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GIANT NEURONS AND AXON PATHWAYS IN THE BRAIN OF *TRITONIA*

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INTRODUCTION

Much of the recent work on the giant cells in the central nervous system of gastropods is related to the activity and the electrical properties of individual cells (Tauc, 1955, 1957, 1960, 1962; Arvanitaki & Chalazonitis, 1956; Kerkut & Walker, 1961). Although this has greatly advanced our understanding of the physiology and pharmacology of the individual neurons, there is still much to be learned of the nature and organization of the integrative pathways between the ganglia and the topography of the brain in relation to the behaviour of the animal.

Some progress towards this end was made by the earlier workers using techniques of lesion and stimulation of the nerves combined with recording the electrical and mechanical responses, particularly from the foot (Herter, 1931; Postma, 1943). More recently, transmission across molluscan ganglia has been studied by Turner & Nevius (1951), Horridge (1961) and Nisbet (1961). Although this work has yielded some information of a general nature on the pathways through the brain, it is complicated by the large number of units involved and gives no clear picture of the events taking place inside the ganglia. Hughes & Tauc (1962, 1963) have combined stimulation techniques with intracellular recording from ganglion cells to study the branching axons of the giant cells and cells from the pleural ganglion of *Aplysia*. As yet, these investigations have not provided the answer to the question of their function.

The brain of *Tritonia* seems to offer some advantages over *Aplysia* for studies of this nature, with its more generous allocation of giant cells and the compact nature of the ganglia. The present paper describes some of these features and the distribution of axons from some of the giant cells to nerves on both sides of the brain.

MATERIAL AND METHODS

Tritonia hombergii (Cuvier) is the largest British nudibranch, the largest specimens attaining a length of about 14 cm. It is found sublittorally in association with *Alyconium digitatum* on which it feeds. The animals can be kept in the laboratory for several weeks if they are collected without damage and provided with food and running sea water.

The brain of *Tritonia* is 4–5 mm. across, lying on top of the buccal mass, and is easily exposed by a cut made along the dorsal mid-line. The brain and nerves are surrounded by a vascular connective tissue sheath and these are removed entire, care being taken to avoid damage to the cerebro-buccal connective. The brain is then pinned out in a smaller dish and the connective tissue surrounding the nerves and

ganglia is removed with fine needles and scissors under a dissecting microscope. When this has been done the outlines of a number of large neurons can be recognized in the ganglia.

The methods used to establish whether a nerve contained an axon of a particular cell were similar to those used by Hughes & Tauc (1962). The first method was to stimulate the nerve and look for the subsequent antidromic action potential in the soma, using the criteria suggested by Tauc (1957). This method also provides information on orthodromic pathways to the cell. The second method, which was found more suitable, was to use the rising edge of the intracellular spike to trigger the sweep of a dual beam oscilloscope while displaying action potentials recorded externally from the nerves on the second beam. The latter were obtained using a suction electrode connected to the input of a high-gain differential amplifier. The action potential associated with the cell containing the micro-electrode could be recognized by its constant delay of a few milliseconds and its 1:1 relationship with the intracellular spike. Where the pathway involves a synapse the delay is longer and variable and susceptible to fatigue, particularly at higher frequencies.

Intracellular recordings were obtained with glass microcapillary electrodes filled with 3 M-KCl and having a resistance of about 5 MΩ. The electrode formed one arm of a bridge circuit on the grid of a cathode follower (Koketsu, Cerf & Nishi, 1955) which allowed recordings to be made from the cell whilst passing a stimulating current through the electrode. To locate a particular cell a small hole was made in the epineurium covering the brain with a sharpened tungsten needle, and the electrode was inserted into the cell through the hole.

ABBREVIATIONS

A system of abbreviations has been used in the text and figures for reference to individual giant cells and the nerves arising from the ganglia. The cells are identified by the capital of the first letter of the ganglion in which they occur, *Pe* distinguishing pedal from pleural. This is followed by a numeral which refers to a cell in Fig. 2. Reference to nerves is made in the same way except that *N* follows the capital letter indicating the ganglion. The numbering of the nerves refers to Fig. 1. Left and right sides are indicated by *L* and *R* placed before the abbreviation.

MORPHOLOGY OF THE BRAIN

A considerable volume of literature has accumulated on the morphology and evolution of the brain of gastropods. The nervous system of dorids is of particular interest in that it shows an intermediate condition between a primitive opisthobranch such as *Aplysia*, where the ganglia are well separated by long commissures and connectives, and an advanced pulmonate type such as *Helix*, where concentration and fusion of the original ganglia has occurred to form a 'brain' and a pro-cerebrum has been added to the cerebral lobes.

A good general account of the anatomy of *Tritonia* is given in Alder & Hancock (1909). A shortening of the commissures and connectives has occurred between the three main pairs of ganglia that form the brain, with the exception of the pedal

commissure which has lengthened to allow the ganglia to move dorsally and lie alongside the cerebral and pleural ganglia. The cerebral and pleural ganglia of the same side have fused, but retain their identity as rounded lobes, whilst the cerebral commissure forms a ridge joining the left and right cerebral ganglia closely together. Three large nerves arise from the lateral border of the cerebral ganglia (Fig. 1) and innervate the areas above, lateral to and below the mouth, including the oral tentacles. The nerves branch extensively towards the periphery and carry both sensory and motor elements. The fourth nerve arises ventrally from the cerebral ganglia and is the cerebro-buccal connective.

