The cycle rate of the oscillator was a continuous function of the depolarization of the command neuron: this relation is plotted for several current strengths in Fig. 2D. Hyperpolarization of the command neuron was without effect on the motor output. Action potentials were effective in activating the oscillator only if delivered in trains with a frequency high enough to build up a sufficient average depolarization. Again currents of far greater amplitude than those shown did not produce cycling once the electrode was withdrawn just far enough to lose the resting potential. On the late part of the depolarization plateau of Fig. 2C, as well as after termination of the current pulses in Fig. 2, B and C, there is a variation of the membrane potential in time with the motor rhythm. This oscillation within the command neuron may be due to feedback, from a later stage, that serves to entrain the command neuron and assure rhythmicity; yet it is clearly not a necessary part of the oscillatory mechanism (the early part of the record in Fig. 2C shows no oscillation on the plateau, and in Fig. 2B there is no sign at all of oscillation in the command neuron). Although the local oscillator may have the ability to entrain the command unit's transmitter output to its rhythm, oscillation does not require periodic input from the command element; rather, its rate appears to be a direct function of the steady-state input. (Since I could not record simultaneously from command and oscillator neurons, there is no way to be certain that the command neuron makes a direct connection to the oscillator. Yet if any stages intervene between the two they must be nonperiodic as well, since, as previously indicated, the oscillator displays no sign of rhythmic input. Thus the conclusion that the oscillator can convert various levels of steady drive into different frequencies is still valid.)

The complete absence of action potentials in the oscillators is not a surprising finding. It is becoming increasingly clear that many short neurons, with total lengths comparable to their space constants, do not employ action potentials since electronic propagation of signals is sufficient to convey information with acceptably small loss (*5*). In one case a coxal receptor located outside but close to the thoracic ganglion of a crab was shown to convey excitation into the ganglion and elicit a complete reflex entirely without spikes (*6*). Because the oscillators have not been visually identified and measured, their size is not known; but since the ganglion itself is not over 200 to 300 μm in half-width their processes cannot be any longer than those of the coxal receptors, and a similar mode of operation is entirely possible.

It is more difficult to account for the complementary actions of depolarization and hyperpolarization of the oscillator on the motor neurons. No interneurons were encountered with properties that would fit them to be intermediates between oscillators and motor neurons. It is worth speculating that this is another case of an interneuron with a dual action whose followers are either excited or inhibited according to specific postsynaptic responsiveness to the driver neuron's transmitter (*7*). In such a system one group of motor neurons would be depolarized and excited and the other group inhibited by increased transmitter output from the oscillator on its depolarization phase. Then as the oscillator repolarized, its transmitter release would decrease, the excited cells would become silent while the inhibited cells fired on rebound depolarization; the relation of a particular motor neuron's spike pattern to the input drive would be set by its own characteristics of refractoriness and accommodation (*8*). Thus command interneurons and oscillator interneurons both appear to be able to control periodic activity of their follower cells with a smooth, graded release of transmitter whose duration greatly exceeds the cycle period of the controlled output. Others have reported unsuccessful attempts to detect local oscillator cells, which has led to the proposal that all oscillator systems consist of diffuse networks of neurons operating in concert (*9*). Finding discrete oscillator neurons in lobsters and hermit crabs encourages the belief that such neurons are present elsewhere and again demonstrates the parsimony of the crustacean nervous system in utilizing the available neurons.

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Developmental Behaviors: Delayed Appearance in Monkeys Asphyxiated at Birth

Abstract. *Developmental behaviors were studied in monkeys subjected to asphyxia at birth. Visual depth perception, visual placing, and locomotion appeared significantly later than in nonasphyxiated monkeys. After these behaviors had been established in asphyxiates, however, there was little difference from those observed in normal monkeys. These results were compared with reports of permanent learning deficits that occur in monkeys asphyxiated at birth for similar periods of time. Such comparison suggests that the neural structures responsible for the developmental behaviors studied are not damaged by asphyxia to the same extent as those for acquisition. Delay in development may be an early indication of brain damage with subsequent mental retardation.*

A regularly occurring pattern of structural brain damage by birth asphyxia of 10 to 17 minutes duration in rhesus monkeys has been established by Windle and his associates (*1*). Similar lesions have been seen in human infants after birth asphyxia (*2*). In this respect the infant monkey appears

to be more like the human infant at birth than do the offspring of other laboratory animals and provides a better model for experiments of the kind described herein. The results we report here have been obtained with animals physiologically manipulated by asphyxia *at* birth and are therefore not comparable with the results of other experiments obtained with animals in which the physiological manipulation occurs *after* birth.

