. 59). Only a few scattered primary follicles were seen in the cortex. There were only a few isolated well developed secondary follicles. Most of the follicles showed degeneration of the granulosa cells

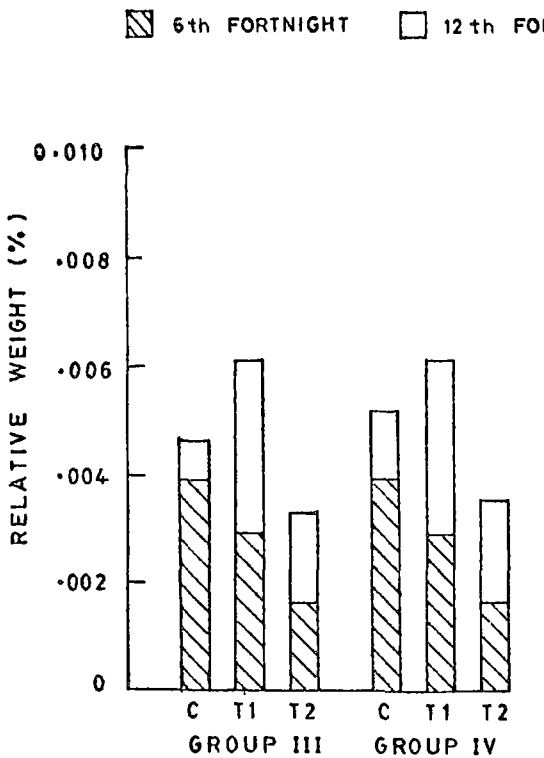


FIG 56 RELATIVE WEIGHT OF THE OVARY IN EXPERIMENTAL HYPOTHYROIDISM

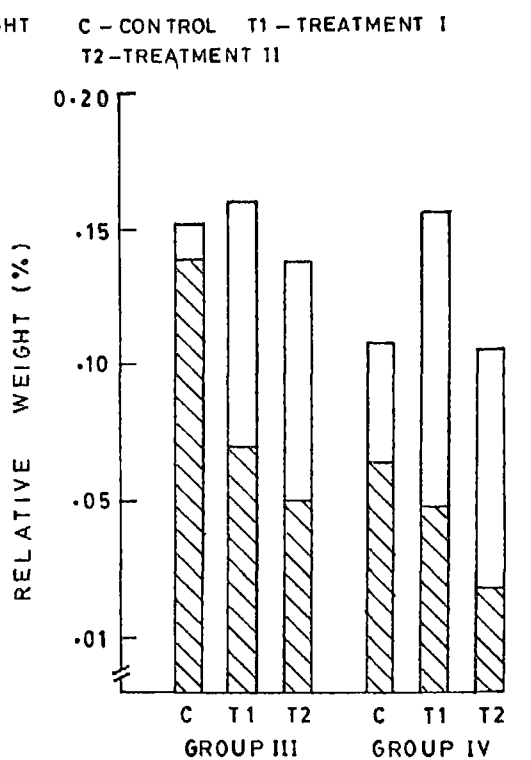


FIG. 63. RELATIVE WEIGHT OF THE UTERUS IN EXPERIMENTAL HYPOTHYROIDISM

(Fig. 60). Mature follicles were relatively few. In some of the follicles degenerating ovum was seen. The stroma was slightly oedematus. There were many degenerated follicles with partial or complete hyalinisation of granulosa cells and the ovum in T2 animals (Fig. 61). While in T1 animals the graffian follicles were small in size and contained only little liquor folliculi. Cells in the theca externa and interna were few. Stromal tissue consisting of closely packed elongated spindle shaped cells was abundant (Fig. 62) in T2 animals than in T1 animals in group IV. In animals after discontinuation of thiourea administration the histological picture of ovaries revealed active stroma with many developing follicles. Mature follicles contained ovum. In T2 animals the ovaries revealed slight fibrosis and a few mature follicles.

4.2.6.9. <u>The uterus</u>

The data on the relative weights of the uterus are given in figure 63.

All the experimental animals of group III and IV dosed with thiourea showed marked decrease in the relative weight of the uterus. In experimental animals of

group IV the reduction in weight was pronounced. The decrease was greater in T2 animals than in T1 animals. The relative weight of the uterus in animals after discontinuation of treatment with thiourea was almost the same as the weight of the uterus in control animals. The weight of the uterus was higher in T1 animals when compared to T2 animals on discontinuation of treatment with thiourea.

Gross appearance

The uterus was small and resembled infantile uterus (Fig. 64 and 65).

Histopathology

The muscular coat was relatively thin and the mucosal glands were few and inactive (Fig. 66). The lining epithelial cells did not show active mitosis. The glands were small in size and were lined with low cuboidal type of eipthelium (Fig. 67). There was no evidence of secretory activity. Moderate degree of submucosal and interstitial oedema was evident. In T2 animals the changes were severe and the glands revealed no secretory activity in the female kids. The lands were slit like and the lining

cells were flattened epithelial cells (Fig. 66). There was slight interstitial oedema. In animals on discontinuation of treatment with thiourea, the uterus showed well developed active endometrial glands lined by tall columnar epithelium. There was evidence of secretory activity. The glands were active and dividing cells were observed.

CHAPTER V

DISCUSSION

DISCUSSION

Survey studies on the thyroid status in goats employing protein bound iodine (PBI) as the marker have established the prevalence of hypothyroid state in goats. Further, the study has yielded valuable information on the incidence of reproductive disorders in hypothyroid animals and has helped to focus attention on the role of hypothyroidism in reproductive disorders. A significant increase in the PBI level in pregnant animals when compared to normal males and females has been documented. In this context it may be pointed out that an increase in the PBI level has been reported in the pregnant laboratory animals (Van Zyl, 1957; Wills and Schindler, 1970), women (Singh and Morton, 1956, Stoffer et al. 1957) and cows (Vzaimosuyaz, 1973). This increase in the concentration of PBI during pregnancy has been attributed to the increased metabolic demand. It is relevant to point out that Galton (1968) observed increased thyroid activity as evidenced by the rate of uptake of iodine in pregnant rats. Significantly low level of PBI was recorded in goats with reproductive disorders. Animals with postpartum anoestrum had the lowest level of PBI. Similar observation has been reported in buffaloes (Noble and Gupta, 1980). In repeat breeders and in animals with a

history of delayed puberty also the level of PBI was low. Similar low PBI values have been reported in repeat breeder cows (Lewis and Kalton, 1953), in infertile human patients (Horne and Thibult, 1961) and in infertile cattle (Allcroft et al. 1954; Vzaimosuyaz, 1973). A perusal of the literature did not reveal any report on this aspect in goats and this appears to be the first report. The results of the present study have indicated a subnormal thyroid function in animals with reproductive disorders and these observations have convincingly established the relationship between hypothyroidism and reproductive disorders. This would substantiate the experimental studies undertaken to assess the effect of hypothyroidism on reproductive organs in this investigation. Based on this observation it can be suggested that serum PBI level of goats particularly with reproductive disorders should be estimated and iodine supplementation or thyroxine therapy should be instituted if the level of PBI is found low, as a treatment measure, in reproductive disorders. This is a finding that should be applied in the field and adequate data should be gathered to elucidate the effect of such a line of therapeutic approach in the treatment of sub-fertility and infertility cases.

Experimentally hypothyroidism was induced in goats

employing low doses of thiourea to study the effects of hypothyroidism on reproductive organs. There are no reports on experimental hypothyroidism in goats except that of Sreekumaran (1976). He employed thiourea at the rate of 100, 150, 200 and 250 mg/kg body weight and did not undertake detailed investigation on the changes in the reproductive system. In the present study thiourea was administered at the rate of 50 mg/kg and 100 mg/kg body weight to induce hypothyroidism and to study its effects on reproduction. The study has yielded valuable information on the manner in which the growth and reproduction are affected in goats by hypothyroidism. Thiourea and related compounds have been used to induce experimental hypothyroidism in different species of animals. In this study it has been possible to induce hypothyroidism of varying degree experimentally by feeding thiourea to goats at low dose levels. The observations made during the course of this investigation have clearly shown that thiourea could be used as an experimental goitrogen in goats to induce a model of hypothyroid state even at very low dose levels without any side effects. Clinically, experimentally induced hypothyroid state was characterised by disturbance in growth and health. There was stunted growth

and weight loss in all the groups of animals dosed with thiourea. This observation clearly points out that normally functioning thyroid is essential for normal physiological growth and development to take place in goats. Similar observations were made in goats by Sreekumaran (1976). However, Lombardi et al. (1962) did not observe any deleterious effect on the growth in dogs dosed with thiourea. This was attributed to the fact that metabolic processes in the dog are less dependent on the thyroid hormone.

Subcutaneous oedema of varying degree was observed in all the animals dosed with thiourea. Only a few reports have appeared describing such myxoedematous changes in goats in hypothyroidism although this is common change in hypothyroid state in man. Sreekumaran (1976) also observed similar changes in hypothyroid kids. Clinically the coat of the animals was rough and matted even at the dose level of 50 mg/kg body weight. The epidermal layer was considered as an important target organ to the action of thyroxine (Freedberg, 1971) and therefore significant pathological changes could be expected in the skin.

The poor condition and gelatinisation of body fat observed in experimental goats would again highlight the

deleterious effects of hypothyroidism. Similar finding has been reported in kids by Sreekumaran (1976). This might have been due to the reduced feed consumption and feed conversion in the absence of thyroxine. Russell (1943) indicated that most energy demands were met from preformed lipid in hypothyroid rats. Therefore, gelatinisation of body fat observed in hypothyroid goats might be due to utilisation of fat for vital functions of the body and energy requirements of the animal.

It is note worthy that all goats showed dilatation and hypertrophy of the left ventricle and showed moderate degree of hydropericardium. Similar findings have been reported in human beings in myxoedema (Zondek, 1918). Pericardial effusion presumbly resulting from the increased capillary permeability might have caused hydropericardium. Cardiac hypertrophy and dilatation could be considered as pathological changes resulting from the effort on the part of the heart to compensate the function in the face of reduced cardiac output and decreased velocity of blood flow in hypothyroidism.

A few of the animals besides being lethargic, weak and depressed showed a tendency to hold the head down as reported by Sreekumaran (1976). He attributed cerebral

oedema as the cause for these clinical manifestation. The disappearance of symptoms and lesions in animals after discontinuation of treatment with thiourea indicated the establishment of normal metabolic process in these animals in the presence of thyroxine synthesis and clarified that the changes are transient and reversible if the causative factors are eliminated.

The reproductive behaviour of hypothyroid goats was studied in detail. Hypothyroidism caused decreased libido in males. This is in agreement with the reports in bulls by Bignett (1952) and Jovanoic et al. (1953). The loss of libido can only be a clinical manifestation of lowered testosterone production in hypothyroidism. The histological demonstration of sparse distribution of interstitial cells in the testis of hypothyroid goats would support this conclusion. The observation of Kumaran and Turner (1949) that there is reduced output of ICSH in hypothyroidism would also support this explanation.

Manifestation of oestrum was weak in does. The failure of normal function of the thyroid-pituitary and ovarian axis in hypothyroidism might be responsible for this. In this context it may be pointed out that Brownstand and Fowler

(1959) reported low ovulation in sows fed thiouracil. This is an observation in support of the present finding. Weak oestrum has also been reported in hypothyroid cows by Spielman et al. (1946) and Calderbank (1963). The appearance of normal oestrum in animals on discontinuation of treatment with thiourea clearly established the necessity for a normal functioning thyroid for clear manifestation of symptoms of oestrum. This observation has clinical significance because in practice silent heat is an important problem in livestock. Hypothyroid state might be responsible for such weak oestrum and iodine supplementation might help in inducing well defined symptoms of heat and to detect heat.

It is significant to observe that animals in almost all experimental groups recorded gain in body weight during the first fortnight. This would suggest that there has been some anabolic effect at lower dose level causing gain in weight. The slow onset of hypothyroidism lowers the BMR and causes the reduction in catabolism of proteins and utilization of energy for body functions and this would normally lead to a transient positive anabolic effect causing gain in weight. Thus it would appear that in low doses thiourea has transient beneficial effect in increasing body

weight when given for shorter periods. It is relevant to mention here that in pigs thiourea in low doses has been used for fattening (Pearson, et al. 1966). The observation made in this study, therefore, indicates that in goats also low doses of thiourea could be of use in fattening goats intended for slaughter. During the subsequent fortnights stunted growth characterised by reduction in weight gain and stature was a constant feature in all goats dosed with thiourea. Continuous suppression of thyroxine production would naturally cause lowered BMR, cell growth and consequent weight loss. It is relevant to mention here that goitrogens have been found to retard growth in sheep (Lascelles and Setchell, 1959) and poultry (Singh et al. 1968). Retardation of growth in hypothyroidism has been attributed to defective synthesis of new proteins by Metzgear and Freinkel (1971). Kimberg (1971) has reported diminution of absorption of nutrients in human beings in the absence of thyroxine. There was reduction in feed intake of animals dosed with thiourea and this might again be a cause for retardation of growth. Similar observation has been reported in pigs dosed with thiourea (Pearson et al. 1966). Stunting in growth in experimental hypothyroidism has been reported in lambs (Marston and Peirce, 1932), in

rats (Green et al. 1974) and in goats (Sreekumaran, 1976). It may be pointed out that there was been a consistant increase in weight gain and growth of animals after discontinuation of treatment with thiourea. The blocking effect of hypothyroidism on growth rate has been clarified by this observation. This would suggest that there has been synthesis of thyroxine following discontinuation of treatment with thiourea and the effect of thiourea was transient and reversible.

The serum protein bound iodine level, in animals dosed with thiourea was significantly low. This observation is in close agreement with the results of experimental hypothyroidism in sheep (Lascelles and Setchell, 1959), in bull (Lewis, 1956) and in kids (Sreekumaran, 1976). From the observations made during the course of this investigation it is reasonable to conclude that the PBI level could be used as a reliable marker to screen the existence of hypothyroid state in goats. The data will truly reflect the degree of hypothyroid state in the animal. Thiourea inhibits the organification of iodide and iodination causing reduced synthesis and release of thyroid hormone (thyroxine) from the gland (Calderbank, 1963). Reduction in the PBI level was very much pronounced in the high dose level group.

suggesting that thiourea administered at the higher dose has markedly suppressed iodination in the thyroid gland and caused pronounced deficiency of thyroxine. A significant observation, worth noting is that kids and females in all the groups showed lower values than the males. This is an observation which would support the view that kids and females are more prone to develop hypothyroidism and are more susceptible to the action of the antithyroid drugs (Underwood, 1971; Sreekumaran, 1976). There was increase in the PBI level after discontinuation of administration of thiourea. This would suggest that thiourea is an effective antithyroid drug and it blocks the biosynthesis and release of thyroid hormone. The blocking effect was identified to be reversible since on discontinuing the administration of thiourea the PBI level came back to the normal.

There was increase in the total serum protein level in animals dosed with thiourea. Similar increase in the serum protein level was reported in hypothyroidism in human beings (Lamberg and Grasbeck, 1955). An increase in the serum globulin level was reported in thyroidectomised Merino rams (Belonje, 1967) and in experimental hypothyroidism in chicken and goats (Nangia et al. 1975; Sreekumaran,

1976). Crispell and Wilson (1964) documented reduction in both anabolism and catabolism of protein in hypothyroidism in man and they observed that catabolism was more reduced than anabolism of protein in hypothyroidism. This may be pointed out as an explanation for the increased protein level in hypothyroidism. Besides this, the defective utilisation of nutrients in the absence of thyroxine causes accumulation of protein (Mangia et al. 1975). A decrease in the serum protein level to normal values after discontinuation of treatment with thiourea is indicative of synthesis of thyroxine in the thyroid gland. In the presence of adequate quantity of thyroxine there is perforce a higher metabolic rate and this will undoubtedly cause acceleration of enzyme turnover with a resultant increase in the demand for coenzymes and proteins as well as an increase in the break down of these essential materials. The stimulating effect of thyroprotein on the metabolism has been convincingly established by McCartney and Shaffner (1950).

Hypercholesteraemia was a characteristic finding in animals dosed with thiourea and could be considered as an important biochemical parameter in hypothyroidism. Increase in the serum cholesterol level has been reported in experimental hypothyroidism in sheep (Lascelles and Setchell, 1959;

Belonje, 1976) in chicken (Mangia et al. 1975) and in goats (Sreekumaran, 1976). The present finding, therefore, is in agreement with the observations of earlier workers. The increase in the serum cholesterol level has been considered as a specific change in lipid metabolism by Peters and Man (1950) in human myxoedema. Fletcher and Myant (1956) indicated that although in hypothyroid rats the hepatic synthesis and release of cholesterol from acetate was subnormal but the peripheral break down and biliary excretion is lowered still more and this they ascribed as the reason for increase in the serum cholesterol level. It is reasonable to assume that the same mechanism prevails in goats also.