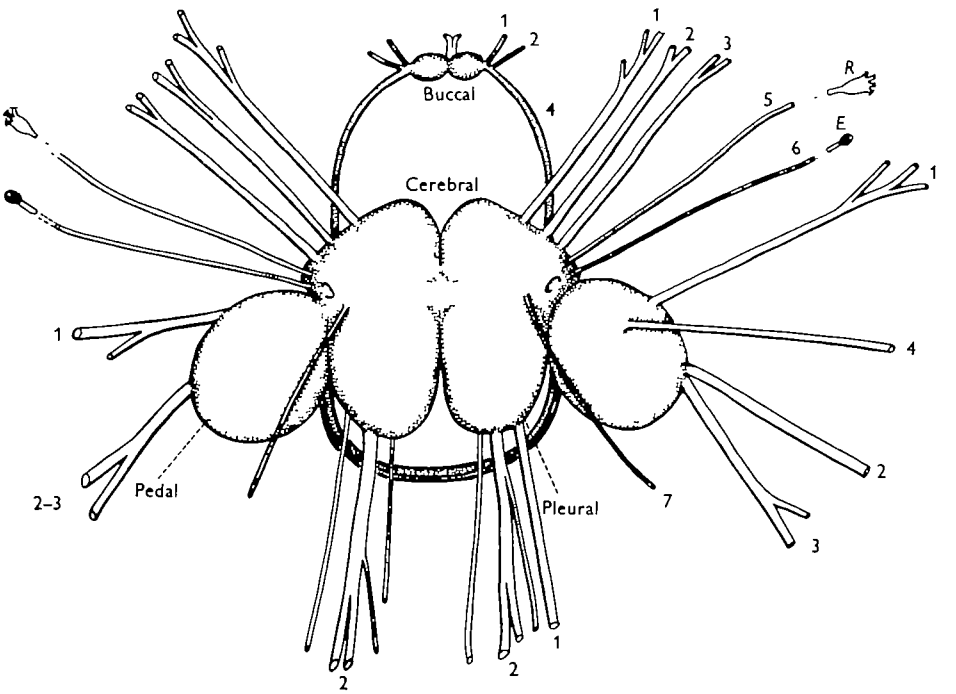

Fig. 1. Dorsal view of the brain and nerves of *Tritonia hombergii*. This figure provides the key to the abbreviations used for referring to nerves in the text. *R*, rhinophore ganglion; *E*, eye.

The paired connectives are normally 3–4 mm. long and run under the connective tissue covering the buccal mass to the buccal ganglia. The remaining nerves from the cerebral ganglia are small but include the connectives with the rhinophore ganglia, the optic nerves and small nerves to the pallium.

In the primitive arrangement, where the ganglia are separate, the pleural ganglia are unique in having connectives but no nerves. Where they do have nerves the suggestion has been made that this is due to their fusion with other ganglia of the visceral loop. Russell (1929) has suggested that in the dorids the parietal, subintestinal and visceral ganglia have fused with the right pleural ganglion and the supra-intestinal and left parietal with the pleural ganglion on the left. This would fit the case in *Tritonia* where the largest nerves from the right pleural ganglion innervate the lateral body wall and the region round the anus, which are typically the areas supplied by the parietal and visceral ganglia in more primitive forms. Similarly, on the left side the

largest nerve supplies the visceral hump which is the region innervated by the supra-intestinal ganglion.

Three large nerves arise from the pedal ganglia to supply the anterior, median and posterior regions of the foot. On the right these nerves are separate, but on the left the second and third nerves have a common origin and separate shortly after leaving the ganglion. The genital nerve arises from the dorsal side of the right pedal ganglion and passes to the base of the penis. The connectives between the pedal and the cerebral and pleural ganglia are very short and can only be seen clearly in horizontal sections of the brain. There is a stout pedal commissure passing under the oesophagus to join the pedal ganglia of each side. The buccal ganglia lie on top of the buccal mass under the oesophagus. Two nerves arise from the point where the cerebro-buccal connectives join the ganglia, and enter the lateral musculature of the buccal mass. A paired radular nerve originates from the short commissure and there is also a small gastro-intestinal nerve passing along the oesophagus.

ANATOMY AND HISTOLOGY OF THE GANGLIA

In the freshly dissected brain the outlines of a number of large cells can be recognized within the ganglia (Dorsett, 1965). The cells appear to be constant in position and number, enabling specific cells to be repeatedly located in different preparations. The giant cells of *Tritonia diomedia* Bergh have been studied by Sakharov, Borovyagin & Zs.-Nagy (1965) and are similar histologically to those described for other opistho-branchs (Bullock, 1961). Vertical and horizontal sections were made of a number of brains and were used to reconstruct cellular maps to show the number and position of the giant cells (Fig. 2). The cerebral ganglia each have a single giant cell lying ventrally in the rounded anterior lobe, similar to that described for Helix by Kunze (1921). This cell, which frequently attains a diameter of 400 μ in large animals, is surrounded dorsally and laterally by intermediate-sized ganglion cells with diameters of 40–50 μ. The lateral and posterior parts of the neuropile are mostly occupied by neuropile containing the axon tracts of the cerebral nerves, the commissure and connectives with the pedal and pleural ganglia (Fig. 3). Many of the smaller cells can be seen sending axons into the commissure but cannot be followed any distance. The axons of the cerebral giant cells can, however, be traced to the ipsilateral buccal connectives, although the contralateral branch of the axon which is indicated in the physiological results, can only by inferred from the traces of two large axons seen at different levels in the commissure. The posterior region of the cerebral ganglia has only a thin rind of small ganglion cells covering the central neuropile.

As the right pleural ganglion is thought to contain the visceral ganglion (Russell, 1929), one might expect asymmetry in the number and arrangement of giant cells on the right and left sides. This does not seem to be so, there being nine giant cells on each side concentrated at the posterior end of the ganglion, which is occupied entirely by their somata. Although the arrangement of cells on each side is frequently not identical (Fig. 2), it is sufficiently so for identification of corresponding cells on a stereotaxic basis, which is important for subsequent physiological comparisons. The pleural ganglia contain the largest cells in the brain (Fig. 2, *RP* 1, *LP* 1) which may have a diameter of 500 μ and lie ventrally near the origin of the second pleural nerve.