We examined the development of three types of behavior in normal and neonatally asphyxiated monkeys (*Macaca mulatta*): visual depth perception, visual placing, and independent locomotion. Eight infant monkeys were used. Four, delivered by cesarean section between day 156 and day 159 of gestation (*3*), were subjected to asphyxia for 15 minutes and resuscitated by methods previously described (*4*). The asphyxiated monkeys after resuscitation were kept separately in incubators and given intensive nursing care until their recovery was assured. The other four monkeys were nonasphyxiated controls. Two of them were delivered by cesarean section and two were born spontaneously. Three of the controls and the four asphyxiated animals were housed separately in nursery cages. One control infant was permitted to remain with its mother after birth except during testing.

Development of visual depth perception, visual placing, and locomotion were tested in all the monkeys, beginning on the first postnatal day and continuing daily until criterion was reached. A modified visual cliff apparatus, similar to that described by Gibson and Walk, was used to evaluate the onset of visual depth perception (*5, 6*). It consisted of a Masonite chamber with four walls, each 76 by 76 by 61 cm. The inside was painted in 5-cm red and white checkers. A plate glass shelf in two equal parts each 76 by 38 cm was laid completely across the chamber at a level 20 cm beneath the top of the walls. It provided a solid surface for the animals to walk on. A 10-cm-wide wooden runway, the length of the apparatus, was centered exactly between the two glass shelves and raised 4 cm above them. This divided the chamber into equal halves. Two Masonite panels, each the size of one glass shelf, were also painted in red and white checkers. To convey the impression that there was a shallow surface to step down onto, one panel was placed with the checkered part flush with the underside of one glass shelf. To convey the impression of a deep surface, the second checkered panel was placed 41 cm below the second glass shelf. When viewed from the runway, a sharp drop or "cliff" appeared to be present on that side of the chamber. The testing room was evenly illuminated with fluorescent lights.

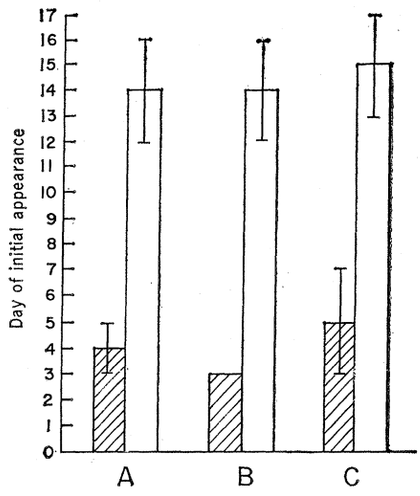

Fig. 1. The effect of neonatal asphyxia on the development of (A) visual depth perception, (B) locomotion, and (C) visual placing. Hatched columns represent normal monkeys; open columns represent asphyxiated monkeys. Number of days are expressed as mean values; line segments show standard deviations. The differences in development on all three behaviors between normal and asphyxiated monkeys are significant at the level of confidence $P < .01$ (t-test).

At each trial the monkey was placed in the center of the runway and left there until it descended to the shallow or deep side or until it had remained on the runway for 2 minutes. A shallow or deep response was scored only when the monkey left the runway within 2 minutes and, with use of all four limbs, moved its body completely onto either surface. A no-descent response was scored if the monkey remained on the runway for 2 minutes. At that time the animal was removed from the apparatus and the trial was over. Six trials were given each day: three with the shallow side on the left and three with the shallow side on the right. Right and left sides were alternated in random sequence. The apparatus was reoriented daily so that the visual environment in relation to the runway was constantly changed. Criterion for the onset of visual depth perception was taken as the first day on which 100 percent choice of the shallow side was reached.

Visual placing responses were tested by two methods immediately before or after depth perception sessions. The first method was to hold the monkey so that its head and limbs were unrestrained and to move it slowly toward the edge of a table until it extended its forelimbs and placed them on the table in anticipation of "landing." Three trials a day were given to evaluate this response. The second method employed the visual cliff apparatus. Again, the monkey was supported so that its head and limbs were unrestrained; it was held in this position 10 to 13 cm above, and directly over, first the shallow side and then the deep side of the apparatus. The object of this test was to see when the monkey perceived the difference between the two sides and placed its limbs only on the shallow side. A positive response consisted of placing its limbs when it was over the shallow surface and flexing them away from the surface when it was over the deep side. Three trials a day were given, each consisting of one presentation of the shallow side and one of the deep side. These presentations were in random order. Criterion for visual placing was considered to have been reached on the first day that 100 percent positive responses were attained with both methods.

