Changes in Electrophysiological Properties of Lamprey Spinal Motoneurons During Fictive Swimming

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Martin, Michelle, M. Changes in electrophysiological properties of lamprey spinal motoneurons during fictive swimming. J Neurophysiol 88: 2463–2476, 2002; 10.1152/jn.00725.2001. Electrophysiological properties of lamprey spinal motoneurons were measured to determine whether their cellular properties change as the spinal cord goes from a quiescent state to the active state of fictive swimming. Intracellular microelectrode recordings of membrane potential were made from motoneurons in the isolated spinal cord preparation. Electrophysiological properties were first characterized in the quiescent spinal cord, and then fictive swimming was induced by perfusion with calcium-free solution (cobalt substituted for calcium) was tested in the quiescent spinal cord. It was found that, in addition to reducing the amplitude of the slow after-spike hyperpolarization (sAHP). Serotonin is known to be released endogenously from the spinal cord, and then fictive swimming was induced by perfusion with D-glutamate and the measurements were repeated. During the depolarizing excitatory phase of fictive swimming, the motoneurons had significantly reduced rheobase and significantly increased input resistance compared with the quiescent state, with no significant changes in these parameters during the repolarizing inhibitory phase of swimming. Spike threshold did not change significantly during fictive swimming compared with the quiescent state. During fictive swimming, the slope of the spike frequency versus injected current (F-I) relationship decreased significantly as did spike-frequency adaptation and the amplitude of the slow after-spike hyperpolarization (sAHP). Serotonin is known to be released endogenously from the spinal cord during fictive swimming and is known to reduce the amplitude of the sAHP. Therefore the effects of serotonin on cellular properties were tested in the quiescent spinal cord. It was found that, in addition to reducing the sAHP amplitude, serotonin also reduced the slope of the F-I relationship and reduced spike-frequency adaptation, reproducing the changes observed in these parameters during fictive swimming. Application of spiperone, a serotonin antagonist, significantly increased the sAHP amplitude during fictive swimming but had no significant effect on F-I slope or adaptation. Because serotonin may act in part through reduction of calcium currents, the effect of calcium-free solution (cobalt substituted for calcium) was tested in the quiescent spinal cord. Similar to fictive swimming and serotonin application, the calcium-free solution significantly reduced the sAHP amplitude, the slope of the F-I relationship, and spike-frequency adaptation. These results suggest that there are significant changes in the firing properties of motoneurons during fictive swimming compared with the quiescent state, and it is possible that these changes may be attributable in part to the endogenous release of serotonin acting via reduction of calcium currents.

INTRODUCTION

The activity of neuronal networks depends both on the properties of the component nerve cells and their synaptic interactions. Several invertebrate preparations have offered key insights into how nerve cells and their synaptic and cellular properties interact to produce the neuronal activities that underlie motor behavior (Getting 1989; Marder and Calabrese 1996). Similar approaches are being applied to the study of vertebrate locomotor networks in a variety of preparations, including the frog (Dale and Kuenzi 1997), turtle (Perrier and Hounsgaard 1999), and mammals (Kiehn et al. 2000). One of the best-studied adult vertebrate locomotor networks is that of the lamprey (Buchanan 2001; Grillner et al. 2001). In the lamprey, the locomotor network can be activated in the isolated spinal cord with an excitatory amino acid (fictive swimming) (Cohen and Wallén 1980; Wallén and Williams 1984), and several classes of nerve cells that participate in swimming activity have been characterized by their morphology and their synaptic interactions (Buchanan 2001). In addition, the electrophysiological properties of these classes of lamprey spinal neurons have been described in the quiescent spinal cord (Buchanan 1993).

It is likely, however, that the electrophysiological properties of nerve cells undergo changes during network activity, and thus the cellular properties characterized in the quiescent state may not be the same as during network activity. For example, in the cat spinal cord it has been demonstrated that the firing properties of motoneurons change in locomotor activity (Brownstone et al. 1992; Krawitz et al. 2001). Changes in cellular properties could result from activity-dependent changes in voltage-gated channels and ligand-gated channels. For example, during fictive locomotion the membrane potentials of lamprey spinal neurons exhibit rhythmic membrane potential oscillations so that voltage-gated channels will be in varying states of activation and inactivation (Buchanan and Cohen 1982). This rhythmic activity is due to rhythmic excitatory and inhibitory synaptic inputs acting via ionotropic glutamate and glycine receptors, which will also affect the membrane properties of the cells (Moore and Buchanan 1993). In addition, activation of N-methyl-D-aspartate (NMDA) receptors can induce oscillatory activity even in the presence of tetrodotoxin (Wallén and Grillner 1987), and the voltage-dependence of the NMDA channel can generate large increases in input impedance (Moore et al. 1995). Neuronal networks are also under the influence of multiple neuromodulators that are selectively released during activity versus quiescence. These modulators have been shown to affect many of the ion channels.
regulating cellular and synaptic activity (Harris-Warrick and Marder 1991; Nusbaum et al. 2001). In lamprey, several neuromodulators, including serotonin and dopamine, have been shown to alter the locomotor network by actions on both cellular and synaptic properties (Buchanan and Grillner 1991; Harris-Warrick and Cohen 1985; Kemnitz 1997; Matsushima and Grillner 1992; Takahashi et al. 2001; Van Dongen et al. 1986; Wallén et al. 1989). These transmitters are present in neurons and processes within the spinal cord (McPherson and Kemnitz 1994; Schotland et al. 1995; Van Dongen et al. 1985), neurons and processes within the spinal cord (McPherson and Kemnitz 1994; Schotland et al. 1995; Van Dongen et al. 1985), and there is evidence that serotonin is released during fictive swimming (Christensen et al. 1989).

Thus, if we are to fully understand the operation of the lamprey locomotor network, or any neuronal network, it will be necessary to characterize the electrophysiological properties of the neurons during network activity. Also, understanding how the properties change in individual cells as they go from a quiescent state to an active state may give insight into the types of modulation of the network neurons that are occurring. In the present work, we have first characterized the electrophysiological properties of motoneurons in a quiescent state and then induced fictive swimming and measured the same properties again to determine whether they change. While some properties do not change significantly, others do change, such as the amplitude of the slow after-spike hyperpolarization (sAHP) and the slope of the firing frequency versus current relationship. Additional experiments suggest that these changes may be due to the release of serotonin and reduction of calcium currents.

