CHAPTER 13

UNILATERAL ADRENALECTOMY AND TAIL REGENERATION IN THE GEKKONID LIZARD, HEMIDACTYLUS FLAVIVIRIDIS

Although considerable interest has been directed towards the study of hormonal control of regeneration, specific role of endocrine glands is yet to be clarified. The suspicion that cellular properties did not play so dominant a role in regeneration became a near certainty when endocrinological factors in regeneration were reexamined in the light of discoveries made by mammalian endocrinologist in regard to the pituitary-adrenal synergism under stress conditions (Selye, 1947; Selye and Stone, 1950). Schotte and Hall (1952) proposed, that, in a manner akin to mammalian endocrinology, the role of pituitary in urodele regeneration was probably confined to the stimulation of adrenal cortex after amputational stress. On the basis of a series of experiments, Schotte et al. (1961) postulated that the pituitary adrenal synergism is a major controlling factor especially in the early stages of limb regeneration in urodeles, which includes wound healing and differentiation. Contradictory views were reported regarding the involvement of adrenal on regeneration. In frogs, regenerative process was induced by an hyperadrenal state (Schotte and Wilber, 1958), while in the rat, an enhanced liver regeneration occurred due to adrenalectomy (Lucia, 1974; Trepstra et al., 1979). Due to the relative lack of information on these aspects in reptiles, it was deemed important to study the involvement of adrenal in lacertilian tail regeneration. Metapirone reportedly failed to reduce adrenocorticosteroids in fishes (Liversage et al., 1971). So, usage of metapirone to supress the secretion of adrenocorticosteroid was not thought fruitful. Further, bilateral adrenalectomy and even minute doses of aniline were found to be lethal. Hence, in the present study, unilateral adrenalectomy was performed to study the possible alterations, if any, in the rate of regeneration in the house lizard, Hemidactylus flaviviridis under hypoadrenalism, as a preliminary step.

It has been reported, that adrenal steroid hormones exert marked effect on carbohydrate and protein metabolisms (Britton and Silvette, 1932). Adrenalectomy results in reduced responsiveness of glycogenolysis and phosphorylase to glucagon activation (Timothy et al., 1979), decreased serum insulin level (Losert et al., 1970), induces hypoglycemia and low tissue glycogen content (Long et al., 1940) and reduce absorption of glucose from the intestine (Chin and Wang, 1979). Contradictory views were reported regarding the influence of adrenal steroids in protein synthesis (Ronald et al., 1979; Lafont and Pilon, 1975).

Adrenalectomy resulted in reduced synthesis and increased degradation of ascorbic acid in the liver and kidney of rats (Nathani et al., 1971). Alkaline phosphatase activity changed after corticosteroid administration (Simmons and Kunin, 1967; Silbermann et al., 1977b; Majumudar, 1981). The importance of adrenal gland on repair and regeneration and that of ATPase in repair synthesis (Robert and Michael, 1979) and of regeneration (Wolburg and Kurtz, 1981) have also been suggested. In the light of the above mentioned relations between adrenal secretion and various physiological parameters, a quantitate evaluation of blood glucose, tissue glycogen, protein, ascorbic acid, phosphatase and ATPase were also undertaken during various stages of regeneration under the condition of unilateral adrenalectomy.

MATERIALS AND METHODS

Adult H. flaviviridis (10-12 grams weight), collected from the local animal dealer, were kept in the laboratory for a fortnight prior to experimentation. They were divided into three groups. First group of animals (6 males and 6 females) was subjected to surgery under hypothermia for unilateral adrenalectomy (UAX) by a lateral abdominal incision. Second group was subjected to sham operation. Third group was kept as such to serve as the intact controls. After 12 days postoperation (time alloted for the healing of the surgical wound), tails of all the lizards were autotomised two to three segments away from the vent. The rate of growth of the regenerates in all the three groups were measured along with the quantitative estimations of blood glucose (Folin and Malmros, 1929) tissue glycogen (Seifter et al., 1950), protein (Lowry et al., 1951), ascorbic acid (Roe, 1954), acid and alkaline phosphatase (Sigma Technical Bulletin No. 104 using PNP as substrate) and Na^+ K^+ ATPase (Stastry, 1971; Fiske and SubbaRaw, 1925).

RESULTS

Although there was a 24 hrs delay in the formation of a blastema in UAX lizards, the regenerate was well formed in size when compared to the normal (unoperated) ones.

Blood glucose was very much reduced in UAX animals and showed an increase till the 3rd day post-autotomy, in contrast to the other two groups, where the glucose level decreased. Apart from this, for the rest of the periods of tail regeneration, the changes in blood glucose level were identical in all the three groups of lizards. Hepatic glycogen content also showed a lower level in unautotomised-UAX lizards with a similar pattern of changes post-autotomy and ensuing tail regeneration as that of the normal, except

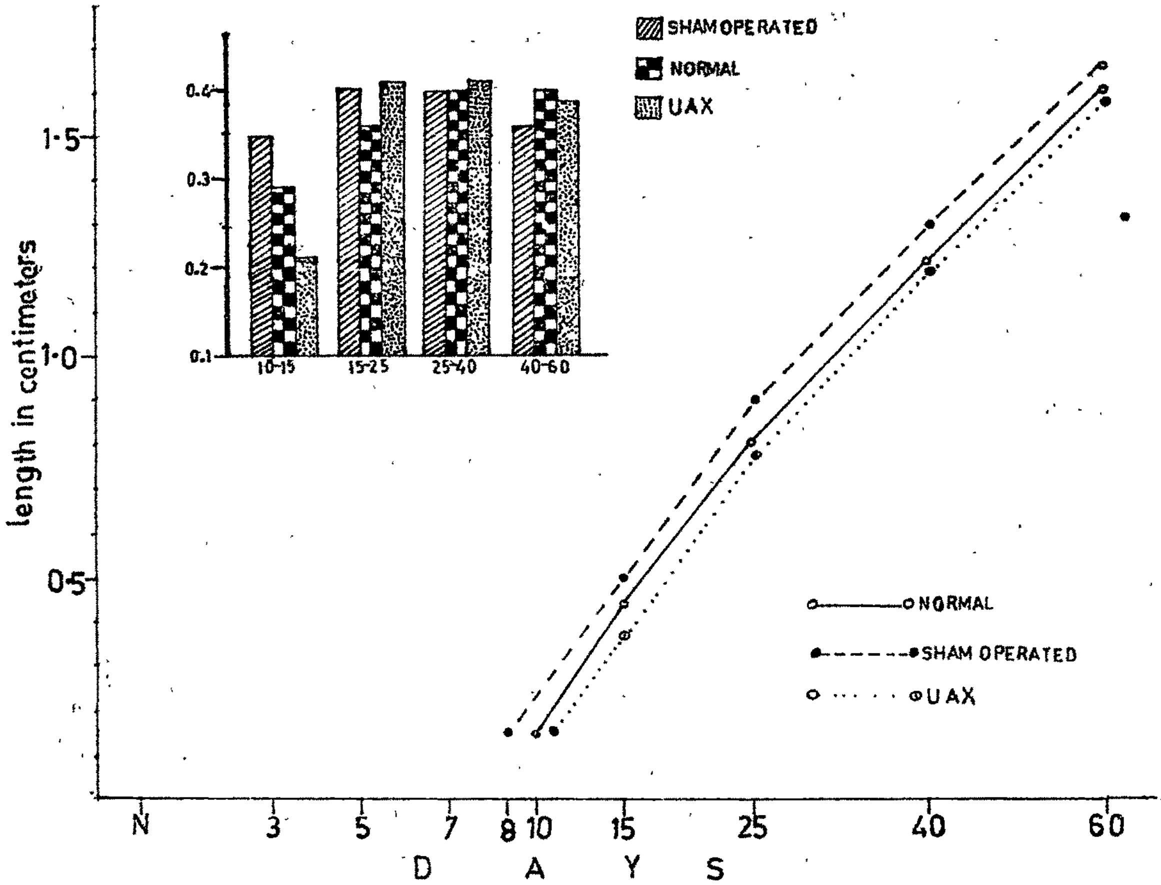

Fig. 1a: Graphic representation of the length of regenerate attained at different time periods, (inset) Rate of growth during different time periods in the normal, sham operated and unilaterally adrenalectomised lizards, H. flaviviridis.

UAX - Unilaterally adrenalectomised.

Table 1. Levels of glucose in blood (mg/100 ml blood) during tail regeneration in normal, sham operated and unilaterally adrenalectomised lizard, *H. flaviviridis*.

Periods of regeneration in days	N	1	2	3	5	7	10	15	25	40	60
Normal intact (IC)	74.00 ±5.82	65.00 ±4.321	67.00 ±7.40	21.00 ±2.85	55.00 ±7.67	69.00 ±6.53	63.00 ±8.32	26.00 ±3.45	26.00 ±5.20	35.00 ±2.89	51.00 ±6.37
Sham operated (SUAX)	36.00 ±4.18	47.00 ±5.35	35.00 ±2.17	15.00 ±1.432	46.00 ±4.36	53.00 ±7.84	50.00 ±6.44	27.00 ±2.11	40.00 ±3.33	31.00 ±5.37	52.00 ±7.20
Unilaterally adrenalectomised (UAX)	22.00 ±4.40	25.00 ±3.70	42.00 ±5.28	65.00 ±9.23	48.00 ±1.40	25.00 ±3.94	57.00 ±6.35	44.00 ±1.19	11.00 ±1.11	55.00 ±7.10	26.00 ±2.45

± S. D.

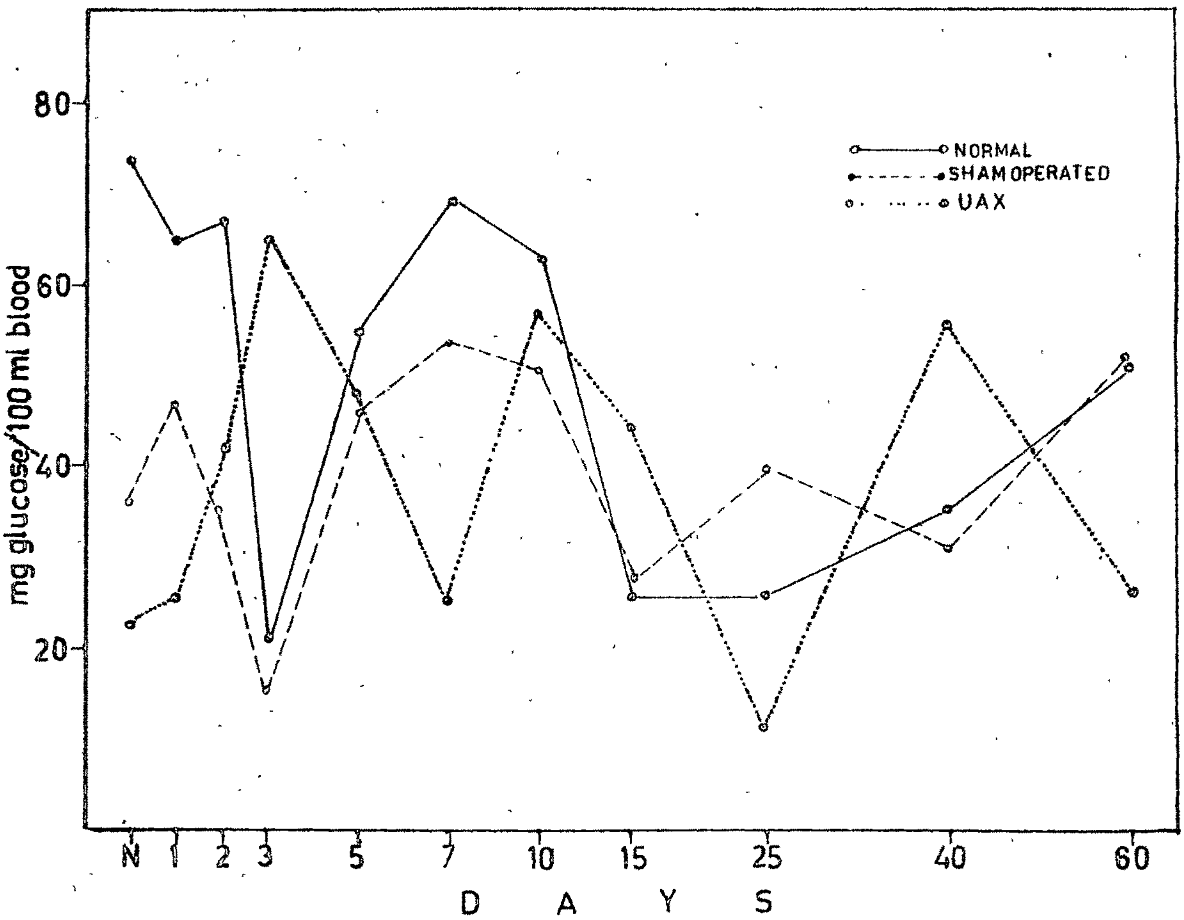

Fig. 1b: Graphic representation of the levels of blood glucose during tail regeneration in the normal, sham operated and unilaterally adrenalectomised lizards, H. flaviviridis

UAX - Unilaterally adrenalectomised.

