INTRODUCTION

Motor activity is one of the most important outputs an animal's nervous system can produce, as various motor behaviors are of critical importance for survival and reproduction. Examples of such behaviors include moving the body and extremities for spatial translation, orienting sense organs for perception, actuating sound-producing organs for acoustic communication, moving stomatic organs for chewing and swallowing food, cleaning and grooming, or breathing (Hooper & Büschges, 2017; Orlovsky, Deliagina, & Grillner, 1999). Almost all these behaviors are rhythmic. Locomotion, such as walking, crawling, swimming, or flying, is a typical example for rhythmic behavior. Already at the beginning of the twentieth century, it became clear that alternation between activity of the flexor and extensor muscles for leg stepping could be generated in the absence of sensory signals and descending commands.
the cat spinal cord (Brown, 1911). The neural circuits that are capable of generating a rhythmic and patterned output similar to that observed in locomotion, in the absence of external rhythmic input are called central pattern generators (CPGs) Marder & Bucher, 2001). Because of their role in rhythm generation, CPGs are also cited in the literature as central rhythm generators. Based on their neuronal connectivity, CPGs enable nervous systems to reliably and robustly generate patterned motor output of particular frequencies and phases (summaries in: Ayali & Lange, 2010; Grillner, 1975; Marder & Rehm, 2005). Notably, mechanisms of CPG-related pattern generation remain crucial even in systems, in which the functional rhythmic motor output results from highly distributed neural networks acting in concert with central neural circuits, intersegmental coordinating pathways, and external sensory signals (Bidaye, Bockemühl, & Büschges, 2018; Büschges, Scholz, & Manira, 2011; Orlovsky et al., 1999). In such cases, it is the organization and topology of a CPG network that serves the generation of transitions between different phases of activity of a rhythmic motor pattern. In this article, we have selected three forms of locomotion in insects: flying, walking, and crawling (Figure 1). We will use these forms of locomotion to give an overview of the organization of CPG networks and their function.

2 | INSECT FLIGHT

Flight was the first locomotor behavior in insects, for which it was shown that the alternating activity between the muscles serving elevation and depression of the wing was generated by a thoracic neural circuit in the central nervous system. In 1961, Donald Wilson published a highly influential paper (Wilson, 1961), according to which continuous wind stimulation of the sensory hairs on the head of a locust resulted in ongoing rhythmic and alternating activity between elevator and depressor flight muscles in the otherwise deafferented thoracic nervous system. This activity pattern resembled the flight motor output of the intact flying locust, and thereby was the first description of fictive locomotion (Grillner & Zangger, 1979) in insects. The 10-Hz frequency of this fictive locomotor pattern is slower compared to the frequency of 25–30 Hz observed in in vivo flight (Wilson, 1961). Thus, the motor output resembles the pattern expressed by the intact behaving animal only to some extent, as the frequency of the motor activity is reduced by deafferentation and the phase relationships differ. However, the findings in the locust flight system provided clear evidence that the neural basis of rhythmic locomotor activity in insects can arise from the operation of a CPG, which resides in the central nervous system (here the thoracic ganglia), anatomically close to the locomotor organs.

From 1960 onward, neurophysiological research on the locust flight system provided further findings regarding the role of CPGs in motor control in insects. These studies provided insights into: the topological organization of the CPG for locust flight (e.g., Robertson & Pearson, 1983, 1985); the role of afferent feedback in tuning CPG-generated neural activity and transforming it into a functional locomotor output (Pearson, Reye, & Robertson, 1983; Reye & Pearson, 1987; Wendler, 1974; Wolf & Pearson, 1988); the role of neuro-modulation in selection, initiation, and tuning of locomotor networks, including the modulation of intrinsic neuronal properties for the generation of rhythmic locomotor activity (e.g., Ramirez & Pearson, 1991a, 1991b) and the mechanisms underlying reorganization of neural circuit interactions contributing to functional recovery after partial deafferentation (Büs cgbes, Ramirez, Driesang, & Pearson, 1992; Büschges, Ramirez, & Pearson, 1992; Wolf & Büschges, 1997). The following section will introduce the reader to the most relevant findings on the locust flight system with a particular focus on these four aspects (for reviews see also: Burrows, 1996; Hooper & Büschges, 2017; Orlovsky et al., 1999).
2.1 | Topology of locust flight CPG

Keir G. Pearson and collaborators unraveled the neural circuitry of the CPG underlying the generation of flight motor output in the migratory locust, *Locusta migratoria* (e.g., Pearson & Robertson, 1987; Robertson & Pearson, 1983, 1985; Robertson & Reye, 1988). Using paired sharp electrode recordings, they showed that the neural kernels responsible for generating alternating activity between depressor and elevator motor neurons in the locust flight system consist of pairs of interneurons forming specific connections to each other (Figure 2a). Activity in interneuron 301 induces delayed excitation of interneuron 501, which in turn terminates the activity of 301 via an inhibitory synaptic connection (Pearson & Robertson, 1987; Robertson & Pearson, 1983, 1985). Rhythmic activity in this kernel was found to rely on interneuron 301’s ability to generate plateau potentials, resulting in bursting (Ramirez & Pearson, 1991a, 1991b; see below). Overall, five of these kernels were identified in the locust flight CPG. Figure 2b depicts the current knowledge with regard to the specific connectivity between the different classes of thoracic interneurons, that have been identified in each segment as part of the CPG network for flight. Knowledge on the neurotransmitters used for synaptic transmission within the CPG network is still sparse. Some of the inhibitory interactions were found to be GABA mediated (Robertson & Wisniowski, 1988). It is important to note that the neural network forming the locust flight CPG for forewings and hindwings is composed of intersegmental interneurons distributed in both the meso- and metathoracic ganglia. Evidence for this came from lesion experiments, in which meso- and metathoracic ganglia were longitudinally transected. Most operated animals could still fly after the mesothoracic ganglion had been longitudinally lesioned. However, no animal was able to fly following longitudinal lesioning of either the metathoracic ganglion or both the meso- and metathoracic ganglia (Ronacher, Wolf, & Reichert, 1988). This comes in contrast to findings concerning other CPGs, for example, walking CPGs, which as we shall review later, appear to reside within a ganglionic hemisegment (Figure 2b).

2.2 | The role of afferent feedback

Since the 70s it is known that sensory feedback signals from the appendages used for locomotion can shape the centrally generated motor activity toward a functional motor output (Grillner, 1975; Orlovsky et al., 1999; Wendler, 1974). Explicit evidence on how sensory input signals affect CPGs to generate functional motor output was first found by studying the locust flight system (Wolf & Pearson, 1989, for conceptual review see: Pearson, 1987).

In the locust flight system, two sense organs located at the basis of each of the four wings provide sensory feedback to the flight CPG. Those are the wing stretch receptors (e.g., Gettrup, 1962; Pabst, 1965) and the wing tegulae (Fischer, Wolf, & Büschges, 2002; Wolf & Pearson, 1988). Wing stretch receptors signal the upper position of each wing (Altman & Tyrer, 1977; Gettrup, 1962) and wing tegulae signal the downstroke of each wing (Fischer et al., 2002; Neumann, 1985). Interestingly, in the adult locust solely the forewing stretch receptors and the hindwing tegulae affect CPG activity and are sufficient for the generation of a functional activity of elevator and depressor muscles (e.g., Büschges & Pearson, 1991; Pearson & Ramirez, 1990).