METHODS

Animals and preparation

Thirty-four adult silver lampreys (*Ichthyomyzon unicuspis*), 21–33 cm in length, were used in these experiments. The animals were kept in freshwater tanks at 7°C without feeding. For dissection, the animals were anesthetized by immersion in a 0.1 mg/ml solution of tricaine methanesulfonate (Sigma). The details of the dissection have been previously described (Rovainen 1974). Dissections and experiments were done in cooled Ringer solution (9–10°C) containing (in mM): 91 NaCl, 2.1 KCl, 2.6 CaCl_2_, 1.8 MgCl_2_, 4 glucose, 20 NaHCO_3_, 8 HEPES-free acid, and 2 HEPES sodium salt. The pH was adjusted to 7.4, and the solution was bubbled continuously with 98% O_2_–2% CO_2_ during the experiment. The preparation typically consisted of 8 to 12 segments of spinal cord from the midbody region between the last gill and the beginning of the fin. The notochord was split down the ventral midline and pinned to the floor of a cooled chamber lined with silicone elastomer (Sylgard, Dow Corning). The dorsal meninges, including the meninx primativa, were then removed from the spinal cord. The preparation was viewed with a stereomicroscope with illumination from below. All experimental procedures were approved by the institutional animal care and use committee.

Recording techniques

Intracellular recordings of membrane potential were made using microelectrodes pulled on a horizontal puller (P-87, Sutter) from glass pipettes containing a filament and backfilled with 4 M potassium acetate. The electrodes typically had resistances of 30–60 MΩ. On impalement, an action potential was elicited by depolarizing current injection, and the cell was identified as a motoneuron by the presence of one-for-one extracellular spikes occurring with fixed latency in a nearby ventral root as recorded with a glass suction electrode (Fig. 1A). The cell was allowed to recover until the membrane potential remained stable and the action potential was >80 mV in amplitude. Cells that did not maintain an action potential >80 mV as measured from near resting potential throughout the experiment were excluded from analysis. Once the cell stabilized after impalement, various electrophysiological properties of the cell were measured under three conditions: quiescent, fictive swimming, and wash. After the final set of recordings, the electrode was removed from the spinal cord and the amount of electrode polarization was measured. If polarization was >2 mV, then the voltage measurements were corrected. The wash recordings were corrected by the full amount of polarization, and the fictive swimming recordings were corrected by one-half of the polarization. No correction was made in the quiescent recordings, as the electrode potential was nulled just prior to cell impalement. The average polarization at the end of the experiment (1–2 h) was ~9 mV.

Measurement and analysis of electrophysiological properties

Intracellular recordings were done in current clamp using an Axoclamp 2B amplifier (Axon Instruments). The signals were low-pass filtered at 1 kHz with a CyberAmp 320 DC amplifier (Axon Instruments) and digitized at 3–4 kHz using a micro1401 computer interface with Spike2 software (Cambridge Electronic Design). Action potential properties were measured in bridge mode by eliciting action potentials with a 1-ms current pulse repeated at 2-s intervals. An average of 29.5 ± 10.1
(mean ± SD) action potentials were elicited to make each measurement. Spike amplitude and half-amplitude duration were measured individually for each of the action potentials and the population average was calculated from these measurements. For the sAHP, all the action potentials including the sAHP were averaged first, and then the amplitude of the sAHP was measured once from the averaged trace.

Electrophysiological properties that required current injection were done in discontinuous current clamp mode (DCC) at a sampling rate of 1–2 kHz. The adequacy of the sampling rate was monitored throughout the experiment and occasionally adjusted as needed. Input resistance was measured with 200-ms negative current pulses delivered at 2-s intervals. Five or six levels that hyperpolarized the cell to a maximum of 20 mV below resting potential were given, and three to four traces at each level were averaged (Fig. 3A). Steady-state voltage in the last 50 ms of the pulse was measured and plotted versus the injected current (Fig. 3B). A linear regression was fitted to the voltage-current relationship, and the slope of the regression was taken as the input resistance of the cell. Rheobase was defined as the minimal current level required to elicit an action potential and was measured in DCC mode using depolarizing pulses of 200-ms duration (Fig. 2, A and B). Because synaptic inputs could cause spontaneous spike generation during fictive swimming, four or more pulses were given and rheobase was considered to be the lowest current level that could initiate a spike during >50% of the pulses. Spike threshold was measured as the membrane potential at the inflection point of the action potential initiated at rheobase.

The relationship between firing frequency and the depolarizing current level ($F-I$) was measured in DCC mode using 200-ms pulses repeated at 2-s intervals. For this procedure, 15 to 20 current levels were incremented from rheobase to the approximate maximum firing frequency of the cell. At each current level, the intervals between the first and second action potentials of two to six pulses were averaged. The instantaneous firing frequency was calculated as the inverse of this interval, and the instantaneous firing frequency of the first interval was plotted versus the injected current. To characterize the $F-I$ relationship, a three-parameter sigmoidal curve was fitted to the data

$$f = \alpha/[1 + \exp(-(x - x_0)/\beta)]$$  \hspace{1cm} (1)

where $f$ is the instantaneous firing frequency in Hz and $x$ is the injected current in nA. All fitted curves had $R^2 > 0.8$. Three characteristics of the curve fits were extracted: the slope at the inflection point ($\alpha/4\beta$), the $x$-axis value of the inflection point ($x_0$), and the saturation firing frequency ($\alpha$). The first and second spike intervals were used to determine the degree of spike frequency adaptation. Spike frequency adaptation was defined as the difference between the average instantaneous firing frequencies of the first and second spike intervals, normalized to the instantaneous frequency of the first interval (Fig. 6A). Expressed in terms of interspike intervals

$$\text{spike-frequency adaptation} = \{1 - \left(\frac{1}{2}\right)\} \times 100\% \hspace{1cm} (2)$$

The calculation of spike frequency adaptation was done over the first interval frequency range of 40 to 80 Hz. The values for spike frequency adaptation are expressed as percentages.

During fictive swimming, the motoneurons receive phasic periods of synaptic excitation and inhibition that coincide approximately with the timing of the burst and silent period in the ipsilateral ventral root, respectively (Kahn 1982; Russell and Wallén 1983). In these experiments, oscillations of the membrane potential within each motoneuron follow the bursting pattern of ventral root as well, so that the peak level of depolarization is always at or near the center of the burst. Thus the properties were measured at two time points during the swim cycle: during the depolarized peak (excitatory phase) and the hyperpolarized trough (inhibitory phase) of the oscillating membrane potential. The ventral root burst was used as the criterion for the time of measurement; a current pulse given in the middle of the ventral root burst was considered to be in the excitatory phase, and a pulse given outside of the burst was in the inhibitory phase. The burst occupies about 30% of the entire cycle during swimming, and the average cycle period in these experiments was 4.0 s. Pulses given in the excitatory phase were required to be $\geq \frac{1}{2}N$ within the ventral root burst. Because the inhibitory phase was longer, the pulses were required to be completely outside of the burst; the beginning or end of a pulse was always $>100$ ms away from edge of an adjacent burst.

