

ANATOMY OF MOTONEURONES INNERVATING MESOTHORACIC INDIRECT FLIGHT MUSCLES IN THE SILKMOTH, *BOMBYX MORI*

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SUMMARY

1. The anatomy and innervation of mesothoracic indirect flight muscles, dorsal longitudinal (DLM) and dorsoventral muscles (DVM) are described. The major DLMs depress the forewing, the major DVMs elevate it.

2. The morphology of the motoneurones and unpaired median neurones (UM-neurone) innervating these muscles was investigated by back-filling with cobalt. The DLMs are innervated by eight paired motoneurones and one UM-neurone: one of these motoneurones has its cell body in the contralateral side of the mesothoracic ganglion, and the others have theirs in the ipsilateral side of the prothoracic ganglion. The DVMs are innervated by nine paired motoneurones and one UM-neurone, with their cell bodies in the ipsilateral and the median portion of the mesothoracic ganglion, respectively.

3. All of the paired DLM and DVM motoneurones have many dendritic processes, mainly in the dorso-ipsilateral neuropile of the ganglia, but the DLM motoneurones also have processes in the contralateral neuropile.

4. Each UM-neurone has its cell body in the postero-medial cortex of the mesothoracic ganglion, extending a single median neurite anteriorly to the dorsal neuropile where it bifurcates to form a right and left axon.

5. The arrangement of the mesothoracic indirect flight motoneurones in different insect orders is discussed in terms of the neural organization of the flight system.

INTRODUCTION

It is well known that the male silkmoth, *Bombyx mori*, performs a mating dance in response to the female sex pheromone. The major behavioural components of the dance are walking, wing vibration, and occasional abdominal curvature. The mating dance plays an essential role in locating the calling female (Obara, 1979). Close observations have revealed the behavioural properties of wing vibration in the mating dance, and extracellular recordings from identified mesothoracic indirect flight muscles have elucidated the electrical activity of the muscles during the dance (Y. Obara & Y. Kondoh, in preparation). The motor pattern of wing vibration during the mating dance is similar to that during flight. Deviation from the normal

flight motor pattern is caused by irregular inhibitory inputs from legs that occasionally contact the ground during the dance. Surgical experiments suggest nevertheless that some neuronal command elements for the mating dance exist in the supraoesophageal ganglia (Y. Obara & Y. Kondoh, in preparation). These command elements are activated by stimulation with a pheromone and remain active for as long as 4–5 min afterwards. The neural mechanisms controlling the mating dance, however, remain to be demonstrated at the cellular level.

The present study was carried out first to elucidate which of the indirect flight muscles is involved in wing vibration and then to reveal the anatomy of their motoneurons. The results will be discussed in terms of the motoneuronal architecture of the insect flight system.

MATERIALS AND METHODS

(1) *Animals*

Males of the TW2-strain of *Bombyx mori* were used. This strain lacks the dark pigments on the surface of ganglia in the ventral nerve cord, present in the wild type, which make it difficult to observe the fine dendritic processes of the motoneurons. The moths were reared in mass culture at 26 °C under a long day (16L:8D) photoperiod regime. They were kept at 26 °C for two days after eclosion and subsequently at 4 °C until use.

(2) *Anatomy*

The anatomy of the mesothoracic musculature and nervous system was investigated by dissecting moths which had been preserved for 24 h in a fixative consisting of 25 ml of 40% formalin, 1.25 ml of acetic acid and 10 g of chloral hydrate in 100 ml distilled water (Chauthani & Callahan, 1966). The nervous tissues were stained with aniline blue (1% in distilled water). The designations of the muscles and nerve trunks were based on those given by Eaton (1974) for *Manduca sexta*.

The number and sizes of axons in the nerve trunks and the cell bodies of the flight motoneurons within the thoracic ganglia were determined by examining serial sections in the light microscope. The tissue was fixed for 12 h in 2% glutaraldehyde in phosphate buffer, washed for 12 h in buffer containing glucose (3.6 g per 100 ml), dehydrated, and then embedded in epoxy resin through *n*-butyl glycidyl ether. Serial sections 1.5–2 μ m thick were cut and stained with toluidine blue.

The location of cell bodies and the arborization patterns of flight motoneurons were determined by the cobalt axonal filling method (Sandeman & Okajima, 1973; Tyrer & Altman, 1974). The pro- and pterothoracic ganglia and their nerves were dissected out in a *Bombyx* saline (H. Tsujimura, personal communication) which was based on Naoumoff & Jeuniaux's (1970) analysis of haemolymph. Axons were exposed to a 400 mM cobalt chloride solution containing 13 mg of bovine serum albumen per 100 ml (Strausfeld & Obermayer, 1976), for 24–72 h at 4 °C. The tissue was rinsed in saline, developed in ammonium sulphide (1 drop in 3 ml saline), fixed, dehydrated, cleared in methyl benzoate, and then mounted in Canada balsam. On some preparations, Timm's silver-intensification (Bacon & Altman, 1977) was

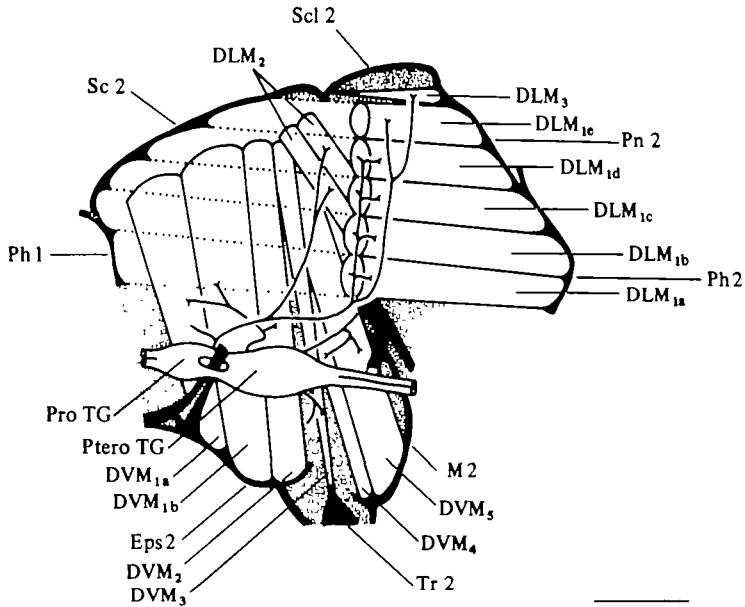


Fig. 1. Lateral view of right half of mesothorax in the moth, *Bombyx mori*, showing indirect flight muscles, ganglia, and nerve trunks to these muscles. Other nerves to sense organs, direct flight muscles, and leg muscles are not shown. Pterothoracic ganglion consists of fused, meso- and metathoracic ganglia and 1st and 2nd abdominal ganglia. Pro TG: prothoracic ganglion, Ptero TG: pterothoracic ganglion, Ph: phragma, Sc: scutum, Scl: scutellum, Pn: postnotum, Eps: episternum, Tr: trochanter, M: meron, DLM: dorsal longitudinal muscle, DVM: dorsoventral muscle. Subscripts 1 and 2 indicate 1st and 2nd thoracic segments, respectively. Scale bar: 1 mm.

performed to enhance details of the dendritic processes before dehydration. The stained neurones were drawn using a camera lucida drawing tube attached to a microscope. A total of 103 successful preparations were examined.

