SUMMARY

CHAPTER I :

Influence of two different breeding seasons as well as thyroid hormone on tail regeneration in the Scincid lizard, Mabuya carinata has been evaluated by measuring the rate of growth of the regenerate at various time intervals. The analysis has revealed no apparent seasonal alteration in the final length attained at the end of 60 days. However, a differential per day rate of growth is indicated during the late non-breeding and early breeding phases. A better per day rate of growth in the first fortnight of tail regeneration corresponding to blastema formation and early differentiation was noticeable in the recrudescent period, while the growth rate during 25 to 60 days of tail regeneration which corresponds to late differentiation and growth was faster in the breeding period. These observations are correlated with differential physiological and endocrine status characteristic of the two seasons. With reference to thyroid influence on regeneration, hypothyroidic animals recorded a 71% retardation of regenerative ability as compared to euthyroidic animals which was however rectified by T4 replacement. Both direct as well as indirect modes of action of thyroxine on lizard tail regeneration is discussed.

CHAPTER II :

Apart from local responses, systemic involvements too have been implicated during tail regeneration in lizards. Hence in the present study alterations in carbohydrate metabolism have been assessed at the local site as well as in the visceral organs (liver and skeletal muscle) by quantitative evaluation of glycogen content and phosphorylase activity during tail regeneration in the Scincid lizard, Mabuya carinata. Significant and correlatory alterations in both the parameters could be observed in all the three tissues during various phases of tail regeneration. Increased phosphorylase activity and depletion in glycogen content are the features during the early half of regeneration in liver and skeletal muscle; whence the tail depicted decreased phosphorylase activity and increased glycogen content. The later half of regeneration was marked by a reversed trend of changes. These observations have led to the conclusion of dependence of the process of regeneration on systemic stores of carbohydrates, especially during the early periods i.e., wound healing, blastema formation and early differentiation. With the commencement of differentiation the regenerating system is purported to make use of its own carbohydrate reserves. Further the alterations in hepatic and muscle glycogen stores have also been discussed in terms of related and essential metabolic adjustments of the body as a whole in meeting the exigencies of tail regeneration.

CHAPTER III :

Local and systemic alterations in lactate and succinate dehydrogenases (LDH and SDH) have been carried out quantitatively during tail regeneration in the scincid lizard, Mabuya carinata. Increased levels of LDH activity during the first five days followed by decreased levels upto about 25 days of tail regeneration were noted. Supranormal levels of SDH were however, the feature between 7th and 25th days of tail regeneration. Post-differentiation periods of regeneration depicted gradual settling down of the enzyme activities to the respective pre-autotomic state. These changes are construed to indicate a generalized in loco and systemic anaerobiosis during the wound healing period, active aerobic metabolism during blastemic and differentiation phases and reversal to the normal pattern during the growth period of tail regeneration.

CHAPTER IV :

Electrophoretic separation of lactate dehydrogenase isozymes together with the quantitative evaluation of muscle type (A) and heart type (B) subunits were carried out, during different phases of tail regeneration, in the regenerate as well as liver and skeletal muscle of the scincid lizard, Mabuya carinata. Both, the electrophoretic pattern of isozymes as well as the analogue ratio (ratio of B type to A type subunits) indicate increased anaerobiosis during the period of

wound healing in all the three tissues. Blastemic and early differentiation phases are marked by an aerobic pattern of metabolic process as revealed by the distinctly increased proportion of B subunits as well as the appearance of well defined fast moving (anodic) LDH 1 and 2 isozymes. Since then, from differentiation phase onwards there was a gradual reversal of this pattern, which finally with the completion of regeneration got set to the original anaerobic pattern of isozymic profile as well as analogue ratio, characteristic of the corresponding tissues of normal animals (with intact tails). These changes in LDH activity are discussed in terms of the overall subcellular metabolic adaptations pervading the animal body in response to the regenerative process.

CHAPTER V :

Quantitative alterations in total protein content and activity levels of alanine and aspartate aminotransferases (GPT and GOT) in the regenrate, liver, muscle and serum of the lizard, Mabuya carinata have been presently evaluated. Increased positive nitrogen balance marked by supranormal protein content in all the three tissues is recorded. The two transaminases were noted to remain subnormal in the regenerate while they recorded supranormal levels in the skeletal muscle during tail regeneration. This has been taken to indicate a dependence by the regenerate on systemic

source(s) of amino acids. A detailed discussion on these aspects is undertaken in the text.

CHAPTER VI :

Changes in the levels of cyclic AMP in tail regenerate, liver and skeletal muscle of the Scincid lizard, Mabuya carinata have been monitored indirectly by assaying the activity of cyclic AMP phosphodiesterase during various phases of tail regeneration. Though changes in the levels of activity of this enzyme have been observed in all the three tissues, the regenerating tail depicted maximum variation. During the first week of regeneration (corresponding to the formation of blastema) there was a significant decrease in phosphodiesterase activity in the regenerate, followed by an increase to the normal level by 10th day and remaining so till 25th day. On day 40, there was again a significant fall in enzyme activity which by day 60, however, settled down to the normal level. These changes in the tail regenerate are correlated with the changing levels of cyclic AMP associated with cell proliferation, differentiation and growth. With reference to liver and skeletal muscle, though both did show fluctuations in enzyme activity, the changes in the liver were statistically non-significant thus indicating the maintainence of a steady level of cyclic AMP in this organ. However, in the case of skeletal muscle, there were two

phases of significant decrease in phosphodiesterase activity; an early short lived one during the first 5 days post-autotomy and a second decrease during the terminal phases of regeneration lasting from 25th to 60th days post-autotomy. The increased level of cyclic AMP in skeletal muscle during these two periods of tail regeneration is rather interesting and may have to be looked upon from the point of view of significant metabolic upheavals affecting this tissue.

