cal signs indicating risk for SIDS are manifested in the newborn, these signs may not be due solely to postnatal environmental influences. Early neonatal deaths and death from SIDS share a variety of common epidemiological factors (*14*). The antecedents of SIDS might therefore lie in a disturbance in fetal development.

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### References and Notes

1. A. B. Bergman, C. G. Ray, M. A. Pomeroy, P. W. Wahl, J. B. Beckwith, *Pediatrics* **49**, 860 (1972).
2. T. Anders, R. Emde, A. Parmelee, *A Manual of Standard Terminology, Techniques, and Criteria for Scoring of States of Sleep and Wakefulness in Newborn Infants* (UCLA Brain Information Service/BRI Publications, Los Angeles, 1971).
3. P. G. F. Swift and J. L. Emery, *Arch. Dis. Child.* **48**, 947 (1973).
4. Evidence for a higher incidence of SIDS among SSIDS is derived from a study of 162 SIDS victims in Northern Ireland [P. Froggatt, M. A. Lynas, G. MacKenzie, *Br. J. Prev. Soc. Med.* **25**, 119 (1971)] corroborated in America [D. R. Peterson, N. M. Chinn, L. D. Fisher, *J. Pediatr.* **97**, 265 (1980)]. Anecdotal data support this evidence [J. B. Beckwith, *Curr. Probl. Pediatr.* **3**, 1 (1973)]. Epidemiological considerations for this study are described in detail elsewhere [C. R. Stark, in *Ontogeny of Sleep and Cardiopulmonary Regulation: Factors Related to the Sudden Infant Death Syndrome*, M. B. Sterman, J. Hodgman, H. Hoffman, C. R. Stark, Eds., in preparation.]
5. E. Hofmann, B. Havens, S. Geidel, T. Hoppenbrouwers, J. E. Hodgman, *Acta Paediatr. Scand. Suppl. 266* (1977).
6. R. M. Harper, T. Hoppenbrouwers, M. B. Sterman, D. J. McGinty, J. Hodgman, *Pediatr. Res.* **10**, 945 (1976).
7. R. M. Harper, R. J. Sclabassi, T. Estrin, *IEEE Trans. Autom. Control* **19**, 932 (1974); J. W. Tukey, *Exploratory Data Analysis* (Addison Wesley, Reading, Massachusetts, 1977). Smoothing of the spectral curves was accomplished with a three-point moving median, repeated once.
8. M. A. Valdes-Dapena, *Pediatr. Clin. North Am.* **10**, 693 (1973); D. R. Peterson, *Am. J. Epidemiol.* **84**, 478 (1966).
9. R. M. Harper and E. K. Sauerland, in *Sleep Apnea Syndromes*, C. Guilleminault and W. C. Dement, Eds. (Liss, New York, 1978), pp. 219–234.
10. E. A. Phillipson and C. E. Sullivan, in *ibid.*, pp. 47–64.
11. J. B. Beckwith, in (*4*).
12. R. L. Naeye, *Science* **186**, 837 (1974); *Am. J. Clin. Pathol.* **66**, 526 (1976). The evidence for hypoxia in SIDS victims includes increased heart weights, increased muscle tissue in pulmonary arteries, increased volume of chromaffin cells in the adrenal medulla, and abnormal retention of brown fat cells above the adrenals.
13. R. M. Harper, B. Leake, T. Hoppenbrouwers, M. B. Sterman, D. J. McGinty, J. Hodgman, *Pediatr. Res.* **12**, 778 (1978); R. M. Harper, B. Leake, J. Hodgman, T. Hoppenbrouwers, M. B. Sterman, in preparation.
14. D. R. Peterson, G. Van Belle, N. M. Chinn, *Am. J. Epidemiol.* **110**, 699 (1979).
15. Supported by NIH grants RO1 HD 14608-01, HD-2-2777, and HD4-2810. We thank S. Geidel, B. Havens, J. R. Mason, L. Miyahara, D. Taube, and S. Vigil.