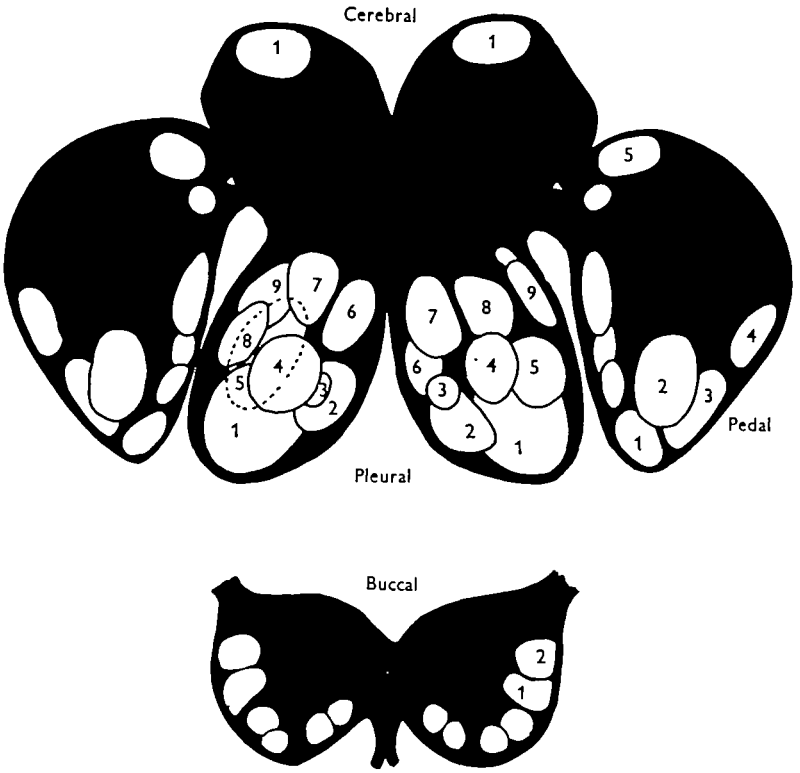

Fig 2 Reconstruction of the brain to show the position of the giant cells within the ganglia. Numerals provide the key to cells mentioned in the text.

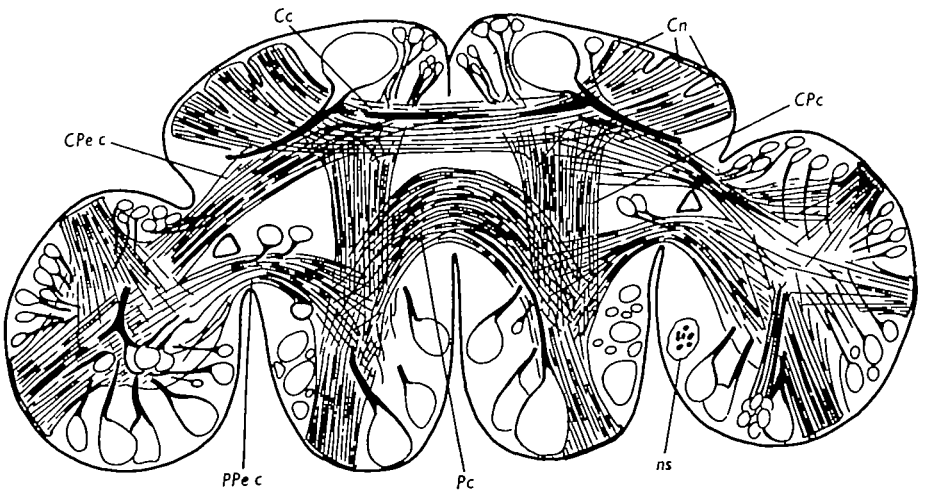

Fig. 3. Diagram of fibre tracts and axon pathways seen in horizontal and vertical sections. *Cc*, cerebral commissure; *Cn*, cerebral nerves; *CPc*, cerebro-pleural connective; *CPe.c*, cerebro-pedal connective; *ns*, neurosecretory cell; *Pc*, pleural commissure; *PPe.c*, pleuro-pedal connective.

In these opisthobranchs where the ganglia are widely separated the pleural ganglia may only communicate through the cerebral and pedal commissures. A new feature which is consequent upon the fusion and centralization of the cerebral and pleural ganglia is the establishment of a separate pleural tract or commissure (Fig. 3, *Pc*) providing a direct communication between the pleural ganglia within the brain. One of the branches of the axon of the giant cell *RP* 1 has been followed in sections using this tract to cross to the left side. The cerebro-pleural and pleuro-pedal connectives still remain as identifiable tracts of orientated fibres projecting into the anterior region of the pleural lobe. These junctional areas of the neuropile are always surrounded by small ganglion cells with diameters of 15–30 μ.

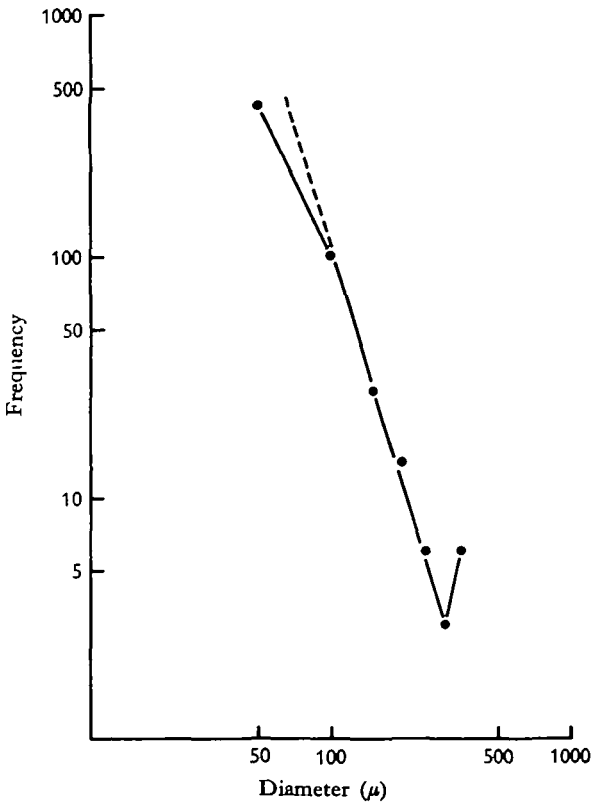

Fig. 4. Logarithmic size-frequency plot of cells in the brain showing the discontinuous nature of the giant cells.