Conditions induced for measurement of electrophysiological properties

The main goal of these experiments was to measure the effect of fictive swimming on the electrophysiological properties of lamprey spinal motoneurons and to compare these changes with those induced by serotonin or calcium-free solution. Several different protocols were used to induce these conditions. For experiments testing the effects of fictive swimming, quiescent recordings were done after the membrane potential stabilized following impalement. Fictive swimming was then
induced by bath perfusion of 0.75 mM d-glutamate, and the electrophysiological properties were remeasured about 10 to 15 min later when the ventral root showed regular bursting activity. During fictive swimming, the recordings were repeated twice: during the excitatory phase and the inhibitory phase of swimming. The d-glutamate was then washed out with normal Ringer solution, and the measurements were repeated after 10 to 20 min when the ventral root was again silent. During the wash, the electrophysiological properties were measured at resting potential and while a depolarization was imposed on the cell to match the membrane potential observed during the excitatory phase of fictive swimming. For experiments using spiperone, measurements were first done in quiescence and in normal fictive swimming, then spiperone (10 μM), a blocker of serotonin receptors in lamprey (Wikström et al. 1995), was added during fictive swimming and the measurements were repeated. There was no discrimination between fictive swim phases because spiperone had a tendency to disrupt ventral root firing patterns. For experiments using serotonin or calcium-free solution, the measurements were repeated after 10 to 20 min when the ventral root was again fully active.

The electrophysiological properties were remeasured about 10 to 15 min later during the wash. The electrophysiological properties were remeasured about 10 to 15 min later during the wash. Therefore, the Wilcoxon signed-rank test was used for statistical analysis. Significance was considered to be 0 ≤ 0.05. Values are given as the mean ± SD.

**RESULTS**

**Cell excitability**

As reported previously (Buchanan and Cohen 1982; Buchanan and Kasicki 1995; Kahn 1982; Russell and Wallén 1983), the membrane potentials of lamprey spinal motoneurons exhibited oscillations during fictive swimming (Fig. 1, B and C and Table 1). The changes in membrane potential were significant when comparing the excitatory phase of swimming versus quiescence and the excitatory phase versus the inhibitory phase (Fig. 1D) (P < 0.001 for both).

As a basic measure of cell excitability, rheobase decreased significantly during the excitatory phase of fictive swimming compared with the quiescent state, 0.8 ± 0.8 versus 2.5 ± 1.5 nA (n = 15; P < 0.001) (Fig. 2, A, C, and D), but did not differ significantly when comparing the inhibitory phase versus quiescence, 2.7 ± 1.6 versus 2.5 ± 1.5 nA (n = 15) (Fig. 2, B, C, and D). Rheobase also differed significantly between the two phases of swimming (n = 15; P < 0.001) (Fig. 2, C and D).

Rheobase is determined in part by membrane potential but is also influenced by input resistance and spike threshold. In the quiescent state, the mean input resistance of the motoneurons was 11.0 ± 4.7 MΩ and increased significantly to 14.7 ± 7.6 MΩ (n = 15; P = 0.01) during the excitatory phase of swimming (Fig. 3, C and D). In the example motoneuron in Fig. 3, A and B, the input resistance increased from 12.7 to 21.9 MΩ in the excitatory phase. The input resistance during the inhibitory phase of swimming was also larger than in quiescence but this difference was not significant. There were no significant changes in spike threshold when comparing the quiescent state versus fictive swimming (Fig. 4, A and B). Thus

**Statistics**

For each measured property, significance was determined from the population of motoneurons, as opposed to assessing whether changes were significant in individual cells. Multiple measurements in individual cells were not done because of the necessity to average many individual measurements in the face of large membrane potential fluctuations during fictive swimming. In about two-thirds of the measured properties, quiescent values did not show a normal distribution. Therefore the Wilcoxon signed-rank test was used for statistical analysis.

<table>
<thead>
<tr>
<th>Property</th>
<th>Q</th>
<th>ES</th>
<th>IS</th>
<th>W</th>
<th>D</th>
<th>Q-ES</th>
<th>ES-IS</th>
<th>IS-W</th>
<th>W-D</th>
</tr>
</thead>
<tbody>
<tr>
<td>Membrane potential (mV)</td>
<td>−78.3</td>
<td>± 5.1</td>
<td>−68.8</td>
<td>± 5.5</td>
<td>−79.1</td>
<td>± 6.2</td>
<td>−79.3</td>
<td>± 3.9</td>
<td>−67.8</td>
</tr>
<tr>
<td>Rheobase (nA)</td>
<td>2.5</td>
<td>± 1.5</td>
<td>0.8</td>
<td>± 0.8</td>
<td>2.7</td>
<td>± 1.6</td>
<td>2.5</td>
<td>± 1.4</td>
<td>1.6</td>
</tr>
<tr>
<td>Input resistance (MΩ)</td>
<td>11.0</td>
<td>± 4.7</td>
<td>14.7</td>
<td>± 7.6</td>
<td>12.9</td>
<td>± 7.3</td>
<td>11.4</td>
<td>± 4.0</td>
<td>12.4</td>
</tr>
<tr>
<td>Spike threshold (mV)</td>
<td>−55.8</td>
<td>± 2.8</td>
<td>−57.1</td>
<td>± 4.6</td>
<td>−58.4</td>
<td>± 3.3</td>
<td>−55.3</td>
<td>± 2.9</td>
<td>−58.0</td>
</tr>
<tr>
<td>Action potential peak (mV)</td>
<td>16.2</td>
<td>± 10.5</td>
<td>14.7</td>
<td>± 13.7</td>
<td>13.3</td>
<td>± 13.4</td>
<td>19.0</td>
<td>± 14.7</td>
<td>21.9</td>
</tr>
<tr>
<td>Action potential amp. (mV)</td>
<td>94.5</td>
<td>± 13.3</td>
<td>83.6</td>
<td>± 14.8</td>
<td>92.7</td>
<td>± 14.7</td>
<td>98.4</td>
<td>± 16.3</td>
<td>89.7</td>
</tr>
<tr>
<td>Action potential dur. (ms)</td>
<td>2.27</td>
<td>± 0.66</td>
<td>2.10</td>
<td>± 0.60</td>
<td>2.13</td>
<td>± 0.73</td>
<td>2.10</td>
<td>± 0.35</td>
<td>1.90</td>
</tr>
<tr>
<td>F-I slope (a/4βf) (Hz/μA)</td>
<td>26.9</td>
<td>± 11.5</td>
<td>18.7</td>
<td>± 6.9</td>
<td>19.1</td>
<td>± 6.7</td>
<td>25.7</td>
<td>± 8.3</td>
<td>21.4</td>
</tr>
<tr>
<td>F-I inflection point (μA)</td>
<td>4.8</td>
<td>± 2.3</td>
<td>3.5</td>
<td>± 1.5</td>
<td>5.7</td>
<td>± 2.7</td>
<td>4.9</td>
<td>± 2.1</td>
<td>3.5</td>
</tr>
<tr>
<td>F-I saturation (a) (Hz)</td>
<td>84.2</td>
<td>± 5.3</td>
<td>80.2</td>
<td>± 7.0</td>
<td>81.1</td>
<td>± 8.7</td>
<td>87.7</td>
<td>± 11.6</td>
<td>84.1</td>
</tr>
<tr>
<td>Adaptation (%)</td>
<td>42.0</td>
<td>± 15.2</td>
<td>34.6</td>
<td>± 13.8</td>
<td>37.1</td>
<td>± 7.7</td>
<td>41.5</td>
<td>± 16.5</td>
<td>32.6</td>
</tr>
<tr>
<td>Slow AHP amplitude (mV)</td>
<td>3.0</td>
<td>± 0.7</td>
<td>4.0</td>
<td>± 1.7</td>
<td>2.4</td>
<td>± 0.9</td>
<td>3.3</td>
<td>± 0.8</td>
<td>5.3</td>
</tr>
</tbody>
</table>