Forewing stretch receptors synapse onto individual CPG interneurons or directly onto depressor motor neurons, and establish the in vivo cycle period (Ausborn, Stein, & Wolf, 2007; Pearson & Ramirez, 1990; Reye & Pearson, 1988) and determine the switch from elevator to depressor activity within the wingbeat cycle (Wolf & Pearson, 1988; Figure 2b,c). Similarly, specific connections of hindwing tegulae afferents with CPG interneurons initiate the opposite switch from depressor to elevator activity in the wingbeat cycle and increase wingbeat frequency (Büschges & Pearson, 1991; Fischer et al., 2002; Pearson & Wolf, 1988; Wolf & Pearson, 1988; Figure 2b,c). The role of this sensory feedback in the locust flight system is best exemplified by the influence that hindwing tegulae signals have on the flight motor output generated. In the intact animal, during tethered flying the phase of elevator activation takes place on average at phases 0.39 and 0.49 in the wingbeat cycle, for the fore- and hindwing, respectively, while in the absence of signals from the hindwing tegulae, elevator gets active later at phases 0.49 and 0.61 (Büschges & Pearson, 1991). This late activation of elevator muscles in the flight cycle would not allow for functional flight. The synaptic efficacy of signals from wing sense organs, that is, the wing tegulae, is modulated in the wingbeat cycle, with the highest efficacy being observed around the phase of trough potential in wing elevator motor-neurons. The cyclic change in synaptic efficacy is based on phase-dependent presynaptic modulation of tegula afferents mediated by GABA (Büschges & Wolf, 1999).

Interestingly, sensory control of wingbeat activity in holometabolous insects apparently shares the contribution of the same sense organs: in both *Locusta migratoria*, a holometabolous insect (see above), and *Manduca sexta*, a hemimetabolous insect (Frye, 2001; Möhl, 1985) the forewing stretch receptor has an important role in controlling flight motor activity. In addition, wing campaniform sensilla in both the locust and the moth affect wing beat activity (Ando, Wang, Shirai, Kiguchi, & Kanzaki, 2011; Elson, 1987).

To date, the knowledge about the sensory systems contributing to the generation of functional flight motor output...
FIGURE 2  Organization of the neural machinery underlying the locust flight system. (a) Schematic depiction of a two-cell neural circuit between interneurons 301 and 501 forming the kernel of the locust flight CPG (open triangle—excitatory synapse; filled circle—inhibitory synapse; D—delay). On top of the scheme and below the scheme evidence for synaptic interactions between the interneurons is given (Robertson & Pearson, 1983, 1985) (b) Schematic presentation of the topology of the locust flight CPG distributed in the meso- and metathoracic ganglion. For clarity, only the left side of the network is shown. Black lines and interneurons denote connectivity within the CPG, gray lines and motor neurons denote connections of the CPG interneurons to flight motor neurons (Dep—depressor motoneuron; Elev—elevator motoneuron). Rhythmic alternating activity of Elev (upper trace) and Dep (lower trace) motor neurons is shown exemplarily underneath the network scheme. Please note that in the metathoracic ganglion only one copy of each CPG interneuron is depicted (Robertson & Pearson, 1983). Direct excitatory input from wing sense organs onto CPG interneurons is depicted in red for connections from the hindwing tegula and in green for connections from the forewing stretch receptor. (c) Schematic depiction of the role and interaction of wing hinge sense organs with the locust flight CPG (Wolf & Pearson, 1988; Ramirez & Pearson, 1990). Arrows denote neural influences, open triangles and filled circles denote excitatory and inhibitory synaptic connections. (d) Schematic depiction of the modifications in connectivity between forewing and hindwing tegulae and flight interneurons (schematized with bilateral symmetrical dendritic arbor) in the metathoracic ganglion following unilateral removal of one hindwing tegula (Wolf & Büschges, 1997). Left: intact connectivity between forewing and hindwing tegulae and flight interneurons (each synapse depicts 10% frequency of connection; size of terminals denote relative strength of synaptic efficacy). Right: connectivity between forewing and hindwing tegulae and flight interneurons 2 weeks after surgical removal of one hindwing tegula.
in insects is incomplete. For example, Stevenson (1996) reported on proprioceptors located at the base of the wings, which provide sensory feedback in response to muscle contractions and strongly affect flight motor activity. However, the underlying network mechanisms remain elusive.

2.3 The role of aminergic modulation in selection, initiation, and tuning of flight motor network activity

Activation of locomotor circuits by descending signals from the brain of an animal is mediated by fast synaptic transmission via descending interneurons, and also includes longer term influences due to neuromodulation of downstream circuits (Kim, Su, & Wang, 2017; Nässel & Zandawala, 2019; Pflüger & Büschges, 2004). The adult locust locomotor system can produce locomotor behaviors as diverse as jumping, walking, swimming, or flying. The selection of a specific locomotor behavior, such as flight activation, has been shown to be strongly affected by biogenic amines acting on neural networks that generate the specific motor activity. Octopamine released by dorsal median unpaired (DUM) neurons in the central nervous system (e.g., Bräunig & Pflüger, 2001; Roeder, 2005; Verlinden et al., 2010) has been shown to promote flight activity, but not walking activity (e.g., Rillich, Stevenson, & Pflueger, 2013). This is true even in the immature flight circuits at larval stages. Already at the larval stage, CPG activity for flight can be elicited by octopamine, even though larvae have no functional wings and, therefore, no need of a system for flight activation (Stevenson & Kutsch, 1988). To date, evidence suggests that octopamine serves two functions in the locust flight system: it sets the flight muscle metabolism to anaerobic operation (e.g., Mentel et al., 2003; review in: Pflüger & Duch, 2011) and contributes to the generation of plateau potentials in elevator interneurons of the flight circuitry (Ramirez & Pearson, 1991a, 1991b). The latter function of octopamine contributes to a timely activation and synchronization of elevator activity in the wingbeat cycle.

2.4 Mechanisms of network reorganization and functional recovery after partial deafferentation

Plasticity of neural circuits can be essential for survival as exemplified by neural circuit reorganization after injury. Well-established behavioral examples in insects include network modifications for re-establishing the escape response, after loss of cercal sensory feedback (Chiba & Murphey, 1991; Vardi & Camhi, 1982), and the plasticity in the auditory system implicated in acoustic communication (Huber, 1987; Huber, Kleindienst, Weber, & Thorson, 1984; Schmitz, 1989).

In the locust flight system, plasticity was investigated by experimental elimination of sensory signals generated by the sensory organs used for flight (Büschges & Pearson, 1991; Büschges, Ramirez, Driesang, et al., 1992; Büschges, Ramirez, & Pearson, 1992; Wolf & Büschges, 1997). Removing tegulae of the hindwing or all wings led to an acute cessation of flight ability. However, about 2 weeks post lesion the flight system was able to recover and full flight ability was re-installed (Büschges & Pearson, 1991; Büschges, Ramirez, Driesang, et al., 1992). The underlying modifications in the flight circuitry were studied in particular detail following the loss of the hindwing tegulae. In the intact locust, interneurons of the flight CPG received almost exclusively input from the hindwing tegulae. Within 2 weeks after ablation of the hindwing tegulae, forewing tegulae sensory neurons sprout new collaterals and connect to all flight interneurons relevant for elevator activation in the wingbeat cycle (Büschges, Ramirez, Driesang, et al., 1992; Büschges, Ramirez, & Pearson, 1992). The capacity for reorganization in the locust flight circuitry becomes in particular obvious after experimentally induced ablation of only a single hindwing tegula (Figure 2d): First, afferents of the remaining hindwing tegula sprout toward the contralateral lesioned side; second, dendrites of the flight motor neurons on the lesioned side sprout across the midline, toward the intact side; third, the forewing tegula afferents on the lesioned side connect to flight CPG interneurons, whereas the number and strength of similar connections on the intact side gets reduced. All of these modifications contribute in re-establishing connectivity between wing tegula afferents and the locust flight network, which is essential for proper activation of elevator motor neurons shortly after initiation of the wing downstroke (Wolf & Büschges, 1997), thereby assuring a functional flight motor output. Ample potential for recovery after lesion has been also found in the leg muscle control system of insects (e.g., Page & Matheson, 2009).

