

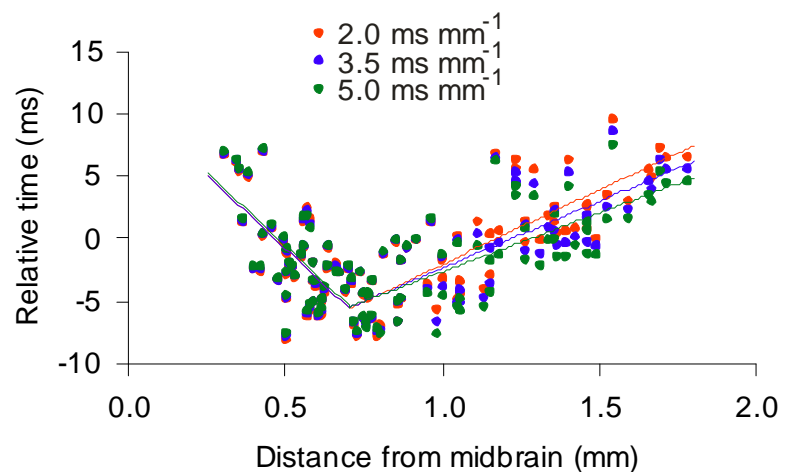
FOR PUBLICATION

Supplemental material

Adjustment of spike timing measurements according to ventral root location

Timing measurements for spikes based on loose patch recordings were made relative to the onset of ventral root bursts at the level of the 5th post-otic segment (1.36 caudal to the midbrain). To compare between animals, where some ventral root recordings were made at other rostro-caudal levels, timing was adjusted assuming a mean rostro-caudal, ventral root burst delay of 3.5 ms mm^{-1} . To check that this assumption did not produce a misleading interpretation, analysis was repeated using the minimum (2.0 ms mm^{-1}) and maximum (5.0 ms mm^{-1}) reported values for delay (Tunstall & Roberts, 1991). For each of the three delays, timing measurements are well described by two contiguous linear regression lines with a changeover 0.725 mm from the midbrain. Differences between these regression lines are very small except at relatively caudal positions (Supplemental Fig. 1).

Supplemental Figure 1.
Differences in the relative timing of spikes during swimming using different estimates for rostro-caudal delay in ventral root burst onset.



Modelling the relative timing of dIN spikes and EPSCs.

Data from our sample of individual recorded spike and EPSC times were used to estimate the timings within a whole reticulospinal dIN population. We started with the descriptions of timings in samples of each:

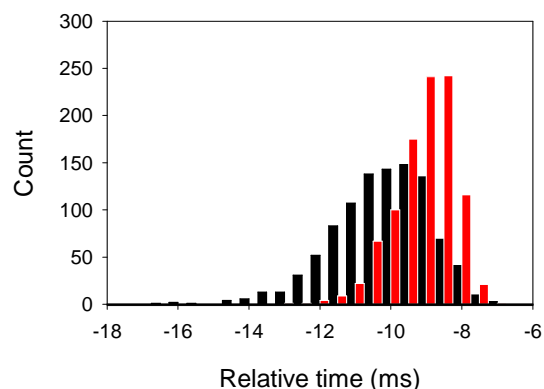
dIN spikes: mean = $-3.964 \pm 2.877 \text{ ms}$, $n = 32$ neurons
 dIN EPSCs: mean = $-5.343 \pm 1.766 \text{ ms}$, $n = 16$ neurons

Neither of these distributions deviated significantly from normal. (Anderson-Darling: $p = 0.074$ and $p = 0.827$ respectively)

We then assumed a population of 50 dINs (a conservative estimate) and generated normally distributed datasets (each $n = 50$) for spike and EPSC timings with means and S.D.s obtained from measured samples. For each distribution, the minimum value was recorded; that is, the time of the first spike and the first EPSC. This was then repeated 1000 times to generate a distribution of "earliest times" (Supplemental Fig. 2)

In this modelled population of dINs, the first spikes (-10.457 ± 1.366 ms) significantly preceded the first EPSCs (-9.348 ± 0.839 ms; two sample t -test: $t = -21.88$, $p < 0.0001$) (difference: -1.109 ± 1.565 ms).

Supplemental Figure 2. Distribution of relative times for the first spike (black) and the first EPSC (red) in populations of 50 dINs; 1000 repeats.



This was then repeated for a larger (probably more realistic) dIN population ($n = 100$). The difference in timing between the earliest dIN spikes and EPSCs increased: the earliest spikes (-11.167 ± 1.238 ms) preceded the earliest EPSCs (-9.735 ± 0.758 ms; two sample t -test: $t = -31.2$, $p < 0.0001$) even further (difference: -1.432 ± 1.432 ms).

Reference

Tunstall MJ & Roberts A. (1991). Longitudinal coordination of motor output during swimming in *Xenopus* embryos. *Proc R Soc Lond B* **244**, 27-32.