Values are means ± SD. Numbers in parentheses reprint cells. P values represent differences. Significance determined using the Wilcoxon signed-rank test. Q, quiescent; ES, excitatory swim; IS, inhibitory swim; W, wash; D, depolarization (imposed on a quiescent cell to match value in the excitatory phase of swimming).
the changes in rheobase associated with fictive swimming appeared to have been mainly due to changes in membrane potential and perhaps also to the change in input resistance. When the changes in rheobase were compared with changes in membrane potential, input resistance, and spike threshold, the only significant correlation was between rheobase and membrane potential. For rheobase versus membrane potential, the correlation or \( r^2 \) of the linear regression was 0.57, for rheobase versus input resistance, 0.08, and for rheobase versus spike threshold, 0.01. However, when rheobase was compared during quiescence and fictive swimming at the same membrane potentials (depolarization was imposed on the quiescent motoneuron by continuous current injection), the reduction observed in fictive swimming was significantly greater. During the excitatory phase of swimming, rheobase was reduced from 2.5 \( \pm \) 1.5 to 0.8 \( \pm \) 0.8 nA, while depolarization reduced it to a lesser extent, to 1.6 \( \pm \) 1.2 nA. The difference was statistically significant \((n = 14; P = 0.05)\). Thus the change in rheobase cannot be accounted for solely by membrane depolarization.

In these experiments, the speed of swimming was typically slow; the mean cycle period was 4.0 \( \pm \) 3.9 s \((n = 24)\) or 0.25 Hz. The slower swimming was probably because of the short time allowed for the glutamate to activate swimming. Of the 24 motoneurons studied during fictive swimming, 10 showed spiking during the excitatory phase.

Action potential properties

Three properties of the action potential were measured (Table 1): the membrane potential of the peak, the base-to-peak amplitude, and the half-amplitude duration. While the membrane potential of the peak of the action potential did not change significantly going from quiescence to the excitatory phase of fictive swimming, the base-to-peak amplitude significantly decreased from 94.5 \( \pm \) 13.3 to 83.6 \( \pm \) 14.8 mV \((n = 15; P = 0.002)\) due to the depolarization of the base membrane potential. The base-to-peak amplitude in quiescence was not significantly different from the inhibitory phase of fictive swimming, 94.5 \( \pm \) 13.3 versus 92.7 \( \pm \) 14.7 mV \((n = 14)\). The half-amplitude duration of the action potential did not change significantly during swimming.

F-I relationship

In the example motoneuron of Fig. 5, A and B, when comparing the excitatory phase of swimming (ES) versus quiescence (Q), there is a decrease in the slope of the F-I relationship and a leftward shift of the curve along the x-axis. During the inhibitory phase in Fig. 5B, the F-I curve also showed a decreased slope, but a rightward shift in \( x_e \). There also appears to be a decrease in the
saturation level of firing frequency for this neuron, but saturation did not change significantly across the population. The slopes of the F-I curves are plotted in Fig. 5C for all the motoneurons. In the quiescent state, the mean slope of the F-I relationship was $26.9 \pm 11.5 \text{ Hz/nA}$ and decreased significantly during fictive swimming to $18.7 \pm 6.9 \text{ Hz/nA} (n = 13; P = 0.001)$ during the excitatory phase and to $19.1 \pm 6.7 \text{ Hz/nA} (n = 12; P = 0.015)$ during the inhibitory phase (Fig. 5D).

During the excitatory phase of fictive swimming there was a significant decrease in $x_0$, which is the $x$-axis value of the inflection point of the sigmoidal curve. In the quiescent state, the mean $x_0$ was $4.8 \pm 2.3 \text{ nA}$ and it decreased to $3.5 \pm 1.5 \text{ nA}$ during excitatory swim ($n = 13; P = 0.005$) (Table 1). During the inhibitory phase, the F-I curve shifted to the right compared with the quiescent state $x_0$. 4.8 $\pm$ 2.3 versus 5.7 $\pm$ 2.7 nA ($n = 12; P = 0.005$) in inhibitory swim. When a depolarization was imposed on the cell during quiescence to match the depolarization of excitatory swim, the mean $x_0$ was 3.5 $\pm$ 1.7 nA ($n = 10$), not significantly different from the swimming value of 3.5 $\pm$ 1.5 nA (Table 1). This suggests that the leftward shift is due primarily to the depolarization associated with the excitatory phase of swimming. The saturation level ($\alpha$), which represents the maximum firing frequency of the cell, decreased but not significantly during fictive swimming (Table 1).