RESULTS

1. The forewing indirect flight muscles

The forewing indirect flight muscles in the mesothorax are shown in Fig. 1 and the sclerites to which they attach are shown in Table 1. These muscles are morphologically divided into two groups. The first, the dorsal longitudinal muscles (DLM), attach to the notum, or the sclerites derived from it, at both ends. The DLMs are further subdivided into three muscles, DLM₁, DLM₂ and DLM₃. Of these, the DLM₁, consisting of five bundles (DLM_{1a-e}), is the largest. All run longitudinally in the dorsal portion of the mesothorax. The DLM₂, consisting of two muscle bundles, runs lateral to DLM₁. Although both clearly belong morphologically to the DLMs, they run obliquely, or nearly dorsoventrally, with their posterior end ventral. The DLM₃ is a small muscle bundle which is dorsal to the DLM₁ and runs longitudinally.

The second group of indirect flight muscles, the dorsoventral muscles (DVM),

Table 1. *Mesothoracic indirect flight muscles and their function in forewing movement*■

Muscle	Attachment sclerites	Function
DLM _{1 a-e}	1st phragma, scutum and pre-scutum to postnotum and 2nd phragma	Wing depressor
DLM ₂ (= dorsal oblique)	Scutum to laterophragma	?
DLM ₃	Scutum to postnotum	?
DVM _{1 a, b} (= tergo-sternal)	Scutum to episternum	Wing elevator
DVM ₂ (= anterior tergo-coxal)	Scutum to basicosta	Wing elevator
DVM ₃ (= tergotrochanteral)	Scutum to trochanter	Wing elevator
DVM ₄ and DVM ₅ (= posterior tergo-coxal)	Scutum to meron	Wing elevator

consists of six muscle bundles, all of which run dorsoventrally. Three of them (DVM_{1 a, b} and DVM₂) are anterior to the coxa, while the other two (DVM₄ and DVM₅) are posterior to it. Another thin DVM (DVM₃) arises at the mesonotum and attaches to the trochanter.

The function of some of these indirect flight muscles was determined by pressing the sclerites to which they attach along their long axis. Shortening the mesonotum along the axis of DLM₁ resulted in depression of the forewing, while pressing down the mesonotum ventrally along the DVM long axis resulted in wing elevation (Table 1). These results indicate that DLM₁ functions as a forewing depressor and DVM as a forewing elevator. The same kind of mechanical movement of the sclerites to which the DLM₂ and DLM₃ attach did not cause any observable movement of the wing, and thus their function in moving wings could not yet be established.

2. Innervation of the indirect flight muscles

The thoracic ganglia from which nerve trunks emerge to innervate the relevant indirect flight muscles are shown diagrammatically in Figs. 1 and 2. There are two thoracic ganglia, a pro- and a pterothoracic ganglion. The latter is composed of the mesothoracic ganglion and a series of ganglia shown ontogenetically to be the meta-thoracic, and the first and the second abdominal ganglia (H. Tsujimura, personal communication).

All the DLMs are innervated by the nerve IIN_{1c}. It arises from the dorsal side of the pro-mesothoracic connective, together with the thick nerve trunk IIN_{1b} extending to the wing base. It then deviates from the latter and runs dorsolaterally. After branching to the mesoscutum, the IIN_{1c} splits into a further two nerve trunks. One runs postero-dorsally and laterally to DLM₁ and innervates DLM₂, while the other extends dorsally medially to DLM₁ and innervates DLM_{1e} and DLM₃. The main nerve trunk of IIN_{1c} enters the DLM_{1a} and then runs dorsally while dividing into many branches that innervate DLM_{1a-d}.

The DVMs are innervated by three nerve trunks, IIN₄, IIN_{5b} and IIN₆. Nerve IIN₄ arises together with IIN₂ on the lateral side of the mesothoracic ganglion. It immediately runs between DVM_{1b} and DVM₂ and then divides into many branches that innervate DVM_{1a, b} and DVM₂. Nerve IIN_{5b} is the second branch of the median nerve trunk, IIN₅, which arises from the posterolateral portion of the mesothoracic ganglion. It extends posterolaterally between DVM₂ and DVM₃ and

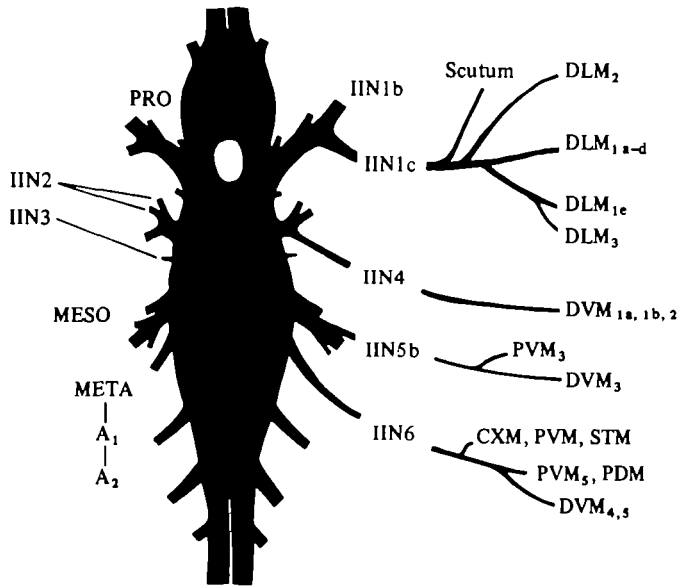


Fig. 2. Innervation of indirect flight muscles in the mesothorax in *B. mori*. The peripheral nerves are designated after Eaton (1974). PRO: prothoracic ganglion, MESO: mesothoracic ganglion, META-A₁-A₂: the complex of the metathoracic and 1st and 2nd abdominal ganglia, PVM: pleuroventral muscle, CXM: coxal muscle, STM: sternopedal muscle, PDM: pleurodorsal muscle. See also Fig. 1.

divides into a few branches to innervate DVM₃ and the pleuroventral muscle (PVM₃). Nerve IIN6 arises on the posterolateral portion of the fused mesothoracic ganglion, runs posteriorly on the medial surface of the DVM₄ and DVM₅ and sends a ventral branch to coxal (CXM), sternopedal (STM), and pleuroventral muscles (PVM). The main nerve trunk of IIN6 passes to the posterior side of DVM₄ and DVM₅ and innervates them with a second branch. It then innervates one of the subalar muscles (PVM₅) and the axillary muscles (PDM).

