PART - II

ENDOCRINE PHYSIOLOGY OF TAIL REGENERATION

CHAPTER VIII

THYROID AND CARBOHYDRATE METABOLISM IN RELATION TO TAIL REGENERATION IN THE SCINCID LIZARD, MABUYA CARINATA : A LOCAL AND SYSTEMIC ANALYSIS

Carbohydrates in general as instant source of energy for living systems is a well documented fact. Importance of glycogen, the chief stored carbohydrate of animal tissues, in the energetics and metabolic activities of developmental processes too is by now well recognized (Borghese, 1957; O'Conner, 1957; Engle, 1961; Falin, 1961). Involvement of glycogen and its metabolism in processes associated with vertebrate appendage regeneration have also gained attention in the recent past. Schmidt (1968) and Connely et al. (1974) have given some insight about the functional significance of carbohydrate metabolism in amphibian limb regeneration. Similarly Shah and Chakko (1967), Radhakrishnan and Shah (1973), Shah and Hiradhar (1974) have extended such studies to reptilian tail regeneration. Most of their studies were restricted to the local site of regeneration. However, Procaccini et al. (1973), Shah et al. (1977 a, b) have explored the involvement of visceral carbohydrate source in amphibian and reptilian regeneration respectively. The earlier study of Shah et al. (1977 b) has shown the dependence

of the tail regeneration process on hepatic glycogen and blood glucose in M. carinata. In fact, blood glucose was considered to be the principal source of energy for the blastemal cells which had been further stressed by the reported elevated activity of hexokinase in the regenerating blastema (Shah et al., 1980 b). Findings of Chapter II have demonstrated dimetrically opposite pattern of changes in glycogen content and phosphorylase activity of the liver as well as adaptive alterations of these two parameters in the regenerate during tail regeneration in M. carinata. The findings reported therein have also shown significant involvement of muscle glycogen during regeneration and suggested thereby a strong systemic response involving carbohydrate metabolism in the molecular intricacies underlying tail regeneration.

Thyroid hormone which is known to be involved in modulations of carbohydrate metabolism (McNabb, 1969; Hoch, 1974), has been reported to have varying influence in amphibian limb regeneration (see Schmidt, 1968). However, studies of Turner and Tipton (1971) and Chapter I have both yielded an identical result of inhibition of tail regeneration under conditions of hypothyroidism. Though Turner and Tipton (1971), have attributed this inhibition to a delay in the outgrowth of ependyma, observations of Chapter I have suggested both indirect as well as direct actions of thyroxine on tail

regeneration in Mabuya carinata. Since thyroxine has been shown to influence tail regeneration and as it is also intimately associated with systemic metabolic aspects which is a factor of significance in lizard tail regeneration, an evaluation of thyroid in relation to regeneration, vis-a-vis local and systemic carbohydrate metabolism was thought pertinent. In this light a detailed quantitative analysis of total glycogen content and phosphorylase (EC 2.4.1.1) activity during tail regeneration in liver, muscle and regenerating tail of the Scincid lizard, Mabuya carinata under normal euthyroidic, hypothyroidic, and thyroxine supplemented conditions was planned and executed. To make the study more complete, levels of blood glucose too were estimated in the above said three groups of animals during various stages of tail regeneration.

MATERIALS AND METHODS

Healthy Mabuyas of both sexes obtained from Hyderabad, India and allowed to get acclimated to the laboratory conditions were kept on insect diet. Animals were subjected to chemical thyroidectomy and replacement therapy as described in Chapter I. Tails were autotomized by pinching off at about 1.5-2.0 cms distal to the vent. Animals were then sacrificed under mild anaesthesia at fixed intervals of 3, 5, 7, 10, 12, 15, 25, 40 and 60 days post-autotomy.

Blood was immediately collected by cardiac puncture. Normal or regenerating tail, as the case may be, and liver and skeletal muscle were quickly removed and homogenized in ice-cold redistilled water. A 2% homogenate was prepared for liver and skeletal muscle whereas in the case of tail a 4% homogenate was found satisfactory.

Estimations of glycogen, phosphorylase, inorganic phosphate and protein content were done according to the methods described in Chapter II.

Blood glucose estimation :

Glucose content in the blood was estimated according to the micromethod of Folin and Malmros (1929).

For each day, each tissue and each blood sample specified a total of five to seven estimations were made. The mean and standard deviation were obtained and students' 't' test was used to determine statistical significance.

RESULTS

Figs. 1-7 and Tables 1-3 depict the levels of glycogen content and phosphorylase activity in the regenerate, liver and skeletal muscle and blood glucose level in euthyroid controls, PTU treated, and T4 administered Mabuyas during various phases of tail regeneration.

Tail Glycogen and Phosphorylase :

The glycogen content of normal unautotomized tail is higher in euthyroidic (Group A) animals and lower in the PTU fed (Group B) animals. Animals replaced with T4 (Group C) showed an intermediate level. Correspondingly high phosphorylase activity was recorded in the group B animals and lower in group A animals. Again, C group animals showed an intermediate level of the enzyme activity. During tail regeneration control euthyroidic animals showed an initial depletion on the 3rd day which got soon reverted to an above normal level on the 5th day. Since then there was a continuous increase in the glycogen content in the regenerate till a maximal level was attained on the 15th day. Thereafter a continuous and gradual decrease was recorded through 25th to 40th and ultimately 60th day at which stage the more or less fully regenerated tail had a slightly above normal level of glycogen. Quite in agreement with the glycogen content is the phosphorylase activity which remains subnormal till 10th day. Since then on 15th and 25th days it showed increased levels of activity in ascending order. Ultimately phosphorylase activity returned to normal level on 60th day, with a slight subnormal level on 40th day.

