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Studying Non-Human Primates: A Gateway to Understanding Anxiety Disorders

By Ned H. Kalin, MD

ABSTRACT *~ Non-human primates, such as the rhesus monkey, provide excellent models of human fear and anxiety because of similarities in behavioral responses and brain function. Studies of rhesus monkeys demonstrate that animals with an anxious temperament exhibit inappropriately exaggerated responses to fearful situations, extreme asymmetrical electrical activity in the right prefrontal cortex, and dysregulation of the corticotropin-releasing factor system. Similar findings have been observed in anxious or behaviorally inhibited children who are at greater risk of developing anxiety disorders later in life. Characterization of distinct behavioral and neurobiological features in anxious rhesus monkeys may one day form the basis of tools to identify children who are at risk to develop anxiety disorders and other stress-related problems later in life.* Psychopharmacology Bulletin. *2004;38(Suppl 1):8-13.*

INTRODUCTION

Preclinical studies are an important component in building a knowledgeable base for psychiatric disorders. In particular, animal models that closely mimic human behavior and physiology are pivotal to understanding the neurobiological basis of mental illness as well as for understanding the effects of drug therapy. The rhesus monkey (*Macaca mulatta*) is an excellent model of human behavior, especially fear and anxiety, because of similarities in behavioral brain functions between species.

Temperament is a trait that can be described as an individual's stable behavioral, emotional, and physiological disposition. Individual temperament is believed to be determined by both genetics and early-life experiences. An anxious, fearful temperament has been identified in the rhesus monkey that is similar to anxious temperament in humans.[1] The findings of studies in non-human primates and human children suggest that anxious temperament is associated with inappropriately exaggerated responses to fearful situations, extreme asymmetrical electrical activi-

Dr. Kalin is Hedberg Professor and chair of the Department of Psychiatry at the University of Wisconsin Medical School in Madison, WI.

To whom correspondence should be addressed: Ned H. Kalin, MD, Hedberg Professor and Chair, Department of Psychiatry, University of Wisconsin Medical School Department of Psychiatry, 6001 Research Park Boulevard, Madison, Wisconsin 53719; Tel: (608) 263-6079; FAX: (608) 263-9340; email: nkalin@facstaff.wisc.edu.

ty in the right prefrontal cortex, and dysregulation of the corticotropin-releasing factor (CRF) system.[2] Evidence suggests that children with extremely anxious temperament are at greater risk to develop anxiety disorders and further characterization of these behavioral and neurobiological features in early life may one day be a tool to identify children at risk for development of anxiety disorders and other stress-related problems later in life.

BEHAVIORAL RESPONSE TO FEAR

The developmental timeline for infant monkeys to mount and appropriately regulate defensive behaviors in response to anxiety-provoking environmental cues is well-documented. In a series of experiments designed to evoke responses to anxiety-provoking stimuli (APS) (eg, maternal separation or intruder threat), infant rhesus monkeys were shown to engage in adaptive defensive responses by about 2 months of APS. For example, when a young monkey is separated from its mother, it makes coo calls that identify the young monkey's location so that it can be retrieved by its mother. If a potential predator enters the environment, such as a human intruder, the monkey modulates its defensive response. If the intruder does not have eye contact with the monkey, the monkey engages in freezing behavior and reduces its coo calls. This response functions to help the monkey remain inconspicuous in the face of a threat while allowing it to prepare to take action. If the intruder has direct eye contact with the monkey, in order to protect itself the monkeys stops freezing and engages in threatening behaviors attempting to ward off a potential attack.[3] The ability to adapatively regulate defensive behaviors follows a developmental time course. Very young infants were not able to appropriately regulate these responses. For example, instead of remaining still and silent (ie, freezing) when confronted with a human intruder who did not appear to notice them, young infants often barked or shrieked, thus unnecessarily calling attention to themselves. However, by the time most infant monkeys were 3 months of age, they were able to adaptively regulate defensive responses that were appropriate to the type of threat.[4,5] The 3-month rhesus brain is developmentally similar to the human brain at 1 year of age. This is of interest because human infants develop stranger anxiety at this time.[5]

The findings from autoradiography studies in 3-month-old infant monkeys suggest that the ability to regulate defensive behavior corresponds with maturation of the CRF system in the hippocampus. This is of interest because the CRF system has been implicated in mediating adaptive and maladaptive stress responses and fearful behaviors. While the defensive reactions observed in infant monkeys are adaptive, exaggerated or extreme responses may suggest a predisposition to developing

anxiety-related problems. A possible human correlate to excessive and/or inappropriate freezing behavior may be extreme behavioral inhibition in young children.

NEUROBIOLOGIC CORRELATES OF ANXIETY

The human intruder paradigm described above was used in a series of pharmacologic challenge studies to assess involvement of neurochemical systems in mediating the defensive responses in infant monkeys. The findings demonstrated that distinct neurochemical pathways were involved in mediating the attachment distress induced by maternal separation versus the anxiety-related response elicited during the intruder threat. With maternal separation, low doses of morphine blunted infants' cries for their mother, and the opiate antagonist, naltrexone, increased this behavior, suggesting involvement of the opiate system in the response to disruption of attachment bonds and perhaps separation anxiety.[6] Conversely, GABA-ergic systems were found to be involved in regulating threat-induced defensive behaviors, such as freezing and barking, which were decreased following administration of benzodiazepines.[7]

Although defensive responses to anxiety-provoking stimuli are ubiquitous in non-human primates, there is considerable inter-individual variation.[8] As with humans, some monkeys are more anxious than others. One neurobiologic correlate of exaggerated defensive behaviors in rhesus monkeys that has been well-documented in humans is asymmetrical electrical activity in the frontal brain regions. Young animals with extreme right frontal brain electrical activity exhibit overly intense defensive behaviors, elevated levels of plasma cortisol, and a hyperactive CRF system, defined by increased CRF cerebraospinal fluid levels. These correlates are stable over time and some persist from infancy into adolescence.[2,9] Similar findings have been reported in 6-month old children, where elevated salivary cortisol levels and extreme right frontal electroencephalographic asymmetry were associated with behavioral inhibition.[10] Further study in young children is warranted to determine if these correlates are clinically useful in the early identification of children who are at risk of developing anxiety disorders later in life.

