

SHORT COMMUNICATION

SOME MOTOR NEURONES OF THE ABDOMINAL
LONGITUDINAL MUSCLES OF GRASSHOPPERS AND
THEIR ROLE IN STEERING BEHAVIOUR

BY A. BAADER

Zoologisches Institut der Universität, Basel, Switzerland

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The abdomen is used by acridid grasshoppers for steering during flight (Camhi, 1970a; Gewecke & Philippen, 1978; Taylor, 1981; Altman, 1982; Arbas, 1986; for a review see Rowell, Reichert & Bacon, 1985). The aim of the present study is to characterize some of the various motor neurones innervating the dorsal and ventral longitudinal muscles of the previously undescribed second abdominal segment (ASG2) of *Locusta* and to describe their activity during steering.

Abdominal movements have been intensively studied in tethered *Schistocerca gregaria* (Cyrtacanthacridinae), flying in a wind stream in the dark (Camhi, 1970a,b). When this wind jet was rotated around the vertical axis of the animal some degrees away from the frontal direction (i.e. simulating yaw), the abdomen was moved laterally to the same side. In a free-flying animal such movement would tend to neutralize the perceived yaw. Abdominal movement occurred within 300 ms of stimulation onset, and mainly as a result of the contraction of longitudinal musculature in the first and second abdominal segments. Visually induced yaws resulted in a similar deflection of the abdomen which, however, also involved more posterior segments. None of these abdominal movements took place if the animal was not flying. Basically similar observations have been made by the other authors cited above, using a variety of genera.

The ultrastructure and electrophysiology of the nerve supplying the longitudinal muscles of the first abdominal segment (ASG1) was studied by Camhi & Hinkle (1974). Fifteen axons including five axons with diameter $>10\ \mu\text{m}$ were found using the light microscope. In quiescent animals only some fibres were active, in phase with respiratory pumping movements of the abdomen. In flight, more fibres were active and some modulated at flight frequency. This modulation continued after the destruction of all peripheral thoracic nerves, suggesting that it derived from the flight oscillator rather than from sensory feedback (Camhi & Hinkle, 1972).

The innervation of the homologous muscles of ASG4 in *Melanoplus* (Melanoplinae) was studied by Tyrer (1971a,b). He described eight axons innervating the dorsal median internal muscles (m182) and two more innervating the median

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external muscle as well as 11 presumed sensory axons coming from the heart (total 21). Each of the five dorsal median internal muscles was innervated by all eight motor axons. Further description of the innervation of the dorsal longitudinal muscles of segment four (Lewis, Miller & Mills, 1973) and segment one (Yang & Burrows, 1983) agree in general with the results of Tyrer.

The peripheral distribution of the dorsal nerve of ASG2, Ab2DNV (nomenclature of Tyrer & Gregory, 1982) was revealed with intensified orthograde cobalt fills, and was similar to that described for Ab4DNV of *Melanoplus* (Tyrer, 1971a). The branching pattern of Ab2DNV is shown in Fig. 1.

Ab2DNV was back-filled with 5% CoCl₂ solution distal to its innervation of muscle 151, to stain the motor neurones of the external lateral muscle 149 and the lateral internal dorsal muscle 150. Seven to nine neurones distributed in the first and second abdominal neuromere of the fused metathoracic ganglion were repeatedly filled (Fig. 2). The dendritic fields and axons of these cells were located just below the dorsal surface of the ganglion, whereas the somata were ventral, about 260 μm below the dorsal surface. The size of the somata was 10–30 μm . Four had ipsilateral somata; their ipsi- and contralateral arborizations extended into the neuromere of abdominal ganglion one (AG1). Four had contralateral somata. One neurone had its soma in the dorsal midline at a depth of 150 μm , but only a unilateral axon. Further cobalt backfills of the whole nerve close to the ganglion revealed up to 16 additional neurones and showed that six motor neurones of the dorsoventral and ventral muscles were very similar in location and shape to those of the dorsal muscles.

Motor neurones innervating the ventral and dorsal muscles of ASG2 were also stained intracellularly with Lucifer Yellow (Fig. 3). AG1 contains motor neurones that are presumably homologous, which are active during the expiratory phase of ventilation (Yang & Burrows, 1983) but whose role in steering has not been examined.