The pedal ganglia contain about eight large cells, mostly at the posterior end, with individual cells recognizable by their position. The axons of most of the cells pass into the central neuropile where it is difficult to follow them further. The buccal ganglia have two giant cells and a row of smaller cells along the posterior border (Fig. 2). Both the pedal and the buccal ganglia are being studied more fully and will form the subject of another account.

The central ganglia of molluscs are notable for possessing cells which have an unusually wide range of cell diameters. It has never been satisfactorily determined

whether the giant cells represent one extreme of a continuous spectrum of cell size, or are discontinuous in that they occur in a separate part of the size-frequency range. Fig. 4 is a logarithmic size-frequency plot of the diameters of approximately 600 cells taken from fifteen different vertical planes through the brain of *Tritonia*. The frequency decreases exponentially with size up to 300 μ when it suddenly increases. This increase represents the giant cells which can therefore be said to represent a distinct component of the central ganglia of molluscs.

THE GIANT CELLS *RC* 1, *LC* 1

The cerebral ganglia are closely associated with the sensory structures of the head and it is generally assumed from their innervation that they play some part in relaying and processing incoming sensory information to appropriate motor centres in the buccal or pedal ganglia. As yet, no physiological information exists on the nature of this integrative function, which makes the study of the cerebral giant cells particularly interesting. When the cerebral giant cells in an isolated brain are penetrated by a micro-electrode they have without exception been shown to exhibit spontaneous activity (Fig. 6A). The cells fire regularly with a frequency that varies between 1·1 and 1·7 impulses/sec., the membrane potential following the spikes showing a slow depolarization typical of a pacemaker potential. In undisturbed preparations this activity is maintained for several hours. Simultaneous independent recordings from the left and right giant cells suggest that there are no direct physiological connexions between the two. Hyperpolarization of the soma of one cell lowers the spike frequency of that cell without affecting the frequency of the other.

Electrophysiological recordings confirm histological evidence that the axon of the giant cell passes into the ipsilateral cerebro-buccal connective. Apart from the connectives no other nerves arising from the cerebral, pleural or pedal ganglia show any action potentials associated with the intracellular spike of the cerebral giant cells. Occasionally, the spike in *LC* 1 was followed by an action potential in *LPeN* 2–3, but the long and variable delay of 120 msec. suggests that the pathway is at least mono-synaptic. The intracellular spike in each giant cell is followed by an action potential in the ipsilateral buccal connective after a delay of 7 msec., the recording electrode being placed 2–3 mm. from the cerebral ganglia. When the external electrode is transferred to the opposite connective, one also finds an action potential following the intracellular spike but the transmission time is increased to 15 msec. The constant delay and 1:1 relationship of the two action potentials indicate that the giant cell sends a branch of its axon into the contralateral connective and that synaptic transmission is not involved in crossing the brain. The mean conduction velocity of the axon for a number of experiments varied between 0·3 and 0·5 msec., which is relatively rapid for gastropod axons. The further course of the axons of the giant cells was followed in the nerves arising from the buccal ganglia. Action potentials from *LC* 1 were found in both *LBN* 1 and *LBN* 2 (Fig. 5), which innervate the left side of the buccal mass. The action potential in *LBN* 1 has a transmission delay of 7 msec., while that of *LBN* 2 has a longer delay of 22 msec. The short delay in *LBN* 1 must mean that the axon of *LC* 1 passes directly into the nerve from the connective. The longer delay in the second nerve has two possible explanations. First, a branch of the axon may

enter the ganglion and synapse with a smaller secondary neuron whose post-synaptic fibre emerges in the second buccal nerve. Alternatively, the branch of *LC* 1 in the right connective may cross the buccal ganglia and leave directly in *LBN* 2. Of these possibilities the second seems more likely. It has been established that the time taken for the action potential of *LC* 1 to reach the right connective is 15 msec., of which 7 msec. is taken to cross the cerebral commissure. If a similar transmission delay is allowed for crossing the buccal ganglia the total delay would be 22 msec. This figure is in agreement with the experimental time.

The right giant cell is symmetrical to the left in the distribution of its axons (Fig. 8). So far no activity associated with the giant cells has been found in the radular nerve or the gastro-oesophageal nerves.

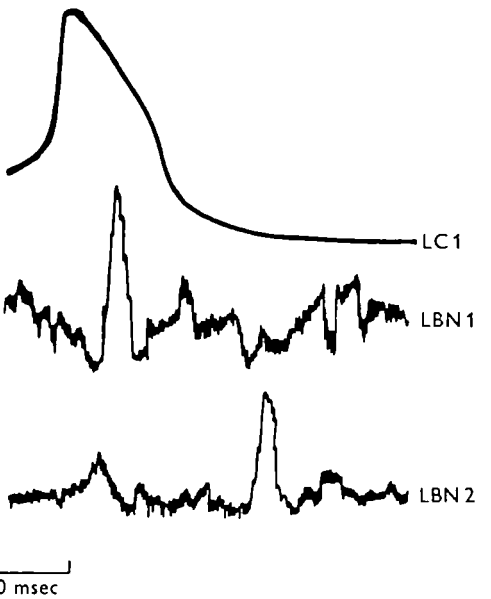

Fig. 5. The intracellular spike in the left cerebral giant cell and the subsequent action potentials in the ipsilateral buccal nerves

ORTHODROMIC PATHWAYS TO THE CEREBRAL GIANT CELLS

Difficulties in restricting the muscular movements of the buccal mass and foot of whole animals have so far prevented the effective use of micro-electrodes in the investigation of the natural sensory input to the cerebral giant cells. To overcome this, stimulation of the cerebral nerves was used to indicate those which contained axons making orthodromic connexions with the giant cells.

