

Indian Grassland and
Fodder Research Institute
Jhansi (U. P.), India

K. C. PANDEY and S. A. FARUQUI

Studies on the neuro-endocrine complex of *Bagrada hilaris* BURM. (Hemiptera: Pentatomidae)

With 6 text figures

Introduction

In the recent past, the work on the structural details of the associated structures in the retro-cerebral neuroendocrine complex has advanced tremendously. 'Lame fibrogliale' (CAZAL, 1948) or 'Cardiogliale' (JUNQUA, 1956) or 'Carpus cardiacum' was for long considered as the main neurohaemal organ for cerebral NSM. (NAYAR, 1956; EWEN, 1962; WIGGLESWORTH, 1964; GUPTA, 1970). But the latter workers, (DOGRA, 1967 a, b, 1969; SRIVASTAVA, 1970; AWASTHI 1973 a, b and FARUQUI, 1974, 1975 a, b, 1977 a, b) have suggested that it is the aorta and not CC that acts as a neurohaemal organ for the NSC of Median group. NORMAN, 1965 and THOMSEN, 1969, in *Calliphora erythrocephala* and AWASTHI, 1972 b, in *Sarcophaga ruficornis* have attributed the same function to the aorta in insects.

In the present work an attempt has been made to work out the distribution of different types of NSC in the brain, sub-oesophageal ganglion, transport of NSM and structural details of the endocrine glands. These findings will be later on correlated with the various physiological and behavioral aspect of this important pest insect.

Material and techniques

The adults and the developing nymphs were collected in February—March. In situ stained preparations were made with Performic acid-victoria blue technique — PAVB (DOGRA & TANDON, 1964). The histological preparations were made using, i. paraldehyde fuchsin- PF (Harker's modification-pers. communication) ii. chrome alum-haematoxyline-phloxine -CAHP, (GOMORI, 1941), iii. alcian blue-phloxine, ABP, (SLOPER, 1957). The classification of NSC is based on the classification proposed by GUPTA & FARUQUI (1984).

Observations

The morphological association of different structures are almost the same as described for gymnoceretan Heteropterans (FARUQUI, 1975 a, b). However the components show the differences as described below.

a. Median Neurosecretory cells (MNC): These are the most conspicuous cells located in the pars intercerebralis on either side of the mid line just below the peri-

