Gating of steering signals through phasic modulation of reticulospinal neurons during locomotion

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The neural control of movements in vertebrates is based on a set of modules, like the central pattern generator networks (CPGs) in the spinal cord coordinating locomotion. Sensory feedback is not required for the CPGs to generate the appropriate motor pattern and neither a detailed control from higher brain centers. Reticulospinal neurons in the brainstem activate the locomotor network, and the same neurons also convey signals from higher brain regions, such as turning/steering commands from the optic tectum (superior colliculus). A tonic increase in the background excitatory drive of the reticulospinal neurons would be sufficient to produce coordinated locomotor activity. However, in both vertebrates and invertebrates, descending systems are in addition phasically modulated because of feedback from the ongoing CPG activity. We use the lamprey as a model for investigating the role of this phasic modulation of the reticulospinal activity, because the brainstem–spinal cord networks are known down to the cellular level in this phylogenetically oldest extant vertebrate. We describe how the phasic modulation of reticulospinal activity from the spinal CPG ensures reliable steering/turning commands without the need for a very precise timing of on- or offset, by using a biophysically detailed large-scale (19,600 model neurons and 646,800 synapses) computational model of the lamprey brainstem–spinal cord network. To verify that the simulated neural network can control body movements, including turning, the spinal activity is fed to a mechanical model of lamprey swimming. The simulations also predict that, in contrast to reticulospinal neurons, tectal steering/turning command neurons should have minimal frequency adaptive properties, which has been confirmed experimentally.

large-scale modeling | compartmental modelling | full-scale model | MLR

In many vertebrate and invertebrate motor systems, a phasic modulation occurs in the descending control system determining the level of activity (1–3) during rhythmic movements. The physiological role of this modulation has remained enigmatic, because it has been shown that tonic activity is sufficient to effectively drive motor activity like locomotion. One example is the reticulospinal neurons in the brainstem that serve as the major interface between higher level commands and the networks in the spinal cord in all vertebrates from lamprey to primates (4–6). In this study, we investigate the motor system of the lamprey, belonging to the most ancient group of vertebrates that has been investigated in considerable detail not only at the brainstem–spinal cord level but also with regard to the forebrain systems underlying the control of action (2, 7, 8). A bilateral symmetric activation of reticulospinal neurons will activate the locomotor networks in the spinal cord, resulting in coordinated swimming movements (2, 9–11). The reticulospinal neurons act on the excitatory and inhibitory network interneurons in the spinal cord through NMDA and AMPA receptors (12). Most reticulospinal neurons can be involved in several motor patterns (13). Whereas a bilaterally symmetric activation leads to locomotion, a unilateral addition of excitation to one side will enhance motor activity on this side and result in turning. This is the basis of steering during locomotion (14).

The phasic modulation of reticulospinal neurons is most pronounced in the fastest-conducting group involved in steering (15). It results from feedback from the network neurons in the rostral segments of the spinal cord during locomotor movements, so that the reticulospinal neurons become active in phase with those segments, and during the inactive period they are instead inhibited (15–19). This feedback is conveyed to reticulospinal neurons via ascending spinobulbar neurons (15–20) that provide an "efference copy" regarding the cycle-to-cycle activity in the locomotor network. Spinobulbar neurons (21) provide the excitatory and inhibitory drive to reticulospinal neurons, resulting in modulation of their activity in phase with the ipsilateral rostral parts of the spinal cord (21, 22) and this forms a closed spino-reticulo-spinal loop (17).

Visuomotor coordination (23) and steering results from activation of tectal output neurons that monosynaptically activate reticulospinal neurons (24, 25), which represent the interface between tectum and the spinal cord networks. Here, we focus on the role of the phasic modulation of the reticulospinal neurons. We show that the phasic modulation of the reticulospinal cells is advantageous in that the steering commands from tectum become gated and thereby arrive in the correct phase of the locomotor cycle. The tectal commands, therefore, need not be timed very precisely in relation to the locomotor cycle. The reticulospinal modulation ascertains that the command signal will be accurately timed provided that the tectal command signal itself remains constant and thus has a limited spike-frequency adaptation, which indeed applies to the tectal output neurons as shown here experimentally (25). We explore the effect of steering signals through a combined simulation-experimental

Significance

In many vertebrate and invertebrate species descending motor commands are phasically modulated in synchrony with rhythmic movements. The physiological role of this modulation has remained enigmatic. We report here, from the lamprey locomotor system, that steering signals from for instance tectum can be gated by a downstream locomotor-related modulation of the reticulospinal command neurons. Such gating will ascertain that the steering commands will be transmitted in the appropriate phase of a swimming cycle and be suppressed out-of-phase. Another consequence of this mechanism is the relative independence of the motor response on the timing of the steering signal.

