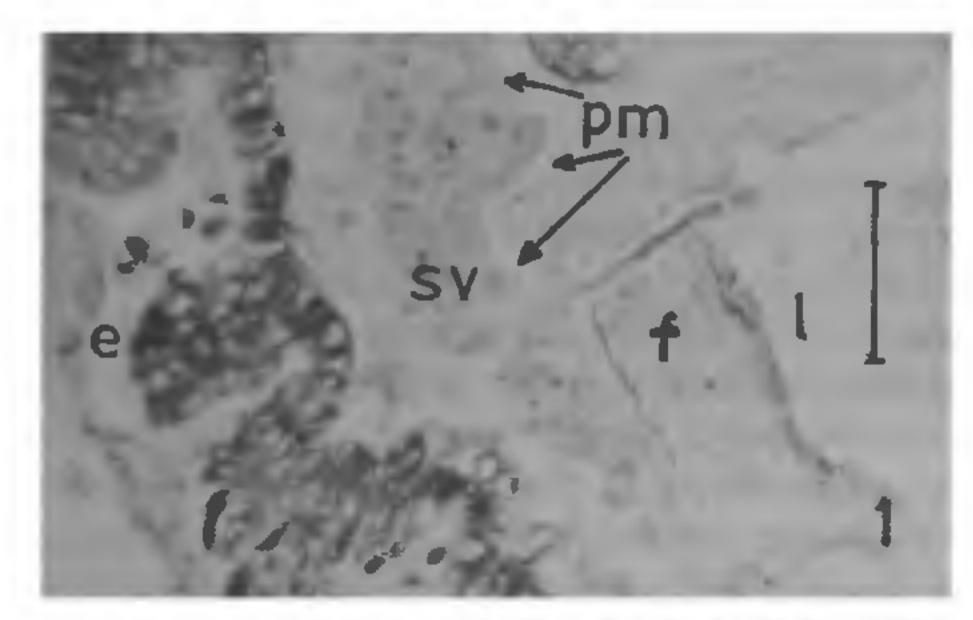
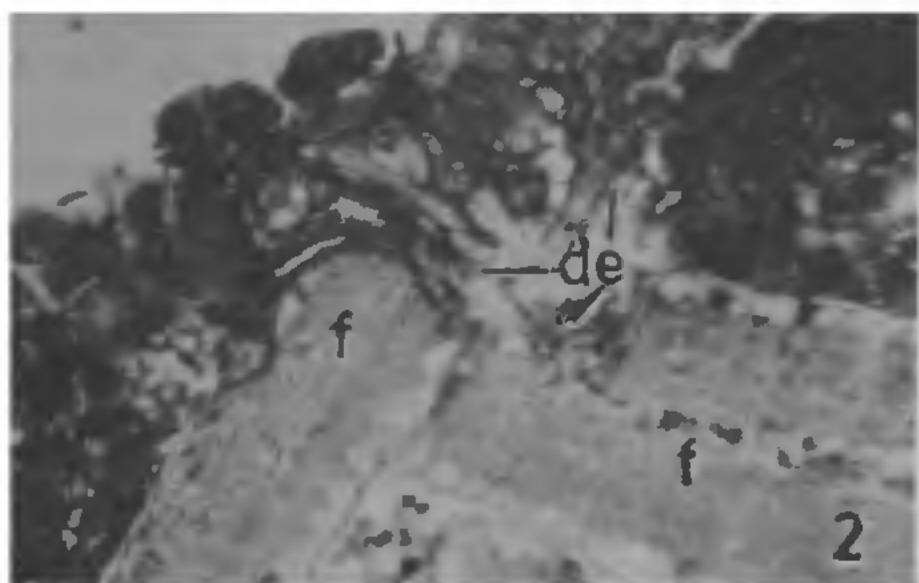
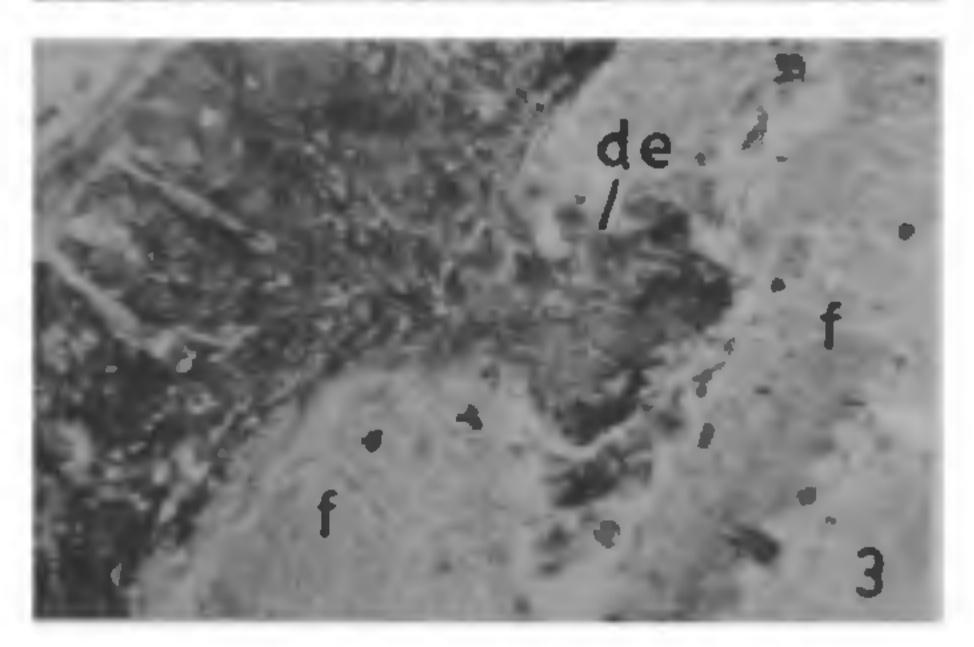
comprised by angular fragments of mulberry leaves; there is no direct contact between the food particles and the epithelium; digestive secretions and the







