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Recruitment of reticulospinal neurones and steady locomotion in lamprey

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Abstract

In lamprey, the supraspinal control of velocity is mainly accomplished by the reticulospinal (RS) system. During locomotion, RS neurones are rhythmically active with a cycle duration corresponding to the duration of the swim cycle. While the velocity of the muscular contraction wave changes as swimming velocity changes, the conduction velocity of RS axons remains constant. Thus, an action potential generated during a specific phase of the swim cycle will, depending on swimming velocity, provide input to a particular downstream segment during different phases of its rhythmic activity. In order to investigate the importance of this effect for the control of locomotion, the temporal and spatial characteristics of the propagation of the population of action potentials along RS axons in the spinal cord were investigated. The results suggest that if RS neurones are recruited independently of their sizes and conduction velocities, a phasic wave of action potentials in these fibers will reach some segments during the inhibited phase of their rhythmic activity. Such an input could hinder a smooth propagation of the contraction wave and disrupt swimming. In contrast, by recruiting successively larger and hence more rapidly conducting neurones for successively more rapid swimming, the phasic wave of action potentials may propagate with the same velocity as that of the muscular contraction wave. Under such conditions, reticulospinal activity would support and stabilise locomotion.

Keywords: Conduction velocity; Lamprey; Locomotion; Motor control; Reticulospinal system; Size principle

1. Introduction

In the lamprey, a phylogenetically ancient vertebrate, the reticulospinal (RS) system forms the main pathway conveying information from brain to spinal cord. This pathway plays a key role in the control of locomotion, in particular in the control of swim velocity. Whereas the anatomy and synaptic physiology of RS neurones is fairly well characterised (Brodin et al., 1988), the mechanisms underlying the generation and control of specific movements remain unknown. The control of swim velocity, in particular, poses an interesting problem which arises from three factors. First, the RS neurones are phasically active during locomotion (Kasicki and Grillner, 1986; Kasicki et al., 1989). Second, the conduction velocity of individual RS axons remains nearly constant over the entire extent of the cord (Rovainen, 1982). Consequently, a spike generated in the soma of a RS neurone reaches a particular spinal segment S_n with a constant delay. This delay depends only on the conduction velocity of the RS neurone and on the distance between the soma and S_n. Third, during constant velocity swimming, a wave of muscular contraction propagates with a constant velocity along the body and the swim velocity is determined by the velocity of that contraction wave. In other words, the contraction wave starting in the first segment (S_1) reaches the nth segment (S_n) with a delay depending on the swimming velocity and on the distance between S_1 and S_n.

As a consequence, action potentials generated in one RS neurone in the same phase during a slow or rapid swim cycle reach a caudal segment S_n, while its target neurones have reached a different state in their rhythmic activity. Therefore, the excitatory drive provided to the segment S_n may support or counteract the generation of regular rhythmic activity.

A qualitative model for the control of the swimming velocity accounting for these observations has been proposed by Kasicki et al. (1989). The model asserts that during swimming RS neurones generate bursts of action potentials which are phase-locked with the swim cycle and which alternate between both sides of the brain. Since the distance to S_1 is small, these bursts are still coherent when passing

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the first segments.This provides a phasic input to the rostral spinal cord. More caudally, differences in the conduction velocity of the RS axons induce a dispersion of the action potentials, thereby providing a tonic input to middle and caudal segments. According to Kasicki et al. (1989) this tonic input increases the excitation level of the caudal segments and activates the local networks for locomotion. In addition, the inputs received from rostral segments via intersegmental connections pace the rhythmic activity of the motoneurones. The characteristics of the spinal circuits interconnecting subsequent segments cause the contraction wave to propagate with constant velocity along the whole body.

In this study we investigate the influence of the RS neurones onto steady locomotion in the lamprey and find that, in contrast to Kasicki's suggestion (Kasicki et al., 1989), if the recruitment of RS neurones is independent of their conduction velocity, a phasic wave of action potentials propagates along the whole spinal cord. This phenomenon is particularly significant for slow swimming velocities, because the tonic activity building up caudally remains proportionnally small and because the phasic wave of action potentials propagates more rapidly than the muscular contraction wave, rapidly reaching segments in which their target neurones are passing through the inhibited portion of their activity cycle and providing them with an excitatory input. This situation is likely to disrupt the smooth propagation of the muscular contraction wave. In contrast, we found that a recruitment by size of the RS neurones would prevent such a perturbation. A recruitment by size implies that larger and thus faster RS neurones would be recruited at higher swimming velocities. In this situation, the propagation velocity of the phasic wave of RS action potentials increases with increasing swimming speed. We show with a model based on biological data that the recruitment could be tuned in such a way that both the phasic wave of RS activity and the wave of muscular contraction propagate with the same speed along the lamprey. In this case, the phasic component observed for all swimming speeds would stabilize the propagation of the muscular contraction wave.

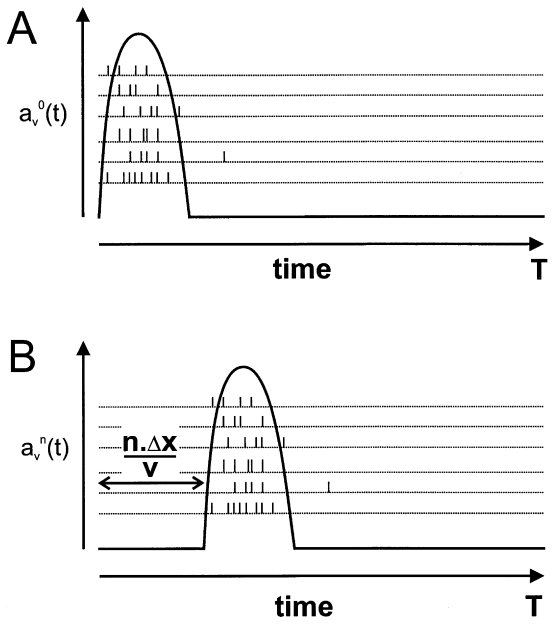

Fig. 1. (A) For a group of RS axons of conduction velocity v, the function $a_v^0(t)$ gives the total number of action potentials generated in the reticular formation as a function of time. The dotted lines schematise the activity of six imaginary RS neurones of conduction velocity v during one swim cycle, each vertical bar symbolising the occurrence of an action potential. (B) For the same group of neurones, the function $a_v^n(t)$ gives the total number of action potentials crossing segment S_n as a function of time. The shape of the function is the same as for the first segment, but the action potentials arrive with a delay $(n\cdot\Delta x)/v$. Here, v is the conduction velocity of the RS neurones and $n\cdot\Delta x$ is the distance between the reticular formation (assumed to be at the place of the 0th segment) and S_n.

