

the neurobiology of FEAR

RESEARCHERS ARE TEASING APART THE PROCESSES IN THE BRAIN THAT GIVE RISE TO VARIOUS FEARS IN MONKEYS. THE RESULTS MAY LEAD TO NEW WAYS TO TREAT ANXIETY IN HUMANS

BY NED H. KALIN

OVER THE YEARS, most people acquire a repertoire of skills for coping with a range of frightening situations. They will attempt to placate a vexed teacher or boss and will shout and run when chased by a mugger. But some individuals become overwhelmed in circumstances others would consider only minimally stressful: fear of ridicule might cause them to shake uncontrollably when called on to speak in a group, or terror of strangers might lead them to hide at home, unable to work or shop for groceries. Why do certain people fall prey to excessive fear?

At the University of Wisconsin–Madison, my colleague Steven E. Shelton and I are addressing this problem by identifying specific brain processes that regulate fear and its associated behaviors. Despite

the availability of noninvasive imaging techniques, such information is still extremely difficult to obtain in humans. Hence, we have turned our attention to another primate, the rhesus monkey (*Macaca mulatta*). These animals undergo many of the same physiological and psychological developmental stages that humans do, but in a more compressed time span. As we gain more insight into the nature and operation of neural circuits that modulate fear in monkeys, it should be possible to pinpoint the brain processes that cause inordinate anxiety in people and to devise new therapies to counteract it.

Effective interventions would be particularly beneficial if they were applied at an early age. Growing evidence suggests overly fearful youngsters are at high risk for later emotional distress. Jerome Ka-

gan and his colleagues at Harvard University have shown, for example, that a child who is profoundly shy at the age of two years is more likely than a less inhibited child to suffer from anxiety and depression later in life.

This is not to say these ailments are inevitable. But it is easy to see how excessive fear could contribute to a lifetime of emotional struggle. Consider a child who is deeply afraid of other children and is therefore taunted by them at school. That youngster might begin to feel unlikable and, in turn, to withdraw further. With time the growing child could become mired in a vicious circle leading to isolation, low self-esteem, underachievement, and the anxiety and depression noted by Kagan.

There are indications that unusually fearful children might also be prone to

physical illness. Many youngsters who become severely inhibited in unfamiliar situations chronically overproduce stress hormones, including the adrenal product cortisol. In times of threat, these hormones are critical. They ensure that muscles have the energy needed for “fight or flight.” But some evidence indicates long-term elevations of stress hormones may contribute to gastric ulcers and cardiovascular disease.


Further, through unknown mechanisms, fearful children and their families are more likely than others to suffer from allergic disorders. And in rodents and nonhuman primates, persistent elevation

of cortisol has been shown to increase the vulnerability of neurons in the hippocampus to damage by other substances; this brain region is involved in memory, motivation and emotion. Human neurons probably are affected in a similar way, although direct evidence is awaited.

When we began our studies two decades ago, Shelton and I knew we would first have to find cues that elicit fear and identify behaviors that reflect different types of anxiety. With such information in hand, we could proceed to determine the age at which monkeys begin to match defensive behaviors selectively to specific cues. By also determining the

parts of the brain that reach maturity during the same time span, we could gain clues to the regions that underlie the regulation of fear and fear-related behavior.

The experiments were carried out at the Wisconsin Regional Primate Research Center and the Harlow Primate Laboratory, both at the University of Wisconsin–Madison. We discerned varied behaviors by exposing monkeys between six and 12 months old to three related situations. In the alone condition, an animal was separated from its mother and left by itself in a cage for 10 minutes. In the no-eye-contact condition, a person stood motionless outside the cage and avoided



RHESUS MONKEY REGISTERS ALARM (*right*) as another monkey approaches her baby. The mother's fear is evident in her “threat” face: the open mouth and piercing stare serve to intimidate would-be attackers and intruders.

looking at the solitary infant. In the stare condition, a person was again present and motionless but, assuming a neutral expression, peered directly at the animal. These conditions are no more frightening than those that primates encounter frequently in the wild or those that human infants encounter every time they are left at a day-care center.