3 INSECT WALKING

Walking differs from flying or crawling, because it involves rhythmic intermittent contact of a species-specific number of legs with a solid substrate. Thus, during walking there is strong mechanical coupling between the legs and this has specific consequences for the organization of the underlying CPGs.

Most insects are vigorous walkers. Their legs are pair-wise attached to the three thoracic segments, the pro-, meso-, and metathorax. Each leg consists of five main segments which are, from proximal to distal relative to the thorax, the coxa, the trochanter, the femur, the tibia, and the tarsus.
The three main leg joints contributing to leg stepping in the stick insect and other insects are the thorax–coxa (ThC), the coxa–trochanter (CTr), and the femur–tibia (FTi) joint. Each of the main joints is controlled by sets of antagonistic muscles. The protractor and retractor coxae control leg movement in the horizontal plane, the levator and depressor trochanteris control leg movement in the vertical plane, and the flexor and extensor tibiae are responsible for movement of the tibia away or toward the body. For the purpose of this review the known movability of the ThC-joint orthogonal to its rotational axis will not be considered (Cruse & Bartling, 1995). Flexion of the tarsal segments is generated by the tripartite retractor unguis muscle, whose apodeme extents from the proximal femur through the tibia and along two further tibial muscle parts until the last tarsal segment. Extension is mediated by passive joint forces between the tarsal segments (Radnikow & Bässler, 1991). Elevator and depressor muscles that reside at the distal tibia and the first tarsal segment allow for tarsal elevation and depression (Fischer, Schmidt, Haas, & Büschges, 2001).

### 3.1 Control of stepping in one leg

Stepping of one leg is a rhythmic biphasic behavior consisting of the stance and the swing phase. During the stance phase the leg has ground contact and creates the force needed to move the body into a specific direction. During the swing phase the leg is lifted off the ground and moves back to its starting position to complete the step and initiate the next stance phase. Individual steps consisting of one stance and swing cycle need to be coordinated among legs of an insect to allow for efficient walking in various behavioral conditions. Depending on the walking speed insects express a continuum of interleg coordination patterns ranging from the slow wave gait (or pentapod), where only one leg is in swing phase, while all other legs touch the ground, to the faster tetrapod and tripod coordination patterns, in which two or three legs are in swing at the same time, respectively (Berendes, Zill, Büschges, & Bockemühl, 2016; Graham, 1972; Graham, 1985; Hughes, 1952; Mendes, Bartos, Akay, Márka, & Mann, 2013; Wendler, 1966; Wilson, 1966; Wosnitza, Bockemühl, Dübbert, Scholz, & Büschges, 2013).

Understanding how walking behavior is generated requires deciphering the neural mechanisms of single-leg stepping first. The neural mechanisms controlling single-leg stepping have been thoroughly described for the middle leg of the stick insect (e.g., Büschges, Akay, Gabriel, & Schmidt, 2008). In brief, a number of sensory structures located in and on the insect leg (Büschges & Gruhn, 2008; Tuthill & Wilson, 2016) organize and couple the activity of distinct CPGs for each leg joint (Büschges, Schmitz, & Bässler, 1995), which generate and/or support alternating muscle contraction between antagonistic muscle groups (see Figure 3), ultimately resulting in coordinated movement of the leg during stepping (Bidaye et al., 2018).

### 3.2 The topology and organization of CPGs in insect walking systems

The role of central neural networks capable of generating alternating activity of motor neuron pools that control antagonistic muscles of the insect leg has been known since the 1970s from experiments on partially or completely deafened cockroach and stick insect preparations (Bässler & Wagner, 1983; Pearson, 1972; Pearson & Iles, 1970). When activated by application of the muscarinic acetylcholine receptor agonist pilocarpine on the thoracic ganglia, these central networks are capable of generating rhythmic motor activity in leg motor neuron pools in the absence of phasic
descending or sensory inputs (Büschges et al., 1995; Fuchs, Holmes, Kiemel, & Ayali, 2011; Johnston & Levine, 2002; Ryckebusch & Laurent, 1993). Such experiments have been routinely used to study central neuronal mechanisms of walking pattern generation (Daun-Gruhn & Büschges, 2011).

For the stick insect, it was shown that transection along the ganglionic midline does not impair the pharmacologically induced rhythmicity, implying the existence of hemisegmental CPGs (Büschges et al., 1995). Calcium oscillations accompany the CPG-driven activity in leg motor neuron pools (Baden & Hedwig, 2009; Goldammer, Mantziaris, Büschges, & Schmidt, 2018), indicating the contribution of calcium transients to the rhythmic activity generated in leg motor neurons. In the stick insect deafferented preparation, after pilocarpine application brief patterns of coordinated activity among motor neuron pools supplying the three main leg joints may spontaneously occur (Büschges et al., 1995). These are reminiscent of the transitions occurring during stepping from swing to stance phase and vice versa. However, no ongoing and systematic cycle-to-cycle coupling of the motor neuron pools of the three leg joints can be observed, which would resemble the motor activity in leg stepping. This appears to differ from what Ryckebusch and Laurent (1993) reported in regard of the locust thoracic ganglia that apart from substantial variability, periods of coordinated and “stepping-like” motor neuron pool activity could be observed in the metathoracic ganglion (see also Baudoux, Duch, & Morris, 1998). Studies on the locust and the cockroach have resulted in connectivity models based on autonomous hemisegmental CPGs (David, Holmes, & Ayali, 2016; Ryckebusch & Laurent, 1994). Taken together, the data indicate that there are distinct and largely independent CPG modules in each hemisegment that control the motor neurons and muscles of each of the main leg joints.

The topology and organization of CPGs in the insect thoracic ganglia still remains elusive. At present, the role of individual premotor interneurons in pattern generation has been unraveled only in the stick insect. Three identified nonspiking interneurons (NSI), the NSI E4, NSI I4, and NSI5, can reset the phase of the pharmacologically induced motor neuron activity and affect the frequency of the rhythmic activity in leg motor neurons (Büschges, 1995). Insights from semi-intact preparations support the notion that these interneurons also serve in generating leg movements in the intact stepping animal. Activation of NSI E4 terminates motor activity during stance phase and initiates the swing phase of a middle leg during stepping of a semi-intact preparation (Bässler & Büschges, 1998; Büschges, Kittmann, & Schmitz, 1994). NSI I4 was found to play a role in both searching and walking movements, which show distinct kinematics of the stick insect leg (Berg, Hooper, Schmidt, & Büschges, 2015). NSI I4, whose membrane potential is tonically depolarized and periodically oscillates during walking, is necessary and sufficient to induce searching movements under the condition that the leg has ground contact. Taken together, these results show that CPG interneurons in insect walking contribute to the generation of the motor output for leg stepping in the intact animal and play a role in the generation and execution of various motor behaviors.

These experimental findings have prompted modeling studies aiming to the development of hypotheses on the neural basis of insect walking pattern generation. In a model that successfully reproduced neurophysiological observations in the stick insect (Toth & Daun-Gruhn, 2011), the CPG neurons are of Hodgkin–Huxley type, nonspiking, and are connected to each other via reciprocal inhibitory synapses. Each half of the CPG excites independent interneurons, which also integrate sensory input from the periphery, and make inhibitory synapses with motor neurons. Neurophysiological data on the stick insect walking system showed that extensor and flexor motor neuron bursting is based on descending tonic depolarization shaped by phasic inhibitory input from the respective CPG (Büschges, 1998; Büschges, Ludwar, Bucher, Schmidt, & DiCaprio, 2004; Ludwar, 2005).

In another model based on neurophysiological experiments on the cockroach walking system, a CPG was assumed to asymmetrically excite the interneuronal networks driving the depressor and levator motor neuron pools of a leg (David et al., 2016). More specifically, the time constant for exciting the depressor interneurons is lower and at the same time they receive inhibitory input from the levator interneurons. This allows the depressor motor neurons to be briefly activated before levator interneurons become active and switch them off by premotor inhibition.