2. The model

The following assumptions based on biological observations were made on the RS activity. During swimming at constant velocity, the firing behaviour of a RS neurone is rhythmic with a cycle duration equal to the swim cycle T. The motoneurones are known to be primarily active during one third of each swim cycle, independently of its duration T (Wallén and Williams, 1984). Most often, we assumed the same activity pattern for the RS neurones, but other patterns were tested as well. Let us denote by $a_v^o(t)$ the total number of action potentials generated at the reticular formation at time t and propagating caudally with velocity v (cf. Fig. 1A). The scaling property of the RS-activity is expressed by writing it in the form $a_v^o(t) = a_v(t/T)$ with a periodic function $a_v(\tau)$ with period 1. The variable $\tau = t/T$ represents the relative time within the swim cycle. The activation functions $a_v(\tau)$ are assumed to be convex on the interval [0, 1/3] and vanishing on the interval [11/3, 1] (see Fig. 2). The conduction velocity of RS neurones ranges from $v_0 = 0.1$ to $v_1 = 3$ m/s, and for each neurone this velocity is constant along the whole extent of the spinal cord. We assumed a body length of $L = 0.3$ m corresponding to the typical size of adult specimens of *Lampetra fluviatilis*. The spread of RS-activity will be compared with the spread of the muscular contraction wave which is supposed to be a single peak of activity propagating with swimming speed along the spinal cord.

2.1. Spread of the reticulospinal activity

The action potentials generated by the RS neurones travel along the segments $S_1,\ldots,S_N$ $(N = 100)$ and the total number of action potentials with conduction velocity v encountered in segment n at time t is $a_v^n(t) = a_v^o(t - n\cdot\Delta x/v)$, where Δx $(= 3$ mm) is the unit lenght of a segment. In our simulation we assumed 300 groups of RS neurones with equally distributed velocities from v_0 to v_1. The total number of action potentials in segment n at time t is the sum of the group

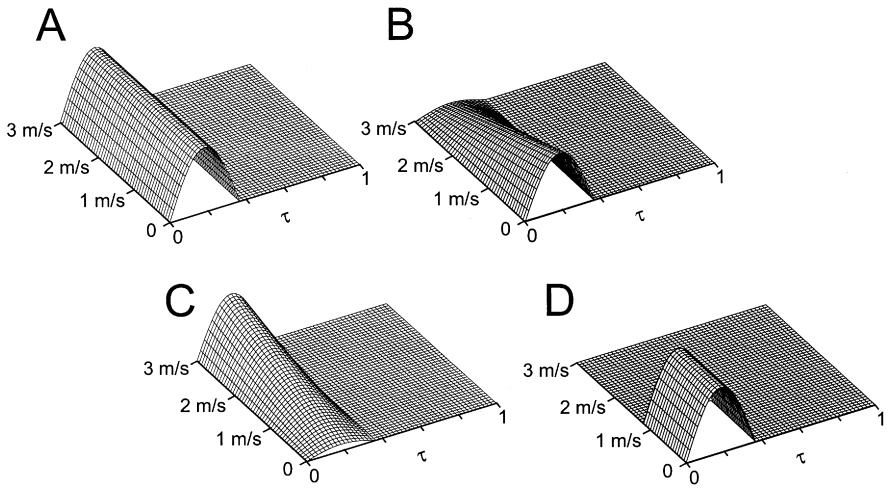

Fig. 2. Examples of activity functions $a_v(\tau) = a_v^o(t)$ studied. Each surface fully describes the RS activity in the first segment as a function of the relative time $\tau = t/T$ within a swim cycle of duration T. (A) Positive half of a sine function with an activity duration of one third of the cycle duration. The same firing activity is given to all RS groups. (B) As in (A), except that the slowly conducting neurones produce more action potentials than the rapidly conducting groups of RS neurones. (C) As in (A), except that the rapidly conducting neurones produce more action potentials than the slowly conducting groups of RS neurones. (D) As in (A), except that only the neurones with a conduction velocity less than or equal to 1.2 m/s were recruited.

activities,

$$A^n(t) = \sum_{v=v_0}^{v_1} a_v^n(t) = \sum_{v=v_0}^{v_1} a_v^o\left(t - \frac{n\cdot\Delta x}{v}\right),$$
$$(n = 1, \ldots, N; t \in [0, T]). \quad (1)$$

With respect to the relative time $\tau = t/T$ the total RS-activity in the nth segment is expressed by $A^n(\tau) = \Sigma a_v[\tau - n\cdot\Delta x/(Tv)]$, where the sum is taken over all the discrete velocities from v_0 to v_1 and where a_v without superscript represents the RS-activity with respect to the relative time. To analyze this model we replace the discrete segments by a spectrum of continuous positions x ranging from the location of the first segment, Δx, to the tail of the lamprey at $L = N\cdot\Delta x$. Moreover, the RS groups are described by a continuous range of axonal conduction velocities v. The activity at location x and relative time τ is then given by

$$A^x(\tau) = \int_{v_0}^{v_1} a_v\left(\tau - \frac{x}{Tv}\right)\mathrm{d}v, (x \in [\Delta x, L], \tau \in [0, 1]), \quad (2)$$

with $a_v(\tau)$ as defined above. Note that at a given site x there may be activity of previous swimming cycles contributing to the present total activity. This is the case for fast swimming (small T) and slow conduction velocity (large x/v) for which the argument of the activation functions may be negative, i.e. $\tau - x/(Tv) < 0$. We shall see that it is exactly this superposition of slowly propagating RS action potentials which may lead to a tonic component at high swimming speed in the lamprey's tail, while for slow swimming speeds there is less superposition and thus a phasic component predominates.

Next we want to estimate for each location x the relative time $\tau^x_{\max}$ within the swim cycle when the total RS activity $A^x(\tau)$ is maximal. This time is physiologically relevant, since it determines when neurones at segment x have the highest chance of being activated by the descending RS-activity. Unfortunately, $\tau^x_{\max}$ is not a linear function of the location x. Nevertheless, $\tau^x_{\max}$ is nearly linear for large x and one can estimate (see Appendix A)

$$\tau^x_{\max} \approx \frac{x}{T\cdot v_1} + \frac{1}{6}\left(1 + e^{-\alpha\frac{Tv_1}{x}}\right) \quad (3)$$

with $\alpha > 0$ and depending on the shape of the functions $a_v(\tau)$. According to this formula the peak activity propagates towards the tail with nearly constant velocity, namely with $x/(T\cdot(\tau^x_{\max} - 1/6)) = v_1$. Thus, the propagation velocity of the maximum RS activity corresponds to the highest axonal conduction velocity v_1 of the activated RS neurones. Slower RS groups do only have a transient influence onto the propagation speed of the peak activity. While this result was deduced for a homogeneous distribution of propagation speeds between v_0 and v_1, the simulations of Eq. (1) reveal that this remains approximately true even if the relative participation RS neurones changes with v (cf. Figs 5 and 6).