Three Typical Fear Behaviors

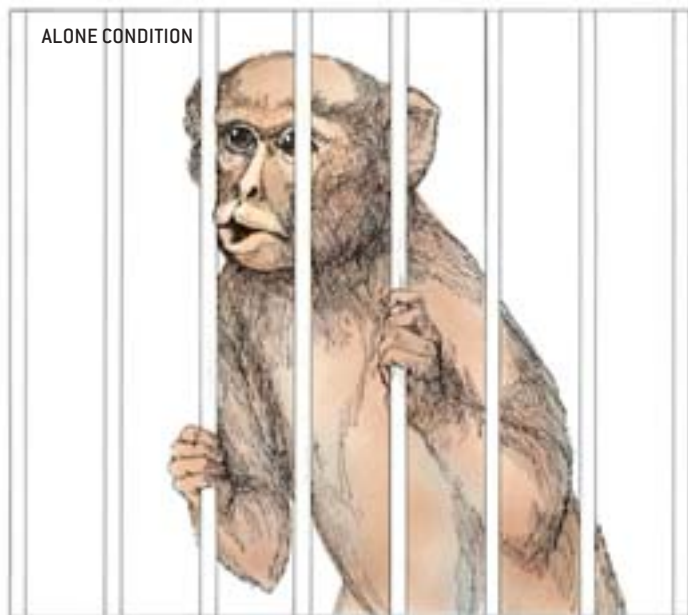
IN THE ALONE CONDITION, most monkeys became very active and emitted frequent “coo” calls. These fairly melodious sounds are made with pursed lips. They start at a low pitch, rise higher and then fall. More than 40 years ago Harry F. Harlow, then at Wisconsin, deduced that when an infant monkey is separated from its mother, its primary goal is affiliative—it yearns to regain the closeness and security provided by nearness to the parent. Moving about and cooing help to draw the mother’s attention.

In contrast, in the more frightening no-eye-contact situation, the monkeys reduced their activity greatly and sometimes “froze,” remaining completely still for prolonged periods. When an infant spots a possible predator, its goal shifts from attracting the mother to becoming inconspicuous. Inhibiting motion and freezing—common responses in many species—reduce the likelihood of attack.

If the infant perceives that it has been detected, its aim shifts to warding off an attack. So the stare condition evoked a third set of responses. The monkeys made several hostile gestures: “barking” (forcing air from the abdomen through the vocal cords to emit a growl-like sound), staring back, producing so-called threat faces, baring their teeth and shaking the cage. Sometimes the animals mixed the threatening displays with submissive ones, such as fear grimaces, which look something like wary grins, or grinding of the teeth. In this condition, the animals cooed more than they did when alone. (We have come to think the cooing displayed in the stare condition may serve a somewhat different function than it does in the alone situation.)

Monkeys, by the way, are not unique in becoming aroused by stares and in us-

THREE experimental conditions elicit distinct fear-related behaviors in rhesus monkeys older than about two months. When isolated in a cage (*left*), youngsters become quite active and emit “coo” sounds to attract their mothers. If a human appears but avoids eye contact (*center*), the monkeys try to evade discovery, such as by staying completely still (*freezing*) or hiding behind their food bin. If the intruder stares at the animals (*right*), they become aggressive.



ing them reciprocally to intimidate predators. Animals as diverse as crabs, lizards and birds all perceive staring as a threat. Some fishes and insects have evolved protective spots that resemble eyes; these spots either avert attacks completely or redirect them to nonvital parts of the body. In India, field-workers wear face masks behind their heads to discourage tigers from pouncing at their backs. Studies of humans show that we, too, are sensitive to direct gazes: brain activity increases when we are stared at, and people who are anxious or depressed tend to avoid direct eye contact.