Stimulation of the first nerve was found to have two effects. The first was obtained when *RCN* 1 was stimulated with single shocks, while *RC* 1 was hyperpolarized through the micro-electrode to stop its spontaneous activity. The cell responded to each stimulus pulse with an e.p.s.p. that resulted in an action potential (Fig. 6C). The spike latency varied between 40 and 100 msec.

The second effect was seen when stimulation was delivered to the first nerve in the form of a short burst of pulses whilst recording from micro-electrodes inserted into

both giant cells (Fig. 7A). Prior to stimulation the cells showed their normal spontaneous rhythm but as the stimulus was applied small i.p.s.p.'s were seen and the spontaneous activity was inhibited. After stimulation ceased the normal rhythm was resumed without a post-inhibitory rebound.

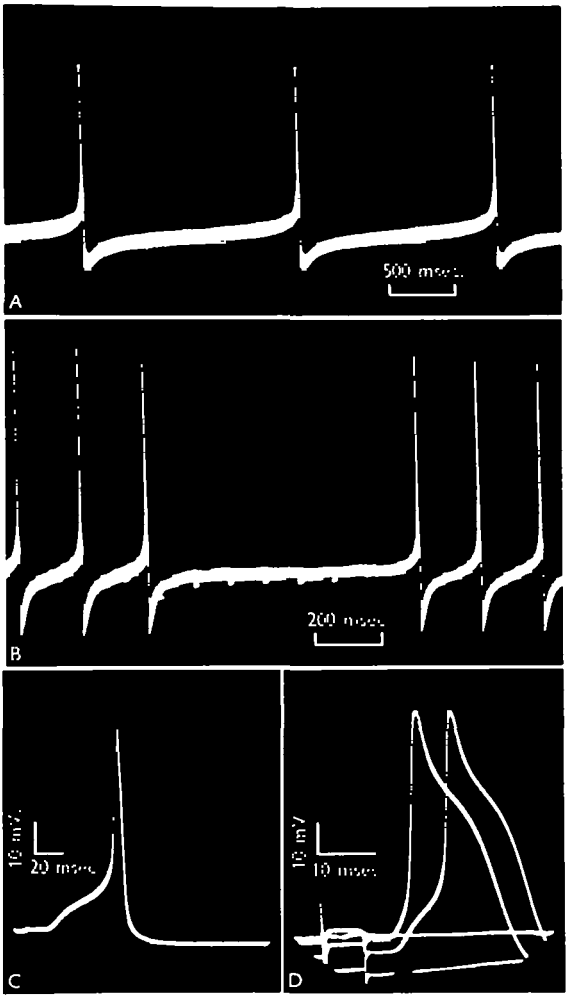

Fig. 6. (A) Spontaneous activity in a cerebral giant cell. Note the absence of e.p.s.p.'s or i p.s.p.'s. (B) Inhibition of the spontaneous activity in *RC* 1 by stimulation of *RCN* 3. (C) Orthodromic excitation of *RC* 1 by stimulation of *RCN* 1. The cell was hyperpolarized to stop it firing. (D) Antidromic and orthodromic spikes in *LP* 1 after stimulating *LPN* 2. Slight hyperpolarization blocks the antidromic spike and reveals the e.p.s.p.

Of the other cerebral nerves only the third has proved effective in modulating the activity of the cerebral giants. Stimulation of this nerve produced inhibition of the ipsilateral giant cell (Fig. 6B). It has not yet been determined whether the inhibitory fibres in this nerve have inputs to the giant cell of the opposite side.

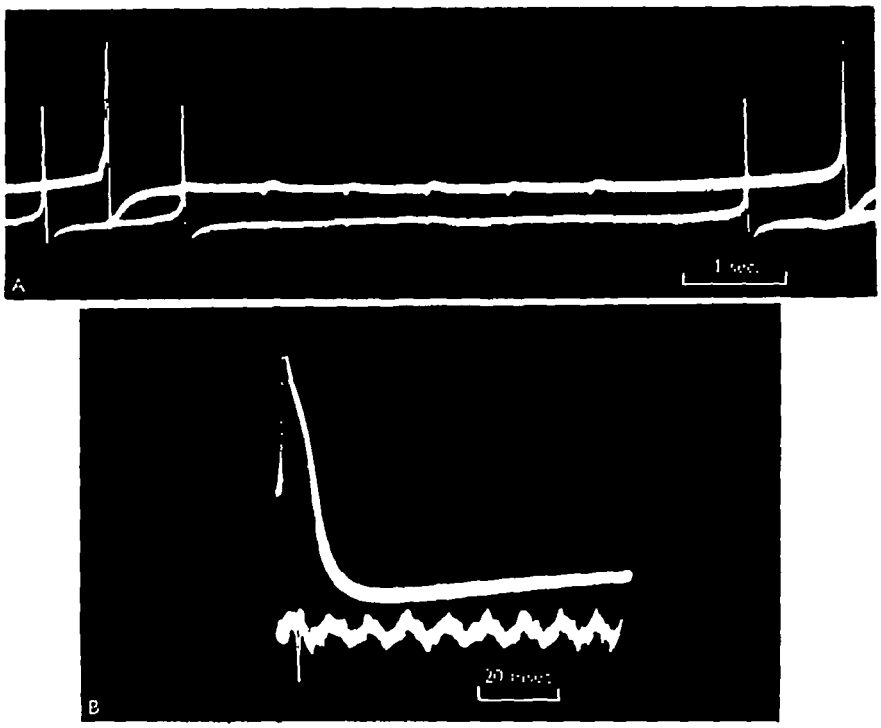

Fig. 7. (A) Simultaneous recordings from the cerebral giant cells showing bilateral inhibition resulting from stimulation of *RCN* 1. *RC* 1 is recorded on the upper trace. (B) The intracellular spike of *LP* 2 and the subsequent action potential recorded from the pedal commissure.