Taken together, it is still unclear, whether there is a common neural mechanism among insect walking systems underlying CPG drive of leg motor neurons.

### 3.3 The effect of sensory input on CPG activity

Signals from various leg sensory organs reporting leg movement or load and force applied on the leg have been shown to affect centrally generated motor neuron activity, that is, to reset or entrain segmental CPG activity generated in the presence of the muscarinic agonist pilocarpine (Akay, Haehn, Schmitz, & Büschges, 2004; Akay, Ludwar, Goritz, Schmitz, & Buschges, 2007; Bucher, Akay, DiCaprio, & Buschges, 2003; Hess & Büschges, 1999). For example, movement signals from the FTo-joint reported by the femoral chordotonal organ (fCO) access the CPG of the neighboring CTr-joint. Flexion signals from the fCO initiate a switch from depressor to levator trochanteris activity, whereas extension signals induce the opposite transition. Ongoing flexion and extension stimuli to the fCO can entrain the activity of the
CTr-CPG driving rhythmic activity in trochanteral motoneurons (Bucher et al., 2003; Hess & Büschges, 1999). Similarly, load signals from the leg can entrain the rhythmic motor neuron activity induced by pilocarpine and affect its phase (Akay et al., 2004, 2007). The individual influences of leg sense organs on the activity of the CPGs corresponding to each of the three main leg joints are sufficient to explain the generation of the motor output for leg stepping (Büsschges et al., 2008; Daun-Gruhn & Büschges, 2011).

Furthermore, sensory signals arising from stepping movements of single legs have been found to influence CPG activity in adjacent segments. Two independent studies using semi-intact stick insect and cockroach preparations have reported that neural signals from a stepping front leg can entrain CPG activity in the posteriortly located mesothoracic ganglion controlling movement of the middle leg, a neuronal influence that may serve interleg coordination (Borgmann, Hooper, & Büschges, 2009; Fuchs et al., 2011). In the stick insect, in-phase activity of homologous CPGs was observed in the next caudal segment (Borgmann et al., 2009), while in the cockroach the intersegmental influence appeared to result in motor activity typical for tripod coordination (Fuchs et al., 2011). However, intersegmental influence from the rostral neighboring leg appear to be rather weak, as it can be overridden by local sensory input, for example, from load sensors on the leg (Borgmann et al., 2009). At present it can therefore be concluded that insect walking segmental CPGs are more susceptible to local than to intersegmental sensory signals.

The critical role of sensory input for CPG activity and coordination has also been highlighted in two modeling studies with contradictory conclusions: According to the first study, CPG phase is asymmetrically influenced by direct sensory input on the central descending command to the CPG, thus resulting in a gait transition, through a more general change in phase relationships among all CPGs (Daun-Gruhn & Tóth, 2011). The second study demonstrates an alternative mechanism that acts locally at the level of sensory feedback and modifies a physiological assistance reflex (active reaction) (Szczecinski & Quinn, 2017). This reflex modification entrains the CPG and affects its phase, ultimately resulting in gait transition.

### 3.4 Intersegmental influences between segmental CPGs

In the complete absence of phasic sensory input, preparations of all well-studied insects (i.e., cockroach, locust, stick insect, and hawk moth) resulted in distinct CPG coordination patterns after pharmacological activation of the thoracic networks. Thus, it appears that CPGs centrally interact with each other. Interestingly, results so far have shown that coordination patterns are species-specific. In the cockroach and the hawk moth pilocarpine application and activation of the thoracic motor system results in CPG coordination that resembled the tripod walking pattern (David et al., 2016; Johnston & Levine, 2002), whereas in the stick insect and the locust centrally generated motor patterns rather showed a tendency for in-phase coordination (Knebel, Ayali, Pfüger, & Rillich, 2017; Mantziaris et al., 2017). The latter study in the stick insect argued that CPGs in at least two of the thoracic ganglia are weakly coupled in the absence of phasic afferent and descending input. Cockroaches and moths tend to walk at faster speeds in comparison to locusts and stick insects (Burns, 1973, Full & Tu, 1991, Graham, 1972). Thus, there is a notion that coordination in slow-walking animals may be largely based on sensory input, while central coupling is weak. Conversely, fast-walking insects might rely more strongly on centrally coupled coordination. In line with this hypothesis, running cockroaches show rapid recovery from leg movement perturbations (Couzin-Fuchs, Kiemel, Gal, Ayali, & Holmes, 2015) and the stump of an amputated leg in fruit flies is strongly coordinated with the other walking legs only during fast walking (Berendes et al., 2016).

Taken together, current evidence suggests that CPGs for insect leg stepping interact with each other both centrally and with sensory input contribution. All the above considered, interleg coordination appears to result from a task specifically tuned interaction between central and peripheral influences, whose neural underpinning, however, still remains largely elusive.

### 4 CRAWLING

Crawling is a rhythmic behavior and is the primary form of locomotion in many classes of terrestrial animals. In contrast to other forms of locomotion, crawling animals move the whole body or trunk to locomote across the substrate; crawling can therefore be generally categorized as limbless locomotion.

Crawling behavior in larval insects, involves longitudinal waves of anterograde muscle contractions, also called peristalsis (Tanaka, Ito, Nakagaki, & Kobayashi, 2012), whose neuronal basis can be traced back to CPG activity. Peristaltic locomotion strongly relies on friction between the body and the substrate. It can most prominently be found in vermiform insect larval stages (i.e., limbless maggots), but also plays an important role for locomotion in many caterpillars, which have short true legs and additional prolegs for attachment to the substrate. Peristaltic locomotion in insects has been mainly studied in the caterpillar of the tobacco hawkmoth Manduca sexta (e.g., Griethuisen & Trimmer, 2014; Trimmer & Issberner, 2007) and in the vermiform larva of the fruit fly Drosophila melanogaster (Berrigan & Pepin, 1995; Heckscher, Lockery, & Doe, 2012). The larvae of these
two insect species will be discussed in greater detail in this section; all following references to these species correspond to their larval stages, unless otherwise noted.

4.1 CPGs for crawling in *M. sexta* and *D. melanogaster*

Prior to pupation, the larvae of *M. sexta* exhibit wandering behavior characterized by extended periods of sustained crawling (Dominick & Truman, 1984). The first evidence that the ventral nervous system of these animals contains all the necessary neural machinery for crawling was described in a study by Dominick and Truman (1986). These authors showed that decerebrated individuals of *M. sexta* crawl normally, and do so even more vigorously. Based on their findings, they concluded that the motor pattern responsible for crawling was produced in the segmental ganglia and that pattern generators were probably involved. More conclusive evidence for the existence of an actual CPG was provided in a study that used pilocarpine, a muscarinic acetylcholine receptor agonist (see also previous section on walking), and the deafferented and isolated nervous system of *M. sexta* (Figure 4a, Johnston & Levine, 1996). This study showed that the isolated nervous system in *M. sexta* is able to produce rhythmic segmental motor output that resembles the muscle activity pattern observed during crawling in intact animals, that is, *fictive locomotion* (see above). More specifically, the intrasegmental temporal coordination of motor neuron activity responsible for the movement of an individual segment closely matched the coordination observed in intact animals. Intersegmental coordination in these preparations showed similar anterograde succession of motor activity throughout the body, although at a slower rate compared to intact animals; a general phenomenon observed in many deafferented CPG preparations (see above).