2.2. Interference of reticulospinal and muscular activity

The wave of RS-activity $A^x(\tau)$ propagating along the lamprey must now be compared to the wave of muscular contraction $M^x(\tau)$ propagating in the same direction, but not necessarily with the same speed, along the body. Behavioural studies have shown that a single wave of muscular contraction propagates with constant velocity on one side of the body per swim cycle, and that the time needed to travel from the head to the tail corresponds to the duration of the swim cycle (Wallén and Williams, 1984). At the 'beginning' of the swimming cycle ($\tau = 1/6$) the muscular activity reaches a peak at $x = 0$ and propagates with swimming

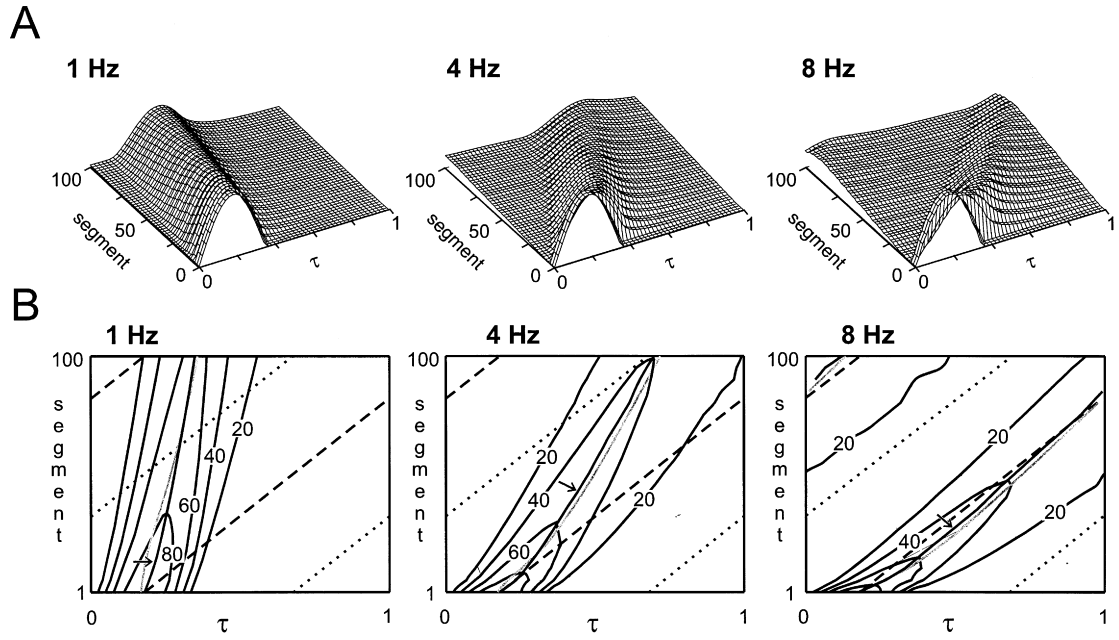

Fig. 3. Propagation of action potentials along RS axons for three swimming velocities. The firing function $a_v(\tau)$ assigned to all groups was the positive half of a sine function lasting one third of the cycle duration, and had the same amplitude for all groups [see Fig. 2A]. (A) Propagation at swim frequencies of 1, 4 and 8 Hz. (B) Contour plots of the data presented in (A). The isoclines indicate levels where the activity reaches 20, 40, 60 or 80% of the maximal value. An arrow points to the curve following the maximum of the wave, and a grey line indicates the estimated position of the maximum using $\alpha = 0.06$ in Eq. (3). Dashed lines: maximum of the muscular contraction wave propagated on the side ipsilateral to the RS axons simulated. Dotted lines: maximum of the inhibited period in the activity cycle of spinal neurones positioned in the hemisegments ipsilateral to the reticulospinal axons simulated.

velocity towards the end at $x = L$. With respect to the normalized time the peak muscular activity reaches the point x at $\tau = x/L + 1/6$. We thus have for the muscular activity $M^x(\tau) = 1$ if $\tau = x/L + 1/6$ and $M^x(\tau) = 0$ else. In the $x - \tau$ diagram the propagation of the peak muscular activity corresponds to a straight line from $(\tau, x) = (1/6, 0)$ to $(\tau, x) = (1 + 1/6, L)$, where τ has to be taken modulo 1 (cf. Fig. 3). The propagation velocity of the muscular activity is L/T which corresponds to 0.3 m/s at a swimming frequency of 1 Hz. On the other hand, Eq. (3) states that the propagation velocity of the RS peak activity roughly corresponds to the fastest RS group which is activated, i.e. to $v_1 = 3$ m/s.To exclude interference between the two activities these two velocities should be the same. One way to achieve equality is to recruit the fast RS groups only for fast swimming. According to Eq. (3), a swimming speed v_s would require the recruitment of the RS groups up to a conduction velocity of v_s. Only then can the coherency between the RS peak activity and muscular peak activity be maintained. We, therefore, postulate a size principle governing the RS neurones at any swimming spead and requiring that all RS neurones with axonal propagation speed up to that swimming speed v_s are recruited.

2.3. Simulation results

Since no data are currently available on the activity of RS neurones during free swimming in lamprey, the spatiotemporal characteristics of the propagation of action potentials along the spinal cord was investigated for different hypothetical patterns of RS neurone activities. Some of the functions $a_v(\tau)$ investigated are depicted in Fig. 2. The parameters which were varied were time course [half sine wave (Fig. 2), square wave (not shown),...] and duration in proportion to the cycle (e.g. 33% or 50%). All groups of RS neurones were considered to receive a similar synaptic input during swimming and to fire with a function having the same time course. In order to account for a differential participation of the various groups of RS neurones, due for instance to differences in the number of neurones belonging to each group, the relative amplitude of the activity function of each group could be scaled (Fig. 2B, C).

The amplitude of the firing function is determined by two factors: first, by the number of neurones in each group, and second, by the activity of each neurone. Because these parameters are unknown, the units for the amplitude of the function are arbitrary, the relative amplitude between RS populations of different conduction velocity reflecting the relative participation of each population to the wave of RS action potentials propagating along the spinal cord.