The PTU fed animals showed in contrast a gradual fall in glycogen content right from tail autotomy till on 25th day when the lowest subnormal level was reached. On day 40, the

Table 1. Levels of glycogen in the regenerate, liver and muscle during tail regeneration under euthyroidic, hypothyroidic and T4 replaced condition in M. carinata.

(Values expressed as mg glycogen/100 mg of tissue weight).

Periods of tail regeneration in days	TAIL			LIVER			MUSCLE		
	Control Group (A)	PTU Group (B)	PTU + T4 Group (C)	Control Group(A)	PTU Group (B)	PTU + T4 Group (C)	Control Group (A)	PTU Group (B)	PTU + T4 Group (C)
N	0.086 ±0.014	0.063 ±0.018	0.076 ±0.010	2.34 ±0.28	3.56 ±0.72	3.30 ±0.55	0.665 ±0.040	1.31 ±0.35	0.925 ±0.055
3	0.056@ ±0.004	0.051 ±0.012	0.063 ±0.009	1.67* ±0.22	3.36 ±0.31	2.58 ±0.44	0.450⁂@ ±0.04	1.29 ±0.18	0.892 ±0.050
5	0.105 ±0.013	0.043 ±0.010	0.055* ±0.007	0.98⁂@ ±0.16	3.21 ±0.25	2.20⁂@ ±0.30	0.352⁂@ ±0.060	1.03 ±0.50	0.845 ±0.050
7	0.165* ±0.005	0.041 ±0.014	0.066 ±0.007	1.85 ±0.23	3.44 ±0.44	2.34* ±0.15	0.247⁂@ ±0.30	1.16 ±0.29	0.430⁂@ ±0.083
10	0.209⁂@ ±0.019	0.046 ±0.020	0.096 ±0.015	1.78* ±0.17	2.97 ±0.44	2.40 ±0.45	0.255⁂@ ±0.037	1.10 ±0.31	0.465⁂@ ±0.065
12	0.278⁂@ ±0.061	0.038 ±0.013	0.111* ±0.014	1.38@ ±0.15	2.97 ±0.51	1.87⁂@ ±0.18	0.277⁂@ ±0.068	0.87 ±0.19	0.350⁂@ ±0.054
15	0.305⁂@ ±0.024	0.033 ±0.017	0.125⁂ ±0.015	1.36@ ±0.18	2.58 ±0.36	1.68⁂@ ±0.15	0.300⁂@ ±0.040	0.66⁂@ ±0.09	0.282⁂@ ±0.060
25	0.266⁂@ ±0.026	0.029 ±0.010	0.111* ±0.010	2.03* ±0.24	3.36 ±0.37	1.62⁂@ ±0.46	0.357⁂@ ±0.057	0.58⁂@ ±0.14	0.335⁂@ ±0.060
40	0.190⁂@ ±0.019	0.037 ±0.020	0.103* ±0.013	2.71 ±0.36	3.57 ±0.38	1.78⁂ ±0.45	0.360⁂@ ±0.120	0.40⁂@ ±0.15	0.402⁂@ ±0.090
60	0.114 ±0.024	0.039 ±0.020	0.093 ±0.020	2.15 ±0.41	3.45 ±0.39	2.46 ±0.26	0.397⁂@ ±0.167	0.65@ ±0.21	0.820 ±0.130

* $P < 0.01$; ⁂ $P < 0.005$; @ $P < 0.025$; @ $P < 0.001$; ⁂@ $P < 0.0005$

N - Normal (Pre-autotomy state)

Table 2. Quantitative levels of Phosphorylase in the regenerate, liver and skeletal muscle during tail regeneration under euthyroidic, hypothyroidic and T4 replaced conditions in M. carinata.

(Values expressed as μg phosphorus liberated/mg protein)

