

EFFECTS OF TEMPERATURE ON GROWTH IN THE REGENERATING TAIL OF THE SCINCID LIZARD, *MABUYA STRIATA*

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ABSTRACT

Experiments on effects of temperature on growth in the regenerating tail of *Mabuya striata*, artificially autotomized, suggest a faster rate of regeneration during hot weather than cold weather.

INTRODUCTION

Limbs of newts and salamanders can generally be replaced both in adults and larvae. Amongst reptiles, however, regeneration of limbs is not a common phenomenon. In lizards, for example, limb regeneration occurs only as abnormal outgrowths (Goss 1969). A few lizards are known to restore lost tails, and the regenerated tail is almost a true substitute for the original tail, not only with respect to structure and function, but also to size. The period taken by a regenerating tail to achieve its original length varies from species to species as well as individual to individual. Such variation in the rate of growth of regenerates seems to be correlated with several factors, viz. the amount of tail autotomized, pressure applied at the time of autotomy, temperature, humidity, hormonal levels and diet (Moffat & Bellairs 1964; Bryant & Bellairs 1967; Licht 1967; Maderson & Licht 1968; Shah & Chakko 1968; Balinsky 1970; Magon 1975a, 1975b). Much more experimentation will be necessary, however, before the effects of these factors can be quantified. The experiments described in this paper examine the influence of temperature on the growth in the regenerating tail of the scincid lizard, *Mabuya striata*, during the different seasons of the year, viz. hot season: December - February; rainy season: March - May & September - November; and cold season: June - August.

MATERIAL AND METHODS

Adult *M. striata* of both sexes with original tails, i.e. normal tails, collected from Kenyatta University College campus, and maintained in the terraria in the laboratory, were used as experimental animals. These animals were collected monthly and the data were analysed

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seasonally. Daily maximum and minimum temperature was recorded in the terraria between December 1974 and November 1975. The mean temperatures were then grouped according to the seasons, viz. hot, rainy and cold (Table 1).

The body length of *M. striata* was measured from snout to tip of tail, and tail length from vent to tip of the tail, in order to select animals of same size, i.e. average total length of 15 cm. The tail autotomy was induced by pinching off tails leaving two or three basal segments intact as stumps. The average length of the autotomized tail was 6,5 cm. The experimental lizards were regularly fed on an unlimited supply of cockroaches. The growth in centimetres of the regenerating tail was determined at 10-day intervals starting from the day of autotomy until attainment of the original length, i.e. 15 cm.

For histological observations, regenerating tails with at least one or two segments of the original tail stump were autotomized and fixed in Bouin's fluid for about 24 hours at room temperature. Sections of 10 to 15 μm were cut on a freezing microtome and stained in haematoxylin-eosin.

TABLE I

Time required (in days) for the different phases of the tail regeneration in the scincid lizard, *Mabuya striata*.

	<i>Hot season</i> (<i>Dec.-Feb.</i>)	<i>Rainy season</i> (<i>Mar.-May &</i> <i>Sept.-Nov.</i>)	<i>Cold season</i> (<i>Jun.-Aug.</i>)
Wound-healing	3	5	9
Blastema	6	9	14
Differentiation	15	18	29
Growth	20	35	50
Fully-regenerated tail	50	70	90
Climatic conditions:			
Temperature (°)			
Mean maximum*	31	22,8	17,2
Mean minimum*	14,6	16,7	10,2

* Daily maximum and minimum temperature was recorded in the terraria from December 1974 to November 1975. The mean temperatures were then grouped according to the seasons, viz. hot, rainy and cold.

OBSERVATIONS

The data for number of days taken by the tail to reach different stages of regeneration are presented in Table 1. For convenience of description the entire period of tail regeneration has been divided histologically into four different periods: wound-closing stage, blastema stage, differentiation stage and growth stage. However, it must be borne in mind that these phases of regeneration are arbitrarily defined. The process of regeneration is in fact a continuous one.

1. *Wound-closing stage.* In this period the first noticeable event to occur is an accumulation of blood cells in the clot on the cut surface of the tail stump. This is followed by formation of a new epithelium and migration of cells from the cut edge of the original tail epidermis to form a cover over the cut area of the wound. Injured tissues get detached from the cut surface as soon as new epithelium is formed. Another salient feature of this stage is that the nerve cord, vertebral column, muscle and fat layers at the cut end of the original tail show a tendency for de-differentiation.
2. *Blastema stage.* During this stage, more epithelial cells are formed and thus the wound epithelium increases in thickness. There is also a formation of cone, regeneration bud or blastema, at the regenerating end of the cut tail. Further, two different types of cells could be differentiated at this stage: (1) the epithelial cells which form the outer margin of the blastema, and (2) the undifferentiated mesenchyme cells which occupy the core of the blastema cone. This stage is characterized by a state of active cell division.
3. *Differentiation stage.* Differentiation in the regenerating tail proceeds proximo-distally. The tendency for differentiation of the epidermis is seen in six-day old regenerates. Various types of epidermal cells, viz. cells of malphigian layers and layers of alpha and beta cells are well-formed by the 15th day of regeneration. By this time only at the base of the regenerate is the lamellar pattern (formation of dermal papillae) observed in the skin. By the 20th day, the dermal papillae have increased in size, pushing the epidermis, with scale as the projecting body. In the dermis region of the regenerating tail, the skin is mainly composed of fibroblasts. By the 30th day, differentiation of fibroblasts into fibrocytes is completed and the latter occupy the dermis. The connective tissue is derived from the fibrocytes occurring at the base of the dermis.
The mesenchyme cells adjacent to the original muscle layers in the tail stump differentiate into myoblasts, and those in the plane of the vertebral column into chondroblasts. The myoblasts later transform into myofibres via myocytes, and chondroblasts into chondrocytes.
4. *Growth period.* During this period, all the differentiated tissues of the regenerate start to grow and finally attain morphological and physiological maturity.