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## Neural Control of Swimming in a Vertebrate

Abstract. *An excitatory drive to the spinal cord neurons supplying rhythmic locomotor output in swimming was demonstrated in paralyzed late* Xenopus *embryos. The motor system for a single muscle can independently generate rhythmic motor discharge with the normal swimming cycle period.*

In many groups of animals, including mammals, the basic alternating pattern of motor discharge required for locomotion can be generated within the central nervous system without reflex feedback from the movement itself (*1*). Current hypotheses favor the idea that the rhythmic activity underlying locomotor patterns in vertebrates is generated within the spinal cord. Higher centers are thought to activate and modulate this spinal cord pattern generator by means of a descending tonic excitatory drive. In most hypotheses, reciprocal inhibition between antagonistic motor systems in the spinal cord is crucial, either being fundamental to the rhythmicity of the spinal cord pattern generator (*2*) or being necessary for the coordination of phase relationships between separate, inherently rhythmic motor systems (*3*, *4*). Despite the long history of most of these proposals, direct evidence for excitatory drive during locomotion and on the role of reciprocal inhibition is not available for vertebrates. We have therefore sought a simple vertebrate preparation in which to explore the origins of locomotory rhythms and their control by higher centers.

The behavior of embryos of fish and amphibians is limited, and they are neuroanatomically simple. They therefore offer a model system in which to explore fundamental features of nervous organization. We have studied late embryos of the clawed toad (*Xenopus laevis*) at stage 37/38 (*5*) (Fig. 1A). When released from their egg membranes, embryos will swim spontaneously or in response to mechanical stimuli or dimming of the light (*6*); swimming can be stopped by bumping the head or cement gland (*7*). Filming

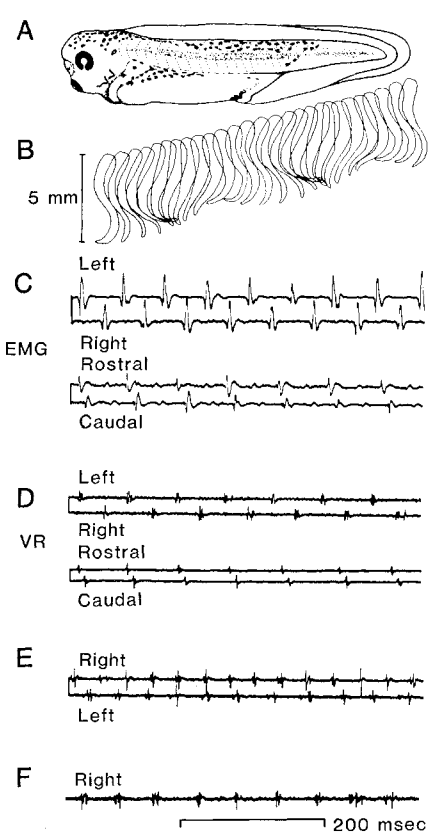


Fig. 1. (A) Side view of a late embryo of *Xenopus laevis* at stage 37/38 (*5*). (B) Tracings of swimming movements from cine films taken at 300 frames per second. Waves of bending pass caudally down the body. (C) Muscle potentials during swimming in restrained embryos were recorded by placing saline-filled suction pipettes on exposed myotomal swimming muscles. Activity on left and right sides of one segment alternated (upper traces). Activity was delayed at the more caudal of two recording sites on the same side, 2 mm apart (lower traces). (D) Motoneuron discharge in the ventral roots (*VR*) of paralyzed embryos (*9*) recorded with suction pipettes placed at the junctions between segmented myotomes where muscle fibers are innervated. Rhythmic discharge, evoked by dimming illumination (*6*), alternated on left and right of one segment (upper traces). Discharge was delayed at the more caudal of two recording sites on the same side, 1.5 mm apart (lower traces). (E) Rhythmic ventral root bursts on both sides of a paralyzed embryo lacked strict alternation when left and right halves of the nervous system were surgically separated caudally from the first postotic myotome (*15*). (F) Rhythmic ventral root bursts recorded from the isolated right half of the hindbrain–spinal cord (*16*).