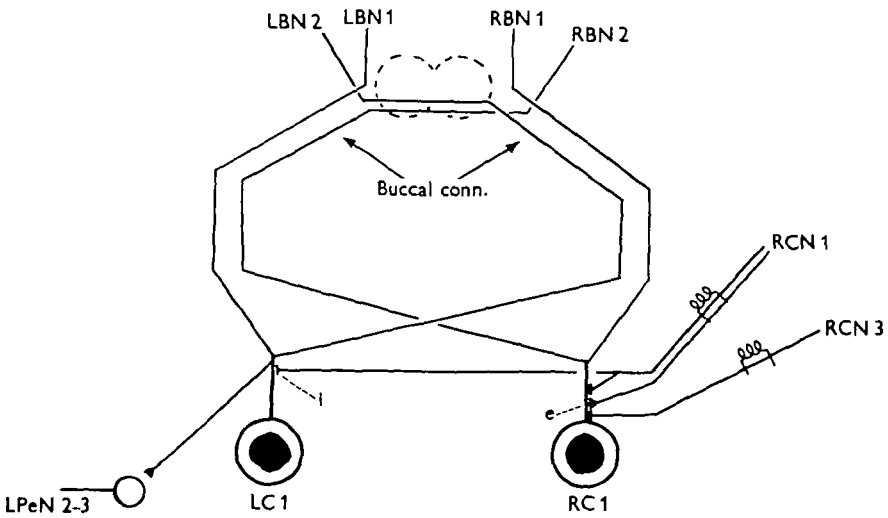

Fig. 8. A diagram of the distribution of the axons from the cerebral giant cells in the buccal connectives and nerves. Orthodromic pathways to the cells in the cerebral nerves are also shown, i, e, excitatory and inhibitory synapses.

THE PLEURAL GIANT CELLS, *RP* 1, *LP* 1

There are eighteen giant cells in the two pleural ganglia and twenty large nerves leaving the brain, so that tracing the direct and synaptic pathways of the axons of the pleural giant cells becomes a considerable task. The first efforts have been directed at the two largest cells *RP* 1 and *LP* 1, which are found near the origin of the second pleural nerves. In contrast to the cerebral giants, *RP* 1 and *LP* 1 do not show spontaneous activity. In order to trace the ramifications of the axon the cell membrane was slightly depolarized through the micro-electrode which caused a slow continuous production of action potentials. Action potentials associated with the intracellular

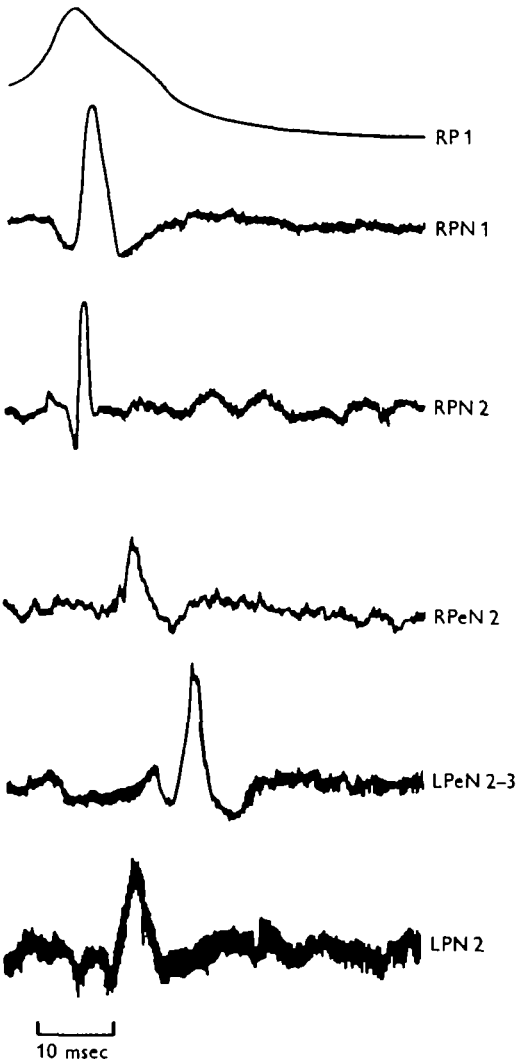

Fig. 9. The intracellular spike of *RP* 1 and the subsequent action potentials in nerves from both sides of the brain. The lower traces are aligned by super-imposing the intracellular spikes obtained on the upper trace of each recording. The noise levels give a comparative indication of the amplification in each case.

spike of *RP* 1 were found in several of the nerve trunks leaving the brain (Fig. 9). Branches pass directly into the two large nerves of the right pleural ganglion, the action potentials in the nerves having a short delay of 2 msec. This is to be expected as the soma lies close to the origin of the nerves. Another branch leaves the right pedal ganglion in the second nerve, the delay increasing to 8 msec. The axon also crosses to the left side of the brain (Fig. 11) where it further subdivides, entering *LPN* 2 and *LPe* 2–3. The transmission delay is 8 msec. in the pleural and 16 msec. in the pedal nerve, but these figures include the time taken travelling along a few mms of nerve as well as crossing the neuropile of two or three ganglia, and are too short for synaptic transmission to be involved.