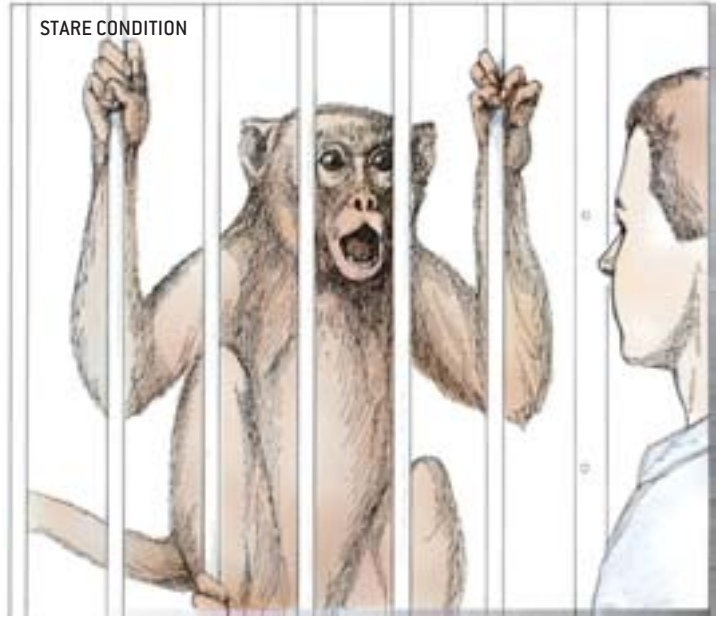
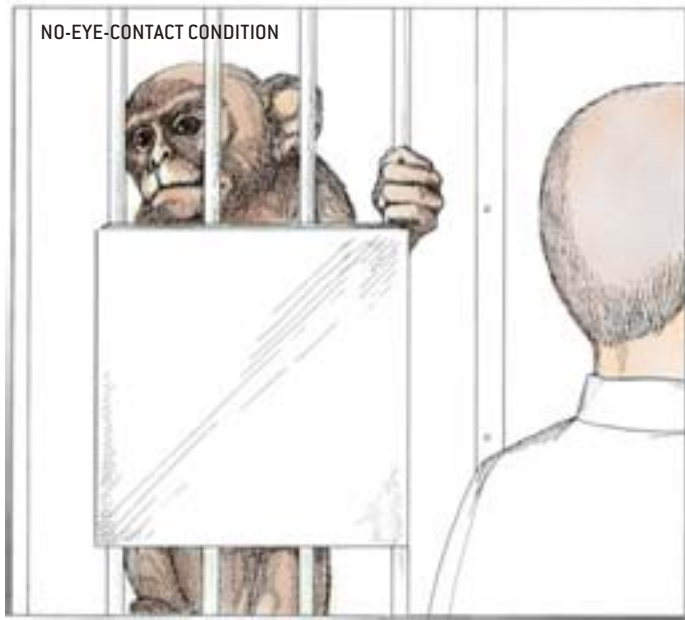
Having identified three constellations of defensive behaviors, we set about determining when infant monkeys first begin to apply them effectively. Several lines of work led us to surmise that the ability to make such choices emerges sometime around an infant’s two-month birthday. For instance, rhesus mothers generally permit children to venture off with their peers at that time, presumably because the adults are now confident that the infants can protect themselves reasonably

TYPICAL BEHAVIORS induced by the alone, no-eye-contact and stare conditions in the laboratory—such as cooing (*left*), freezing (*center*) and hostile display of the teeth (*right*)—are also seen in frightened infants and adults living in the wild. In this case, the setting is Cayo Santiago, an island off the mainland of Puerto Rico.

well. We also knew that by about 10 weeks of age infant monkeys respond with different emotions to specific expressions on other monkeys’ faces—a sign that at least some of the innate wiring or learned skills needed to discriminate threatening cues are in place.

To establish the critical period of development, we examined four groups of monkeys ranging in age from a few days to 12 weeks old. We separated the babies from their mothers and let them acclimate to an unfamiliar cage. Then we exposed





them to the alone, no-eye-contact and stare conditions. All sessions were videotaped for analysis.

We found that infants in the youngest group (newborns to two-week-olds) engaged in defensive behaviors. But they lacked some motor coordination and seemed to act randomly, as if they were oblivious to the presence or gaze of the human intruder. Babies in our two intermediate-age groups had good motor control, but their actions seemed unrelated to the test condition. This finding meant motor

control was not the prime determinant of selective responding.

Only animals in our oldest group (nine- to 12-week-olds) conducted themselves differently in each situation, and their reactions were both appropriate and identical to those of mature monkeys. Nine to 12 weeks, then, is the critical age for the appearance of a monkey's ability to adaptively modulate its defensive activity to meet changing demands.

Studies by other workers, primarily with rodents, suggested that three inter-

connected parts of the brain regulate fearfulness. We suspected that these regions become functionally mature during the nine- to 12-week period and thus give rise to the selective reactivity we observed. One of these regions is the prefrontal cortex, which takes up much of the outer and side areas of the cerebral cortex in the frontal lobe [see illustration on next page]. A cognitive and emotional area, the prefrontal cortex is thought to participate in the interpretation of sensory stimuli and is probably a site where



NED H. KALIN (photographs); CAROL DONNER (illustrations)

the potential for danger is assessed.

The second region is the amygdala, a part of a primitive area in the brain called the limbic system (which includes the hippocampus). The limbic system in general and the amygdala in particular have been implicated in generating fear.

instance, during this time the formation of synapses (contact points between neurons) has been shown to reach its peak in the prefrontal cortex and the limbic system (including the amygdala), as well as in the motor and visual cortices and other sensory areas. Patricia S. Goldman-

strangers. They also become adept at what is called social referencing; they regulate their level of fear based on the expressions they observe on a parent's face.