Periods of tail regeneration in days	TAIL			LIVER			MUSCLE		
	Control Group (A)	PTU Group (B)	PTU + T4 Group (C)	Control Group (A)	PTU Group (B)	PTU + T4 Group (C)	Control Group (A)	PTU Group (B)	PTU + T4 Group (C)
N	8.31 ±0.64	15.2 ± 1.06	9.45 ±0.92	8.1 ±0.69	11.24 ±0.99	9.37 ±0.57	10.87 ±0.81	16.03 ±0.94	10.53 ±0.67
3	7.17 ±0.38	13.9 ±1.38	12.1 @ ±0.56 @	10.78 ±4.57	8.01 @ ±0.68 @	11.23 ±0.61	14.13 @ ±1.17 @	13.86 ±1.43	11.94 ±1.09
5	4.97 * @ ±0.38 @	16.99 ±0.87	14.01 * @ ±0.59 @	18.82 * @ ±0.59 @	9.95 ±1.24	17.64 * @ ±0.87 @	18.53 * @ ±0.9 @	13.22 ±0.83	12.5 * ±0.73
7	4.22 * @ ±0.41 @	12.83 ±2.74	11.19 ±0.91	9.35 ±0.51	9.27 @ ±0.47 @	15.98 * @ ±0.75 @	28.82 * @ ±1.77 @	10.83 * @ ±0.79 @	9.42 ±0.71
10	5.24 * @ ±0.9 @	14.77 ±1.03	8.56 ±1.0	11.2 @ ±0.51 @	13.02 ±1.5	16.43 * @ ±0.95 @	28.78 * @ ±3.81 @	16.23 ±1.47	16.09 * @ ±0.91 @
12	8.89 ±0.78	19.4 @ @ ±0.85 @ @	9.74 ±1.24	14.69 * @ ±0.81 @	11.18 ±0.77	19.3 * @ ±4.19 @	34.66 * @ ±3.66 @	22.61 * @ ±2.22 @	20.21 * @ ±1.32 @
15	14.66 * @ ±0.98 @	24.02 ±1.25	14.78 * @ ±1.2 @	14.31 * @ ±0.81 @	11.31 ±0.96	21.37 * @ ±3.66 @	39.07 * @ ±3.4 @	32.73 * @ ±1.77 @	30.99 * @ ±3.89 @
25	17.16 * @ ±1.34 @	21.4 ±0.9	21.34 * @ ±3.78 @	8.86 ±0.61	9.48 * ±1.22	26.03 * @ ±4.51 @	31.66 * @ ±3.55 @	24.74 * @ ±4.49 @	33.11 * @ ±2.22 @
40	6.64 ±0.98	25.56 * @ ±1.21 @	22.94 * @ ±2.15 @	7.47 ±0.39	10.66 ±0.71	18.18 * @ ±1.07 @	21.04 * @ ±1.46 @	40.17 * @ ±2.47 @	29.06 * @ ±1.13 @
60	9.17 ±0.61	25.69 * @ ±1.34 @	18.7 * @ ±0.97 @	8.12 ±0.69	11.57 ±0.91	11.64 ±0.61	18.71 * @ ±0.97 @	43.57 * @ ±1.86 @	21.67 * @ ±1.66 @

* P < 0.01; * P < 0.0005; @ P < 0.0025; @ P < 0.001; * @ P < 0.0005

N - Normal (Pre-autotomy state)

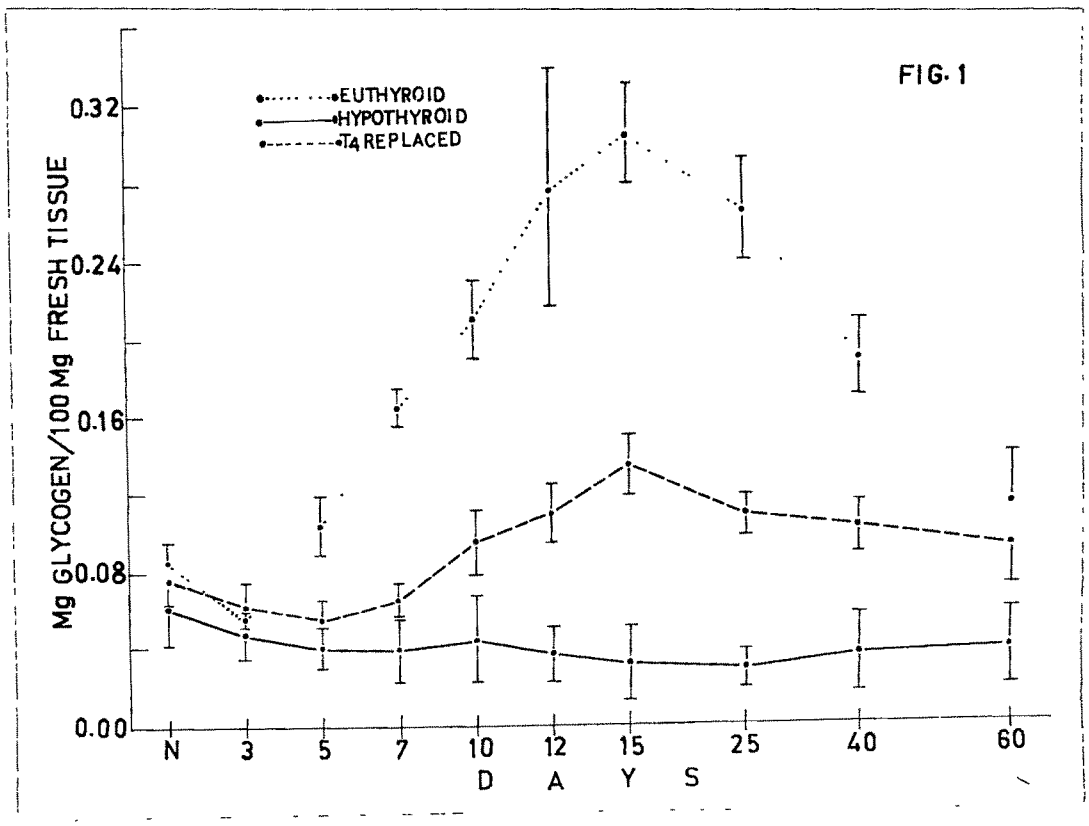

Fig. 1. Changes in glycogen content in the regenerate during tail regeneration under control euthyroidic, PTU induced hypothyroidic and T4 replaced conditions in M. carinata.

tail glycogen content in the group B animals showed a very slight increase and remained steady at that level thereafter. The corresponding phosphorylase activity depicted a level in the range of the elevated preautotomy level characteristic of this group. However, after the 10th day phosphorylase activity showed significantly increasing levels and on 40th and 60th days maximal level of enzyme activity was registered.

The glycogen content of T4 replaced group (Group C) of animals showed a pattern similar to that of the control animals. The phosphorylase activity too, though very much comparable with that of the control, showed an initial increase on the 3rd and 5th days, a fall during 7th and 10th days and a further increase during 12th to 40th days. On day 60, though the activity had shown a significant fall, it was still very much above normal.