Many intersegmental dorsal longitudinal muscles (e.g. flight muscles 81 and 112) are innervated by two sets of motor neurones: neurones with contralateral somata in the posterior ganglion, and a cluster of neurones with ipsilateral somata in the anterior ganglion. Similarly, motor neurones of the dorsal longitudinal muscles of AG4 have somata located partly in the contralateral half of AG4 and partly in the ipsilateral half of AG3 (Lewis *et al.* 1973) and Yang & Burrows (1983) also showed that the dorsal longitudinal muscles of AG1 are innervated from both contralateral and ipsilateral cell bodies, some of which are in the metathoracic, and some in the first abdominal, neuromere. The same pattern was found in the present study, where neurones with contralateral somata were located in the posterior neuromere (AG2) and some of the neurones with ipsilateral somata were in the anterior neuromere (AG1, see Fig. 2). The axons of both, however, ran in Ab2DNV. This suggests, therefore, that the external fusion of nerve 6 of the anterior neuromere with nerve 1 of its posterior neuromere, seen in other segments, has been internalized in these neuromeres. The abdominal neurone with the medial cell body has no obvious homologue in the motor neurones of the longitudinal muscles of other segments of the locust.

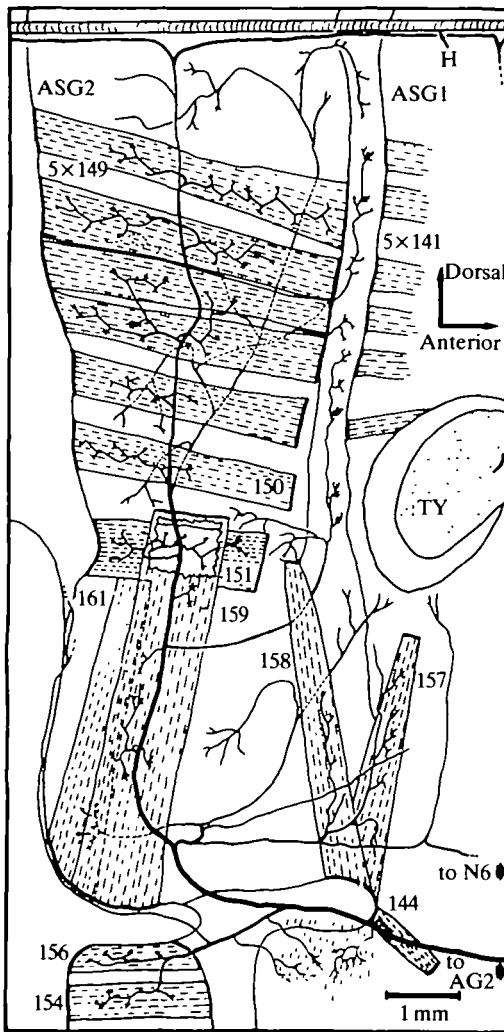


Fig. 1. The course of left nerve Ab2DNV within the second abdominal segment (ASG2) revealed by CoCl_2 staining. Relevant muscles are numbered (Snodgrass, 1935). The nerve leaves the fused metathoracic ganglion and branches above the anterior apodeme of the sternum, supplying the ventral muscles 154 and 156. The next branch innervates the dorsoventral muscles 157, 158 and 161. It also innervates the ventrolateral side of ASG2 (probably sensory), and gives off a further branch which fuses with metathoracic nerve six (N6). More distally, the main trunk gives rise to several small branches to the inner tergosternal muscle 159, the paradorsal muscle 151 and a sensory branch to epidermis between ASG1 and ASG2. At this point a tonic stretch receptor can be recorded extracellularly; it is excited by bending the abdomen contralaterally and inhibited by the opposite movement. Between muscle 151 and the lateral internal dorsal muscle 150 another sensory branch enters, which transmits pressure information from much of the lateral and dorsal surface of ASG2. The nerve then innervates the five bundles of the external lateral 149 and finally joins the median dorsal nerve of the heart. H, heart; TY, tympanal organ; AG2, abdominal ganglion 2.

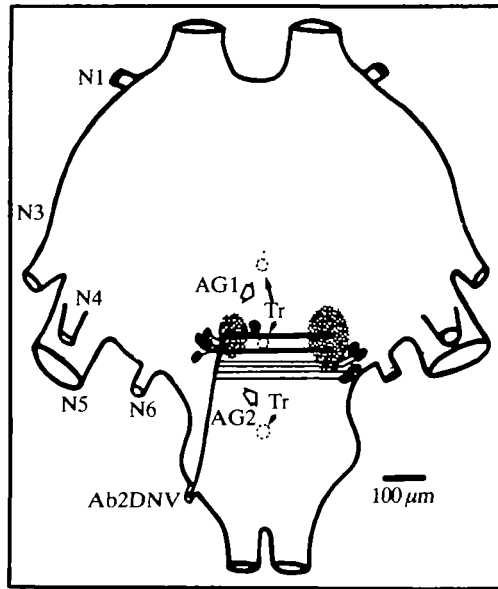


Fig. 2. Schematic reconstruction of nine motor neurones which were identified by filling the dorsal nerve of abdominal ganglion 2 (Ab2DNV of AG2) with CoCl_2 distal to its innervation of paradorsal muscle 151. Somata, axons and dendritic fields (shaded areas) are partly located in the neuromere of abdominal ganglion 1 (AG1). Abdominal neuromeres are divided according to the criteria of Tyrer & Gregory (1982 and personal communication). Tr, median trachea.