Table 2. Levels of caudal glycogen content (mg/100 mg fresh tissue) during tail regeneration in normal, sham operated and unilaterally adrenalectomised lizard, *H. flaviviridis*

Periods of regeneration in days	N	1	2	3	5	7	10	15	25	40	60
Normal intact (IC)	0.565 ±0.156	0.31 ±0.007	0.462 ±0.021	0.306 ±0.18	0.538 ±0.009	0.221 ±0.10	0.289 ±0.08	0.144 ±0.025	0.524 ±0.035	0.288 ±0.009	0.323 ±0.050
Sham operated (SUAX)	0.418 ±0.027	0.554 ±0.017	0.164 ±0.005	0.343 ±0.026	0.366 ±0.11	0.266 ±0.051	0.288 ±0.019	0.144 ±0.006	0.191 ±0.024	0.251 ±0.008	0.334 ±0.014
Unilaterally adrenalectomised (UAX)	0.052 ±0.003	0.047 ±0.002	0.118 ±0.014	0.146 ±0.015	0.524 ±0.001	0.108 ±0.020	0.289 ±0.018	0.338 ±0.019	0.322 ±0.023	0.545 ±0.046	0.508 ±0.054

± S. D.

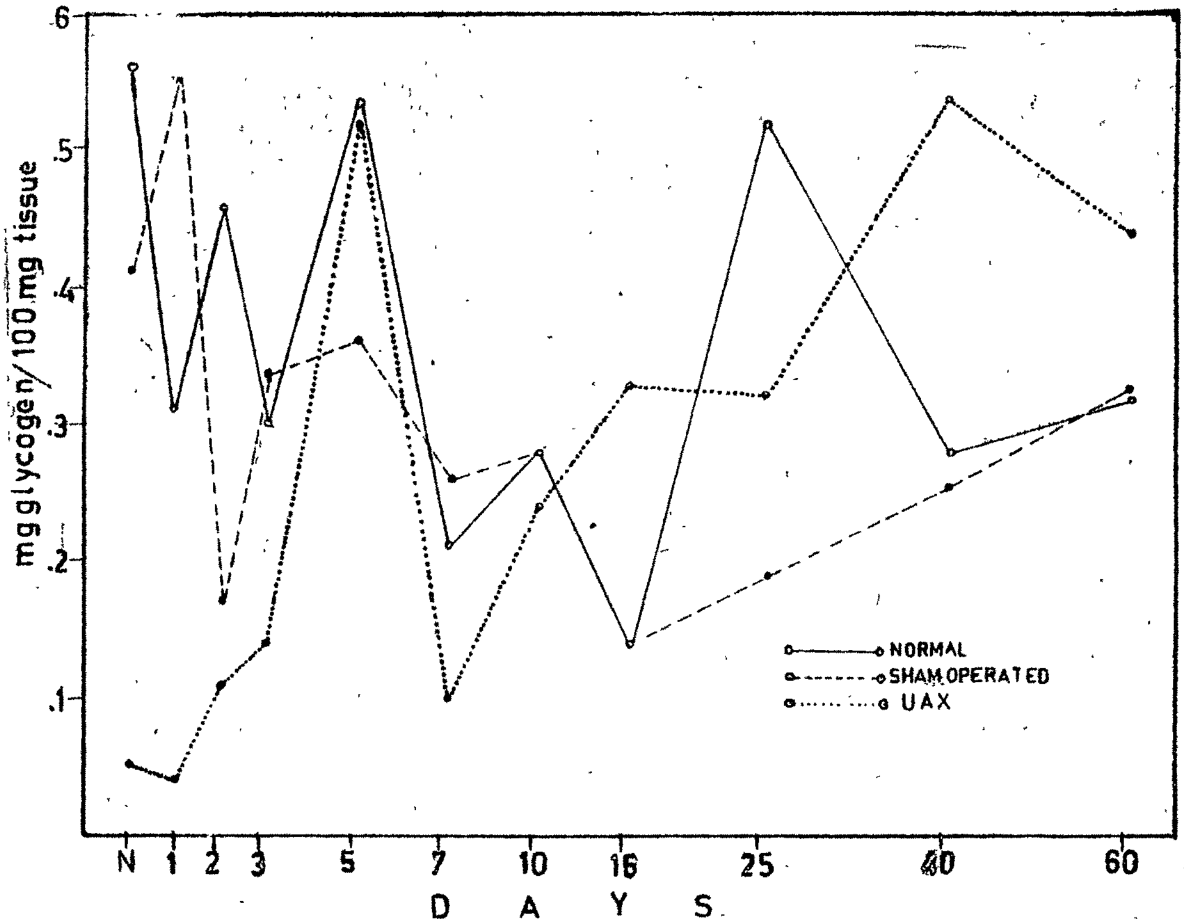

Fig. 2 : Graphic representation of the levels of glycogen in the regenerate during tail regeneration in the normal, sham operated and unilaterally adrenalectomised lizards, H. flaviviridis

UAX - Unilaterally adrenalectomised.

Table 3. Levels of hepatic glycogen content (mg/100 mg fresh tissue) during tail regeneration in normal, sham operated and unilaterally adrenalectomised lizard, *H. flaviviridis*.

Periods of regeneration in days	N	1	2	3	5	7	10	15	25	40	60
Normal intact (IC)	2.37 ±0.008	1.55 ±0.011	0.305 ±0.003	1.22 ±0.04	3.38 ±0.14	1.676 ±0.112	1.10 ±0.007	0.771 ±0.004	0.391 ±0.010	0.483 ±0.021	1.56 ±0.03
Sham operated (SUAX)	1.52 ±0.031	2.24 ±0.134	0.448 ±0.001	2.61 ±0.013	1.16 ±0.10	1.724 ±0.21	2.32 ±0.18	1.481 ±0.003	0.916 ±0.15	0.712 ±0.016	0.751 ±0.024
Unilaterally adrenalectomised (UAX)	0.827 ±0.04	0.520 ±0.02	0.342 ±0.019	0.659 ±0.043	2.0 ±0.17	1.49 ±0.114	0.345 ±0.016	1.098 ±0.005	1.48 ±0.107	0.583 ±0.115	0.44 ±0.0001

± S. D.

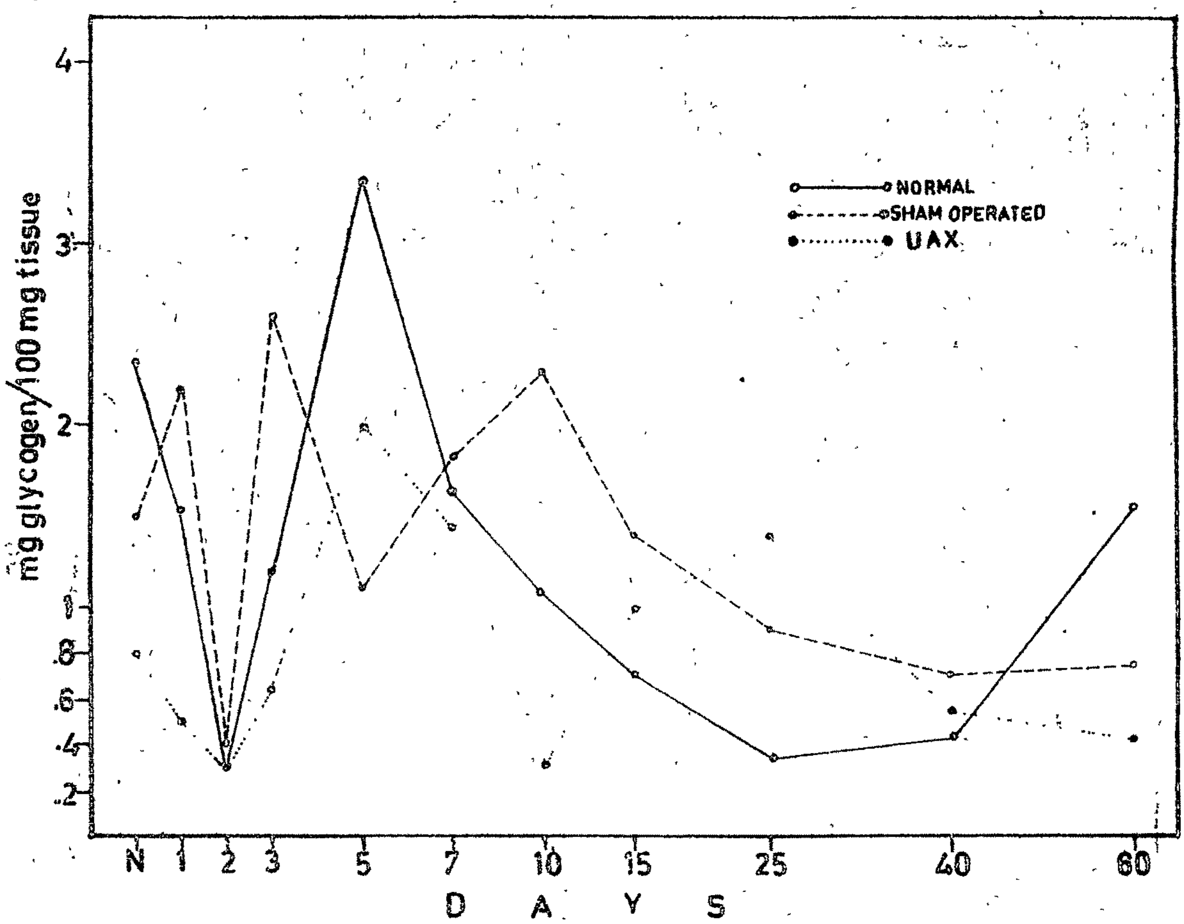

Fig. 3 : Graphic representation of the levels of glycogen in the liver during tail regeneration in the normal, sham operated and unilaterally adrenalectomised lizards, H. *flaviviridis*

UAX - Unilaterally adrenalectomised.

Table 4. Levels of muscle glycogen content (mg/100 mg fresh tissue) during tail regeneration in normal, sham operated and unilaterally adrenalectomised lizard, *H. flaviviridis*.

Periods of regeneration in days	N	1	2	3	5	7	10	15	25	40	60
Normal intact (IC)	0.0815 ±0.003	0.1050 ±0.005	0.09898 ±0.001	0.1235 ±0.014	0.1539 ±0.02	0.1729 ±0.018	0.2160 ±0.007	0.1397 ±0.011	0.1787 ±0.032	0.2317 ±0.017	0.2545 ±0.015
Sham operated (SUAX)	0.0794 ±0.006	0.605 ±0.001	0.1136 ±0.019	0.1353 ±0.016	0.117 ±0.009	0.125 ±0.010	0.154 ±0.012	0.125 ±0.021	0.26 ±0.03	0.285 ±0.008	0.225 ±0.034
Unilaterally adrenalectomised (UAX)	0.0179 ±0.008	0.028 ±0.007	0.072 ±0.010	0.078 ±0.005	0.220 ±0.024	0.249 ±0.017	0.113 ±0.032	0.1060 ±0.009	0.006 ±0.002	0.093 ±0.016	0.136 ±0.004

± S. D.

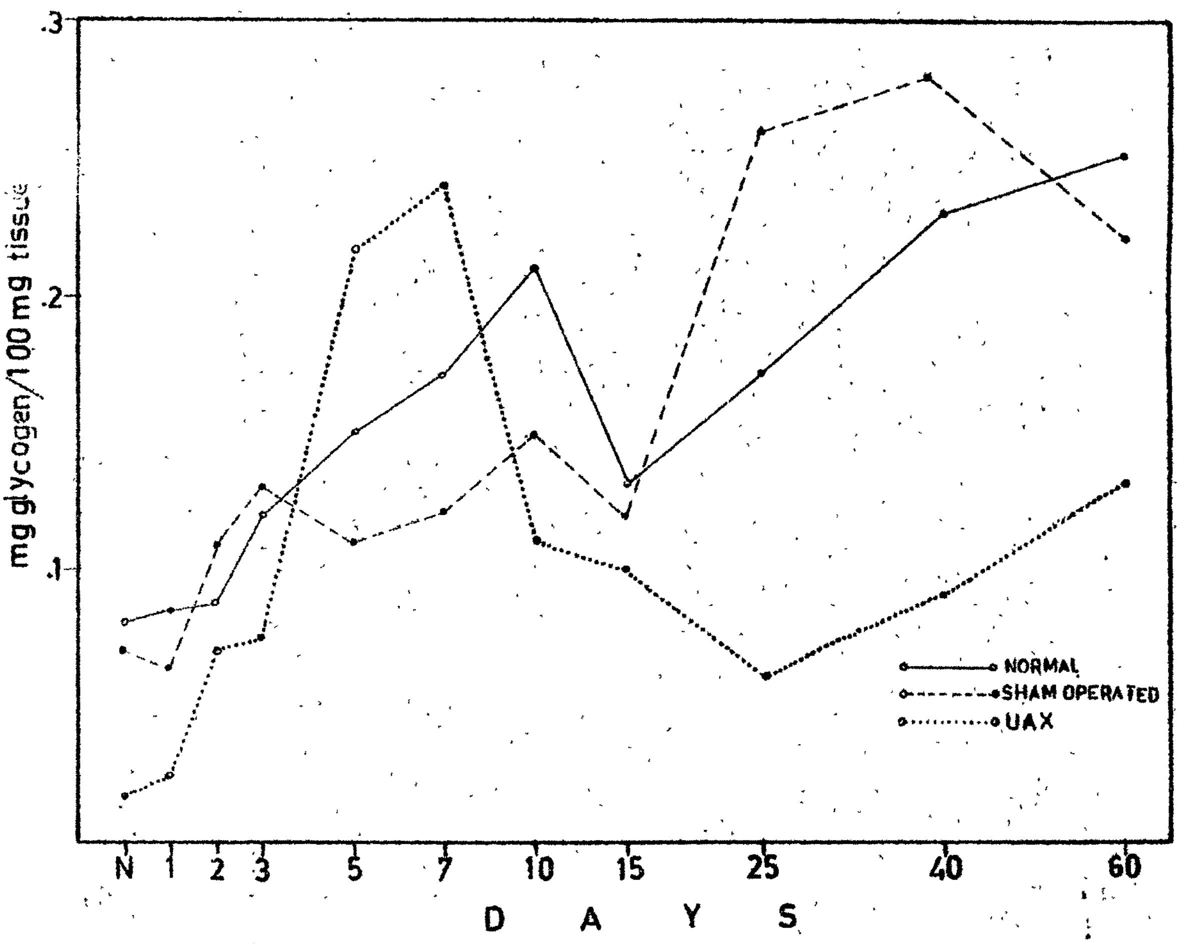

Fig. 4 : Graphic representation of the levels of muscle glycogen during tail regeneration in the normal, sham operated and unilaterally adrenalectomised lizards, H. flaviviridis

UAX - Unilaterally adrenalectomised.

during the period between 10-25 days whence they showed an increase in the liver glycogen content. Muscle glycogen in normal and SUAX (sham operated) continuously increased till the 10th day with minor fluctuations. On day 15th, the muscle glycogen in the above two groups fell, and again increased on the 25th day. The level remained very much above normal till the 60th day. The UAX animals showed a continuous increase till 7th day. Thereafter, a gradual fall was noted till 25th day followed by a gradual increase. Tail glycogen in all the three groups remained below normal and showed a similar pattern in the later stages. Both the SUAX and normal lizards showed minor fluctuations during regeneration, while the normal lizards showed first peak on the 5th day, and a second one on the 25th day, the SUAX controls showed the first peak on 1st day and a minor second one on the 5th day. On 25th day, there was an increase in the unlike glycogen in the normal lizards followed by a low level on 40th, and a gradual increase thereafter, while in the case of sham operated after a decrease between 7th and 15th day the level went on increasing till the 60th day. On the other hand, the UAX animals which had a low level of caudal glycogen before autotomy, showed an increasing trend till the 5th day. After a slight fall on 7th day, it was followed by a gradual increase lasting up till the 40th day. The concentration gradually decreased thereafter, to be more or less at the normal level by 60th day.