We will first consider the case in which the firing activity $a_v(\tau)$ was set to the positive half of a sine function lasting for the first third of the cycle followed by a silent period for the rest of the cycle and in which all RS groups produced the same number of action potentials (Fig. 2A). Fig. 3A depicts the total number of action potentials $A^n(\tau)$ encountered along the cord in relation to the time in the swim cycle for three swimming frequencies. All calculations were

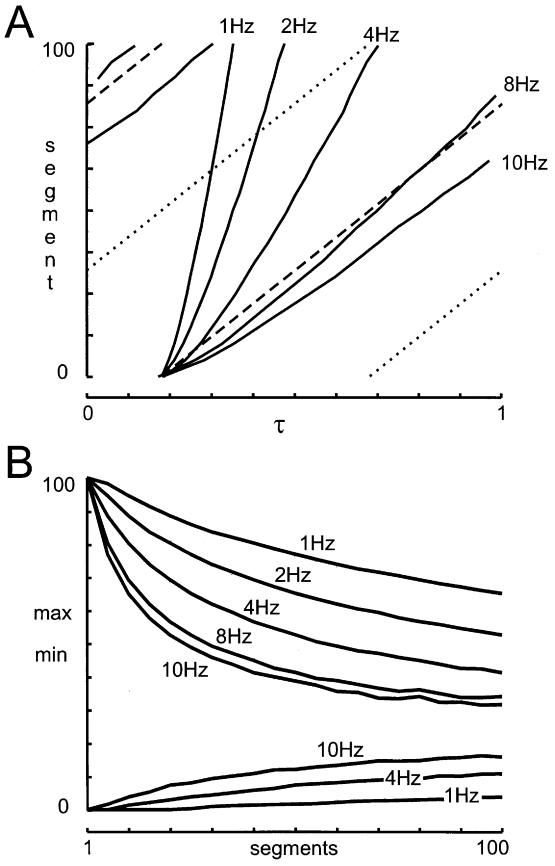

Fig. 4. (A) The peak propagation of the wave of action potentials for different swimming velocities is indicated by continuous lines. Note that during slow swimming (1 and 2 Hz), the RS wave of action potentials crosses segments which are inhibited on the ipsilateral side (dotted lines), and that for a swim frequency of 8 Hz, the peak of the wave of action potentials propagates with the same velocity as the muscular contraction wave (dashed lines). (B) Relative maximal and minimal number of action potentials crossing a given segment for various swim frequencies.

performed by means of Eq. (1) and the coordinates were then scaled by the cycle duration and the body length, respectively. It can be seen that the action potentials cross the first segments in a phasic wave in both slow and rapid swimming conditions. A more complicated pattern evolves in the caudal segments, where the time course with which action potentials cross the segments strongly depends on the swim velocity. At a swim frequency of 8 Hz, the characteristics of the function becomes complex, consisting of a small phasic wave superimposed on tonic activity.

The same results are shown with contour plots in Fig. 3B. The isoclines depicted in the graphs indicate where along the cord and at which time of the cycle 20, 40, 60 or 80% of the maximal number of spikes are encountered. From these figures, it can be seen that for both slow and rapid swimming, a phasic wave of action potentials propagates from rostral to caudal segments. In order to characterise the propagation of this phasic wave for different swim velocities, the time $\tau^{x}_{\max}$ of the swim cycle when the maximum number of action potentials is encountered in each segment and the evolution of the amplitude of the wave as it propagates along the cord were investigated. The peak of the phasic wave propagates with an almost constant velocity corresponding to the fastest recruited RS group along the cord (Fig. 4A). Given in segments per cycle, the velocity of the phasic wave of action potentials can easily be compared with the propagation of the muscular contraction along the body which in these units is maintained at 100 segments per cycle for all swim velocities. For swim frequencies below 8 Hz, the propagation velocity of the phasic wave of action potentials is greater than that of the contraction wave, having a value of 748 segments per cycle at 1 Hz, 347 segments per cycle at 2 Hz and 201 segments per cycle at 4 Hz. At 8 Hz, both waves propagate with the same velocity of 100 segments per cycle. If the lamprey swims at higher frequencies, the propagation velocity of the wave of action potentials will then be less than that of the muscular contraction, being 83 segments per cycle at 10 Hz.

As a consequence of the velocity differences between the waves of muscular contraction and of action potentials, the peak of the wave of action potentials may reach segments at a time when the hemisegments crossed by the RS axons bearing the action potentials are in the inhibited period of the swim cycle (Fig. 4A).

In Fig. 4B, the relative maxima and minima of the number of action potentials $A^{n}(\tau)$ met at each segment is depicted for various swimming frequencies. For low swimming frequencies, the maxima remain proportionally high for the entire extent of the cord. For instance, for swimming at 1 Hz, in the last segment the maximum number of action potentials is still maintained at 65% of the value encountered in the first segment. On the other hand, the minima are kept low along the whole cord, reaching only 3% of the maximal activity in the last segment.

For a high swimming frequency, the situation changes considerably. For instance, at 8 Hz the maxima decrease rapidly in intensity over the first third of the cord, reaching 51% of the value of the first segment in segment 30. More caudally, the maxima fluctuate in a narrow interval of values, and reach 43% of the maximum value of the first segment in the last segment. At the same time, the minima increase from rostral to caudal, reaching up to 11% of the maximal activity. The presence of relatively high minima in the last segments paralleled by relatively low maxima indicates that for high swimming frequencies the caudal portion of the spinal cord receives a tonic input on which a phasic wave of proportionally smaller amplitude propagates.

2.4. Differential participation of the various neuronal groups

In the previous section, all groups of RS neurones sharing the same conduction velocity were considered to fire with the same intensity. To investigate the consequences of a differential participation of RS neurones of various conduction velocity, the amplitudes of the firing functions of the

