Motor Neurons that Multitask

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Animals use a form of sensory feedback termed proprioception to monitor their body position and modify the motor programs that control movement. In this issue of *Neuron*, Wen et al. (2012) provide evidence that a subset of motor neurons function as proprioceptors in *C. elegans*, where B-type motor neurons sense body curvature to control the bending movements that drive forward locomotion.

Motor neurons are most often viewed from the perspective of their efferent actions on muscles. This highly specialized function is a hallmark feature of vertebrate motor neurons, although some mammalian motor neurons also provide feedback to the motor system via inhibitory interneurons known as Renshaw cells (Alvarez and Fyffe, 2007). Generally speaking, however, vertebrate motor neurons lack the ability to "walk and chew gum at the same time." By contrast, many of the motor neurons found in simple invertebrate motor systems are multifunctional, the motor neurons of the crustacean stomatogastric ganglia (STG) being a case in point. In addition to innervating the gut musculature, they also synapse with other STG motor neurons and play critical roles in establishing the rhythmic motor patterns that are necessary for feeding and digestion (Harris-Warrick et al., 1992).

Locomotor movements in most organisms are produced by central pattern generator networks that are strongly influenced by sensory feedback (Büschges et al., 2008; Rossignol et al., 2006). Proprioception, the internal monitoring system that senses body position and movement, is particularly important for motor control. Proprioceptive signals are typically gauged by specialized sensory neurons that measure the stretch or dynamic forces generated by the musculoskeletal system. The monosynaptic reflex first described by Charles Sherrington is a classical example of a simple proprioceptive feedback pathway (Sherrington, 1906). It is comprised of sensory neurons with specialized spindle endings that encircle the intrafusal fibers of a single muscle. These sensory neurons directly report stretch to the alpha motor neurons innervating that same muscle. While the monosynaptic reflex has long been considered a model of cellular economy, it now appears to be trumped by the motor neurons that generate forward movements in the nematode *C. elegans*. In this issue of *Neuron*, Wen and colleagues (Wen et al., 2012) have examined the proprioceptive feedback pathway that worms use to coordinate and propagate forward locomotor movements. Their findings point to motor neurons themselves functioning as proprioceptive sensors, an idea that was first proposed by Richard Russell and Lou Byerly many years ago.

C. elegans propel themselves by rhythmic serpentine movements that are driven by dorsal-to-ventral bending movements of the torso. These movements are generated by four bands of longitudinally aligned body wall muscles that sit beneath the outer cuticle (Altun and Hall, 2008; Figure 1). Worms can either move forward or backward, with A- and B-type cholinergic motor neurons being the primary effectors of backward and forward locomotion, respectively. The motor neurons and muscles that control these movements are arranged somatotopically along the length of the body (Figure 1A), and they are activated in wave-like fashion during forward or backward locomotion. Using an elegant combination of molecular genetics, biomechanical manipulation, and imaging, Wen et al. (2012) asked how worms propagate the bending movements that drive forward locomotion. To do this, they partially immobilized the worms in microfluidic chambers while monitoring their motile behavior. By constraining the worm midway along its torso in a specially etched channel, they were able to isolate the head and tail so that these two body regions could move independently. They then undertook a series of experiments in which they changed the curvature of the channel, noting that increasing the degree of curvature in the middle of the worm's body caused more posterior regions to bend accordingly (Figure 1B); moreover, the greater the curvature of the torso, the greater the bending of the tail region. Two optogenetic approaches were used to show this bending is an active process. The first involved expressing the chloride channel NpHR in body wall muscle cells and hyperpolarizing the cells with light. This caused the tail region to relax and lose its curvature. A second more telling experiment used the genetically encoded calcium reporter GCaMP3 to show that the body wall muscles are more active on the inside of the curve than on the outside of the curve. They then did something rather clever. They used a pneumatic microfluidic device to change the curvature of the trapped worm and examine what happens to more posterior segments. Rapidly changing the channel curvature from a dorsal to ventral bias, or vice versa, resulted in a corresponding change in the curvature of the posterior body (Figure 1B). From these experiments, they concluded that the body of the worm senses curvature and is able to relay this information to more posterior segments, which then follow suit.