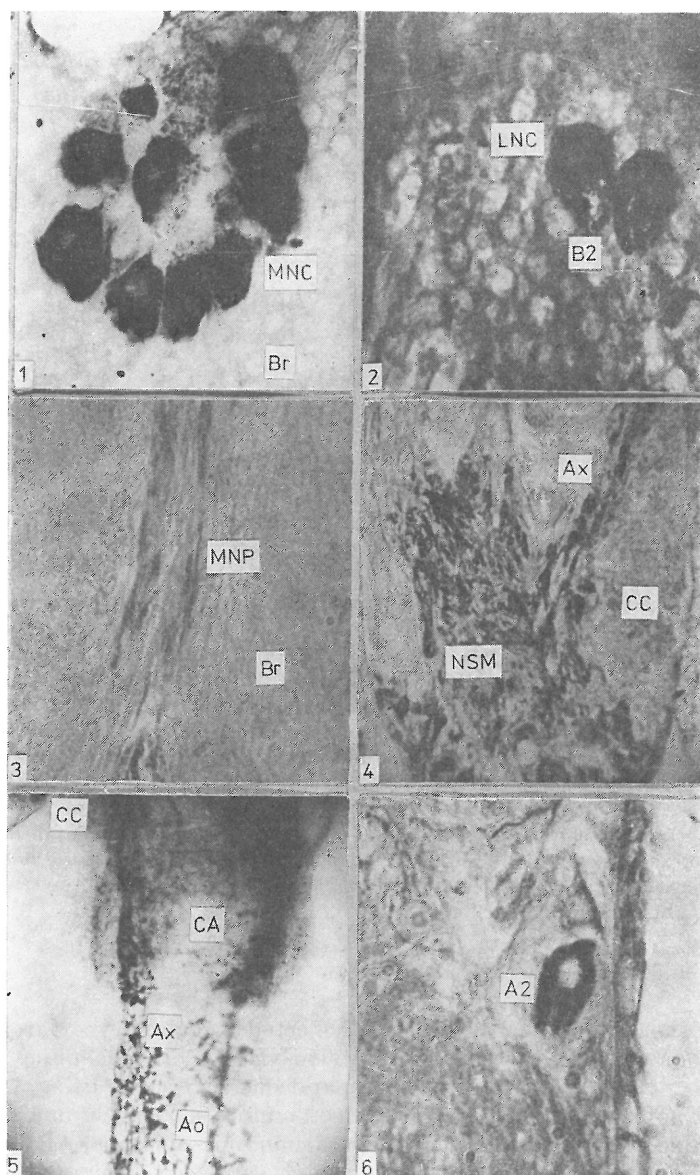


Plate I. 1-6: 1 - Section through pars-inter cerebralis medialis of *B. hilaris*, showing MNC, (type A₁ & B₁) (PF stain $\times 450$). 2 - Lateral neurosecretory cells of *B. hilaris* showing A₂ & B₂ type NSC. (PF $\times 450$). 3 - Section brain of *B. hilaris* showing median neurosecretory pathways. (CAPH $\times 450$). 4 - Section through retrocerebral endocrine complex showing axons running along the margin of CC with NSM in the aorta. (PF $\times 450$). 5 - In situ preparation of endocrine-aorta complex of *B. hilaris*, showing relationship and path structure of the axons. (PAVB $\times 100$). 6 - Section through pars-intercerebralis lateralis of *B. hilaris* showing lateral NSC (PF $\times 450$).

lemma and above the central body. There are about 5–6, A_1 type cells and 2–3, B_1 cells forming a group of about 8 cells in each of the sides (Fig. 1). Average cell measurement are: cell diameter $28\ \mu$, nuclear diameter $14\ \mu$ for A_1 type and $25\ \mu$ – $10\ \mu$ for type B_1 cells.

These cells are noted in different stages of synthesis, secretion and storage. The nucleus in these cells is centrally placed with varying amount of chromatin material. The nucleus is large and marginal in position. The axons are conspicuous and the paths are traceable.

b. Lateral Neurosecretory cells: This group is located in the pars-intercerebralis lateralis of the protocerebrum. They are in group of 4–6 cells belonging to the A_2 and B_2 type of *NSC*. They measure $26\ \mu$ – $12\ \mu$ and $24\ \mu$ – $10\ \mu$ respectively in their cell and nuclear diameters (Fig. 2).

The nucleus is centrally placed, but axonal or abaxonal positions are not very uncommon. The axons are not very conspicuous and it is difficult to trace out their pathways.

These cells show definite secretory activity in late nymphal and adult stages of the insect.

c. Tritocerebral Neurosecretory cells: Few cells of B_2 , C and D are seen to be scatteredly located in the tritocerebral region. They measure $26\ \mu$ – $12\ \mu$, $32\ \mu$ – $18\ \mu$ and $30\ \mu$ – $15\ \mu$ respectively in the diameter (cell-nucleus).

The nucleus is mostly centrally placed in these cells. Only the B_2 cells show secretory activity. Axons are not traceable after a while.

d. *NSC* of sub-oesophageal ganglion: Two very prominent cells stainable with PAVB are seen lying on either side of mid line on the ventral surface (Fig. 6). These cells are similar to the A_1 cells of the median group in staining behaviour but are smaller in size. They measure $20\ \mu$ – $7\ \mu$ in cell, nuclear diameter. Both of these cells give off one axon each, which are directed first towards the dorsal side and then travel anterior wards. Few cells of C and D type scattered were also noticed.

e. The neurosecretory pathway: There are two distinct pathways, that carry the *NSM* elaborated from *NSC* to the neurohaemal organ and return to the target organs.