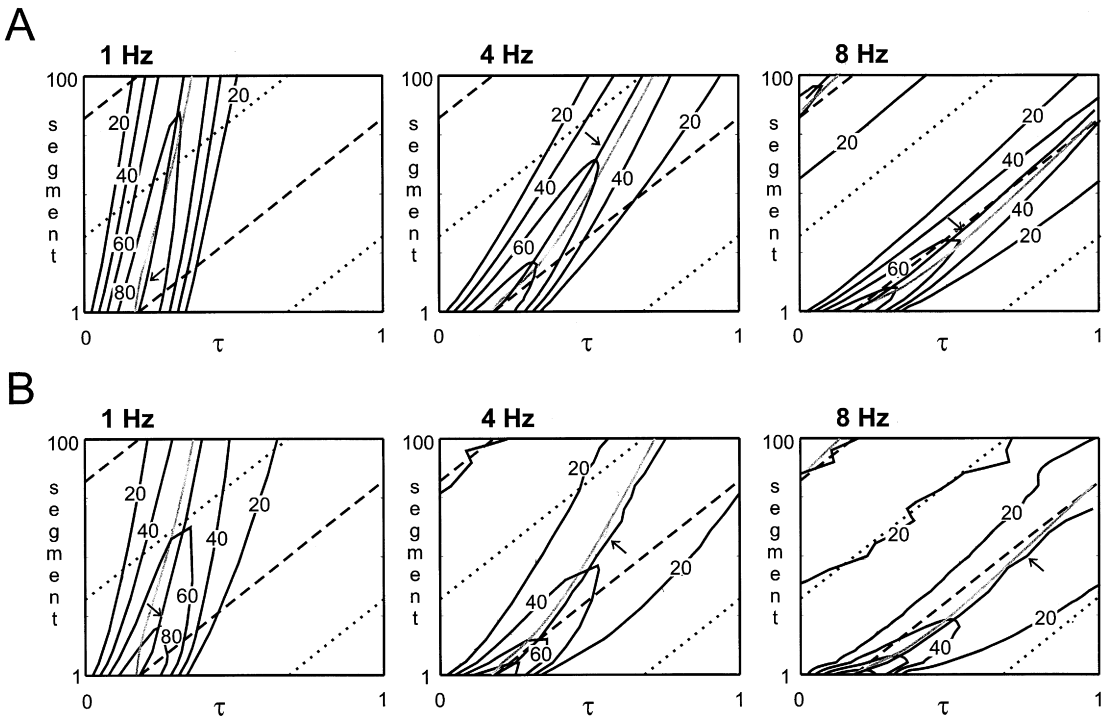

Fig. 5. Same plots as in Fig. 3B, but with RS-activity functions depending on the conduction velocity *v*. (A) Propagation of action potentials along the cord when the relative participation of fast vs slow conducting neuronal groups was differentially weighted. In this example, the group conducting with a velocity 3 m/s was 290 times more active than the group conducting with a velocity of 0.1 m/s [see Fig. 2C]. Note that the action potentials propagate in a well characterised wave for all swimming frequencies, providing a phasic input to all segments. (B) Propagation of action potentials along the cord when the amplitude of the firing functions was increased linearly from fast to slow conducting neuronal groups. In this example, the group conducting with a velocity 0.1 m/s being 290 times more active than the group conducting with a velocity of 3 m/s [see Fig. 2B]. Note that for slow swimming frequencies, the action potentials propagate as a well characterised wave. For rapid swimming, the wave is well characterised for the first segments, but rapidly blurs out; the activity in caudal segments is then rather tonic. Same figure composition as in Fig. 3B; $\alpha = 0.06$.

different cell groups were differentially weighted. The relative amplitudes of the firing functions were linearly varied in relation to the conduction velocity, the proportionality factor between the maximum activity of the slowest to that of the fastest RS group set to up to 300 (Fig. 2B, C). A comparison of these results with those described in the previous sections indicate that if the total number of action potentials produced by rapidly conducting neurones is higher than that produced by slowly conducting neurones (Fig. 2C), the phasic character of the wave of action potentials becomes more explicit for all swimming velocities (Fig. 5A). The relative decrease of the wave amplitude along the spinal cord becomes less pronounced (Fig. 6A), but the velocity remains nearly the same (Fig. 6B). Conversely, if the total number of action potentials produced by rapidly conducting neurones is lower than that produced by slowly conducting neurones (Fig. 2B), the phasic character of the wave of action potentials tends to blur for all swimming velocities, particularly in the caudal portion of the cord at high swimming frequencies (Fig. 5B). The relative decrease of the wave amplitude along the spinal cord becomes more pronounced (Fig. 6A), but in this case too, the velocity remains nearly constant (Fig. 6B).

2.5. Firing functions with different time course

The influence of the time course of the firing function on these results was studied by using different functions corresponding to different firing behaviour of RS neurones. The firing functions $a_v(\tau)$ studied were: (a) the positive half of a sine function lasting 50% of the cycle, while no action potentials are fired during the rest of the cycle; (b) an early phasic activity followed by a tonic activity (respectively 33, 63, 89 and 96% of the action potentials during the first 10, 20, 36 and 50% of the swim cycle), and (c) square waves lasting 36 or 50% of the cycle. The results were basically similar to those described above.

2.6. Recruitment by size

The results presented above suggest that in the absence of a recruitment of specific categories of RS neurones for different swim velocities, a phasic wave of action potentials propagates along the cord with a velocity which often differs from that of the muscular contraction wave (Fig. 4A). The excitatory input thus provided may disrupt the regular propagation of the muscular contraction wave by activating spinal neurones in the inhibited period of their rhythmic activity.

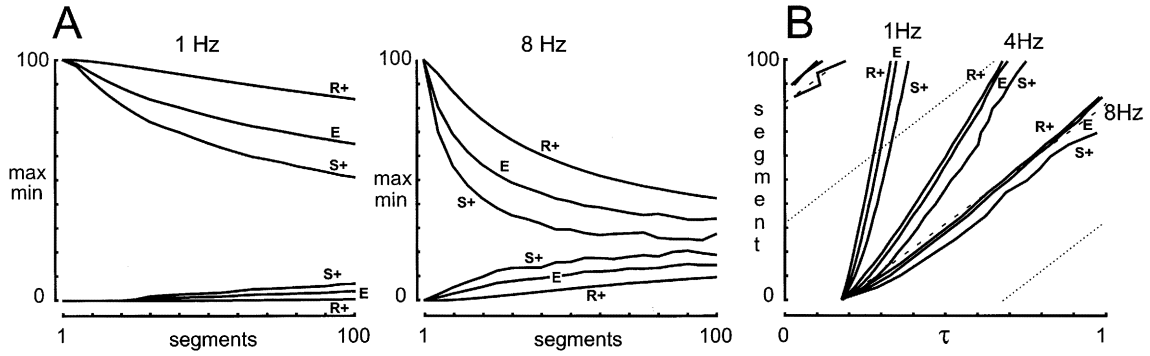

Fig. 6. Different decays of maximal RS-activity along the cord. (a) For slow swimming (1 Hz), the maximal amplitude of the wave of action potentials remains high along the whole cord, and virtually no tonic background activity is present in caudal segments. For rapid swimming (8 Hz), the amplitude of the wave decreases rapidly along the first third of the cord, and the caudal segments receive high background activity during the whole cycle duration. The firing activity for the groups of rapidly conducting neurones was higher (R +), equal (E) or lower (S +) than that of the groups of slowly conducting neurones [see Fig. 2A–C). (B) The propagation of the peak of the wave of RS action potentials was little influenced by changing the relative participation of the different groups of RS neurones. In the case of a relative increase of the participation of slowly conducting RS neurones, at rapid swimming frequencies (8 Hz), the tonic activity in caudal segments becomes large and can locally surpass the wave of action potentials.

At a swim frequency of 8 and 10 Hz, the wave of action potentials and the muscular contraction wave propagate at approximately the same velocity (Fig. 4A). Under the conditions chosen for analysis, this velocity also roughly corresponds to the conduction velocity of the most rapid RS neurones. This suggested that the propagation velocity of the wave of action potentials and that of the muscular contraction wave could be equated for all swim velocities by selectively activating RS neurones of a conduction velocity less than or equal to that of the velocity of the muscular contraction wave. The effects of such a recruitment on the propagation of the wave of action potentials and on its amplitude was calculated for swim frequencies of 1, 2, 4 and 8 Hz by activating all RS neurones of conduction velocity respectively less than or equal to 0.3, 0.6, 1.2 and 2.4 m/s (Fig. 2D).