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(Fig. 1B) has shown that the period of the swimming cycle is 40 to 100 msec, and waves of bending propagate caudally down the body. Muscle potentials recorded (*8*) during swimming in restrained embryos alternate on left and right sides and appear after a delay at more caudal segments (Fig. 1C). In embryos paralyzed by the neuromuscular blocking agent curare (*9*), stimuli that normally initiate swimming evoke alternating bursts of ventral root activity on either side of the trunk (Fig. 1D). Motor root activity is usually delayed in caudal segments relative to that in the more rostral roots. The cycle period of the rhythmic motor root discharge is similar to that of unrestrained swimming movements and in both tends to increase during episodes of activity. Like the swimming movements, the alternating ventral root bursts can be stopped by bumping the head and can also be obtained in spinal embryos (with the brain removed), although episodes of activity are then present only during stimulation. This evidence indicates that the paralyzed embryo can generate a pattern of motor activity similar to that observed during normal swimming locomotion. *Xenopus* embryos are therefore similar to adult vertebrates and invertebrates, in which the basic locomotory pattern can be generated within the central nervous system or spinal cord without sensory feedback. Pattern generation is not a property of a unique site, since separated rostral and caudal segments can generate swimming activity independently.

To provide an anatomical basis for our electrophysiological studies, we have retrogradely labeled spinal cord neurons with horseradish peroxidase (HRP) (*10*). Motoneurons can be distinguished by a ventral soma, lateral and dorsal dendrites in the lateral longitudinal tracts, and by their peripheral axons, which usually project caudally before leaving the cord to innervate the myotomes (Fig. 2, A and B). Using HRP, we have also seen interneurons in the spinal cord whose axons cross to the contralateral side of the cord by way of the ventral commissure (there is no dorsal commissure) and which could mediate reciprocal inhibition.

Using paralyzed animals, we have made intracellular recordings (*11*) from cells in the ventral part of the cord, which we know to contain predominantly motoneurons, at levels from the third to ninth postotic myotome. During "swimming," these putative motoneurons fire rhythmic spikes in phase with the motor root discharges on the same side of the cord (Fig. 2, D and E). Furthermore, iontophoresis of Lucifer yellow dye (*12*) into some of the recorded neurons showed their anatomy to be typical of motoneurons (Fig. 2C). We therefore call these cells motoneurons until further evidence indicates otherwise.

When impaled with a microelectrode, motoneurons are normally silent and have a resting potential of −50 to −67 mV. A single episode of "swimming" generally starts with a gradual depolarization of the motoneuron membrane potential and terminates with its gradual repolarization (Fig. 2, D and E). Between these lies a period when the membrane potential is tonically depolarized (*13*), usually by 10 to 20 mV from the resting potential. Throughout the episode, motoneuron membrane conductance increases, as indicated by a decrease in the amplitude of hyperpolarizing constant current pulses, injected into cells through the microelectrode. A higher amplitude of tonic depolarization is associated with a larger increase in conductance. This shows that the tonic depolarization is a genuine excitatory drive and not, rather, a release from inhibition.

Superimposed on the tonic depolarization during an episode is rhythmic activity whose cycle period is similar to that of unrestrained swimming movements. Each cycle of this rhythmic activity consists of a period of phasic excitation, usually evoking a single spike, alternating with phasic inhibition (*14*) (Fig. 2E). Rhythmic spikes occur in phase with motor root discharges on the same side, and the onset of the period of inhibition is in phase with the motor root discharges on the opposite side. The cycle period, which tends to lengthen during an episode (Fig. 2E), is related to the amplitude of the tonic depolarization. A higher amplitude of tonic depolarization is associated with a shorter cycle period.

