

Zusammenfassung. Mittels des Meideverhaltens wurde die Hörkurve einsömmriger Karpfen bestimmt. Als obere Hörgrenze wurden 3000 Hz (26 dB) und als «untere» 75 Hz erhalten. Zwischen 900 Hz (–25 dB) und 400 Hz

(–22 dB) lag das Perzeptionsoptimum. Bei 250 Hz war eine auffällige Empfindlichkeitsverringerng zu beobachten.

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Metamorphic Changes in the Intracerebral Neurosecretory Pathways in the Lemon-Butterfly, *Papilio demoleus* L. (Lepidoptera)

Several authors have described the neurosecretory cells (NSC) and their pathways in Lepidoptera^{1–8}. But they have not reported any difference in their arrangement in the pupal and adult brains. During our investigations on the postembryonic changes in the neurosecretory system of *Papilio demoleus*, we found a considerable topographic variation in the NSC and their intracerebral pathways in the pupal and adult brains. This has been made possible by the use of the recently developed in situ staining technique of DOGRA and TANDAN⁹. This technique makes it possible to observe the complete neurosecretory pathway even within a single preparation. Three groups of NSC stain in the pupal brain of this insect: a median group, a lateral group and an anterior group. The median group of NSC is located in the median region of the pars intercerebralis close to the intercerebral midline, the lateral group, on the mid-dorsal region of the protocerebrum and the anterior group, on the anterior margin of the pars intercerebralis (Figure 1). In other lepidopterous insects this last mentioned group is reported on the posterior margin of the brain and has been considered characteristic for the order Lepidoptera^{10–13}. The median NSC give rise to 2 sets of axons: those that run outwards constituting the lateral neurosecretory pathway (LNSP) and those that run inwards and forwards constituting the median neurosecretory pathway (MNSP) (Figure 1). The distal portion of the LNSP is not stainable, possibly because it takes a deeper course inside the brain and is not available to the stain in bulk-preparations. On the other hand, the MNSP, being superficial, is fully stainable and could be traced from its origin to emergence from the brain. In addition

to the axons of the median NSC, the MNSP is also joined by the axons of the lateral NSC (Figure 1, extreme left, broken arrow), a feature not common in other insects and also whose implication is not immediately clear. A thick band of axons, thus formed, runs inwards and forwards on the pars intercerebralis and decussates with its counterpart from the other side to form the chiasma. Thereafter, the fibres cross into opposite hemispheres and run in close proximity upto the anterior NSC. At the level of the anterior NSC, they make a bend (Figure 2, solid arrows) and possibly, accompanied by the axons of these cells, run backwards traversing the entire length of the protocerebrum before emerging from the brain as the nervi corporis cardiaci I (NCC I) (Figure 2, broken arrow). The portion of the MNSP between the median and anterior NSC is reckoned as the ascending limb (Figure 1) and

¹ B. HANSTRÖM, K. svenska Vetensk.-Acad. Handl. 78, 1 (1940).

² B. HANSTRÖM, Biologia gen. 15, 485 (1942).

³ C. M. WILLIAMS, Biol. Bull. 90, 234 (1946).

⁴ L. ARVY, J. J. BOUNHIOL and M. GABE, C.r. Acad. Sci, Paris 236, 627 (1953).

⁵ J. J. BOUNHIOL, M. GABE and L. ARVY, Bull. biol. 87, 323 (1953).

⁶ M. KOBAYASHI, Bull. seric. Exp. Stn Japan 75, 181 (1957).

⁷ J. NISHITSUTSUJI-UWO, Nature, Lond. 188, 953 (1960).

⁸ J. NISHITSUTSUJI-UWO, Z. Zellforsch. 54, 613 (1961).

⁹ G. S. DOGRA and B. K. TANDAN, Q. Jl microsc. Sci. 105, 455 (1964).

¹⁰ J. MITSUHASHI and M. FUKAYA, Jap. J. appl. Ent. Zool. 4, 127 (1960).

¹¹ D. G. R. McLEOD and S. D. BECK, Ann. ent. Soc. Am. 56, 723 (1963).

¹² J. MITSUHASHI, Bull. natn. Inst. agric. Sci. Jap. 16, 67 (1963).