Figures 1-3. Transverse sections of midgut. 1. From normal larva; note the peritrophic membrane which separates the food from the epithelium; the gap is about $20-30\,\mu\mathrm{m}$ in width. The epithelial layer is continuous and composed of one layer of cells which is folded in some regions; 2, 3. From the larva lacking a peritrophic membrane. Note the direct contact between the epithelium and fragments of leaves which have pared off the epithelial cells; the extensive damage and the debris of cells can be seen; the epithelial layer is unusually thick in this larva. e-epithelium; d.e.-damaged epithelium; f-food; l-lumen; p.m.-peritrophic membrane; s.v.-secretory vesicle. The scale line represents $20\,\mu\mathrm{m}$.

digested food pass through the peritrophic membrane; the epithelial layer is continuous and smooth (figure 1).

In the freak larva, there was no peritrophic membrane. The passage of closely packed fragments of leaves aided by the peristalsis of the gut had abraded the epithelium; as a result the cells were found pared and damaged. The cells thus damaged and dislodged collected as irregular heaps within the lumen of the midgut. The integrity of the epithelium was lost and the food particles were jutting against the damaged cells (figures 2 and 3).

It is not clear how the larva survived the repeated abrasion and reached the fifth-instar stage. Probably, the regenerative cells in the epithelium were remarkably active in this larva in repairing the tissues and in coping with the enormous rate of destruction of the epithelial cells as evident from the unusually thick layer of the epithelium.

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- 1. Chapman, R. F., The insects: Structure and function, ELBS & English University Press, London, 1973.
- 2. Imms, A. D., A general text book of entomology, Asia Publishing House, Bombay, 1963.
- 3. Wigglesworth, V. B., The principles of insect physiology, ELBS & Chapman & Hall, London, 1977.
- 4. Mercer, E. H. and Day, M. F., *Biol. Bull.*, 1952, 103, 384.

NEUROENDOCRINE CONTROL OF PROTEIN AND AMINOACID LEVELS IN THE BLOOD OF FRESHWATER CRAB, BARYTELPHUSA GUERINI (H. MILNE EDWARDS) (DECAPODA, POTAMIDEA)

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Studies on Hemigrapsus nudus showed that the eyestalk principle diverts metabolism towards general growth and its loss through sinus gland removal led to a decrease in the whole animal protein content suggesting the acceleration of cata-

bolic processes in the tissues. Eyestalk removal led to a decrease in the total nitrogen content, amylolytic activity and RNA content in Procambarus clarkin² and Barytelphusa cunicularis³, while eyestalk injections into eyestalk ablated animals increased the RNA levels. Histochemical studies on hepatopancreas of Scylla serrata showed that glycoproteins and glycolipids were depleted following eyestalk ablation. Variations in free aminoacid levels in relation to eyestalk factors were also noticed in a few crustaceans⁵⁻⁷. Therefore, the effect of eyestalk ablation and eyestalk injection into ablated animals on total protein and aminoacid levels of blood and the effect of other neuroendocrine structures were studied to understand the neuroendocrine control of protein and aminoacid levels of blood in the freshwater crab, Barytelphusa guerini.