Effect of temperature

The growth in centimetres of the regenerating tail of *M. striata* in the different seasons of the year 1974-75 is summarized in Table 2.

During the hot season, the autotomy wound healed in 72 hours, but during the rainy season and cold season, the wound-healing phase lasted until the 5th and 9th day respectively. The first external signs of a regeneration bud, a blastema, were evident on the 6th day during the hot season, the 9th day in the rainy season and the 14th day in the cold season. The histological characterization of the differentiation phase, as discussed earlier in this paper, took place on the 15th, 18th and 29th day respectively in the three seasons. After this, there was a period of rapid growth with attainment of approximately original length in about 50 days in the hot season, 70 days in the rainy season and 90 days in the cold season. The time interval between the different phases of tail regeneration during the three seasons, however, varied appreciably only between growth and fully-regenerated phases.

TABLE 2

Growth in cm of the regenerating tail of the scincid lizard, *Mabuya striata*, in different seasons of the year 1974-1975.

<i>Days after tail autotomy</i>	<i>Hot season (Dec.-Feb.)</i>	<i>Rainy season (Mar.-May & Sept.-Nov.)</i>	<i>Cold season (Jun.-Aug.)</i>
Wound-healing	3 days	5 days	9 days
10th day	0,77 ± 0,12*	0,46 ± 0,17*	0,14 ± 0,19*
20th day	2,11 ± 0,15	1,54 ± 0,18	0,64 ± 0,16
30th day	3,80 ± 0,18	2,90 ± 0,11	0,87 ± 0,19
40th day	5,48 ± 0,14	3,88 ± 0,21	1,56 ± 0,15
50th day	6,49 ± 0,15	4,76 ± 0,12	2,17 ± 0,17
60th day	**	5,73 ± 0,19	3,09 ± 0,23
70th day	**	6,46 ± 0,22	4,13 ± 0,19
80th day	**	***	5,24 ± 0,26
90th day	**	***	6,47 ± 0,22

* Means ± S.D. (50 animals per season).

** No change in growth after 60th day of autotomy.

*** No change in growth after 80th day of autotomy.

DISCUSSION

From December to February, when the mean maximum temperature was around 31°C, the wound-healing phase appeared within 72 hours after autotomy and the regenerate attained its original length by about 50 days, as compared with 70 and 90 days respectively in the rainy and cold seasons (Table 1). It is suggested that temperature has a profound influence on the early phase of regeneration, *i.e.* the wound-healing stage. These data are in agreement with reports of Barfurth (1891) who found that anuran tadpole tail regeneration was two to two-and-a-half times more rapid at 28°C than at 14°C. In axolotls, regeneration proceeds more effectively at 25°C (Belkin 1934). Regeneration studies on *Anolis carolensis* (Maderson & Licht 1968), *Hemidactylus flaviviridis* (Magon 1975a) and *Planaria torva* (Balinsky 1970) support the contention that temperature influences regenerative processes.

The delay in regeneration in the rainy season and cold season when the mean maximum temperatures were 22,8°C and 17,2°C respectively, may be due to several factors other than low temperature. A similar delay in the tail regeneration of *H. flaviviridis* during the monsoon and winter has been reported by Magon (1975a). It has been suggested by Schmidt (1968) that the rate of enzymatic reactions characteristic of the blastema is reduced at low temperature. The same author also suggested that cold interferes with the metabolic activities in the regenerating forelimb of the adult newt, *Diemictylus viridescens*.

In conclusion, it is difficult to say exactly how temperature influences only two phases of tail regeneration in *M. striata*, *viz.* wound-healing and growth (Table 1). If temperature favours enzymatic reaction (Schmidt 1968), and in turn, control of metabolic activities, then the major variations should have been during the blastema and differentiation phases. However, such variations were not noted in spite of the fact that the blastema phase is characterized by a state of active cell division and that there is an increased glycolytic metabolism during the period of functional differentiation (Magon 1977). Thus it is tempting to speculate that other factors apart from temperature might also play an important role in promoting tail regeneration in *M. striata*.

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