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approach. Biologically detailed lamprey spinal cell models (26) are used in large-scale simulations of the locomotor network (7) to replicate electric activity in command centers and the spinal cord. A mechanical model of swimming (27, 28) is used for a quantitative evaluation of the locomotor response.

Results

In a previous full-scale model of the lamprey spinal locomotor system (7), we used biophysically realistic models of each type of neuron [compartmental model (26)] with a realistic number of cells. Using this network model as a basis (*Materials and Methods*), we now introduce phasic modulation of the reticulospinal neurons (from the spinal locomotor network) and explore the complementary effects that this addition introduces, in particular in relation to the input from the optic tectum related to steering. The compartmental cell model is shown in Fig. 1*A*, and the network scheme in Fig. 1*B* is in detail based on the biological findings (2). In addition to extending the network model, we also add a mechanical model (29) directly controlled by the neural network. Using such a combined neuromechanical system provides critical information on how the neural system accomplishes the steering control (Fig. 1*C* and *Materials and Methods*).

The locomotor network is activated from the mesencephalic locomotor region (MLR) and through bilateral activation of the reticulospinal neurons that activate the spinal central pattern generator network (CPG) (shaded area in Fig. 1*B*). The most common locomotor pattern is a wave of alternating left–right neuronal firing, which travels along the spinal cord from head to tail with a constant intersegmental phase lag and a characteristic wavelength of approximately one body length (Fig. 1).

Fig. 2*A* shows a spike raster plot of neurons in MLR that drive the network activity and are tonically active, whereas the reticulospinal cells on the left and right side are modulated in phase

with the rostral population of excitatory and inhibitory interneurons on either side of the spinal cord. The modulation of the reticulospinal neurons is attributable to the feedback projections from the spinal cord, as shown in Fig. 1*B*. Note the delay from head to tail of the activity of the spinal excitatory interneurons that in turn activate the segmental motoneurons (E and M in Fig. 1*B*).

In the neuromechanical simulation (Fig. 2*B*), the activity of motoneurons is integrated to continuous signals using leaky integrators (10 segmental motor outputs from head to tail are used) and directed further to the "muscles" of the segmental mechanical model (*Materials and Methods*). Waves of the left–right alternating electrical activity is translated to coordinated lateral movements of the body, and it makes the model lamprey swim in simulated water. When a symmetric activation of the reticulospinal system is provided (Fig. 1*B*), forward swimming will result, as in Fig. 2*C*.

Steering in the model is achieved via a recruitment of neurons in the left (TL) or right (TR) parts of the tectum, which project contralaterally to the right and left reticulospinal neurons, respectively (Fig. 1*B*). The turning command is simulated as a depolarizing pulse with fixed amplitude and duration applied to TL or TR neurons. This results in a temporary increase of the firing rate of the corresponding reticulospinal population (bottom trace in Fig. 2*B*) and a lateral turn, as in Fig. 2*D*.

Although the effect of a turning command is reflected in perturbations of the firing rates of the spinal interneurons and related motor outputs, these changes are distributed among multiple signals and may also be very subtle (see, for example, responses to a 150-ms-long turn command in Fig. 2*B*), and thus the integrated role of the activity changes is difficult to estimate quantitatively. Because of this, it is important to use a mechanical model to allow a direct observation and measurement of the