Similar to *M. sexta*, the 3rd instar larval stage of *D. melanogaster* has a wandering phase, during which it crawls around in search for an appropriate location to pupate (Dominick & Truman, 1984). Locomotion in these animals is also based on peristaltic waves of body muscle contraction (Figure 4b); unlike *M. sexta*, however, *Drosophila* larvae have no legs or prolegs. Evidence for the involvement of CPGs in crawling in *D. melanogaster* was presented by studies that used semi-intact preparations. Unlike other insect preparations, reduced *D. melanogaster* preparations often show spontaneous motor rhythmicity without any pharmacological application (Barclay, Atwood, & Robertson, 2002; Jan & Jan, 1976). These findings were complemented by a study that showed that oxotremorine, another muscarinic agonist (see above for pilocarpine), can induce rhythmic motor patterns in decerebrated ventral nervous systems of *D. melanogaster* (Cattaert & Birman, 2001). Further investigation then revealed that coordinated intra- as well as intersegmental motor activity persisted after explicit deafferentation of the complete CNS, suggesting the existence of CPGs in the abdominal and thoracic nerve cord (Fox, Soll, & Wu, 2006; Pulver et al., 2015; Suster & Bate, 2002; Figure 4c); the same could also be shown noninvasively in intact animals (Berni, Pulver, Griffith, & Bate, 2012). Finally, it was shown that a single segment contains the neural circuits sufficient for proper rhythmogenesis (Pulver et al., 2015). As we already mentioned, the frequency of the observed rhythms in these strongly reduced preparations was much lower than in intact
animals. However, the intersegmental phase relationships were retained and resembled those observed in vivo, in contrast to the other deafferented preparations presented in this review.

While in *M. sexta* there is detailed data available on the kinematics and (hydro-)dynamics of crawling, in *Drosophila* the focus has been set on the neuronal composition of the motor system, which has received particular attention in the last years (for comprehensive reviews see also: Clark, Zarin, Carreira-Rosario, & Doe, 2018; Kohsaka, Guertin, & Nose, 2017). Although there is no complete and explicit schematic for the segmental CPG circuit responsible for crawling in *Drosophila* yet, we would like to highlight studies that identify neuron groups likely to either be part of rhythmogenic networks or closely connected to those. Functional intrasegmental coordination during crawling requires the delayed contraction of transversal (T) muscles in relation to longitudinal (L) muscles (Gjorgjieva, Berni, Evers, & Eglen, 2013; Heckscher et al., 2012). Both, excitatory premotor neurons (CIL1 and CIL2, Hasegawa, Truman, & Nose, 2016) and inhibitory premotor neurons (PMSIs and GLVIs) are necessary to establish the centrally generated crawling motor pattern (GVLIs: Itakura et al., 2015; PMSIs: Kohsaka, Takasu, Morimoto, & Nose, 2014). Interestingly, the delay between the two types of muscles is defined by another type of inhibitory premotor neuron, iin1 (Zwart et al., 2016). Silencing this neuron results in cocontraction of the T and L muscles.

### 4.2 Sensory information and intersegmental coordination of crawling

Crawling in *M. sexta* seems to be largely centrally generated. For instance, there is no kinematic difference between vertical and horizontal locomotion in *Manduca*, although gravitational forces acting on the animal in each situation substantially differ (Griethuijsen & Trimmer, 2009). Furthermore, experimental ablation of the longitudinal stretch receptor organ (SRO) in several segments had no discernible effect on the kinematics of *Manduca’s* crawling (Simon & Trimmer, 2009). This is surprising as the SRO’s activity is modulated by contractions of the segment it is located in. These two studies suggest that crawling motor output in *M. sexta* might largely be determined by CPG activity, as the generally slow, stereotypic movement of the animal and its strong mechanical attachment to the substrate allow movement to take place without strong reliance on sensory input (Griethuijsen & Trimmer, 2009). However, more recent investigations using electromyography have revealed that motor output at the level of motor neurons indeed changes depending on the posture of the animal, indicating a role of posture-related sensory input in motor activity (Metallo & Trimmer, 2015; Mukherjee, Vaughn, & Trimmer, 2018). In addition, sensory information can act locally and influence the switch to a different motor program, like burrowing (Dominick & Truman, 1986). Thus, sensory input appears to have an emerging role in shaping crawling CPG activity.

In *D. melanogaster*, our knowledge of the sensory contributions to crawling is more extensive. The general circuit functionality for peristaltic locomotion in *Drosophila* can be established during embryonic development in the absence of sensory information (Suster & Bate, 2002). Once the animal actually starts crawling, however, sensory feedback becomes crucial. For instance, it has been shown that chordotonal organs are important for proper peristalsis (Caldwell, Miller, Wing, Soll, & Eberl, 2003); silencing the chordotonal organs resulted in various coordination defects in crawling, with the overall effect being a reduction in crawling speed. Differential ablation of a large number of different sensory neurons in the peripheral nervous system, consisting of multidendritic (md), external sensory, and chordotonal neurons revealed that they all play a role in crawling. However, the md neurons had the strongest effect on crawling (Hughes & Thomas, 2007). Furthermore, these results suggest that the md neurons inform on the successful contraction of a particular segment, signaling the initiation of its relaxation and the contraction of the next segment. This wave propagation is mediated by the GDL–A27h premotor circuit (Fushiki et al., 2016): when the motor activity in a particular segment is reduced, the A27h neuron in that segment stops activating GABAergic dorsolateral neurons (GDLs), which, in turn, release the next-anterior A27h neurons from their inhibition, thus propagating the peristaltic wave of activity. Therefore, sensory feedback appears to be essential for *Drosophila* larvae crawling.

As outlined here, crawling is an interesting locomotor behavior that, in contrast to flight and walking, does not strongly rely on the action of extremities. Instead it makes use of the anteropropagation of muscle contractions in the animal’s body. Consequently, another defining feature that distinguishes crawling from the two other behaviors discussed in this article is the strong, coordinative coupling between adjacent segments, on which peristalsis is based. However, just like the other two behaviors, crawling at its core utilizes segmental CPGs and it will be interesting to find out how these are situated within and combined with the overarching intersegmental networks that establish the complete behavior.

### 5 Conclusions and future prospects for CPG research

From the above review it becomes clear that central neural networks capable of generating rhythmic motor activity are common neural elements and essentially contribute to locomotion in insects. CPGs can generate a basic patterned motor output, which is then transformed into a functional motor
pattern by sensory input, giving rise to diverse behaviors such as flying, walking, and crawling. Thus, identification of the CPG kernels is of particular importance for understanding how locomotor behavior is generated in insects.

The role of CPGs in each of the three insect locomotor systems reviewed here shows both similarities and differences. (a) Present evidence clearly shows that CPGs contribute to the generation of the motor activity for flying, swimming, and crawling, and that they are capable of generating long-lasting rhythmic motor activity without external phasic input. However, functional locomotor behavior does not purely result from CPG activity. (b) In all three forms of locomotion, the frequency of the motor activity generated after CPG activation in afferented nervous systems is slower than that of the muscle activity patterns recorded in vivo. In any case, the contribution of sensory feedback in controlling frequency of motor activity has been shown. (c) The three systems differ with respect to intersegmental coordination. In the locust flying system, motor activity for fore- and hindwing elevation and depression is generated by a distributed CPG network along the meso- and metathoracic ganglia, thereby establishing a firm coupling in activity between both segments. Similarly, the generation of the crawling pattern is centrally organized by distributed coupled networks along the body segments. In contrast, no such firm and predictable cycle-to-cycle coupling between segmental networks has been found in insect walking, where a particular situation appears to be present with multiple CPGs in each hemisegment of the thoracic ganglia.