What is the cellular mechanism that allows worms to sense body curvature and propagate the bending movement to more posterior segments? Could the body wall muscles themselves propagate this proprioceptive signal, given that they are coupled by gap junctions? A combination of classical genetics and

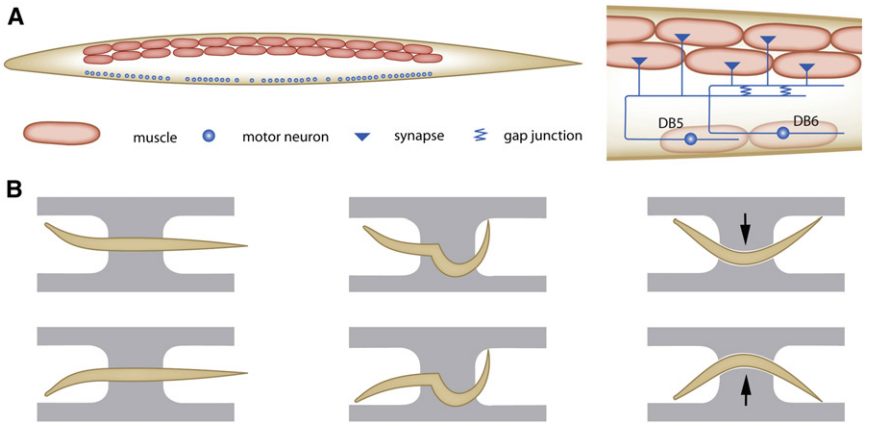

Figure 1. Proprioception in *C. elegans*

(A) Schematic showing the anatomical arrangement of motor neurons and body wall muscles in *C. elegans*. (Left) Motor neurons (blue) are located ventrally and synapse with 95 body wall muscles that are arranged in two dorsal and two ventral bands that encircle the body. A single dorsal muscle band comprised of 24 muscles arrayed along the anterior-posterior axis of the worm is shown. (Right) High magnification schematic showing the innervation of dorsal muscles by two DB neurons. Motor neurons have elongated ventral processes and are electrically coupled via gap junctions.

(B) Placing worms in the channel of a microfluidic device allows free movement of their head while the tail adopts the same curvature as the body region positioned within the microfluidic device (left and middle). Altering the curvature of the channel causes a corresponding change in the curvature of the tail (right).

optogenetic manipulation was employed to rule this possibility out. Mutant worms that lack functional gap junctions between muscle cells were still able to sense and propagate bending. There was also no change in the curvature of the posterior body when muscles located within the channel were hyperpolarized with NpHR, nor did localized channelrhodopsin-induced contraction of these muscles cause the flanking body segments to bend either ventrally or dorsally. This makes it highly unlikely that the muscle cells themselves signal stretch or curvature to each other. What about motor neurons, as these cells have elongated cellular processes that could potentially function as a stretch organ? Not surprisingly, expression of NpHR in all cholinergic neurons (A- and B-type) abolished the bending of the body, while mutations that alter the dorsal B-type motor neurons caused the ventral bias in the bending. Most tellingly, inactivation of B-type motor neurons as opposed to A-type or D-type motor neurons disrupted the correlation between the curvature of the trapped region and more posterior segments, indicating that the worms can no longer sense curvature. Using calcium imaging, they observed a very close correlation between bending and the activation of the B-type motor neurons. The evidence, while largely correlative, clearly points to B-type motor neurons being the cellular substrate for this proprioceptive signal.

A number of questions still remain. Is the proprioceptive signal transferred directly from motor neuron to motor neuron? One way of testing this might be to inactivate or ablate a single B-type motor neuron in the chain and ask if posterior propagation of the signal is disrupted. Another unresolved issue concerns the mechanosensitive machinery that transduces the proprioceptive signal in B-type motor neurons. Mutations in the mechanosensitive channel encoded by *unc-8* do not disrupt proprioceptive coupling, suggesting another channel might serve this purpose. Whether B-type motor neurons express additional mechanosensitive channels is not known. Some uncertainty also exists as to whether other neurons might also contribute to proprioceptive signaling. One potential candidate is the AVB interneuron, a command neuron for forward locomotion whose axon runs the length of the ventral nerve cord and synapses with B-type motor neurons and with AS motor neurons that are also part of the forward locomotion circuit. Finally, what is the role for proprioception in backward locomotion, and if it does play a role, what is the cellular nature of this signal?

Nonetheless, the evidence indicating B-type motor neurons can function both as drivers of forward locomotion while at the same time providing an efferent copy of these actions is striking. It suggests that in an organism with a limited number of neurons, individual neurons need to be able to multitask. It also suggests that the B-type motor neuron itself is able to perform the complex computational task of directly measuring and using proprioceptive information to regulate motor neuron excitability. The challenge ahead is to discover how this computational task is performed by B-type motor neurons and how the proprioceptive signal is then propagated in a directional manner.

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