The blocking effect of thiourea on thyroxine synthesis was eliminated on discontinuation of treatment with thiourea and this was soon followed by decrease in the serum cholesterol level. This observation elucidates the stimulatory effect of thyroxine on degradation of lipids. According to Mangia et al. (1975) metabolic reactions of lipids are stimulated by thyroid hormones. From the observations made during the course of this investigation it is reasonable to conclude that the level of cholesterol in conjunction with PBI level could be used as reliable test to screen the

existance of hypothyroid state in goats. In practice these diagnostic parameters can be effectively exploited to screen goats to detect hypothyroid state.

There was decrease in the total erythrocyte count, haemoglobin level and packed cell volume in all the experimental animals. The blood picture revealed microcytic hypochromic anaemia. This is in accordance with the findings of Sreekumaran (1976) and indicates that thyroxine deficiency would lead to anaemic state. Rivlin (1971) observed that the most significant effect of hypothyroidism in man is reduction of intestinal absorption of Vitamin B_{12}. Howfar this will be applicable to ruminants has to be clarified by further detailed investigations. It is relevant to mention here that Adamson and Finch (1966) have demonstrated decreased production of erythropoietin in hypothyroidism. On discontinuation of thiouren there was improvement in the haemogram values and almost reached the normal level. This can be attributed to the stimulatory influence of thyroxine on haemopoiesis.

A significant decrease in the quantity and quality of semen in experimentally induced hypothyroid state in goats is a significant observation and is a direct evidence to establish the effects of hypothyroidism on the reproductive

performance in goats. Decrease in the ejaculate volume, sperm concentration, motility and an increase in the incidence of abnormal spermatozoa were the important adverse effects observed in the semen in hypothyroid state. Similar deterioration in semen quality has been reported in iodine deficient bulls (Mignett, 1952) and in hypothyroid bulls (Swanson and Boatman, 1953). There was significant pathological changes in the accessory sexual glands of hypothyroid goats on histological examination and these structural abnormalities would naturally lead to impaired secretory activity. The decrease in the ejaculate volume, could therefore, be explained as an effect of impaired secretory activity of the accessory sexual glands in hypothyroid state. Significant reduction in sperm concentration, motility and viability are the manifestations of the derangement in the testicular function and the presence of head abnormalities in sperms is an indication of disturbed spermatogenesis. According to Rollinson (1951) and Roberts (1971) increased number of free loose heads and a high incidence of middle piece and tail abnormalities are suggestions of testicular degeneration and epididymial dysfunction. The high incidence of sperms with proximal protoplasmic droplets according to Roberts

(1971) is definitely an indication of failure of maturation of spermatozoa. The histopathological examination of the testis and accessory sexual organs have revealed significant pathological changes and these changes would account for the clinical abnormalities. The evaluation of semen characteristics of hypothyroid goats has clearly indicated that the testis, epididymis and accessory sexual organs are adversely affected in the absence of adequate quantity of thyroxine. There was significant improvement in the quality and quantity of the semen in hypothyroid animals after discontinuation of treatment with thiourea. This observation is in close agreement with the results reported in iodine deficient rams and in rams following iodine supplementation (Baicoianuo, et al. 1963; Mahltiev, 1966). It is relevant to point out that thyroxine supplementation has been shown to improve the semen quality in bulls (Maqsood, 1952).

The results obtained during the present investigation, therefore, draws attention to the fact that hypothyroidism could be an important factor associated with poor quality of semen. Further the reversibility of the damaging effects clarifies the scope for instituting corrective measures.

It is pertinent to point out that the changes observed in the semen were significant and could adversely effect

the reproductive efficiency of the animals. The sub-fertility and infertility observed in goats in natural situations might also be due to hypothyroid state induced by absolute iodine deficiency or due to intake of goitrogens. This is an observation which has significant practical application and should be thought of when chalking out programmes to control infertility in livestock.

There was significant increase in the relative weight of the thyroid gland in all the goats dosed with thiourea. The increase was more pronounced in goats fed lower dose of thiourea. This is an evidence to show that there has been reactive hyperplastic response in the thyroid under the influence of thiourea. The increase in the weight of the thyroid is a manifestation of a compensatory hyperplastic response mediated through the pituitary under the influence of low thyroxine level. It would appear that the stimulatory effect was more effective when the thyroxine level was not very low. It is significant to observe that there was correlation between the age, dose and degree of enlargement. There was pronounced enlargement of the thyroid gland in kids than in adults demonstrating that growing animals are more

thyroxine dependent than adults. Similarly symptoms of hypothyroidism were more severe in kids when the same dose was administered to kids and adults. These are facts which would support the conclusion that the effect of thiourea is dose and age dependent. No doubt irrespective of other factors there has been compensatory hyperplastic thyroid response mediated efficiently through pituitary. However, functionally it was not found to be compensated since the PBI values in the thiourea dosed goats were much lower when compared to euthyroid animals. Although there was significant increase in the weight of the gland in experimental animals, the enlargement was not appreciable on palpation during the clinical examination. This observation is pertinent and has to be stressed since this points to the fact that subclinical hypothyroidism can exist in animals without gross evidence of thyroid enlargement. Therefore, palpable thyroid enlargement, cannot be taken as a criterion for diagnosing subclinical hypothyroid state in animals. However, thyroid enlargement has been reported in experimental hypothyroidism in different species of animals (Kennedy, 1942; Jones et al. 1946; Harkness et al. 1954; Goldberg et al. 1957; Lascelles and Setchell, 1959; Lazo-wasem,

1960; Sreekumaran, 1976) and in spontaneous hypothyroidism in sheep and goats (Southcott, 1945; Lall, 1952; Dutt and Kehar, 1959). Enlargement may be a feature when the condition is long standing. There was reduction in the weight of the thyroid gland after discontinuation of treatment with thiourea. This observation shows that increase in the level of thyroxine following discontinuation of thiourea administration has prevented the pituitary from releasing TSH and stimulation of thyroid.

Reactive hyperplasia was the characteristic histological picture observed in the thyroid gland. In hypothyroid state this is an expected pathological change. This is a direct histological evidence which shows that pituitary mediated compensatory hyperplasia has taken place. Thyroid hyperplasia has been reported in spontaneous hypothyroidism in sheep (Growth, 1962; Wallach, 1965; George *et al.* 1966) in goats (Lall, 1952; Dutt and Kehan, 1959; Roy *et al.* 1964) and in experimental hypothyroidism in laboratory animals (Jones, 1946; D'Angelo *et al.* 1951; Durlach *et al.* 1954; Goldberg *et al.* 1957) and in goats (Sreekumaran, 1976). The hyperplastic response was more severe in females than in males. This

is another evidence which would support the conclusion that females are more sensitive to hypothyroidism than males. The most important histological observation was the complete absence of colloid in most of the follicles of the thyroid. It was difficult to identify the tissue as thyroid on histological examination. This would suggest that, although there was stimulation by TSH and hyperplasia of thyroid epithelium, there was no synthesis of thyroglobulin due to the non-availability of iodine in the presence of thiourea. This would support the observation that thiourea has effectively blocked the thyroglobulin production and has lowered the PBI level. Formation of new small follicles without having colloid indicates that TSH stimulation was very severe and blocking of iodide uptake and incorporation of iodine were effective. Distended follicles lined with cuboidal to low columnar epithelial cells were the characteristic histological picture of the thyroid gland in animals after discontinuation of thiourea administration. This is an evidence to conclude that synthesis of thyroglobulin has resumed when the blocking factor was removed and iodine was available.

There was increase in the relative weight of the

pituitary gland in all the animals dosed with thiourea irrespective of the dosage. Similar observations have been reported in experimental hypothyroidism in laboratory animals (Kennedy and Purves, 1941; Griesbach et al. 1941; Goldberg et al. 1957; Lasc-wasen, 1960), in goats (Sreekumaran, 1976) and in spontaneous hypothyroidism in goats (Lalli, 1952). The increase in the relative weight of the pituitary was much more in animals given higher dose of thiourea. The thiourea interferes with organic binding of iodine and consequent deficiency of thyroxine stimulates the basophil cells to produce more TSH. The stimulation leads to hyperplasia and hypertrophy of the basophil cells to meet the increased demand for TSH. This hyperplastic response is reflected in the increase in the weight of the pituitary. There was decrease in the relative weight of the pituitary in animals after discontinuation of thiourea administration. This is an anticipated change since feed back inhibition of pituitary is bound to happen when the thyroxine level reaches the normal level.

In the pituitary glands there was hypertrophy and hyperplasia of the basophils. Basophil cell hyperplasia is an evidence to support the conclusion that thyroid

activity was diminished by the administration of thiourea to goats because basophil hyperplasia is a physiological response to stimulate the thyroid in the absence of adequate quantities of thyroxine in the blood. With increasing thyroid dysfunction in the goats the initial hypertrophy of the basophil cells associated with storage of granules was followed by the loss of granules in many of the cells and finally complete degranulation and vacuolation of the basophil cells. These vacuolated basophils have been described in rats as "Thyroidectomy cells" (Zeckwer et al. 1935). The cytoplasmic vacuolation according to them represents an exhaustion stage in the reactive hyperplastic process. These changes have been described in experimental hypothyroidism in dogs (Lippincott et al. 1957), in rats (Goldberg and Chaikoff, 1951) and in goats (Sreekumaran, 1976). The hypertrophy of basophil cells has also been reported in spontaneous hypothyroidism in goats and sheep (Lall, 1952; Dutt and Vasudeva, 1963).

The acidophils showed degranulation and degeneration. This might be due to the feed back inhibition of the acidopuils resulting from inefficient utilisation of growth hormone produced by the pituitary in the absence of thyroxine. Similar histological changes were described

in the pituitary acidophil cells in hypothyroidism, induced by thyroidectomy and goitrogens in laboratory animals (Zeckwer et al. 1935; Goldberg et al. 1957; Contopoulos et al. 1958) and in goats (Sreekumaran, 1976).

There was significant increase in the relative weight of the adrenal glands in animals treated with thiourea. This observation is in contrast to the reports of atrophy of adrenal glands in laboratory animals dosed with thiouracil and allied compounds by Baumann and Marine (1945), Barrow and Money (1949) and McCarthy et al. (1959). However, the present observation is in agreement with the findings of Durlach et al. (1954) who have reported an increase in adrenal weight in guinea-pigs dosed with propylthiouracil. Similar observations have been reported in experimental thyroidectomy in laboratory animals (Gley, 1923) and in goats (Sreekumaran, 1976). The animals with induced hypothyroid state was under the influence of stress and this stress might have been responsible for the enlargement of the adrenal glands. Microscopic changes like depletion of fat, focal areas of haemorrhage, degeneration and necrosis of zona fasciculata seen in the adrenal glands of goats dosed with thiourea are all histological features described by

Symington (1969) in stress reaction in the adrenal gland. The relatively more increase in the adrenal weight in females would suggest that the stress reaction is more severe in females than in males. The reduction in the relative weight of the adrenal glands in goats after discontinuation of thiourea suggests that the animals were relieved of stress response when thyroxine synthesis was normalised.

There was significant decrease in the relative weight of the testis and epididymis in all the goats dosed with thiourea, an observation which has not been previously reported and has a direct bearing on the reproductive performance of goats. The decrease in the weight was more pronounced in kids and adults given higher dose of thiourea. Although no reports have appeared describing the changes in the epididymis of goats in hypothyroidism, similar observations were made by Del Rio et al. (1979) in thyroidectomised rats. On the contrary Prasad and Singh (1971) and Sharma and Singh (1975) recorded increase in the weight of the testis in chicks dosed with thiourea. However, the present findings are in agreement with the observation of Peczely et al. (1979) in quails. They observed low level of testosterone in hypothyroid state

in quails and postulated that this would cause failure of synergism between thyroxine and testosterone and this they attributed as the reason for reduced growth and development of testis in hypothyroidism. There was increase in the relative weight of the testis in hypothyroid animals after discontinuation of administration of thiourea. This is an observation which would support the conclusion that thyroxine is essential for the normal growth and development of the gonads.

Histologically the testis revealed only a few spermatogonial cells and scattered spermatocytes in the seminiferous tubules and there was no evidence of mitotic activity in the spermatogonial layer. Coupled with this arrested activity there was degenerative changes. The histological changes would therefore suggest a basic defect in spermatogenesis and maturation of sperms. The seminiferous tubules were small in size and the sertoli cells were sparse and interstitial cells were only few and scattered. These observations provides histological evidence to conclude that all the cellular components of the testis are affected in hypothyroid state. Similar changes have been reported in experimental hypothyroidism induced by thiouracil in rams and rabbits (Maqsood, 1951).

According to Gorbman and Bern (1974) thyroxine has a priming effect on the action of hormones on cells and they indicated that in the absence of thyroxine, the gonadotrophic hormones of the pituitary will not function effectively and this may lead to degenerative changes in the testicular tissue. Brooks et al. (1964) observed reduction in gonadal and gonadotrophic hormones in thyroidectomised ewes. Besides this the lowered protein synthesis and basal metabolic rate in hypothyroidism may also contribute to the development of degenerative changes in the gonads. In this context it is pertinent to point out that no reports have appeared describing the changes in gonads of both young and adult goats in experimental hypothyroidism of sub-clinical nature and this would be the first report on this aspect. These observations are of utmost importance. Although clinical symptoms may not be pronounced in hypothyroid state of low degree there could be significant degenerative changes in the testis and this basic defect can cause subfertility in goats.

On discontinuation of administration of thiourea there was evidence of organisation of spermatocytes and spermatids in the seminiferous tubules and actively dividing spermatogonial cells. Therefore, the changes in

the male sex organs were also transient and reversible as in the case of other organs and the regenerative changes were almost complete. This is an important observation which would clarify the scope for therapy in reproductive disorders associated with hypothyroidism.

There was significant reduction in the relative weight of the accessory sexual glands; the seminal vesicles and the prostate gland in animals dosed with thiourea. This observation has clearly demonstrated the adverse effect of hypothyroidism on the structure and function of the accessory sexual organs. The reduction in the weight of the glands was associated with degenerative changes in the gland. The changes observed could be caused by the failure of thyroid, pituitary and gonadal axis. It is relevant to mention here that low levels of FSH, LH and testosterone were reported in thyroidectomised hypothyroid rats by Del Rio et al. (1979). Further, on discontinuation of thiourea the accessory glands showed secretory activity and the lining cells were normal confirming this surmise.

There was significant decrease in the relative weight of the ovaries in all the animals dosed with thiourea. In contrast Prasad and Singh (1971) reported an increase in

the weight of the ovary in experimentally induced hypothyroidism in chicken. The increase in the weight was attributed by them to the presence of large number of growing follicles under the influence of low dose of propylthiouracil. However, no such reports are available describing the ovarian changes in hypothyroidism in goats. The ovaries in hypothyroid goats contained only a few scattered developing primary and secondary follicles in contrast to large number of follicles seen in hypothyroid chicken. Absence of mature follicles and corpora lutea would suggest that the ovaries were inactive and this inactivity can be reasonably assumed to have been caused by the inadequate synthesis of thyroxine in the presence of thiourea. The failure of thyroxine synthesis and consequent failure of the pituitary ovarian axis might have caused poor growth and development of the ovaries. In this context it may be pointed out that Wong et al. (1980) have reported low LH and FSH levels in hypothyroidism induced by propylthiouracil in rats. On discontinuation of thiourea there was significant increase in the weight of the ovaries and this was associated with normal development of follicles and formation of corpora lutea under the influence of gonadotrophic and gonadal hormone in the

presence of normal thyroxine synthesis. Similar ovarian regenerative activity has been reported in anoestrum associated with iodine deficiency in cows after iodine supplementation by Kovalskii et al. (1970). The reversibility of ovarian degenerative changes indicates that reproductive disorders associated with hypothyroidism can be corrected. This finding has great significance because if reproductive disorders associated with hypothyroidism are detected and a systematic approach for control is drawn up in the country economic loss due to subfertility and infertility caused by hypothyroidism can be controlled.

A significant reduction in the relative weight of the uterus in all the animals dosed with thiourea was another important finding. This is an observation similar to that reported in hypothyroid rats by Kirkland et al. (1981). The muscular layer was relatively thin and the mucosal glands were few and inactive and revealed no secretory activity. These are clear indications of arrested growth and development of the uterus in the absence of adequate quantity of thyroxine. These findings are similar to that reported by Sreekumaran (1976) in hypothyroid kids. The lining cells of the glands did not show active mitosis as observed by Kirkland (1981) in

the uterus of hypothyroid rats. The reduction in weight according to them was due to the poor growth response of the tissue caused by lowered DNA synthesis and low mitotic index in hypothyroid animals. There was increase in the relative weight of the uterus after discontinuation of treatment with thiourea. Histologically the uterine glands were active and showed mitotic figures. This can be attributed to the effect of onset of thyroxine synthesis and its synergesism with pituitary and gonadal hormones. Wong et al. (1960) observed an increase in the pituitary concentration of FSH and LH in hypothyroid rats following treatment with thyroxine. The same mechanism will operate in goats when the administration of thiourea is discontinued.

