Primate Neurobiology: Neurosurgery with Fetuses

Although experimental sciences advance as much by the development and application of techniques as by new theories, word of a new technique sometimes fails to spread beyond a small group of researchers, even when the method could be widely used by scientists in related fields. Such a lack of communication occurred when investigators studying pregnancy and fetal development in primates devised ways to remove fetuses from the womb, replace them, and deliver them at term. This method is just what many neurobiologists have needed in their efforts to study the origins of behavior in primates, but more than a decade passed between the time the technique became feasible and the time the neurobiologists actually used it.

Among the first neurobiologists to operate on primate fetuses are Ronald Myers and his associates at the National Institute of Neurological and Communicative Disorders and Stroke. More than 10 years ago, Myers clamped the umbilical cords of fetal monkeys at midgestation in order to study the consequences of the deprivation of oxygen to the brain. Although Myers produced changes to the fetuses' nervous systems indirectly, his work raised the possibility that the nervous systems of primate fetuses could be altered directly. Myers speculates that neurobiologists were slow to adopt the fetal surgery techniques because researchers generally believe that the primate fetus is inaccessible—that if the uterus is opened the pregnancy will end.

No single methodological breakthrough is responsible for the feasibility of prenatal neurosurgery in primates. Instead, many small improvements in techniques developed by a number of investigators made this work possible. For example, researchers learned to select an appropriate uterine incision that would not damage the fragile attachment of the placenta to the uterus. They learned the best ways to preserve the amniotic fluid. And they learned to use smooth-muscle relaxants to inhibit uterine contractions so the monkeys do not expel their fetuses after surgery. By using these methods, investigators can completely remove a fetus from the uterus for considerable periods of time, subject it to complicated surgery, and return it to the uterus.

Some neurobiologists now say that surgery on primate fetuses will provide them with a new window on the complex interrelationships between the central nervous system, the environment, and behavior. For years they have relied on the standard experimental approach of excising a region of an animal's central nervous system and then observing how its behavior changes in various environments and at various stages of its life. Neurobiologists find that when they carry out such an experiment at different stages of the development of the central nervous system, they can obtain information that sometimes helps them discriminate between the effects of genes and environmental feedback on behavior patterns. Since so much of the structure of the primate central nervous system is developed before birth, many neurobiologists say that prenatal neurosurgery is essential for the further advancement of knowledge in this field.

Relevance to Human Disorders

A likely spin-off from the neurobiologists' work on primate fetuses is a new understanding of nervous-system damage in humans. Edward Taub of the Institute for Behavioral Research in Silver Spring, Maryland, points out that babies often suffer damage to the central nervous system at birth or before. This damage can occur, for example, as a result of a forceps delivery, too little or no oxygen for a period during or before birth, genetic defects, or spasms in blood vessels. The current neurobiological research can help medical scientists understand the mechanisms that enable (or prevent) recovery of function when a fetus or a baby's central nervous system is injured. For these purposes, it is essential that such investigations be carried out in a species whose brain is phylogenetically close—in size, organization, and tempo of development—to the human brain.

Some years ago, Myers introduced the technique of fetal surgery to Taub and his associate Gilbert Barro. For more than a decade, Taub and Barro had been studying the control of movement in monkeys. A commonly believed hypothesis has been that monkeys and other primates (including humans) are able to learn to use their limbs as a result of both spinal reflex activity and sensory feedback from movement. For example, an animal's forward movement during walking would result from spinal reflexes produced by the previous step. It would learn to make an appropriate turn in a maze because it received sensory feedback from the correct movement.

Taub and Barro tested this hypothesis by cutting the dorsal, or sensory, roots of monkeys' spinal nerves. After these nerves are cut, the animals have no spinal reflexes or sensations in their limbs. Adult, juvenile, and even newborn monkeys with neither sensory feedback nor spinal reflexes were found to still be able to learn a full array of movements. It remained possible that the animals learned movements through visual cues, so Taub and Barro blinded the newborn monkeys after severing their dorsal roots. Once again, the monkeys learned to move and use their extremities. For example, a blinded infant monkey with severed dorsal roots could use its limbs to walk and bring raisins to its mouth.

As a result of these studies, Taub, Barro, and David Martin of Litton-Bionetics Laboratories in Rockville, Maryland, became interested in seeing whether fetuses whose dorsal roots were cut still learn to use their limbs. The newborn monkeys may have learned these movements in the womb, where they had sensory feedback from their own movements. They found, however, that when they operated on fetuses that were two-thirds of the way through gestation, the animals subsequently learned all appropriate limb movements. Last year, Taub, Barro, and Martin operated on monkey fetuses that were two-fifths of the way through gestation. Once again, the monkeys, when born, learned all limb movements. Taub doubts that they can go back much further in fetal development to study this problem because the structures of the nervous system of interest to them would be too small to operate on. Two-fifths of the way through gestation, the dorsal roots are not visible to the naked eye. Consequently, they perform surgery with the aid of a microscope that magnifies the surgical field as much as 25 times. However, before the monkeys are two-fifths of the way through gestation, the nerve roots, although visible under magnification, are so small that, Taub says, "the tremors of the hand—even a rock-steady hand—would render the surgery impossible."

Taub interprets his results as indicating that many motor programs are

0083-8075/78/0303-0960$00.50/0

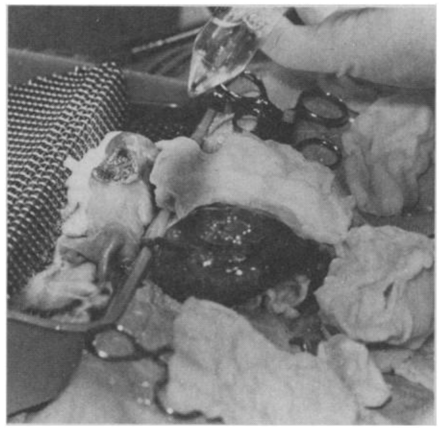
part of a primate's genetic endowment. No sensory feedback or spinal reflex loops are necessary for learning the repertoire of movements, but, since the animals with severed dorsal roots are clumsy, sensory feedback is necessary for "fine tuning," according to Taub.