From our intracellular recordings, we

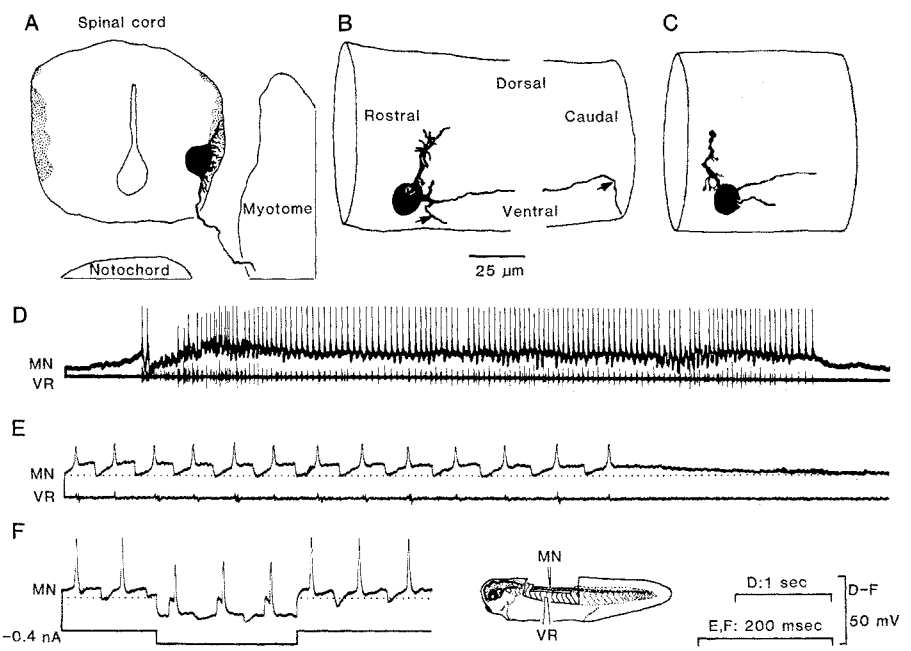


Fig. 2. (A) Horseradish peroxidase—filled, spinal cord motoneuron with its dendrites in the longitudinal axon tract (dotted) and its axon leaving the ventral part of the cord. The dorsal extent of the myotomes and the notochord are also indicated. Drawn from 15-μm transverse wax section. (B) HRP-filled motoneuron with characteristic dorsal dendrites and an axon running caudally before branching and leaving the spinal cord in two places (arrows). Lateral view, drawn from whole mount. About 100 μm of caudal axon is omitted. (C) Lucifer yellow–filled neuron showing anatomical features characteristic of motoneurons, such as in (A) and (B), drawn from whole mount. Orientation as in (B). (D) A single spontaneous episode of "swimming" in a paralyzed embryo, recorded intracellularly from a motoneuron (*MN*) and extracellularly from an adjacent ventral root (*VR*), as indicated in the diagram. The episode starts with a smooth, gradual depolarization and terminates with a smooth, gradual repolarization of the motoneuron membrane potential. Between these, the rhythmic motoneuron spikes are superimposed on a background tonic depolarization. (E) The rhythmic motoneuron activity consists of single spikes, in phase with ventral root bursts on the same side, alternating with midcycle IPSP's. The level of the resting potential is indicated by dots. Swimming cycle period tends to elongate at the end of the episode and finally after rhythmic activity ceases, the background tonic depolarization smoothly returns to the resting potential. (F) Injection of hyperpolarizing current pulse into a motoneuron during a "swimming" episode in a curarized embryo delays the rhythmic spikes sufficiently to reveal underlying depolarizing potentials and reduces the amplitude of the midcycle IPSP's. Note that two IPSP's after the current pulse undershoot the resting potential.

are able to draw some conclusions regarding the nature of the rhythm generator within the spinal cord. In each cycle of a "swimming" episode, motoneurons spike once. Injection of hyperpolarizing current reveals a phasic depolarizing potential that increases in amplitude with hyperpolarization and underlies each spike (Fig. 2F). While motoneurons can be made to fire by injecting depolarizing current, they show no inherent rhythmicity. It appears, then, that the depolarizing potentials underlying motoneuron spikes represent phasic drive from a separate spinal cord rhythm generator.