Under these conditions, the characteristics of the distribution of action potentials along the cord become similar for all swimming velocities (Fig. 7) and the propagation velocity of the wave of action potentials is similar to that of the muscular contraction wave for all swim velocities (Fig. 8A). Moreover, the amplitude of the wave decreases from rostral to caudal in a similar manner for all swim velocities (Fig. 8B). More precisely, the slope of the total RS activity

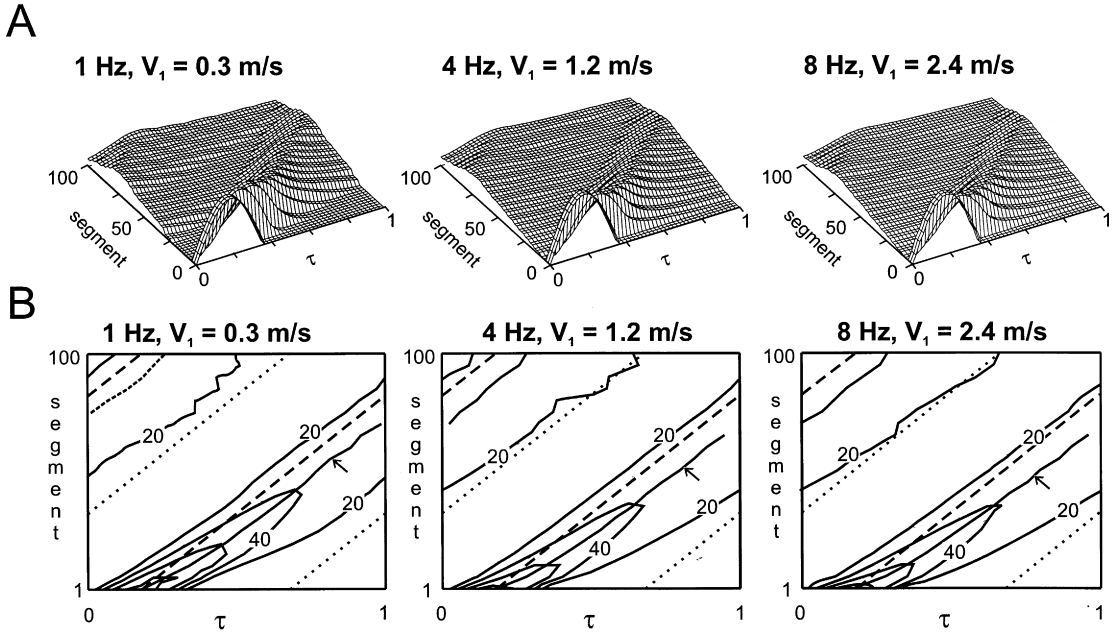

Fig. 7. Effect of a recruitment by size on the propagation of RS-activity. For the groups with a conduction velocity inferior or equal to v_1, the situation was kept as in Fig. 2A and the firing function $a_v(\tau)$ assigned to all those groups was the positive half of a sine function lasting one third of the cycle duration and had the same amplitude for all groups. For the groups with a conduction velocity greater than v_1 we put $a_v(\tau) = 0$. (A) propagation at swim frequencies of 1, 4 and 8 Hz. (B) Contour plots of the data presented in (A). Same figure composition as in Fig. 3.

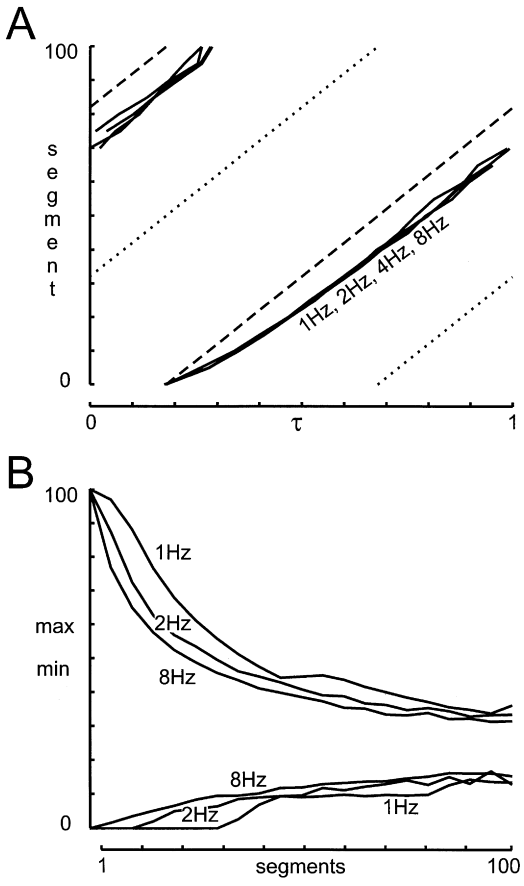

Fig. 8. Effects of recruitment by size on the RS maximum activity. (A) By adjusting the level of recruitment, the peak of the wave of RS action potentials can be forced to propagate with the same velocity as the muscular contraction wave. The full lines represent the propagation of the maximum RS-activity and the dashed lines indicate the propagation of the muscular contraction wave. (B) Relative maximal and minimal number of action potentials crossing a given segment for various swim frequencies.

$A^x(\tau)$ is, up to scaling with $1/T$, roughly independent of the swimming velocity v_s if we assume recruitment of the RS neurones up to v_s (cf. Appendix A). A recruitment of successively more rapid RS neurones for controlling more rapid swimming would thereby presumably support the smooth propagation of the muscular contraction wave.

3. Discussion

This investigation suggests that under the assumption that during swimming the RS neurones are activated independently of their conduction velocities, the descending signals provided by supraspinal centers propagate in a phasic wave along the cord. During slow swimming, the velocity of this phasic wave exceeds that of the wave of muscular contraction, and may provide an excitatory input to spinal neurones during the inhibited phase of their rhythmic activity. In the caudal segments, the late arrival of action potentials propagating along slowly conducting axons only provides a low level of tonic activity. Increasing the swimming velocity increases the velocity of the phasic wave of descending action potentials and increases the amplitude of the tonic background activity observed in the caudal segments.

If recruitment of RS neurones of successively higher conduction velocities is postulated during successively more rapid swimming, the velocity of the wave of action potentials can be kept to the same value as the velocity of the muscular contraction wave. Let us point out that such a size principle is implemented in motoneurones pools where at increasing input motoneurones with successively higher propagation speed and size are recruited (see Henneman and Mendell, 1981).