Significant pathological changes were observed in the reproductive organs of both males and females in hypothyroid state and these observations have categorically established the role of hypothyroidism in reproductive disorders. It was also established that young animals are more susceptible to the effects of hypothyroidism than adults. From the results of this investigation it can be concluded that in the goat population sub-clinical hypothyroid state is existing and this is bound to cause

infertility problem in both males and females. The importance of hypothyroidism in subfertility and infertility in goats has been established and the reversibility of the condition has been convincingly proved by this investigation. This is an important contribution and this will greatly help in chalking out programme to identify and institute therapeutic and prophylactic measures for controlling infertility in livestock.

CHAPTER VI

SUMMARY

SUMMARY

Employing the protein bound iodine level in the serum as a marker a survey study was conducted to assess the thyroid status of goats with different types of reproductive disorders. Hypothyroidism was encountered in postpartum anoestrus animals, repeat breeders and in animals with a history of delayed puberty.

Hypothyroidism was induced in both male and female goats of different age groups employing low doses of thiourea (50 mg/kg and 100 mg/kg) to study the effects of subclinical hypothyroidism on the reproductive organs.

Thiourea was found to be a useful experimental goitrogen to induce a model of hypothyroid state in goats at very low dose levels without any side effects.

Weakness, lethargy, depression reduction in feed intake, subcutaneous oedema of varying degree, loss of libido in males and anoestrum in females were the important clinical features observed in hypothyroid animals. All the clinical manifestations gradually disappeared on discontinuation of treatment with thiourea.

Stunted growth and appreciable reduction in weight was a consistent feature in all hypothyroid goats.

However, in the first fortnight a gain in weight was recorded. This indicated that feeding thiourea for short periods was useful for fattening goats. On discontinuation of treatment with thiourea goats recorded progressive gain in weight.

A significant decrease in the serum PBI was recorded in all groups of animals dosed with thiourea. Young animals showed much lower values of PBI than adults. There was increase in the serum protein and cholesterol level in hypothyroid goats. On discontinuation of thiourea administration the level of serum PBI, total protein and total cholesterol reached the normal level. The results of the study indicated that estimation of serum cholesterol level and PBI could be used as reliable markers for detecting hypothyroidism in goats.

The haemogram values revealed microcytic hypochromic anaemia in hypothyroid goats. There was improvement in haemogram values on discontinuation of treatment with thiourea.

There was significant decrease in the quantity and quality of semen in hypothyroid goats. There was decrease in ejaculate volume, sperm concentration, motility, and

viability with an increase in the incidence of abnormal spermatozoa. There was significant improvement in the quality and quantity of the semen when the animals became euthyroid after withdrawal of thiourea. This established that the seminal changes were reversible.

Hydropericardium, left ventricular hypertrophy and dilatation of varying severity were observed in hypothyroid goats. Gelatinisation of subcutaneous fat was also a consistent feature at autopsy in hypothyroid animals. These lesions were not seen in animals sacrificed after discontinuation of administration of thiourea.

There was significant increase in the relative weight of the thyroid gland in hypothyroid goats. However, the thyroid enlargement was not palpable by gross examination. Histologically the increase in weight was associated with hyperplastic changes. Hyperplasia was characterised by formation of colloid depleted microfollicles. Lining cells were hypertrophic and concomitant degenerative changes were also observed. Absence of colloid formation was considered as a definite indication of blocking of thyroxine formation by thiourea. The effect of thiourea was dose and age dependent. Distended follicles containing pale thin colloid

and lined with cuboidal to low columnar epithelial cells were characteristic histological feature of the thyroid in animals after discontinuation of administration of thiourea. This indicated synthesis of thyroglobulin in the gland.

There was increase in the relative weight of the pituitary gland in all the animals dosed with thiourea. The predominant histological picture was hypertrophy and hyperplasia of basophils. Vacuolation and formation of cysts in focal areas were also evident. On discontinuation of thiourea there was reversal of histological lesions.

In all the animals dosed with thiourea adrenal glands showed significant increase in the relative weight. The pathological changes in the adrenal glands were similar to that seen in stress reaction. It was therefore concluded that goats dosed with thiourea were in a state of stress and on discontinuation of treatment with thiourea the stress response in the adrenal gland was not evident.

A significant decrease in the relative weight of the testis was observed in all the goats dosed with thiourea. Histologically the seminiferous tubules were small and were lined by a single layer of spermatogonial cells.

Spermatogenesis was completely absent. Some tubules, contained degenerative and desquamated cells without any evidence of sperms. These degenerative changes were transient and on discontinuation of administration of thiourea there was spermatogenesis and degenerative changes were not evident. Therefore it was concluded that the testicular changes were reversible.

There was decrease in the relative weight of the epididymis in all the goats dosed with thiourea. Histologically the tubules were small in size, the lining epithelium showed deciliation and desquamation. There was no evidence of sperms in some of the tubules. On withdrawal of thiourea treatment there was increase in the weight of the epididymis and the tubules appeared normal.

The accessory sexual glands(seminal vesicles and prostate) weighed less in hypothyroid goats. Histologically the cells lining the glands were small, inactive, vacuolated and hyalinised. The degenerative changes in the accessory sexual glands were considered as the causative factor for reduced quantity of semen. The reparative process was almost complete on discontinuation of administration of thiourea.

There was significant decrease in the relative weight of the ovaries in all the animals dosed with thiourea. The ovaries were smooth and inactive. Histologically the ovaries contained only a few scattered immature follicles and developing follicles. The follicles contained degenerated ovum. On discontinuation of treatment with thiourea there was increase in the weight of the ovary and histologically mature follicles and corpora lutea were evident.

The weight of the uterus was significantly low in all the goats dosed with thiourea. Histologically the muscular coat was relatively thin and glands were few and inactive. The lining epithelial cells did not show active mitosis. There was no evidence of secretory activity. On discontinuation of treatment with thiourea, the weight of the uterus increased and histologically the muscular coat and the endometrial glands appeared normal.

Significant pathological changes were observed in the reproductive organs of both males and females in hypothyroid state. It was also established that kids and female goats were more susceptible than adults and males

to the effects of hypothyroidism. The importance of hypothyroidism in inducing subfertility and infertility was clarified and the reversibility of the changes was proved.

CHAPTER VII

REFERENCES

REFERENCES

*Adams, A.E. (1946) Variations in the potency of thyrotropic hormone of the pituitary in animals. Quart. Rev. Biol. 21 : 1-32.

Adams, D.D. (1958) The presence of an abnormal thyroid stimulating hormone in the serum of some thyroid patients. J. clin. Endocr. Metab. 18 : 699-712.

*Adamson, J.W. and Finch, G.A. (1966) Mechanism of erythroid marrow activation. Trans. Ass. Am. Physns. 73 : 419.

Akiba, Y. and Matsumato, T. (1977) Effect of graded doses of a goitrogen in rapeseed, on synthesis and release of thyroid hormone in chicks. Japanese J. zootechnical Sci. 48 : 757-765.

Allcroft, R, Scarnell, J. and Bignett, S.C. (1954) A preliminary report on hypothyroidism in cattle and its possible relationship with reproductive disorders. Vet. Rec. 66 : 367-371.

Anderson, R.R. and Harness, J.M. (1975) Thyroid hormone secretion rates in growing and mature goats. J. Anim. Sci. 40 : 1131-1135.

*Andrews, F.N., Shrewsbury, C.L., Harper, C., Vestal, C.M. and Doyle, C.P. (1948) Iodine deficiency in new born sheep and swine. J. Anim. Sci. 7 : 298-310.

Annison, E.F. and Lewis, D. (1959) Thyroid metabolism in sheep during pregnancy. J. Agric. Sci. 52 : 79-86.

*Aronow, L., Angle, D.T. and Sperry, W.M. (1946) Some effects of administration of thiouracil to monkeys. Endocrynology. 38 : 331.

Armed Forces Institute of Pathology (1968) Manual of Histology Staining Methods. McGraw-Hill Book Company, New York. 3rd ed. pp. 12-184.

*Astood, E.B., Bissdi and Hughes, M. (1945) Further studies on the chemical nature of compounds which inhibits the function of the thyroid gland. Endocrinology. 37 : 456-461.

*Astwood, E.B., Greeder, M.A. and Bthlonger, M.C. (1949) The antithyroid factor of yellow turnip. Science. 109 : 631.

*Ayoub, I.A.L.L. (1968) Effect of radio iodine in thyroid function in goats. Zt. tier. arztle. wsch. 75 : 341-344.

*Baicoianuc., Zamfirescu, N. and Dumitresue, I. (1963) Increased semen production in rams by feeding iodinated oats. Zivotrovodstvo. 25 : 85-89.

*Barakat, M.Z., Osman, A.M., Salah, M.A. and Abdel-Aziz, F.I. (1971) A study on the thyroid iodine content of buffalo. Zentrablall vet. Med. 18A : 731-737.

*Barker, M.M., Lindbug, W.A. and ald, M.M. (1941) Further experience with thiocynate, clinical and experimental observation. J. Am. Med. Assoc. 117 : 1591-1594.

Barker, S.B. (1951) Mechanisms of action of thyroid hormone. Physiol. rev. 31 : 205.

Barker, S.B. (1971) "Chemistry and cellular effects" The Thyroid: A Fundamental and Clinical Text. Edited by Warner, S.C. and Ingbar, S.H. Harper and Row, New York. 3rd Ed. pp. 79-90.

*Baryseau, V.M. (1966) The correlation between functional activity of thyroid gland and reproductive character of stud bulls. Zhirot. vet. 467 : 106.

*Baumann, E. (1895-'96) Über das normale vorkommen Von Jod in Thiekorper. Z. Physiol. Chem. 21 : 319.

*Baumann, E.J. and Marine, D. (1945) Involution of adrenal cortex in rats fed thiouracil. Endocrinology. 36 : 400.

*Baumann, E. (1948) Goitre and Myxoedema of new born goats. Mien. tierarztl. Mschr. 55 : 585-592.

*Belonje, P.C. (1967) A report of some long term effect on thyroparathyroidectomy in a Merino wether. J. S. Afr. vet. Med. Ass. 38 : 225-226.

Berman, A.J. (1960) Peripheral effects of L-thyroxine on hair growth and colouration of cattle. J. endocr. 22 : 288-292.

*Blokhina, E.I. (1970) Proc. 1st. Int. Symp. Trace Elements metab. Anim. Edited by C.F. Mills. Livingstone, Edinburg. pp. 426.

Blom, E. (1950) A simple rapid staining method for the differentiation between live and dead sperm cells by means of eosin and nigrosin. Nord. vet. Med. 2 : 58.

Blood, D.C. and Henderson, J.A. (1968) Veterinary Medicine. The english language book society and Bailliere, Tindall and Cassell, London. 3rd Ed. pp. 711.

Brooks, J.R. and Rose, C.V. (1962) Effect of ambient temperature and thyroxine therapy on semen quality of rams. J. Anim. Sci. 21 : 700-705.

Brooks, J.R., Rose, C.V. and Turner, C.W. (1964) Effect of thyroidectomy on reproductive performance of ewes and semen quality of rams. J. Anim. Sci. 23 : 54-58.

Brownstead, G.H. and Fowler, S.H. (1959) Thyroid status and embryonic mortality in swine. Am. J. Physiol. 196 : 287-290.

*Buhler, C.W., Alax, D.S., Peterson, G.E., Wright, J.W., Glenday, A.C. and Johnson, J.M. (1957) Goitrogenic effect of white clover. N. Z. J. Sci. Tech. Sect. A. 38 : 793-802.

Burstein, P.J., Braznin, E., Johnson, J. and Schalch, D.S. (1969) The effect of hypothyroidism on growth, serum growth hormone, the growth hormone-dependent somatomedin, insulin like growth factor and its carrier protein in rats. Endocrinology. 104 : 1107-1111.

Bush, M.M. (1969) Thyroid diseases in the dog. A Review. J. Small Anim. Pract. 10 : 95-109.

Bustad, L.K., Thorne, S., George, C.A. and Seigneur, L.J. (1957) Thyroid adenomas in sheep administration of I^{131} daily. Nature, Lond. 179 : 677.

Calderbank, G. (1958) Thyroid dysfunction and infertility. Vet. Rec. 70 : 731.

Calderbank, G. (1963) "Iodine", Animal Health, Production and Pasture. Longmen's Green Co., London. pp. 250.

Catt, K.J. (1970) ABC of Endocrinology. II. The Thyroid gland. Lancet. 1 : 1383-1389.

*Chensey, A.M., Clawson, T.K. and Webster, H. (1928) Endemic goitre in rabbits. Incidence and characteristic. Bull. John. Hopkin. Hospital. 43 : 261-277.

*Chu, T.P. and Lou, S.S. (1944) The role of thyroid gland and oestrogen in regeneration of gonadotrophic activity of anterior pituitary. J. Endocrinol. 46: 115.

*Clausen, H.J. (1954) Experimental production of struma fibrosa. Arch. Path. 58 : 222-226.

Cohrs, P. (1966) Text book of the special Pathological anatomy of the domestic animals. Pergamon Press. Oxford. pp. 919-922.

Cone, J.M., Umezu, M. and Timiras, P.S. (1975) Developmental and patterns of pituitary and plasma TSH in the normal and hypothyroid female rats. Endocrinology. 97 : 237-240.

Contopoulos, A.N., Simpson, M.E. and Koneff, A.A. (1958) Pituitary function in thyroidectomised rat. Endocrinology. 63 : 643-653.

*Cooper, A.F. (1836) Notes on the structure of the 'thyroid gland'. Guy. hosp. Rep. 1 : 448.

Crispell, K.R. and Wilson, D.C. (1964) "Pathology of myxoedema". The Thyroid. Edited by Hazard, J.B. and Smith D.E. Williams and Wilkins Company, Baltimore. 236-238.

*D'Angelo, S.A., Paschkis, K.E., Cantarow, A. Siegal, A.M. and Rivero Fontan, J.C. (1951) Hyperplasia and I131 uptake of guinea pig thyroid during and after chronic prophylthiouracil treatment. Endocrinology. 49 : 624-636.

Danowski, T.S. (1962) Clinical endocrinology. Thyroid. Williams and Wilkins, Baltimore, Vol. II. pp. 223.

Davidson, B., Soodak, M., Strout, H.V., Neary, J.T. Nakamur, C. and Maloof, F. (1979) Thiourea and cyanamide as inhibitors of thyroid peroxidase. The role of iodide. Endocrinology. 104 : 917-924.

Delesi, C.A., Astier, H.S. and Burke, M.I. (1979) Kinetics of thyrotrops and somatotrops during development of hypothyroidism and d-triiodo thyronine treatment of hypothyroid rats. Endocrinology. 104 : 1172-1180.

Del Rio, A.G., Valdez Toledo, C.S. and Quiros, L.C. (1979) Thyroid gland and epididymal function in rats - Histological study. Arch. Androl. 3 : 19-22.

*De Robertis, L. (1941) Proteolytic enzyme activity of colloid extracted from single follicles of the rat thyroid. Anat. Rec. 80 : 219-230.

Dhoble, A.L. and Gupta, S.K. (1980) Protein bound iodine in blood plasma of buffaloes in relation to post-partum anoestrum. Indian J. Dairy Sci. 33 : 124-125.

Disbrey, B.D. and Rack, J.H. (1970) Histological Laboratory Methods. E & S Livingstone, Edinburgh. pp. 99-101.

*Dokumow, S.J., Miriams, S., Stanczeva and Spasova, S.A. (1971) Protein bound iodine and normal menstrual cycle. Endokrinol. pol. 22 : 113-122.

*Durlach, J., Bouvet, P. and Levilliain, A. (1954) Influence of propylthiouracil on the adrenaline content of guinea-pig adrenals. Annls. Endocr. 15 : 164-171.

Dutt, B. and Kehar, N.D. (1959) Incidence of goitre in goats and sheep in India. Brit. vet. J. 115 : 176-179.

Dutt, B. and Vasudeva, B. (1963) A clinical case of hypothyroidism in a ram. Indian vet. J. 40 : 505-506.

*Edelhock, H. (1960) The properties of thyroglobulin: I. The effects of alkali. J. biol. Chem. 235 : 1326-1334.

*Edelhock, H. and Hall, J.W. (1964) "The proteins and enzymes of thyroid gland". The Thyroid Gland. Edited by Pitt. Rivers, R. and Trotter, W.R. Butterworth., London pp. 113-130.

Elwishy, A.B., Ablou, M.S.S., Ablou, M.S. and El-Sawaf, S.A. (1973) Hormonal variations in the blood of buffaloes and cows during post partum period. Gonadotrophic, thyrotrophic, and adreno-corticotrophic hormones. J. Anim. Morph. Physiol. 20 : 49-54.

Falconer, I.R. (1966) Studies on congenitally goitrous sheep. The iodinated compound of serum and circulatory TSH. Biochem. J. 100 : 190-196.

FAO/IDA. (1968) A guide to Autopsy Techniques. Stockholm. Vol. IV. pp. 1-10.