i. Median *NS* pathway (*MNP*) and Nervi corporis cardiaci I (*NCCI*)— The axons of Median *NSC* converging to form the *MNP*. These pathways after running over a very short distance cross over each other so that the axons of left lobe run on the right side of proto and deutocerebrum emerge out of brain from tritocerebrum as Nervi corporis cardiaci I (*NCCI*) (Fig. 3). The *NCCI* after running free for a very short distance enters the dorsal aorta. There is no connection axonal or otherwise between *NCCI* and the *CC* (Fig. 4). The axons after entering in the aorta gives off a number of secondary axons which stores the neurosecretory material in form of granules running some distance in the inner thick layer of the loose tissues of the aorta (Fig. 5).

ii. Lateral *NS* Pathway (*LNP*) and Nervi corporis cardiaci II (*NCCII*) —: The axons emerging out of the lateral groups of *NSC* join to form the *LNP*. These converged axons travel the distance of the brain and emerge out of the brain from the tritocerebrum on outer side of *NCCII*. These axons are noticed to enter the corpus cardiacum from the apical end on each side.

f. The endocrine glands: A pair of corpus cardiacum (*CC*) and a single corpus allatum (*CA*) are seen lying ventro-lateral to the aorta and dorso-lateral to the gut. The *CC* of the two sides get fused on the poterio-ventral surface at the point of attachment with *CA*.

i. Corpus cardiacum: Histologically the CC consists of three type of cells, namely nerve cells, chromophobe cells and chromophil cells.

The chromophobe cells are irregular in shape with chromatin rich small nuclei. They measure about 6–8 μ with a nucleus of about 3 μ .

The chromophil cells are of two types on the basis of their size, arranged on the periphery of the glands, both on the anterior and posterior side. They measure 18–20 μ with a nucleus of 8–9 μ and 12–14 μ with a nucleus of 5–6 μ respectively. The chromophil cells show definite changes in nuclear size as also accumulation of the granular material in the cytoplasm. These cells stain intensely purple with PF but are phloxophilic with CAHP.

The CC thus secretes its own hormone along with storing secretion of the LNC, (Fig. 4).

ii. Corpus allatum (CA): It is a bilobed structure at the postero-dorsal part of CC. Histologically it is seen to consist of a single type of cells which measure between 10–14 μ with a nucleus of 6–7 μ . These cells have distinct cell boundaries with a centrally placed nucleus, varying amount of chromatin material is noticed in the nuclei. The cytoplasm is acidophilic. Secretory granules are seen in the cytoplasm around the nucleus.

g. Innervation of the endocrine glands: The CC is clearly seen to have receiving axons of NCC II. These are seen running in the intercellular spaces of the gland. The axons of NCC I are however seen to run attached to this gland but do not penetrate. They are however seen in the CA along with the axons of NCC II which enter the gland via nervi corporis allati (NCA).

Discussion

The retro-cerebral neuro-endocrine complex of *Bagrada hilaris*, as a whole falls in general pattern described for other Heteropteran insects (DOGRA, 1967a, b; SRIVASTAVA, 1970; FARUQUI, 1974, 1975a, b, 1977a, b).

The median and lateral NSC are distinct groups in this insect and not like those described for *Cyclopelta siccifolia* (FARUQUI, 1977a) in which both the groups lie very close to each other to form a single group of cells. This position is in agreement with those of other insects described by SAINI, (1966, 1971).

The type of cells described in this insect is based on a new classification proposed by GUPTA & FARUQUI (1984). Type A₁ and B₁ are present in the median group, type A₂ and B₂ in lateral group, B₂, C and D in tritocerebrum group and A₁ in suboesophageal ganglion, NAYAR (1955), EWEN (1962), DOGRA (1967) and GUPTA (1970), have reported two types i.e. A and B cells in median group in *Imphita limbata*, *Dysdercus koenigii*, *D. similis*. These cells are parallel to the present A₁ and B₁ cells as by their staining characteristics. The presence of lateral group of NSC is seen in the *Dysdercus similis*, GUPTA (1970), *Leptocoris acuta*, FARUQUI (1975a) as has been observed in the case of *Bagrada hilaris*. However these cells are reportedly absent in some insects like *Belostoma indica*, DOGRA (1969), *Halys dentatus* (SRIVASTAVA 1970), *Nezara viridula*, *Metochus uniguttatus* and *Sphaerodema rusticum* (AWASTHI 1972a, 1973a, b).