But what of the hypothalamus, the third brain region we assumed could participate in regulating fear-related behav-

Levels of stress hormones influence how appropriately animals and people behave in the face of fear.

The final region is the hypothalamus. Located at the base of the brain, it is part of the hypothalamic-pituitary-adrenal system. In response to stress signals from elsewhere in the brain, such as the limbic system and other cortical regions, the hypothalamus secretes corticotropin-releasing hormone. This small protein spurs the pituitary gland, located just below the brain, to secrete adrenocorticotropic hormone (ACTH), which prods the adrenal gland to release cortisol, which prepares the body to defend itself.

In neuroanatomic data collected in other laboratories, we found support for our suspicion that maturation of these brain regions underlies selective responding in the nine- to 12-week period. For

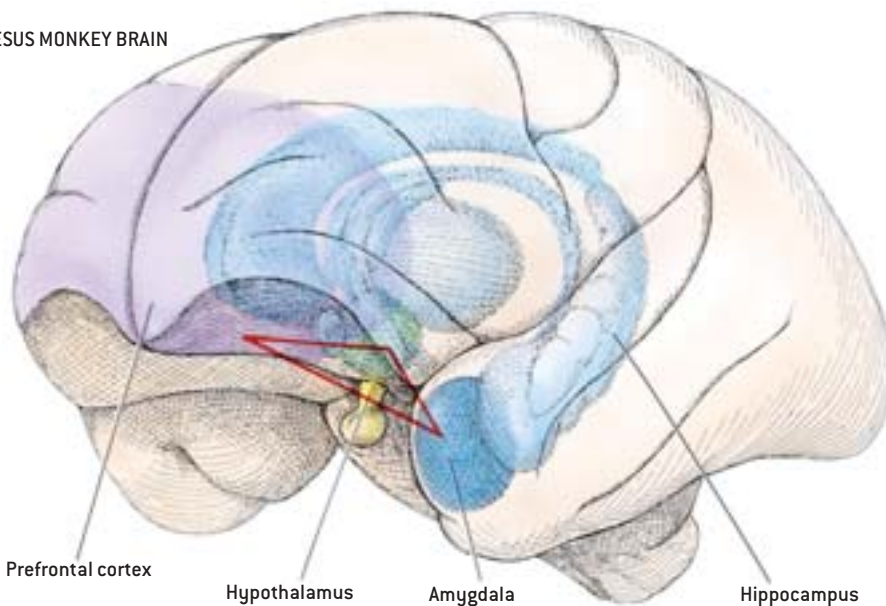
Rakic of Yale University has also established that as the prefrontal cortex matures in rhesus monkeys, the ability to guide behavior based on experience emerges. This skill is necessary if one is to contend successfully with danger.

Maturation of the prefrontal cortex likewise seems important for enabling humans to distinguish among threatening cues. Harry T. Chugani, then at the University of California at Los Angeles, and his co-workers have shown that activity in the prefrontal cortex increases when human offspring are seven to 12 months of age. During this span—which appears to be analogous to the time when monkeys begin to respond selectively to fear—children begin to display marked fear of

Published research did not tell us much about its development or about the development of the complete hypothalamic-pituitary-adrenal system in monkeys. Our own investigations, however, revealed that the full system matures in parallel with that of the prefrontal cortex and the limbic system.

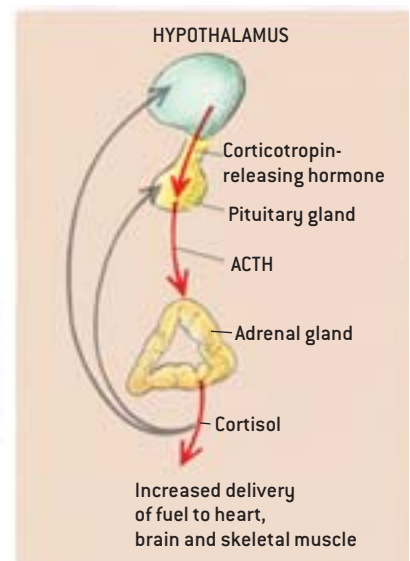
In these studies, we used the pituitary hormone ACTH as a marker of the system's function. We again examined four groups of rhesus infants from a few days old to 12 weeks old. From each subject, we measured ACTH levels in blood drawn while the baby was with its mother. This reading provided a baseline. We also measured ACTH levels in blood samples obtained 20 minutes after the infant

RHESUS MONKEY BRAIN



THREE BRAIN REGIONS that are interconnected by neural pathways (shown schematically by red lines) are critically important in regulating fear-related behaviors. The prefrontal cortex (purple) participates in assessing danger. The amygdala (dark blue) is a major constituent of the emotion-producing

limbic system (light blue). And the hypothalamus (green), in response to signals from the prefrontal cortex, amygdala and hippocampus, directs the release of hormones (red arrows in box) that support motor responses to perceived threats. (Gray arrows represent inhibitory activity by cortisol.)



was separated from its parent. Hormonal levels rose in all four age groups during separation, but they jumped profoundly in the oldest (nine- to 12-week-old) monkeys.