3. Axons in the relevant nerve trunks

The number and diameter of axons in the nerve trunks innervating the indirect flight muscles is shown in Table 2. These measurements were obtained from examination of sections taken from each nerve trunk where they enter each muscle. It was, however, impossible to determine the number and diameter of axons innervating the muscle bundles of DLM_{1a-d}, DVM_{1a, b}, DVM₂, DVM₄ and DVM₅, because the nerve trunks do not branch until they have entered these groups of muscle bundles. Table 2, therefore, gives only an indication of the minimum number of axons which innervate these groups of muscle bundles.

All but two of the axons innervating DLMs were relatively thick, ranging from 7 to 15 μm in diameter (Table 2 and Fig. 3). The two thin axons were between 2–6 μm in diameter. One innervates DLM₃ and the other, which was presumed to be an unpaired median neurone (UM-neurone), innervates DLM_{1a-d} muscle group. The DVM muscle groups are also innervated by relatively thick axons ranging from

Table 2. *The axons innervating DLMs and DVMs*

Muscle	Number of axons	Diameter (μm)
DLM _{1a-d}	4 (+ 1 small)	10-15 (3-6)
DLM _{1e}	1	8-10
DLM ₂	2	7-8
DLM ₃	1	2-3
DVM _{1a, b} and DVM ₂	4	8-13
DVM ₃	3	6-8
DVM ₄ and DVM ₅	2	8-10

6 to 13 μm in diameter. From these results and the muscle anatomy described above (Fig. 1), it was found that the larger muscles were innervated by the thicker axons. All these axons including that of the UM-neurone were ensheathed by a tissue stained heavily with toluidine blue, which seemed to be glial cells (Fig. 3).

4. *Anatomy of the indirect flight motoneurones*

(1) *General remarks*

Axonal filling with cobalt chloride through the cut end of each of the peripheral nerves revealed the number, location and dendritic arborization of the flight motoneurones (Fig. 4). Fig. 5 summarizes the positions of all motoneurones and unpaired median neurones innervating mesothoracic indirect flight muscles. When nerve IIN_{1c} was filled unilaterally, 9 cell bodies appeared stained: 7 of them are located ipsilateral in the prothoracic ganglion and 2 in the mesothoracic ganglion ($N = 21$). One of the latter 2 cell is located contralateral and the other near the midline of the boundary between the meso- and metathoracic ganglia. It was shown by selectively filling each of the branches of nerve IIN_{1c} ($N = 9$) that 4 large cells of the 7 cells in the prothoracic ganglion innervate DLM_{1a-d}, and 2 intermediate and one small cell innervate DLM₂ and DLM₃, respectively. The contralateral cell in the mesothoracic ganglion innervates DLM_{1e}. The median cell, a UM-neurone, has a single neurite in the anterior portion of the ganglion which then bifurcates to send paired right and left axon to the main trunk of IIN_{1c} innervating DLM_{1a-d}.

Filling nerve IIN₄ resulted in the staining of five cell bodies ($N = 13$). Four of these are located ipsilaterally near the ventral surface of the mesothoracic ganglion. The fifth has a T-shaped axon and is located near the dorsal median surface of the ganglion. Filling a branch of IIN_{5b} which extends to DVM₃ ($N = 24$), and a branch of IIN₆ to DVM₄ and DVM₅ ($N = 4$), revealed that these muscles are innervated by three and two motoneurones, respectively. Their cell bodies are ipsilateral, in the mesothoracic ganglion. These results agree well with the number of axons seen in sections of the relevant nerve trunks.

The cell bodies of these motoneurones are located in the peripheral layer or cortex of the ganglia. All of the paired motoneurones, except those of DVM₃, which are located on the lateral side of the mesothoracic ganglion, have their cell bodies on the ventral surface of the ganglia. This was also confirmed cross sections of pro- and mesothoracic ganglia. Most cell bodies are seen to be located in the ventral or lateral portion of the ganglia, with only a small number on the dorsal median surface.