Table 5. Quantitative alterations in caudal protein content (mg/100 mg) during tail regeneration in normal, sham operated and unilaterally adrenalectomised lizard, H. flaviviridis.

Periods of regeneration in days	N	1	2	3	5	7	10	15	25	40	60
Normal intact (IC)	5.267 ±1.32	4.454 ±1.108	5.343 ±1.17	1.479 ±0.54	4.882 ±1.327	4.469 ±1.101	5.199 ±1.02	5.877 ±0.762	3.137 ±0.88	5.086 ±1.44	4.195 ±1.05
Sham operated (SUAX)	6.836 ±0.481	5.104 ±0.785	9.469 ±2.45	6.310 ±1.113	4.951 ±0.463	5.050 ±1.07	4.186 ±1.061	4.126 ±1.151	3.94 ±0.65	3.476 ±0.93	4.353 ±0.47
Unilaterally adrenalectomised (UAX)	4.213 ±0.924	5.658 ±0.33	6.223 ±1.16	5.042 ±0.95	4.363 ±0.784	4.348 ±0.96	4.807 ±1.35	4.597 ±1.54	1.650 ±0.008	4.234 ±0.53	4.5 ±1.001

± S. D.

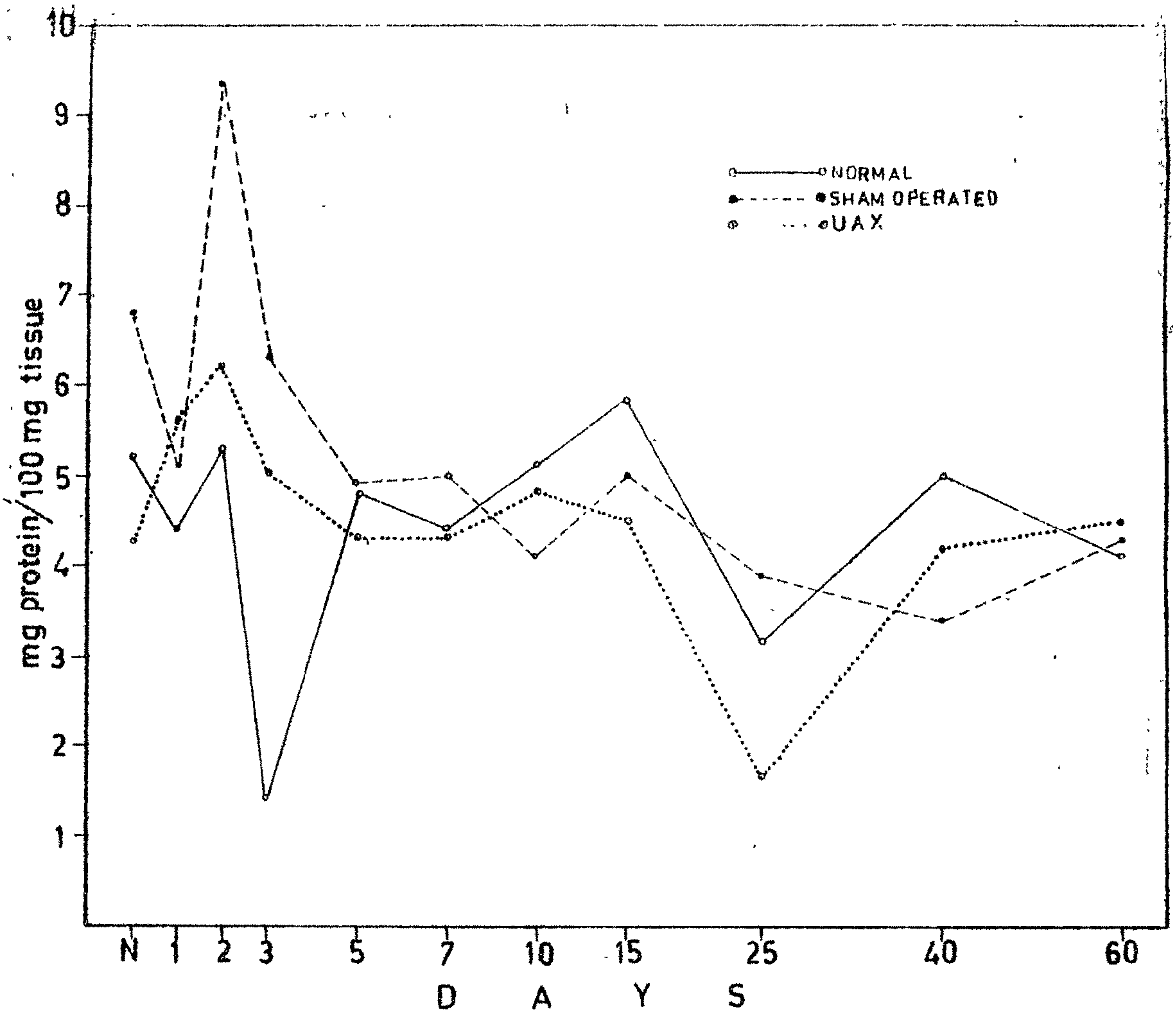

Fig. 5 : Graphic representation of the levels of caudal protein during its regeneration in the normal, sham operated and unilaterally adrenalectomised lizards, H. *flaviviridis*

UAX - Unilaterally adrenalectomised.

Table 6. Quantitative alterations in hepatic protein content (mg/100 mg) during tail regeneration in normal, sham operated and unilaterally adrenalectomised lizard, *H. flaviviridis*.

Periods of regeneration in days	N	1	2	3	5	7	10	15	25	40	60
Normal intact (IC)	23.945 ±3.832	13.386 ±2.481	12.014 ±2.483	10.645 ±1.467	11.540 ±0.899	13.631 ±4.324	15.148 ±2.35	14.565 ±2.11	13.49 ±0.99	15.064 ±3.397	20.961 ±5.32
Sham operated (SUAX)	23.233 ±3.341	16.695 ±3.333	14.336 ±4.872	15.456 ±2.281	14.702 ±3.75	14.107 ±1.88	12.768 ±3.66	13.158 ±2.705	12.85 ±1.183	18.460 ±4.463	19.04 ±4.526
Unilaterally adrenalectomised (UAX)	12.553 ±2.82	14.965 ±4.450	18.357 ±1.97	11.911 ±1.924	11.786 ±2.918	10.672 ±0.58	14.004 ±1.117	14.515 ±3.18	11.333 ±2.46	13.179 ±1.47	16.969 ±1.457

± S. D.

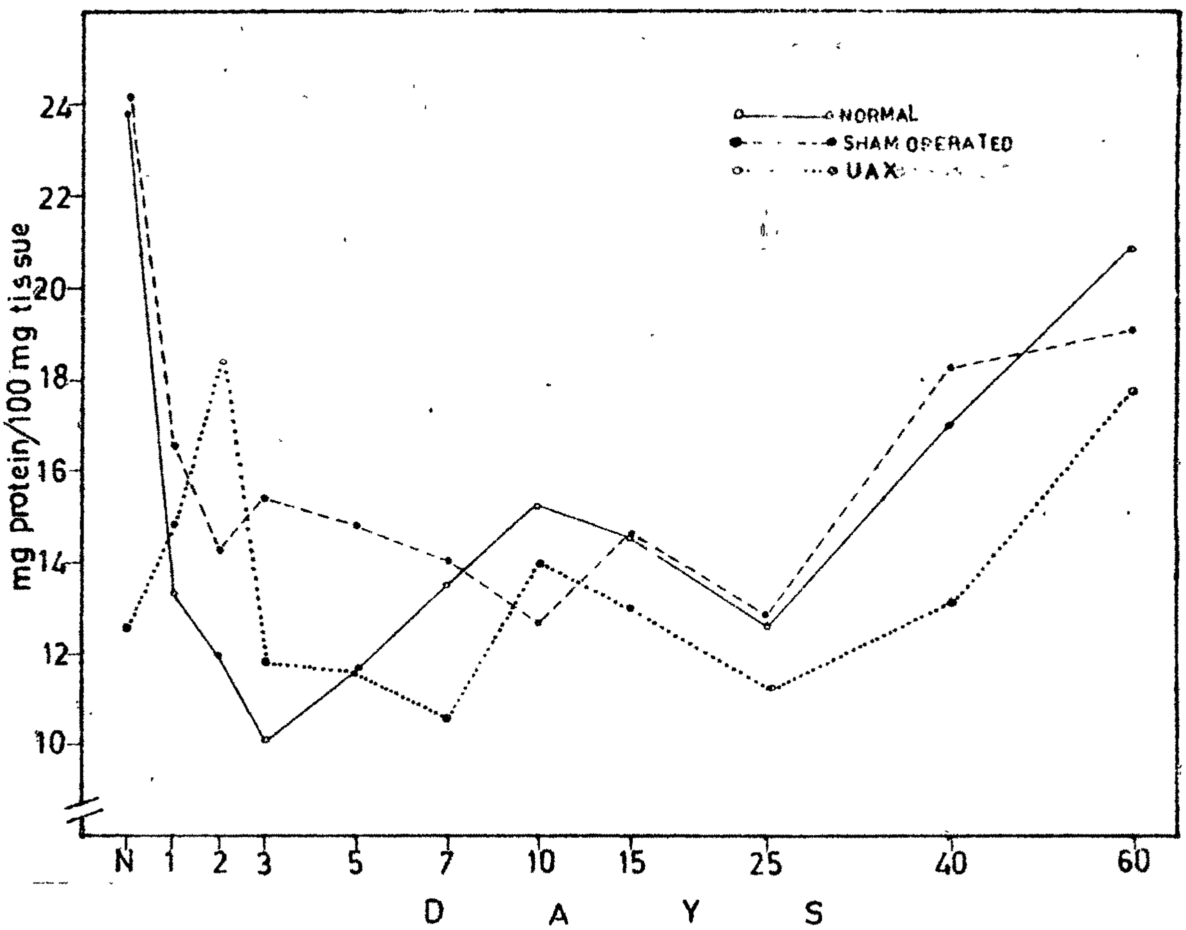

Fig. 6 : Graphic representation of the levels of hepatic protein in the normal, sham operated and unilaterally adrenalectomised lizards, H. flaviviridis

UAX - Unilaterally adrenalectomised

Table 7. Quantitative alterations in muscle protein content (mg/100 mg) during tail regeneration in normal, sham operated and unilaterally adrenalectomised lizard, *H. flaviviridis*.

