CHAPTER 4

CERTAIN CYCLIC CHANGES IN THE THYROID OF THE MIGRATORY STARLING, STURNUS ROSEUS (LINNAEUS)

Seasonal changes in the thyroid of several avian species, migratory as well as non-migratory, have been the subject of extensive studies (Riddle and Fisher, 1925; Hacker, 1926; Watzka, 1934; Küchler, 1935; Miller, 1937; Winchester, 1940; Turner, 1948; Höhn, 1949, 1950; Davis and Davis, 1954; Oakeson and Lilley, 1957, 1960; Swift, 1959; Wilson and Farner, 1960). It is now well known that birds are no exception to the general rule that thyroidal activity is increased in winter and decreased in summer (Reineke and Turner, 1954; Höhn, 1961; Dorst, 1962). Besides, thyroidal activity is considerably increased during moult (Glazener and Jull, 1946; Höhn, 1949). However, there is some difference of opinion regarding the role of the thyroid in bird migration.

Riddle, Smith and Benedict (1932) observed in the migratory Mourning Dove, Zeniadura macroura, that inspite of the low activity of the thyroid towards winter, the birds did migrate. Administration of thyroid extract have been observed to stimulate 'Zugunruhe' in the white throats (Wagner, 1930). Similarly Merkel (1937, 1938, 1940) found that in white throats and European robins, small doses of thyroxin injections stimulated 'Zugunruhe', but not larger doses. He (1958) suggested that the thyroid could have an integral function in the annual stimulation for migration, and that a slight increase in the thyroid activity could well be responsible for the initiation of fall as well as spring migrations. He also suggested that a higher level of thyroxin causes moulting. This was also in agreement with the data obtained by previous workers (Häcker, 1926; Küchler, 1935; Elterich, 1936).

Groebbels (1930) had suggested that decreased thyroid activity might be involved in the development of the necessary metabolic conditions stimulating migration. On the other hand, others believed that an increase in thyroid activity causes migration (Putzig, 1937, 1938a). Putzig (1938b), Schildmacher (1952), Wilson and Farner (1960) suggested that the response of the thyroid to temperature changes may be an intrinsic rhythm which could be important in migration. Merkel (1958) after confirming his earlier results, showed that migratory activity could be inhibited by a dose of 0.2 cm³ of a solution of 5% methyl thiouracil.

In the light of the works cited above it is clear that the actual role of the thyroid in bird migrations is a controversial subject. The present work on the seasonal variations in the activity of the thyroid in relation to corresponding changes in the other endocrine glands and organs in the Rosy Pastor was carried out with a view to a better understanding of the problem (Chapters, 1, 3, 5 to 9).

The Rosy Pastors arrive in Baroda (India) about September and leave by late April to return to their breeding grounds abroad.

Material and Methods

Birds of both sexes were collected every month from

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October to April. The thyroids were immediately dissected out and fixed in Bouin's, Zenker formol and formol saline. Paraffin blocks were prepared and sections of 5 to 6 μ cut and stained with the following stains. (1) Heidenhain azan stain as described by Gurr (1956). (2) Haematoxylin and eosin. The histological observations made every month are presented in Table I. The height of the epithelial cells of the follicle and the relation between the inner diameter and the number of cells in the follicle were measured in order to obtain the ratio, diameter/number of cells (d/n), according to Lever (1948, 1950) and Stolk (1957).

The average ambient environmental temperature (Maximum and Minimum) for every month during the period under investigation (1960-1961) was obtained from the Meteorological department of the University (Table I).

The estimation of the protein-bound iodine in the thyroid was done by the method adopted by Joshi (1962) from the original methods of Salter and Jonston (1948) and Barker (1951). The birds (both sexes) were taken in two sets, one in the first week of April and the other a few days prior to migration. After the birds were killed, both the thyroids were removed carefully without rupturing them and weighed on a Mether balance. The tissues were transferred into chilled pyrex test tubes and homogenized in 10 ml of cold acetone using a glass rod. The homogenate was centrifuged at 3000 r.p.m. for 10 minutes. The supernatant was decanted off. Again 10 ml of cold acetone was added and the procedure was repeated. The rest of the procedure was as followed by Joshi (1962).

Observations

The Heidenhain azan stained preparations showed acidophilic colloid in the follicles of the thyroid in different grades of colour intensity from October to April. In very few cases, however, the basic colloid was observed in the peripheral follicles of the thyroid. The activity of the thyroid in these birds could be classified broadly into four phases on the basis of the epithelial height of the follicle cells, the d/n ratio, follicle diameter and the amount of the colloid present.