Fig. 1. Neuromechanical simulation of planar swimming in lamprey. (*A*) Biological neuron model consists of 16 cylindric electrical compartments with multiple ionic channel subtypes. Synaptically activated channels are placed on the reticulospinal (RS) cell soma and dendrites; inhibitory (glycinergic) synapses are shown in blue, excitatory (mixed AMPA/NMDA glutamatergic) synapses are shown in red. (*B*) Spinal and supraspinal locomotor networks are built of populations of neurons. Excitatory and inhibitory synaptic projections are shown in red and blue, respectively. Tectal neurons from the left and right sides (TL and TR), as well as symmetrically projecting neurons from the mesencephalic locomotor region (MLR), activate RS neurons on both sides. RS neurons provide excitation to locomotor circuits in the spinal cord, left and right sides shown in gray, constructed of locally projecting excitatory cells (E), inhibitory cells (I), and motoneurons (M). Local projections have asymmetric rostrocaudal extension with longer caudal projections, as shown by white rectangles. Firing activity in model motoneurons is integrated and directed to 10 pairs of motor outputs. Information about locomotor activity in the rostral part of the spinal cord is fed back to RS populations (large circles) via excitatory and inhibitory afferents (red and blue lines). (*C*) Segmental model of the lamprey body consists of 11 cylindric links (skin and muscles are not shown). For illustration purposes, variable thickness of the links reflects stiffness of the body decaying toward the tail. Activation of a muscle at joint i is simulated as two opposite torques applied to the adjacent links. (*D*) Activity of model neural populations in the brainstem and rostral part of the spinal cord during swimming. MLR neurons fire tonically. Half of RS cells are tonically active (solid lines), and another 50% are modulated (dashed lines) in phase with ipsilateral activity in the left and right sides of the rostral spinal cord (integral firing rate of M populations from 10 most rostral spinal segments are shown for the left and right sides, respectively).

Fig. 2. Electrical and mechanical dynamics in the model. (*A*) Initiation of spiking activity in the neural populations. Spikes in reticulospinal (RS) cells and spinal interneurons (INs) (i.e., E and I cells) from the left and right sides of the body are shown below and above the spikes in MLR (*Middle*) and ordered from head to tail according to arrows. (*B*) Phasic and tonic components of RS and motor outputs from the right side. An output signal of neuron population is the firing frequency computed by leaky integrator with time constant equal to the cell membrane time constant. Timing of the turn command is shown with horizontal bar. (*C* and *D*) Shape of the body during forward swimming and a lateral turn caused by dc stimulation of TL (0.5 nA, 150 ms). (*E*) Turn is characterized by turn phase and turn angle. The turn phase is defined as delay relative to the last unperturbed cycle of RS modulation on the turning side (in radians). Turn angle is an instantaneous change of the body direction measured one cycle before and one cycle after the turn command (in degrees). Solid black circles (two dots before and two dots after the turn) show position of the head at the same phase of locomotion and define direction of swimming.

resulting motor responses to different steering commands. The magnitude of the response to a turning command is defined as the turning angle measured between swim directions one cycle before and one cycle after the turning cycles (during which the turn command is applied), as shown in Fig. 2*E*. In the present study, the magnitude of the turning response is analyzed as a function of cellular and network properties (spike-frequency adaptation in tectal neurons and feedback modulation of reticulospinal neurons) and parameters of the turning command: phase, duration, and amplitude.

Role of Spike-Frequency Regulation in the Tectal Turning Command.
The turning command from tectum is conveyed via the reticulospinal neurons, which are in turn modulated in phase with the rostral spinal segments on the same side (Fig. 2*A*). The tectal

command would need to arrive during the active phase of reticulospinal neurons to be effective, because a brief signal applied in the phase dominated by inhibition would not change their firing significantly. However, if the tectal command signal lasts during a full cycle this would ascertain that it covers the phase during which the reticulospinal neurons are active. There would be one further requirement; the command signal from tectum would need to be constant throughout this period. This provides the possibility of a simple command, which would give the same effect irrespectively of whether it affects one cycle or two parts of consecutive cycles of reticulospinal activity.

In lamprey tectal neurons, there is merely a weak adaptation. Fig. 3*A* shows the firing response of a tectal output neuron to long current pulses and the corresponding model neurons. Spike-frequency adaptation is much less pronounced than in lamprey motoneurons or interneurons (30, 31). Also in frogs or *Xenopus* tadpoles (32, 33), tectal output neurons show little or no spike-frequency adaptation.