¹³ W. S. HERMAN and L. I. GILBERT, Nature, Lond. 205, 926 (1965).

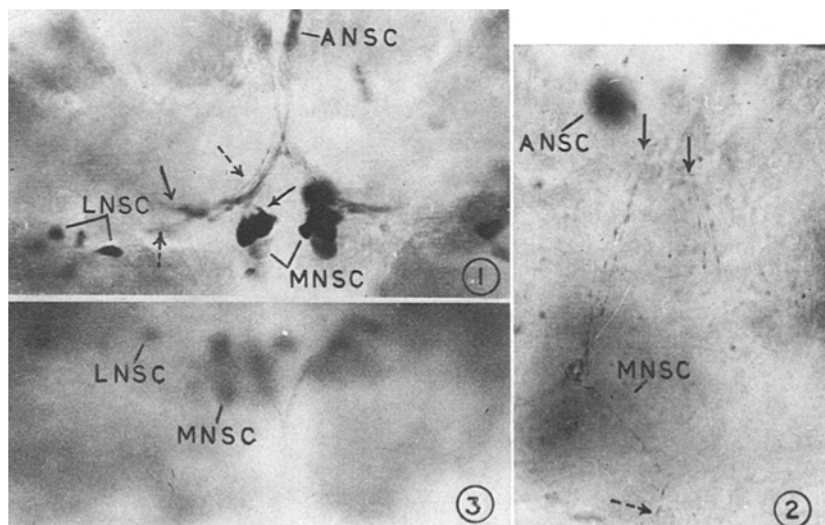


Fig. 1. Bulk-stained preparation of the pupal brain showing 3 groups of NSC and their axons. Solid arrows indicate outwardly directed axons of the LNSP and the broken arrows, inwardly running fibres of the MNSP (median NSC, contributing to MNSP in the left hemisphere, are unstained), ANSC, LNSC, MNSC, anterior, lateral and median groups of NSC, respectively. AF, $\times 180$.

Fig. 2. Bulk-stained preparation of the pupal brain showing descending limbs of the MNSP in the 2 hemispheres (ascending limbs being more superficial are out of focus). Solid arrows point to the bend in the pathway and broken arrow to the emergence of NCC I. AF, $\times 400$.

Fig. 3. Bulk-stained preparation of the adult brain showing the formation of the chiasma by the descending limb of the MNSP (whose post-chiasmatic portion unstained). AF, $\times 150$.

that between the anterior NSC and the place of emergence of the NCC I, as the descending limb of the MNSP (Figure 2). In the adult, however, the condition is greatly changed. Firstly, the axons of even the MNSP are not as readily and fully stainable in the adult as they are in the pupa and secondly, the adult MNSP is not distinguishable into its ascending and descending limbs. Instead, its fibres run inwards and backwards (not forwards) to constitute only one (descending) limb which after forming the chiasma emerges directly as the NCC I (Figure 3). Consistent with the demands of a metamorphic stage, the pupal brain synthesizes and transports through its intracerebral (neurosecretory) axons larger amounts of the neurosecretory material (NSM). This is reflected in

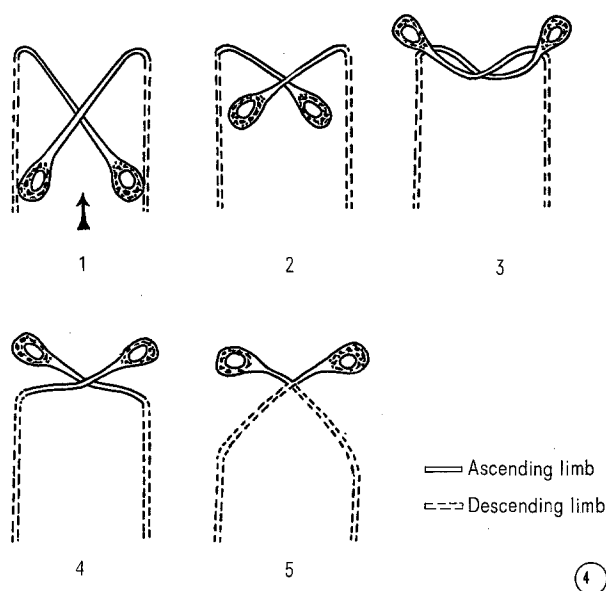


Fig. 4. Hypothetical diagrams depicting the possible steps in the change from the pupal (diagrams 1-3) to adult (diagrams 4-5) condition of the axonal pattern under a supposed anteriorly directed squeeze (arrow).