Hepatic Glycogen and Phosphorylase activity :

In general PTU fed, and PTU + T4 treated animals showed an increased hepatic glycogen content in that order in the normal unautotomized condition. With respect to phosphorylase, activity was higher in the PTU fed animals, lower in the euthyroidic animals and intermediate in the T4 replaced animals. The hepatic glycogen content in the A group of animals showed a depletion subsequent to tail autotomy during 3rd and 5th days and on 7th day it registered an

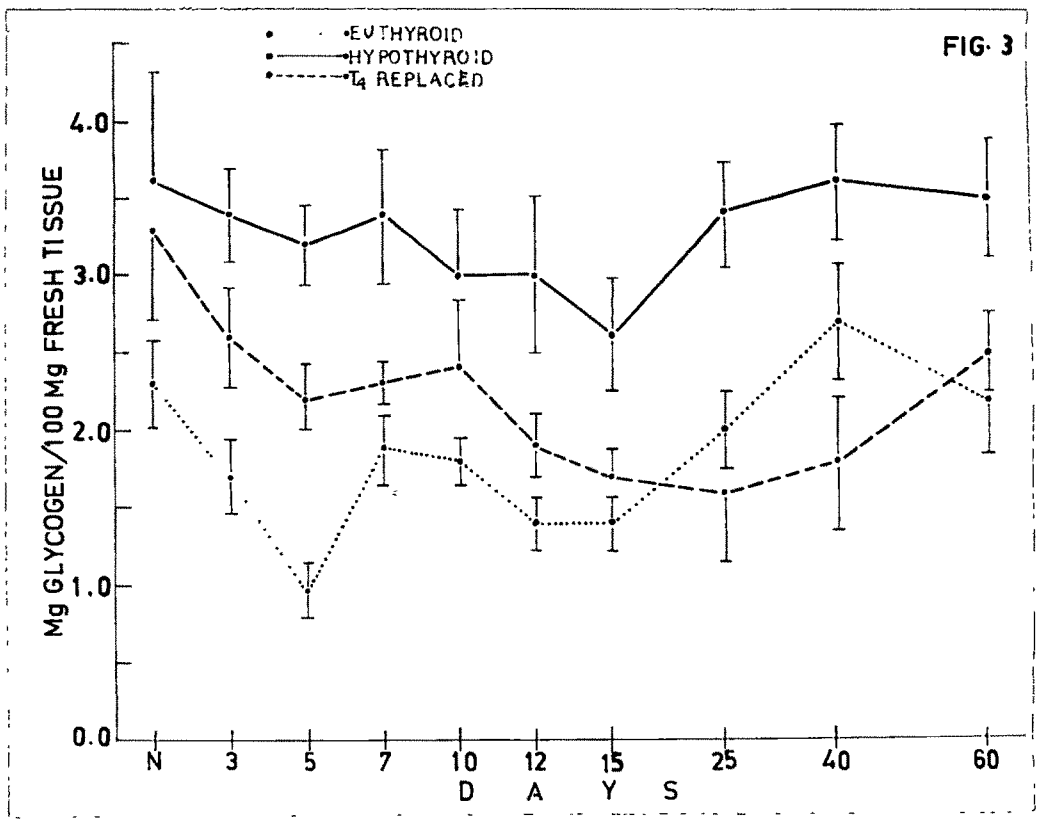

Fig. 3. Changes in glycogen content in liver during tail regeneration under control euthyroidic, PTU induced hypothyroidic and T4 replaced conditions in M. carinata.