In contrast, in lamprey reticulospinal and spinal neurons, spike-frequency adaptation is prominent and determined to a large degree by the afterhyperpolarization following the action potential, which is attributable primarily to calcium-dependent potassium channels (K_{CaN}) and slow sodium-dependent potassium channels (K_{NaS}) and facilitated by low-voltage activated Ca^{2+} channels ($Ca_{v1.3}$) (26, 31, 34). Output of a matching tectal model to the neuron in Fig. 3*A* is shown in Fig. 3*B*, with only 10% of the standard value (26) of K_{CaN} and K_{NaS} conductances and no Ca_{LVA} current. Properties of the spinal and reticulospinal neurons have been matched in previous studies (7) and have not been changed in the present simulations.

To quantify the influence of spike-frequency adaptation, we simulated turning responses in the model network for different levels of spike-frequency adaptation in tectal neurons (Fig. 3*C*). The turning angle (Fig. 2*E*) was measured for the same turning command (duration around one cycle) with an onset at different phases of the locomotor cycle. With no spike-frequency adaptation (Fig. 3*C*, upper graph), the resulting turning angle was more uniform across different turn phases and the variability was much reduced compared with the case when a substantial spike-frequency adaptation was used (Fig. 3*C*, lower graph). For the remaining part of the study, if not mentioned otherwise, tectal neurons without spike-frequency adaptation were used.

Turning Angle as a Function of Tectal Command Amplitude and Duration in Relation to the Cycle Duration.
Fig. 4 shows the variability in turn angle for turn commands with a constant amplitude and a duration of 0.7 (Fig. 4*A*), 1.0 (Fig. 4*B*), and 1.4 (Fig. 4*C*) times the cycle duration (see also Figs. S1–S3). The variability is much lower when the time coincides with the cycle duration (*T* in Fig. 4*B*) but substantially larger at both 1.4 and 0.7 *T*. At 1.4 *T*, the command will affect not only one full cycle but also part of the following cycle, and therefore the turning angle becomes larger, and conversely with a shorter command the turning angle will be reduced (Fig. 4*A*). The average turn angle increases linearly with the duration of the turn command (Fig. 5*A*), and the variability is the smallest at *T* and 2*T*, as expected from Fig. 4 (compare with Fig. S4).

The average turning angle increases almost linearly with turn command amplitude above a threshold, as shown in Fig. 5*B*. Turn variability is more uniform along the curve and does not change much until at the largest turning amplitudes.

Locomotion Elicited by a Tonic or Modulated Reticulospinal Activity.
Well-coordinated locomotor activity can be induced via a tonic excitatory drive to spinal networks, without the phase-dependent modulation of the reticulospinal neurons. To explore whether the phase-dependent modulation of the reticulospinal neurons would affect the characteristics of the locomotor activity, we

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A Lamprey tectal output neuron

Frequency (Hz) vs Interval — curves labeled 50, 28, 18, 10

Adaptation index vs Mean frequency (Hz)

8 Hz   20 Hz

250ms   250ms

B Tectal model neuron

8 Hz   20 Hz

250 ms   250 ms

C

No SFA

Turn angle (degrees) vs Turn phase (radians)

SFA

Turn angle (degrees) vs Turn phase (radians)

Fig. 3. Spike-frequency adaptation (SFA) in tectal neurons and the corresponding variability of the turns in lamprey. (*A*) Spikes elicited in an output tectal neuron induced by moderate and strong dc stimulation resulting in a maintained frequency of 8 and 20 Hz, respectively. The diagrams show the spike-frequency adaptation (*Upper Left*) to a depolarizing step current of 2 s at different current strength. The upper right diagram shows that the adaptation index increases somewhat with tonic frequency but from a very low level. The adaptation index is the average of the two first spike interval minus the average of the last two intervals divided with the average of the first two. An adaptation index of 0.1–0.2 represent a very low adaptation. (*B*) Spikes in the corresponding model neuron. Conductances of the ionic channels responsible for SFA (K_{CaN} and K_{NaS}) are reduced to 10% of the standard values. No Ca_{LVA} current is present. (*C*) Value of the turn angle depends on the phase of reticulospinal modulation at which the turn command is applied (the turn phase). Turn angle is a 2π-periodic function of the turn phase. A sine function fitted to the data points (filled circles) is plotted with solid line. Simulation data are shown without SFA (0% of K_{CaN}, K_{NaS}, and Ca_{LVA} conductances) and with SFA (10% of K_{CaN} and K_{NaS} and 100% of Ca_{LVA}) in the tectal neurons and fixed turn command applied to TL neurons at random phases (0.5 nA dc stimulation of TL for 115 ms; 32 simulations in each set). Variability of the turn response increases with SFA (*Lower*). Small inset plots show firing frequency of the tectal population in response to the turn command.

compared the amplitude of the locomotor bursts in the rostral section (mostly affected by the phasic drive). It is clear from Fig. 6 that the amplitude of the locomotor bursts were significantly increased with reticulospinal modulation, and therefore

this cyclic modulation of the reticulospinal neurons during locomotion also contributes to the performance and stability of the locomotor network (compare Fig. 5*A* and Fig. S5).

