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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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.
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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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mocouple was implanted about 2.5 mm deep in the left parietal lobe under ketamine-HCl anesthesia (22 mg/kg, subcutaneously). (Studies were performed 2 to 3 hours after implantation, when recovery from the anesthesia appeared complete.) The second thermocouple was inserted at least 1 cm into the rectum. The thermocouples were connected to a two-channel chart recorder, and reference junctions were placed in ice water. After each experiment, the animal was killed and appropriate placement of the thermocouples was confirmed by autopsy. Thermopotentials were calibrated over the range 32° to 46°C with a mercury thermometer in a bath kept at constant temperature.

The following behavioral changes and seizure characteristics were observed in pups made hyperthermic for the first time at different ages. Two-day-old animals remained quiet while rectal temperatures rose from 32° to 36°C. At about 37°C seizure activity began, with the pups rolling onto their backs and making independent clonic movements with all four legs. When the temperature was reduced, all the pups immediately resumed normal nursing behavior. Five-day-old pups remained quiet while being warmed from 32° to 38°C but became agitated at 38° to 40°C. At 40° to 41°C, generalized seizure activity appeared, manifested by loss of upright posture, tonic extension of the trunk, and rapid synchronous clonic movements of the extremities and mouth. Then the animals became limp and unreactive for 2 to 3 minutes. Similar seizures occurred in 7-day-old pups at about 43°C, with the postictal depression lasting about 5 minutes. All 5- and 7-day-old pups survived and appeared normal after the postictal period. At 10 days, only half the pups survived seizures similar to those at 5 and 7 days. Pups older than 10 days showed no seizure-like activity until rectal temperatures reached 44° to 45°C. No animals survived this extreme temperature.

Figure 1 shows baseline rectal temperatures, rectal temperatures at seizure thresholds, and concurrent brain temperatures in pups of different ages. Rectal temperatures at which seizures appeared increased significantly between 2 and 5 days and between 5 and 7 days of age. Threshold temperatures in individual animals at each age were very uniform despite variations in rates of temperature rise (0.3° to 0.7°C per minute). The maturational increase in basal rectal temperature was much less than the rise in seizure threshold temperatures in pups 2 to 7 days of age. The maturational

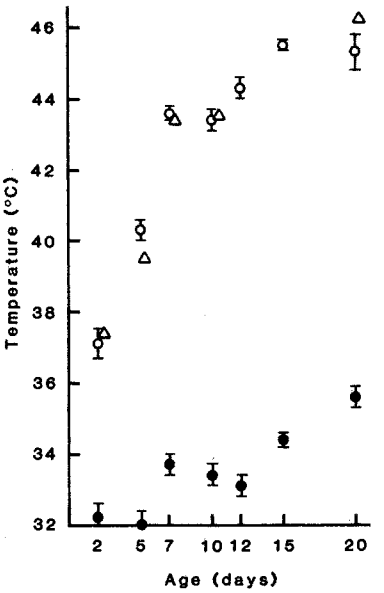

Fig. 1. Seizure threshold temperatures in rat pups. Rectal temperatures are shown as measured in pups equilibrated to room temperature before warming (●) and at seizure onset (○). At least six animals were studied at each age. The increases in seizure threshold temperature were significant between 2 and 5 days ($P < .001$, Student's t-test) and between 5 and 7 days of age ($P < .001$). Brain temperature (△) at the seizure threshold was measured in at least four additional animals at the ages indicated. Data are means ± standard errors.

increase in seizure threshold temperatures measured in the brain was comparable to that measured rectally (*8*).