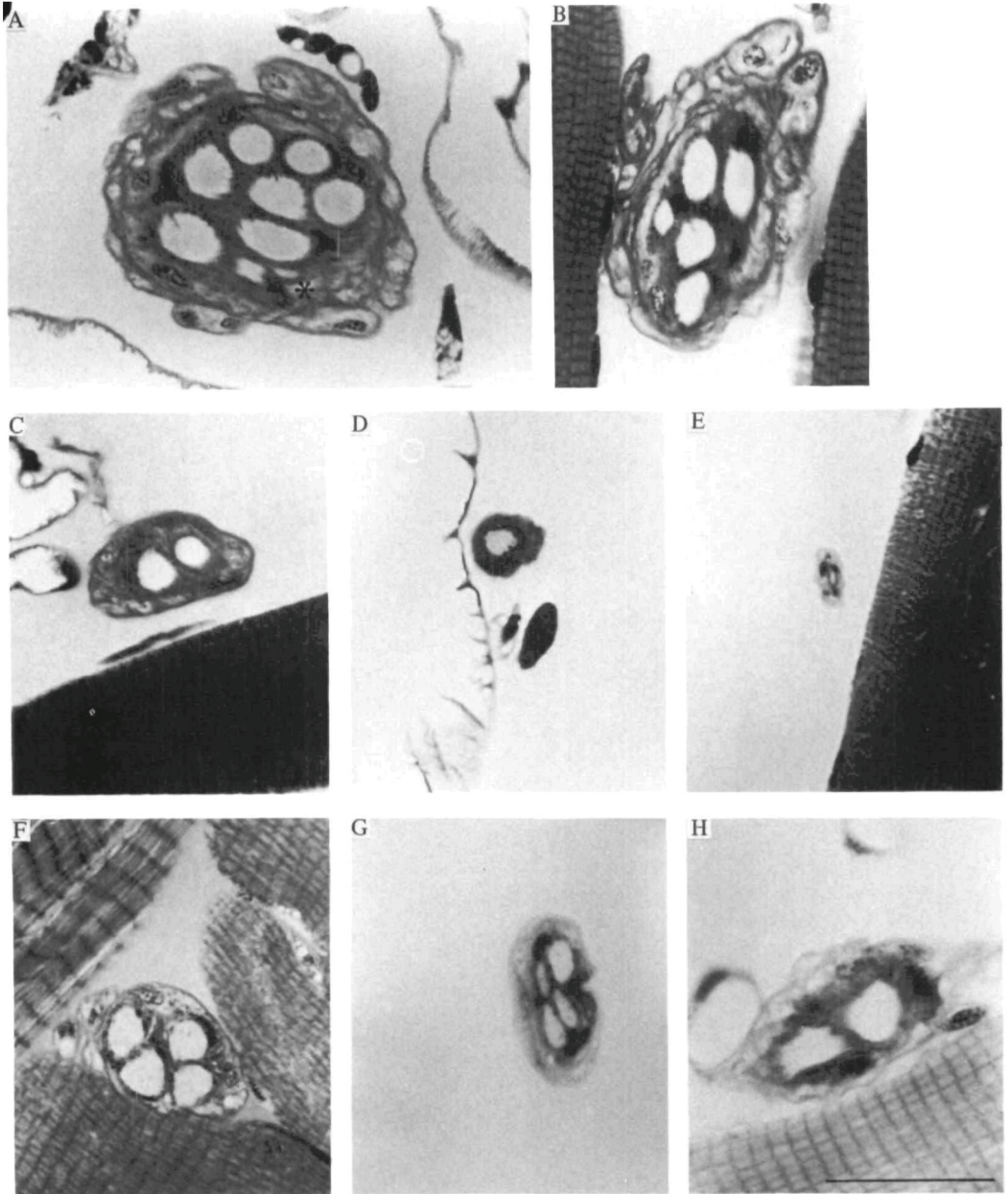


Fig. 3. Cross section of nerves supplying mesothoracic indirect flight muscles. (A) IIN_{1c} containing 9 axons. Serial sections show that an axon-like profile (asterisk) is not any of axons associated with DLMs. (B) The main nerve trunk of IIN_{1c} as it enters DLM_{1a-d}. Four thick (motor) and one thin (UM) axons are recognizable. (C) A lateral branch of IIN_{1c} innervating DLM₂ which contains 2 axons. (D) A median branch of IIN_{1c} which contains one axon innervating DLM_{1e}. (E) A very thin axon innervating DLM₃. (F) IIN₄ as it enters the DVM_{1a, b} and DVM₂. Four thick axons are recognizable. (G) IIN_{5b} after it arises from the mesothoracic ganglion. Three of the axons innervate DVM₃ and the other PVM₃. (H) The second branch of IIN₆ containing two axons as it enters DVM₄ and DVM₅. Scale bar: 30 μ m.

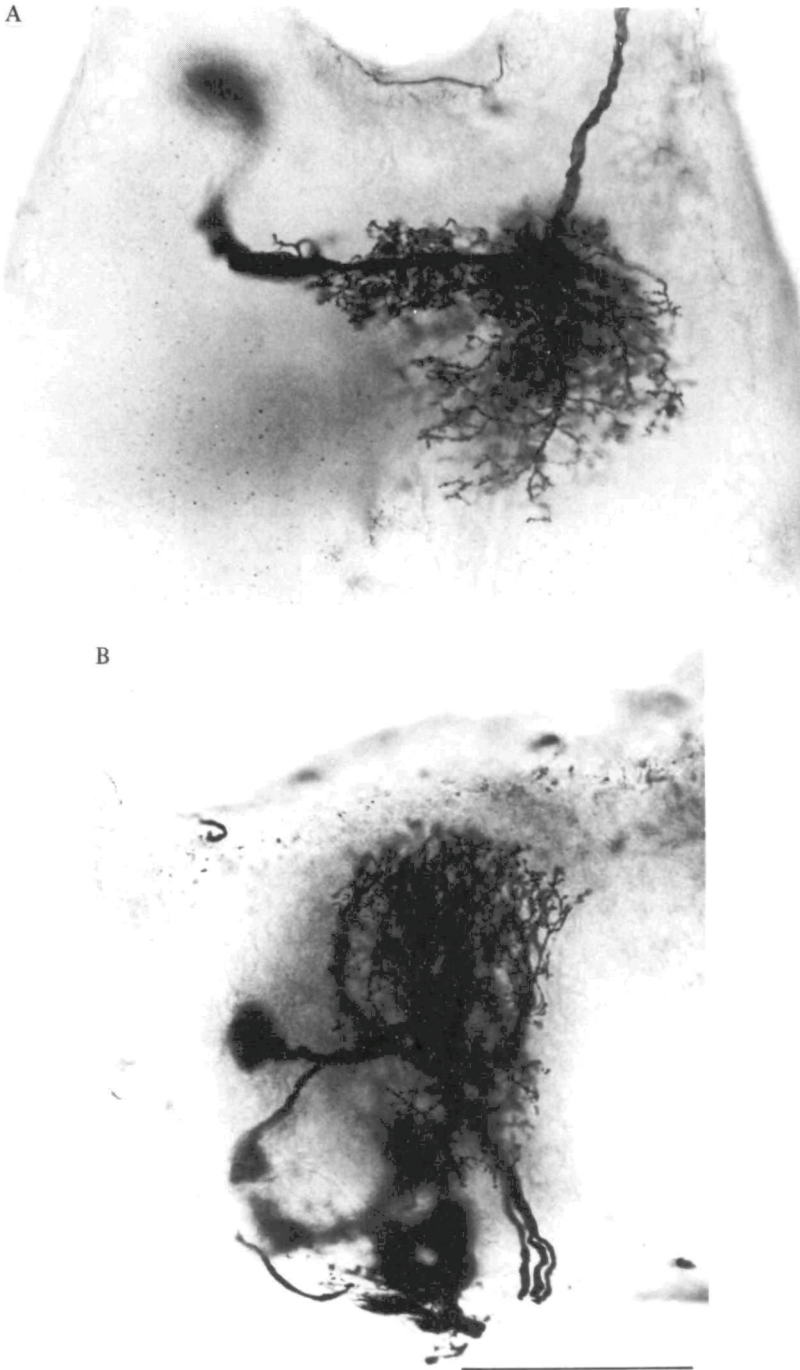


Fig. 4. Photographs of wholemount intensified preparations. (A) Dorsal view of DLM1e motoneurone in the mesothoracic ganglion. (B) Lateral view of DVM3 motoneurons in the mesothoracic ganglion. Scale bar: 100 μ m.