Discussion

In this study, we have simulated the brainstem–spinal cord network underlying locomotion with realistic numbers of cells with appropriate cellular properties. Moreover, we have used this neural network to control a mechanical model of the swimming lamprey. This has been important for elucidating the characteristics of the commands underlying steering. We have focused on one particular problem, the role of the cyclic modulation of the reticulospinal neurons that occur during locomotion. This is a characteristic of many motor systems, but the potential advantage of the cyclic modulation has remained enigmatic.

During steering of the locomotor movements to the left or right, the reticulospinal activity is transiently elevated on one side leading to a bending of the trunk and a change of the direction of swimming. The facilitation of the reticulospinal activity must be correlated to the ongoing locomotor activity and in particular the motoneuronal activity in the rostral segments. A steering command from for instance the optic tectum would have to be timed to occur in the appropriate phase of the movement, particularly if the reticulospinal activity was tonic and not phasically modulated. The problem of the timing would be an additional challenge for the neural control system, which would require more complicated processing. By incorporating a phasic modulation of the reticulospinal neurons that project to a large part of the spinal cord, the problem of timing is substantially

A Turn command duration = 0.7 T

Turn angle (degrees) vs Turn phase (radians)

B Turn command duration = T

Turn angle (degrees) vs Turn phase (radians)

C Turn command duration = 1.4 T

Turn angle (degrees) vs Turn phase (radians)

Fig. 4. Variability of the turn angle in simulations with fixed amplitude of the turn command (0.5 nA dc pulse) and varied duration. Duration of the turn command is given in terms of the period *T* of reticulospinal modulation, 115 ms: 0.7 *T* or 80 ms (*A*), *T* or 115 ms (*B*), and 1.4 *T* or 160 ms (*C*). Experimental setup as in Fig. 3*C*, with 16 simulations in each dataset. Amplitude of the turn variability is markedly reduced if the turn command duration is close to the period *T* of reticulospinal modulation, as can be seen in *B*.

Fig. 5. Turn control curves. Turn angle increases with the turn command duration (*A*) and the turn command amplitude (*B*). Points represent the mean turn angle, and error bars are SD. Each data point is calculated for 16 simulations with random turn phases. Width of the shaded area in *A* shows phasic variability of the turn response (amplitude of the sine fit as in Fig. 4). Turn variability is smaller if the turn command duration is approximately a multiple of the cycle duration, here *T* and *2T* (average cycle duration *T* during the turn is 115 ms, as in Fig. 4).

reduced. The steering command signal will be gated at the reticulospinal level, so it will automatically facilitate the reticulospinal activity in the appropriate phase.

We discovered an additional restriction in the tectal command signal in that it needs to be at a similar level throughout the command. If the command signal were to show substantial spike-frequency adaptation, the effect on the reticulospinal neurons would differ in the early, compared with the late part of the tectal command signal. Therefore, an additional requirement suggested by the simulations would be that the neurons that provide the tectal command display little or no frequency adaptation. The output neurons of tectum responsible for monosynaptic activation of the reticulospinal neurons and the steering commands indeed have a very limited spike-frequency modulation in both lamprey and other vertebrates. We also used simulations to show that with such a control signal, the steering commands were quite reproducible, as long as the steering command had a duration of approximately one cycle. This means that reproducible steering movements can be achieved by a steering command that does not need to be timed very accurately: the phasic modulation of the reticulospinal neurons ascertains that the timing will be appropriate.

We have also shown that the cyclic modulation of the reticulospinal neurons, which occurs in phase with the most rostral

segments, also influences the stability and amplitude of the motoneuronal activity and also has a pacing effect on the spinal excitatory neurons that are responsible for the segmental burst generation.