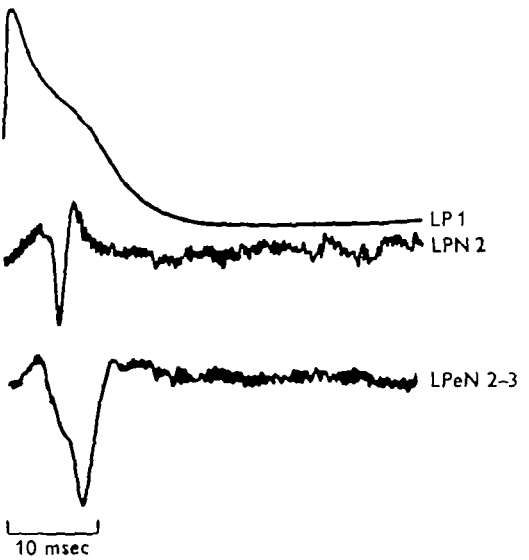

Fig. 10. Recordings from the left pleural giant cell *LP* 1, and the action potentials in the pleural and pedal nerves.

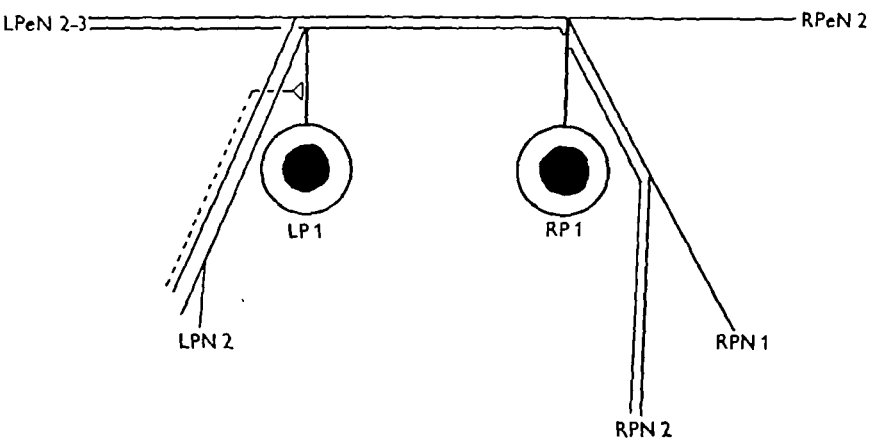

Fig. 11. Diagram of the axon distribution of the pleural giant cells *RP* 1. *LP* 1, An orthodromic excitatory pathway to *LP* 1 is indicated by the dotted line.

The axon from *RP* 1 crosses to the left pleural ganglion in the pleural commissure or tract. The pedal commissure, which has been suggested as a pathway between the pleural ganglia, is not involved, as no action potential associated with the *RP* 1 spike is found in this nerve.

The left pleural giant cell, *LP* 1, appears to be symmetrical with its partner on the right. It has branches in its axon in *LPN* 2 and *LPe* 2–3 on the left side and *RPN* 2 on the right (Figs. 10, 11). Its presence in the first pleural and second pedal nerves on the right has yet to be confirmed.

The second pedal nerve on the left side contains fibres which synapse with the giant cell *LP* 1. Stimulation of this nerve (Fig. 6D) produces an antidromic spike, as would be expected. If the soma is slightly hyperpolarized the antidromic spike can be blocked, revealing the e.p.s.p. and action potential produced by the orthodromic pathway.

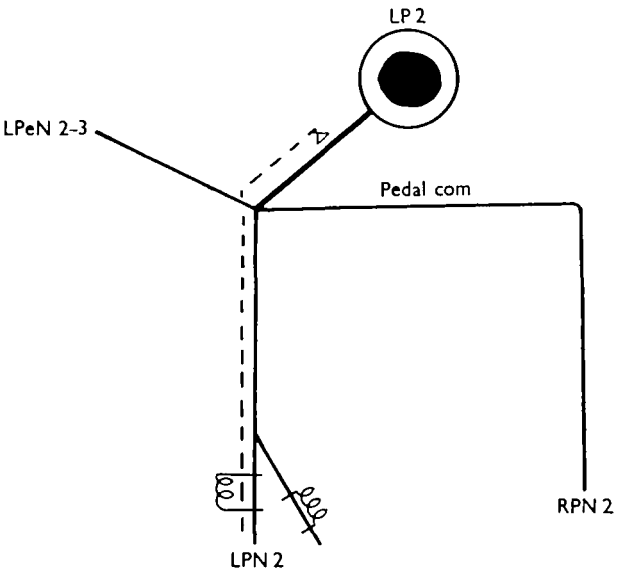

Fig. 12. Diagram of the axon distribution of the pleural giant cell *LP* 2. This cell is notable for having a branch of its axon crossing the brain in the pedal commissure. An orthodromic excitatory pathway is shown by the dotted line.

PLEURAL GIANT CELL *LP* 2

This provides the first experimental demonstration of a cell with an axon which uses the pedal commissure rather than the cerebral commissure in crossing from one pleural ganglion to the other. The axon has two branches which leave in the second pleural nerve on each side (Figs. 7B, 12).

DISCUSSION

Recent work on the isolated ganglia and whole animal preparations of *Aplysia* (Hughes & Tauc, 1962, 1963) has revealed the variety of activity exhibited by cells in the abdominal and pleural ganglia. Many cells show pacemaker activity which is unaffected by stimulation of nerves leaving the ganglia, whereas other cells show

excitation or inhibition of the spontaneous rhythm. Units similar to these have also been found in crustaceans (Wiersma, 1958) and insects (Fielden & Hughes, 1962), where they have been classified as interneurons.

The activity of the cerebral giant cells of *Tritonia* is influenced by stimulation of the sensory nerves in a way that is similar to a cell described from the abdominal ganglion of *Aplysia* (Hughes & Tauc, 1962). This unit was inhibited by mechanical stimulation of the head. A train of stimulating pulses (possibly representing a normal sensory input) applied to the first cerebral nerve of *Tritonia* produced inhibition of the spontaneous rhythm in both cerebral giant cells. The suggestion has been made many times and for many different groups of animals that the cells of the cerebral ganglia exert a moderating influence on the activity of the other ganglia, and it is interesting to speculate whether the inhibition of the spontaneous rhythm by stimulation of the sensory nerves from around the mouth may serve to release some pattern of motor activity concerned with the feeding response.