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(1).	October	Period	of	high activity.
(2).	November to March	17	Ħ	moderate activity.
(3).	April (1st to 21st)	11	Ħ	very high activity.
(4).	April end (3 or 4 days prior to migration)	și,	Ħ	release of the colloid

During the first phase (October), the thyroid on the basis stated above, was found to be quite active (Table I). The amount of the colloid present could be determined by the intensity of the staining obtained. When the colloid is thick it is separated from the cell boundary (Figs. 1 & 2). The histological preparations showed an increase in new follicles having numerous dense nuclei with compact cytoplasm. During this month (October) though food was available in plenty they did not accumulate much fat in the body (Chapter, 1).

The second phase started in November with a decrease in the amount of the colloid stored as well as in the height of

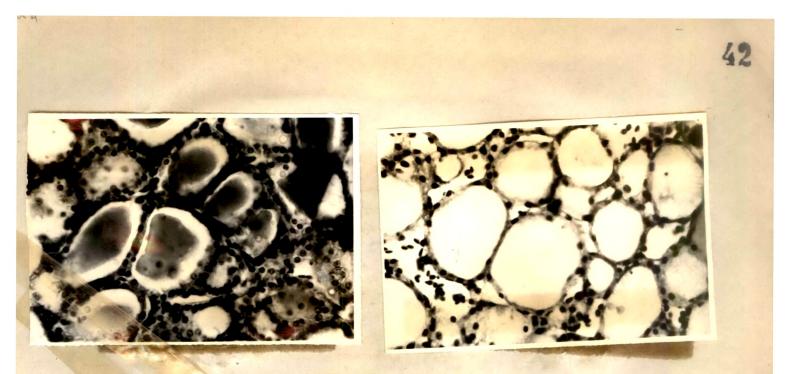
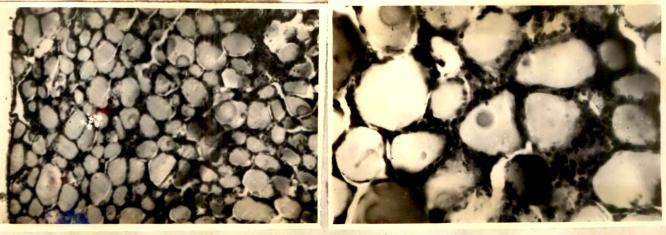


Fig. 1



Fig.1. T.S. of thyroid of Rosy Pastor in the month of October. Mean height of the epithelium is 3.4 u. The follicles are seen to contain large accumulation of the colloid.(Heidenhain azan stained) Fig.2. A portion of Fig. 1 magnified.



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- Fig. 3. T.S. of thyroid in the first week of March. Mean height of the epithelium = 3.0 u. The follicles contain less colloid. (Heidenhain azan stained)
- Fig. 4. A portion of Fig. 3 magnified.

Table I

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Showing changes in the thyroid activity of Rosy Pastor during October to April.

	Average mont temperature.	Average monthly temperature. °C.	Number of birds		Thyroid activity		
Months	Max1- mum	Mini- mum	studied	I Ht. of the follicular cells. u	Diameter of the follicle. u	No. of cells of folli ≤	Ratio d/n
October	35 . Č	20.2	œ	3.42 ± 0.14	38.7 ± 4.56	16	2.42 ± 0.20
November	33 . 8	16.0	S.	2.76 ± 0.16	32.64± 4.15	11	2.96 ± 0.31
December	30.3	13.2		3.30 ± 0.24	40.55 ± 3.69	17	2.32 + 0.20
January	30.3	12.0	œ	3.34 ± 0.14	41.39 ± 4.84	17	2.43 ± 0.13
February	33.3	12.8	00	3.01 ± 0.17	41.58 ± 5.16	17	2.44 ± 0.30
March (1 tó 15)	34.7	15.0	10	3.07 ± 0.17	41.19 ± 6.38	17	2.42 ± 0.16
March (16 to 31)	38°3	18.2	œ	3.17 ± 0.17	42 . 85 ± 6.54	17	2.52 ± 0.02
pr11	38 .3	20.3	01	3.47 ± 0.16	47.55 ± 4.73	50	2.35 ± 0.10
(16 to 22) 40.4) 40 c4	22.4	œ	4.28 + 0.25	56.70 ± 4.08	26	2.18 ± 0.17
(23 to 26) 40.4	40.4	22.4	თ	2.42 ± 0.12	60.38 ± 5.78	16	3.47 ± 0.24

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Fig. 5. T.S. of thyroid in the third week of April. Mean height of the epithelium is 4.3 u. Follicles contain very large amount of the colloid. (Heidenhain azan staimed)
Fig. 6. A portion of Fig. 5 magnified.