Materials and Methods

Cell Model. A detailed compartmental model of lamprey locomotor network neurons (26) is used with simulation of the different ion channel subtypes found experimentally. The original model is in ModelDB (available at http://senselab.med.yale.edu/modeldb; accession no. 93319). The cell morphology is simplified in that the small adjacent membrane compartments are lumped together to make longer cylinders. The cell membrane of the simplified model contains 16 compartments: a soma, axon initial segment, and 2 primary, 4 secondary, and 8 tertiary dendrites (Fig. 1*A*). Input resistance is 52 MΩ. Ion current kinetics and conductance distribution are unchanged, for a template cell. The same template cell is used for all types of neurons in the simulation, with necessary modifications, as stated.

Synaptic channels are distributed uniformly among specified compartments, as shown in Fig. 1*A*. Inhibitory (glycinergic) synapses are located on the soma and proximal primary dendrites, with synaptic conductance g_{syn} = 1 nS. Excitatory (AMPA and NMDA) synapses are placed on all compartments except axon initial segment, with synaptic conductance (g_{syn}) of 0.25 and 0.12 nS for AMPA and NMDA synapses, respectively. Other synaptic parameters are set according to (7, 35).

Neural Network. The neural network is built of inhomogeneous populations of neurons (19,600 in total) distinguished by the types of synaptic projections they make and receive. Random uniform variability of ±50% is applied to passive membrane parameters (R_m and C_m), decay time constants of ionic pools (Na+ and Ca2+), and channel conductances responsible for spike-frequency adaptation (K_{CaN} and K_{NaS}) as in ref. 7. The code is available at http://senselab.med.yale.edu/modeldb (accession no. 151338).

Neurons are distributed randomly within a volume corresponding to the spinal cord (100 mm × 2 mm × 0.25 mm) and parts of the brainstem they occupy (4 mm × 2 mm × 0.25 mm), which ensures variability of synaptic delays. Conduction velocity is 0.7 m/s for excitatory projections and 1 m/s for inhibitory projections; minimal synaptic delay is 1 ms.

Populations of 4,000 motoneurons (M), 6,000 excitatory (E), and 4,000 inhibitory (I) interneurons are distributed symmetrically along the left and the right sides of the spinal cord. The spinal network is conventionally divided to 100 segments, each with its own pair of ventral roots. As shown in Fig. 1*B*, E cells make ipsilateral asymmetric projections up to four segments rostrally and eight segments caudally. Inhibitory cells make contralateral projections, 5 segments rostrally and 15 segments caudally. Motoneurons integrate excitatory and inhibitory activity and provide motor output to the muscles. In the simulation, 10 motor output nodes built of a single passive compartment with synaptic channels are set on each side to integrate spikes from M cells

Fig. 6. Dynamic range and amplitude of the locomotor oscillations with cyclic and tonic reticulospinal (RS) drive. Locomotor patterns are generated at different levels of TS stimulation. Amplitude [in arbitrary units (a.u.)] and frequency of oscillations are measured in the third spinal motor output on the right side of the model spinal cord with and without RS modulation (filled and open circles, respectively).

in 20 neighboring segments and provide continuous output signals to the model muscles.

Spinal interneurons and motoneurons are activated synaptically via ipsi-lateral projections from the left and right reticulospinal neurons, 800 cells in each population. A subset of reticulospinal cells (50%) receive synaptic feedback from the spinal interneurons of the most rostral part of the spinal cord within the length of the rostral projections of interneurons, as shown in Fig. 1B. This provides modulation of their activity in phase with oscillations in rostral segments. Synaptic strength of descending spinal projections of modulated reticulospinal neurons decays exponentially with distance from the brainstem (characteristic decay length, 8 mm).

In the simulation, activity of reticulospinal neurons is controlled by the neurons in tectum and in mesencephalic locomotor region (MLR), 4,000 cells in total. Within the tectum, left and right tectal neurons (TL and TR) are considered which project to the right and left reticulospinal populations, respectively. Stimulation of MLR evokes tonic bilateral activation of the network and causes forward swimming (at 8.1 Hz for 0.5 nA dc). Stimu-lation of either TL or TR evokes turning (0.4–0.6 nA dc for the duration of 40–300 ms).