Consecutive motoneuron spikes are separated by a period of inhibition (*14*), which starts in phase with motor root discharges on the opposite side of the cord. In order to investigate reciprocal interactions in the swimming pattern generator, we made motor root recordings in surgically altered embryos. Using a fine needle, we completely separated the two sides of the nervous system to interrupt any crossed commissural pathways between the two sides. The roof of the hindbrain was opened along the midline and the cut continued caudally along the neurocoele into the spinal cord. Ventral connections between the two sides were then cut until the notochord was clearly visible between the left and right sides. Normally alternating activity, evoked by stimulation, was present provided that commissural connections in about 150 μm of the rostral spinal cord were intact. Complete midline separation of the left and right sides from the midbrain caudally (*15*) did not upset the rhythmic burst production on either side, but the normally strict alternation between the two sides was lost (Fig. 1E). When one whole side of the hindbrain and spinal cord was removed (*16*), the remaining half could still generate rhythmic burst activity (Fig. 1F).

Our experiments indicate that a capability to generate rhythmic bursts of motoneuron discharge with normal swimming cycle periods lies on either side of the central nervous system. This rhythmic pattern generation does not depend on mutual interconnections between antagonistic motor systems of the left and right sides. Models of pattern generation in which reciprocal inhibition is fundamental to rhythmicity (*2*) are therefore not applicable here. However, reciprocal inhibitory interactions are implicated in the organization of the usual strict alternation in activity of the left and right sides that underlies swimming. We suggest that spinal interneurons involved in swimming pattern generation are activated, like the putative motoneurons, by a tonic excitatory drive which is the result of the descending excitation previously hypothesized as an important component of locomotor pattern generation. The nature and source of this excitation remains to be ascertained.