The collection and maintenance of the freshwater crab, B. guerini for experimentation were described earlier⁸. Males weighing between 30 and 50 g were used for the experiments carried out at room temperature (26–28°C). The total protein and aminoacid levels of blood were estimated quantitatively^{9,10} in the normal animals with intact eyestalks. Another group of laboratory adapted crabs was employed for eyestalk ablation. After 48 h of eyestalk ablation, they were divided into five batches.

One batch of eyestalk ablated animals was used to estimate the total protein and aminoacid levels to serve as control (C). Extracts of eyestalk, sinus gland, brain and thoracic ganglionic mass were injected into the remaining four experimental batches constituting E₁, E₂, E₃ and E₄ groups respectively. Protein and aminoacids were analysed after 48 h of extract injection.

Eyestalk ablation and preparation of different neuroendocrine extracts were according to the procedure outlined elsewhere⁸. Blood (1 ml) was drawn with the help of hypodermic syringe rinsed with sodium oxalate from the base of the chelate leg. The total protein content was determined⁹ and aminoacids were quantified¹⁰ with deproteinized blood. A minimum of 6 animals were used for each observation and the results statistically analysed using Student's t test.

Bilateral extirpation of eyestalks led to a decrease in total protein (45%) and aminoacid (51.42%) levels of the blood after 48h of eyestalk ablation. This decrease was highly significant (P < 0.01) when compared with normal values. The injection of eyestalk extracts into eyestalk ablated animals ele-

vated the protein and aminoacid levels by about $48\,\mathrm{h}$. The recovery in levels was almost complete, such that the variations in levels of experimental animals (eyestalk extract injections received) were insignificant (P > 0.1), when compared with normal animals with intact eyestalks and were highly significant (P < 0.01) in respect of the control eyestalk ablated animals (figure 1A, B).

Injections of extracts of sinus gland, brain and thoracic ganglionic mass into the eyestalk ablated animals did not alter the protein and aminoacid levels of blood. Hence, the two levels obtained in the experimental animals (extract injections received) after 48h of extract injections were significantly (P < 0.01) different, when compared with normal intact animals and were insignificant (P > 0.1), when compared with eyestalk ablated control animals (figure 1A, B).

The principal protein of the blood is haemocyanin and its decrease after eyestalk ablation may be correlated with the decreased metabolic rate after eyestalk ablation and the lesser need for oxygen transportation through blood⁸. Earlier studies on Astacus astacus⁵ and Callinectes sapidus⁶ showed that eyestalk ablation facilitates incorporation of aminoacids into tissue proteins and their utilization

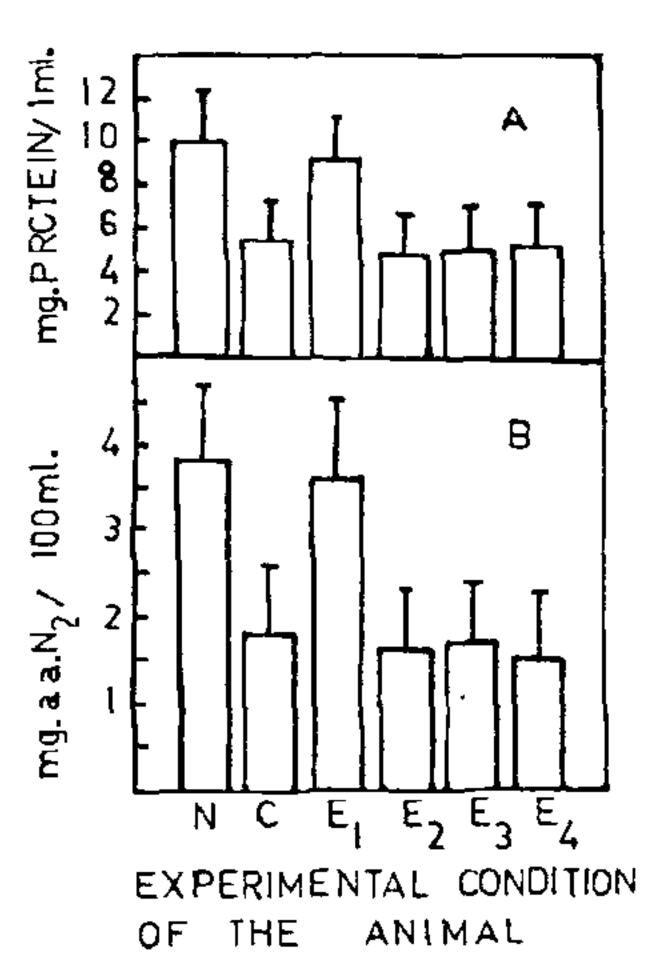


Figure 1. Protein (A) and total free aminoacids (B) contents in the blood of the crab in the normal intact eyestalks condition (N), on eyestalk ablation (C) and injection of extracts of eyestalks (E_1) , sinus gland (E_2) , brain (E_3) and thoracic ganglionic mass (E_4) into eyestalk ablated animals. (Values shown are the average of 6 observations \pm S.D.)