From the F-I relationship, spike-frequency adaptation was measured as the decrease in firing frequency between the first and second spike intervals (Fig. 6A). There was a significant decrease in the degree of spike-frequency adaptation in the excitatory phase of swimming compared with quiescence: 34.6 $\pm$ 13.8 versus 42.0 $\pm$ 15.2% ($n = 15; P = 0.008$), but not in the inhibitory phase versus quiescence. Depolarization alone also decreased the adaptation, from 42.0 $\pm$ 15.2 to 32.6 $\pm$ 14.7% ($n = 13; P = 0.003$) (Fig. 6, B and C).

Slow AHP

An important ionic current that influences the F-I relationship and spike-frequency adaptation is the calcium-activated potassium current that underlies the sAHP (Hill et al. 1985). Therefore the amplitude of the sAHP was measured during fictive swimming. Due to the significant changes in membrane potential during swimming and the proximity of the sAHP reversal potential to the resting membrane potential, there will be significant changes in driving force on the sAHP during fictive swimming. Therefore, to compare the amplitude of the sAHP during quiescence and during fictive swimming, it was necessary to make the comparison of amplitudes at the same resting membrane potential (Fig. 7, D and E). In the example motoneuron illustrated in Fig. 7, A and B, there was a decrease in the amplitude of the sAHP over a range of resting membrane potentials during the excitatory phase of swimming compared with the quiescent state. Plots of the amplitude of the sAHP versus the imposed membrane potentials revealed that the slope of this relationship decreased significantly during fictive swimming from 0.34 $\pm$ 0.18 in quiescence to 0.14 $\pm$ 0.09 ($n = 8; P < 0.001$) (Fig. 7C) during the excitatory phase of fictive swimming. Since the membrane potential in quiescence was not significantly different from that during the inhibitory phase (Fig. 1D), it was possible to directly compare the sAHP amplitude under these two conditions (Fig. 7, D and F). The mean quiescent sAHP amplitude of 3.0 $\pm$ 0.7 mV was significantly reduced to 2.4 $\pm$ 0.9 mV during the inhibitory phase of fictive swimming ($n = 14; P = 0.005$). When the quiescent cells were depolarized to the same membrane potential observed during the excitatory phase of fictive swimming, the sAHP amplitude was 5.3 $\pm$ 1.8 mV, which was significantly larger than the 4.0 $\pm$ 1.7 mV measured during the excitatory phase ($n = 14; P = 0.015$) (Fig. 7, E and F).

Effects of serotonin

As has been reported previously (Buchanan and Grillner 1991), bath perfusion of serotonin (5 μM) produced a significant hyperpolarization of the motoneurons from $-72.1 \pm 3.8$ to $-77.1 \pm 6.0$ mV ($n = 12; P = 0.004$).
Serotonin did not produce significant effects on membrane potential, input resistance, spike threshold, or action potential amplitude and duration. As previously reported (Van Dongen et al. 1986), serotonin significantly reduced the sAHP amplitude from 2.3 ± 1.2 to 0.5 ± 0.5 mV (n = 12; P < 0.001) (Fig. 8A). However, the serotonin-induced hyperpolarization alone could have caused this reduction in sAHP amplitude due to a reduction in driving force on the sAHP potassium current. Therefore the amplitude of the sAHP was measured at several membrane potential levels imposed by current injection and the sAHP amplitude was plotted versus membrane potential. A linear regression fitted to this relationship exhibited a significant decrease in slope from 0.23 ± 0.11 to 0.09 ± 0.05 (n = 8; P < 0.001), indicating that the membrane potential versus sAHP amplitude relationship was reduced by the applied serotonin (Fig. 8B).

The effect of applied serotonin on the F-I relationship was also examined. As shown in the example motoneuron of Fig. 8C, serotonin decreased the slope of the F-I relationship, as was also observed in fictive swimming (Fig. 7). Overall, applied serotonin reduced the F-I slope from 43.9 ± 14.0 to 34.7 ± 10.7 Hz/nA (n = 12; P = 0.03) (Fig. 8D). Serotonin had no significant effects on x₀ or saturation (α) (Table 2). Spike-frequency adaptation was also reduced by applied serotonin, similar to the changes in adaptation observed during fictive swimming (Fig. 6). The mean adaptation was reduced from 33.4 ± 19.9 to 21.4 ± 9.3% (n = 12; P = 0.02) with the application of serotonin (Table 2).

Effects of spiperone during fictive swimming

If the changes that are observed during swimming were due in part to the endogenous release of serotonin, then blocking the action of serotonin during fictive swimming would be expected to reverse some of these changes. This was tested using spiperone, a blocker of 5HT₁A receptors that has been shown to be effective in blocking the reduction of the sAHP amplitude by serotonin in lamprey (Wikström et al. 1995). As a control, spiperone (10 μM) was tested on quiescent motoneurons to determine whether spiperone alone had any effects on their electrophysiological properties in the absence of applied serotonin. The only property that changed was rheobase, which increased in all four cells tested with a mean change of 2.0 ± 1.1 to 2.5 ± 1.4 nA (data not shown).

Spiperone was tested on fictive swimming by first measuring the electrophysiological properties in the quiescent cell, then during fictive swimming, and finally during fictive swimming with bath-applied spiperone. Before adding spiperone, the quiescent versus swim differences were similar to those reported above (Table 2). When spiperone was added, the only significant change in cellular properties was an increase in the sAHP amplitude (Fig. 9, A and B). As shown for the example motoneuron in Fig. 9A, the slope of the sAHP amplitude versus membrane potential relationship was reduced during swimming compared with the control. Addition of spiperone partially reversed this reduction. The mean sAHP amplitude increased significantly from 2.3 ± 0.9 to 3.6 ± 1.1 mV (n = 9; P = 0.03) with the addition of spiperone (Fig. 9B) while membrane potential was not significantly altered by spiperone (−70.4 ± 5.8 versus −70.5 ± 6.6 mV) (Table 2). The mean slope of the F-I relationship was increased by spiperone in seven of nine cells, but the difference was not statistically significant (Fig. 9, C and D). Before the addition of spiperone, the slope of the F-I relationship was reduced during fictive swimming from 48.1 ± 18.6 to 31.2 ± 10.7 Hz/nA (n = 9). After addition of spiperone, the slope was 34.2 ± 11.2 Hz/nA (n = 9; P = 0.15). During fictive swimming, spiperone did not have significant effects on membrane potential, input resistance, rheobase, spike threshold, x₀, saturation firing frequency of the F-I relationship, adaptation, or action potential amplitude and duration (Table 2).