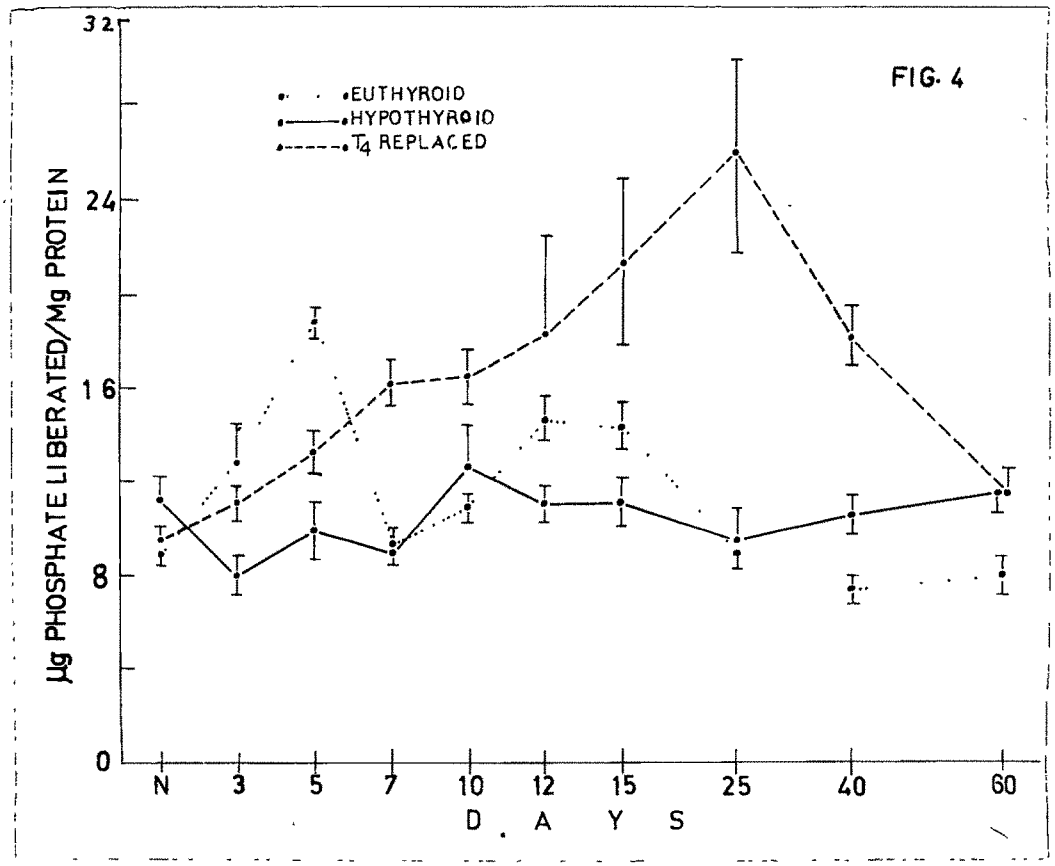

Fig. 4. Changes in phosphorylase activity in liver during tail regeneration under control euthyroidic, PTU induced hypothyroidic and T4 replaced conditions in M. carinata.

increase though the level was subnormal. There was a second phase of glycogen depletion during 10th to 15th days post-autotomy. By the 25th day the glycogen level had increased back to near normal level and remained at that level thereafter. The pattern of modulations in the phosphorylase activity was strictly on inversely reciprocal basis with the changes in the glycogen content described above.

The hepatic glycogen content of the B group of animals showed very little alteration and remained more or less in the elevated pre-autotomy level during the first week of tail regeneration. There was a marginally decreased content on 10th to 15th days post-autotomy which was however, statistically non-significant. From 25th day onwards the glycogen content was once again in the same range recorded in the first week. Similarly the quantitative level of phosphorylase activity too showed very little alterations and remained in the activity range of the preautotomy level characteristic of these animals. In the immediate post-autotomy periods i.e. during 3rd and 5th days the enzyme activity tended to be slightly subnormal.

The glycogen content of liver in the C group of animals showed variations ; however, the changes were of a continuous pattern devoid of the modulations shown by the A group of animals. Accordingly the glycogen content depicted an immediate decrease on the 3rd day post-autotomy and this level was more or less maintained till the 10th day, following which there

was a further gradual decrease during 12th, 15th and 25th days of tail regeneration. Subsequently during the 40th and 60th days a tendency towards a gradual increase in glycogen content was recorded. The corresponding changes in the phosphorylase activity showed an exactly identical pattern as that observed in the control animals during the first 12 days post-autotomy. However, unlike in controls the enzyme activity continued to increase even on the 15th and 25th days in this group of animals and thus registered a maximal level on the 40th day. Since then by day 60, the enzyme activity showed a considerable drop towards the normal level though it was still above normal.

Muscle Glycogen and Phosphorylase :

The hypothyroid group of animals once again showed an increased muscle glycogen content as compared to the control animals and intermediate level in the T4 replaced group of animals in the unautotomized condition. Just as in the case of liver, muscle phosphorylase activity also was elevated in the hypothyroid animals as compared to euthyroid and T4 replaced group of animals.

The muscle glycogen too showed alterations during tail regeneration and was characterized by a gradual decrease during the first 10 days and a very gradual increase during 12th to 60th days in the euthyroidic group of animals. Still the level

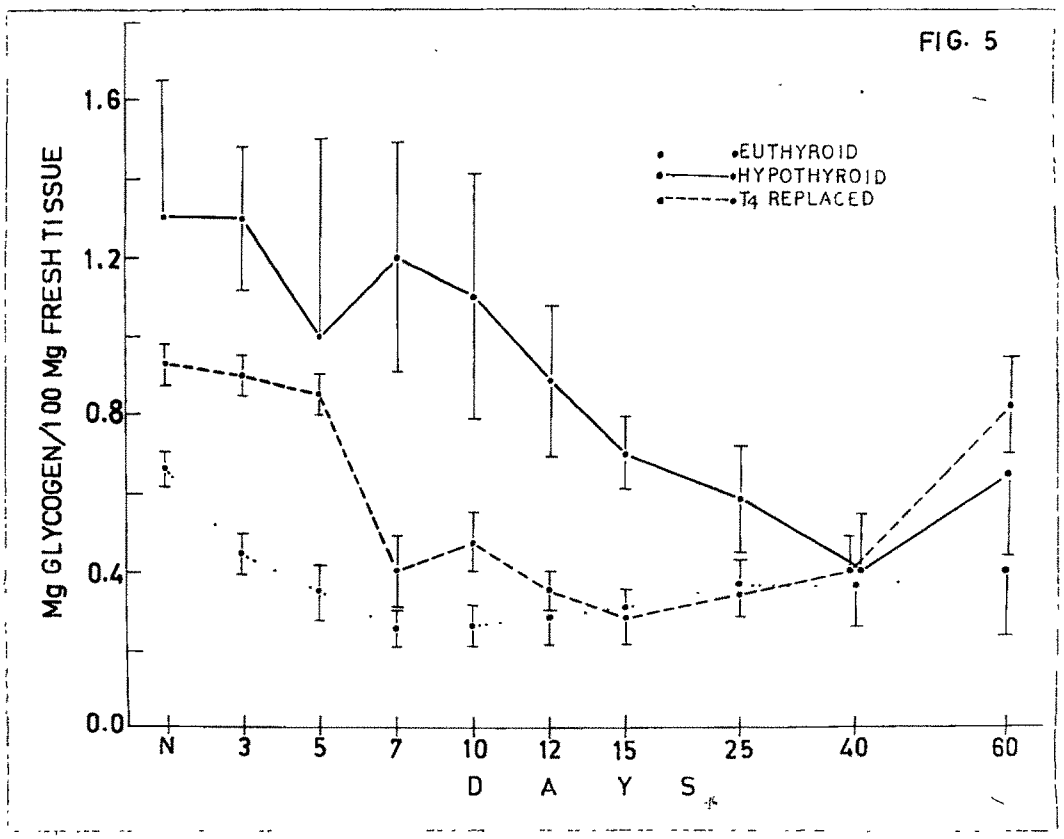

Fig. 5. Changes in glycogen content in muscle during tail regeneration under control euthyroidic, PTU induced hypothyroidic and T4 replaced conditions in M. carinata.