Faulkner, L.W., Levy, A.P., and Leonards, J.R. (1961) Simplified technique for the determination of serum protein bound iodine. Clin. Chem. 7 : 637-645.

*Ferguson, K.A., Schinckel, P.G., Carter, H.B. and Clarke, W.J. (1956) The influence of thyroid and wool follicle development in the lamb. Aust. J. biol. Sci. 9 : 575-585.

Fletcher, K. and Myant, N.B. (1958) Influence of thyroid on the synthesis of cholesterol by liver and skin in vitro. J. Physiol. 144 : 361-372.

Follis, R.H., Jr. (1959) Experimental colloid goitre produced by thiouracil. Nature, Lond. 183 : 1817-1818.

Freedberg, I.M. (1971) 'Skin', The Thyroid: A fundamental and clinical text. Edited by Werner, S.E. and Ingbar, S.H., Harper and Row, New York. 3rd Ed. pp. 722.

Galton, V.A. (1968) Thyroxine metabolism and thyroid function in pregnant rat. Endocrinology. 82 : 282.

George, J.M., Marleigh, C.A. and Harris, A.L.A. (1966) Occurrence of goitre in summer born lambs of different breeds. Aust. vet. J. 42 : 1-4.

*Gley, E. (1923) The physiology of Adrenals. Rev. med. 40 : 193-221.

*Goldberg, R.C. and Chaikoff, I.L. (1950) The cytological changes that occur in anterior pituitary glands in rats injected with various doses of I^{131} and their significance in the estimation of thyroid function. Endocrinology. 46 : 91-104.

*Goldberg, R.C. and Chaikoff, I.L. (1951) On the nature of hypertrophied pituitary gland induced in mouse by I^{131} injections and mechanism of its development. Endocrinology. 48 : 1-5.

Goldberg, R.C., Wolf, J. and Greep, R.O. (1957) Studies on the nature of thyroid pituitary on the relationship. Endocrinology. 60 : 38-52.

*Goldschmidt, V.M. (1954). Geochemistry. Calaenon, Oxford. pp. 602-620.

Gorbman, A. and Bern, H.A. (1974). A Text Book of Comparative Endocrinology. Wiley Eastern private Limited., New Delhi. pp. 159.

*Gottschalk, A. and Ada, G.L. (1954). The separation and quantitative determination of component sugars of mucoproteins. Biochem. J. 62 : 681-685.

Green, C.R., Ham, K.N. and Tange, J.D. (1974) Glomerular lesions in experimental hypothyroidism. Arch. Path. 24 : 156-160.

*Greer, M.A. (1950) Nutrition and goitre. Physiol. Rev. 30 : 513-548.

*Greer, M.A. and Whallon, J. (1961) Antithyroid effects of barbarian (phenylthiooxazolidone) a naturally occurring compound from barbares. Proc. Soc. Expt. Biol. Med. 107 : 802-804.

*Greer, M.A. (1962) The natural occurrence of goitrogenic agents. Recent. Progr. Hormone. Res. 18 : 167-212.

*Griesbach, W.E., Kennedy, J.C. and Purves, H.D. (1941) Studies on the experimental goitre II. The effect of goitrogenic diet on hypophysectomised rat. Brit. J. Exp. Path. 22 : 249-254.

*Gross, J and Pitt-Rivers, R. (1952) The identification of 3:5:3-L-Triiodothyroxine in human plasma. Lancet. 1 : 439-441.

*Growth, A. (1962) Pathology of goitre and thyroid tumours in domestic animals. Dtsch. tierarztl. wschr. 69 : 707-713.

* Holik J. and Zavadsky T. (1978) Mass outbreak of goitre in breeding rams Vet. Lnarstvi 28 68-69

*Hall, W.P.K. (1952) Effect of methylthiouracil on liver, Vitamin A, Spermatogenesis and calcium metabolism of Merino rams. Qd. J. Agric. Sci. 9 : 301-306.

Hancock, J.L. (1957) The morphology of Boar spermatozoa. J. Roy. Micro. Soc. 76 : 84.

*Harington, C.R. and Barger, G. (1927) Chemistry of thyroxine :II. Constitution and synthesis of thyroxine. Biochem. J. 21 : 169-183.

*Harkness, M.L.R., Harkness, R.D. and Bantler, J.E. (1954) Changes in collagen content of thyroid in rats treated with thiouracil. J. Physiol. 125 : 51-55.

Hasnan, A. and Akram, A.L. (1980) Plasma protein bound iodine and lactational performance of cross-bred cows during pregnancy. Indian J. Dairy Sci. 33 : 304-309.

Hignett, P.G. (1952) Animal health production and posture. Edited by Calderbank, G. Longmann Green and Co. London. pp. 250.

*Mojer, J.A. (1931) Goitre studies. The prevalance of endemic goitre in Sweden. Svenska. Lak. Sallsk. Forhandl. 57 : 1-104.

Lorne, R.L. and Thibult, J.P. (1961) Protein bound iodine and reflex speed measurements in infertile patients. Int. J. Fertil. 6 : 385-389.

*Munt, J.D. (1944) Mitotic activity in the thyroid gland of female rats. Anat. Rec. 90 : 133-138.

Nichiona, N.A. (1964) Direct Biuret determination of total protein in tissue homogenates. J. Lab. Clin. Med. 63 : 319-324.

*James, V.H. (1967) A simple silver method for the determination of reticulin fibers. J. med. lab. Tech.: 24 : 49.

*Jones, R.P. (1946) Studies on the effect of thiourea and allied substance on thyroid gland and other organs in rats and mice. J. Path. Bact. 58 : 483-493.

*Jones, G.E.J., Delfa, E. and Foote, W.E. (1946) The effect of thiouracil hypothyroidism on reproduction in rat. Endocrinology. 38 : 337-344.

*Jovanovic, M., Puntic, V. and Markovic, D. (1953) Goitre in domestic animals in Serbia. Acta vet. Belgrade. 3 : 31-51.

*Jovanovic, M. (1955) Histology of thyroid in endemic goitre in domestic animals. Acta vet. Belgrade 5 : 13-52.

Jubb, K.V.F. and Kennedy, P.C. (1970) Pathology of Domestic Animals. Academic Press, New York. 2nd Ed. Vol. I. pp. 407-415.

Kaneko, J.J. (1970) "Thyroid function" Clinical Biochemistry of Domestic Animals. Edited by Kaneko, J.J.

and Cornelius, C.E. Academic Press, New York. 2nd Ed. Vol. I. pp. 27-233.

Kapoor, P.C., Sharma, B.P. and Mangia, P.C. (1974) Studies on correlation between protein bound iodine and different characteristics of buffalo semen and amongst its different characters. Indian J. Anim. Sci. 44 : 223-228.

*Kelly, F.C. and Snedden, J.... (1960) "Prevalence and geographical distributio. of endemic goitre", Endemic goitre. World health Organisation monograph series No. 44., Geneva. pp. 27-233.

Kelly, S.T., Colme, P.L. and Brandt, G.W. (1974) Measurement of thyroid gland function during oestrus cycle of nine mares. Vet. Med. Small. Anim. Clin. 69 : 1531-1533.

*Kendall, E.C. (1915) The isolation in Crystalline form of the compound containing iodine which occurs in the thyroid. Its chemical nature and physical activity. Trans. Ass. Am. Physns. 30 : 420-449.

*Kennedy, T.H. and Purves, H.D. (1941) Experimental goitre. I The effects of brassica seed diets on rat. Brit. J. exp. Path. 22 : 241-244.

*Konneay, T.... (1942) Thiourea as goitrogenic substance. Nature, Lond. 150 : 233-234.

*Kimbal, O.P. (1937) Prevention of goitre in Michigan and Ohio. J. Am. med. Ass. 108 : 860-864.

Kimberg, D.V. (1971) "Gastrointestinal tract" The Thyroid: A fundamental and clinical text. Edited by Werner, S.C. and Ingbar, S.H. Harper and Row, New York. 3rd Ed. pp. 736.

*King, F.H. (1836) Observation on the thyroid gland. Guy. Hosp. Rep. 1 : 423-447.

*Kocher, T. (1883) Ueber Kropfexstirpation und ihre Folgen. Arch. Klin. Chir. 29 : 254.

Koutras, D.A. (1971) " Non toxic goitre" The thyroid; A fundamental and clinical text. Edited by Werner, S.C. and Ingbar, S.H. Harper and Row, New York. 3rd ed. pp. 409.

*Kovalskii, V.V., Struck, M.I. and Ioon, A.I. (1970) Role of iodine in eradicated cow infertility in area of iodine deficiency in the Amur region. Dokl. Vses. Akad. sel. Khoz. Nauk. VI. Lenina. pp. 24-25.

Krikland, J.L. Gardna, W.M., Nanku, V.K. Akhter, M. and Stancel, G.M. (1981). Hormonal control of uterine growth. The effect of hypothyroidism on estrogen stimulated cell division. Endocrinology. 108 : 2346-2351.

*Krohn, P.L. (1947) The effect of propylthiouracil on estrus cycle in mice. J. Endocr. 5 : 53.

Krohn, P.L. and Waible, M.C. (1950) The effect of hypothyroidism on reproduction in female albino rat. J. Endocr. 6 : 375-385.

Kumaran, J.D.S. and Turner, C.W. (1949) The endocrinology of spermatogenesis in birds. Effect of hypo and hyper thyroidism. Poultri. Sci. 28 : 653-665

Lall, M.K. (1952). A case of congenital goitre in kids. Indian vet. J. 29 : 133-135.

Lamberg, B.A. and Grasseck, R. (1955) The serum protein pattern in disorders of thyroid function. Acta Endocr. 19 : 91-100.

Lascelles, A.K. and Setchell, B.P. (1959) Hypothyroidism in sheep. Aust. J. Biol. Sci. 12 : 455-464.

*Lazo-wasem, E.A. (1960) Pituitary ACTH level during adrenal involution following thiouracil administration. Proc. Soc. exp. Biol. Med. 103 : 300-302.

Lennon, H.D.Jr. and Mixner, J.P. (1958) Relationships between plasma protein bound iodine and certain measures of reproductive and lactional performance in dairy cattle. J. Dairy Sci. 42 : 327-332.

*Levine, I., Remington, R.E. and vonVolwitz, R. (1933) Studies on the relation of diet to goitre. I. A dietary technic for the study of goitre in the rat. J. Nutr. 6 : 325-345.

Lewis, R.C. and Malaton, R.P. (1953) Protein bound iodine levels in dairy cattle plasma. J. Dairy Sci. 36 : 33.

Lewis, R.C. (1956) The effect of thyroidectomy and iodine supplementation on the plasma PBI of a Jersey bull. J. Dairy Sci. 39 : 610-611

*Lippincott, G.W., Lewallen, C.G. and Sneelaberger, C.J. (1957) Pathology of radio isotopic ablation of the thyroid in the dog. Arch. Path. 63 : 540-556.

Lombardi, M.A., Collar, C.L. and Kirk, R.W. (1962) Diagnosis of thyroid gland functions in dog. Am. J. vet. Res. 23 : 412-420.

Louvet, J.P., Govarre, M., Solandin, A.M. and Soulard, C. (1979) Hypothyroidism and amovulation (Letter). Lancet. 8124 : 1032.

*Love, J.G. (1942) Parenchymatous goitre in new born goats and kids. J. Am. vet. med. Ass. 101 : 484-481.

Mackenzie, C.G. and Mackenzie, J.B. (1941) Effects of sulphanamides and thiourecas in the thyroid gland and basal metabolism. Endocrinology. 32 : 185-209.

*Maltiev, S.M. (1966) The effect of potassium iodide and iodo-microfertiliser on reproduction in sheep in iodine deficient zones. Dokl. Vser. Akad. sel. khoz. Nauk. VI. 1 : 38-41.

Malkastan, G.D. and Mayberry, W.E. (1969) Serum total and free thyroxine and thyroprotein in normal pregnant women, neonates and women receiving progesterone. Am. J. Obstet. Gynaec. 1061 : 1234.

Maqsood, M. (1951) Influence of thyroid status on spermatogenesis. Science. 114 : 693-694.

Maqsood, M. (1952) Thyroid function in relation to reproduction of mammals and birds. Biol. Rev. 27 : 281-319.

*Marine, D. and Lenhart, C.H. (1910) On the occurrence of goitre (active thyroid hyperplasia) in fish. Bull. John Hopkins. Hosp. 21 : 95-98.

*Marks, S. Bocken, H.C. and Eastad, C.V. (1957) Histopatology of thyroid gland of sheep in prolonged administration of I^{131} Amer. J. Path. 33 : 219-249.

*Marston, H.R. and Peirce, A.W. (1952) The effect of following thyroidectomy in a Merino sheep. Aust. J. Exp. Biol. med. Sci. 10 : 203-213.

Mason, K. and Wilkinson, J.S. (1973) Thyroid gland - A review. Aust. vet. J. 49 : 44-49.

Mayberry, W.E. and Astwood, E.B. (1961) Advances in Thyroid Research. Pergamon Press, New York. pp. 42.

McCarrison, R., Newcomb, C., Viswanath, B., and Norris, R.V. (1927) The relation of endemic goitre to the iodine content of soil and drinking water. Indian J. Med. Res. 15 : 207-246.

*McCarrison, R. (1933) Recent researches on the aetiology of goitre. Report of second international conference Berne. Verlag. Hans. Huber. Berne. pp. 354-403.

McCarthy, J.C., Corley, A.C. and Zarrow, M.X. (1969) Effect of goitrogen and adrenal gland of rat. Am. J. Physiol. 197 : 693-698.

McCartney, M.G. and Shaffner, C.S. (1950) The influence of altered metabolism upon fertility and hatchability in the female fowl. Poult. Sci. 29 : 67-77.

McIntosh, G.H., Baghurst, K.I., Potter, B.J. and Hetzel, B.S. (1979) Foetal thyroidectomy and brain development in the sheep. Neuropath. Appl. Neuro. biol. 5 : 363-376.

*McIntosh, R.A. (1943) Goitre of new born. Can. J. comp. Med. 7 :263-265.

*McKenzie, F.F. and Berlier, V. (1937) The reproductive capacity of rams. Missouri Agr. Expt. Sta. Res. Bull. pp. 265.

McKenzie, J.M. (1958) Delayed thyroid response to serum from thyrotoxic patients. Endocrinology. 62 : 865-868.

Metzger, B.E. and Freinkel, N. (1971) "Metabolic changes" The Thyroid. A fundamental and clinical text. Edited by Werner, S.C. and Ingbar, S.H. Harper and Row, New York. 3rd. Ed. pp. 746.

*Miale, J.B. (1967) Laboratory Medicine Haematology. The C.V. Morby Company, Scient Louis. 3rd Ed. pp. 1143-1144.

*Moberg, R. (1959) Possible influence of iodine-deficiency in reproductive performances in cattle with special reference to retained placenta. Proc. III World Cong. on Fertand Steril. Amsterdam.

Mukherjee, D.P., Roy, A. and Bhattacharya, P. (1953) The effect of feeding thyroprotein on semen quality and on some physiological conditions of goats. Effect on reaction time and semen characters. Indian J. vet. Sci. 23 : 1-7.

*Murray, M.M., Mule J.A., Simpson, B.W. and Wilson, D. (1948) Thyroid enlargement and other changes related to the mineral content of drinking water (with a note on goitre prophylaxis). Medical Research Council Memorandum. No. 18, London.

Nangia, O.P., Dixit, U.P. and Agarwal, V.K. (1975) Studies on some blood constituents in chicken with modified thyroid activity. Haryana. Agric. Univ. J. Res. 5 : 254-259.

Nangia, O.P. and Gulati, D.P. (1976) Influence of thyroid status on growth of chickens. Indian vet. J. 53 : 185-189.

Pierce, E. (1851) The Thyroid. Edited by Beach Hazard, J. and Smith, D.E. The Williams & Wilkins Company, Baltimore. pp. 76.

Otomatus, T. (1954) Studies on goitre of farm animals in Japan, Studies on goitre in sheep. Jap. J. vet. Sci. 16 : 53-64.

Pearson, A.M., Heinke, E.P., Hopper, J.A. and Morrow, H.E. (1966) Effect of environmental temperature and thiouracil feeding upon growing fattening pigs. J. Anim. Sci. 25 : 894-899.

Peczely, P., Astier, E. and Jallageas, M. (1979) Reciprocal interactions between testis and thyroid in male Japanese quail. Gen. Comp. Endocr. 37 : 400-403.

*Peters, J.P. and Man, E.B. (1950) The significance of serum cholesterol in thyroid disease. J. Clin. Invest. 29 : 1-7.

*Potter, G.D., Lindsay, S. and Chaikoff, I.L. (1960) Introduction of neoplasms in rat by low doses of radioiodine. Archs. Path. 69 : 257-269.

Prasad, O. and Singh A. (1971) Thyroid gland relationship in desi chicken under hypo and hyper thyroid conditions. Indian J. Anim. Sci. 41 : 867-872.