Locomotor development was evaluated daily by taking the animal out of its cage or incubator and placing it on a low table. The appropriate level of locomotion was accepted when the animal used all four limbs to crawl and could change its position independently.

The results are given in Fig. 1. Depth perception in the infant monkey normally appeared between postnatal days 3 and 5, but its onset was delayed in the asphyxiated monkeys until 12 to 16 days of age. Appearance of locomotion was almost parallel with that of depth perception: 3 days for the normal and 12 to 16 days for the asphyxiated monkeys. Visual placing responses appeared between days 3 and 7 in normal monkeys but not until days 13 to 17 in asphyxiated monkeys. These threefold to fourfold differences between the two groups of animals are significant ($P < .01$).

The time of appearance of depth perception and independent locomotion in normal infant monkeys confirms that reported by others (*7*). We believe ours to be the first report of development of these three types of behavior in monkeys subjected to birth asphyxia. Although these functions were delayed in the asphyxiated monkeys, there was little difference from the normal monkeys in the effectiveness with which the responses were made after they had become established.

Previous investigators have suggested (*6*) that visual depth perception is adaptive and that one of the keys to survival of a species is development of visual depth perception by the time locomotion becomes independent. Such an ability appears to be innate and does not seem to depend on learning during early life. Evidence for this was found in the time of onset of depth perception in other animals: the chick and goat, 1 day; the rat and cat, 3 or 4 weeks; the human infant, 6 to 10 months (*6, 7*). According to our own observations, the onset of these two responses is nearly coincident with the development of visual placing.

The distribution of focal lesions in rhesus monkeys subjected to asphyxia at birth was found to involve afferent systems, such as somethetic, auditory, and vestibular, but the visual pathways were spared (*1*). Although the brains of the still living monkeys of the present study will not be examined for some time, we assume that they have been injured and that the pattern of damage is the same as that in other animals subjected to birth asphyxia for a comparable period of time. In spite of this presumed brain damage, these developmental behaviors were fully expressed in the asphyxiated monkeys, although significantly later than in nonasphyxiated monkeys.

Monkeys surviving neonatal asphyxia after 8 to 10 years showed severe memory deficits for events in the immediate past (*8*). Short-term memory deficits also occurred within the same time limits in monkeys at 10 months after birth asphyxia (*9*). In addition, Saxon and Ponce (*10*) found evidence of performance deficits of asphyxiated monkeys at 6 to 7 months of age; these were observed in learning set problems, delayed response, and perseveration tasks, all of which depend upon visual stimuli. When compared with the establishment of these developmental behaviors, deficits in learning and memory (*10, 11*) (although significantly delayed) suggest that brain damage by neonatal asphyxia can result in a degree of dissociation of developmental and acquired behavior. Whatever organic basis there may be to explain our findings remains to be discovered. Nevertheless, the late appearance of developmental behaviors may be one of the first observable signs of brain damage and mental retardation. Rather than regarding these events as transient and negligible, they should be systematically evaluated in the human infant with a view toward early diagnosis.

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Malaria Resistance: Artificial Induction with a Partially Purified Plasmodial Fraction

D'Antonio *et al.* (*1*) have described a fraction of *Plasmodium berghei* that confers a measure of protection against subsequent challenge by the organism. However, we now call attention to an impression given by their paper which we regard as misleading. They state: "In contrast to other methods, plasmodia isolated in this manner [French pressure cell] have been shown by serologic and ultrastructural examinations to be free from significant amounts of contamination by constituents of the host cell." They then refer to an article by us (*2*) and one by Killby and Silverman (*3*), implying that these articles support their statement on ultrastructural grounds. In point of fact, neither article supports their contention. For example, in the article by Killby and Silverman, figure 5 shows two parasites surrounded by large amounts of cell debris, including many membrane vesicles. Our results [figure 2b in (*2*)] showed the same thing—that is, the complete fragmentation of most parasites by French pressure cell lysis of the parasitized erythrocytes. It is clear that many of the membrane vesicles in these micrographs originated from the host erythrocyte since they are not "solubilized." Otherwise, D'Antonio *et al.* would not have been able to prepare normal "erythrocyte sediment" by the same technique.

D'Antonio *et al.* have lowered the initial lysis pressure from that used to obtain the preparations examined by Killby and Silverman and by us. However, in the absence of published low power electron micrographs showing both (i) many intact parasites and (ii)