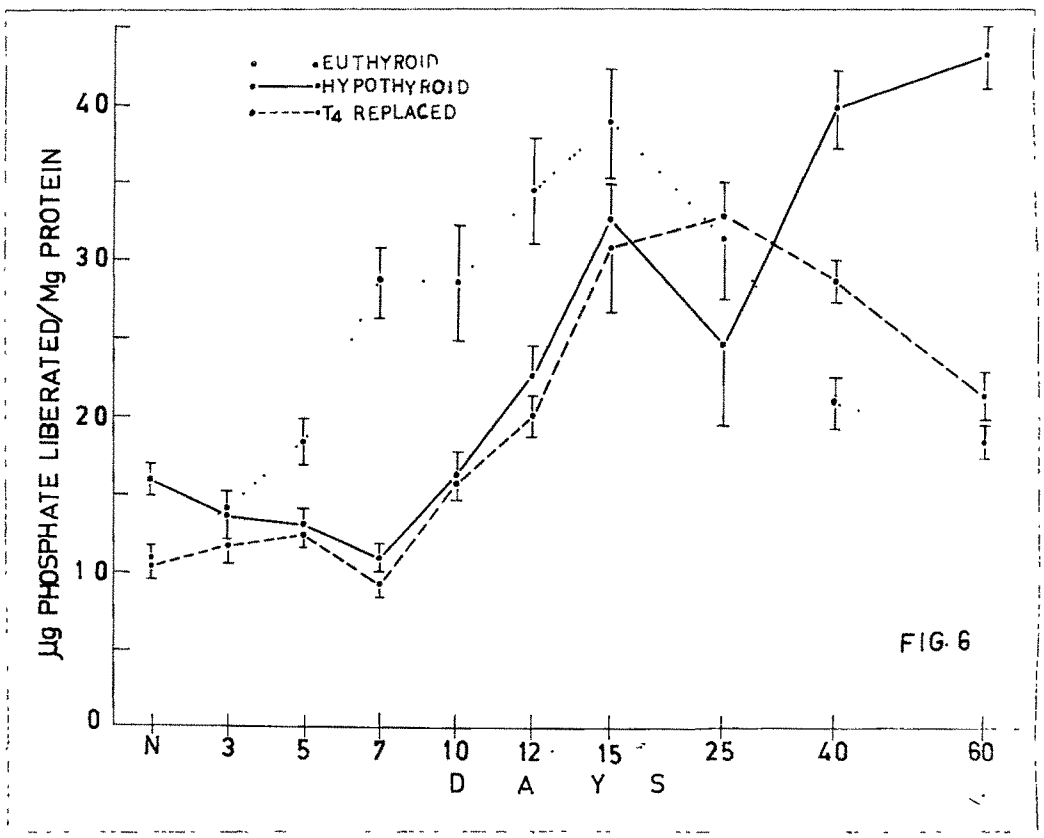

Fig. 6. Changes in phosphorylase activity in muscle during tail regeneration under control euthyroidic, PTU induced hypothyroidic and T4 replaced conditions in M. carinata.

as it stood on the 60th day is very significantly below normal. In perfect accordance with glycogen content the phosphorylase activity showed a continuous increase during the first 15 days and a gradual decrease from the 25th day onwards. Once again the level of the enzyme activity on the 60th day was significantly supranormal.

The muscle glycogen content in the hypothyroid group of animals showed a slow decreasing trend during the first 10 days and a sharper decreasing trend during 12th to 40th days of tail regeneration. On day 60, the glycogen level tended to show a slightly increased level. The muscle phosphorylase activity in hypothyroid group of animals tended to show a slight decrease from the elevated pre-autotomy level during the first 7 days and thereafter the enzyme activity registered significantly increasing levels from 12th to 60th days.

The alterations in glycogen content and phosphorylase activity in the skeletal muscle of T4 replaced group of animals were very much parallel to the changes observed in the control group of animals; the only difference being with respect to the individual values.

Blood Glucose :

The blood glucose level was higher in the group A

Table 3. Quantitative levels of blood glucose during tail regeneration under euthyroidic, hypothyroidic and T4 replaced conditions in M. carinata.

(Values are expressed in mg glucose/100 ml of blood)

Periods of tail regeneration in days	G L U C O S E		
	Control Group (A)	PTU Group (B)	PTU + T4 Group (C)
N	94.83 ±1.04	81.45 ±3.60	91.2 ±1.43
3	123.75*@ ±3.41	92.83* ±4.55	107.85*@ ±3.37
5	108.48*@ ±2.81	104.03*@ ±4.12	123.2 *@ ±7.25
7	103.23 ±5.34	85.63 ±2.06	126.65*@ ±3.07
10	120.48*@ ±2.33	87.05 ±5.65	131.65*@ ±5.84
12	118.03*@ ±6.41	87.7 ±2.55	140.15*@ ±5.07
15	130.03*@ ±6.66	97.05@ ±4.91	141.68*@ ±4.21
25	101.93* ±3.59	101.78*@ ±4.18	136.95*@ ±3.71
40	103.43 ±7.25	89.28 ±4.72	119.51*@ ±4.21
60	104.2 ±3.29	87.41 ±2.61	114.55*@ ±7.54

* $P < 0.01$; * $P < 0.005$; @ $P < 0.0025$,
@ $P < 0.001$; *@ $P < 0.0005$.