These results rely on the validity of the assumptions made and therefore a discussion of their agreement with biological data is necessary. The first assumption asserts that the RS neurones are rhythmically active during swimming. In the lamprey, data on the activity of RS neurones during free swimming are not currently available, but rhythmic activity of RS neurones is suggested by several observations. For instance, the RS neurones receive sensory information from the vestibular system whose components are rhythmically active during fictive locomotion (Rovainen, 1967; Rovainen, 1979; Wickelgren, 1977; Bussières and Dubuc, 1992a; Bussières and Dubuc, 1992b). Furthermore, afferents from the spinal cord provide a source of rhythmic input, transmitting information about segmental activity and intervening in a feedback loop to the spinal cord (Kasicki and Grillner, 1986; Kasicki et al., 1989; Vinay and Grillner, 1993). Oscillations of the membrane potential in phase with the locomotor activity have been observed in both large and small RS neurones of all reticular nuclei (Kasicki and Grillner, 1986; Kasicki et al., 1989). In other vertebrates, few data are available for freely moving animals. In the cat, the activity of RS neurones during free locomotion is modulated with the periodicity of the locomotor rhythm (Orlovsky, 1970; Drew et al., 1986)

In addition to the rhythmic phasic component, a tonic background activity is expected in some RS neurones. This activity was not integrated in the simulation reported here since tonic activity would influence all segments equally and therefore not affect the existence of a phasic wave of action potentials propagating along the cord. The main influence of tonic activity will rather be to set a background activation level of the spinal cord, and thereby to alter the ratio between the maximal number of spikes at the peak of the phasic wave and that of the minimal activity.

The second assumption concerns the range of conduction velocities encountered in RS axons. The axons in the lamprey spinal cord are not myelinated, and their diameter ranges from 0.1 to 100 μm (Bertolini, 1964). For unmyelinated axons, the conduction velocity is proportional to the square root of the diameter of the axon (Jack et al., 1975). In electrophysiological experiments on ca. 30 cm long adults of *Lampetra fluviatilis* or *Ichthyomyzon unicuspis* most RS neurones have conduction velocities ranging between 0.1 and 3 m/s (Kasicki et al., 1989; Wannier, 1994). Higher

conduction velocities have been reported, but these appear to be rather exceptional (Wannier, 1994). In the present model, the conduction velocities ranged between 0.1 and 3 m/s, which are realistic values for lampreys of ca. 30 cm body length.

The third assumption states that for a given RS axon, the conduction velocity of the action potentials is constant along the cord. Anatomical data indicate that the diameter of RS axons slowly decreases as the axons extend more caudally, and therefore the conduction velocity of RS axons also decreases (Rovainen et al., 1973). However, direct measurements have shown that the variations are slight, and therefore the conduction velocity can be accurately approximated by considering it to be constant along the whole length of the RS axons (Rovainen, 1982).

The propagation of action potentials has been studied for swim frequencies between 1 and 10 Hz. Although few data are available on the range of swimming frequencies performed by free swimming lampreys, this range seems representative. In laboratory conditions, *Ichthyomyzon unicuspis* swimming at frequencies between 1.5 and 7.6 Hz have been observed (Wallén and Williams, 1984). In the *in vitro* preparation of the spinal cord, the minimum frequency of fictive locomotion lies at ca. 0.1 Hz, while for rapid swimming values above 8 Hz were not observed (Brodin et al., 1985).

The time course of the function describing the firing activity of a RS cell group was selected in consideration of the fact that, for most RS neurones, the firing behaviour is intimately related to that of the motoneurones placed ipsilaterally in the rostral segments (Kasicki et al., 1989). For the first simulations, the duration of RS activity was set to one third of the cycle duration, the proportion of the cycle duration during which motoneurones are active (Wallén and Williams, 1984). This proportion is independent of the swim frequency (Wallén and Williams, 1984). Increasing the burst duration to 50% of the cycle duration does not markedly alter the behaviour of the wave and the conclusions of this study.

The model describes the distribution of action potentials along RS axons as a function of the time in the swim cycle, when the system has reached a steady state of swimming. This is achieved when the burst of action potentials elicited during the first swim cycle and propagating along the axons sharing the lowest conduction velocity has reached the last segment. Since the lowest conduction velocity considered in this model is 0.1 m/s, an action potential travelling on an axon of this conduction velocity will need 3 s to traverse the 30 cm length of the body. For swimming at 1 Hz, the steady state is then achieved after three cycles, but is already approximated during the first cycle since all the action potentials travelling with a conduction velocity above 0.3 m/s will have passed through the whole length of the body. Theoretically, for swimming at 8 Hz, the steady state is achieved after 3 s as well and, therefore, requires 24 cycles to complete. In this case, at the end of the first cycle, only the action potentials travelling at a conduction velocity above 2.4 m/s have traversed the whole length of the body, and the distribution of action potentials along the cord during the first cycle would differ markedly from that observed when the steady state is reached. However, considering that such high velocities are presumably reached after an acceleration phase involving a gradual increase of velocity, this situation probably does not occur.

The relative amplitude of the function describing the firing behaviour of two groups of neurones is determined by two factors: first, the number of neurones in each group, and second the firing activity of each neurone. The number of RS neurones in the brain of the lamprey has recently been estimated to range from 1250 (Davis and McClellan, 1994) to 2500 (Bussières, 1994). Because most of these neurones are small, a dominant participation of slow conducting neurones may prevail. However, since no data are presently available on the conduction velocities and on the proportion of neurones effectively participating in the control of velocity, the possibility exists that the participation of rapidly conducting neurones predominates over that of slowly conducting neurones. Both situations have been considered and they differ more in their strength than in the qualitative aspects of the effect.

In the experiments of Kasicki et al. (1989), both slowly and rapidly conducting RS neurones were modulated during fictive locomotion, but the majority of those producing action potentials were rapidly conducting neurones. Interestingly, 41% of the reticulospinal neurones located in the posterior rhombencephalic reticular nucleus (a nucleus containing mainly small neurones) recorded in this study were not modulated during fictive locomotion (Kasicki et al., 1989). In the model, strong participation of slowly conducting neurones determines the strength of the tonic activity reaching caudal segments during the whole duration of the swim cycle. If the activity of rapidly conducting neurones prevails, the relative importance of the tonic component decreases, and the phasic wave gains in amplitude.

An important observation of the study is that in the case of slow swimming, a phasic wave of action potentials propagates along the whole length of the cord, at a velocity greater than that of the muscular contraction wave. Such a wave was found for all firing functions investigated, and will reach levels of the cord crossing the inhibitory phase of the swim cycle. Because the majority of RS neurones have an excitatory action on their target neurones, this input would tend to entrain the activity of these segments and cause a shift in their phase in relation to the first segments. This would presumably disrupt the smooth propagation of the muscular contraction wave along the body.

However, if successive recruitment of RS neurones of increasing conduction velocity is introduced for the generation of increasing swimming velocities, the velocity of the wave of action potentials and that of the muscular contraction wave can be kept to similar values. Because conduction velocity and size of the soma are positively correlated in the

lamprey (Kasicki et al., 1989), this recruitment order would basically correspond to the recruitment by size observed for motoneurones in many species (see Henneman and Mendell, 1981). The mechanisms underlying the recruitment order of motoneurones remain a subject of debate, but those mechanisms proposed are also applicable for RS neurones (Henneman and Mendell, 1981; Lüscher and Clamann, 1992; Senn et al., 1997). A size-dependent recruitment order would, therefore, provide a simple physiological means for controlling swimming velocities.