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### References and Notes

1. S. Grillner, *Physiol. Rev.* **55**, 247 (1975).
2. T. G. Brown, *Q. J. Physiol.* **5**, 237 (1912); W. O. Friesen and G. S. Stent, *Biol. Cybern.* **28**, 27 (1977); S. Miller and P. D. Scott, *Exp. Brain Res.* **30**, 387 (1977); G. Szekely, *Acta Physiol. Acad. Sci. Hung.* **27**, 285 (1965).
3. R. L. Calabrese, *J. Exp. Biol.* **82**, 163 (1979); E. von Holst, *Ergeb. Physiol. Biol. Chem. Exp. Pharmakol.* **42**, 228 (1939).
4. D. M. Wilson and I. Waldron, *Proc. IEEE* **56**, 1058 (1968).
5. P. D. Nieuwkoop and J. Faber, *Normal Tables of* Xenopus laevis *(Daudin)* (North-Holland, Amsterdam, 1956).
6. A. Roberts, *Nature (London)* **273**, 774 (1978); _____ and B. P. Hayes, *Proc. R. Soc. London Ser. B* **196**, 415 (1977).
7. A. Roberts, *J. Comp. Physiol.* **135**, 341 (1980); _____ and A. R. Blight, *Proc. R. Soc. London Ser. B* **192**, 111 (1975).
8. For recording, embryos were in saline containing 115 m*M* NaCl, 2.5 m*M* KCl, 1.8 or 10 m*M* $CaCl_2$, 2.4 m*M* $NaHCO_3$, pH 6.8 to 7.2. Glass suction electrodes were applied to exposed muscles or intermyotome clefts for ventral root recording.
9. *d*-Tubocurarine, $10^{-4}$*M*, in physiological saline.
10. To label motoneurons, crystalized HRP was crushed onto axons in myotomes. Processing was conventional.
11. Intracellular recordings were made from 62 cells in 33 embryos with micropipettes filled with 2*M* potassium acetate (100 to 200 megohm). Current was passed through the recording electrode. To allow penetrations, the side of the spinal cord was exposed in paralyzed embryos.
12. W. W. Stewart, *Cell* **14**, 741 (1978).
13. Evidence that the background depolarization is tonic is as follows: (i) there was no repolarization of membrane potential in the portion of each swimming cycle between the spike and the midcycle inhibitory postsynaptic potential (IPSP) (*14*); (ii) the cell remained tonically depolarized between spikes, where the midcycle IPSP was not visible; (iii) episodes generally started with a gradual smooth depolarization and finished with a gradual smooth repolarization; and (iv) when swimming stopped during an episode, the background membrane potential remained tonically depolarized.
14. Evidence that the midcycle hyperpolarizations are IPSP's and not simply due to a fall in the level of tonic excitation is as follows: (i) they had a fast rise time; (ii) they could undershoot the resting potential (Fig. 2F); and (iii) when the intracellular [$Cl^-$] was raised by the use of 3*M* KCl microelectrodes, the midcycle IPSP's were reversed to become depolarizing potentials.
15. Ventral root recordings were made rostral to the fourth to eighth postotic myotome in 15 animals [operations were checked and confirmed histologically in (*1*)]. Spinal cord caudal to this was removed from the animal to simplify operations. Activity was evoked by dimming illumination or stroking the skin.
16. As for (*15*). Recordings were made from 13 animals [checked and confirmed histologically in (*1*)]. Activity was evoked by stroking the skin.
17. We thank the Royal Society, Science Research Council, and Medical Research Council for support and B. K. Follett, S. Miller, D. M. Armstrong, J. W. Dodson, J. Simmers, and R. de G. Weevers for their comments on this report.

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## Hyperthermia-Induced Seizures in the Rat Pup: A Model for Febrile Convulsions in Children

Abstract. *Seizures were produced in rat pups by ambient hyperthermia. Seizure threshold temperatures, measured rectally and intracerebrally, increased between 2 and 10 days of age. Electrocortical paroxysmal discharges were confirmed in hyperthermic 6- and 10-day-old pups. The increasing resistance to hyperthermic seizures with maturation and the electroencephalographic changes induced by hyperthermia are similar to those in young children.*

Febrile convulsions occur in about 3 to 5 percent of children between 6 months and 5 years of age (*1–3*). Victims are normal children with a genetic predisposition and children with brain damage or idiopathic epilepsy (*1–3*). Clinical studies have led to strong disagreement as to whether febrile seizures contribute to brain damage in very young children (*3–5*). Seizures induced by hyperthermia have been observed in the rat pup (*6*). We have further characterized hyperthermia-induced seizures in the immature rat as a model for studying the pathogenesis of febrile seizures and their sequelae.

Litters of Sprague-Dawley albino rats were culled to eight pups each on the day of birth and maintained (one litter and mother per cage) at 22° to 23°C. To determine seizure threshold temperatures, the pups were warmed individually in a Lucite chamber measuring 13 by 13 cm at the base and 6.5 cm in height. The walls and base were 0.6 cm thick and the chamber was covered by a black copper top 0.3 cm thick. The chamber floor was covered with paper towels. There were spaces in the walls for wires and air movement. A 250-W infrared lamp was held 10 cm above the chamber until seizure activity appeared and then turned off immediately.

For measurement of rectal temperatures, a plastic-insulated 1/16-inch thermistor probe was inserted at least 1 cm beyond the anus and connected to a recording thermometer (Yellow Springs Instrument Co.). For concurrent measurement of brain and rectal temperatures, bifilar, nylon-insulated, copper-constantan thermocouples (California Fine Wire Co.) approximately 0.2 mm in diameter were used (*7*). The brain ther-


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