N - Normal (Pre-autotomy state)

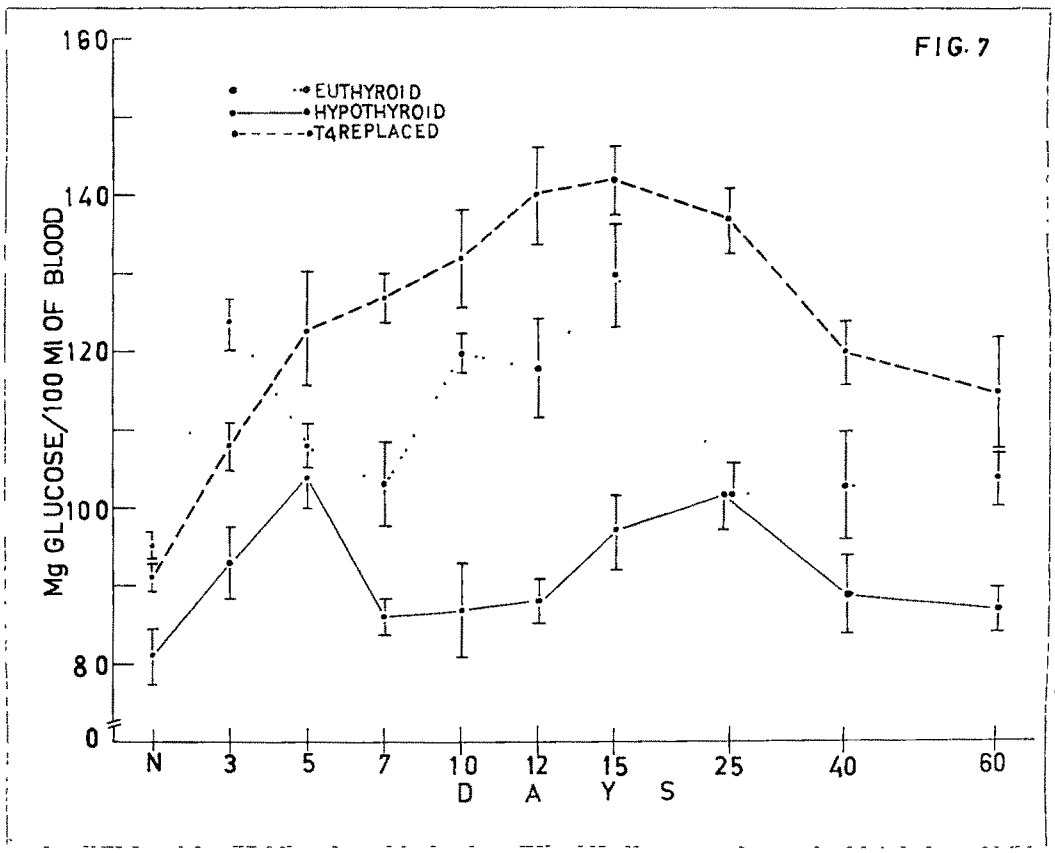

Fig. 7. Changes in blood glucose level during tail regeneration under control euthyroidic, PTU induced hypothyroidic and T4 replaced conditions in M. carinata.

animals and lower in the group B animals. T4 replaced group of animals once again showed an intermediate condition. Both A and C groups of animals showed significant alterations in the blood glucose level during tail regeneration. Whereas the A group of animals showed an increase on the 3rd day, a decrease on the 5th and 7th days and again increase during 10th to 15th days and a subsequent fall towards normal level, the C group of animals showed a continuous increase right from 3rd day till the 15th day. Thereafter the level of glucose in the blood in this group of animals showed a decreasing trend but remaining above normal during 25th, 40th and 60th days. In contrast the B group of animals tended to maintain the preautotomy level of glucose except for a slight increase initially during 3rd and 5th days and a second increase during the 15th and 25th days.

DISCUSSION

In a previous chapter, influence of PTU induced hypothyroidism on tail regeneration in Mabuya carinata, in the form of an initial delay in the formation of a blastema and later inhibition of the further development of the blastema was reported. Further, abolition of the negative effects of thyroid hormone deficiency was also reported by T4 replacement in PTU fed animals. Now we show that hypothyroidism affects carbohydrate metabolism by inducing

alterations in the steady state levels of glycogen, phosphorylase and blood glucose. A tendency towards rectification of these alterations by T4 replacement is also evident. Increased hepatic glycogen content and lowered blood glucose level - observations similar to the present study, have been reported by other investigators too in different vertebrate groups under hypothyroidic state (see Hoch, 1974; Suryavanshi and Dubeyer, 1977; Raheja and Linscheer, 1978). The present study reports in addition the effect of hypothyroidic condition in Mabuya carinata on muscle and tail glycogen content in the form of an increase, and also on phosphorylase activity of liver, muscle and tail which is also uniformly increased in all the three tissues. Within the framework of increasing tissue glycogen content, increased phosphorylase activity is apparently contradictory. Possible explanation that could be offered in this connection is the activation of adenylate cyclase and the resultant increase in cAMP and phosphorylase activation under the influence of the elevated blood TSH concentration to be expected under PTU induced hypothyroidism. Substantial evidence in this respect is available on TSH mediated adenylate cyclase activation and cAMP formation in thyroid tissue (see Tong, 1974; Bidey et al., 1980). Though there is an increased phosphorylase activity, the glycogenolytic action of the enzyme is presumably inhibited by the absence of

thyroxine as is evident by the increased tissue glycogen content. The increased glycogen content also reflects the increased glycogenesis known to occur under hypothyroidism (see Hoch, 1974).