Effects of calcium-free solution

It has been reported that serotonin reduces calcium currents in lamprey spinal neurons (El Manira et al. 1997). To test whether a reduction of calcium currents may be contributing to
the effects of serotonin on motoneurons, the electrophysiological properties of motoneurons were measured before and after substituting cobalt for calcium. Calcium-free solution produced similar changes observed in swimming and with the addition of serotonin for the slope of the $F-I$ relationship, the amplitude of the sAHP, and spike-frequency adaptation (Table 2). As shown for the example motoneuron in Fig. 10A, the amplitude of the sAHP was reduced by calcium-free solution as would be expected for a calcium-activated current. The mean sAHP amplitude was significantly reduced from $2.6 \pm 1.2$ to $1.0 \pm 0.9$ mV ($n = 11; P = 0.001$) in the calcium-free solution with no significant change in the mean membrane potential, $-70.0 \pm 6.2$ versus $-71.6 \pm 6.4$ mV (Table 2). The slope of the sAHP amplitude versus membrane potential relationship was reduced in all five cells tested from a mean level of $0.13 \pm 0.10$ to $0.02 \pm 0.008$ in the calcium-free solution (Fig. 10B). The mean $F-I$ slope was reduced from a control value of $43.6 \pm 14.7$ to $34.0 \pm 10.4$ Hz/nA ($n = 11; P = 0.03$) in the calcium-free bathing solution (Fig. 10, C and D). Spike-frequency adaptation was reduced from $37.7 \pm 18.4$ to $21.0 \pm 11.1\%$ ($n = 11; P = 0.002$) in the calcium-free solution (Table 2). The other parameters of the $F-I$ relationship, $x_0$ and the saturation firing frequency, did not change significantly. There were no significant changes in calcium-free solution for membrane potential, input resistance, action potential properties, rheobase, and spike threshold (Table 2).

**DISCUSSION**

The activity of neuronal networks is a function not only of the synaptic connectivity but also of the cellular properties that determine how neurons transform synaptic inputs into an output of action potentials. In these experiments, intracellular recordings of membrane potential were used to characterize the changes that occur in the electrophysiological properties of lamprey motoneurons when the spinal cord goes from a quiescent state to fictive swimming. The most significant changes observed were a decrease in the slope of the $F-I$ relationship, a decrease in the degree of spike-frequency adaptation, and a decrease in the amplitude of the sAHP. In an attempt to identify possible mechanisms underlying these changes, it was found that serotonin and low-calcium solutions both produced similar changes in these three parameters. These results suggest that the changes in these parameters may be due in part to the endogenous release of serotonin during fictive swimming, with a subsequent reduction in calcium currents.

The only previous thorough characterization of the electrophysiological properties of lamprey spinal neurons was done in the quiescent spinal cord, without the addition of an excitatory amino acid to induce fictive swimming (Buchanan 1993). The quiescent state values reported here for motoneurons are in good agreement with those of the previous study. However, it was necessary to do similar measurements during fictive swimming because network activity represents a significantly dif-
TABLE 2. Values for electrophysiological properties in the presence of serotonin, spiperone, and calcium-free Ringer solution

<table>
<thead>
<tr>
<th>Property</th>
<th>Experiment 1</th>
<th>Experiment 2</th>
<th>Experiment 3</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>C1</td>
<td>SHT</td>
<td>SWIM</td>
</tr>
<tr>
<td>Membrane potential (mV)</td>
<td>–72.1 ± 3.8</td>
<td>–77.1 ± 6.0</td>
<td>–70.4 ± 5.8</td>
</tr>
<tr>
<td>Rheobase (nA)</td>
<td>1.5 ± 1.6</td>
<td>1.5 ± 1.3</td>
<td>2.9 ± 3.0</td>
</tr>
<tr>
<td>Input resistance (MΩ)</td>
<td>18.2 ± 9.3</td>
<td>17.9 ± 9.0</td>
<td>8.9 ± 5.0</td>
</tr>
<tr>
<td>Spike threshold (mV)</td>
<td>–54.9 ± 4.9</td>
<td>–57.5 ± 4.7</td>
<td>–57.7 ± 5.1</td>
</tr>
<tr>
<td>Action potential duration (ms)</td>
<td>19.5 ± 12.1</td>
<td>15.2 ± 11.0</td>
<td>20.7 ± 5.1</td>
</tr>
<tr>
<td>Action potential amplitude (mV)</td>
<td>93.4 ± 13.3</td>
<td>93.4 ± 14.7</td>
<td>98.0 ± 5.6</td>
</tr>
<tr>
<td>F-I slope (a)</td>
<td>43.9 ± 14.0</td>
<td>34.7 ± 10.7</td>
<td>48.1 ± 18.6</td>
</tr>
<tr>
<td>F-I inflection point (μA)</td>
<td>3.0 ± 3.2</td>
<td>2.5 ± 2.4</td>
<td>4.3 ± 3.0</td>
</tr>
<tr>
<td>F-I saturation (a)</td>
<td>89.2 ± 14.5</td>
<td>95.2 ± 16.6</td>
<td>96.9 ± 5.9</td>
</tr>
<tr>
<td>Adaptation (%)</td>
<td>33.4 ± 19.9</td>
<td>21.4 ± 9.3</td>
<td>46.0 ± 14.7</td>
</tr>
<tr>
<td>Slow AHP amplitude (mV)</td>
<td>2.3 ± 1.2</td>
<td>0.5 ± 0.5</td>
<td>2.6 ± 1.4</td>
</tr>
</tbody>
</table>

Values are means ± SD. Numbers in parentheses reprint cells. P represent differences. Significance determined using the Wilcoxon signed-rank test. Experiment 1, control Ringer solution (C1) and after the addition of 5 μM serotonin (SHT). Experiment 2, quiescent (Q), during fictive swimming (SWIM), and during fictive swimming with 10 μM spiperone added (SPIP). Experiment 3, control Ringer solution (C2) and in calcium-free Ringer solution (CaFREE).

In addition, the motoneurons are depolarized compared with quiescence, and they exhibit oscillating membrane potentials, often with spiking during the depolarizing phase (Buchanan and Cohen 1982; Buchanan and Kasicki 1995). In addition, the motoneurons receive much greater levels of both excitatory and inhibitory synaptic inputs during fictive swimming (Kahn 1982; Russell and Wallén 1983). Finally, any neuromodulators that are selectively released during fictive swimming may be acting to change the properties of ion channels and other cellular processes.

