Influence of Recurrent Nerve Severance on Haemolymph Proteins in Periplaneta americana L.

The median neurosecretory cells, as well as the corpus allatum, have been found to influence protein metabolism in insects 1-5. The role of the recurrent nerve which innervates the gut of insects⁶ has not been investigated in this connection, although it is an important pathway to the gut for neural and hormonal stimuli⁷⁻⁹. Hence it was thought worthwhile to investigate the effect of recurrent nerve severance on the haemolymph protein concentration in an insect, as haemolymph is the likely medium to be affected by any change in protein meta-

Female cockroaches (Periplaneta americana) under ether anaesthesia were surface sterilized with 80% ethyl alcohol at the site of operation. The recurrent nerve was severed just behind the nervi cardio-stomatogastrici through an incision made on the dorsal sclerite of the neck. After introducing a crystal of streptomycin sulphate, the wound was sealed with molten paraffin. Controls were kept in which all procedures were carried out except nerve severance. The animals had free access to biscuits and water. All animals were bled between 115 and 145 days after the operation by pleural puncture. The blood was collected in capillary tubes, sealed with plasticine and centrifuged at 0 °C in a refrigerated centrifuge. The clear serum was used for disc electrophoresis after the method of Reisfeld et al.10. Stained electrophorograms were scanned using a Canalco microdensitometer and the quantities of proteins in different fractions estimated against similar preparations of a crystalline bovine serum albumin standard.

The experimental animals were fed as well as the controls. The haemolymph protein patterns of the experimental and control animals are shown in the Figure, and the concentrations of proteins in different fractions are summarized in the Table. Of the total of 9 protein fractions found in the haemolymph of the female cockroach¹¹, in fractions 3 and 4 which are the major fractions, there is a conspicuous fall in concentration of proteins. Fractions 2, 4, 5, 6, 7 and 8 are not at all recognizable in the experimental animal. The average

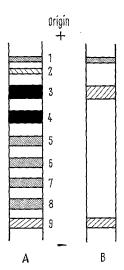


Diagram representing electrophorograms of the haemolymph proteins of Periplaneta. (A) control female; (B) female 143 days after recurrent nerve severance. Dark shading indicates highest concentration of proteins in a fraction; cross hatching and stipples, lesser and east concentration respectively.

Concentration of proteins in $\mu g/\mu l$ in different fractions of the haemolymph of Periplaneta americana 115-145 days after recurrent nerve severance

Experimental groups	Major fractions 3 and 4	Minor fractions	Total
I. Females after recurrent	11.3 (7.8, 3.5)	6.7	18.0
II. Sham operated controls	35.6 (30.3, 5.3)	17.7	53.3

Crystalline bovine serum albumin was taken as standard. Each value represents an average of at least 5 animals.

total protein concentration in the experimental animal is 18.0 μ g/ μ l which is only 34% of the control value.

The haemolymph protein concentration in insects is known to decrease after extirpation of the neurosecretory cells 3, 12, 13, as these components of the neuroendocrine complex of insects stimulate the synthesis of gut proteases⁵ or protein synthesis by the fat body. The gut in insects, especially in cockroaches, is well innervated by the stomatogastric nervous system. Neurosecretory material is present along these nerves in aphids and cockroaches 8, 9. These authors also suggest the possibility of direct release of neurosecretory material contained in the stomodaeal nerves at the site of action, i.e. the gut. Hence it is presumed that the low protein concentration in the haemolymph of Periplaneta observed in the present study is due to a nervous or a hormonal disturbance of the synthesis of enzymes involved in protein digestion. It is concluded that recurrent nerve severance in female cockroaches results in a decrease of the haemolymph protein concentration due to decreased protease activity of the

Zusammenfassung. Es wird gezeigt, dass bei der weiblichen Schabe Periplaneta americana Durchtrennung des Nervus recurrens zu einer Reduktion der Hämolymphproteine führt.

V. K. K. Prabhu and P. Hema

Department of Zoology, University of Kerala, Kariavattam, Trivandrum (India), 5 May 1969

- ¹ F. Engelmann, Ann. Rev. Entomol. 13, 1 (1968).
- ² F. Engelmann and D. Penney, Gen. comp. Endocrin. 7, 314
- ³ K. C. Highnam, O. Lusis and L. Hill, J. Insect Physiol. 9, 587 (1963).
- ⁴ K. K. Thomas and J. L. Nation, Biol. Bull. 130, 254 (1966).
- ⁵ E. Thomsen and I. Möller, J. exp. Biol. 40, 301 (1963).
- ⁶ R. B. WILLEY, J. Morph. 108, 219 (1961).
- ⁷ K. G. Davey and J. E. Treherne, J. exp. Biol. 40, 775 (1963).
- ⁸ B. Johnson, J. Insect Physiol. 9, 727 (1963).
- B. Johnson and B. Bowers, Science 141, 264 (1963).
 R. A. Reisfeld, V. J. Lewis and D. E. Williams, Nature 195, 281 (1962).
- 11 K. G. ADIYODI and K. K. NAYAR, Curr. Sci. 35, 587 (1966).
- ¹² L. Hill, J. Insect Physiol. 8, 609 (1962).
- ¹³ R. R. MILLS, F. C. GREENSLADE and E. F. COUCH, J. Insect Physiol. 12, 767 (1966).
- 14 We thank Prof. K. K. NAYAR for guidance, the Ford Foundation for a generous grant, and the University of Kerala for a research training fellowship to one of us (PH).