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Identification of inhibitory premotor interneurons activated at a late phase in a motor cycle during *Drosophila* larval locomotion

Abstract

Muscles, motor neurons, central neurons and peripheral sensory neurons all have distinct important functions in coordinating animal movements. Whereas muscles, motor neurons and sensory neurons have been well investigated, far less is known about the identities and functions of interneurons, which makes it difficult to understand how networks of the cells produce motor outputs. I used *Drosophila* motor circuit underlying larval peristaltic locomotion as a model to identify such interneurons and tried to elucidate the operational principle of the motor circuit. The locomotion is driven by rhythmic waves of muscular contraction that propagate through multiple body segments anteriorly. Here, I identified a class of glutamatergic premotor interneurons called Glutamatergic Vento-Lateral Interneurons (GVLs). I used calcium imaging to search for interneurons that show rhythmic activity and identified GVLs as interneurons showing wave-like activity during peristalsis. Paired GVLs were present in each abdominal segment A1-A7 in the ventral nerve cord and locally extended an axon towards a dorsal neuropile region, where they formed GRASP-positive putative synaptic contacts with motor neurons. The interneurons expressed vesicular glutamate transporter (vGluT) and thus likely secrete glutamate, a neurotransmitter known to inhibit motor neurons in this system. These anatomical results suggest that GVLs are premotor interneurons that locally inhibit motor neurons in the same segment. Consistent with this, optogenetic activation of GVLs with the red-shifted channelrhodopsin, CsChrimson ceased ongoing peristalsis in crawling larvae. Simultaneous calcium imaging of the activity of GVLs and aCCs, one class of motor neurons, showed that GVLs' wave-like activity lagged behind that of aCCs by several segments. Thus, GVLs may inhibit motor neurons locally when the front of a forward motor wave reaches the second or third anterior segment. I propose that GVLs are part of the feedback inhibition system that terminates motor activity once the motor wave proceeds to anterior segments. A possible model for this motor circuit will be discussed based on the results and a recent study on another group of glutamatergic premotor interneurons, PMSIs (period-positive median segmental interneurons).

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