These findings are in accordance with the environmental setting and behavioral requirements for each of these three forms of locomotion. Flying of an insect in the air, a rather homogenous environment, and crawling of a caterpillar or a larval insect on a highly predictable substrate give rise to relatively stereotypic behavioral patterns. However, insect walking in a demanding environment with six multisegmented legs results to the generation of a more complex locomotor pattern. Each leg has to generate two different phases of activity, which are linked by nonsteady transitions, that is, variable timing between stance offset and swing onset and vice versa. In addition, for each leg safe operation in both predictable and unpredictable environment needs to be ensured during stepping. Taken all together into consideration, it is quite conceivable that the organization of neural networks is more decentralized for insect walking as compared to flight and crawling. Among the three locomotor systems described here, the flight system in the locust is arguably the most thoroughly described. Its CPG kernels have been identified at the cellular and network level and can now be placed into an extensive connectivity diagram, including coordinate and sensory influences. Walking is well understood with regard to the overall coordinated behavior and sensory feedback contribution. However, although some of the interneurons that participate in walking pattern generation have been identified, we are far from being able to sketch a CPG network consisting of specific neurons. For crawling, network diagrams illustrating the connectivity of identified neurons or neuronal classes have started to emerge. Intersegmental coordination has been thoroughly described in this system, whereas the explicit cellular structure of the CPG kernel is still missing. Taken together, to understand how rhythmic locomotor behavior in insects is generated across locomotor tasks, being it flying, walking or crawling, it is necessary to further identify and characterize the respective neural circuits and interactions among them at the connectivity and cellular level. This will also help us unravel the neural basis of motor flexibility, that is, the known task-dependent flexibility in locomotor pattern generation (e.g., Bidaye et al., 2018; Ritzmann & Büschges, 2007), best exemplified by the modulation in leg kinematics of a walking animal, when changing heading, surpassing obstacles or dealing with unpredictable environments.

Today, electrophysiological approaches on large insects like cockroaches, locusts and stick insects, offering the possibility of simultaneous single cell recordings combined with mechanical stimulation of sensory organs and semi-intact preparations of the locomotor system, can be combined with interesting approaches on genetically accessible organisms, such as the fruit fly. In line with this, around 1,000 transgenic *D. melanogaster* lines have been generated that target specific cell types in the ventral nerve cord, allowing for targeted anatomical and optogenetic studies (Minegishi, Feng, & Dickson, 2019). In addition, the consequent use of modeling will enable us to test, substantiate and define the role of single neurons in walking behavior and pin down the underlying neural networks.

CPGs are common neural elements for locomotion in insects. As we have discussed here, they are crucial for the generation of rhythmic motor output in behaviors as diverse as flight, walking, and crawling. Given the task-specific flexibility of locomotor function to serve the varying behavioral needs, and the reliance of locomotor activity on sensory input across systems, we inevitably need to explore how the animal’s ecology affects nervous system function with respect to generating functional locomotor activity.

**ACKNOWLEDGEMENTS**

We thank S. Seeliger and A. Hagenbruch for technical and administrative support. Research reported in this review was supported by the Alberta Heritage Foundation for Medical Research, DFG grants Bu857, the RTG 1960 funded by the Deutsche Forschungsgemeinschaft—233886668/GRK1960, and the University of Cologne.

**ORCID**

Charalampos Mantziaris [https://orcid.org/0000-0003-2280-1733](https://orcid.org/0000-0003-2280-1733)
REFERENCES

Akay, T., Haehn, S., Schmitz, J., & Büschges, A. (2004). Signals from load sensors underlie interjoint coordination during stepping movements of the stick insect leg. *Journal of Neurophysiology*, 92, 42–51.

Akay, T., Ludwar, B. C., Goritz, M. L., Schmitz, J., & Buschges, A. (2007). Segment specificity of load signal processing depends on walking direction in the stick insect leg muscle control system. *The Journal of Neuroscience*, 27, 3285–3294.

Altman, J. S., & Tyrer, N. M. (1977). The locust wing hinge stretch receptors. I. Primary sensory neurones with enormous central arborizations. *Journal of Comparative Neurology*, 172, 409–430.

Ando, N., Wang, H., Shirai, K., Kiguchi, K., & Kanzaki, R. (2011). Inhibitory synaptic drive patterns motoneuronal activity in rhythmic preparations of isolated thoracic ganglia in the stick insect. *Brain Research*, 1245–1255.

Borgmann, A., Hooper, S. L., & Büschges, A. (2009). Sensory feedback induced by front-leg stepping entrains the activity of central pattern generators in caudal segments of the stick insect walking system. *Journal of Neuroscience*, 29, 2972–2983.

Buchter, D., Akay, T., DiCaprio, R. A., & Buschges, A. (2003). Interjoint coordination in the stick insect legcontrol system: The role of positional signaling. *Journal of Neurophysiology*, 89, 1245–1255.

Büschges, A. (1995). Role of local nonspiking interneurons in the generation of rhythmic motor activity in the stick insect. *Journal of Neurobiology*, 27, 488–512.

Büschges, A. (1998). Inhibitory synaptic drive patterns motoneuronal activity in rhythmic preparations of isolated thoracic ganglia in the stick insect. *Brain Research*, 783, 262–271.

Büschges, A., Akay, T., Gabriel, J. P., & Schmidt, J. (2008). Organizing network action for locomotion: Insights from studying insect walking. *Brain Research Reviews*, 57, 162–171.

Büschges, A., & Gruhn, M. (2008). Mechanosensory feedback in walking: From joint control to locomotor patterns. *Advances in Insect Physiology*, 34, 193–230.

Büschges, A., & Pearson, K. G. (1991). Adaptive modifications in the flight system of the locust after the removal of wing proprioceptors. *Journal of Experimental Biology*, 157, 313–333.

Büschges, A., Ramirez, J.-M., Driesang, R., & Pearson, K. G. (1992). Connections of the foreleg tegulae in the locust flight system and their modification following partial deafferentation. *Journal of Neurobiology*, 23, 44–60.

Büschges, A., Ramirez, J.-M., & Pearson, K. G. (1992). Reorganization of sensory regulation of locust flight after partial deafferentation. *Journal of Neurobiology*, 23, 31–43.

Büschges, A., & Wolf, H. (1999). Phase-dependent presynaptic modulation of mechanosensory signals in the locust flight system. *Journal of Neurophysiology*, 81, 959–962.

Büschges, A., & Wolf, H. (1999). Phase-dependent presynaptic modulation of mechanosensory signals in the locust flight system. *Journal of Neurophysiology*, 81, 959–962.

Caldwell, J. C., Miller, M. M., Wing, S., Soll, D. R., & Eberl, D. F. (2003). Dynamic analysis of larval locomotion in *Drosophila* chordotonal organ mutants. *Proceedings of the National Academy of Sciences of the USA*, 100, 16053–16058.

Cattaert, D., & Birman, S. (2001). Blockade of the central generator of locomotor rhythm by noncompetitive NMDA receptor antagonists in *Drosophila* larvae. *Journal of Neurobiology*, 48, 58–73.
Chiba, A., & Murphey, R. K. (1991). Connectivity of identified central synapses in the cricket is normal following regeneration and blockade of presynaptic activity. *Journal of Neurobiology*, 22, 130–142.

Clark, M. Q., Zarin, A. A., Carreia-Rosario, A., & Doe, C. Q. (2018). Neural circuits driving larval locomotion in *Drosophila*. *Neural Development*, 13, 6.

Cruse, H., & Bartling, C. (1995). Movement of joint angles in the legs of a walking insect, Carausius morosus. *Journal of Insect Physiology*, 41(9), 761–771.

Couzin-Fuchs, E., Kiemel, T., Gal, O., Ayali, A., & Holmes, P. (2015). Intersegmental coupling and recovery from perturbations in freely running cockroaches. *The Journal of Experimental Biology*, 218, 285–297.

Daun-Gruhn, S., & Büschges, A. (2011). From neuron to behavior: Dynamic equation-based prediction of biological processes in motor control. *Biological Cybernetics*, 105, 71–88.

Daun-Gruhn, S., & Tóth, T. I. (2011). An inter-segmental network model and its use in elucidating gait-switches in the stick insect. *Journal of Computational Neuroscience*, 31, 43–60.

David, I., Holmes, P., & Ayali, A. (2016). Endogenous rhythm and pattern-generating circuit interactions in cockroach motor centres. *Biology Open*, 5, 1229–1240.