Periods of regeneration in days	N	1	2	3	5	7	10	15	25	40	60
Normal intact (IC)	19.538 ±5.08	9.010 ±0.485	10.877 ±3.11	8.017 ±1.77	8.012 ±2.43	8.556 ±1.38	9.875 ±1.43	12.350 ±3.62	7.220 ±1.45	9.01 ±1.08	10.250 ±0.597
Sham operated (SUAX)	16.072 ±3.365	14.824 ±4.55	10.137 ±2.75	9.785 ±1.53	9.440 ±1.56	10.314 ±0.978	9.669 ±2.66	11.261 ±2.43	10.16 ±0.97	10.487 ±2.34	11.074 ±1.356
Unilaterally adrenalectomised (UAX)	9.402 ±2.35	10.947 ±1.65	13.223 ±1.99	12.131 ±1.34	10.694 ±1.44	11.840 ±2.56	9.680 ±0.721	7.676 ±2.482	7.760 ±1.64	8.610 ±1.73	6.07 ±1.05

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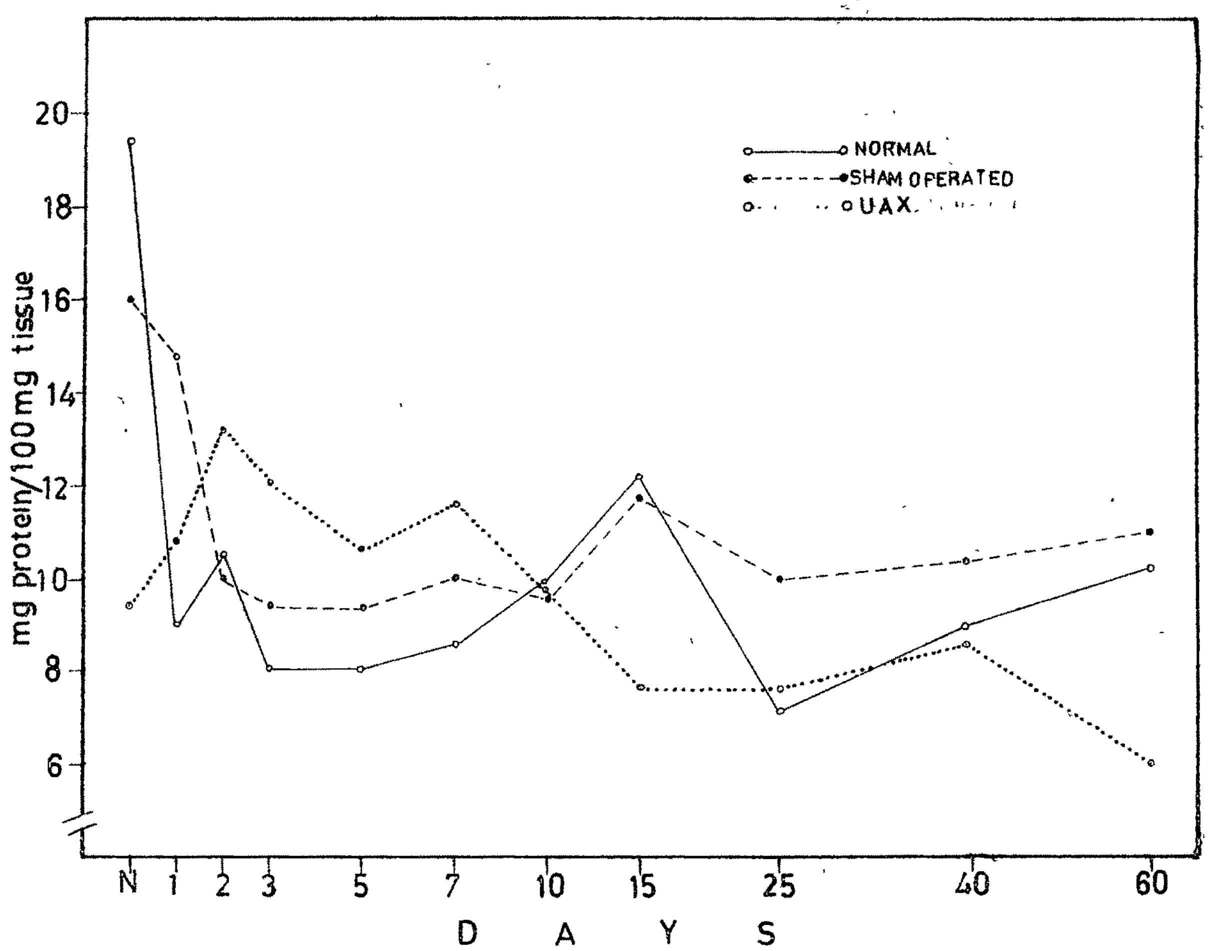

Fig. 7 : Graphic representation of the levels of muscle protein during tail regeneration in the normal, sham operated and unilaterally adrenalectomised lizards, *H. flaviviridis*

UAX - Unilaterally adrenalectomised.

Table 8. Alterations in the levels of hepatic ascorbic acid (mg/100 gm fresh tissue) during tail regeneration in the normal, sham operated and unilaterally adrenalectomised lizard, H. *flaviviridis*.

Periods of regeneration in days	N	1	2	3	5	7	10	15	25	40	60
Normal intact (IC)	21.9 ±5.08	21.4 ±3.24	20.9 ±1.77	18.2 ±3.11	23.1 ±3.52	32.6 ±8.35	20.5 ±4.08	23.1 ±3.43	27.8 ±9.72	31.0 ±6.45	23.6 ±5.02
Sham operated (SUAX)	16.5 ±1.72	28.1 ±5.55	25.5 ±6.05	32.9 ±8.34	11.1 ±2.22	26.7 ±5.87	27.3 ±4.54	25.0 ±1.99	25.0 ±5.04	31.4 ±9.47	32.1 ±5.47
Unilaterally adrenalectomised (UAX)	21.7 ±3.47	22.5 ±3.11	23.1 ±2.17	22.1 ±5.61	19.7 ±2.80	28.8 ±7.13	40.1 ±10.08	27.6 ±9.19	31.7 ±6.11	28.9 ±6.24	20.8 ±2.27

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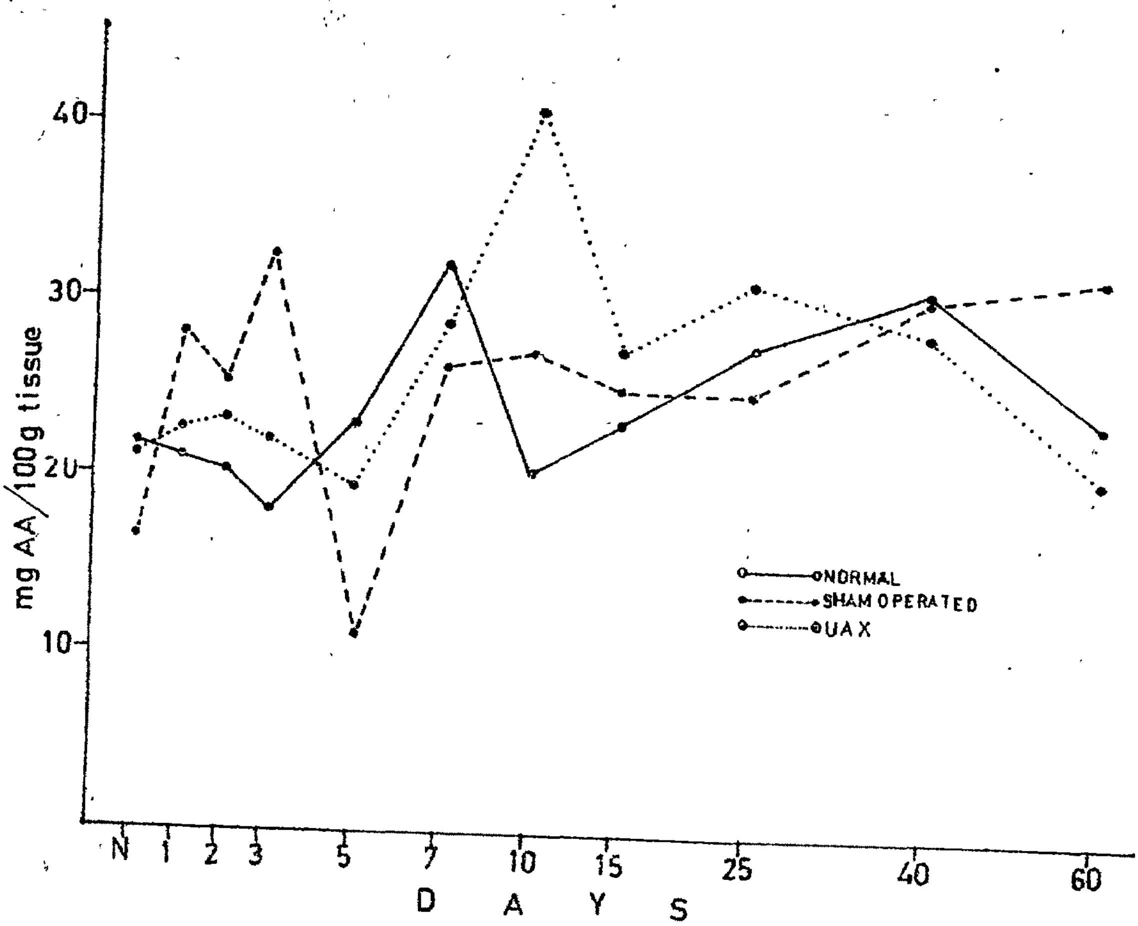

Fig. 8 : Graphic representation of the levels of hepatic asrorbic acid during tail regeneration in the normal,sham operated and unilaterally adrenalectomised lizards, H. flaviviridis.

UAX - Unilaterally adrenalectomised.

Table 9. Alterations in the levels of renal ascorbic acid (mg/100 gm fresh tissue) during tail regeneration in the normal, sham operated and unilaterally adrenalectomised lizards, *H. flaviviridis*.

Periods of regeneration in days	N	1	2	3	5	7	10	15	25	40	60
Normal intact (IC)	17.5 ±1.87	22.3 ±3.14	20.9 ±2.92	18.0 ±3.11	14.2 ±1.74	15.8 ±2.24	14.2 ±1.99	28.2 ±5.85	19.0 ±4.01	18.7 ±4.16	27.0 ±5.97
Sham operated (SUAX)	11.3 ±1.44	15.0 ±2.66	10.3 ±1.43	33.1 ±6.67	10.6 ±1.08	24.6 ±5.32	23.3 ±4.15	34.8 ±8.25	24.6 ±6.21	30.6 ±8.82	26.6 ±7.10
Unilaterally adrenalectomised (UAX)	14.5 ±2.13	21.3 ±5.04	17.3 ±2.16	21.1 ±3.43	15.5 ±1.01	22.3 ±3.97	24.8 ±6.04	15.8 ±1.77	33.0 ±9.38	25.5 ±6.45	28.6 ±4.21

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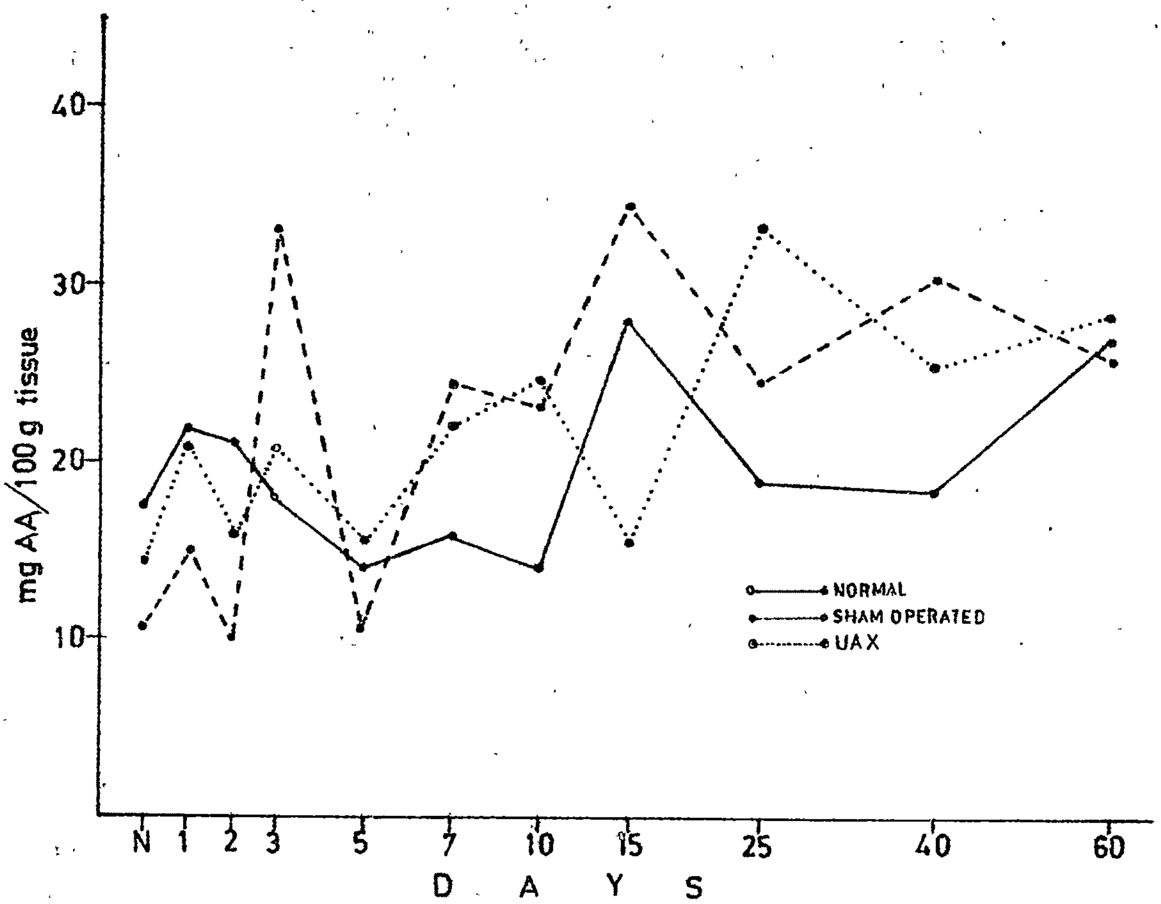

Fig. 9 : Graphic representation of the levels of renal ascorbic acid during tail regeneration in the normal, sham operated and unilaterally adrenalectomised lizards, H. flaviviridis

UAX - Unilaterally adrenalectomised.

Table 10. Alterations in the levels of caudal ascorbic acid (mg/100 gm fresh tissue) in the normal, sham operated and unilaterally adrenalectomised lizard, H. *flaviviridis*.