Some cell bodies are strikingly large ranging from about 40–60 μm in diameter and occur in the regions of the ganglia where the cell bodies of indirect flight motoneurons and UM-neurons were found to be located by cobalt filling.

The cell body of the UM-neurone sending an axon to DLM1 *a-d* is located in either the ventral or dorsal surface of the mesothoracic ganglion. In 25 preparations

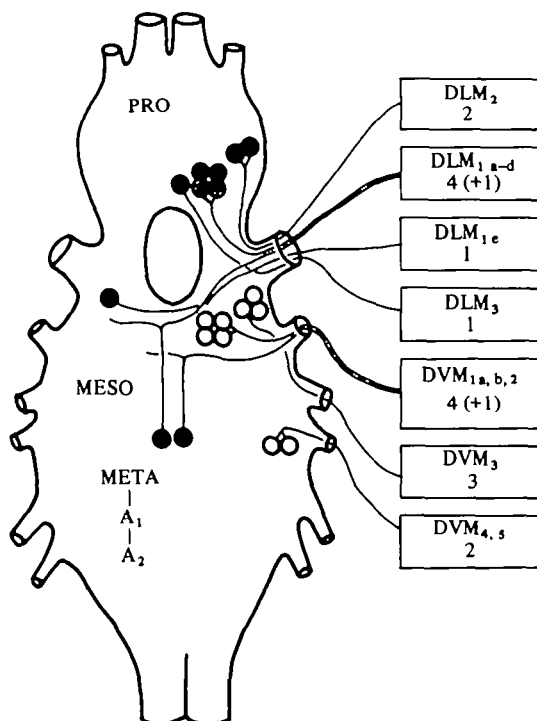


Fig. 5. Motoneurons and unpaired median neurons innervating the mesothoracic indirect flight muscles, showing the location of their cell bodies. The number of neurons innervating each muscle are indicated below the abbreviated name of the muscles. Hatched cells: DLM motoneurons, open cells: DVM motoneurons, filled cells: unpaired median neurons.

filled unilaterally, the cell body appeared on the ventral surface in 15 and on the dorsal surface in 10. Only one such cell body was seen in 9 preparations filled bilaterally. Another UM-neurone with its axon in the IIN₄ innervating DVM1 *a, b* and DVM₂ has its cell body on the dorsal median cortex of the mesothoracic ganglion ($N = 14$).

(2) *DLM* motoneurons

The cell bodies of the four motoneurons innervating DLM1 *a-d* appear clustered on the ventral and posterior portion of the prothoracic ganglion (Fig. 6A, B). They are among the largest (40–60 μm in diameter) in the ganglion. The main neurites from their cell bodies extend into the dorsal neuropile where they branch into many secondary processes. Most of the processes extend towards the dorsal portion of the ganglion and some of them cross the midline. There appears to be a few