The neurosecretory pathways i. e. NCC I and NCC II are separate structures all along in this insect, like those seen in case of *Dysdercus similis* (GUPTA 1970), *Nezara viridula* (AWASTHI 1972a). The presence of NCC III (*Ranatra elongata*, DOGRA 1967b), is not noticed in *Bagrada hilaris*. Similarly, fusion of paracardiacum internus (NCC I & II of the present case), lie in form of a common structure as described by CAZAL (1948) and BENWITZ (1956) is not seen in this insect.

The axons of *NCC I* terminate in the aorta after forming a network as seen in other gymnoceretans, *Nezara viridula* (AWASTHI 1972a), *Leptocorisa acuta*, *Cyclopelta siccifolia*, *Rhodnius prolixus* (FARUQUI 1975b, 1977a, b) and *Oncoccephalus* sp. (GHOSH & FARUQUI 1977). In case of cryptoceratans, the axons are reported to run parallel to each other (FARUQUI 1974, 1975b).

The aorta is seen to serve as the neuro-haemal organ for median cells *NSM* in this insect as has been reported in other Heteropterans by SESHAN and ITTYCHERIAH (1966), DOGRA (1967a, b), SRIVASTAVA (1970), AWASTHI (1972a, b, 1973a, b), FARUQUI (1975a, b). The lateral cells *NSM* is transported and stored in *CC* like that reported in *Leptocorisa acuta* and *Cyclopelta siccifolia* (FARUQUI 1975b, 1977a).

Morpho-histologically the endocrine glands in *Bagrada hilaris* are similar to that described for *Cyclopelta siccifolia* (FARUQUI 1977). The chromophilic cells of *CC* shows secretory activity and intrinsic secretion as also stated by SCHARER (1963), NOVAK (1966), SMITH and SMITH (1966), FARUQUI (1974, 1975a, b). The *CA* cells also shows secretory cells and variable amount of secretory granules with in the cells. The *A* cell *NSM* seen in the intercellular spaces of the gland seems to act as raw material for the production of allatotropic hormone (SAINI 1971).

Acknowledgement

The authors wish to acknowledge the valuable suggestions received from Dr. B. D. PATIL, Director of the Institute during the course of study and also for various facilities made available to them.

Zusammenfassung

Bagrada hilaris BURM., ein verbreiteter Schädling auf Kreuzblütlern, wurde mit histologischen Methoden und mit in situ Färbung eingehend auf Verteilung und Typen von neurosekretorischen Zellen (*NSC*), auf neurosekretorische Bahnen (*NSP*), auf Speicherorgane und auf endokrine Drüsen untersucht. Der Pars intercerebralis des Gehirns besteht aus zwei Gruppen von *NSC* in jedem Lappen, die mediale und die laterale Gruppe bestehen aus verschiedenen Typen von *NSC*. Das Tritocerebrum des Gehirns enthält auch einige neurosekretorische Zellen. Das Sekret (*NSM*) der medialen Gruppe wird von *NCC I* zur Aorta transportiert, die als neurohämales Organ für dieses Sekret dient, während das *NSM* von den lateralen Zellen von *NCC II* zur Aorta gebracht wird, die als neurohämales Organ für dieses Material wirkt. Zwei den medialen Zellen ähnliche *NSC* wurden im subösophagealen Ganglion festgestellt. Das Sekret dieser Zellen wird zur Aorta transportiert. Die histologischen Einzelheiten der endokrinen Drüsen, d. h. *CC* und *CA*, werden ebenfalls erörtert. Ihre Verbindung miteinander und mit den *NSC* wird beschrieben.

Summary

Bagrada hilaris Burm., a common pest of cruciferous plants has been studied for the distribution and types of neurosecretory cells (*NSC*), the neurosecretory pathways (*NSP*), storage organs and the endocrine glands in detail using histological and in situ staining techniques. Pars intercerebralis of the brain consists of two groups of *NSC* on each lobe-median and lateral group consists of different types of *NSC*. The tritocerebrum of brain also contains some neurosecretory cells. The secretion (*NSM*) of the median group is carried to the aorta by *NCC I* which serves as the neurohaemal organ for this secretion whereas the *NSM* from the lateral cells is carried to the corpora cardiaca by *NCC II*, which functions as the neurohaemal organ for this material. Two *NSC* similar to the median cells are observed in sub-oesophageal ganglion. The secretion of these cells is transported to the aorta. The histological details of endocrine glands i. e. *CC* and *CA* is also discussed. Their association with each other and *NSC* is described.