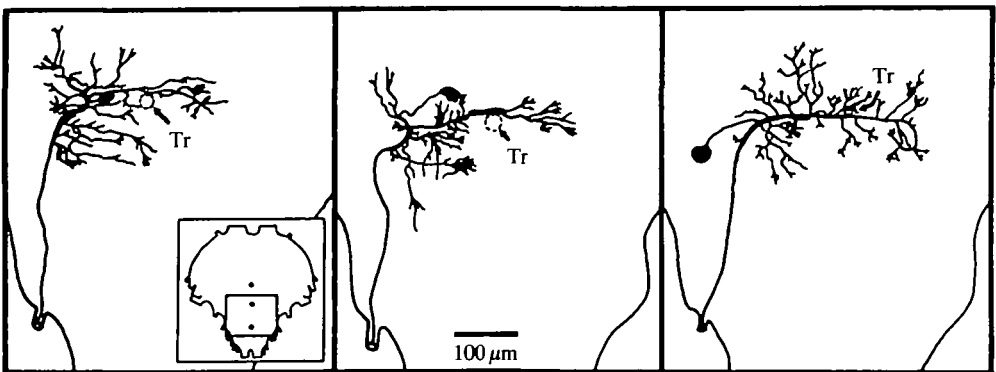


Fig. 3. *Camera lucida* reconstruction of three motor neurones of ASG2 filled intracellularly with Lucifer Yellow. The left-hand neurone was only excited during flight periods and corresponded to a small unit in the nerve Ab2DNV. The central neurone, the response of which to horizontal roll turns is shown in Fig. 4D, also corresponds to one of the small extracellular units. The third motor neurone innervates ventral muscles of ASG2; it responded to horizontal roll stimuli and was tonically excited in flight. Note that neurone branches extend anterior to the median trachea which divides AG1 from AG2. Tr, median trachea.

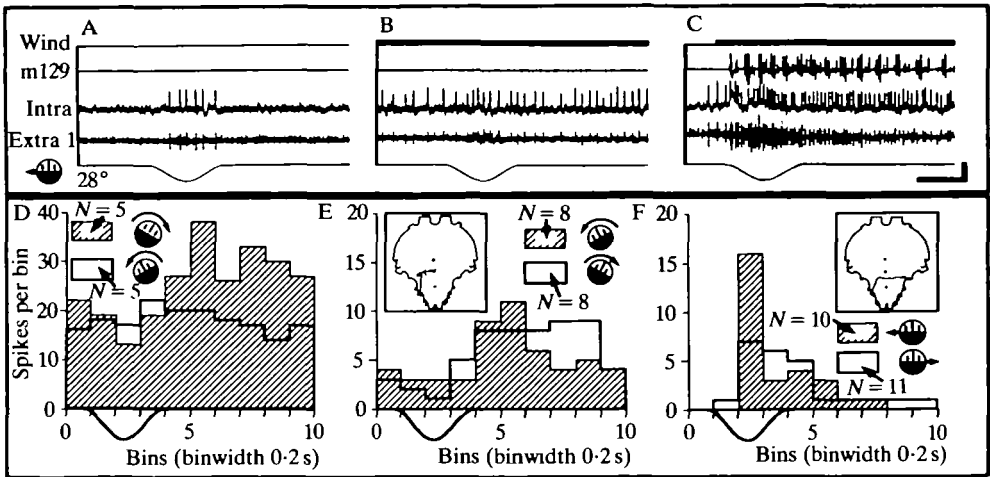


Fig. 4. (A–C) Recordings from a motor neurone with a contralaterally located soma (drawn in inset of F). It was excited during rest by a horizon yaw (20° deflection, A) and by wind (B) and increased its activity when the preparation was flying (C). Time bar for A–C: 0.5 s, voltage bar: 12 mV. (D–F) Post-stimulus time histogram for three examples of responses of abdominal motor neurones to horizon stimuli. Intra, intracellular recording of motor neurone; Extra 1, extracellular recording of Ab2DNV. (D) This neurone (drawn in Fig. 3 centre) was inhibited during a horizon roll to the right (contralateral) side and subsequently excited; a reversed stimulus led to no strong reaction. A similar response was produced by another neurone (E): inhibition and post-inhibitory excitation to roll turns of the horizon but without any preference for a particular direction. The post-stimulus time histogram in F (the same neurone as in A–C) shows a pronounced excitation to yaw to the ipsilateral side and a less prominent excitation to yaw to the contralateral side, in a non-flying animal.