Despite these altered conditions of network activity compared with the quiescent state, there were relatively few significant changes in the electrophysiological properties of the population of sampled motoneurons, and these changes were modest in magnitude. For example, no changes were observed in spike threshold or in action potential amplitude and duration. No attempt was made here to assess significant changes in individual motoneurons due to the inherent difficulties associated with the experiments (see METHODS). Thus it remains possible that individual changes were not indicated in the population statistics. Using population statistics could also obscure possible sub groupings in parameter changes, e.g., one subgroup of motoneurons might always have an increase in a particular parameter while another subgroup of motoneurons would always have a decrease in that parameter. If such subgroups existed, one might expect correlations to changes in other parameters as well. However, no such subgroups or correlations among parameters were observed in the data. There did not appear to be any properties in which the changes were clearly divided into subsets, in which groups of cells showing relatively large changes in opposing directions made the mean change insignificant. Also, any cells whose changes were not indicative of the population mean did not show a correlation to changes in any other property.

Possible mechanisms of change

The motoneurons become depolarized by about 10 mV during the excitatory phase of the fictive swim cycle compared with the quiescent membrane potential. It is possible that this level of depolarization alone is sufficient to induce some of the observed changes in electrophysiological properties by altering voltage-dependent ion channels. In an attempt to assess the contribution of depolarization alone to the observed changes, two strategies were used. First, the measurements of the properties during both the excitatory phase and the inhibitory phase of fictive swimming were compared with the quiescent state because the membrane potential in the inhibitory phase was not significantly different from the quiescent membrane potential. The reduction in the slope of the F-I relationship was present in both swim phases, suggesting that membrane potential alone could not account for this change. While spike-frequency adaptation did decrease in both phases of swimming, it was only significant in the excitatory phase. For the amplitude of the sAHP, there was no significant
change during the excitatory phase compared with quiescence despite the greater driving force due to the depolarization of the membrane. This was supported by the observation of a significant reduction of sAHP amplitude in the inhibitory phase, when the driving force was not significantly different from in the quiescent state. This indicates that there was either a decrease in the overall resistance of the cell or specifically in the sAHP conductance. Since the resistance of the motoneurons increased or showed no change during swimming, there must have been a decrease in the conductance of the sAHP. The second strategy was to impose depolarization on quiescent neurons to a similar level as observed during the excitatory phase of fictive swimming. The depolarizations alone did not produce a significant change in the F-I slope but did significantly decrease spike-frequency adaptation. Imposed depolarization increased the amplitude of the sAHP to a greater degree than that observed during the excitatory phase of swimming. This was supported by the observation of a significant reduction of sAHP amplitude in the inhibitory phase, when the driving force was not significantly different from in the quiescent state. This indicates that there was either a decrease in the overall resistance of the cell or specifically in the sAHP conductance.

FIG. 8. Serotonin reduces the sAHP amplitude and the slope of the F-I relationship. A: sAHP amplitude of individual motoneurons before (control) and after (5-HT) bath application of 5 μM serotonin (5-hydroxytryptamine, 5-HT). B: changes in the slope of the sAHP amplitude versus Vm relationship in 8 motoneurons. C: example of the change in the F-I relationship with the addition of serotonin. In this motoneuron, the slope of F-I relationship was reduced from 29.8 Hz/nA (5-HT, gray triangles) to 23.0 Hz/nA (5-HT, black circles).

FIG. 9. Spiperone partially reversed the swimming-induced reduction in the sAHP amplitude but had no significant effect on the slope of the F-I relationship. A: sAHP amplitude was measured at several membrane potentials in quiescence, swimming, and swimming with spiperone, and data points were fitted with a linear regression. Slope of the regression decreased from 0.36 to 0.12 when the cell went from quiescence to swimming and then increased to 0.25 after spiperone was added. B: sAHP amplitude of 9 motoneurons during swimming in the absence (swim) and presence (+spiperone) of spiperone. Symbols with error bars represent the mean ± SD of the amplitudes for the 9 motoneurons. There was no significant change in membrane potential, so the amplitudes may be directly compared. C: slope of the F-I relationship was reduced from 42.9 Hz/nA in quiescence (black circles) to 20.0 Hz/nA during swimming (light gray triangles). Spiperone partially reversed this change (dark gray squares), bringing the slope to 27.4 Hz/nA. D: F-I slopes in the 9 motoneurons exposed to spiperone. *P ≤ 0.05, signed-rank test.
fictive swimming but did not change the input resistance, again showing that there had been a decrease in the underlying conductance of the sAHP during fictive swimming. These results suggest that the reductions in the slope of the F-I relationship and the amplitude of the sAHP were not due solely to the depolarization associated with fictive swimming, but the case for spike-frequency adaptation is less clear.

It is also possible that the activation of excitatory and inhibitory neurotransmitter ionotropic receptors contributed to some of the observed changes. The membrane potential oscillations themselves are due to phasic activation of glutamate and glycine receptors (Buchanan 1982; Buchanan and Grillner 1987; McPherson et al. 1994; Russell and Wallén 1983). Activation of non-NMDA glutamate receptors and glycine receptors would be expected to reduce the input resistance of the neurons. However, an increase in input resistance during fictive swimming was observed. It is possible that activation of NMDA receptors by glutamate may have contributed to the increase in input resistance due to the voltage dependence of the NMDA channel. The resulting negative-slope conductance can produce an apparent increase in input resistance over certain membrane potential ranges (Moore and Buchanan 1993). Alternatively, resting or leak membrane conductances may be inactivated during locomotion, thereby producing the increase in resistance.

The activation of NMDA receptors can induce oscillatory membrane potentials in the presence of tetrodotoxin (TTX) (Wallén and Grillner 1985), and these NMDA-induced, TTX-resistant potentials contribute to the shape of the membrane potential oscillations during fictive swimming (Wallén and Grillner 1987). If any of the changes observed in this study were due to NMDA-mediated conductances, then one would expect a difference in the measurements made in the excitatory versus inhibitory phases that cannot be reproduced by imposed depolarization of the membrane. For example, the increased calcium conductance via NMDA channels would be expected to increase the sAHP amplitude in the excitatory phase, but not in the inhibitory phase due to the voltage-dependent block of the NMDA channels. Rather, the sAHP amplitude decreased in both phases of locomotion, suggesting that this change is not the result of NMDA receptor activation. With the exception of input resistance, all changes observed here were either present in both phases of the cycle or were reproducible with imposed depolarization. For rheobase, the decrease during the excitatory phase of swimming could be reproduced by imposed depolarization, but the depolarization-induced change was only 50% of the magnitude of the quiescent-swim change. Thus the possibility that NMDA-mediated plateau potentials may have some contribution cannot be excluded.