the maximum intensity that the pupal NSC show in the aldehyde-fuchsin (AF) stain. Since, NSM is the actual stainable component of the axons, there is greater possibility of the latter's staining in this stage than in the adult where the synthesis and transport of the material are relatively less. This, therefore, may account for the first difference. The cause of the second difference may lie in the changes that take place during metamorphosis in the anatomy of the brain itself. It is seen that the pupal brain is dorsally much flatter compared to the adult brain. This increase in the surface-convexity of the adult brain – obviously the result of an increase in the size and complexity of the brain¹⁴ – seemingly brings into play an anteriorly directed squeeze on the dorsum of the brain under which the superficially located NSC move forwards and in so doing dispense with the ascending limb of the MNSP. This hypothesis is supported by the fact that both median and lateral groups of NSC in the adult brain are indeed very much anteriorly displaced (compare Figures 1 and 3). Such a change from the pupal to adult condition could also be reconstructed on an axonal model prepared out of thread pieces. Diagrams of Figure 4, depicting the steps possibly involved in this transformation, are based on such a model¹⁵.

Zusammenfassung. Untersuchungen am Gehirn von Puppen und Imagines des Schmetterlings *Papilio demoleus* zeigen, dass die Axone der neurosekretorischen Zellen der Pars intercerebralis in den Puppen vorerst nach innen vorwärts, dann nach rückwärts laufen, in Imagines aber direkt nach innen rückwärts ziehen. Die notwendigen ontogenetischen Lageänderungen der NSZ und ihrer Axone werden an einem Modell demonstriert.

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¹⁴ R. H. NORDLANDER and J. S. EDWARDS, *J. Morph.* 126, 67 (1968).

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Protection of a Hermit Crab by its Symbiotic Sea Anemone *Calliactis tricolor*

For over a century, the symbiosis between hermit crabs and sea anemones has been studied by various workers, the results of which have recently been reviewed¹. In these partnerships, one or more anemones, usually of the same species, attach by their pedal disc to a gastropod shell inhabited by a hermit crab. The sea anemone has been suggested to protect the hermit crab from predators²⁻⁴, but virtually no experimental work has been conducted to test this hypothesis, with the exception of Ross' recent study⁴.

The purpose of the present study is to present experimental evidence that the presence of a sea anemone on its shell does appear to protect a hermit crab from one of its natural predators.

Materials and methods. *Pagurus pollicaris* SAY, which bears the sea anemone *Calliactis tricolor* (LESEUR) on its shell, is found in the shallow inshore waters of the Western Atlantic and the Gulf of Mexico. Occurring with this hermit crab is its remarkable predator, the oxystomatid crab, *Calappa flammea* (HERBST). One cheliped of *Calappa* (usually the right one) is modified for snipping open gastropod shells and in this fashion the crab is able to very

effectively remove and eat hermit crabs, as well as gastropods⁵.

Numerous preliminary observations in our laboratory suggested that *Calappa* was unable to prey successfully on those hermit crabs carrying anemones, while those without anemones appeared to be readily eaten, thus the reason for the present study.

It was necessary to standardize as closely as possible the 'hunger levels' of the various *Calappa* used. Each *Calappa* was maintained in a 20 gallon recirculating water aquarium and was fed to satiation with frozen squid once per day for 5 days (average of 4.94 g of squid per crab per day) and then starved for the 24 h preceeding an experimental

¹ D. M. ROSS, *Oceanogr. Marine Biology Annual Review* (Ed. H. BARNES; George Allen and Unwin Ltd., London 1967), vol. 5, p. 291.

² L. BERNER, *Bull. Soc. Zool. fr.* 78, 221 (1953).

³ B. B. BOYCOTT, *Publ. Staz. zool. Napoli* 25, 67 (1954).

⁴ D. M. ROSS, *Nature, Lond.* 230, 401 (1971).

⁵ J. B. SHOUR, *Science* 160, 887 (1968).