The relatively weak response in the younger animals, particularly in those under two weeks old, is consistent with findings in rat pups, whose stress hormone response is also blunted during the first two weeks of life. The development of the rodent and primate stress hormone system may well be delayed during early life to protect young neurons from the potentially damaging effects of cortisol.

Assured that the hypothalamic-pituitary-adrenal system becomes functionally mature by nine to 12 weeks, we pressed the inquiry forward to determine whether levels of cortisol and ACTH might partly account for individual differences in defensive behavior. We were also curious to know whether the responses of the infants resembled those of their mothers; a correspondence would indicate that further analyses of mothers and their infants could help reveal the relative contributions of inheritance and learning to fearfulness. We mainly examined the propensity for freezing, which we had earlier found was a stable trait in our subjects.

Maturing Fear Response

IN ONE SET OF STUDIES, we measured baseline levels of cortisol in monkeys four months to a year old and then observed how much time the youngsters froze in the no-eye-contact condition. Monkeys that started off with relatively low levels of cortisol froze for shorter periods than did their counterparts with higher cortisol levels—a pattern we also noted in separate studies of adult females. In other studies, we observed that as youngsters pass through their first year of life, they become progressively like their mothers hormonally and behaviorally. By the time infants are about five months old, their stress-induced rises in ACTH levels parallel those of their mothers. And by the time they are a year old, the duration of freezing in the no-eye-contact condition also corresponds to that of the mother.

Strikingly, some of these results echoed those obtained in humans. Extremely in-

	 COOING	 FREEZING	 BARKING
MORPHINE (OPIATE)	Decreases	No effect	No effect
NALOXONE (OPIATE BLOCKER)	Increases	No effect	No effect
DIAZEPAM (BENZODIAZEPINE)	No effect	Decreases	Decreases

EFFECTS ON COOING, FREEZING AND BARKING were evaluated some years ago for three drugs that act on neurons responsive to opiates (*top two rows*) or to benzodiazepines (*bottom row*). The results implied that opiate-sensitive pathways in the brain control affiliative behaviors (those that restore closeness to the mother, as cooing often does), whereas benzodiazepine-sensitive pathways control responses to immediate threats (such as freezing and barking). Newer evidence generally supports this conclusion but adds some complexity to the picture.

hibited children often have parents who suffer from anxiety. Moreover, Kagan and his colleagues have found that basal cortisol levels are predictive of such children's reaction to a frightening situation. They measured cortisol concentrations in saliva of youngsters at home (where they are presumably most relaxed) and then observed the children confronting an unfamiliar situation in the laboratory; high basal cortisol levels were associated with greater inhibition in the strange setting.

These similarities between humans and monkeys again imply that monkeys are reasonable models of human emotional reactivity. The link between basal cortisol levels and duration of freezing or inhibition suggests as well that levels of stress hormones influence how appropriately animals and people behave in the face of fear. (This effect may partly be mediated by the hippocampus, where the concentration of cortisol receptors is high.) And the likeness of hormonal and behavioral responses in mothers and infants implies that genetic inheritance

might predispose some individuals to extreme fearfulness, although we cannot rule out the contribution of experience.

No one can yet say to what extent the activity of the hypothalamic-pituitary-adrenal system controls, and is controlled by, other brain regions that regulate the choice of defensive behavior. We have, however, begun to identify distinct neurochemical circuits, or systems, in the brain that affect different behaviors. The two systems we have studied most intensely seemed at first to have quite separate functions. But more recent work implies that the controls on defensive behavior are rather more complicated than the original analyses implied.