Prasad, O. and Singh, A. (1979) Effect of altered thyroid status in PBI, Blood glucose, Plasma cholesterol, liver glycogen and oxygen consumption by liver and gonads of poultry. Indian J. Anim. Sci. 49 : 446-450.

*Purves, H.D. (1964) Thyroid gland. Edited by Pitt-Rivers and Trotter, W.R. Butterworth., London. pp. 1-38.

Rao, K., Hill, G.N., Iain, R.W. and Mulhearn, C.J. (1968) Congenital goitre in Marino sheep due to an inherited defect in the synthesis of thyroid hormone. Res. vet. Sci. 9 : 209-233.

Ragowitsch, N. (1989) The Thyroid. Edited by Beach Hazard, J. and Smith D.E. The Williams and Wilkins Company. Baltimore. pp. 76.

Rajkumar, S.S. (1970) A case of congenital goitre in kids. Indian vet. J. 29 : 133-135.

*Rall, J.E., Robbins, J. and Lewallen, C.G. (1964) The Thyroid. The hormones. Edited by Thimann, K.V. and Astwood, E.B. Academic Press., New York. Vol. V. pp. 159.

Ramalingaswami, V., Subramanian, T.A.V. and Rao, M.G. (1961) The aetiology of Himalayan endemic goitre. Lancet. 1 : 791-794.

* Reineke, E.P., Bergman, A.J. and Turner, C.W. (1941) Effect of thyroidectomy on young male goats upon certain anterior pituitary hormones. Endocrinology. 29 : 306-312.

*Riggs, D.S. (1952) Quantitative aspects of iodine metabolism in man. Pharmacol. Rev. 4 : 284-370

Rivlin, R.S. (1971) 'Vitamins', The Thyroid. A fundamental and clinical text. Edited by Warner, S.C. and Ingbar, S.H. Harper and Row, New York. 3rd Ed. pp. 751.

Roberts, S.J. (1971) Veterinary Obstetrics and Genital Diseases. CBS Publishers and Distributors, New Delhi. Ed. 2. pp. 717.

Robertson, H.A. and Belconer, I.R. (1961) Reproduction and thyroid activity. J. Endocr. 22 : 133-142.

*Rocne, J. and Mussitzky, S. (1960) "Etiology of Endemic Goitre" Endemic Goitre. World Health Organisation monograph series No. 44. Geneva. pp. 351-368.

Rollinson, W.H.L. (1951) Studies on abnormal spermatozoa of bull semen. Brit. vet. J. 107 : 203, 251, 258.

Roy, S., Deo, M.G. and Ramalingaswami (1964) Pathological features of Himalayan endemic goitre. Am. J. Path. 44 : 839-851.

*Russell, J.A. (1943) "The relationship of the anterior pituitary to the thyroid and adrenal cortex in control of carbohydrate metabolism". Essays in Biology. Edited by Farquhar, H.T., Leake, C.D., Lyons, T.R. and Simpson, M.E. University of California Press, Berkely pp. 507-527.

Russel, A.J.F. (1967) A note on goitre in lambs grazing rape (Brassica napus). Anim. Prod. 9 : 131-133.

Schalm, O.W. (1965) Veterinary Haematology. Lea and Febiger., Philadelphia. 2nd Ed. pp. 62, 80, 95-100.

*Schultze, A.B. and Davis, H.P. (1946) The influence of feeding synthetic thyroprotein on fertility of bulls. J. Dairy Sci. 29 : 534-535.

Scott, P.P., Greaves, J.P. and Scott, M.C. (1961) Nutrition of cat. 4. Calcium and iodine deficiency on a meat diet. Br. J. Nutr. 15 : 35-51.

Borimshaw, N.S. (1964) "Geographical pathology of thyroid disease" The Thyroid. Edited by Hazard, J.B. and Smith, D.E. William and Wilkins Company, Baltimore. pp. 107-111.

*Sellers, E.A. and Ferguson, J.K.W. (1949) Exoptnalmus in rats after prolonged administration of propyl-thiouracil. Endocrinology. 45 : 345-346.

Setchell, B.P., Dickinson, D.A., Lascelles, A.K. and Donner, M.B. (1960) Neonatal mortality in lambs associated with goitre. Aust. vet. J. 36 : 159-164.

Sharma, D.N. and Singh, Y. (1975) Histological observations on thyroid gland relationship in male cross-bred (New Hampshire X White Cornish) chicken. Indian J. Anim. Health. 14 : 147-149.

Sharma, D.P. and Sharma, A. (1976) Protein bound iodine levels during oestrus, pregnancy and non pregnant states in goats. Indian J. Physiol. Pharmac. 20 : 242-244.

*Sharpless, G.R., Pearson, J. and Prato, G.S. (1939) Production of goitre in rats with raw and treated soyabean flour. J. Nutr. 17 : 545-555.

Sharpless, G.R. and Metzger, M. (1941) Arsenic and goitre. J. Nutr. 22 : 119-127.

*Silberg, M. and Silberg, R. (1940) The effect of thyroid-ectomy and anterior pituitary extract of cattle. Am. J. Path. 16 : 505-524.

*Sinclair, O.P. and Andrews, E.D. (1958) Prevention of goitre in new born lambs from kale-fed ewes. N. Z. vet. J. 6 : 87-95.

Singh, B.P. and Norton, D.C. (1956) Blood PBI determination as a measure of thyroid function in normal pregnancy and threatend abortion. Am. J. Obstet. Gynaec. 72 : 607.

Singh, A., Meinke, E.P. and Hinger, A.A. (1968) Influence of thyroid status of the chick on growth and metabolism with observation on several parameters of thyroid function. Poult. Sci. 47 : 205-211.

*Smelser, G.K. (1944) Differential concentration of hormones in the central peripheral zones of the bovine anterior pituitary gland. Endocrinology. 34 : 39-43.

*Smith, P.E. and Smith, I.P. (1922) The repair and activation of the thyroid in hypophysectomised tadpole by parenteral administration of fresh anterior lobe of the bovine hypophysis. J. Med. Res. 43 : 267-283.

Smith, H.A., Jones, T.C. and Hunt, R.D. (1972) Veterinary Pathology. Lea and Febiger., Philadelphia. 4th Ed. pp. 1357-1363.

Soliman, F.A. and Meineke, E.P. (1950) Changes in uptake of radio-active iodine by the thyroid of the rat during the oestrous cycle. Am. J. Physiol. 178 : 89-90.

Soliman, F.A. and Meineke, E.P. (1952) Influence in variation in environmental temperature and thyroid status on sexual function of young female mice. Am. J. Physiol. 168 : 405-409.

Soliman, F.A. and Madawi, A.M. (1956) Level of thyroid and thyrotropic hormones in the blood of rats at various stages of the oestrus cycle. Nature, Lond. 177 : 235.

Soliman, F.A., Nasr, H. and Zaki, K. (1963) Levels of thyroid and thyrotrophic hormones in the blood of Friesian cows at various reproductive stages. J. Reprod. Fertil. 6 : 335-340.

Soliman, F.A., Zaki, K., Soliman, M.K. and Abdo, M.S. (1964) Thyroid function of Friesian cows during oestrus cycle and in conditions of ovarian abnormalities. Nature, Lond. 204 : 693.

*Soliman, F.A., Afiefy, M.M., Zaki, K. Abdle, W., Ayoub, L.A. and Fahmy, M.F. (1973) Variations in thyroid status of buffalo during oestrus cycle, pregnancy and in conditions of ovarian abnormalities. Zantial. vet. Med. 20 A. 324-331.

Solomon, J. and Greep, R.D. (1959) The effect of alteration in thyroid function on pituitary growth hormone content and acidophil cytology. Endocrinology. 65 : 158-164.

*Southcott, W.H. (1945) Congenital goitre in lamb in Tasmania. Aust. vet. J. 21 : 35-36.

*Spielman, A.A., Peterson, W.E., Litch, J.B. and Pomeroy, B.S. (1946) General appearance, growth and reproduction of thyroidectomised bovine. J. Dairy Sci. 29 : 329.

*Spisni, D. and Garavaglia, G. (1954) Histology of thyroid gland of cattle fed on cauliflower. Vet. Ital. 5 : 883-895.

Sreekumaran, T. (1976) Pathology of experimental hypothyroidism in goats. M.V.Sc. Thesis. Kerala Agricultural University

Srinivas, V. (1979) Studies on protein bound iodine in the blood serum of the buffaloes in certain physiological and pathological conditions of reproduction. M.V.Sc. Thesis. University of Agricultural Science, Bangalore.

*Stanbury, J.B., Brownell, G.L., Riggs, D.S., Perinetti, H., Itoiz, J. and del Castillo, E.B. (1954) Endemic goitre. The adaptation of Man to iodine deficiency. Cambridge, Mass. Harward University Press.

Stoffer, R.P., Koeneke, L.A., Cherry, V.E. and Hellwing, C.A. (1957) The thyroid in pregnancy. Am. J. Obstet. Gynaec. 74 : 304-308.

*Stott, H., Bhatia, B.B., Lal, R.S. and Rai, K.C. (1930-31) The distribution and cause of endemic goitre in united provience. Indian J. Med. Res. 18 : 1059-1086.

*Suzuki, H., Higuchi, J., Sawa, K., Ohtakis, S. and Horiuchi, Y. (1965) Endemic coast goitre in Hokkaido, Japan. Acta Endocr. 50 : 161-176.

Swanson, E.W. and Boatman, J.F. (1953) The effect of thiouracil feeding upon the seminal characteristics of dairy bulls. J. Dairy Sci. 36 : 246-252.

Symington, T. (1969) Functional Pathology of Human Adrenal Gland. E & S Livingstone Limited., Edinburgh. pp. 70.

Symonds, H.W. (1969) The effect of thyroidectomy and parathyroidectomy on accertion of phosphorus into the skeleton of young goats. Res. vet. Sci. 10 : 218-224.

Symonds, H.W. (1970) The effect of thyroidectomy and thyroparathyroidectomy upon phosphorus homeostasis in goat. Res. vet. Sci. 11 : 260-269.

Tong, W. (1971) " Thyroid hormone synthesis and release" The Thyroid: A fundamental and clinical text. Edited by erner, S.C. and Ingbar, S.H. Harper and ow., New York. 3rd Ed. pp. 24-40.

*Turner, C.W. and Cuops, P.L. (1940) Effect of certain experimental conditions on the thyrotrophic hormone

content of albino rat. Endocrinology. 26 : 1042-1047.

Underwood, E.J. (1971) Trace Elements in Human and Animal Nutrition. Academic Press., New York. 3rd. ed. pp. 300.

Vadodaria, V.P., Jamakiraman, K. and Buch, N.C. (1977) Thyroid activity in relation to reproductive performance of Surti buffalo heifers- Bubalus bubalis. I. Serum PBI. All India symposium on comparative physiology. Nov. 16-18.

Vadodaria, V.P., Jamakiraman, K. and Buch, N.C. (1980) Studies on thyroid follicles in relation to phases of oestrus cycle and breeding periods. Indian vet. J. 57 : 985-990.

Van Dyl, A. (1957) Serum PBI and serum lipid changes in the baboon during pregnancy and lactation. J. Endocr. 14 : 317-324.

*Vzaimosuyaz, U.S.D.I.S. (1973) Relation of protein bound iodine to reproduction in cows. Nutr. Abstr. Rev. 43 : 917, 7593.

Wallach, J.D. (1965) Goitrogenic hypothyroidism in feeder lambs. Vet. Med. small Anim. Clinician. 60 : 1051-1053.

Walton, J.A. and Humphrey, J.D. (1979) Endemic goitre of sheep in high lands of papua New Guinea. Aust. vet. J. 55 : 43-44.

Watson, D.A., Roadbent, G.D. and Kilpatrick, A. (1962) Congenital goitre in Dorset horn lambs. Vet. Rec. 74 : 506-509.

Werner, S.C. (1971) " Male reproductive system" The Thyroid: A fundamental and clinical text. Edited by Werner, S.E. and Ingbar, S.H. Harpor

and Row., New York, 3rd. Ed. pp. 785.

*Wharton, T. (1956) The Thyroid: A fundamental and clinical text. Edited by Werner, S.E. and Ingbar, S.H., Harper and Row, New York. 3rd Ed. pp. 5

Wills, P.I. and Schindler, W.J. (1970) Radio thyroxine turnover studies in mice. Effect of temperature diet, sex and pregnancy. Endocrinology. 86 : 1272-1280.

*Wilson, D.C. (1941) Flourine in the aetiology of endemic goitre. Lancet. 1 : 211-212.

*Wilson, B., Raghupathy, E., Tonoue, T., and Tong, W. (1968) TSH like action of dibutyryl-CAMP on isolated bovine thyroid cells. Endocrinology. 82 : 877.

Wilson, J. (1975) Hypothyroidism in ruminants with special reference to foetal goitre. Vet. Rec. 97 : 161-164.

*Wintrobe, M.M. (1961) Clinical Haematology. Lea and Febiger, Philadelphia. 5th Ed. pp. 381.

Wollman, S.H. and Wodinsky, I.C. (1955) Localisation of protein bound ^{131}I in the thyroid gland of mouse. Endocrinology. 56 : 9-20.

Wong, C.C., Dohler, K.D., Muhlen, A.V. (1980) Effects of Tri-iodothyroxine, thyroxine and isopropyl-Di-Iodothyronine on the thyroid-stimulating hormone in serum and pituitary gland, and on pituitary concentrations of prolactin, growth hormone, luteinizing hormone and follicle stimulating hormone in hypothyroid rats. J. Endocr. 87 : 255-263.

Yatvin, M.B., Wannemacher, J.R. and Banks, W.L. Jr. (1964) Effects of thiouracil and thyroidectomy on liver protein metabolism. Endocrinology. 74 : 874-884.

Zak, B. (1957) A simple rapid micro technique for serum total cholesterol. Am. J. Clin. Path. 27 : 583-588.

*Zarrow, M.X. and Money, W.L. (1949) Involution of the adrenal cortex of rats treated with thiouracil. Endocrinology. 44 : 345-357.

*Zeckwer, I.T., Davison, L.W. Keller, T.B. and Livingood, C.S. (1935) The pituitary in experimental cretinism. I. Structural changes in pituitaries of the thyroidectomised rats. Am. J. med. Sci. 190 : 145-147.

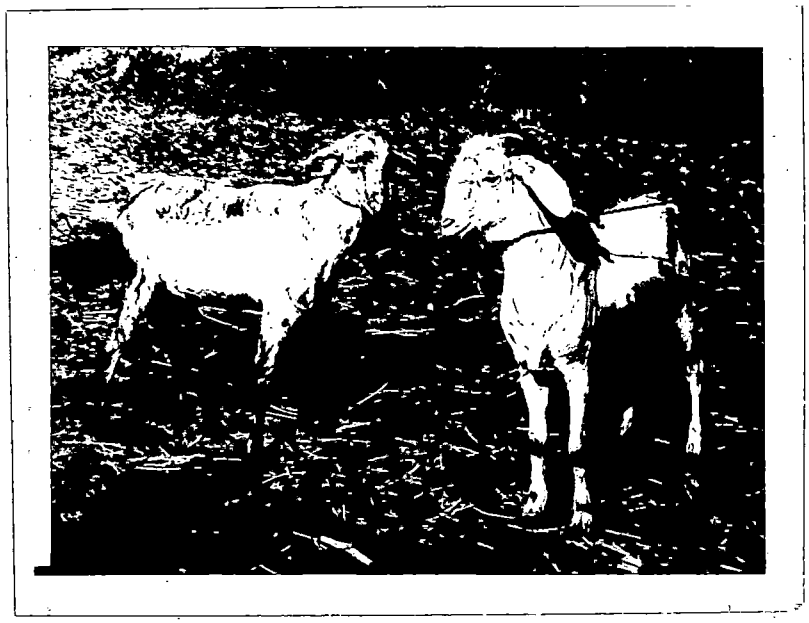
*Zondek, H. (1918) Das Myxodemherz. Munchen. Med. wschr. 65 : 1180.

* Reference not consulted in original.

PHOTOGRAPHS

Fig. 2. Group I - Stunted growth in experimental hypothyroidism - The buck on the left side is stunted in growth and dull and matted hair. The age matched healthy control animal is on the right side.

Fig. 3. Group IV - Stunted growth in kids in experimental hypothyroidism. Experimental kid is stunted in growth compared to the age matched control animal on the left side.