Periods of regeneration in days	N	1	2	3	5	7	10	15	25	40	60
Normal intact (IC)	6.01 ±1.52	7.4 ±2.23	6.12 ±1.66	6.84 ±2.34	7.66 ±2.12	6.37 ±1.77	8.4 ±2.24	11.97 ±3.33	9.02 ±1.93	8.46 ±2.25	5.9 ±0.98
Sham operated (SUAX)	4.89 ±1.01	9.6 ±2.35	7.35 ±1.14	6.34 ±2.51	4.1 ±0.88	9.78 ±2.45	9.02 ±3.00	5.3 ±0.78	7.4 ±2.15	3.6 ±0.44	6.6 ±0.75
Unilaterally adrenalectomised (UAX)	5.95 ±1.82	4.43 ±0.98	4.55 ±1.01	7.17 ±2.43	9.36 ±2.57	7.2 ±1.73	7.83 ±2.66	14.66 ±3.47	5.9 ±1.08	3.9 ±0.55	6.86 ±2.05

± S. D.

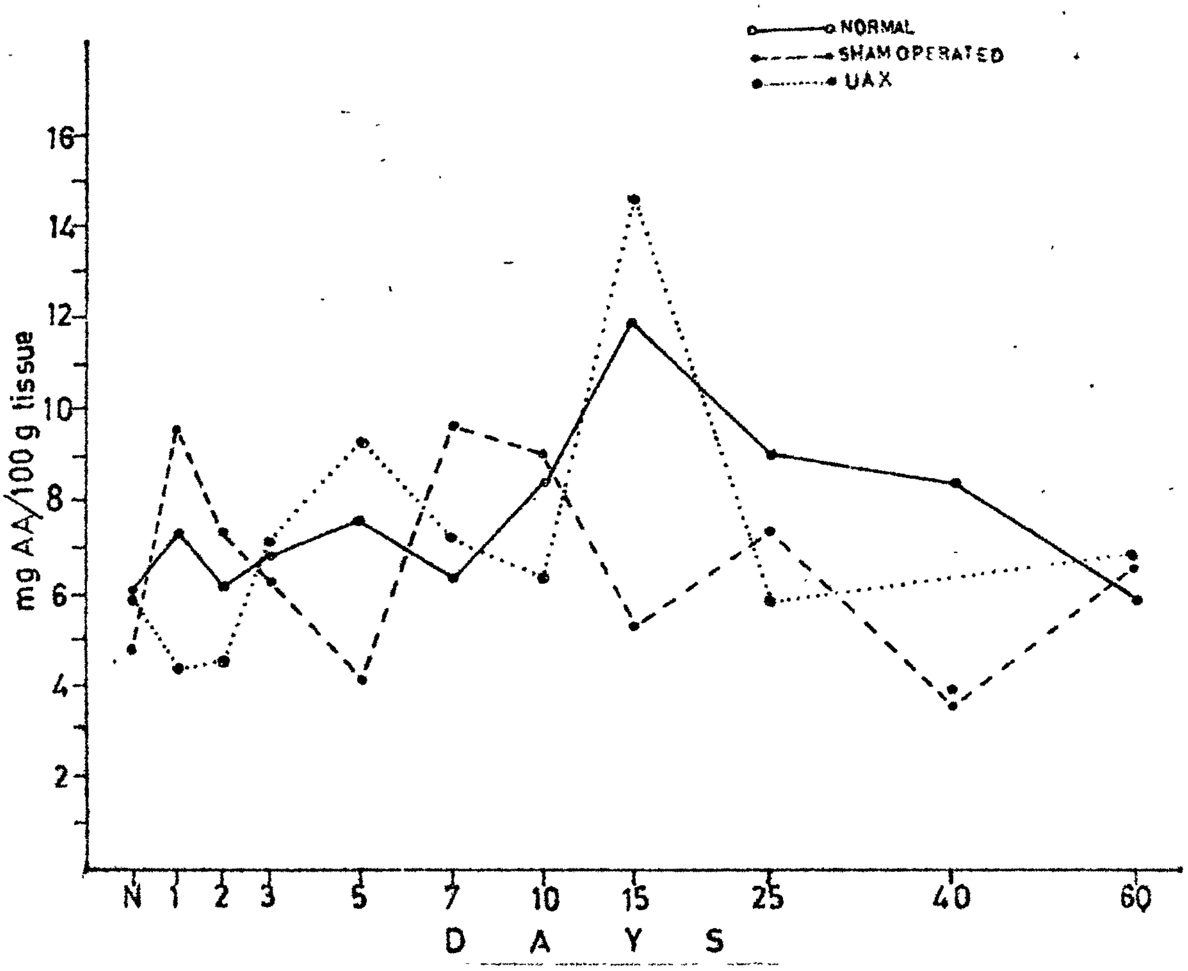

Fig. 10 : Graphic representation of the levels of caudal ascorbic acid during tail regeneration in the normal, sham operated and unilaterally adrenalectomised lizards, *H. flaviviridis*.

UAX - Unilaterally adrenalectomised.

Liver protein of UAX lizards remained very low before autotomy. When the liver protein content of normal and SUAX control lizards showed a decrease, the UAX lizards showed an increase on the 2nd day. For the rest of the periods the pattern remained more or less the same. Muscle protein also showed a reduction in the UAX animals prior to autotomy. Apart from the initial increase on the 2nd day in the case of UAX lizards, the pattern was more or less similar. In the case of tail protein too, apart from slight difference during initial periods, the changes were more or less comparable in all the three groups.

Liver and kidney ascorbic acid tended to remain higher in all the three groups of animals with minor variations in the levels. There was more or less no difference in tail AA content during regeneration among all the three groups.

Alkaline phosphatase in the liver of UAX lizards showed a high activity during tail regeneration as compared to the other two groups, while the muscle and tail alkaline phosphatase showed a moderate activity in UAX. Acid phosphatase activity in liver, muscle and tail was at a higher level in UAX all throughout compared to the other two groups.

Na^{+} K^{+} ATPase activity of liver of normal lizards showed an initial increase lasting upto the 2nd day followed

Table 11. Levels of caudal alkaline phosphatase (μmole p-NP released/mg protein/30 minutes) during tail regeneration in normal, sham operated and unilaterally adrenalectomised lizard, *H. flaviviridis*.

Periods of regeneration in days	N	1	2	3	5	7	10	15	25	40	60
Normal intact (IC)	0.0826 ±0.002	0.1598 ±0.032	0.229 ±0.004	0.192 ±0.041	0.403 ±0.001 0.0005@	0.866 ±0.105 0.0025@	0.157 ±0.028	0.161 ±0.005	0.495 ±0.010	0.404 ±0.001	0.346 ±0.005
Sham operated (SUAX)	0.6669 ±0.003	0.113 ±0.014	0.6807 ±0.006	0.0349 ±0.001	0.428 ±0.104	0.0995 ±0.009	0.124 ±0.048	0.162 ±0.005	0.217 ±0.04	0.268 ±0.018	0.25 ±0.06
Unilaterally adrenalectomised (UAX)	0.096 ±0.018	0.109 ±0.007	0.222 ±0.040	0.125 ±0.032	0.152 ±0.054	0.212 ±0.038	0.171 ±0.004	0.287 ±0.011	0.213 ±0.015	0.215 ±0.020	0.394 ±0.054

± S. D.

@ P value obtained in comparison with IC

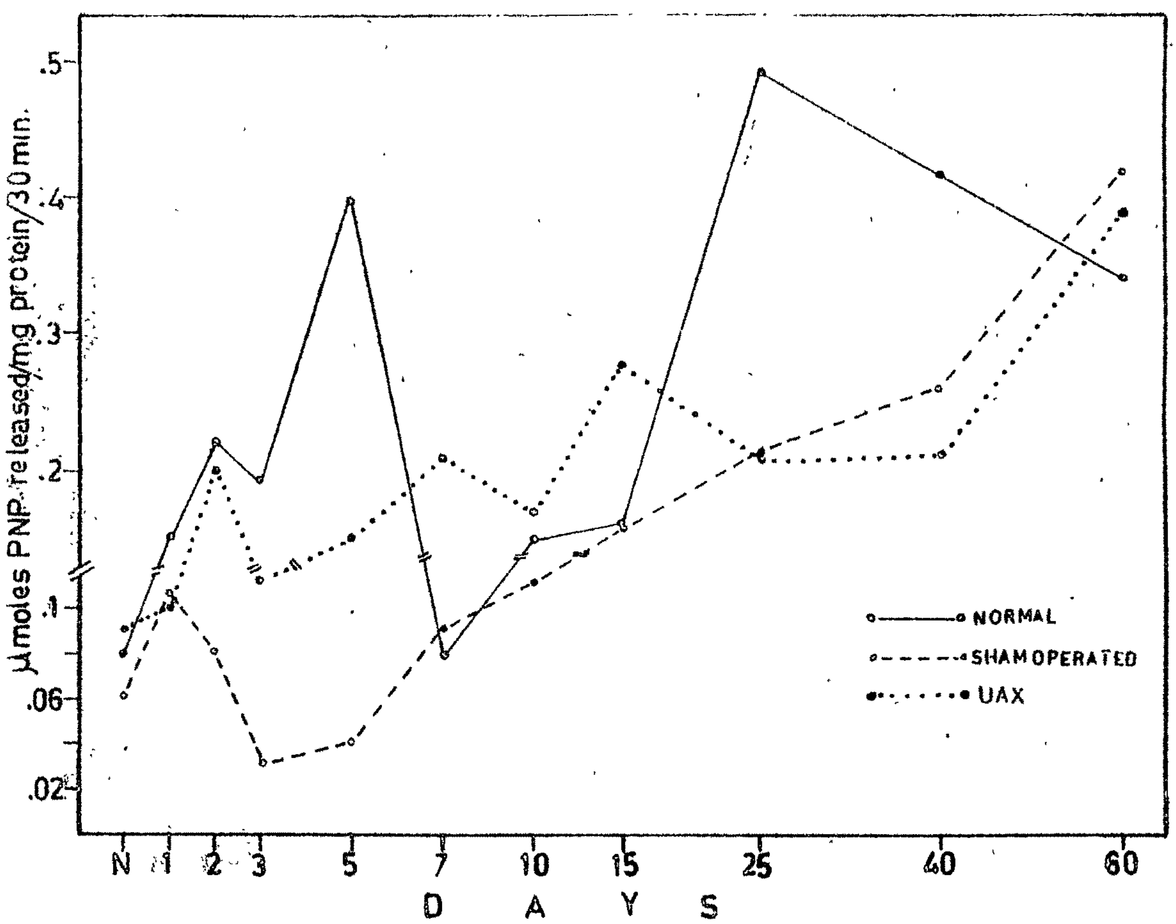

Fig. 11 : Graphic representation of the levels of alkaline phosphatase in the tail during its regeneration in the normal, sham operated and unilaterally adrenalectomised lizards, _H. flaviviridis_

UAX - Unilaterally adrenalectomised.

Table 12. Levels of liver alkaline phosphatase (μ mole p-NP released/mg protein/30 minutes) during tail regeneration in normal, sham operated and unilaterally adrenalectomised lizard, *H. flaviviridis*.

Periods of regeneration in days	N	1	2	3	5	7	10	15	25	40	60
Normal intact (IC)	0.406 ±0.108	0.489 ±0.007	0.414 ±0.076	0.3 ±0.101	0.113 ±0.005	0.373 ±0.001	0.367 ±0.004	0.403 ±0.018	0.481 ±0.034	0.315 ±0.104	0.526 ±0.182
Sham operated (SUAX)	0.474 ±0.005	0.364 ±0.103	0.465 ±0.034	0.289 ±0.007	0.518 ±0.113	0.299 ±0.118	0.443 ±0.009	0.459 ±0.017	0.545 ±0.048	0.715 ±0.008	0.525 ±0.107
Unilaterally adrenalectomised (UAX)	0.379 ±0.006	0.489 ±0.014	0.332 ±0.011	0.461 ±0.005	0.557 ±0.017	0.359 ±0.004	0.375 ±0.054	0.791 ±0.032	0.275 ±0.013	0.479 ±0.012	0.516 ±0.020

± S. D.