We gathered our initial data more than a decade ago by treating six- to 12-month-old monkeys with two different classes of neuroactive chemicals—opiates (morphinelike substances) and benzodiazepines (chemicals that include the anti-anxiety drug diazepam, or Valium). We chose to look at opiates and benzodiazepines because neurons that release or take

THE AUTHOR

NED H. KALIN, a clinician and researcher, directs the Health Emotion Research Institute. He is Hedberg Professor of Psychiatry and Psychology and chairman of the department of psychiatry at the University of Wisconsin—Madison Medical School. He is also a scientist at the Wisconsin Regional Primate Research Center and the Harlow Primate Laboratory at the university. He earned his B.S. degree in 1972 from Pennsylvania State University and his M.D. in 1976 from Jefferson Medical College in Philadelphia. Before joining his current departments, he completed a residency program in psychiatry at Wisconsin and a postdoctoral fellowship in clinical neuropharmacology at the National Institute of Mental Health.



up those chemicals are abundant in the prefrontal cortex, the amygdala and the hypothalamus. The opiates are known to have natural, or endogenous, counterparts, called endorphins and enkephalins, that serve as neurotransmitters; after the endogenous chemicals are released by certain neurons, they bind to receptor molecules on other nerve cells and thereby increase or decrease nerve cell activity. Receptors for benzodiazepines have been identified, and investigators are attempting to characterize endogenous benzodiazepinelike molecules.

Selective Drug Effects

ONCE AGAIN, our subjects were exposed to the alone, no-eye-contact and stare conditions. We delivered the drugs before the infants were separated from their mothers and then recorded the animals' behavior. Morphine decreased the amount of cooing normally displayed in the alone and stare conditions. Conversely, cooing was increased by naloxone, a compound that binds to opiate receptors but blocks the activity of morphine and endogenous opiates. Yet morphine and naloxone had no influence on the frequency of stare-induced barking and other hostile behaviors, nor did they influence duration of freezing in the no-eye-contact

situation. We concluded that opiate-using neural pathways primarily regulate affiliative behaviors (such as those induced by distress over separation from the mother), but those pathways seem to have little power over responses to direct threats.

The benzodiazepine we studied—diazepam—produced a contrary picture. The drug had no impact on cooing, but it markedly reduced freezing, barking and other hostile gestures. Thus, benzodiazepine-using pathways seemed primarily to influence responses to direct threats but to have little power over affiliative behavior.

We still think the opiate and benzodiazepine pathways basically serve these separate functions. But the simple model we initially envisioned grew more interesting as we investigated two additional drugs: a benzodiazepine called alprazolam (Xanax) and a compound called beta-carboline, which binds to benzodiazepine receptors but elevates anxiety and typically produces effects opposite to those of diazepam and its relatives. When we administered alprazolam in doses that lower anxiety enough to decrease freezing, this substance, like diazepam, minimized hostility in the threatening, stare condition. And beta-carboline enhanced hostility. No surprises here. Yet, unlike diazepam,

these drugs modulated cooing, which we had considered to be an affiliative (opiate-controlled) behavior, not a threat-related (benzodiazepine-controlled) one. Moreover, both these compounds decreased cooing. We cannot explain the similarity of effect, but we have some ideas about why drugs that act on benzodiazepine receptors might influence cooing.

It may be that, contrary to our early view, benzodiazepine pathways can in fact regulate affiliative behavior. We favor a second interpretation, however. Cooing displayed in the stare condition may not solely reflect an affiliative need (a desire for mother's comfort); at times, it may also be an urgent, threat-induced plea for immediate help. One behavior, then, might serve two different functions and be controlled by different neurochemical pathways. (This conclusion was strengthened for me when I tried to photograph a rhesus infant that had become separated from its mother in the wild, where we are now conducting additional studies. Its persistent, intense coos attracted the mother, along with a pack of protectors. The strategy worked: I retreated rapidly.)

More generally, our chemical studies led us to suspect that the opiate- and benzodiazepine-sensitive circuits both oper-



RELAXED MOTHER (*left*) barely reacts to the presence of the camera-wielding author, whereas a more sensitive mother (*right*) becomes frightened, as evinced by her "fear grimace." The author hopes explorations of the neural bases for such differences in monkeys will facilitate development of new therapies for excessively anxious humans.

ate during stress; the relative degree of activity changes with the characteristics of a worrisome situation. As the contribution of each pathway is altered, so, too, are the behaviors that appear.

Exactly how neurons in the opiate and benzodiazepine pathways function and how they might cooperate are unclear. But one plausible scenario goes like this: When a young monkey is separated from its mother, opiate-releasing and, consequently, opiate-sensitive neurons become inhibited. Such inhibition gives rise to yearning for the mother and a generalized sense of vulnerability. This reduction of activity in opiate-sensitive pathways enables motor systems in the brain to produce cooing. When a potential predator appears, neurons that secrete endogenous benzodiazepines become suppressed to some degree. This change, in turn, leads to elevated anxiety and the appearance of behaviors and hormonal responses that accompany fear. As the

sense of alarm grows, motor areas prepare for fight or flight. The benzodiazepine system may also influence the opiate system, thereby altering cooing during threatening situations.