for metabolic energy release. In B. guerini eyestalk removal leads to decrease in free aminoacid contents in different tissues like muscle, gill, hepatopancreas and heart, which were incorporated by increase in the total protein contents of the tissues¹¹. The decreased blood glucose level in this crab after eyestalk ablation is possibly due to the stimulation of glycogenesis¹². There is also evidence to show that eyestalk ablation leads to increased lipid synthesis and gluconeogenesis^{4,13–15}. In fact, the utilization of aminoacids as an important source of energy-producing compounds is well-demonstrated amongst the crustaceans¹⁶. It may therefore be suggested that decreased aminoacid contents in the blood after eyestalk ablation may be due to their catabolism for energy release in the absence of sugars, because of its utilization in the process of lipid synthesis and glycogenesis. As such variations in total aminoacids seen in the present study are justifiable and their utilization for energy release and incorporation into tissue proteins seems to be under the control of eyestalk principle.

The sinus gland extracts did not change protein and aminoacid levels suggesting that the factor influencing the protein metabolism is not present in the sinus gland. It is possible that the factor produced in the X-organ complex of the eyestalk is released directly into the blood stream instead of being stored in the sinus gland. The central nervous structures also did not affect the protein and aminoacid levels suggesting the absence of any factors in these structures which would influence protein metabolism. The present study thus shows that the protein and aminoacid levels are under the control of eyestalk.

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- 1. Neiland, K. A. and Scheer, B. T., *Physiol. Comp. Oecol.*, 1953, 3, 321.
- 2. Fingerman, M., Dominiczak, T., Miyawaki, M., Oguro, C. and Yamamoto, Y., Physiol, Zool., 1967, 40, 23.
- 3. Nagabhushanam, R. and Diwan, A. D., J. Anim. Morphol. Physiol., 1974a, 21, 35.
- 4. Rangneker, P. V. and Momin, M. A., Mikrosk. Anat. Forsch. Leipzig., 1974, 88, 874.
- 5. Zandee, D. I., Arch. Int. Physiol. Biochem., 1966, 74, 614.
- 6. Tucker, R. K. and Costlow, J. D. (Jr), Comp.

- Biochem. Physiol. A. Comp. Physiol., 1975, 51, 75.
- 7. Mcwhinnie, M. A. and Mohrherr, C. J., Comp. Biochem. Physiol., 1970, 34, 415.
- 8. Vasantha, N., Gangotri, M. S. and Venkatachari, S. A. T., Indian J. Exp. Biol., 1979, 9, 974.
- 9. Lowry, O. H., Rosebrough, N. J., Farr, A. L. and Randall, R. J., J. Biol. Chem., 1951, 193, 265.
- 10. Oser, B. L., Hawk's physiological chemistry, (ed.) B. L. Oser, McGraw-Hill, New York, 1965, p. 1048.
- 11. Gangotri, M. S., Ph.D. thesis, Marathwada University, Aurangabad, 1980.
- 12. Gangotri, M. S., Venkatachari, S. A. T. and Vasantha, N., Crustaceana, 1987, 53, 5.
- 13. Rangneker, P. V. and Madhyastha, M. N., Indian J. Exp. Biol., 1971, 9, 462.
- 14. Diwan, A. D., Marathwada Univ. J. Sci., 1973, 12, 279.
- 15. Madhyastha, M. N. and Rangneker, P. V., Hydrobiologia, 1976, 48, 25.
- 16. Munday, K. A. and Poat, P. C., Chemical zoology, (eds) B. T. Scheer and M. Florkin, Academic Press, New York, 1971, Vol. 6, p. 191.

RAT UTERINE BIOASSAY FOR THE RESIDUAL EFFECT OF THE ADMINISTERED TESTOSTERONE ACETATE IN THE CARP, CYPRINUS CARPIO (L.)

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To achieve higher growth rate and to induce sex reversal/sterility in fish, gonadal hormones are employed in aquaculture experiments^{1,2}. One of the impediments for the use of steroids in aquaculture is the lack of information on residual concentrations of the used exogenous hormone in fish destined for human consumption.

In the present investigation, bioassay studies were conducted to detect the effect of residual hormonal concentration in the muscle of the sex steroid-treated fish, at the end of the 14-month post-