CHAPTER VII :

A quantitative evaluation of acetyl (AChE) and non-specific (NspChE) cholinesterases in the tail regenerate, liver and skeletal muscle was undertaken during various time periods of tail regeneration in Mabuya carinata. The level of AChE in all the three tissues depicted a parallel decrease during the wound healing and pre-blastemic phases and a parallel maximal level during early differentiation phase. Since then by a gradual decrease during the late differentiation and growth phases it settled down to the normal level in the fully regenerated stage. A similar pattern was depicted by the NspChE too, except for the tremendous increase noted during the 3rd day post-autotomy in the tail. Whereas the high level of NspChE during the early differentiation period may be correlated with lipid metabolism, the concomitant peak level of AChE may be correlated with metabolic alterations

and macromolecular synthesis characteristic of regeneration probably by bringing about an ionic flux. The most significant observation of a short living nine fold increase in NspChE activity in the regenerate on 3rd day post-autotomy is purported to be somehow involved, either directly or indirectly, in the as yet unknown mysterious events associated with the process of initiation of regeneration.

CHAPTER VIII :

Quantitative evaluation of glycogen content and phosphorylase activity in liver, muscle and regenerate together with blood glucose was undertaken during various periods of tail regeneration in the Scincid lizard, Mabuya carinata under different functional status of thyroid gland. Hypothyroidic animals failed to depict depletion of hepatic and muscle glycogen and elevation in blood glucose during the first fortnight of tail regeneration characteristic of euthyroidic controls. Correspondingly phosphorylase activity too of these organs was affected. Changes in the caudal glycogen content and phosphorylase activity too were distinctly different in the hypothyroid lizards. The differences were however, rectified by T4 replacement. These changes are construed to denote the probable indirect mode of action of thyroid on tail regeneration by its influence on regeneration associated adaptive systemic carbohydrate metabolism.

CHAPTER IX :

Quantitative alterations in succinate INT reductase (SDH) were carried out in the tail regenerate, liver and skeletal muscle of the Scincid lizard, Mabuya carinata under euthyroidic, hypothyroidic and T4 replaced conditions. Increased enzyme activity in loco as well as systemically characteristic of tail regeneration during blastemic and differentiation phases was shown by euthyroidic and T4 replaced groups of animals. Hypothyroidic animals, failed to show this increase in enzyme activity. It is construed that the inability to raise up the level of oxidative metabolism might be one of the factors for the observed inhibition of tail regeneration in chemically thyroidectomized lizards. Moreover, the observation of maintainence of slightly below normal basal levels of the enzyme activity in all the three tissues suggests the existence of other points of control of SDH activity in lizards with thyroxine, nevertheless having a permissive role.

CHAPTER X :

Thyroid and protein metabolism in relation to tail regeneration have been evaluated by quantitative analysis of total protein content and activity levels of alanine and aspartate aminotransferases (GPT and GOT) in the regenerate,

liver and skeletal muscle of normal, PTU fed and T4 replaced lizards, Mabuya carinata. Protein content of all the three tissues was noted to increase in euthyroidic and T4 replaced animals, while such an increase was not evident in hypothyroid lizards. However, GPT and GOT activities were not much affected by hypothyroidism. Inability of hypothyroid animals to bring about positive nitrogen balance has been considered to be one of the factors responsible for the observed inhibition of tail regeneration. Moreover, the observed similar pattern of GPT and GOT activities in all the three groups of animals overrules the possible regulatory role of thyroxine on transaminases, though a permissive role has nevertheless been considered possible based on the results obtained and discussed in detail in the text.

CHAPTER XI :

In order to evaluate the functional roles of thyroid and cAMP on lacertilian tail regeneration, cAMP phosphodiesterase, the hydrolytic enzyme of cAMP, was assayed in the tail regenerate, liver and skeletal muscle of control (A), chemically thyroidectomized (B) and thyroidectomized and T4 replaced (C) groups of animals during various periods of tail regeneration. The results obtained indicate elevated above normal levels of phosphodiesterase in all the three tissues

of group B animals. Animals of group C depicted an intermediate level between the controls (A) and experimentals (B). These observations tend to indicate a possible regulatory role of thyroxine in maintaining optimum levels of phosphodiesterase activity in vertebrate tissues and hence cAMP concentration. Presumably, thyroxine induced elevations in cAMP may be mediated by its suppressive action on phosphodiesterase. The retardation in regeneration observable in the hypothyroid group (B) of animals may thus be correlated with the low levels of tissue cAMP. However, the operation of other influencing factors as well, on phosphodiesterase during regeneration can be surmised from the observed tendency to exhibit similar pattern of phase specific modulations in enzyme activity. All these observations have been discussed in the text in terms of phase specific involvement of cAMP in regeneration as well as its role in other metabolic aspects and the possible mode of indirect control exerted by thyroxine on lacertilian tail regeneration.

CHAPTER XII :

Levels of hepatic and renal ascorbic acid contents have been estimated in euthyroidic, hypothyroidic and T4 replaced conditions during tail regeneration in the Scincid

lizard, Mabuya carinata. Pattern of changes in euthyroid lizards seems to indicate significant involvement of systemic stores of ascorbic acid in meeting the exigencies of tail regeneration. Excepting for the modulations to be slightly late, the pattern of changes in T4 replaced animals was more or less similar to that of the euthyroidic controls. In hypothyroid group of animals the pattern of alterations of hepatic and renal ascorbic acid contents was parallel and quite distinct from that of euthyroid and T4 replaced groups. A possible non-utilization of ascorbic acid by hypothyroid animals have been inferred. A detailed discussion on this aspect as well as the possible regulation of systemic ascorbic acid turnover is attempted in the text.

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