Intrauterine surgery is now being used by Patricia Goldman of the National Institute of Mental Health to study the central nervous system. She and her associates are studying the development and modifiability of primate brains.

When part of an animal's brain is destroyed, behavior controlled by that area is altered, but the extent of alteration frequently depends on the age of the animal at the time of injury. In primates, including humans, the effects of brain injury are often less if the injury occurs when the animal is young than when it is mature. It is generally not known, however, whether prenatal brain injuries are also more likely to be corrected or compensated for. It is possible that the fetal brain is, if anything, more resilient than the brain of a newborn. On the other hand, it could be that a brain injury during fetal development would interrupt a chain of essential events and have far more serious consequences than a similar injury occurring after birth.

Goldman and her associates have studied the consequences of injury to the prefrontal cortex, an area of the brain necessary for cognitive functions. This region has undergone an enormous expansion during the course of primate evolution and has reached its peak size in humans. In both monkeys and humans, damage to the prefrontal cortex more severely affects behavior if it occurs during adulthood than during infancy. Monkeys whose dorsolateral prefrontal cortices are removed when they are juveniles or adults do very poorly on learning tests. Monkeys that have undergone surgery when they were 50 days of age perform normally on these tests until they are about 1 year old, after which their performance may deteriorate. Their performance, however, never becomes as poor as the monkeys that had undergone surgery as juveniles or adults. One explanation of this deterioration is that it is only after the animals are 1 year old that the prefrontal cortex is used in these learning tasks.

Goldman and her colleagues report that monkeys whose prefrontal cortices are removed about two-thirds of the way through gestation can apparently completely overcome the effects of the operation. In fact, one such monkey actually performed within the range of normal monkeys on all the learning tests it was



A monkey fetus, two-thirds of the way through gestation, is prepared for surgery. Its spinal cord is exposed so that the dorsal roots can be severed. The fetus is connected by its umbilical cord to the placenta, which can be seen on the right. The size of the fetus can be judged relative to the hand in the upper right-hand corner of the photograph. [Source: E. Taub, P. N. Perrella, E. A. Miller, G. Barro, *Biological Psychiatry* **10**, 619 (1975)]

given during a 2½-year period of postnatal development. On killing the monkey at that age, the investigators found that its brain was morphologically very different from the brains of normal monkeys and even from the brains of monkeys whose prefrontal cortices were removed after birth. It had anomalous folds and indentations far from as well as near the site of the lesion. In addition, neurons from the monkey's thalamus, which normally project to the prefrontal cortex, did not degenerate as they do when the operation is performed after birth. Instead, they seemed to have been rerouted to other parts of the brain. According to Goldman, it is tempting to speculate that some or all of the morphological changes in the monkey's brain allowed it to compensate for the loss of its prefrontal cortex. She points out, however, that further study is necessary to establish the nature and causes of these changes.

In recent years, many neurobiologists have studied the visual system of monkeys in order to determine whether neural connections are modified by experience and, if so, how. For example, David Hubel and Torsten Wiesel of Harvard Medical School found critical periods in primates, occurring shortly after birth, during which vision can be permanently altered by a lack of experience. For example, if one of a monkey's eyes is sutured shut during this critical period, it will lose its ability to see with that eye.

Neurons from the retina of each eye are the first link in a chain of neurons that terminates at the visual cortex, where the brain is organized in columns. One column of cortical cells is served by neurons from the right eye, the adjacent column by neurons from the left eye, the next column by the right eye, and so on. Hubel and Wiesel found that when a monkey is prevented from using one eye during the critical period, the columns in the visual cortex that normally receive neural fibers from that eye are larger than those receiving fibers from the other eye. It seemed possible that fibers from the sighted eye spread and took over the columns originally destined for the deprived eye. Pasko Rakic of Harvard Medical School operated on monkey fetuses in order to investigate this hypothesis and found evidence that contradicts it.

Rakic used the fetuses to study how and when the neural connections of the visual system develop. For example, he injected radioactively labeled compounds into one eye of a fetus, returned the fetus to the womb, and 2 weeks later removed the fetus and examined its brain. The radioactive labeling of the developing nerve fibers permitted Rakic to determine when they reach the visual cortex and how they are organized there.

Rakic discovered that neurons from both eyes terminate in each column of the visual cortex at the midgestational period. About 3 weeks before birth, however, these neurons begin to segregate, but they do not become completely segregated into alternating columns until after birth. Therefore, Rakic concludes, when an eye is sutured shut during the critical period, the neurons from the nondeprived eye simply fail to retract. Rakic points out that the changes in neural connections that occur when an eye is sutured shut could represent an arrest of normal development rather than an active rearrangement of neural connections.

Studies of the visual system may be pertinent to studies of other areas of the brain. For example, Goldman and others have evidence that columns like those in the visual system may be a general form of brain organization. Such columns occur in the motor and prefrontal cortex of the monkey, she finds, even though these areas of the brain receive no direct sensory input from the environment.

Thus far, only a few neurobiologists have used the technique of operating on primate fetuses. Taub predicts that, as word of the success of this method spreads, a new discipline, which he calls "prenatal neuropsychology" or "the study of the prenatal origins of behavior," will become established.

—Gina Bari Kolata