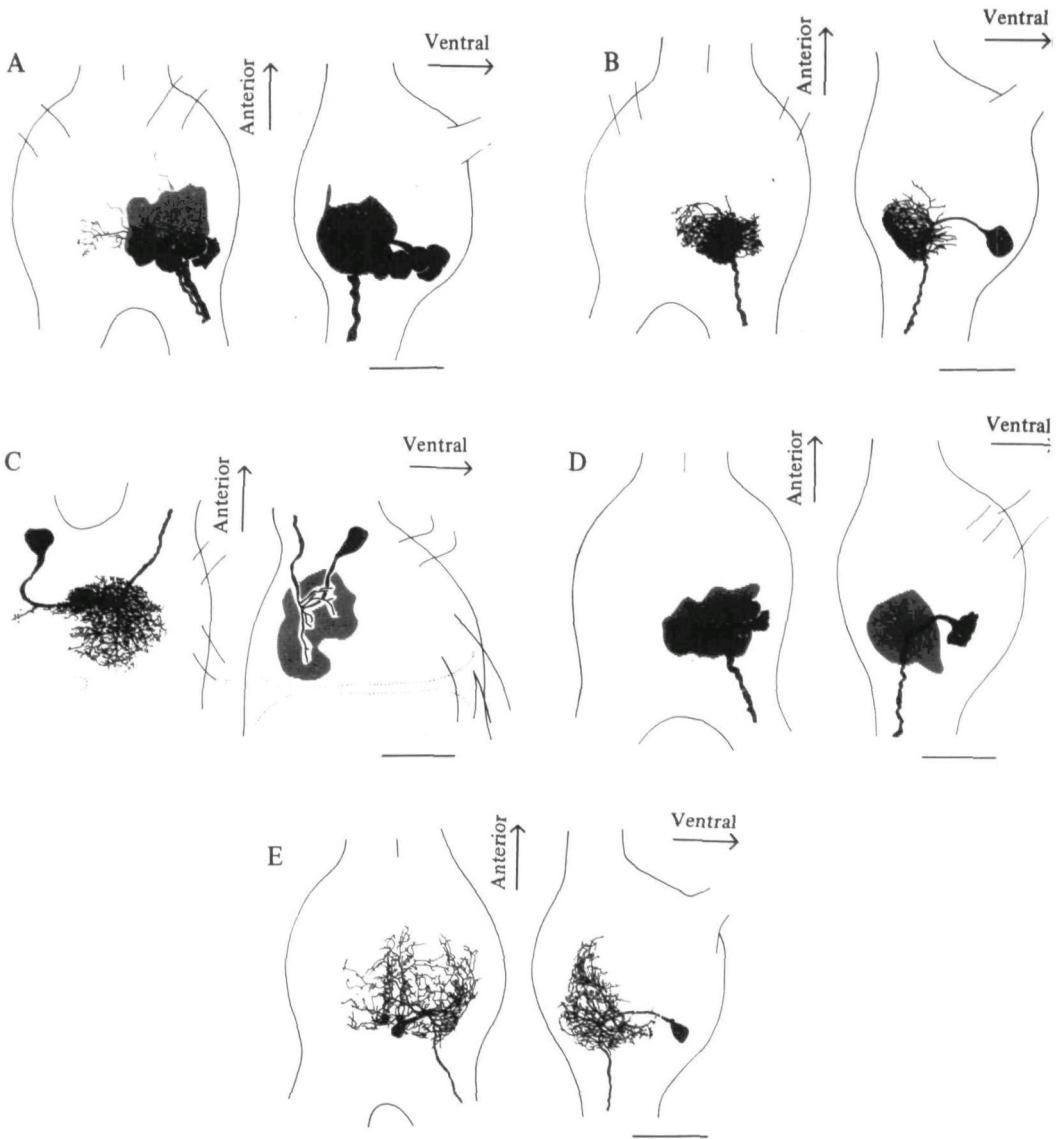


Fig. 6. Dorsal (left) and lateral (right) view of DLM motoneurons. (A) Four motoneurons innervating DLM1*a-d*. The extent of their dendritic processes is indicated by shaded patch. (B) DLM1*a* motoneurone. (C) DLM1*e* motoneurone with its cell body in the contralateral side of the mesothoracic ganglion. The insertion of the neural lamella drawn with dotted line indicates the boundary between meso- and metathoracic ganglia. (D) DLM2 motoneurons. (E) DLM3 motoneurone. Scale bar: 100 μm .

processes extending ventrally (Fig. 6B). The main neurites then run posteriorly into *pro-* and *mesothoracic* connectives.

DLM1*e* motoneurone has a large contralateral cell body, which is 30–45 μm in diameter and located close to the root of the anterior connective in the ventrolateral region of the mesothoracic ganglion (Figs. 4A, 6C). Its neurite runs postero-dorsally

to the dorsal surface of the ganglion and then turns to travel across the midline to the ipsilateral neuropile, where most dendritic branches are located. Short fine dendritic processes are also seen near the midline. In one preparation, some branches were observed contralateral to the muscle (Fig. 6C).

Two DLM₂ motoneurons have cell bodies of an intermediate size (30–40 μm in diameter) and occur slightly anterolateral to DLM_{1a-d} motoneurons in the prothoracic ganglion (Fig. 5). Their main dendritic processes lie ipsilateral to the muscles, although some fine dendritic branches cross the midline (Fig. 6D). The processes, however, are restricted to the posterior half of the ganglion and it seems that the area occupied by them overlaps that of DLM_{1a-d} motoneurons.

DLM₃ motoneurone has the smallest cell body (15–20 μm in diameter) of DLM motoneurons and is near the midline in the posteroventral region of the prothoracic ganglion (Fig. 6E). This motoneurone is characterized by having less dense dendritic processes, compared with those of other DLM motoneurons. A few secondary branches travel across the midline of the ganglion and there give rise to numerous finer branches.

(3) *DVM motoneurons*

The DVM_{1a, b} and DVM₂ muscle bundles are innervated by 4 motoneurons with cell bodies 40–50 μm in diameter (Fig. 7A, B). These neurons are in a close cluster on the ventrolateral surface of the mesothoracic ganglion near the root of the anterior connective. All four neurites run together in a close bundle, extend dorsally and then turn sharply ventrolaterally to the IIN₄ nerve trunk. These motoneurons have dense branches at the point where their neurites turn ventrolaterally. These dendritic branches are confined to the dorsal neuropile ipsilateral to their cell bodies, and have not been observed to travel across the midline of the ganglion. By selectively filling the branch of nerve IIN₄ innervating DVM₂, it was possible to stain the DVM₂ motoneurone alone, so allowing its fine dendritic processes to be observed in detail (Fig. 7B).

The DVM₃ is innervated by three motoneurons which have cell bodies, 20–30 μm in diameter, in the anterolateral surface of the mesothoracic ganglion (Figs. 4B, 7C). The neurites of each of the DVM₃ motoneurons run horizontally to the lateral region of the ganglion, where most of the secondary processes arise dorsally, a few ventrally.

The cell bodies of DVM₄ and DVM₅ motoneurons are 40–50 μm in diameter and are located near the boundary between meso- and metathoracic ganglion (Fig. 7D). Their neurites run dorsally and slightly anteriorly to the dorsal region of the ganglion and there run ventrally. All the dendritic processes are in the posterior two thirds of the ipsilateral side of the ganglion.

(4) *Unpaired median neurones*

Two UM-neurons were found in the mesothoracic ganglion (Fig. 8). Both of those UM-neurons are characterized by these morphological features. (1) their single median neurite bifurcates to form symmetrical left and right axons, (2) dendritic processes are considerably shorter and sparser as compared with paired moto-