Dominick, O. S., & Truman, J. W. (1984). The physiology of wandering behaviour in *Manduca sexta*. I. Temporal organization and the influence of the internal and external environments. *Journal of Experimental Biology*, 110, 35–51.

Dominick, O. S., & Truman, J. W. (1986). The physiology of wandering behaviour in *Manduca sexta*. III. Organization of wandering behaviour in the larval nervous system. *Journal of Experimental Biology*, 121, 115–132.

Elson, R. C. (1987). Interneuronal processing of inputs from the campaniform sensilla of the locust hindwing. *Journal of Comparative Physiology*, 161, 761–776. https://doi.org/10.1007/BF00605017

Fischer, H., Schmidt, J., Haas, R., & Büschges, A. (2001). Pattern generation for walking and searching movements of a stick insect leg. I. Coordination of motor activity. *Journal of neurophysiology*, 85, 341–353. https://doi.org/10.1152/jn.2001.85.1.341

Fischer, H., Wolf, H., & Büschges, A. (2002). The locust tegula: Kinematic parameters and activity pattern during the wing stroke. *Journal of Experimental Biology*, 205, 1531–1545.

Fox, L. E., Soll, D. R., & Wu, C.-F. (2006). Coordination and modulation of locomotion pattern generators in *Drosophila* larvae: Effects of altered biogenic amine levels by the tyramine β hydroxylase mutation. *Journal of Neuroscience*, 26, 1486–1498.

Frye, M. A. (2001). Effects of stretch receptor ablation on the optomotor control of lift in the hawkmoth *Manduca sexta*. *The Journal of Experimental Biology*, 204, 3683–3691.

Fuchs, E., Holmes, P., Kiemel, T., & Ayali, A. (2011). Intersegmental coordination of cockroach locomotion: Adaptive control of centrally coupled pattern generator circuits. *Frontiers in Neural Circuits*, 4, 125.

Full, R. J., & Tu, M. S. (1991). Mechanics of a rapid running insect: Two-, four- and six-legged locomotion. *Journal of Experimental Biology*, 156, 215–231.

Fushiki, A., Zwart, M. F., Kohsaka, H., Fetter, R. D., Cardona, A., & Nose, A. (2016). A circuit mechanism for the propagation of waves of muscle contraction in *Drosophila*. *eLife*, 5, e13253.

Gettrup, E. (1962). Thoracic proprioceptors in the flight system of locusts. *Nature*, 193, 498–499.

Gjorgjieva, J., Berni, J., Evers, J. F., & Eglen, S. J. (2013). Neural circuits for peristaltic wave propagation in crawling *Drosophila* larvae: analysis and modeling. *Frontiers in Computational Neuroscience*, 7, 24.

Goldammer, J., Mantziaris, C., Büschges, A., & Schmidt, J. (2018). Calcium imaging of CPG-evoked activity in efferent neurons of the stick insect. *PLoS ONE*, 13, e0202822.

Graham, D. (1972). A behavioural analysis of the temporal organisation of walking movements in the 1st instar and adult stick insect *Carausius morosus*. *Journal of Comparative Physiology*, 81, 23–52.

Graham, D. (1985). Influence of coxa-thorax joint receptors on retrac- tor motor output during walking in *Carausius morosus*. *Journal of Experimental Biology*, 114, 131–139.

Grillner, S. (1975). Locomotion in vertebrates: Central mechanisms and reflex interaction. *Physiological Reviews*, 55, 247–304.

Grillner, S., & Zanger, P. (1979). On the central generation of locomotion in the low spinal cat. *Experimental Brain Research*, 34, 241–261.

Hasegawa, E., Truman, J. W., & Nose, A. (2016). Identification of excitatory premotor interneurons which regulate local muscle contraction during *Drosophila* larval locomotion. *Scientific Reports*, 6, 1–13.

Hecksher, E. S., Lockery, S. R., & Doe, C. Q. (2012). Characterization of *Drosophila* larval crawling at the level of organism, segment, and somatic body wall musculature. *Journal of Neuroscience*, 32, 12460–12471.

Hess, D., & Büschges, A. (1999). Role of proprioceptive signals from an insect femur-tibia joint in patterning motoneuronal activity of an adjacent leg joint. *Journal of Neurophysiology*, 81, 1856–1865.

Hooper, S. L., & Büschges, A. (eds.). (2017). *Neurobiology of motor control*. Hoboken, NJ, USA: John Wiley & Sons, Inc.

Huber, F. (1987). Plasticity in the auditory system of crickets: Phonotaxis with one ear and neuronal reorganization within the auditory pathway. *Journal of Comparative Physiology*, 161, 583–604.

Huber, F., Kleinidienst, H.-U., Weber, T., & Thorson, J. (1984). Auditory behavior of the cricket. *Journal of Comparative Physiology*, 155, 725–738.

Hughes, C. L., & Thomas, J. B. (2007). A sensory feedback circuit coordinates muscle activity in *Drosophila*. *Molecular and Cellular Neuroscience*, 35, 383–396.

Hughes, G. M. (1952). The co-ordination of insect movements. I. The walking movements of insects. *Journal of Experimental Biology*, 29, 267–285.

Iitaka, Y., Kohsaka, H., Ohyama, T., Zlatic, M., Pulver, S. R., & Nose, A. (2015). Identification of inhibitory premotor interneurons activated at a late phase in a motor cycle during *Drosophila* larval locomotion. *PLoS ONE*, 10, e0136600.

Jan, L. Y., & Jan, Y. N. (1976). Properties of the larval neuromuscular junction in *Drosophila* melanogaster. *Journal of Physiology*, 262, 189–214.

Johnston, R. M., & Levine, R. B. (1996). Crawling motor patterns induced by pilocarpine in isolated larval nerve cords of *Manduca sexta*. *Journal of Neurophysiology*, 76, 3178–3195.

Johnston, R. M., & Levine, R. B. (2002). Thoracic leg motoneurons in the isolated CNS of adult *Manduca* produce patterned activity in response to pilocarpine, which is distinct from that produced in larvae. *Invertebrate Neuroscience*, 4, 175–192.

Kim, S. M., Su, C.-Y., & Wang, J. W. (2017). Neuromodulation of innate behaviors in *Drosophila*. *Annual Review of Neuroscience*, 40, 327–348.
Knebel, D., Ayali, A., Pflüger, H.-J., & Rillich, J. (2017). Rigidity and flexibility: The central basis of inter-leg coordination in the locust. *Frontiers in Neural Circuits*, 10, 112.

Kohsaka, H., Guerin, P. A., & Nose, A. (2017). Neural circuits underlying fly larval locomotion. *Current Pharmaceutical Design*, 23(12), 1722–1733.

Kohsaka, H., Takasu, E., Morimoto, T., & Nose, A. (2014). A group of segmental premotor interneurons regulates the speed of axial locomotion in *Drosophila* larvae. *Current Biology*, 24, 2632–2642.

Ludwar, B. C. (2005). Modulation of membrane potential in mesothoracic moto- and interneurons during stick insect front-leg walking. *Journal of Neurophysiology*, 94, 2772–2784.

Mantziaris, C., Bockemühl, T., Holmes, P., Borgmann, A., Daun, S., Kohsaka, H., Takasu, E., Morimoto, T., & Nose, A. (2014). A group of the stick insect. *Journal of Neurophysiology*, 118(4), 2296–2310. https://doi.org/10.1152/jn.00321.2017.

Marder, E., & Bucher, D. (2001). Central pattern generators and the control of rhythmic movements. *Current Biology*, 11, R986–R996.

Marder, E., & Rehm, K. J. (2005). Development of central pattern generating circuits. *Current Opinion in Neurobiology*, 15, 86–93.

Mendes, C. S., Bartos, I., Akay, T., Márka, S., & Mann, R. S. (2013). Quantification of gait parameters in freely walking wild type and sensory deprived *Drosophila* melanogaster. *eLife*, 2, e00231.