We are now refining our model of brain function by testing other compounds that bind to opiate and benzodiazepine receptors. We are also examining behavioral responses to substances, such as the neurotransmitter serotonin, that act on other receptors. (Serotonin receptors occur in many brain regions that participate in the expression of fear.) And we are studying the activities of substances that directly control stress hormone production, including corticotropin-releasing hormone, which is found throughout the brain.

In collaboration with Richard J. Davidson, here at Wisconsin, Shelton and I have identified at least one brain region in which the benzodiazepine system exerts its effects. Davidson had shown that the prefrontal cortex of the right hemisphere is unusually active in extremely inhibited children. We therefore wondered whether we would see the same asymmetry in frightened monkeys and whether drugs that reduced fear-related behavior in the animals would dampen right frontal activity.

This time we used mild restraint as a stress. As we anticipated, neuronal firing rose more in the right frontal cortex than in the left. When we delivered diazepam in doses we knew lowered hostility, the drug returned the restraint-induced electrical activity to normal. In other words, the benzodiazepine system influences defensive behavior at least in part by acting in the right prefrontal cortex.

Therapeutic Implications

THESE FINDINGS HAVE therapeutic implications. If human and monkey brains do operate similarly, our data would suggest that benzodiazepines might be most helpful in those adults and children who exhibit elevated electrical activity in the right prefrontal cortex. Because of potential side effects, many clinicians are cautious about delivering antianxiety medications to children over a long time. But administration of such drugs during critical periods of brain development might prove sufficient to alter the course of later development. And behavioral training could possibly teach extremely inhibited youngsters to regulate their benzodiazepine-sensitive systems without having to be medicated. Alternatively, by screening compounds that are helpful in monkeys, investigators might discover new

drugs that are quite safe for children. As the workings of other fear-modulating neurochemical systems in the brain are elucidated, similar strategies could be applied to manage those circuits.

Our discovery of cues that elicit three distinct sets of fear-related behaviors in rhesus monkeys has thus enabled us to gain insight into the development and regulation of defensive strategies in these animals. We propose that the opiate and benzodiazepine pathways in the prefrontal

demonstrated that extreme right frontal animals displayed more intense defensive behaviors (freezing and hostility) and had higher levels of cortisol when compared with their extreme left frontal counterparts. Furthermore, the extreme right frontal animals had higher cerebrospinal fluid levels of corticotropin-releasing hormone (CRH), and each individual animal's level of CRH appeared to be relatively stable. CRH not only regulates the release of cortisol but also mediates oth-

ety. We expected that monkeys without a functioning amygdala would display marked reductions in defensive behaviors as well as reductions in cortisol and CRH concentrations in the cerebrospinal fluid. We also expected that, as we had observed earlier with diazepam treatment, these monkeys would display a shift in their pattern of frontal brain activity characterized by an increase in left frontal electrical activity and a decrease in the right.

Consistent with earlier studies, we dis-

We have therefore laid the groundwork for deciphering the relative contributions of various brain systems underlying inordinate fear in humans.

cortex, the amygdala and the hypothalamus play a major part in determining which strategies are chosen. And we are currently attempting to learn more about the ways in which these and other neural circuits cooperate with one another.

Fearful Temperament

TO FOLLOW UP on the finding that humans with a preponderance of right frontal brain electrical activity are more likely to be anxious, we, along with Davidson, examined individual differences in this measure of brain activity in young monkeys. Similar to the observations in humans, we found that each animal's pattern of frontal brain activity was stable over time, such that animals with extreme asymmetric right frontal activity remained this way as they matured. Recall that we previously documented that a monkey's propensity to freeze is also a relatively stable trait.

Using this brain electrical activity measure, we next screened a large number of monkeys and selected two subsets of animals, those with extreme left frontal activity and those with extreme right frontal activity. Without knowing anything about their behavior, we hypothesized that those animals with extreme right frontal activity would be more fearful and also would have higher levels of cortisol. Based on this single measure of brain activity, this experiment

er fear-related behavioral and physiological responses.

Taken together, these findings led us to describe a fearful/anxious temperament or emotional style that is a relatively stable trait of some individuals. This temperament includes excessive fearfulness and critical physiological components: extreme right frontal brain activity, elevated basal cortisol and increased brain CRH. Evidence from other studies suggests that these characteristics will also hold for humans.