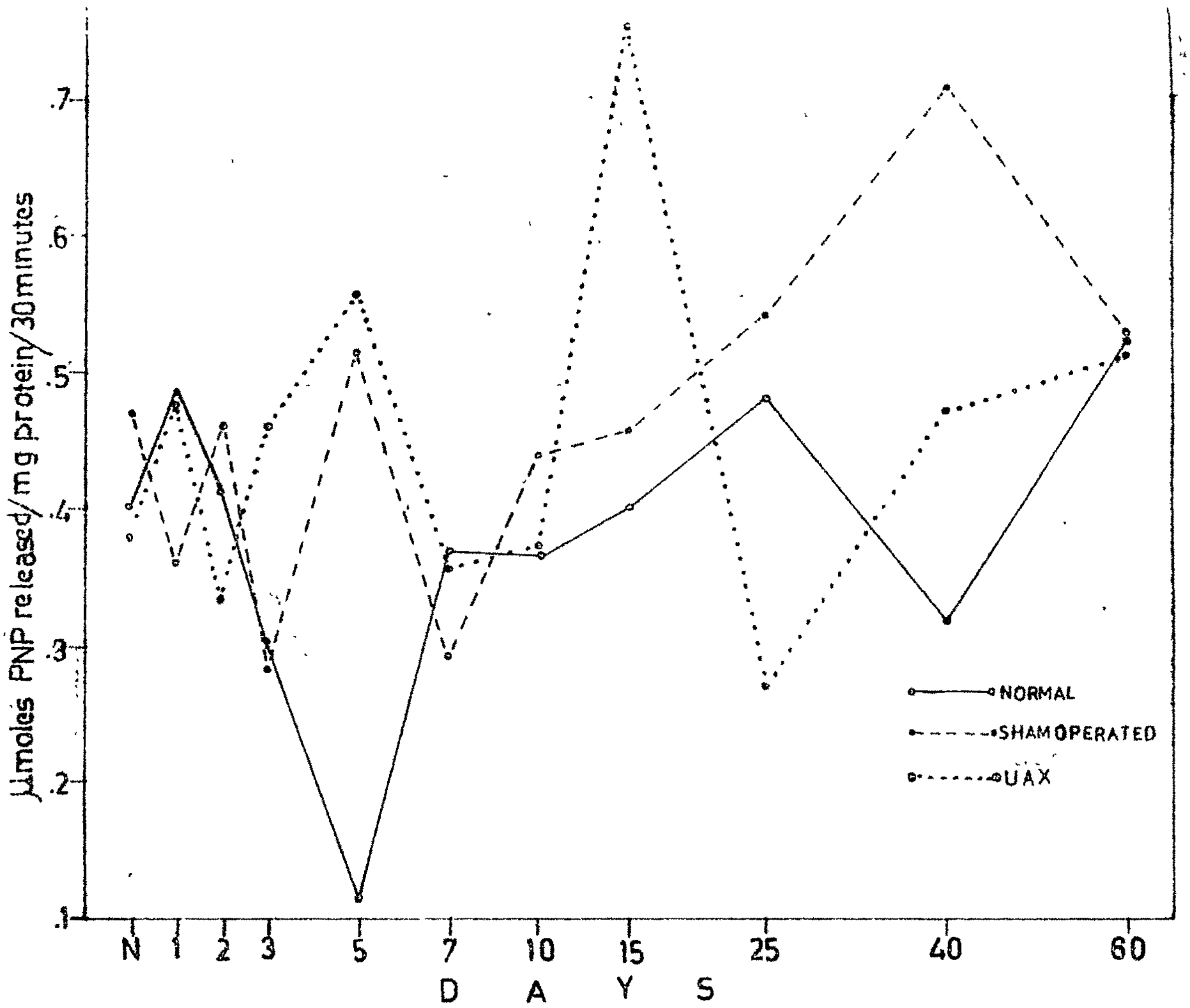

Fig. 12 : Graphic representation of the levels of alkaline phosphatase in the liver during tail regeneration in the normal, sham operated and unilaterally adrenalectomised lizards, H. flaviviridis

UAX - Unilaterally adrenalectomised.

Table 13. Levels of muscle alkaline phosphatase (μ mole p-NP released/mg protein/30 minute) during tail regeneration in normal, sham operated and unilaterally adrenalectomised lizard , _H._ _flaviviridis_.

Periods of regeneration in days	N	1	2	3	5	7	10	15	25	40	60
Normal intact (IC)	0.0306 ±0.005	0.0491 ±0.003	0.08085 ±0.012	0.113 ±0.004	0.1356 ±0.324	0.0168 ±0.001	0.0367 ±0.018	0.0424 ±0.005	0.0308 ±0.001	0.037 ±0.007	0.035 ±0.010
Sham operated (SUAX)	0.0216 ±0.007	0.038 ±0.009	0.1099 ±0.057	0.0022 ±0.0001	0.1051 ±0.034	0.0146 ±0.006	0.0214 ±0.002	0.0362 ±0;001	0.0376 ±0.008	0.033 ±0.005	0.047 ±0.018
Unilaterally adrenalectomised (UAX)	0.0059 ±0.0006	0.026 ±0.001	0.005 ±0.002	0.0055 ±0.0014	0.0066 ±0.001	0.0263 ±0.0015	0.0309 ±0.003	0.0372 ±0.002	0.037 ±0.015	0.0145 ±0.002	0.0498 ±0.0143

± S. D.

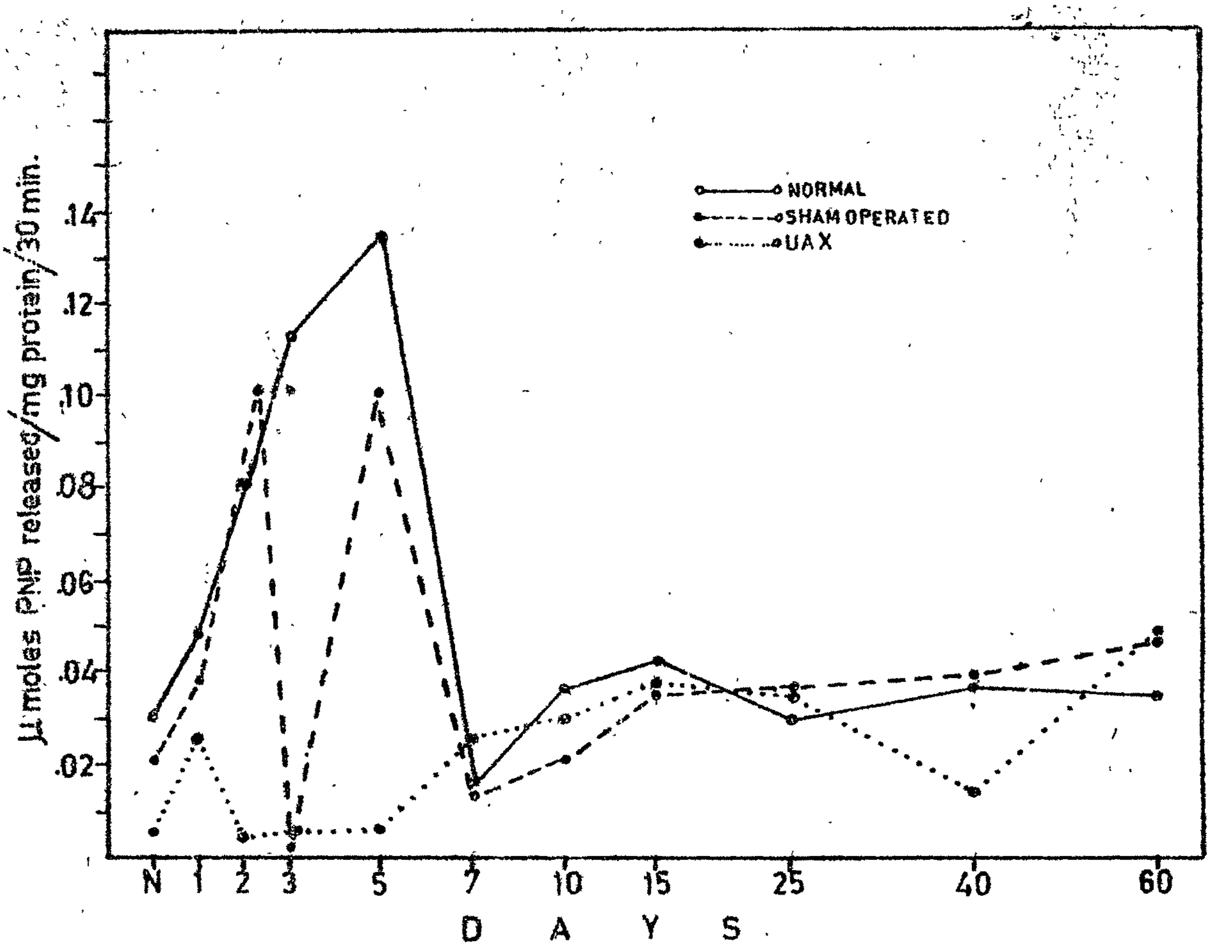

Fig. 13: Graphic representation of the levels of alkaline phosphatase in the muscle during tail regeneration in the normal, sham operated and unilaterally adrenalectomised lizards, *H. flaviviridis*.

UAX - Unilaterally adrenalectomised.

Table 14. Levels of caudal acid phosphatase (μ mole p-ND released/mg protein/30 minutes) during tail regeneration in normal, sham operated and unilaterally adrenalectomised lizard, *H. flaviviridis*.

Periods of regeneration in days	N	1	2	3	5	7	10	15	25	40	60
Normal intact (IC)	0.2357 ±0.052	0.371 ±0.003	0.277 ±0.018	0.207 ±0.001	0.249 ±0.014	0.221 ±0.021	0.329 ±0.004	0.351 ±0.016	0.3374 ±0.0142	0.43 ±0.03	0.351 ±0.019
Sham operated (SUAX)	0.352 ±0.101	0.285 ±0.008	0.198 ±0.012	0.731 ±0.015	0.285 ±0.001	0.303 ±0.004	0.336 ±0.102	0.352 ±0.009	0.301 ±0.122	0.319 ±0;005	0.374 ±0.013
Unilaterally adrenalectomised (UAX)	0.308 ±0.004	0.055 ±0.006	0.492 ±0.13	0.259 ±0.010	0.444 ±0.034	0.536 ±0.040	0.345 ±0.028	0.712 ±0.015	0.49 ±0.026	0.688 ±0.067	0.700 ±0.32

± S. D.

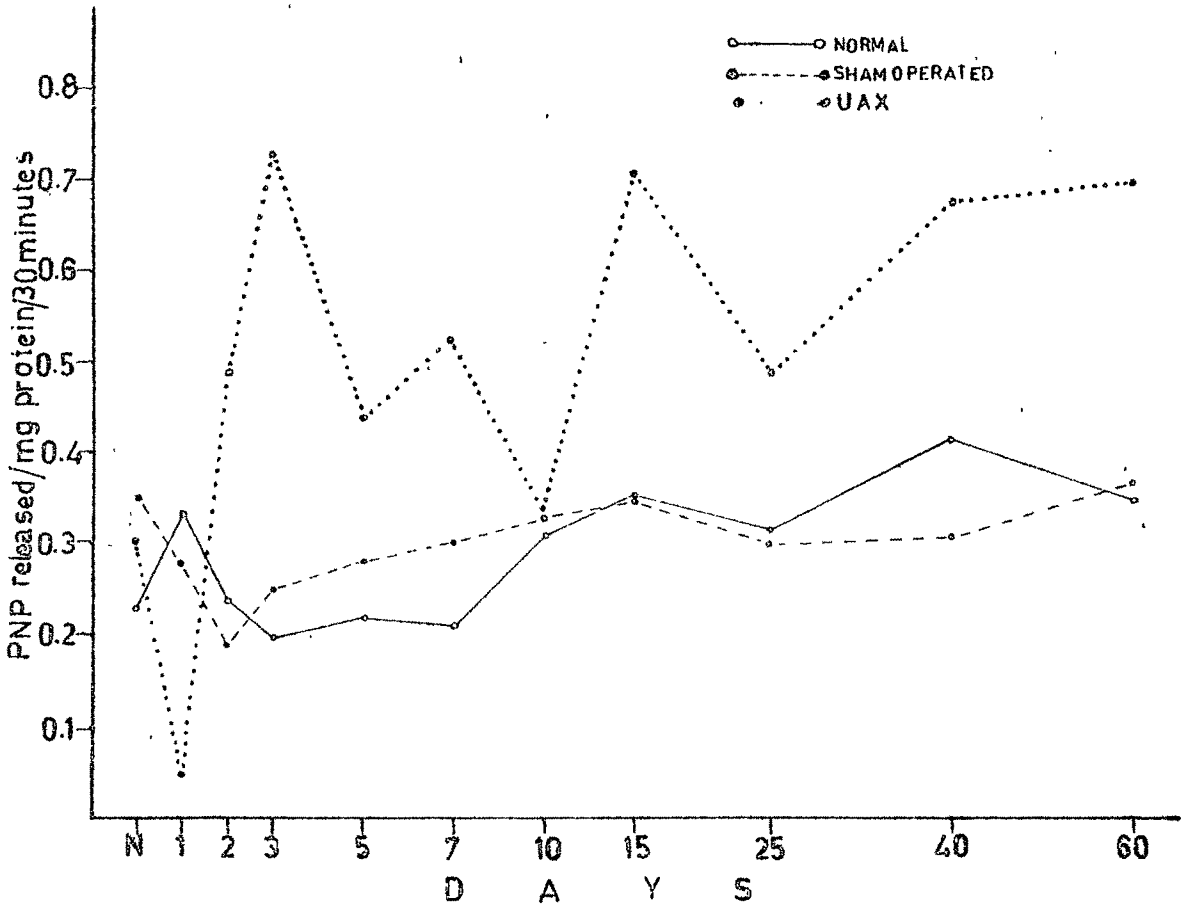

Fig. 14 : Graphic representation of the levels of acid phosphatase in the regenerate during tail regeneration in the normal, sham operated and unilaterally adrenalectomised lizards, *H. flaviviridis*

UAX - Unilaterally adrenalectomised.

Table 15. Levels of liver acid phosphatase (µ mole p-NP released/mg protein/30 minute) during tail regeneration in normal, sham operated and unilaterally adrenalectomised lizard, *H. flaviviridis*.

Periods of regeneration in days	N	1	2	3	5	7	10	15	25	40	60
Normal intact (IC)	0.768 ±0.019	1.1678 ±0.065	1.364 ±0.18	0.210 ±0.005	0.836 ±0.013	0.788 ±0.04	0.764 ±0.15	0.827 ±0.067	0.925 ±0.107	0.635 ±0.092	0.815 ±0.108
Sham operated (SUAX)	0.675 ±0.025	1.1137 ±0.197	1.1214 ±0.42	1.389 ±0.075	0.919 ±0.121	0.684 ±0.132	0.687 ±0.207	0.696 ±0.009	0.768 ±0.153	0.817 ±0.251	0.743 ±0.005
Unilaterally adrenalectomised (UAX)	0.913 ±0.045	0.878 ±0.017	0.7957 ±0.114	1.159 ±0.195	1.092 ±0.42	0.817 ±0.194	1.306 ±0.080	0.949 ±0.324	1.102 ±0.09	0.862 ±0.007	0.89 ±0.042

± S. D.