Extracellular stimulation of the entire Ab2DNV with current pulse trains led to a contraction of the abdominal muscles, whereas suprathreshold intracellular stimulation of single motor neurones of the dorsal longitudinal muscles never resulted in visible twitches. This observation, like the observed morphological convergence of eight axons on single muscles (Tyrer, 1971a), suggests that summation of several neuronal inputs is necessary for contraction.

Wind alone had no effect on most of the recorded neurones. In only one ventral longitudinal muscle motor neurone was a response to wind seen in a non-flying animal (Fig. 4A,B). To study the activity of the motor neurones during steering behaviour, the animal was mounted with its head at the centre of rotation of a movable hemispherical horizon device, equipped with a central wind jet (for details see Rowell & Reichert, 1986): course deviations were simulated both visually and through wind stimulation. First, motor neurones frequently responded to horizon movement when the animal was not flying (Fig. 4A). That is, drive from the flight oscillator was not invariably necessary to bring the abdominal motor neurones to threshold. Second, in most cases the response of the motor neurone to horizon movement, which could take the form of either an inhibition with a postinhibitory

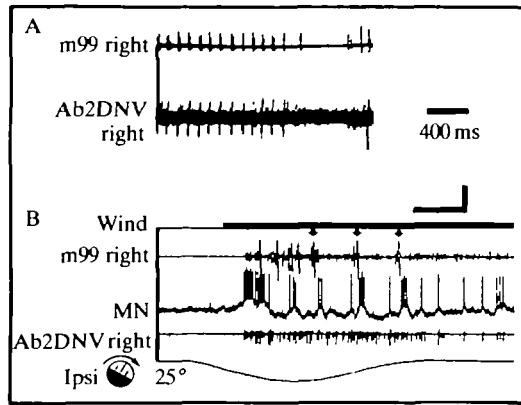


Fig. 5. (A) Extracellular recording of Ab2DNV between m150 and m151 showing that one unit is active in the rhythm of flight frequency (monitored at subalar m99). (B) In this example a clear correlation could be seen between flight-like activity in the right subalar m99 and bursts in an abdominal motor neurone (MN) (see arrows). Flight was elicited by wind and a horizontal roll to the ipsilateral side. Scale bars, 200 ms, 10 mV.

excitation (Fig. 4D,E) or an excitation (Fig. 4F), depending on the neurone and the type of movement, was not reversed when the direction of horizon movement was reversed: that is, the recorded responses were not obviously linked to a compensatory steering behaviour. Third, all the recorded neurones were excited during flight. However, this activity was usually *not* modulated at wing beat frequency.

While many of these results agree with those of previous authors, some require comment. (a) Behavioural experiments (Taylor, 1981) indicate that although abdominal steering movements are much weaker in non-flying animals, they can be recorded in response to visual stimulation as shown here also. A similar observation was made by Kien (1977), who used a moving stripe pattern to see turning reactions in non-flying locusts. Camhi & Hinkle (1974) did not see any such response, but they were simulating yaw with wind alone, which is known to produce less directional information in some of the descending interneurones signalling course deviation than either vision or wind and vision combined (Rowell & Reichert, 1986). (b) The observation that not all responses to horizon displacement were qualitatively directional presumably indicates that the mechanism of abdominal movement is complex. For example, bending the abdomen could be achieved by activating the muscles of both sides, but with different intensities (Fig. 4F). Such a co-contraction would also increase the stiffness of the abdomen. (c) Camhi & Hinkle (1972) showed that a small rhythmic component is present in the drive to the abdominal motor neurones during flight, which is transmitted to the abdominal ganglia neurally. Although corresponding rhythmic activity of motor neurones can be recorded extracellularly in the nerve Ab2DNV (Fig. 5A), it was almost never seen in the motor neurones penetrated intracellularly in this study. The one exception (Fig. 5B) showed a clear modulatory pattern only occasionally, when the flight oscillator was running slowly and erratically.

Summarizing, the present results mostly fit the picture of abdominal steering drawn by previous workers. Sensory information about course deviations reaches the thoracic and abdominal ganglia *via* a population of descending interneurons, and the animals often – though by no means always (see Thüring, 1986) – react with steering movements of wings and abdomen. In the thoracic ganglia the sensory information reaches the flight motor neurones both directly and also *via* premotor interneurons, where it is mixed with both phasic and tonic drive from the flight oscillator circuit (Reichert & Rowell, 1985). A similar hypothesis was proposed by Camhi & Hinkle (1974) for the abdominal motor neurones. The present results require that this hypothesis be modified. Most abdominal motor neurones do not require drive from the oscillator in order to react to deviation-detector inputs, and not all receive oscillatory drive during flight. Indeed, this drive may reach only a few abdominal motor neurones. The response to sensory input can have directional and non-directional components. Finally, it must be emphasized that the interneuronal source of neither sensory nor oscillatory drive to these motor neurones has yet been identified.

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