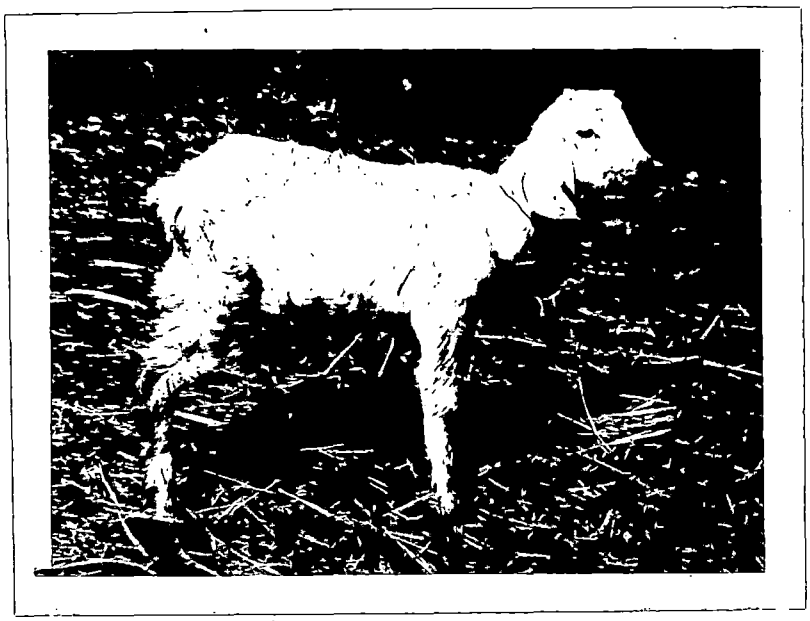


Fig. 4. Group IV - Stunted growth in experimental hypothyroidism. The control animal is on the left side.

Fig. 5. Clinical symptoms - Experimental animal showing periorbital oedema and rough matted hair coat.

Fig. 6. Clinical symptoms - experimental animal - The animal is weak, has loose skin and matted rough hair coat.

Fig. 7. Clinical symptoms - experimental animal showing lacrimation and facial oedema.

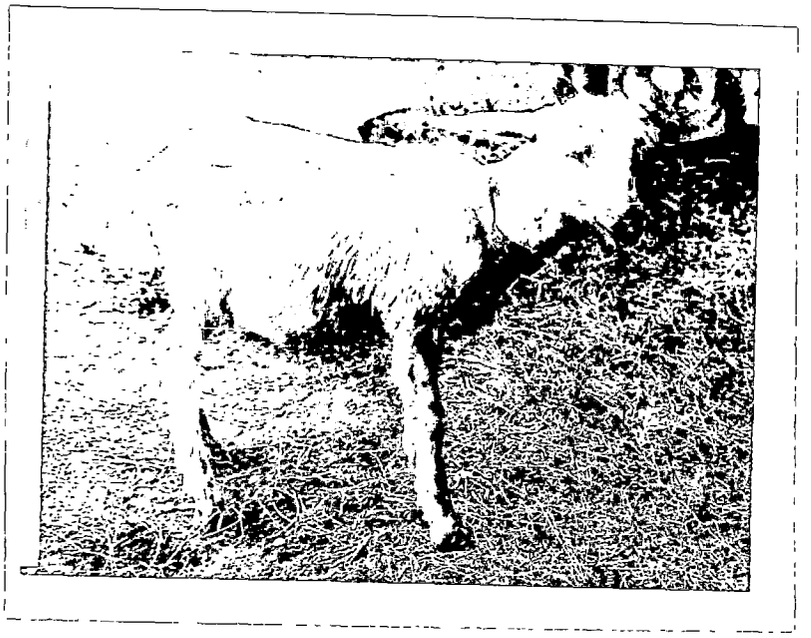


Fig. 22. Abnormal Spermatozoa - Free loose Heads. X 1000.

Fig. 24. Abnormal Spermatozoa - Middle piece abnormality. X 1000.

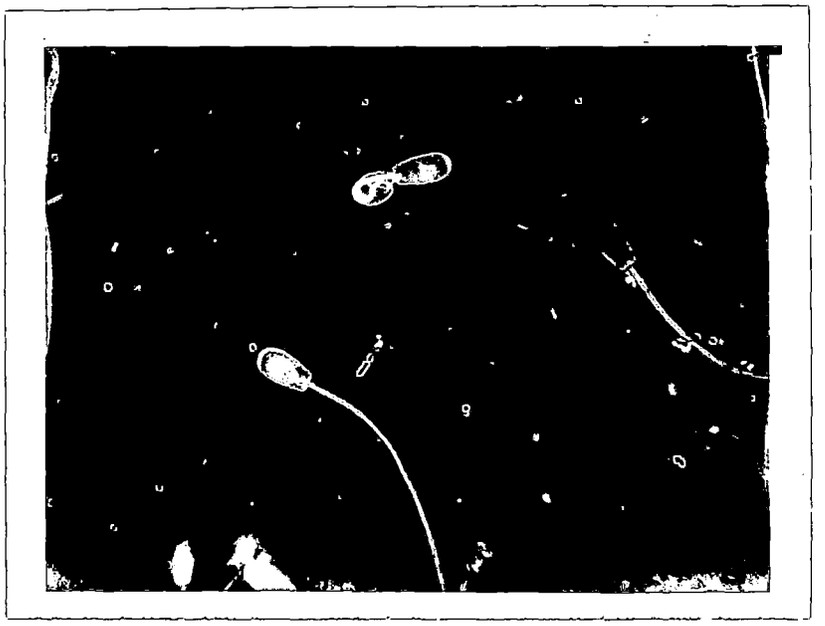
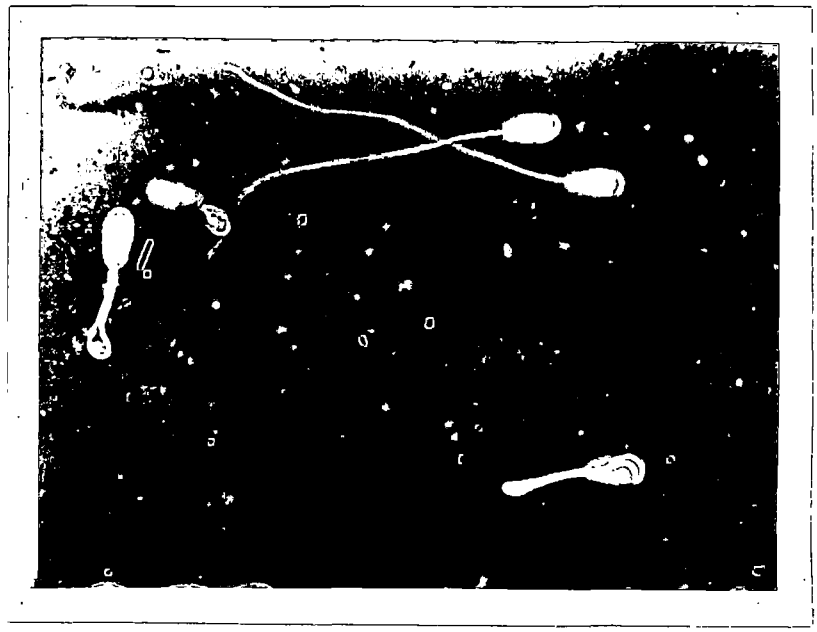


Fig. 26. Abnormal spermatozoa - Abnormalities in tail. X 1000.

Fig. 29. Thyroid gland - Enlarged thyroid glands of the experimental group. The glands of the control animal are seen on the right side.



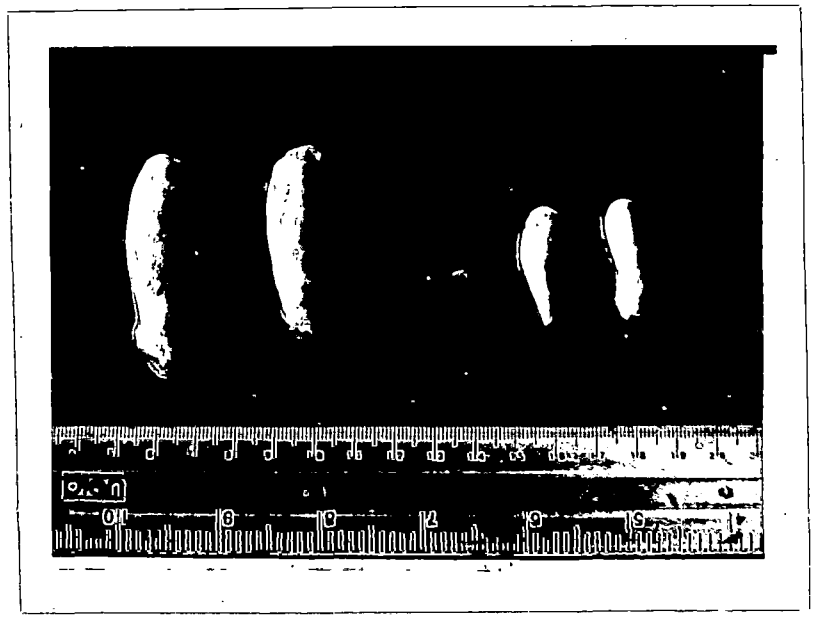


Fig. 30. Group A1 - Thyroid - Follicles are seen lined by tall columnar epithelial cells. Lumen is devoid of colloid. H & E X 160.

Fig. 31. Group A2 - Thyroid - Numerous small follicles are seen lined by tall columnar epithelial cells. Follicular lumen is small and devoid of colloid. H & E X 160.

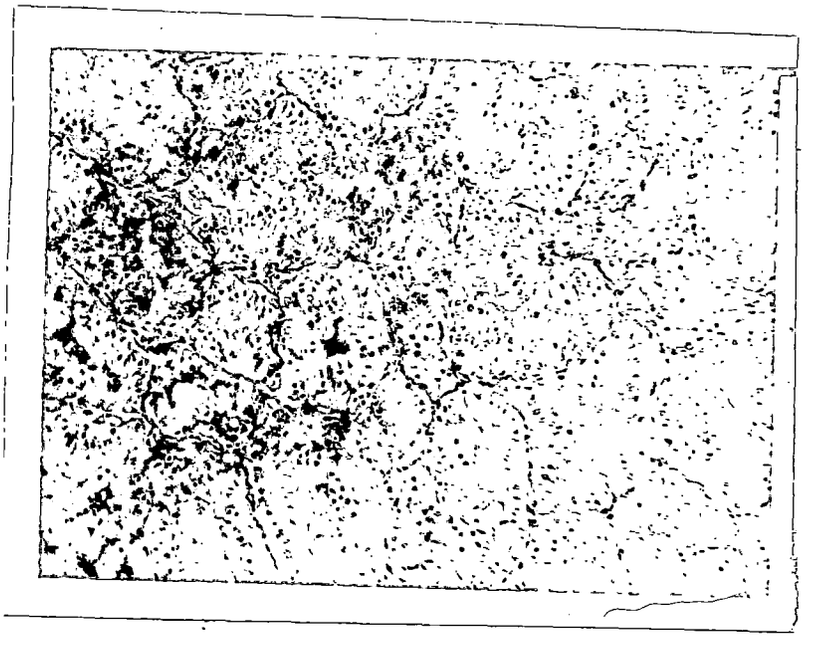
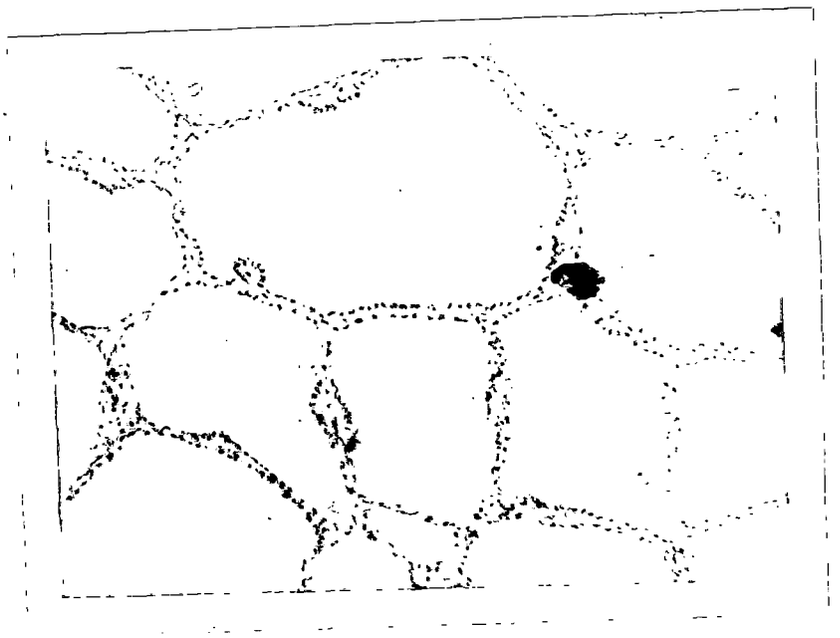


Fig. 32. Thyroid - Discontinuation of treatment with thiourea - Distended follicles are seen lined with flattened epithelial cells. H & E X 160.

Fig. 34. Group 12 - Pituitary - Severe degree of hyperplasia of basophils and chromophobes. Vacuolation and formation of cysts are also evident. H & E X 250.



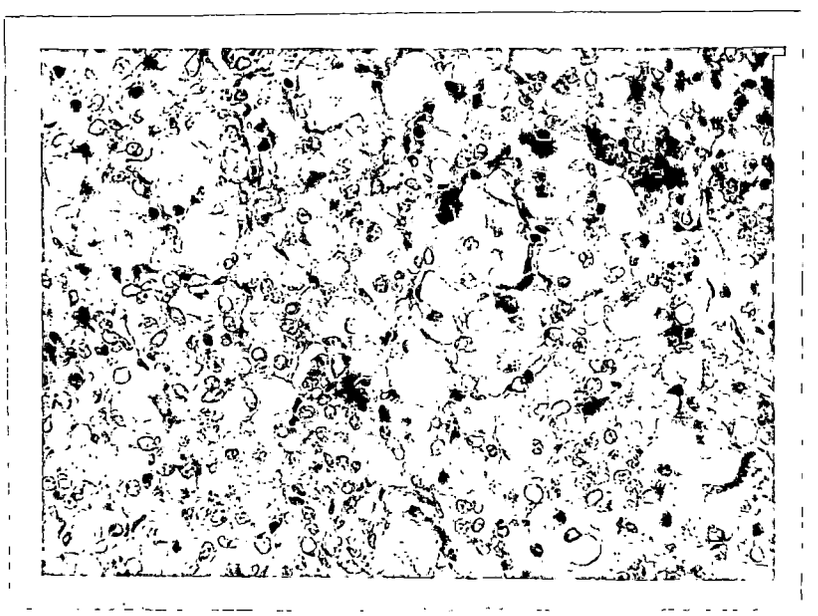


Fig. 35. Pituitary - Acidophils are seen in normal proportion. Vacuolation in few of the basophils is still evident after discontinuation of treatment with thiourea. H & E X 250.

Fig. 37. Adrenal - Group T1 - hyperplasia and hypertrophy of cells of the zona fasciculata. In focal areas the cells were devoid of fat. H. & E X 160.

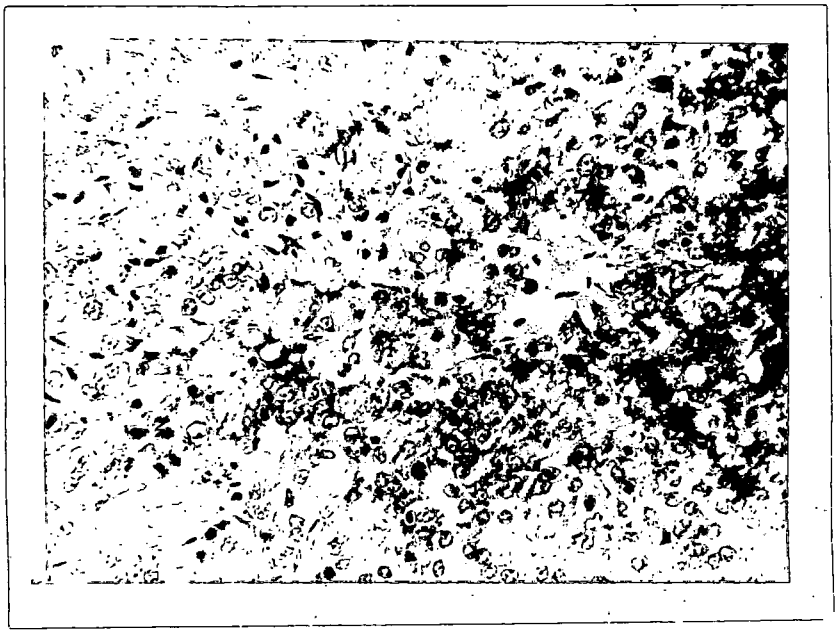


Fig. 38. Adrenal - Group T2 - Hypertrophy and hyperplasia and scattered fatty cysts in the zona fasciculata. H & E - X 160.

Fig. 40. Testis and Epididymis - Group I - Reduction in the size of the testis and epididymis. The testis and epididymis of the control animals are on the left side.