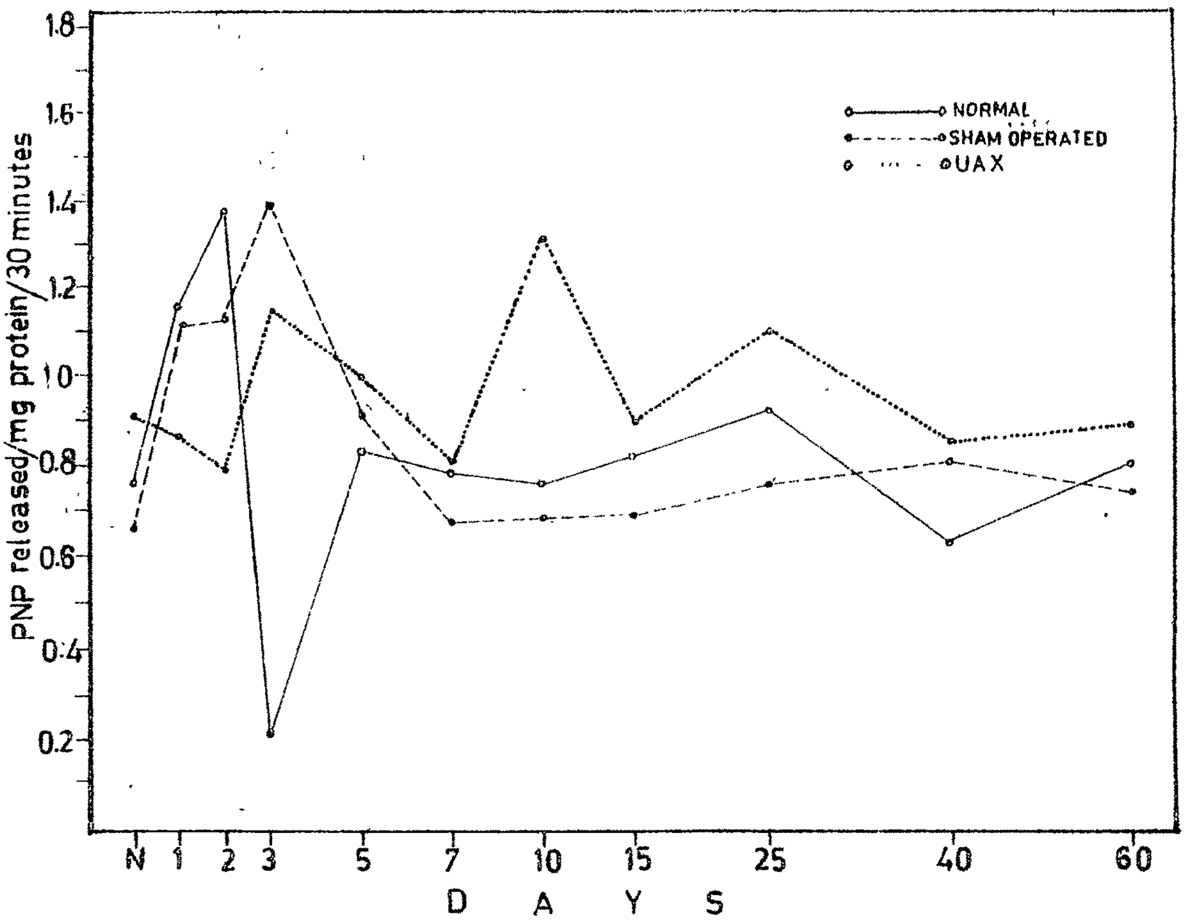

Fig. 15 : Graphic representation of the levels of acid phosphatase in the liver during tail regeneration in the normal, sham operated and unilaterally adrenalectomised lizards, *H. flaviviridis*

UAX - Unilaterally adrenalectomised.

Table 16. Levels of muscle acid phosphatase (μ mole p-NP released/mg protein/30 minute) during tail regeneration in normal, sham operated and unilaterally adrenalectomised lizard, *H. flaviviridis*.

Periods of regeneration in days	N	1	2	3	5	7	10	15	25	40	60
Normal intact (IC)	0.0706 ±0.006	0.2066 ±0.008	0.0344 ±0.0041	0.0191 ±0.001	0.0734 ±0.017	0.089 ±0.012	0.1225 ±0.009	0.109 ±0.087	0.0675 ±0.001	0.063 ±0.007	0.108 ±0.013
Sham operated (SUAX)	0.055 ±0.002	0.4014 ±0.108	0.0417 ±0.019	0.0373 ±0.005	0.1092 ±0.04	0.145 ±0.024	0.096 ±0.004	0.129 ±0.015	0.0789 ±0.014	0.073 ±0.006	0.053 ±0.009
Unilaterally adrenalectomised (UAX)	0.1802 ±0.014	0.1632 ±0.058	0.165 ±0.035	0.177 ±0.011	0.1231 ±0.014	0.162 ±0.08	0.149 ±0.007	0.126 ±0.053	0.208 ±0.018	0.217 ±0.016	0.208 ±0.042

± S. D.

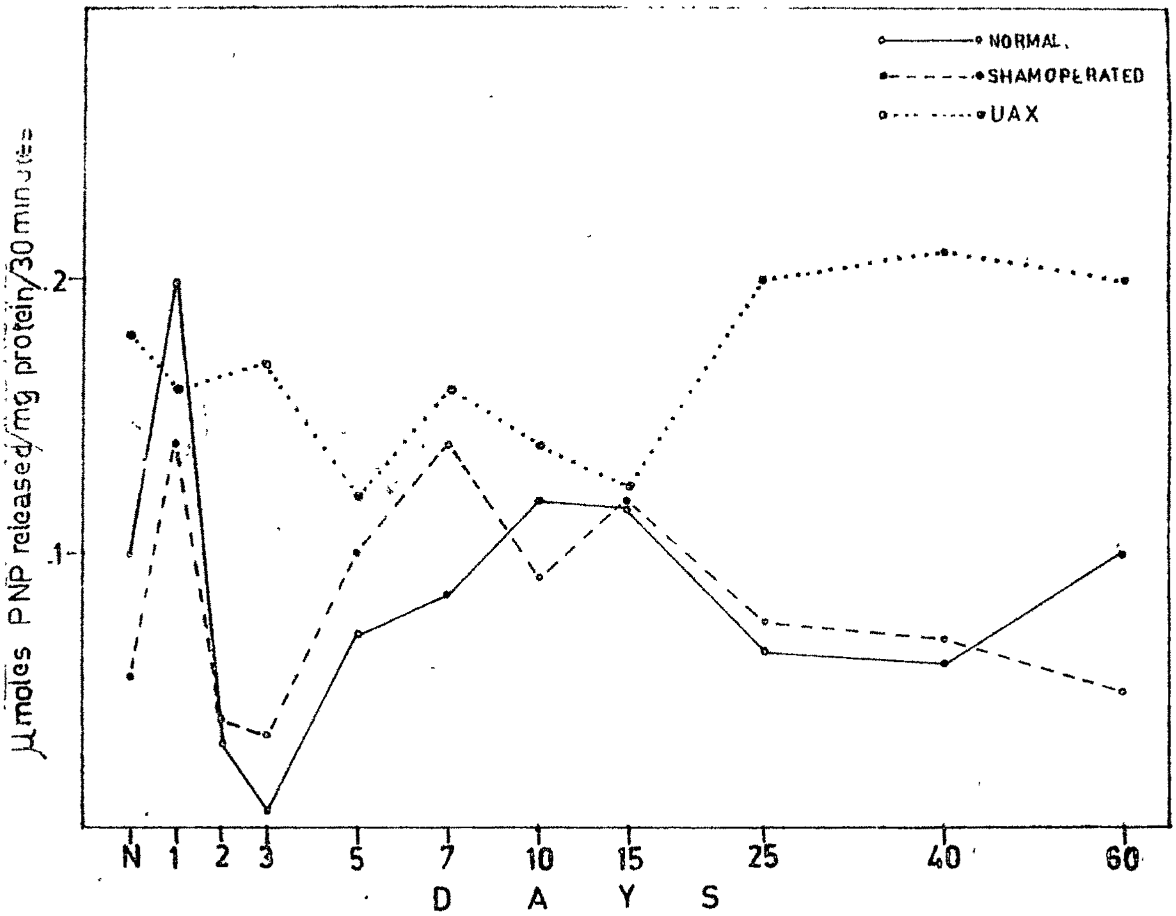

Fig. 16 : Graphic representation of the levels of acid phosphatase in the muscle during tail regeneration in the normal, sham operated and unilaterally adrenalectomised lizards, H. flaviviridis

UAX - Unilaterally adrenalectomised.

by a fall on 3rd day. On 5th day, it attained the maximum peak followed by a smaller peak on 10th day with a fall on the 7th day. Thereafter, the activity level of the enzyme decreased till 25th day followed by an increase. Sham operated ones also showed a more or less similar pattern in fluctuation in the activities of the enzyme. The initial high level activity of ATPase in UAX liver reduced on 1st day post-autotomy followed by a gradual increase till 5th day. There onwards its level gradually decreased till 15th day and then again increased. Muscle $Na^{+}K^{+}$ ATPase showed three peak values on 2nd, 5th and 10th days in the normal animals, while the sham operated animals showed peak values on 1st, 7th and 25th days. The initial high level of the enzyme activity in UAX showed a reduction on the 1st day. Peak levels were noted on 3rd and 7th days and a very minor peak on the 40th day. $Na^{+}K^{+}$ ATPase activity in the tail gradually decreased till the 2nd day in the UAX animals in contrast to the other two groups which showed a slight increase till the 3rd day resulting in a higher peak value. Thereafter, the enzyme level decreased reaching the minimum sub-normal level on 15th and 10th days respectively for the normal and sham operated lizards, and later into a slow rise thereafter. In UAX lizards also, there was a small rise in enzyme activity in their regenerate on 3rd day followed by a much higher peak level on 10th day, a steep fall on 15th day and a slow rise thereafter.

Table 17. Levels of caudal Na^+ K^+ ATPase (mg 'P' radicals liberated/mg protein/ 10 minute) during tail-regeneration in normal, sham operated and unilaterally adrenalectomised lizard, H. *flaviviridis*.

Periods of regeneration in days	N	1	2	3	5	7	10	15	25	40	60
Normal intact (IC)	5.0 ±0.152	4.519 ±0.12	5.028 ±0.21	12.322 ±0.32	5.267 ±0.18	4.299 ±0.14	5.719 ±0.08	1.34 ±0.14	2.498 ±0.11	5.044 ±0.093	3.725 ±0.0912
Sham operated (SUAX)	3.30 ±0.008	3.723 ±0.016	4.206 ±0.24	10.78 ±0.189	4.466 ±0.17	5.40 ±0.114	1.008 ±0.015	3.177 ±0.014	3.7 ±0.217	3.698 ±0.132	4.25 ±0.18
Unilaterally adrenalectomised (UAX)	4.061 ±0.12	2.448 ±0.048	1.559 ±0.034	5.818 ±0.125	4.437 ±0.213	5.10 ±0.18	10.362 ±0.278	2.632 ±0.017	3.65 ±0.043	2.656 ±0.0147	3.924 ±0.0153

± S. D.

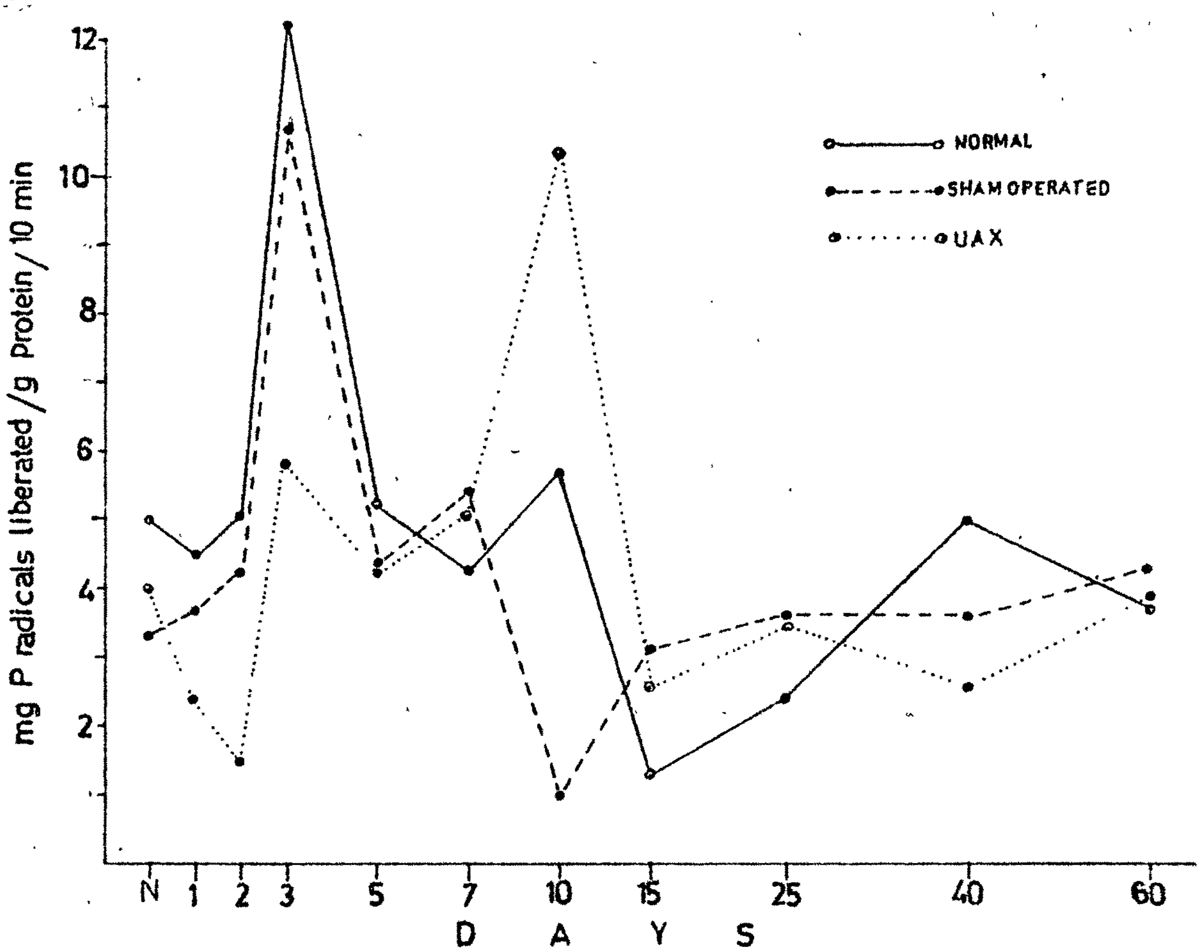

Fig. 17 : Graphic representation of the levels of Na^{+} K^{+} ATPase in the tail of the normal, sham operated and unilaterally adrenalectomised lizards, *H. flaviviridis*.