Electroencephalograms (EEG's) were recorded in eight additional pups 2 to 10 days of age. Two metal eyeglass screws (10 mm in diameter) were secured with dental cement in small holes in the parietal bones. After recovering from anesthesia for 2 to 3 hours, the pup was implanted with a rectal thermistor and secured to the floor of the Lucite chamber with tape and a cardboard headholder, which prevented head movements. Bipolar recordings were obtained in the conventional manner with the use of low- and high-frequency filters of 0.1 Hz and 10 kHz. No electrocortical seizure activity was observed in 2- to 3-day-old animals warmed from 32° to 41°C. In two pups 6 days of age, EEG's showed constant low-amplitude fast activity (9 to 11 Hz) during warming from 32° to 40°C (Fig. 2A). Coincident with the onset of rhythmic jerking of the extremities at about 40.5°C, the EEG's showed large slow waves at a frequency of about one per second (Fig. 2B). At about 43°C, the EEG consisted of sharp waves with about the same frequency (Fig. 2C). In three 10-day-old pups, the baseline EEG's showed low-voltage fast activity (Fig. 2D). Irregular sharp waves appeared at about 41°C (Fig. 2E). Coincident with the appearance of rhythmic jerking of the extremities at 44°C, the EEG's showed high-voltage spike activity (Fig. 2F). No slow waves were seen in recordings from 10-day-old animals.

The increase in seizure threshold temperatures with maturation and the EEG changes during warming in rat pups are similar to the seizure characteristics associated with fever in young children. Seizures associated with hyperthermia in normal children are limited to the first 6 years of life (*3*). In the rat pup the seizure threshold temperature increases until 10 to 12 days of age, when it reaches an extreme that results in irreversible cell damage and death (*9*). Other observers have reported that a longer duration or higher degree of hyperthermia is required to produce seizures in older rat pups compared to younger pups (*6*). The high-voltage slow wave EEG pattern in the hyperthermic 6-day-old pups has been observed in children made hyperthermic by intravenous injection of typhoid vaccine (*10*). This EEG pattern is also characteristic of the immature brain's response to epileptogenic stimuli, such as penicillin (*11*).

We conclude that hyperthermia alone can produce seizures in the rat pup and, by extension, in the human child. Chemical mediators of the febrile response to an infection may also contribute to the genesis of seizures in the young child (*12*). The sensitivity of the immature brain to hyperthermia may be due, in

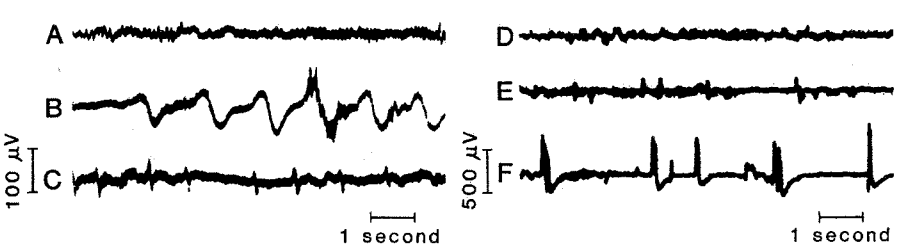

Fig. 2. Electroencephalograms from hyperthermic rat pups. (A to C) Recordings from a 6-day-old pup at 32.5°C (before warming), at 40.5°C, and at 43.7°C. (D to F) Recordings from a 10-day-old pup at 33.7°C (before warming), at 41.3°C, and at 44°C. Temperatures were measured rectally.

part, to a relatively limited capacity to increase cellular energy metabolism at elevated temperatures (*13*). This deficit would be markedly increased during seizures and could contribute to the brain damage associated with febrile-status epilepticus in very young children (*3, 4*). Functional neurological deficits, including decreased maze-solving ability and increased sensitivity to other epileptogenic stimuli, occur as sequelae to hyperthermic seizures in the rat pup (*6*). Further studies of the pathogenesis of hyperthermic seizures and their sequelae in the immature rat and in higher mammals may help to answer important clinical questions concerning febrile convulsions in young children.