Our next step was to identify the brain regions that underlie these behavioral and physiological features. The first brain region we selected was the amygdala because of its well-known involvement in mediating fear responses and emotions. Researchers such as Joseph E. LeDoux of New York University and Michael Davis of Emory University have extensively explored the functions of this brain region in rodents [see "Emotion, Memory and the Brain," by Joseph E. LeDoux, on page 62]. Yet relatively few studies have used modern neurobiological techniques to examine the role of the amygdala in mediating emotion in primates.

Using techniques developed by David G. Amaral of the University of California at Davis and his colleagues, we were able to inactivate cells selectively in the monkey amygdala, allowing us to explore the role of this structure in mediating fear and anx-

covered that the monkeys' acute fear responses were blunted. For example, those without a functioning amygdala displayed a blunted withdrawal response when exposed to a snake and a decrease in submissive gestures when placed in the presence of an unfamiliar, threatening larger monkey. Their stress-induced hormonal response was also muted. But we were surprised to observe that these animals did not show deficits in their ability to freeze or display hostile gestures in the human intruder paradigm, nor were the physiological parameters that we believe make up the fearful/anxious temperament affected. Even among monkeys without a functioning amygdala, the magnitude of an individual's defensive response, the pattern of brain electrical activity, and the level of basal cortisol and CRH in the cerebrospinal fluid remained unaffected. In a recent study targeted at a specific region within the amygdala, we found that inactivation of this region blunted, but did not completely ablate, some of the responses associated with the anxious/fearful temperament.

These findings have led us to speculate that in primates, the amygdala serves to mediate acute fear-related responses and that other brain regions are most likely involved in responses that mark stable temperamental traits. Based on human studies and other animal work, we think that the prefrontal cortex may be instrumen-



tal in mediating the behavior and physiological aspects of the fearful/anxious temperament.

Studies are now under way to examine a specific region of the prefrontal cortex known as the orbitofrontal cortex. Interconnected with the amygdala, this brain region lies above the eyeballs and is much more prominent in primates than in rodents. Various studies have demonstrated the importance of this area in maintaining long-term, habitual behavioral responses, in modulating emotional responses and in enabling the prediction of the consequences of future behaviors.

Using these techniques in the monkeys in conjunction with human functional brain-imaging studies, we are confident that we and others will be able to characterize the brain circuits and the neurochemicals involved in the expression of adaptive fear and anxiety responses in humans, as well as to understand what is different in the brains of those individuals who suffer from exces-

INFANT (*left*) has strayed a short distance from its mother (*center*) and is producing a rudimentary threat face in an attempt to keep a photographer (the author) at bay. Rhesus monkeys become adept at matching their behavior to the severity and type of a threat when they are between nine and 12 weeks old, probably because certain neuronal pathways in three regions of the brain—the prefrontal cortex, amygdala and hypothalamus—reach functional maturity during this same period.

sive and maladaptive responses. We have therefore laid the groundwork for deciphering the relative contributions of various brain systems underlying inordinate fear in humans. We can envision a time

when treatments will be tailored to normalizing the specific signaling pathways that are disrupted in a particular child, thereby sparing that youngster enormous unhappiness later in life. SA

MORE TO EXPLORE

Love in Infant Monkeys. Harry F. Harlow in *Scientific American*, Vol. 200, No. 6, pages 68–74; June 1959.

Stress and Coping in Early Development. Jerome Kagan in *Stress, Coping, and Development in Children*. Edited by N. Garnezy and M. Rutter. McGraw-Hill, 1983.

Defensive Behaviors in Infant Rhesus Monkeys: Environmental Cues and Neurochemical Regulation. Ned H. Kalin and Steven E. Shelton in *Science*, Vol. 243, pages 1718–1721; March 31, 1989.

Stress in the Wild. Robert M. Sapolsky in *Scientific American*, Vol. 262, No. 1, pages 116–123; January 1990.

Defensive Behaviors in Infant Rhesus Monkeys: Ontogeny and Context-Dependent Selective Expression. N. H. Kalin, S. E. Shelton and L. K. Takahashi in *Child Development*, Vol. 62, No. 5, pages 1175–1183; October 1991.

The Primate Amygdala Mediates Acute Fear but Not the Behavioral and Physiological Components of Anxious Temperament. N. H. Kalin, S. E. Shelton, R. J. Davidson and A. E. Kelley in *Journal of Neuroscience*, Vol. 21, No. 6, pages 2067–2074; March 15, 2001.