Fig. 5

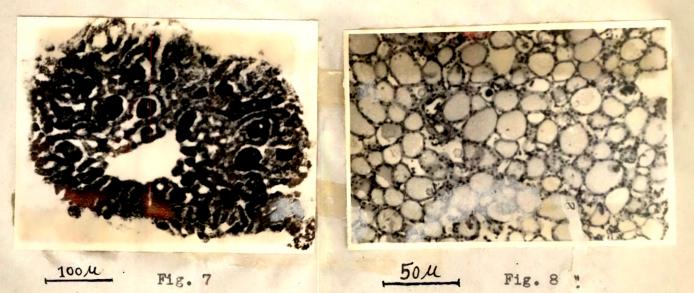


Fig. 7. T.S. of thyroid 2.3 days prior to migration. Mean height of the epithelium = 2.4 u. The follicles are almost empty without the colloid. (Heidenhain azan) Fig. 8. A portion of Fig. 7 magnified. the epithelial cells However, the increase in the d/n ratio (Table I) suggests 1/w thyroid activity. During this month the birds increase in weight and a slight fat accumulation in the liver was noted (Chapter, 1). In the next two months (December and January) there was a gradual increase in thyroid activity (Table I) followed by a slight decrease from the middle of February to the end of March. In the first week of March the thyroid follicle had only little colloid but it was completely filled. The epthelial cell height was also low (Figs, 3 & 4). The diameter of the follicle was moderate.

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In the third phase the thyroid became more active (Table I), the epithelial cells increased in height as well as in number(Figs. 5 & 6). The colloid of the follicle became thicker than that in October and a gradual increase in follicle diameter was recorded. As the migratory period approached nearer and newser, the activity of the thyroid increased with increased storage of the colloid. This condition of the thyroid more or less resembled the goiter type, having excess of the secretion stored in the follicles.

In the last phase, just a few days prior to migration, the colloid from the follicles was released. The follicles became almost empty with hardly any secretion left. The follicle size was found to increase but the cells appeared to decrease in number. The epithelial cells presented a flattened appearance (Figs. 7 & 8). The d/n ratio also increased (Table I).

The amount of protein-bound iodine in the thyroid during

the first week of April was higher than what it was a few days prior to the migration (Table 2). However, when the histological data obtained during these periods are considered, it is clear that the peak thyroid activity was in the third week of April. The results obtained with regard to protein-bound iodine for the first week of April, are not the highest. It is also realized that the lower figure for iodine obtained during the last week of April was due to the the release of the secretion.

Table 2

Protein-bound iodine in the thyroid of the Rosy Pastor during first and last week of April.

Number of birds studied.	, Month	Iodine value, µg protein- bound iodine in 100 mg wet tissue of thyroid.
?	April (First week)	121.17 <u>+</u> 7.94
8	April (Last week)	84.47 ± 6.38

Discussion

Merkel (1940) found a relationship between thyroid activity and body weight. He found that after an injection of a large dose of thyroid extract or of 0.3 ml of thyroxin, the bird W_{f}^{g} lost weight. This effect must have been due to increased oxidation of metabolites particularly the fat reserves. Merkel (1958) also found that administration of 0.1 ml of thyroxin to a bird induced

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heavy deposition of fat. Similar results were obtained later also by Schildmacher and Rautenberg (1952) by giving 0.2 mg of thyroxin to Bramblings and Chaffinches, Fringilla coelebs. This resulted in increased food intake and consequently increase in body weight despite a higher metabolic rate. Langdon (1960) and Menon (1961) have also suggested a role for thyroxin in fat metabolism. In the light of the work cited above, what remained to be shown in migratory birds is that a low release of thyroxin during the premigratory period actually facilitates premigratory fattening and a high release prior to migration helps in the utilization of fat for energy during migratory flight. In the Rosy Pastor during October the slightly increased activity of the thyroid was accompanied by slight hyperphagia accounting for the increased body weight recorded in November (Chapter, 1). During November the thyroid activity decreased while the activity of the islet cells increased, thereby denoting less utilization and greater storage of metabolites.

In November the thyroid activity was less and the islet cells were increasing in number denoting increasing hyperphagia. During this period increased in body weight and increased fat storage in the liver were recorded. This weight increase is therefore to be regarded as for wintering.