Another possible mechanism underlying the observed parameter changes during fictive swimming is neuromodulation. Neurotransmitters acting via metabotropic receptors can alter the properties of ion channels and is well documented in a variety of neuronal networks (Harris-Warrick and Marder 1991). One of the best-studied neuromodulators in lamprey is serotonin, which is known to act both pre- and postsynaptically (Buchanan and Grillner 1991; Takahashi et al., 2001; Wallén et al. 1989). As stated previously, it has been shown that serotonin reduces the sAHP in lamprey neurons (Van Dongen et al. 1986), and this is consistent with the reduction of the sAHP amplitude observed here during fictive swimming. The effect of serotonin on the F-I slope had not been previously documented in lamprey motoneurons. Unexpectedly, serotonin reduced the slope of the F-I relationship. On the basis of the reduction of the sAHP amplitude alone, one would predict an increased F-I slope, the effect observed with selective blockade of the sAHP with apamin (Meer and Buchanan 1992). The reduction of the F-I slope by serotonin corresponds to the reduction of the F-I slope observed during fictive swimming. Applied serotonin also reduced spike-frequency adaptation, a change similar to that observed during fictive swimming. To
test the possibility that endogenously released serotonin was producing these observed changes, spiperone was applied during fictive swimming. In lamprey, it has been shown that spiperone blocks the reduction of the sAHP produced by serotonin (Wikström et al. 1995) and also speeds fictive swimming (Zhang and Grillner 2000), opposite the effect of applied serotonin on fictive swimming (Harris-Warrick and Cohen 1985). While spiperone produced a significant increase in sAHP amplitude during swimming, it produced only a partial recovery of the amplitude compared with quiescence. Further, it had only a slight but not significant effect on the slope of the F-I relationship. This would suggest either that spiperone is not adequate to block the relevant serotonin receptors or that other neuromodulators or factors are involved. Dopamine is another candidate modulator in the lamprey spinal cord, known to alter neuromodulators or factors are involved. Dopamine is another candidate modulator in the lamprey spinal cord, known to alter

Comparison to other vertebrates

Similar studies examining the changes in motoneuron properties during fictive locomotion have been done in the cat. Before these are discussed, it is important to note that the method used to invoke locomotor activity is different in the present study. In this study, locomotion was induced by pharmacological addition of glutamate, whereas in cats brain stem stimulation is usually used. Brain stem stimulation was not used here; the short bouts of swimming that can be generated are not adequate for these experiments. Thus there may be different pathways activated in these two models. It is also important to note that bath glutamate may affect the membrane properties of the cells and thus produce changes in these experiments that are not seen in the cat model.

In these studies, Brownstone et al. (1992) found that the slope of the F-I relationship is reduced in fictive locomotion compared with the quiescent state. In the extreme cases, the F-I slope approached zero, and the firing rate was virtually independent of the membrane potential. Later experiments demonstrated that the F-I relationship during the hyperpolarized phase of fictive locomotion was similar to the quiescent state (Fedirchuk et al. 1998), and it was suggested that plateau potentials occurring during the excitatory phase were responsible for the flattening of the F-I relationship. In lamprey there were much smaller decreases in F-I slope, and these reductions were present in both the excitatory and the inhibitory phase. One important difference to note is that lamprey motoneurons do not show the same type of plateau potentials as demonstrated in cat motoneurons, which can exhibit self-sustained firing that is triggered by a short excitatory input and terminated by a short inhibitory input (Crone et al. 1988; Hoffer et al. 1987). This bistability keeps the membrane potential constant despite various levels of synaptic (or injected) current. Lamprey motoneurons do not show this type of bistability and thus the membrane potential is affected by the summation of synaptic input. Because of this, the firing rate would tend to be unresponsive to changes in current injection in cats (while the plateau potentials are activated) and thus the reduction in the F-I slope much more dramatic. One could speculate that in cat the firing rate during locomotion is controlled by the properties of the cells themselves, but in lamprey it is controlled by the amount of synaptic input.

The amplitude of the sAHP in cat was reduced during fictive locomotion, but to a greater degree during the excitatory phase of fictive locomotion compared with the inhibitory phase (Brownstone et al. 1992; Schmidt 1994). In lamprey there was also a reduction of sAHP amplitude but no clear difference in the degree of reduction of the sAHP amplitude in the two phases.
of fictive swimming was observed. In cat it was found that spike threshold decreased during both excitatory and inhibitory phases of fictive locomotion (Krawitz et al. 2001), while in lamprey no significant changes in spike threshold were observed. In cat most motoneurons showed either a decrease or no change in input resistance during fictive locomotion (Gosgnach et al. 2000; Shefchyk and Jordan 1985), whereas lamprey motoneurons increased their resistance. In Shefchyk and Jordan’s study, 28 of 52 motoneurons showed no change in input resistance, which was similar to this study in that the result was not the expected decrease in resistance due to the synaptic activity during locomotion. However, the reason for the discrepancy of the results in this study versus those in cats is currently unknown.

Serotonin is well established as a neuromodulator that affects vertebrate locomotor activity (Schmidt and Jordan 2000). In the cat, serotonin has been shown to increase step length and the duration and amplitude of hindlimb electromyographic activity (Barbeau and Rossignol 1991). In lamprey and frog tadpole, serotonin has comparable effects of lengthening cycle period and increasing duration and amplitude of ventral root bursting during swimming activity (Harris-Warrick and Cohen 1985; Woolston et al. 1994). In turtle and cat motoneurons, serotonin facilitates the expression of plateau potentials (Crone et al. 1988; Hounsgaard et al. 1984, 1988; Hounsgaard and Cohen 1988). In cat most motoneurons showed either a decrease or no change in input resistance, which was similar to this study in that the result was not the expected decrease in resistance due to the synaptic activity during locomotion. However, the reason for the discrepancy of the results in this study versus those in cats is currently unknown.

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In summary, some electrophysiological properties of lamprey motoneurons change as the locomotor network goes from quiescence to an active state. Overall, the changes increase the excitability of the neurons. While the mechanisms for these changes are not understood, it is clear that depolarization alone cannot account for them and that release of neuromodulators during fictive swimming is a likely candidate for the observed changes. Serotonin, which is known to be released during fictive swimming, can reproduce some of the changes and may act by reducing calcium currents.

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