Резюме

С помощью гистологических методов и окрашивания in situ подробно изучен *Bagrada hilaris* BURM., широко распространенный вредитель на крестоцветных, от-

носителем его распределения и типов нейросекреторных клеток (*NSC*), нейросекреторных полос (*NSP*), запасящих органов и эндокринных желез. *Pars intercerebralis* мозга состоит из двух групп *NSC* в каждой доли; медиальная и латеральная группы состоят из разных типов *NSC*. В *Tritocerebrum* мозга находится также ряд нейросекреторных клеток. Секрет (*NSM*) медиальной группы перемещается с помощью *NCC* и к аорте, которая служит нейрогемальным органом для этого секрета, а *NSM* латеральных клеток перемещается с помощью *NCC* II к аорте, которая служит нейрогемальным органом для этого материала. Установлены две сходные медиальным клеткам *NSC* в субэзофагеальном ганглие. Секрет этих клеток перемещается к аорте. Обсуждаются также гистологические детали эндокринных желез, т.е. *CC* и *CA*. Описывается их связь друг с другом и с *NSC*.

References

- AWASTHI, V. B.: Studies on the neurosecretory system and retrocerebral endocrine glands of *Nezara viridula* LINN. (Heteroptera: Pentatomidae). J. Morphol. **136** (1972a). — S. 337—352.
- Neurosecretory systems of the adult flesh fly, *Sarcophaga ruficornis* F. (Diptera: Sarcophagidae). Int. J. Insect Morphol. Embryol. **1** (1972b). — S. 133—140.
- Neurosecretory system and the retrocerebral endocrine glands of *Metochus uniguttatus* THUMB. (Heteroptera: Lygaeidae). Int. J. Insect Morphol. Embryol. **2** (1973a). — S. 1—12.
- Studies in situ on the neurosecretory system of *Sphaerodema rusticum* FABR. (Heteroptera: Belostomatidae). Zool. Beitr. **19** (1973b). — S. 151—156.
- BENWITZ, G.: Der Kopf von *Corixa punctata* III GEOFFROYI: LEECH (Hemiptera-Heteroptera). Zool. Jb. Anat. **75** (1956). — S. 311—378.
- CAZAL, P.: Les glandes endocrines retro-cerebral des insects (Etude Morphologique). Bull. Biol., Suppl. **32** (1948). — S. 1—227.
- DOGRA, G. S.: Studies on the neurosecretory system and the functional significance of *NSM* in the aorta wall of the bug *Dysdercus koenigii*. J. Insect Physiol. **13** (1967a). — S. 1895—1906.
- Studies on the neurosecretory system of *Ranatra elongata* FABR. (Hemiptera: Nepidae) with reference to the distal fate of *NCC* I and II. J. Morphol. **121** (1967b). — S. 223—240.
- Studies in situ on the neuroendocrine system of the giant water bug, *Belostoma indica*. Serb. Acta Anat. **72** (1969). — S. 429—445.
- DOGRA, G. S. & TANDON, B. K.: Adaptation of certain histological techniques for in situ demonstration of the neuroendocrine system of insects and other animals. Quart. J. Microsc. Sci. **105** (1964). — S. 455—466.
- EWEN, A. B.: An improved aldehyde fuchsin staining technique for neurosecretory products in insects. Trans. Am. microsc. Soc. **81** (1962a). — S. 94—96.
- FARUQUI, S. A.: Studies on the cystine and/or cysteine rich components of the neurosecretory system of an insect, *Sphaerodema rusticum* FABR. (Heteroptera: Belostomatidae). Zool. Jb. Anat. **92** (1974a). — S. 181—187.
- Studies on the neurosecretory system and the retrocerebral endocrine glands of *Ranatra filiformis* FABR. (Hemiptera), using the technique specific for cysteine and/or cystine. Anat. Anz. **137** (1975a). — S. 154—461.
- Neurosecretory system and retrocerebral endocrine glands of *Leptocoris acuta* THUMB. (Heteroptera: Coreidae). Folia Morphol. **22** (1975b). — S. 261—267.
- Neurosecretory cells and the retrocerebral endocrine glands of active and hibernating adults of *Cyclopelta siccifolia* (Hemiptera: Pentatomidae). Acta ent. bohemoslov. **74** (1977a). — S. 375—380.
- Neurohaemal organ in *Rhodnius prolixus* STAL (Heteroptera: Reduviidae). Int. J. Insect Morphol. & Embryol. **6** (1977b). — S. 345—347.
- GHOSH, K. & FARUQUI, S. A.: Retrocerebral neuroendocrine complex of Reduviid, *Oncocephalus* spp. with special reference to neurohaemal organ. Zool. Jb. Anat. **97** (1977). — S. 525—531.
- GOMORI, G.: Observation with differential stains on human islets of Langerhans. Am. J. Path. **17** (1941). — S. 395—406.
- GUPTA, D. P.: Neurosecretory cells in *Dysdercus similis* (Hemiptera). J. Zool. London **162** (1970). — S. 401—411.
- GUPTA, D. P. & FARUQUI, S. A.: Studies on neurosecretory system of Heteroptera-I neurosecretory cell types. 17th. Int. Cong. Entomol. Hamburg, 1984.