UAX - Unilaterally adrenalectomised.

Table 18. Levels of liver Na^+ K^+ ATPase (mg 'P' radicals liberated/ mg protein/10 minute) during tail regeneration in normal, sham operated and unilaterally adrenalectomised lizard, *H. flaviviridis*.

Periods of regeneration in days	N	1	2	3	5	7	10	15	25	40	60
Normal intact (IC)	1.125 ±0.0017	1.685 ±0.005	2.721 ±0.0034	1.867 ±0.001	3.834 ±0.008	1.892 ±0.0012	3.457 ±0.010	2.26 ±0.0076	0.926 ±0.003	1.414 ±0.0015	1.573 ±0.0011
Sham operated (SUAX)	1.071 ±0.0001	2.151 ±0.002	0.866 ±0.003	1.381 ±0.012	2.358 ±0.020	0.1688 ±0.0016	2.936 ±0.0023	1.323 ±0.0018	2.296 ±0.0024	1.542 ±0.0032	2.24 ±0.021
Unilaterally adrenalectomised (UAX)	1.662 ±0.02	0.261 ±0.00114	1.333 ±0.0008	1.652 ±0.002	3.654 ±0.0015	1.874 ±0.004	0.800 ±0.0004	0.525 ±0.0012	0.679 ±0.0007	0.77 ±0.0013	2.293 ±0.0014

± S. D.

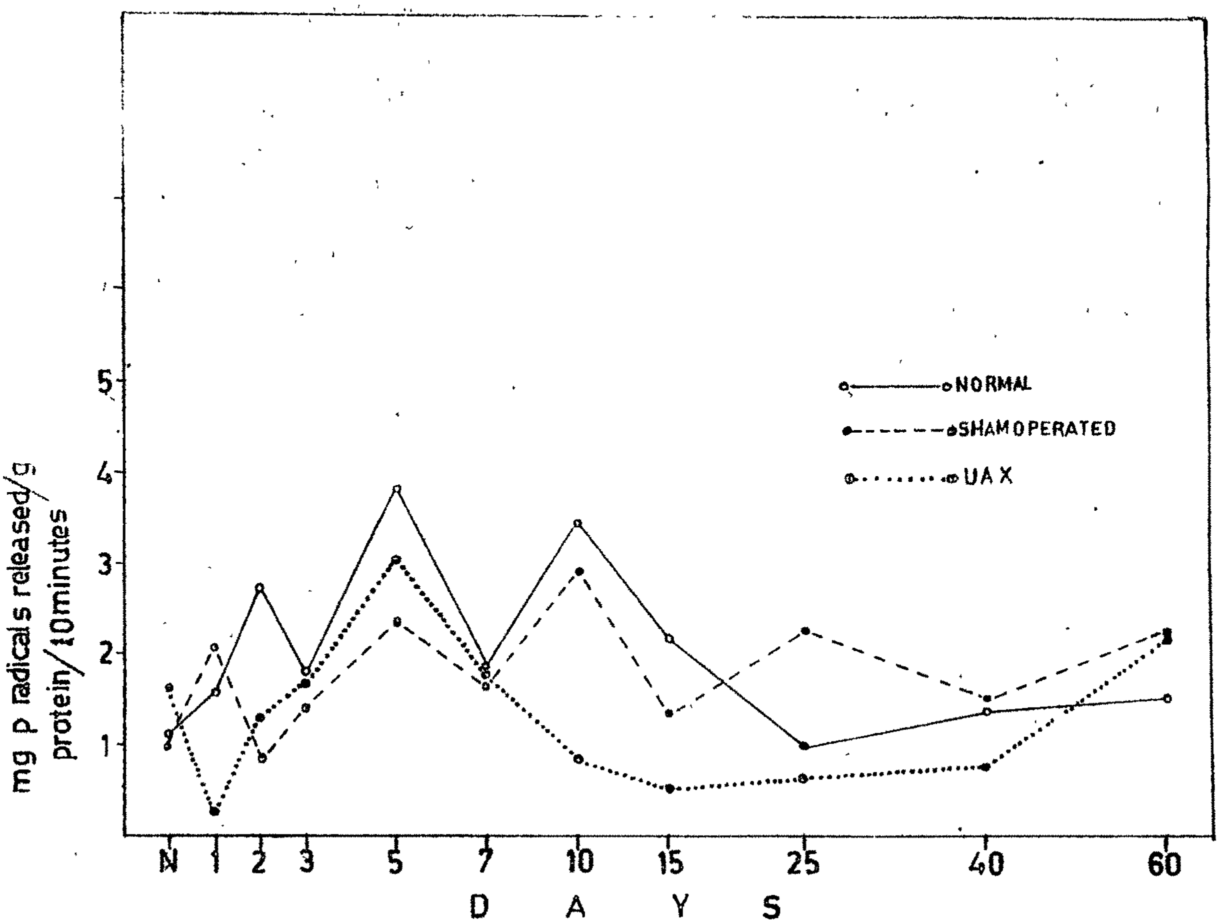

Fig. 18 : Graphic representation of the levels of Na^{+} K^{+} ATPase in the liver during tail regeneration in the normal, sham operated and unilaterally adrenalectomised lizards, H. *flaviviridis*

UAX - Unilaterally adrenalectomised.

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Table 19. Levels of muscle Na^{+} K^{+} ATPase (mg 'P' radicals liberated/ mg protein/10 minute) during tail regeneration in normal, sham operated and unilaterally adrenalectomised lizard, *H. flaviviridis*.

Periods of regeneration in days	N	1	2	3	5	7	10	15	25	40	60
Normal intact (IC)	2.38 ±0.12	2.473 ±0.08	5.73 ±0.007 0.0005@	3.41 ±0.0041	5.391 ±0.0145 0.0005@	2.452 ±0.005	4.593 ±0.0018	3.218 ±0.00192	2.891 ±0.020	1.827 ±0.011	2.315 ±0.0132
Sham operated (SUAX)	1.321 ±0.05	2.785 ±0.101	1.894 ±0.025	2.872 ±0.006	3.923 ±0.047	4.293 ±0.008	3.977 ±0.0156	2.795 ±0.136	4.163 ±0.0058	1.881 ±0.121	2.13 ±0.0155
Unilaterally adrenalectomised (UAX)	2.602 ±0.011	2.31 ±0.0042	2.78 ±0.031	5.26 ±0.0055	1.59 ±0.0172	2.944 ±0.121	2.032 ±0.082	1.524 ±0.0131	1.283 ±0.0124	2.498 ±0.032	1.466 ±0.0062

± S. D.

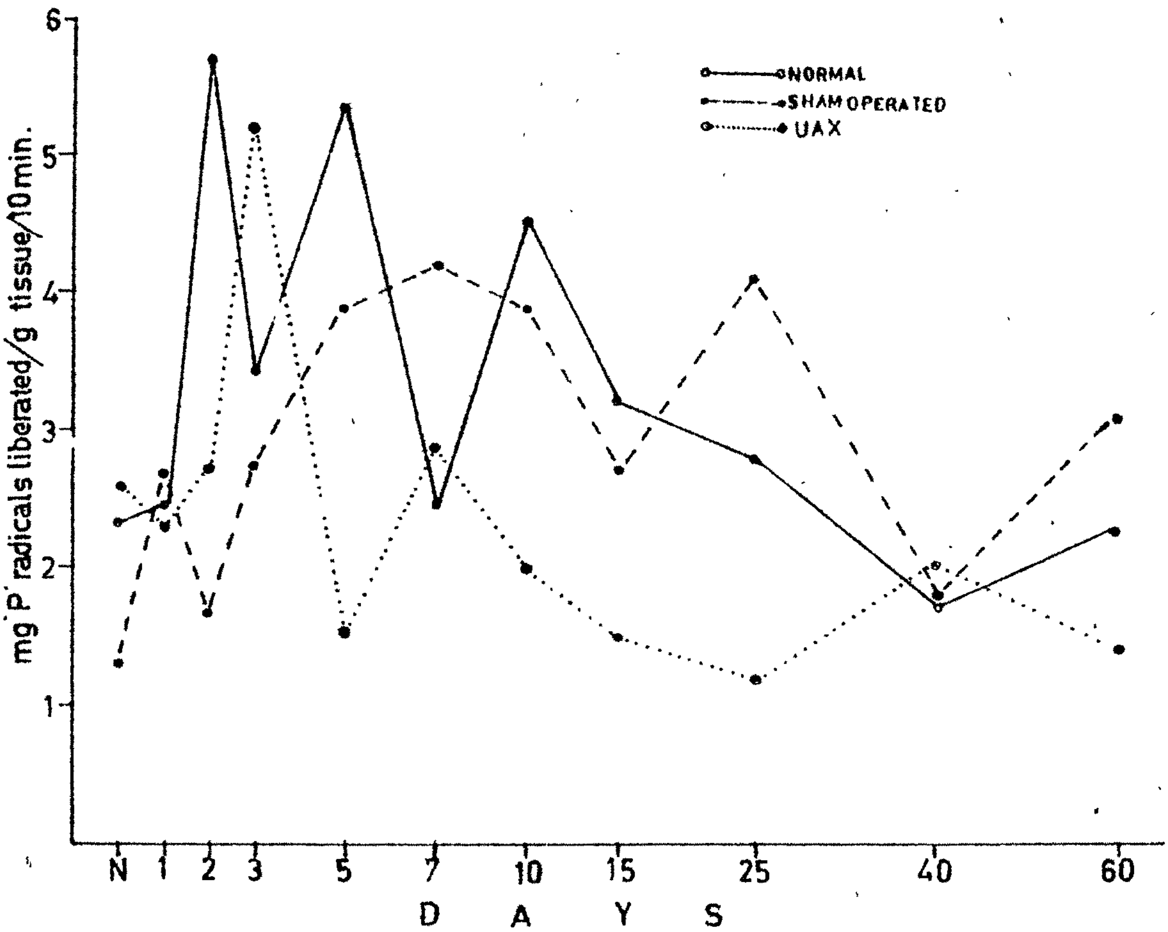

Fig. 19 : Graphic representation of the levels of Na^+ K^+ ATPase in the muscle of the normal, sham operated and unilaterally adrenalectomised lizards, <u>H</u>. <u>flaviviridis</u>

UAX - Unilaterally adrenalectomised

DISCUSSION

The initial alterations in various parameters noted to occur in UAX animals can easily be considered to be due to the induced insufficiency of adrenal hormones. The slight delay in the initial stage during regeneration must in all probability to be due to the insufficiency of the adrenal hormones rendered by unilateral adrenalectomy. By and large, similar pattern of changes noticed in the various parameters studied as well as the attainment of the normal growth rate during the later stages in UAX lizards indicate the compensatory overproduction of hormones by the remaining adrenal. Such compensatory activity of remaining adrenal is known to occur (Banerjee and Quay, 1981; Ray, 1980; Matilde et al., 1980). This was proved by the observed histological and morphologic studies of the remaining adrenal (unpublished). The development of an early blastema in the sham operated lizards must be geared up metabolic activities in the animal due to the abdominal wound. Adrenal steroids (glucocorticoids) are speculated to stimulate the break down of proteins by stimulating the synthesis of membrane proteins essential to the autophagic process in cultured hepatocytes (Hopgood et al., 1981) and also bring about an increase in the number of lysosomes (Carubelli and Griffin, 1970; Hopgood and Ballard, 1980). As phagocytosis is an inherent mechanism during the wound healing process preceeding the formation of blastema, the delayed blastema formation in the UAX group could be due to the prevailing hypoadrenalism in the early days after autotomy.

From the results it is obvious that in the initial period after unilateral adrenalectomy, there was a delay in the regenerative mechanics, while during the later periods, not only did the various parameters under investigation return to the normal pattern but even the tail regenerate was better formed in girth. This has to be due to the hypoadrenalism in the initial periods and the compensatory hypersecretion by the intact adrenal during the later periods. Apparently, involvement of adrenal can be surmised and the idea has to be put to experimental evaluation by studying the impact of total suppression of adrenal cortex by a suitable technique on tail regeneration in H. flaviviridis.