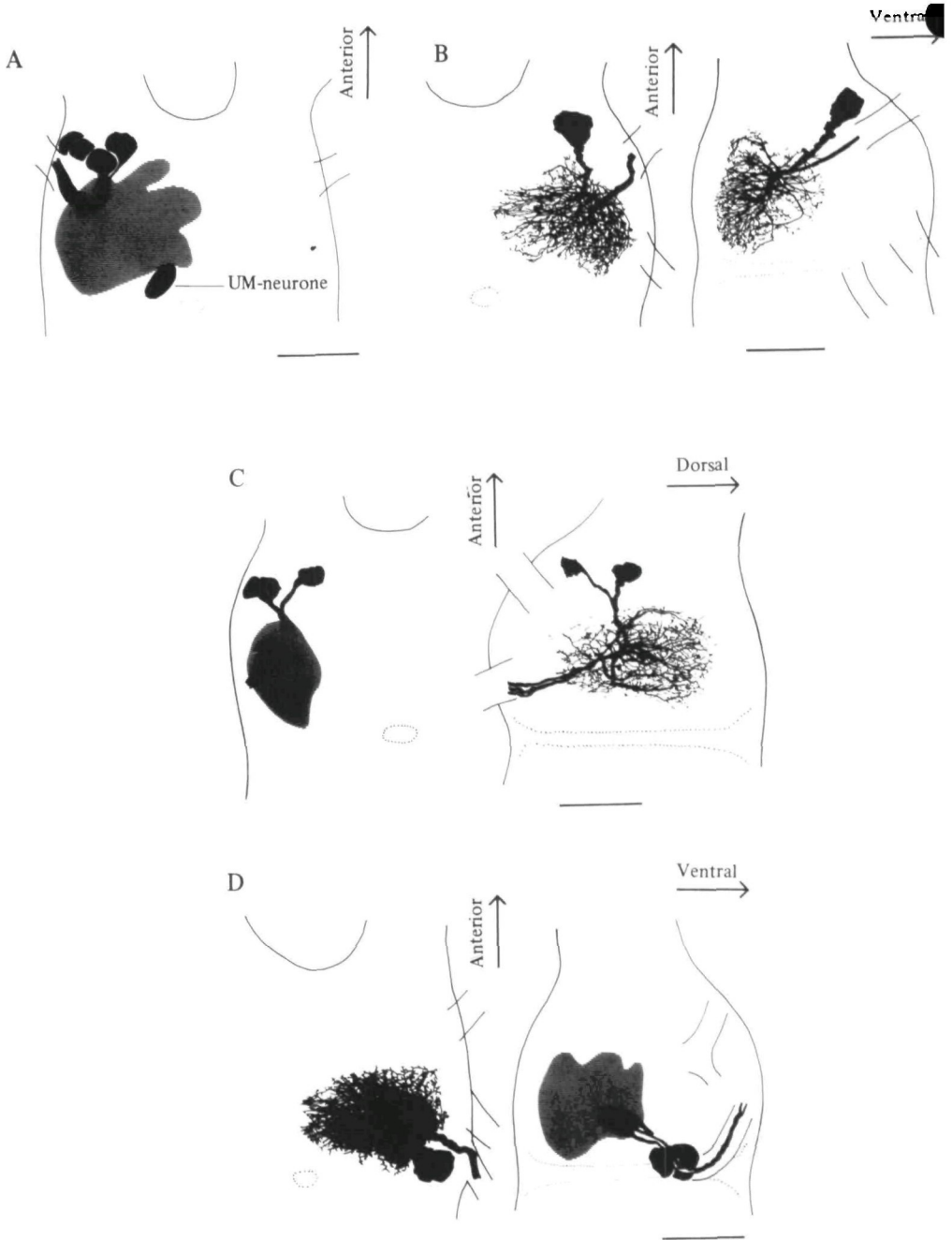


Fig. 7. DVM motoneurons showing the location of their cell bodies and branching pattern. (A) Four motoneurons innervating DVM_{1a}, b and DVM₂. (B) DVM₂ motoneurone. (C) Three tergoprochanteral (DVM₃) motoneurons with the cell bodies in the lateral cortex of the ganglion. (D) Two posterior tergocoxal (DVM₄ and DVM₅) motoneurons with the cell bodies near the boundary of the meso- and metathoracic ganglion. Scale bar: 100 μ m.

neurones examined in the present study, (3) they have relatively large cell bodies ($30\text{--}40\ \mu\text{m}$ in diameter) in the posteromedial surface of the ganglion, and (4) their axons are thin ($3\text{--}6\ \mu\text{m}$) and this seems to be the reason why their cell bodies could not always be filled, and why their axons could not be recognized in some of the microscopical preparations.

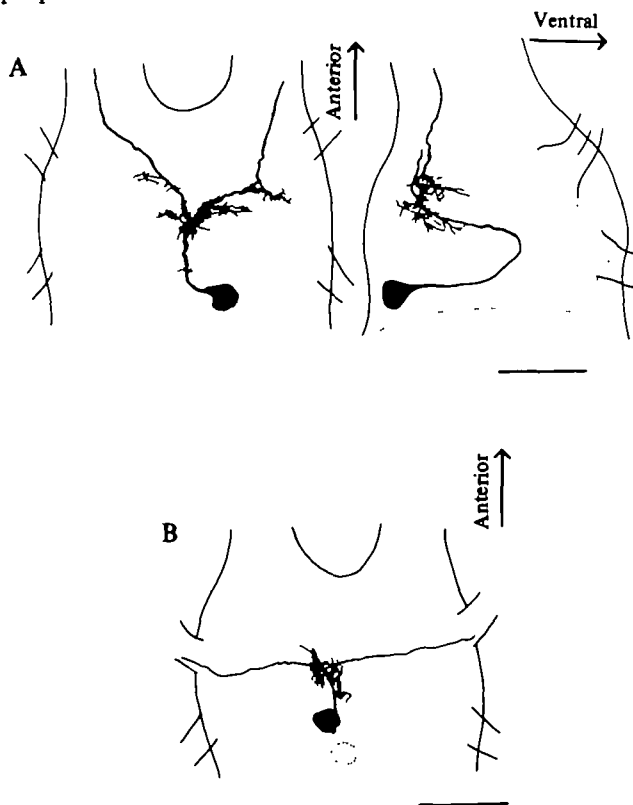


Fig. 8. Camera lucida drawings of unpaired median neurones. (A) UM-neurone associated with DLM_{1a-d} , with the cell body in either dorsal or ventral surface of the mesothoracic ganglion. The cell body is located in the dorsal cortex of the ganglion in this preparation. (B) UM-neurone associated with $\text{DVM}_{1a, b}$ and DVM_2 . Its cell body is located in the dorsal cortex of the mesothoracic ganglion. Scale bar: $100\ \mu\text{m}$.

In spite of the drastic variance in the location of the cell body of the UM-neurone innervating DLM_{1a-d} , there is no observable difference in its basic pattern of dendritic arborization and orientation of neurite. In Fig. 8A a neurone with a dorsal cell body is shown. Its neurite runs ventrally up to a depth of $200\text{--}300\ \mu\text{m}$ from the dorsal surface and then turns anterodorsally to the dorsal neuropile where it bifurcates to form paired right and left axons. A small number of short and fine secondary processes arise near the T-shaped region of the median neurite and also in the paired neurites.

DISCUSSION

1. *Motoneurones innervating indirect flight muscles*

Mesothoracic indirect flight motoneurones have been described in cricket (Bentley, 1973), locust (Bentley, 1970; Burrows, 1973; Altman & Tyrer, 1974; Tyrer & Altman, 1974), dragonfly (Simmons, 1977), Heteropteran (Davis, 1976), and Dipteran insects (Ikeda *et al.* 1975, 1976; Coggshall, 1980). In Lepidopterous insects only the DLM motoneurones in *Manduca sexta* have been described (Casaday & Camhi, 1976). The present study provides the first data on DVM neurones as well as DLM neurones, which will be discussed in terms of their arrangement in the ganglion and morphological features of their arborization.