- JUNQUA, C. Etude morphologique et histophysiologique des organes endocrines de l'*Hydrocyrius colombiae*. SPIN. (Hemiptera: Belostomatidae). Bull. Biol. 90 (1956). — S. 154—162.
- NAYAR, K. K. Studies in the neurosecretory system of *Iphita limbata* STAL. I. Distribution and structure of the neurosecretory cells of the nerve ring. Biol. Bull. 108 (1955). — S. 296—307.
- Studies on the neurosecretory system of *Iphita limbata* STAL III. The endocrine glands and the neurosecretory pathways in the adult. Z. Zellforsch. 44 (1956). — S. 697—705.
- NORMAN, T. C. The neurosecretory system of the adult *Calliphora erythrocephala*-I. The fine structure of the corpus cardiacum with some observations on adjacent organs. Z. Zellforsch. 67 (1965). — S. 461—501.
- NOVAK, V. J. A. Insect Hormones. Methuen: London, 1966.
- SAINI, R. S. Neuroendocrine control of oocyte development in the beetle *Aulacophora foveicollis* LUC. J. Insect. Physiol. 12 (1966). — S. 1003—1008.
- Neuroendocrine control of oocyte development in *Poecilocera picta* (Insecta: Orthoptera). J. Zool. London. 165 (1971). — S. 275—283.
- SCHARRE, B. Neurosecretion XIII. The ultrastructure of the corpus cardiacum of the insect *Leucophaea maderae*. Z. Zellforsch. 60 (1963). — S. 761—796.
- SESHAN, K. & ITTYCHERIAH, P. I. *Iphita limbata* STAL. Components of the neurosecretory material. Science. 153 (1966). — S. 427—428.
- SLOPER, J. C. Presence of a substance rich in protein bound cystine or cysteine in the neurosecretory system of an insect. Nature, London. 175 (1957). — S. 148—149.
- SMITH, U. & SMITH, D. S. Observations on the neurosecretory processes in the corpus cardiacum of the stick insect *Caransius morosus*. J. Cell Sci. 1 (1966). — S. 59—60.
- SRIVASTAVA, R. C. Morphology of the neurosecretory system and retrocerebral endocrine glands of adult *Halys dentatus* (Heteroptera: Pentatomidae). Ann. Ent. Soc. Am. 63 (1970). — S. 1372—1376.
- THOMSEN, M. The neurosecretory system of the adult *Calliphora erythrocephala* IV Histological study of the corpus cardiacum and its connection with the nervous system. Z. Zellforsch. 94 (1969). — S. 205—219.
- WIGGLESWORTH, V. B. Life of Insects. WIDENFIELD and NICOLSON: London, 1964.