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Abdominal Vagotomy Blocks the Satiety Effect of Cholecystokinin in the Rat

Abstract. *The site where peripherally administered cholecystokinin-8 elicits satiety was investigated by injecting rats with cholecystokinin-8 (1 to 8 micrograms per kilogram of body weight, intraperitoneally) after they had received bilateral lesions of the ventromedial hypothalamus or after they had undergone bilateral abdominal vagotomy or selective vagotomies. Abdominal vagotomy or gastric vagotomy abolished or reduced the satiety effect of cholecystokinin, but lesions of the ventromedial hypothalamus did not. These results demonstrate that peripherally administered cholecystokinin acts in the abdomen through gastric vagal fibers and not directly on the brain to produce satiety in the rat.*

Cholecystokinin (CCK), a peptide present in the central nervous system and gut, inhibits food intake in animals and humans after it is centrally (*1–3*) or peripherally administered (*4*). The site of action of this effect is controversial. After central administration, CCK-8 presumably acts on brain CCK receptors (*5*), although Nemeroff *et al.* (*6*) concluded that centrally administered CCK-8 inhibited tail pinch–induced feeding by acting at a peripheral site. It has been suggested that CCK-8 also acts in the brain (*2*), particularly in the ventromedial hypothalamic area, after it is administered peripherally (*3*). However, Kulkosky *et al.* (*7*) observed a normal satiety effect of peripherally administered CCK-8 in rats with bilateral lesions of the ventromedial hypothalamus (VMH). We have further investigated the site of action of peripherally administered CCK-8 (*8–10*) and report here that peripherally administered CCK-8 has a peripheral site of action in an abdominal organ innervated by the gastric vagal nerves.

Male Sprague-Dawley rats (300 to 500 g) were subjected to bilateral abdominal vagotomy and female Sprague-Dawley rats (225 to 275 g) received electrolytic lesions of the VMH (*11*). Testing began 1 month after surgery, when all vagotomized rats had stable intakes of food and water and looked healthy. Ten of 12 rats with VMH lesions were hyperphagic in the first 2 weeks after surgery (daily sweet milk intakes > 2 standard deviations larger than control). Eight of these hyperphagic rats were selected randomly and placed on restricted food intake to maintain body weight close to that of rats that received sham operations.

For each test, the vagotomized rats were offered a test diet (Gibco, EC116)

Table 1. Effect of abdominal vagotomy or ventromedial hypothalamic (VMH) lesions on CCK-induced satiety. The data (means ± standard error) show the percentages of inhibition of food intake. The percentage of inhibition of food intake = 100 × 1 − 30-minute intake after CCK-8/ 30-minute intake after saline.

Group	*N*	Dose of CCK-8 (μg/kg)			
		1	2	4	8
Controls	16*	20 ± 9	31 ± 8	60 ± 6	54 ± 9
VMH lesion	8	22 ± 12	20 ± 14	62 ± 7	60 ± 8
Vagotomy†	8	−14 ± 18	2 ± 15	16 ± 15	7 ± 16

*N = eight controls for hypothalamic lesions and eight controls for vagotomy; since there was no difference in the intakes of the two groups, their data were pooled. †Group mean differs significantly from controls ($P < .01$) and VMH lesion ($P < .05$) by Tukey test after analysis of variance (*14*). Analysis of variance was significant for the group effect, $F(2, 29) = 7.51$, $P < .01$, and for dose, $F(3, 42) = 8.45$, $P < .01$, but not for the interaction ($P > .10$). Negative inhibition indicates rats ate more after this dose of CCK-8.

Table 2. Effect of selective vagotomies on CCK-induced satiety. The data (means ± standard error) show the percentages of inhibition of food intake.

Group	*N*	Dose of CCK-8 (μg/kg)			
		1	2	4	8
Controls	11	22 ± 5	43 ± 6	44 ± 4	67 ± 4
Total*	7	22 ± 6	−13 ± 16	−8 ± 17	17 ± 11
Gastric*	11	4 ± 10	11 ± 11	18 ± 14	10 ± 8
Coeliac	8	14 ± 4	31 ± 6	41 ± 12	56 ± 7
Hepatic	5	16 ± 15	41 ± 10	31 ± 8	56 ± 7
Coeliac plus hepatic	7	22 ± 10	27 ± 15	61 ± 10	47 ± 7

*Total and gastric vagotomies differ significantly from controls (Tukey test, $P < .01$) after analysis of variance. Analysis of variance was significant for the group effect, $F(5, 43) = 13.25$, $P < .01$, and for dose, $F(3, 129) = 7.00$, $P < .01$, but not for the interaction ($P > .10$).

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