Mechanical Simulations. Continuous drive from the output nodes of the neural network is fed to the mechanical model (27) to control body move-ments in the swimming plane. The mechanical model consists of linear links connected by 10 joints. Proportional activation of every muscle is translated to a pair of torques applied to adjacent links, as shown in Fig. 1C. Resulting movement of the body is the net effect of all muscular torques, viscosity, and stiffness of the joints and resistance of the water. As in ref. 27, linear models

for muscles and water resistance are used. Parameters of the mechanical setup are adapted from the salamander simulation (28), with reduced stiffness and damping in the body joints (10%) to replicate the characteristic cone-shaped body profile during forward swimming (Fig. 2C).

Simulation Platform. Simulations of neural activity are done using the GENESIS neural simulator (36). To solve the differential equations, an implicit hsolve method (37) with an integration time step of 50 μs is used. All network simulations are performed on a CRAY XE6 parallel supercomputer. Mechanical simulations and graphical visualization are done on a Linux platform using Python scripting interface to Open Dynamics Engine library (available at www.ode.org).

Electrophysiology. Whole-cell patch recordings from identified middle rhom-bencephalic reticular nucleus (MRRN) projecting tectal cells were obtained from transverse lamprey tectal slices of 350- to 400-μm thickness maintained in cold (5–6 °C) artificial cerebrospinal fluid. Fluorescent dextran dye injections into the area of the reticulospinal cells in the MRRN were performed to retrogradely label cells in the tectum, which were subsequently identified with the use of a mercury amp. Spiking was induced in these cells after progressively increasing positive intracellular current injections (5–10 pA) of 1 s in duration.

1. Orlovsky GN (1970) Work of the reticulospinal neurons during locomotion. *Biophysics (Oxf)* 15:761–771.
2. Grillner S (2003) The motor infrastructure: From ion channels to neuronal networks. *Nat Rev Neurosci* 4(7):573–586.
3. Marder E (1998) From biophysics to models of network function. *Annu Rev Neurosci* 21:25–45.
4. Orlovsky GN (1972) Activity of vestibulospinal neurons during locomotion. *Brain Res* 46:85–98.
5. Drew T, Dubuc R, Rossignol S (1986) Discharge patterns of reticulospinal and other reticular neurons in chronic, unrestrained cats walking on a treadmill. *J Neurophysiol* 55(2):375–401.
6. Perreault MC, Drew T, Rossignol S (1993) Activity of medullary reticulospinal neurons during fictive locomotion. *J Neurophysiol* 69(6):2232–2247.
7. Kozlov A, Huss M, Lansner A, Kotaleski JH, Grillner S (2009) Simple cellular and net-work control principles govern complex patterns of motor behavior. *Proc Natl Acad Sci USA* 106(47):20027–20032.
8. Stephenson-Jones M, Samuelsson E, Ericsson J, Robertson B, Grillner S (2011) Evolu-tionary conservation of the basal ganglia as a common vertebrate mechanism for action selection. *Curr Biol* 21(13):1081–1091.
9. Wallén P, Williams TL (1984) Fictive locomotion in the lamprey spinal cord in vitro compared with swimming in the intact and spinal animal. *J Physiol* 347:225–239.
10. Buchanan JT, Grillner S (1987) Newly identified 'glutamate interneurons' and their role in locomotion in the lamprey spinal cord. *Science* 236(4799):312–314.
11. Brocard F, et al. (2010) The transformation of a unilateral locomotor command into a symmetrical bilateral activation in the brainstem. *J Neurosci* 30(2):523–533.
12. Ohta Y, Grillner S (1989) Monosynaptic excitatory amino acid transmission from the posterior rhombencephalic reticular nucleus to spinal neurons involved in the control of locomotion in lamprey. *J Neurophysiol* 62(5):1079–1089.
13. Deliagina TG, Zelenin PV, Orlovsky GN (2002) Encoding and decoding of reticulospinal commands. *Brain Res Brain Res Rev* 40(1-3):166–177.
14. Fagerstedt P, Orlovsky GN, Deliagina TG, Grillner S, Ullén F (2001) Lateral turns in the Lamprey. II. Activity of reticulospinal neurons during the generation of fictive turns. *J Neurophysiol* 86(5):2257–2265.
15. Kasicki S, Grillner S, Ohta Y, Dubuc R, Brodin L (1989) Phasic modulation of retic-ulospinal neurones during fictive locomotion and other types of spinal motor activity in lamprey. *Brain Res* 484(1-2):203–216.
16. Vinay L, Grillner S (1992) Spino-bulbar neurons convey information to the brainstem about different phases of the locomotor cycle in the lamprey. *Brain Res* 582(1):134–138.
17. Dubuc R, Grillner S (1989) The role of spinal cord inputs in modulating the activity of reticulospinal neurons during fictive locomotion in the lamprey. *Brain Res* 483(1):196–200.
18. Dubuc R, Bongianni F, Ohta Y, Grillner S (1993) Anatomical and physiological study of brainstem nuclei relaying dorsal column inputs in lampreys. *J Comp Neurol* 327(2):260–270.