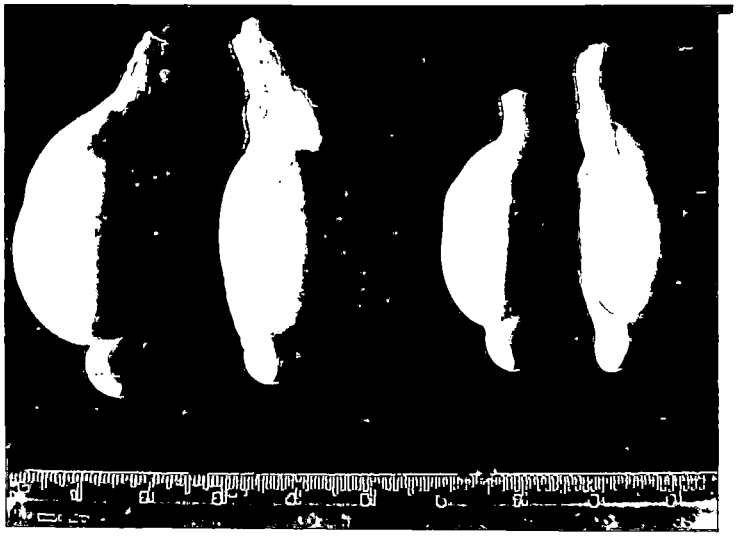
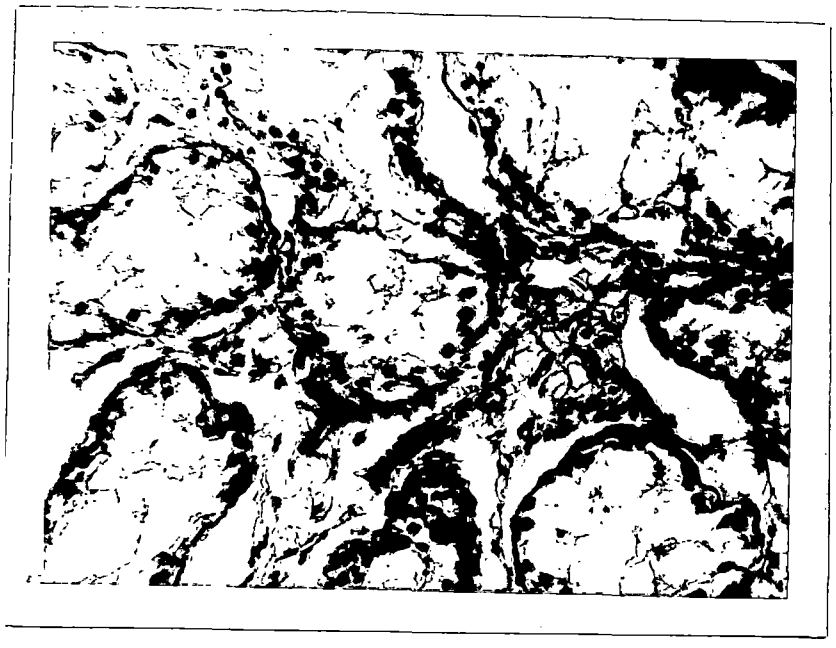


Fig. 41. Testis and Epididymis - Group II - Reduction in the size of the testis and epididymis in experimental animals. Testis and epididymis of the control animals are seen on the left side.

Fig. 42. Testis - Group T1 - Seminiferous tubules showing vacuolar and hyaline degeneration. Primary and secondary spermatocytes are also seen. H & E X 250.

Fig. 43. Testis - Group T2 - A few scattered spermatogonial cells close to the basement membrane are seen in the seminiferous tubules. No evidence of spermatogenesis. H & E X 400.

Fig. 44. Testis - Group II - Severe degree of hyaline degeneration in the seminiferous tubules. Spermatogenesis is absent. A few scattered interstitial cells are seen. H & E X 400.



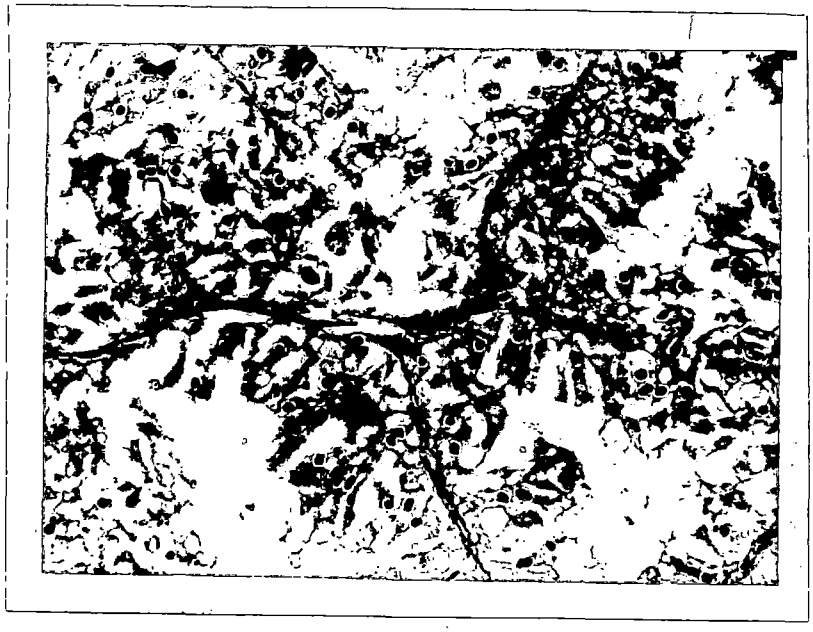


Fig. 45. Testis - After discontinuation of treatment with thiourea - Seminiferous tubules are active and organised layering of spermatocytes, spermatids and sperms is seen. Slight degeneration is still evident. H & E X 400.

Fig. 46. Epididymis - Tubules showing absence of sperms. Interstitial tissue is predominant. H & E X 160.

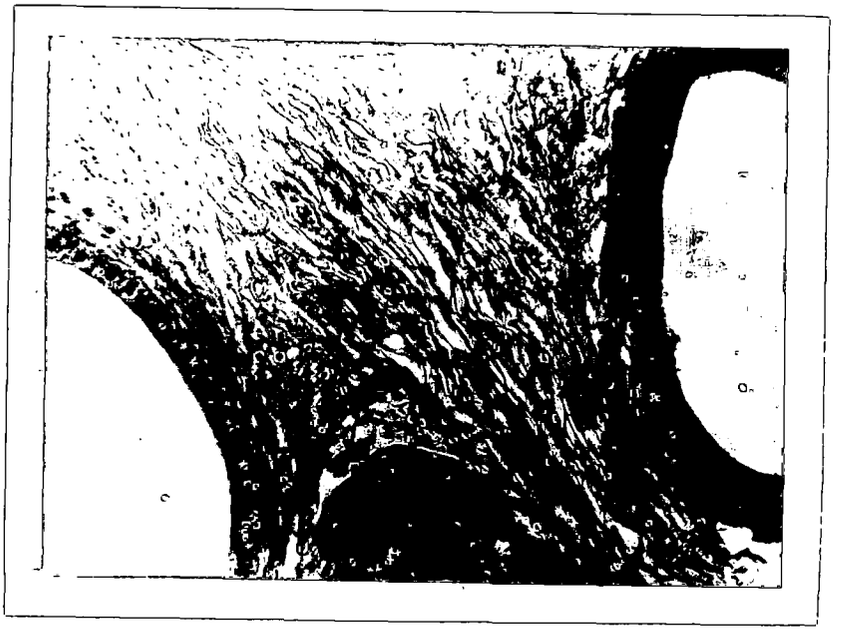
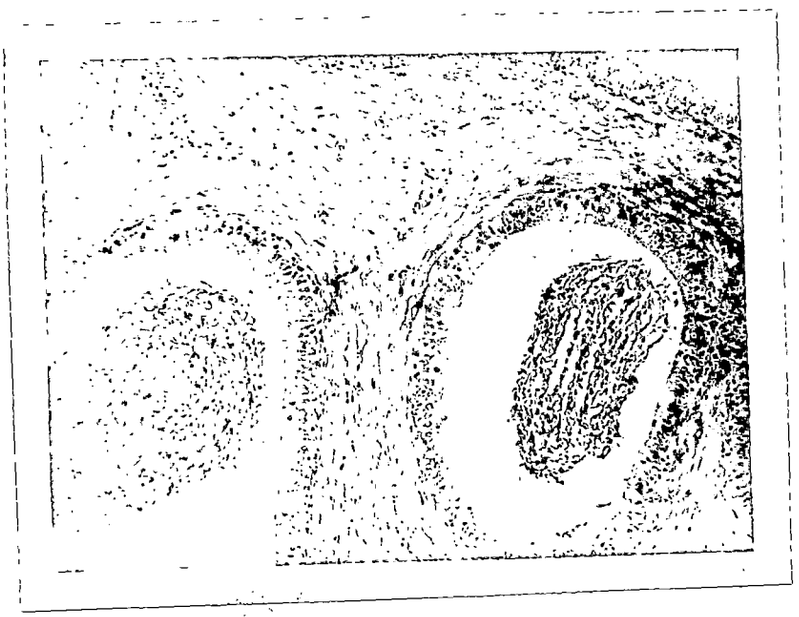


Fig. 47. Epididymis - Tubules showing degenerative changes. There is desquamation of the epithelial lining. The cells are of low cuboidal flattened epithelial type. Interstitial tissue is oedematous. H & E X 150.

Fig. 48. Epididymis - Tubules are intact with mild degeneration. Sperms are seen in the tubules. H & E X 150.



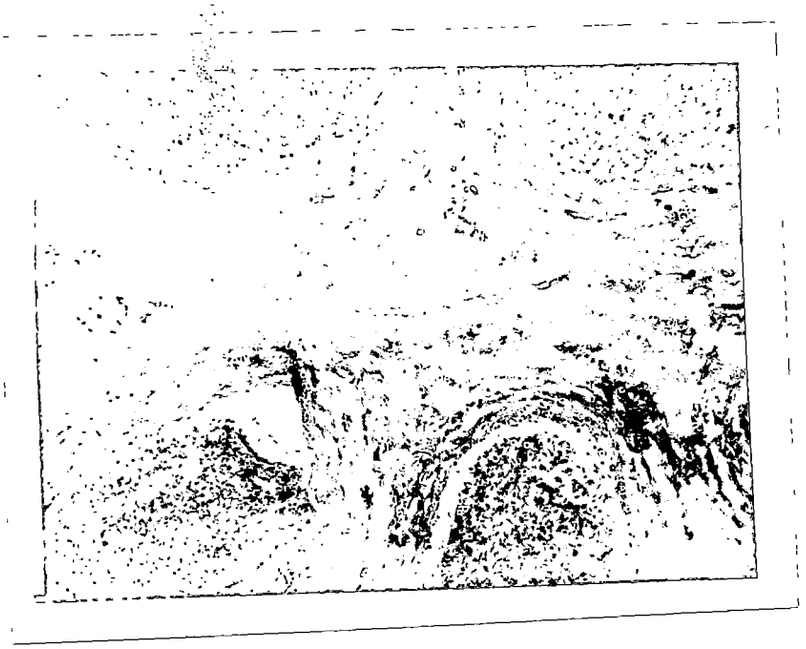


Fig. 49. Caput epididymis - Decilliation of the epithelial lining of the tubules. The lining cells are low cuboidal to flattened epithelial type. H & E X 250.

Fig. 50. Corpus epididymis - Tubules showing degenerative changes. Interstetial tissue is oedematous. H & E X 250.

Fig. 52. Seminal vesicles - Group T1 - The cells lining the glands are small and inactive with no evidence of secretory activity. H & E X 250.

Fig. 53. Seminal vesicles - Group T2 - The glands are dilated and cells lining the glands are vacuolated and hyalinised. No secretory activity is seen. The interstitial tissue is oedematous. H & E X 250.

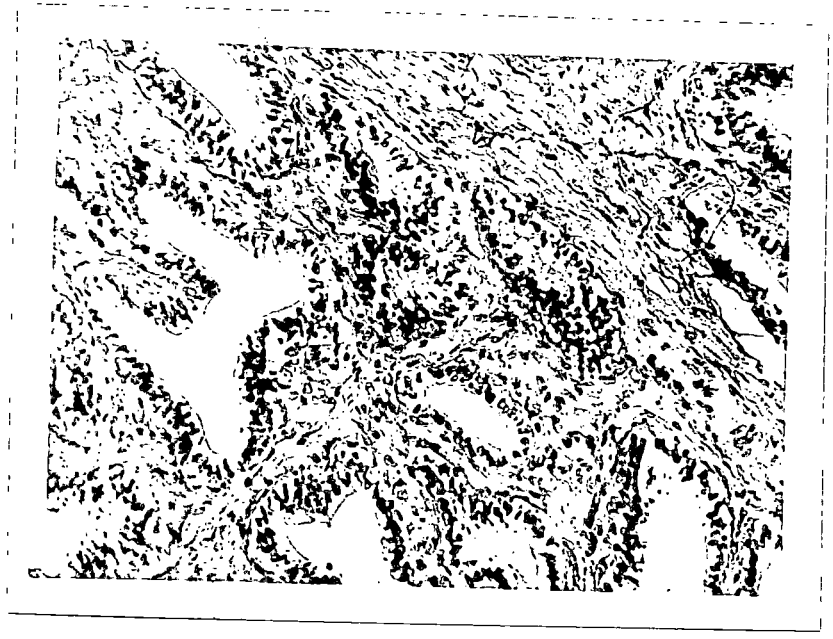
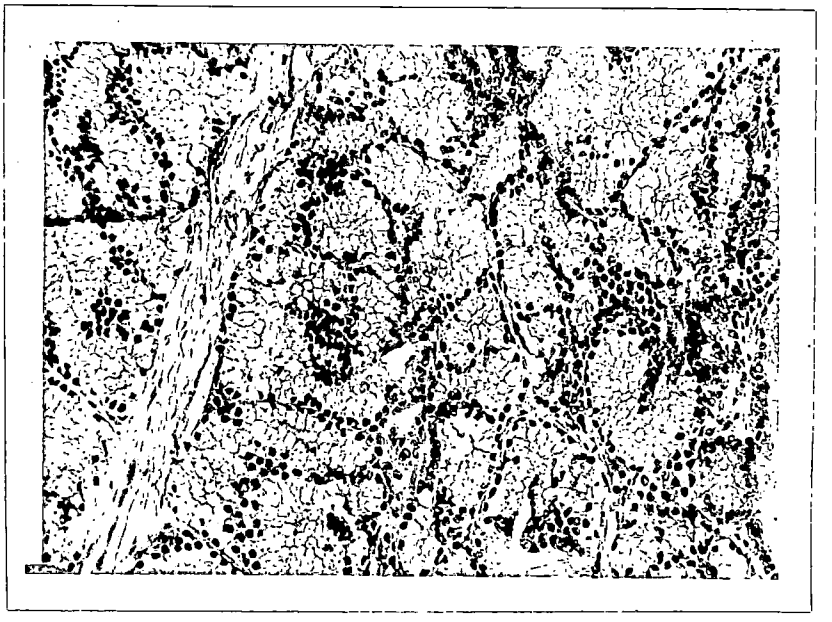


Fig. 54. Prostate - Group T1 - The cells lining the glands show vacuolation. H & E X 250.

Fig. 55. Prostate - Group T2 - The acini are small and degenerative changes are seen. H & E X 250.



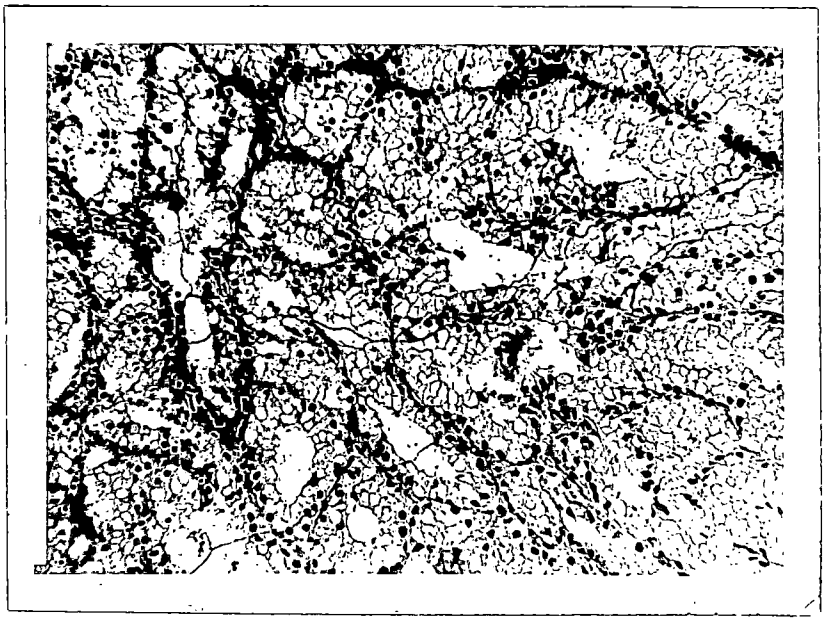


Fig. 57. Ovaries - Inactive ovaries in group III. The small ovaries and enlarged thyroids are seen on the side. The organs are from the control animals in the middle.

Fig. 58. Ovaries - Smooth and small ovaries in group IV are seen at the bottom along with enlarged thyroid glands. Active ovaries and the thyroid glands from the control group are seen on the top.