(1) *Arrangement of the mesothoracic indirect flight motoneurones*

A comparison of the arrangement of DLM and DVM motoneurones in *B. mori* with that of other insects may allow us to imagine the general neural organization of the insect flight system. Thus, the number and location of indirect flight motoneurones in the mesothorax shows some common features among the studied species (Table 3). First, the location of the DLM neurones in the ganglia is the same throughout these insects. Motoneurones innervating the four bundles of DLM (DLM_{1 a-d} in *B. mori*), the dorsal oblique muscle, and another DLM (DLM₃ in *B. mori*) have their cell bodies in the ipsilateral and posterior portion of the next anterior ganglion, while the motoneurone of one DLM (DLM_{1 e} in *B. mori*) is in the anterior of the contralateral half of the ganglion in the same segment as the muscle. In the locust (Neville, 1963; Bentley, 1970) and in *Drosophila* (Coggshall, 1980), the contralateral neurone innervates the dorsal most bundle of the major dorsal longitudinal muscles (DLM_{1 e} in *B. mori*), while the four ipsilateral motoneurones in the prothoracic ganglion innervate the more ventral bundles. Secondly, the number of motoneurones to the DLM is the same in all the insects examined except for DOM in *Dysdercus* which has three motoneurones. Thirdly, the dorsoventral muscles are all innervated by motoneurones of the same segment, whose cell bodies are ipsilateral to the muscles they innervate. Of the DVM motoneurones, those of tergo-sternal, anterior tergo-coxal and tergotrochanteral muscles are at the anterior end of the mesothoracic ganglion, while those to posterior tergo-coxal muscles are at the posterior end.

This basic architecture of motoneurones innervating dorsal longitudinal muscles and dorsoventral muscles is similar to that in the abdominal ganglion of the moth (Taylor & Truman, 1974), supporting the idea that insect segmental ganglia might have a basic plan for the arrangement of motoneurones (Taylor & Truman, 1974).

(2) *Arborization of the indirect flight motoneurones*

One remarkable feature is that the dendritic processes of dorsal longitudinal motoneurones, including that to the dorsal oblique muscles, extend to the contralateral neuropile; whereas dorsoventral motoneurones have a dendritic field restricted to the ipsilateral half of the ganglion. This is also the case in locust (Tyrer & Altman, 1974). The functional meaning of the difference in the extent of arborization between dorsal longitudinal and dorsoventral motoneurones is uncertain. The dorsal longi-

Table 3. Motoneurons innervating mesothoracic indirect flight muscles in the different insects

Muscle	<i>Bombyx</i> (moth) Kondoh & Obara (in the present paper)	<i>Manduca</i> (moth) Casaday & Camhi (1976)	<i>Drosophila</i> (fly) Coggshall (1980)	<i>Dysdercus</i> (Hemiptera) Davis (1976)	<i>Schistocerca</i> (locust) Bentley (1970)	<i>Hemianax</i> & <i>Aeshna</i> (dragonfly) Simmons (1977)
Dorsal longitudinal	DLM _{1a-e} 4-PI 1-MC DLM ₂ 2-PI DLM ₃ 1-PI DVM _{1a, b}	DLM _{1a-e} 4-PI 1-MC DLM ₂ 2-PI DLM ₃ 1-PI DVM _{1a, b} and <i>c</i>	DLM _{1a-f} (45 <i>a-f</i>) 4-PI 1-MC 46 <i>a, b</i> ? — — 47 <i>a, b, c</i>	DLM 4-PI 1-MC DOM 3-PI — —	81 4-PI 1-MC — — — 83, 84	d1m 4-PI 1-MC — — — dvm1
Dorsal oblique	DVM ₂	DVM ₂ ?	?	DVM 2-MI	2-MI 89 2-MI	3-MI dvm6 6-MI —
Another dorsal longitudinal	DVM ₃ 3-MI DVM _{4, 5} 2-MI	DVM ₃ ? DVM _{4, 5} ?	66 ? 48 <i>a, b</i> ?	— — — —	103 3-MI 90 3-MI	dvm7 5-MI
Tergosternal	DVM _{1a, b}	DVM _{1a, b} and <i>c</i>	47 <i>a, b, c</i>	—	83, 84	dvm1
Anterior tergocoxal	DVM ₂	DVM ₂ ?	?	DVM 2-MI	2-MI 89 2-MI	3-MI dvm6 6-MI —
Tergotrochanteral	DVM ₃	DVM ₃ ?	66	—	103	—
Posterior tergocoxal	DVM _{4, 5} 2-MI	DVM _{4, 5} ?	48 <i>a, b</i> ?	— —	90 3-MI	dvm7 5-MI

The number below the abbreviation of each muscle depict the number of innervating motoneurons. PI, MC and MI indicate the position of cell bodies in the ganglion as follows. PI: ipsilateral in the prothoracic ganglion, MC: contralateral in the mesothoracic ganglion, MI: ipsilateral in the mesothoracic ganglion.

tudinal motoneurons on each side of the ganglion, however, may morphologically and consequently physiologically have contact with each other through their branches extending over the contralateral hemiganglion.

By filling the dorsal nerve of the 4th abdominal ganglion which is probably homologous to IIN_{1c} innervating DLMs, Truman & Reiss (1976) revealed one contralateral motoneurone in the ganglion of the same segment as the nerve filled in the pharate adult of *Manduca*. On the basis of homology of the nerve filled and its innervating muscles (H. Tsujimura, personal communication), the 4th abdominal contralateral neurone can be safely judged homologous to the DLM_{1e} motoneurone. This abdominal motoneurone has two dendritic fields on each side of the midline which are nearly of the same size. This is in sharp contrast to the homologous neurone (DLM_{1e}) in the mesothoracic ganglion in *B. mori* which has scarce dendritic processes on the side contralateral to the axon. It is, however, not known whether this difference results from the difference in segment, developmental stage, or species.

2. Unpaired median neurones

In the present study, two unpaired median neurones were revealed. One of them is in either the ventral or dorsal median cortex of the mesothoracic ganglion, while the other is always in the dorsal median cortex. These neurones were shown to be unpaired by the results of bilateral filling. This is also supported by their morphological similarity to DUM neurones in locust (Plotnikova, 1969; Crossman *et al.* 1971, 1972; Hoyle, 1974, 1978).

DUM neurones associated with the dorsal longitudinal muscles (DUMDL) have been reported for Orthopterans (Bentley, 1973; Clark, 1976; Hoyle, 1978), a Heteropteran (Davis, 1978), fruit fly, *Drosophila melanogaster* (Coggshall, 1980) and a saturniid moth, *Antheraea polyphemus* (Heinertz, 1976). Taylor & Truman (1974) have also described six ventral and four dorsal UM-neurones in the 4th abdominal ganglion of *Manduca*. Alternative location, in the dorsal or ventral surface of ganglion, has not been previously described for a UM-neurone. It may arise in *B. mori* simply by developmental accidents.

The UM-neurone associated with the DVM has not been previously observed.

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