19. Antri M, Fénelon K, Dubuc R (2009) The contribution of synaptic inputs to sustained depolarizations in reticulospinal neurons. *J Neurosci* 29(4):1140–1151.
20. Vinay L, Grillner S (1993) The spino-reticulo-spinal loop can slow down the NMDA-activated spinal locomotor network in lamprey. *Neuroreport* 4(6):609–612.
21. Einum JF, Buchanan JT (2006) Spinobulbar neurons in lamprey: Cellular properties and synaptic interactions. *J Neurophysiol* 96(4):2042–2055.
22. Vinay L, Bongianni F, Ohta Y, Grillner S, Dubuc R (1998) Spinal inputs from lateral columns to reticulospinal neurons in lampreys. *Brain Res* 808(2):279–293.
23. Jones MR, Grillner S, Robertson B (2009) Selective projection patterns from subtypes of retinal ganglion cells to tectum and pretectum: Distribution and relation to be-havior. *J Comp Neurol* 517(3):257–275.
24. Saitoh K, Ménard A, Grillner S (2007) Tectal control of locomotion, steering, and eye movements in lamprey. *J Neurophysiol* 97(4):3093–3108.
25. Kardamakis A, Stepheson-Jones M, Robertson B, Grillner S (2012) The tectal micro-circuit – a lamprey perspective. Neuroscience 2012, New Orleans, LA, October 13–17, 2012, abstr 78.25/GG6.
26. Huss M, et al. (2007) Roles of ionic currents in lamprey CpG neurons: A modeling study. *J Neurophysiol* 97(4):2696–2711.
27. Ekeberg Ö (1993) A combined neuronal and mechanical model of fish swimming. *Biol Cybern* 69(5):363–374.
28. Harischandra N, et al. (2011) Sensory feedback plays a significant role in generating walking gait and in gait transition in salamanders: A simulation study. *Front Neuro-robot* 5:3.
29. Ekeberg Ö, Grillner S (1999) Simulations of neuromuscular control in lamprey swim-ming. *Philos Trans R Soc Lond B Biol Sci* 354(1385):895–902.
30. Wallén P, Grillner S, Feldman JL, Bergelt S (1985) Dorsal and ventral myotome mo-toneurons and their input during fictive locomotion in lamprey. *J Neurosci* 5(3):654–661.
31. Wallén P, et al. (1989) Effects of 5-hydroxytryptamine on the afterhyperpolarization, spike frequency regulation, and oscillatory membrane properties in lamprey spinal cord neurons. *J Neurophysiol* 61(4):759–768.
32. Gutmaniene N, Svirskiene N, Svirskis G (2003) Firing properties of frog tectal neurons in vitro. *Brain Res* 981(1-2):213–216.
33. Pratt KG, Aizenman CD (2009) Multisensory integration in mesencephalic trigeminal neurons in Xenopus tadpoles. *J Neurophysiol* 102(1):399–412.
34. Wallén P, et al. (2007) Sodium-dependent potassium channels of a Slack-like subtype contribute to the slow afterhyperpolarization in lamprey spinal neurons. *J Physiol* 585(Pt 1):75–90.
35. Tråvén HG, et al. (1993) Computer simulations of NMDA and non-NMDA receptor-mediated synaptic drive: Sensory and supraspinal modulation of neurons and small networks. *J Neurophysiol* 70(2):695–709.
36. Bower JM, Beeman D (1998) *The Book of GENESIS: Exploring Realistic Neural Models with the General Neural Simulation System* (Springer-Verlag, New York), 2nd Ed.
37. Hines M (1984) Efficient computation of branched nerve equations. *Int J Biomed Comput* 15(1):69–76.

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