Mentel, T., Duch, C., Stypa, H., Wegener, G., Müller, U., & Pflüger, H.-J. (2003). Central modulatory neurons control fuel selection in flight muscle of migratory locust. *Journal of Neuroscience*, 23, 1109–1113.

Metallo, C., & Trimmer, B. A. (2015). Orientation-dependent changes in single motor neuron activity during adaptive soft-bodied locomotion. *BBE*, 85, 47–62.

Minegishi, R., Feng, K., & Dickson, B. (2019). Cellular level analysis of the locomotor neural circuits in *Drosophila* melanogaster. In *Living Machines*, 334–337, Nara, Japan, 9-12 July 2019.

Möhl, B. (1985). The role of proprioception in locust flight control. II. Information signalled by forewing stretch receptors. *Journal of Comparative Physiology A*, 156, 103–116.

Mukherjee, R., Vaughn, S., & Trimmer, B. A. (2018). The neuromechanics of proleg grip release. *Journal of Experimental Biology*, 221, jeb173856.

Nässel, D. R., & Zandavala, M. (2019). Recent advances in neuropeptide signaling in *Drosophila*, from genes to physiology and behavior. *Progress in Neurobiology*, 179, 101607.

Neumann, L. (1985). Experiments on tegula function for flight coordination in the locust. In M. Gewecke & G. Wendler (Eds.), *Control of locomotion: From Mollusc to Man*. (pp. 167–185). Boston, MA: Springer.

Pearson, K. G., & Iles, J. F. (1970). Discharge patterns of coxal levator and depressor motoneurones of the cockroach, *Periplaneta americana*. *The Journal of Experimental Biology*, 52, 139–165.

Pearson, K. G., & Ramirez, J. M. (1990). Influence of input from the forewing stretch receptors on motoneurones in flying locusts. *Journal of Experimental Biology*, 151, 317–340.

Pearson, K. G., Reye, D. N., & Robertson, R. M. (1983). Phase-dependent influences of wing stretch receptors on flight rhythm in the locust. *Journal of Neurophysiology*, 49, 1168–1181.

Pearson, K. G., & Robertson, R. M. (1987). Structure predicts synaptic function of two classes of interneurons in the thoracic ganglia of *Locusta migratoria*. *Cell and Tissue Research*, 250, 105–114.

Pearson, K. G., & Wolf, H. (1988). Connections of hindwing tegulae with flight neurones in the locust, *Locusta migratoria*. *Journal of Experimental Biology*, 135, 381–409.

Pflüger, H. J., & Büsschges, A. (2004). Neuronal control of microcircuits in motor systems: Invertebrates. In *Report of the 93rd Dahlem workshop on microcircuits: The interface between neurons and global brain function*. Berlin.

Pflüger, H.-J., & Duch, C. (2011). Dynamic neural control of insect muscle metabolism related to motor behavior. *Physiology*, 26, 293–303.

Pulver, S. R., Bayley, T. G., Taylor, A. L., Berni, J., Bate, M., & Hedwig, B. (2015). Imaging fictive locomotor patterns in larval *Drosophila*. *Journal of Neurophysiology*, 114, 2564–2577.

Radnikow, G., & Bässler, U. (1991). Function of a muscle whose apodeme travels through a joint moved by other muscles: Why the retractor unguis muscle in stick insects is tripartite and has no antagonist. *The Journal of Experimental Biology*, 157, 87–99.

Ramirez, J.-M., & Pearson, K. G. (1991a). Octopamine induces bursting and plateau potentials in insect neurones. *Brain Research*, 549, 332–337.

Ramirez, J. M., & Pearson, K. G. (1991b). Octopaminergic modulation of interneurons in the flight system of the locust. *Journal of Neurophysiology*, 66, 1522–1537.

Reye, D. N., & Pearson, K. G. (1987). Projections of the wing stretch receptors to central flight neurones in the locust. *Journal of Neurophysiology*, 7, 2476–2487.

Reye, D. N., & Pearson, K. G. (1988). Entrainment of the locust central flight oscillator by wing stretch receptor stimulation. *Journal of Comparative Physiology*, 162, 77–89.

Rillich, J., Stevenson, P. A., & Pflueger, H. J. (2013). Flight and walking in locusts: cholinergic co-activation, temporal coupling and its modulation by biogenic amines. *PLoS ONE*, 8, e62899.

Ritzmann, R. E., & Büschges, A. (2007). Adaptive motor behavior in insects. *Current Opinion in Neurobiology*, 17, 629–636.

Robertson, R. M., & Pearson, K. G. (1983). Interneurons in the flight system of the locust: Distribution, connections, and resetting properties. *Journal of Comparative Neurology*, 215, 33–50.

Robertson, R. M., & Pearson, K. G. (1985). Neural circuits in the flight system of the locust. *Journal of Neurophysiology*, 53, 110–128.

Robertson, R. M., & Reye, D. N. (1988). A local circuit interaction in the flight system of the locust. *Journal of Neuroscience*, 8, 3929–3936.

Robertson, R. M., & Wisniowski, L. (1988). GABA-like immunoreactivity of identified interneurons in the flight system of the locust, *Locusta migratoria*. *Cell and Tissue Research*, 254, 331–340. https://doi.org/10.1007/BF00225805

Roeder, T. (2005). Tyramine and octopamine: Ruling behavior and metabolism. *Annual Review of Entomology*, 50, 447–477.
van Griethuijsen, L. I., & Trimmer, B. A. (2014). Locomotion in caterpillars. *Biological Reviews*, 89, 656–670.

Vardi, N., & Camhi, J. M. (1982). Functional recovery from lesions in the escape system of the cockroach. *Journal of Comparative Physiology*, 146, 299–309.

Verlinden, H., Vleugels, R., Marchal, E., Badisco, L., Pflüger, H.-J., Blenau, W., & Broeck, J. V. (2010). The role of octopamine in locusts and other arthropods. *Journal of Insect Physiology*, 56, 854–867.

Wendler, G. (1974). The influence of proprioceptive feedback on Locust flight co-ordination. *Journal of Comparative Physiology*, 88, 173–200. https://doi.org/10.1007/BF00695406

Wendler, G. (1966). The co-ordination of walking movements in arthropods. *Symposia of the Society for Experimental Biology*, 20, 229–249.

Wilson, D. M. (1961). The central nervous control of flight in a locust. *Journal of Experimental Biology*, 38, 471–490.

Wilson, D. M. (1966). Insect walking. *Annual Review of Entomology*, 11, 103–122.

Wolf, H., & Büschges, A. (1997). Plasticity of synaptic connections in sensory-motor pathways of the adult locust flight system. *Journal of Neurophysiology*, 78, 1276–1284.

Wolf, H., & Pearson, K. G. (1988). Proprioceptive input patterns elevator activity in the locust flight system. *Journal of Neurophysiology*, 59, 1831–1853.

Wolf, H., & Pearson, K. G. (1989). Comparison of motor patterns in the intact and deafferented flight system of the locust. *Journal of Comparative Physiology*, 165, 61–74.

Wosnitza, A., Bockemühl, T., Dübbert, M., Scholz, H., & Büschges, A. (2013). Inter-leg coordination in the control of walking speed in Drosophila. *Journal of Experimental Biology*, 216, 480–491.

Zwart, M. F., Pulver, S. R., Truman, J. W., Fushiki, A., Fetter, R. D., Cardona, A., & Landgraf, M. (2016). Selective inhibition mediates the sequential recruitment of motor pools. *Neuron*, 91, 615–628.

---

**How to cite this article:** Mantziaris C, Bockemühl T, Büschges A. Central pattern generating networks in insect locomotion. *Develop Neurobiol*. 2020;80:16–30. [https://doi.org/10.1002/dneu.22738](https://doi.org/10.1002/dneu.22738)