In December and January again the thyroid activity increased, at the same time the islets were gradually getting disorganized and decreasing in activity (Chapter, 3). It is most likely that during these winter months the accumulated fat was used for the increased basal metabolism required to maintain 47

body temperature. A reduction in the liver fat at this time, was also noted (Chapter, 1).

The end of March saw the beginning of an active phase of thyroid as well as islet cells accompanied by hyperphagia (Chapter, 3). This was followed by the fast fat accumulation and gonadial development (Chapters, 1 & 5). Increased thyroid activity is known to enhance gonadial development (Magsood, 1952, 1954).

In the anterior pituitary two phases of increase in thyrotrop cells were observed: one during December and January and the other in April (Chapter, 7). The increase in thyrotrop cells was followed quickly by increase in thyroid during these periods. It was also observed that at the peak thyroid activity, the colloid in the follicle was very thick and was released three to four days prior to migration. The release of the colloid was followed by the degranulation of the thyrotrop cells as well as decrease in alkaline phosphatase activity in these cells (Chapter, 7 & 8). The observations on changes in protein-bound iodine, also support the possibility of the release of the colloid at that time.

The release of thyroxin three to four days prior to migration should be considered very significant event in the operation of the migratory stimulus because at that time the birds became restless. Lardy <u>et al.</u>, (1960) showed that thyroid feeding induced an almost 20 fold increase in rat liver mitochondria and alpha-glycerophosphate dehydrogenase. George and Talesara (1961) observed a gradual increase towards migration, of succinic dehydrogenase in the <u>Pectoralis major</u> of the Rosy Pastor. George and Vallyathan (1963) observed an increase of phosphorylase in the pectoralis of this bird towards migration. In the light of the above findings there seems to be no doubt that the thyroid has a direct influence in enhancing the oxidative metabolism, thereby promoting utilization of fat in migration. Lardy and Maley (1954) have shown in rats fed on desiccated thyroid tissue, increase in the number of mitochondria per unit of tissue increased oxidation of fat.

Schildmacher (1951) stated that spring migration in some birds was characterized by intense thyroid activity. Similarly Küchler (1935) and Merkel (1958) believed that the determinent 1. of the migratory impulse is in the thyroid cycle. Merkel (1958) states that the functioning of the thyroid, triggers the migratory impulse of birds. He argues that if cold could stimulate the thyroid, a drop in temperature could also be expected to trigger the departure of birds in migration. Caged birds have been observed to show increased migratory restlessness in cold (Merkel, 1958). In the case of the Rosy Pastor on the other hand, increased storage of the colloid in the thyroid preparatory to migration, is seen as the ambient temperature rises. At the time of migration, the temperature continued to be on the upward trend. Yet, the release of the thyroidal colloid took place three or four days prior to migration. This release was accompanied by the release of the neurosecretory material (Chapter, 6) from the neurohypophysis and the median eminence. It therefore seems clear that the release of

the neurosecretory material as well as the thyroidal colloid, acts as a trigger for migration. The possibility of a stress created by the increased cortical activity of the adrenal occurring simultaneously (Chapter, 9), also functioning as one of the triggers for migration cannot be ruled out.

It should be stated here that a drop in the temperature could act in cold countries as a trigger for thyroidal release though not as a direct trigger for migration. Difference in the daily photoperiod in these countries could also be effective in the accumulation of premigratory fat but in the tropical countries it would be temperature rather than light, that is a factor for premigratory fat storage.

Summary

1. Histological changes in the thyroid of the Rosy Pastor were studied during the period October to April. Four distinct phases in thyroid activity were formulated.

2. After the arrival of this bird in Baroda (India) about September, from their breeding grounds abroad, increased activity of the thyroid was observed from October. In November there was slight decrease in the thyroid activity and consequently there was also some accumulation of fat in the body for the coming winter.

3. Slightly increased activity of the thyroid for the maint@hance of body temperature during the winter months(December and January) and later upto March a moderate activity were observed.

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4. The highest thyroid activity was recorded in the first three weeks of April resulting in a large amount of colloid being stored in the follicles resembling the goiter condition.

5. Three or four days prior to migration the degranulation of the thyrotrop cells of the pituitary took place, followed by rapid release of the stored colloid from the follicles.

6. The role of the thyroid and the significance of the release of the thyroidal colloids as the stimulus for migration is discussed in relation to certain simultaneously occurring factors such as neurosecretion and other endocrinal and gonadial activities.