One clear feature that has arisen from the present study is the multiple branching undergone by many of the giant cells in the pleural ganglia. A picture is emerging of cells with multi-branched axons which are symmetrically distributed among the nerves of the pleural and pedal ganglia, while a similar pattern is seen with the cerebral giant cells and the buccal nerves. In this way they resemble the giant cells of *Aplysia* more closely than the other cells in the pleural ganglia whose branching was studied by Hughes & Tauc (1962).

From the functional point of view the passage of the axons through the ganglia creates additional problems. It seems that the axons of the giant cells do not terminate on cells in motor centres such as the buccal and pedal ganglia, although the possibility of synaptic connexions has not been excluded. If the giant cells are to be regarded as interneurons then the nature of their peripheral terminations must be considered. Two alternatives seem possible, either they terminate directly on muscle fibres where they influence muscle tonus, or they provide a central input to the scattered neurons in structures like the sole plexus in the foot or the visceral epithelia.

I should like to thank Prof. G. M. Hughes for reading the manuscript and his many useful comments, and W. R. G. Rowntree for photographing the recordings.

SUMMARY

1. The anatomy and histology of the brain of *Tritonia hombergii* are described.

2. The brain contains giant neurons which are regularly found in the same positions: cerebral ganglia, one in each; pleural, nine; pedal, eight; buccal, two.

3. The two cerebral giant neurons show spontaneous activity which is inhibited by stimulation of the cerebral nerves. The two largest cells in the pleural ganglia are not spontaneously active but have orthodromic excitatory connexions with axons in the pleural nerves.

4. The branching of the axons of the giant cells has been followed using electrophysiological methods. The axons of a particular giant cell are distributed in a symmetrical fashion to nerves arising on both sides of the brain. This pattern of distribution is similar to that of the corresponding cell in the opposite half of the brain.

REFERENCES

ALDER, J. & HANCOCK, A. (1909). *British Nudibranchiate Mollusca.* London Ray Society.

ARVANITAKI, A. & CHALAZONITIS, N. (1956). Activations du soma géant d'Aplysia par voie orthodrome et par voie antidrome (derivation endocytaire). *Arch. Sci. physiol.* **10**, 95–128.

BULLOCK, T. H. (1961). In *Nervous Inhibition.* E. Florey (ed.). New York: Pergamon Press.

DORSETT, D. A. (1965). The brain of the sea slug. *Rep. Challenger Soc.* **3**, no. XVII, 24.

FIELDEN, A. & HUGHES, G. M. (1962). Unit activity in the abdominal nerve cord of the dragonfly nymph. *J. Exp. Biol.* **39**, 31–44.

HERTER, K. (1931). Der Jordansche 'Halbtierversuch'. *Z. vergl. Physiol.* **15**, 261–308.

HORRIDGE, G. A. (1961). The centrally determined sequence of impulses initiated from a ganglion of the clam Mya. *J. Physiol.* **155**, 320–36.

HUGHES, G. M. & TAUC, L. (1962). Aspects of the organization of the central nervous pathways in *Aplysia depilans*. *J. Exp. Biol.* **39**, 45–69.

HUGHES, G. M. & TAUC, L. (1963). An electrophysiological study of the anatomical relations of the giant nerve cells in *Aplysia depilans*. *J. Exp. Biol.* **40**, 469–86.

KERKUT, G. A. & WALKER, R. J. (1961). The effects of drugs on the neurons of the snail *Helix aspersa*. *Comp. Biochem. Physiol.* **3**, 143–60.

KOKETSU, K., CERF, J. & NISHI, S. (1959). Further observations on the activity of frog spinal ganglion cells in sodium free solutions. *J. Neurophysiol.* **22**, 177–249.

KUNZE, H. (1921). Zur Topographie und Histologie des Centralnerven systems von *Helix pomatia*. L. *Z. wiss Zool.* **118**, 87–9.

NISBET, R. H. (1961). Some aspects of the neurophysiology of *Archachatina marginata* (Swainson). *Proc. Roy. Soc.* B **154**, 309–31.

POSTMA, N. (1943). Über den Tonus des Schneckenfusses (*Helix pomatia L.*). VI. Tonus und Zerebralganglion. *K. Akad. Wet. Amst. Afd. natuur. Versl.* **52**, 228–38.

RUSSELL, L. (1929). The comparative morphology of the elysiod and aeolidioid types of molluscan nervous system and its bearing on the ascoglossan nudibranchs. *Proc. Zool. Soc. Lond.* **99**, 197–223.

SAKHAROV, D. A., BOROVYAGIN, V. L. & ZS.-NAGY, I. (1965). Light fluorescence and electron microscope studies on neurosecretion in *Tritonia diomedia* Bergh (Mollusca, Nudibranchiata). *Z. Zellforsch. mikrosk. Anat.* **68**, 660–673.

TAUC, L. (1955). Étude de l'activité élémentaire des cellules du ganglion abdominale de l'Aplysie. *J. Physiol. Paris* **47**, 769–92.

TAUC, L. (1957). Stimulation du soma neuronique de l'Aplysie par voie antidromique. *J. Physiol. Paris* **49**, 974–86.

TAUC, L. (1960). Diversité des modes d'activité des cellules nerveuses du ganglion déconnecté de l'Aplysie. *C.R. Soc. Biol., Paris* **153**, 17–21.

TAUC, L. (1962). The site of origin and propagation of the spike in the giant neuron of Aplysia. *J. Gen. Physiol.* **45**, 1077–97.

TURNER, R. S. & NEVIUS, D. B. (1951). The organisation of the nervous system of *Ariolimax columbianus*. *J. Comp. Neurol.* **94**, 239–56.

WIERSMA, C. A. G. (1958). On the functional connections of single units in the central nervous system of the crayfish *Procambarus clarkii* (Girard). *J. Comp. Neurol.* **110**, 421–72.