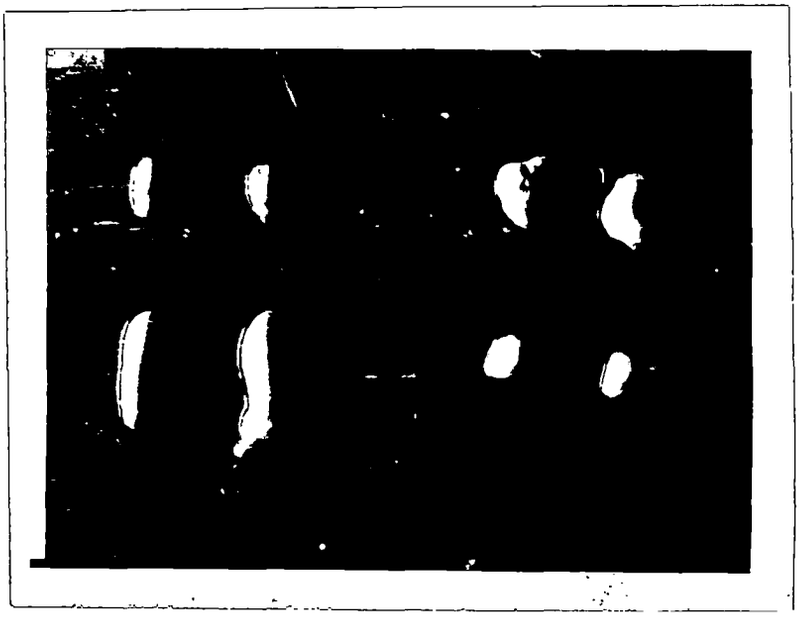


Fig. 59. Ovary - Cortex - showing loose cellular stroma and few atrophic follicles. H & E X 250.

Fig. 60. Ovary - Follicles showing degenerative changes in granulosa cell layer. H & E X 250.

Fig. 61. Ovary - Cortex - loose, oedematous cellular stroma. Degenerating follicles are also seen. H & E X 250.

Fig. 62. Ovary - stromal tissue consisting of closely packed vacuolated elongated spindle shaped cells. Corpus albicans is also seen. H & E X 250.

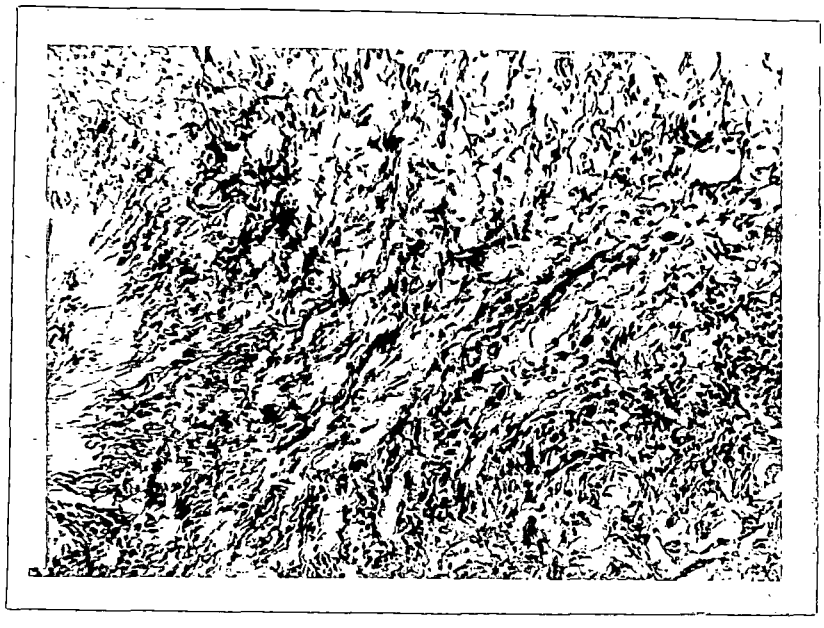
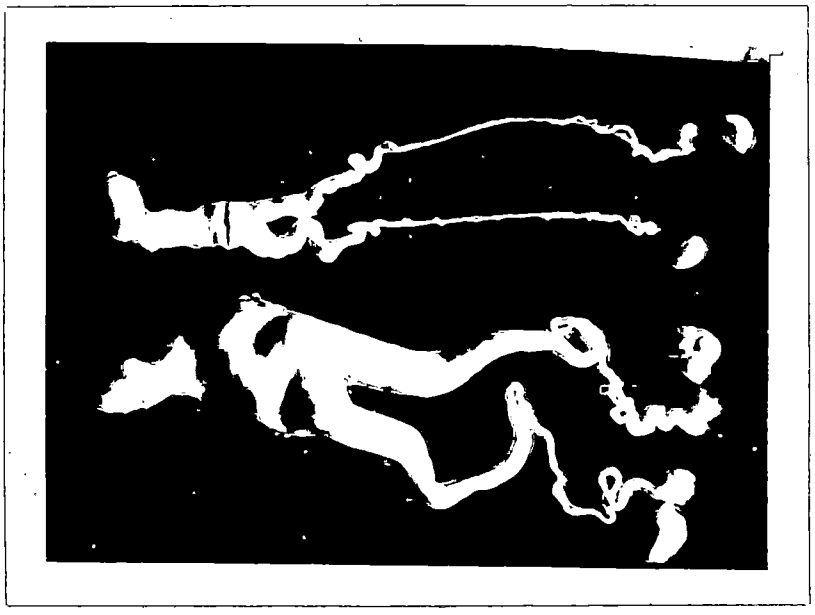


Fig. 64. Group III - The hypoplastic ovaries and uterus of the experimental animals are seen on the top. The normal ovaries and uterus of age matched control are at the bottom.

Fig. 65. Group IV - Hypoplastic uterus from the experimental animal on the top- Uterus from the control animal at the bottom.



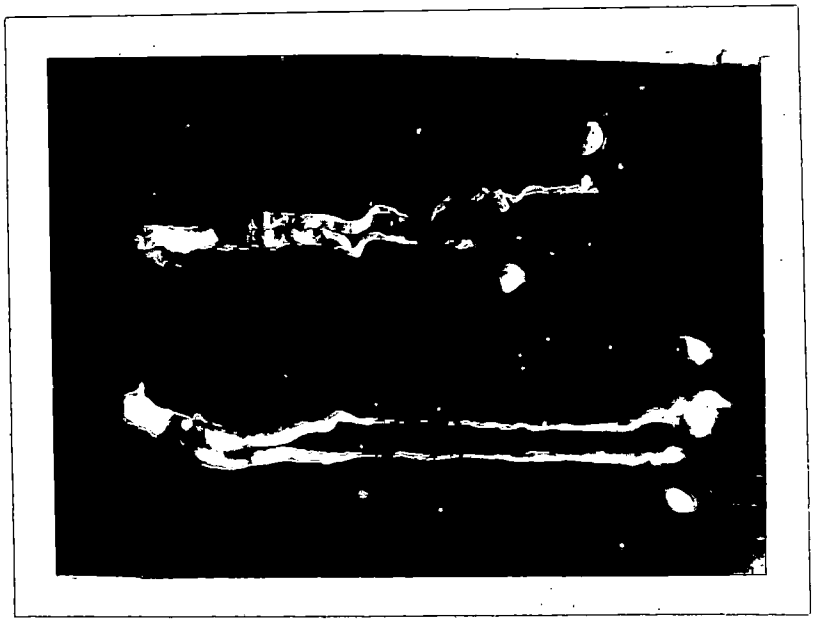
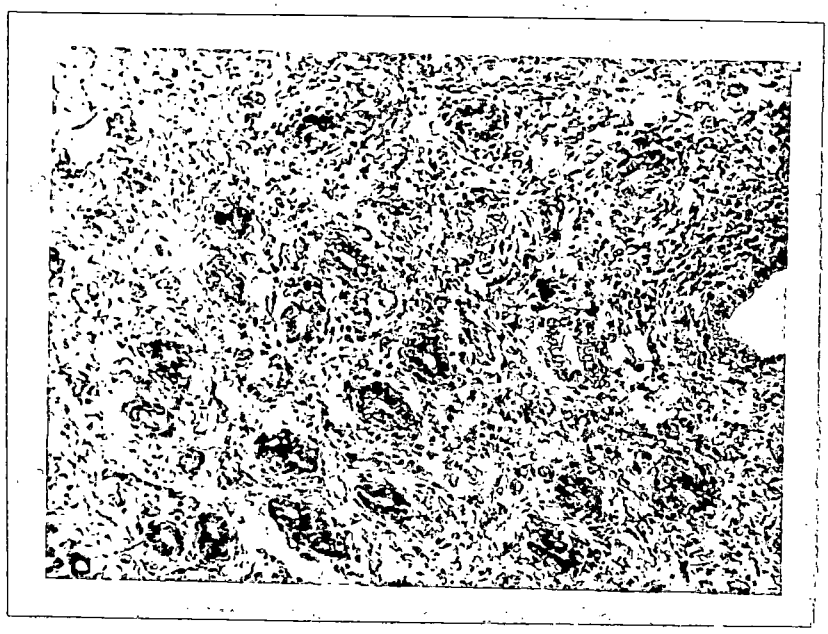


Fig. 66. Group III - T1 - Uterus - The glands are small in size, inactive and lined by low cuboidal type of epithelium. No evidence of secretory activity is seen. Moderate interstitial oedema is also evident. H & E X 160.

Fig. 67. Group III- T2 - Uterus - The glands are small in size and the lining cells are cuboidal to flattened epithelial type. Moderate interstitial oedema is also seen. H & E X 160.



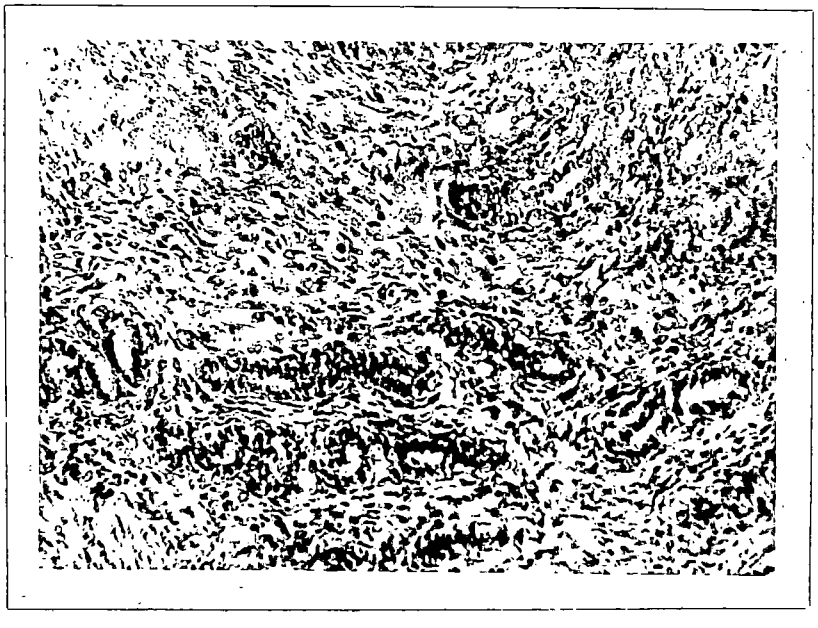
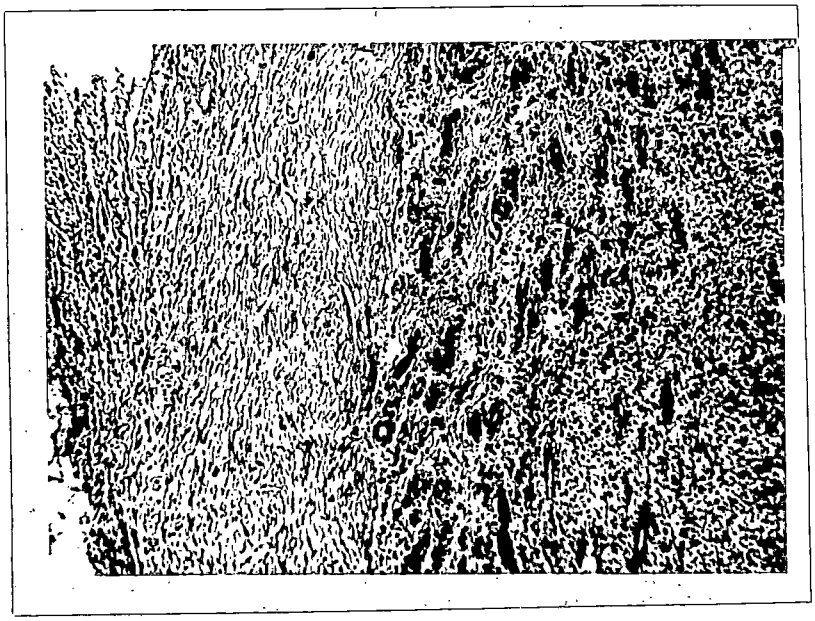


Fig. 68. Group IV - Uterus - Uterine glands are small and slit like. Stroma is relatively more. H & E X 160.



PATHOLOGY OF THE REPRODUCTIVE ORGANS IN EXPERIMENTAL HYPOTHYROIDISM IN GOATS

By
NEMALI MOHAN REDDY

ABSTRACT OF A THESIS

Submitted in partial fulfilment of the
requirement for the degree

DOCTOR OF PHYLOSOPHY

Faculty of Veterinary and Animal Sciences
Kerala Agricultural University

Department of Pathology
COLLEGE OF VETERINARY AND ANIMAL SCIENCES
Mannuthy - Trichur

1982

ABSTRACT

A survey study was conducted to assess the role of hypothyroidism in the etiology of reproductive disorders in goats. Lowered functional activity of the thyroid was associated with postpartum anoestrum, repeat breeding and delayed puberty. An experimental model of controlled hypothyroid state was induced in goats, using different dose regimes of thiourea with the objective of studying the sequence of physiopathological changes in hypothyroidism and its influence on reproduction. Clinically healthy young and adult cross-bred goats of both sexes were employed for the study. The animals were divided into control, Treatment I (T1) and Treatment II (T2) groups. Groups T1 and T2 were administered thiourea orally at the rate of 50 mg/kg and 100 mg/kg body weight respectively. Clinical symptoms and growth rate were studied. Protein bound iodine, total serum protein and serum cholesterol were estimated. Haematological data were collected and the semen characteristics were evaluated. On the 90th day of the experiment both control and experimental animals were sacrificed leaving at least one animal in each group. These animals were maintained without treatment with thiourea

for a further period of 90 days and sacrificed. Gross lesions and histopathological changes in tissues were recorded.

Weakness, lethargy, depression, reduced feed intake, subcutaneous oedema of varying degree, loss of libido in males and anoestrum in females were the important clinical features observed. There was stunting of growth and apparent reduction in weight of the animals dosed with thiourea. On discontinuation of treatment with thiourea all the clinical signs manifested disappeared and the animals gained weight progressively. There was significant decrease in the protein bound iodine, and increase in the total serum protein and cholesterol level in all the animals dosed with thiourea. The values reached the normal level on discontinuation of treatment with thiourea. Macrocytic hypochromic anaemia was evident in hypothyroid goats. There was improvement in haemogram values on withdrawal of thiourea administration. Reduction in quantity and quality of semen was observed in hypothyroid goats. The semen appeared normal in quality and quantity on discontinuation of thiourea administration.

Gelatinisation of subcutaneous fat and hypertrophy

and dilatation of the left ventricle were the common findings at autopsy. There was significant increase in the relative weight of the thyroid, pituitary and the adrenal gland. Histologically the thyroid glands exhibited varying degree of hyperplasia and hypertrophy. Predominant histological changes in the pituitary was hyperplasia and hypertrophy of basophil cells and degenerative changes in the acidophils. Adrenal gland showed hypertrophy and depletion of fat in the zona fasciculata. On discontinuation of treatment with thiourea all the lesions disappeared.

A significant decrease in the relative weight of the testis and epididymis was observed in all the goats dosed with thiourea. Histologically the seminiferous tubules were small and lined by single layer of spermatogonial cells. Some tubules contained degenerated and desquamated cells without any evidence of sperms. The tubules of the epididymis were small in size and the lining cells showed deciliation and desquamation. These degnerative changes were found to be reversible on discontinuation of treatment with thiourea. The accessory sexual glands, the seminal vesicles and the prostate weighed less in hypothyroid goats. Histologically the cells lining the

glands were small inactive, vacuolated and hyalinised. After discontinuation of administration of thiourea the reparative changes were almost normal. There was significant decrease in the relative weight of the ovary and uterus in all the animals dosed with thiourea. Histologically ovaries were found to be inactive and only few scattered immature follicles were seen. Uterine glands were few and non secretory type. The lining epithelial cells did not show active mitosis. On discontinuation of treatment with thiourea the weight of the ovary and uterus increased and histologically ovaries and uterus were found to be normal. Kids and female goats were more susceptible to the effects of hypothyroidism. The significant pathological changes observed in the reproductive organs have clarified the importance of hypothyroidism in inducing subfertility and infertility in goats. The reversible nature of the lesion was also proved.