Previous studies from this laboratory have shown involvement of hepatic glycogen and blood glucose in the regenerative process of Mabuya carinata (Shah et al., 1977 b). Further studies on this line confirmed not only these observations but also demonstrated the involvement of muscle glycogen as well as adaptive alterations in systemic phosphorylase activity (Chapter II). The present study on euthyroidic control animals substantiates the systemic and local alterations in glycogen content, phosphorylase activity, and blood glucose levels noted to occur during tail regeneration in the above referred studies. The unaltered hepatic glycogen content and phosphorylase activity and inhibited glycogenesis in the tail and inability of the blood glucose level to increase in the PTU fed animals as a unit could be one of the reasons for the observed inhibition of the tail regeneration. The reversal of these metabolic features to a near normal pattern and the removal of the blockade effect on tail regeneration by T4 replacement in PTU fed lizards corroborates the above fact. Similarly the increased glycogen content occurring in the regenerate during normal tail regeneration is also abolished in the hypothyroidic condition with the caudal glycogen level

remaining subnormal althroughout, essentially due to elevated phosphorylase activity. In the T4 replaced group of animals after an initial delay, increase in caudal glycogen content was noted to occur though the amount of glycogen was significantly less as compared to the controls. This is probably due to the comparatively higher levels of caudal phosphorylase activity.

From the Figs. 1-7 and Tables 1 to 3 it becomes clear that T4 given animals tended to show the euthyroidic pattern of changes of glycogen and phosphorylase in muscle though the attainment of peak modulations was slightly delayed. However, the hypothyroidic condition served to lead towards significantly increased levels of phosphorylase activity in both muscle and tail after about 10th day post-autotomy with the levels during the first 10 days being slightly lower (statistically insignificant), from the elevated levels characteristic of the unautotomized condition. The data also reveals an initial slow depletion and further pronounced depletion of muscle glycogen during the later phases of regeneration. Clearly, unlike hepatic glycogen, muscle glycogen depletion is not inhibited by lack of thyroid hormone. This might be due to the differing hormonal control of glycogenolysis in the two systems. Apparently, in the present context hypothyroidism renders glucagon mediated hepatic glycogenolysis inoperative, while catecholamine

induced muscle glycogenolysis remains still operative. Interestingly, Moelsen and Sonne (1976) and Sperling et al., (1980) have demonstrated an increase and decrease in the number of receptors in adipose tissue and liver under hyperthyroidic and hypothyroidic conditions respectively. Though this might explain the suppression of liver glycogenolysis observed herein, persistence of muscle glycogen loss, however, appears to be contradictory to the reported depression of catecholamine induced muscle glycogenolysis under hypothyroidic conditions (see Hoch, 1974; Bray and Jacobs, 1974). It is quite likely that in reptiles though thyroxine may have a permissive influence on catecholamine action on muscle glycogen, absence of thyroid hormone may not necessarily inhibit the action. This is evident from the observed alterations in muscle glycogen in the three groups of animals subsequent to tail autotomy. Besides these, a couple of puzzling observations are the continuously increasing level of phosphorylase activity in both tail and muscle uptill 60 days post-autotomy, and the increasing glycogen content of both the organs during 40th to 60th days in spite of the increase in phosphorylase activity. These may be the off-shoot of altered hormonal setup induced due to tail autotomy and the imbalanced interaction thereof in the absence of thyroxine which might involve TSH and TRH. Moreover, the glycogen synthesis observed might also indicate

the operation of other hormonal factors in controlling carbohydrate metabolism secondarily.

Finally, the maintainence of subnormal steady level of blood glucose in hypothyroid animals is mainly due to the reduced glycogenolysis paralleled by reduced peripheral utilization of glucose. Supporting evidence for this comes from the reported positive influence of thyroxine on potentiating the action of insulin on tissue utilization of glucose and the insensitiveness of tissues to insulin in hypothroidic condition(Cohen, 1957; Hagen, 1960; West et al., 1975). Besides, Rall et al. (1964) have shown the depressive effect of hypothyroidism on removal of exogenously administered glucose from blood. Another observation that merits consideration is the similarity in estimated values of glycogen and phosphorylase in T4 replaced animals with that of the PTU fed group. Though the pattern of modulations resembles the euthyroidic group, the dissimilarity in values is probably due to the depressive role of PTU on peripheral T4-T3 conversion which restricts the level of metabolically active T3 to the minimum in the PTU + T4 treated animals; hence the similarity to PTU induced hypothyroidic group. It may be concluded from the present observations that hypothyroidism induced alteration of carbohydrate metabolism from the fixed pattern associated with normal tail regeneration

might be one of the factors for the inhibition of tail regeneration in Mabuya carinata. This might thus represent a probable secondary systemic influence of thyroxine as both primary in loco and secondary systemic actions of thyroxine on tail regeneration in M. carinata have been hinted at previously (Chapter I).

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