If RS neurones are not recruited by size then alternative mechanisms will have to be sought to explain how locomotor activity can remain undisturbed by inputs reaching particular segments during an inappropriate portion of their swim cycle. Because no descending pathway other than the RS system affects middle and caudal segments directly, such a mechanism would presumably have to rely completely on a local spinal circuit. For instance, local control of the synaptic drive of RS neurones by the segmental locomotor circuit could provide a means to gate synaptic input and insure that its effects do not conteract the generation of the locomotor activity. Since presynaptic inhibition of RS neurones has been described in lamprey (see Shupliakov et al., 1995; Cochilla and Alford, 1997) this mechanism is also conceivable. However, in contrast to the size-dependent recruitment hypothesis, such a mechanism requires a complex network and would introduce new limitations in the control of locomotion (e.g. the system would not be accessible to descending control during the whole swim cycle).

4. Conclusions

We investigated the global spatio-temporal characteristics of the propagation of action potentials along RS axons in lamprey. A common assumption derives from the model proposed by Kasicki et al. (1989), according to which RS neurones are activated independently of their conduction velocity during locomotion. The present study indicates that under this condition, the action potentials propagate in a phasic wave. This wave is particularly prominent at slow swimming frequencies, and would provide an excitatory input to spinal neurones which crosses the inhibitory phase of their swim cycle. At high swimming frequencies, a tonic activity builds up in the caudal segments, but a phasic component remains. These results remain valid when biologically realistic variations either of the duration or time course of the firing activity are made, as well as when a weighted participation of the various groups of RS neurones is assumed.

If instead recruitment by size of RS neurones is assumed, then it is possible for each swimming velocity to adjust the level of recruitment in order to bring the velocity of the wave of action potentials to a value similar to the velocity of the muscular contraction wave. We showed that RS and muscular wave propagate in parallel if the speed of the fastest activated RS neurons corresponds to the swimming speed. With such a recruitment, the distribution of the RS activity over the body and a cycle scales linearly with the swimming frequency. This fact may simplify the control of movement for different swimming velocities.

The present study suggests that, in lamprey, the supraspinal control of velocity may rely on a recruitment of RS neurones by size. Otherwise, mechanisms to regulate the effects of the descending neurones on their spinal target neurones as a function of the segmental activity will have to be postulated.

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Appendix A Peak propagation and shape invariance of RS activity

For simplicity we assume that the activation functions $a_v = a$ are the same for all conduction velocities v. Substituting $s = x/(Tv)$ one calculates $\mathrm{d}v = -x/(Ts^2)\mathrm{d}s$ and from Eq. (2) one gets

$$A^x(\tau) = \int_{s_1}^{s_0} a(\tau - s)\frac{x}{Ts^2}\,\mathrm{d}s = \int_{-\infty}^{\infty} a(\tau - s)\rho^{x/T}(s)\mathrm{d}s = a^*\rho^{x/T}(\tau), \tag{A1}$$

where $\rho^{\xi}(s) = \xi/s^2$ for $s_1 < s < s_0$ and $\rho^{\xi}(s) = 0$ else. Here we set $s_1 = x/(Tv_1)$ and $s_o = x/(Tv_o)$. Since $a(\tau)$ is periodic with period 1 we have $A^x(\tau + 1) = A^x(\tau)$ for all τ. Since in addition $\rho^{\xi}(s)$ is monotonically decreasing in s one gets $A^x(s_1 + \Delta\tau) < A^x(s_1)$ for $1/3 < \Delta\tau < 4/3$ and together with the periodicity we conclude that the maximal value of A^x is attained in the range $s_1 + 1/6 \le \tau^x_{\max} \le s_1 + 1/3$. The particular boundaries are due to the fact that the positive values of $a(\tau)$ within a periodicity domain are concentrated onto the interval [0, 1/3] and that the peak of $a(\tau)$ is assumed to be at $\tau = 1/6$. Since for fixed swimming frequency the parameter s_1 increases linearly with x we get the approximated linearity of $\tau^x_{\max}$ asserted in Eq. (3). To motivate the nonlinear term in this equation we restrict ourselves to show that $\tau^x_{\max}$ moves (monotonically) from the upper bound $s_1 + 1/3$ to the lower bound $s_1 + 1/6$ when T increases from 0 to ∞. Let us first consider the case $T \to \infty$. Since the integral over the function $\rho^{\xi}(s)$ is constant ($= v_1 - v_0$), the functions $\rho^{x/T}(s)$ converge (up to scaling) to a Dirac delta-function at $s = s_1$ and the maximum of $A^x(\tau)$ is attained at the maximum of $a(\tau)$ transferred by s_1, i.e. at $\tau^x_{\max} = s_1 + 1/6$. In the case that $T > 0$ is small, the function $\rho^{x/T}(s)$ is (again up to

scaling) close to a step function on the interval $[s_1 - 1/3, s_1 + 1/3]$ with step (from 0 to v_1/s_1) at $s = s_1$. The maximum of $A^x(\tau)$ is, therefore, attained for a value of τ such that $a(\tau - s)$ is as large as possible for all $s \in [s_1, s_1 + 1/3]$. This value has to be $\tau^x_{\max} = s_1 + 1/3$, the smallest value for which $a(\tau^x_{\max} - s) > 0$ for the referred s. To fit $\tau^x_{\max}$ for intermediate values of T we chose an exponential which interpolates between $s_1 + 1/6 \le \tau^x_{\max} \le s_1 + 1/3$. A similar reasoning shows that the approximation Eq. (3) turns towards equality if x or v_1 tend to 0 and ∞.

Let us now infer the shape invariance of the RS-activity $A^x(\tau)$ for the case that the RS neurones are activated according to their size (and thus according to increasing conduction velocity). Thus, if $v_s = L/T$ denotes the swimming velocity, we assume that only those RS neurones with conduction velocity between v_0 and v_s are activated. The total RS activity is then calculated according to Eq. (A1) by an integral from $s_1 = x/(Tv_s) = x/L$ to $s_0 = x/(Tv_0)$. For parameter values $L = 0.3$ m, $v_0 = 0.1$ m/s and swimming frequency between 1 and 8 Hz ($T = 1$ s, …, 0.125 s), the upper bound s_0 is 10–80 times larger than the lower bound s_1. Due to the factor $1/s^2$ the part of the integral in Eq. (A1) from $s_0' = x/v_0$ to s_0 is, therefore, small compared to the integral from s_1 to s_0'. We obtain the approximation of the total activity by the integral from s_1 to s_0',

$$A^x(\tau) = \int_{x/L}^{x/(Tv_0)} a(\tau - s)\frac{x}{Ts^2}\mathrm{d}s \approx \frac{x}{T}\int_{x/L}^{x/v_0} a(\tau - s)\frac{1}{s^2}\mathrm{d}s = \frac{1}{T}f(\tau, x), \tag{A2}$$

where $f(\tau,x)$ does not depend on T. According to this formula, the total RS activity scales roughly linearly with the swimming frequency $1/T$ while the particular shape remains (cf. Figs 7 and 8).

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