The amygdala is a key structure in the processing of fearful stimuli, and amygdalar activation is an important feature of the fear and anxiety response.[11] In a longitudinal series of studies comparing young children who were extremely behaviorally inhibited were followed for 20 years, and at age 22, amygdalar responses to novel stimuli were monitored using functional magnetic resonance imaging (fMRI). In contrast to controls, individuals who were socially inhibited as young children exhibited greater amygdala activity in response to images of novel faces.[13]

The role of the amygdala in mediating the primate's behavioral and physiologic response to threat was assessed in rhesus monkeys that underwent procedures that lesioned, or damaged, the amygdala. The lesions were made in either the whole amygdala or a region of the amygdala called the central nucleus. Unlike earlier work in monkeys, the lesions were very specific and only damaged neurons and not the fibers passing through the amygdala. While these techniques have been used extensively to study fear in rodents, very few studies have been performed in primate species. The results were somewhat surprising in that the effects of the lesions were not dramatic. Some reduction in fearfulness and a decrease in freezing behavior were observed. However, the monkeys maintained their ability to regulate their defensive responses. Also, the lesions blunted stress-induced hormonal responses. In addition, damage to the amygdala did not affect the patterns of right frontal asymmetry.[11] Thus, the primate amygdala appears to be involved in mediating some, but not all features of the anxious temperament. Other brain regions, such as the prefrontal cortex, may be equally important in regulating fearful and anxious responses.

EARLY-LIFE STRESS AND PSYCHOPATHOLOGY

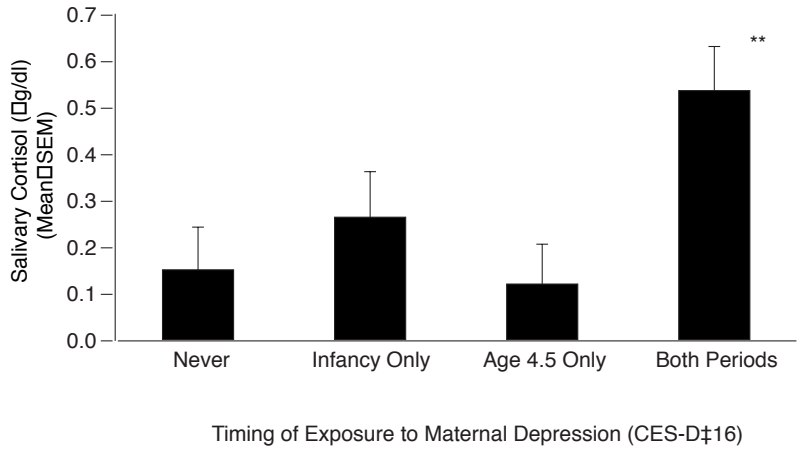
A large body of preclinical literature demonstrates that early-life adversity, such as abuse or parental loss, can permanently change behavioral and hormonal responses to fearful situations which may increase the risk for the development of psychopathology.[14] There also is growing evidence that exposure to stress early in life can alter temperament and increase the risk of mood and anxiety disorders in humans.[15] Our group has demonstrated the effect of maternal stress and depression on hypothalamic-pituitary-adrenal (HPA) axis activity and symptoms of mood and anxiety in pre-school age children. In this study, salivary cortisol levels in preschoolers were measured. Cortisol concentrations were somewhat higher in children who were exposed to their mother's depression at a young age compared with children whose mothers were never depressed or who were currently depressed. In contrast, cortisol levels were markedly higher in children with both current high levels of stress exposure and early-life exposure to maternal depression (Figure 1). In addition, children with the highest levels of salivary cortisol exhibited more pronounced psychiatric symptoms in first grade. This suggests that early-life stress can activate the HPA axis and predispose individuals to develop behavioral and emotional problems later in life .

CONCLUSIONS

Anxious temperament in rhesus monkeys is characterized by overly intense behavioral responses to threatening situations, extreme right frontal electrical brain activity, elevated baseline plasma cortisol, and increased activity of the CRF system. A similar profile of maladaptive responses in behaviorally inhibited children has been observed to persist into adulthood, suggesting that anxious temperament and propensity for anxiety disorders can be predicted early in life. Studies in monkeys and humans implicate the amygdala in the expression of anxious temperament. The underlying causes of anxiety disorders are not known. However, exposure to stress or adversity during childhood is believed to be an environmental factor that is strongly associated with later development of psychiatric illness, including anxiety disorders, in genetically predisposed individuals. Preclinical nonhuman models of human anxiety

FIGURE 1

TIMING OF EXPOSURE TO MATERNAL DEPRESSION AND CHILD CORTISOL LEVELS AT AGE 4.5 YEARS. ADAPTED WITH PERMISSION.[16]



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disorders are valuable scientific tools that will continue to inform our diagnostic, treatment, and preventive efforts. ✤

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