Neurobiological characteristics of rhesus macaque abusive mothers
and their relation to social and maternal behavior

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Abstract

Previous studies have reported hyperactivation of catecholaminergic systems and elevated concentrations of corticotropin-releasing-hormone (CRH) in the cerebrospinal fluid (CSF) of child maltreatment victims or combat veterans with post-traumatic stress disorder (PTSD). This study investigated the CSF concentrations of CRH and monoamine metabolites in rhesus macaque mothers that physically abused their infants and had themselves been abused as infants. Ten abusive mothers and 10 controls served as study subjects. All animals were sampled for CSF during pregnancy and the postpartum period. Focal observations of social and maternal behavior were also made. Abusive mothers had significantly higher CSF concentrations of CRH and 5-HIAA than controls. Across both subjects and controls, higher CRH, 5-HIAA and MHPG concentrations were associated with anti-social behavior patterns including a high frequency of maternal aggression, infant rejection, and a low frequency of contacts received from other individuals. These findings are consistent with those of previous primate and human studies and suggest that the neurobiological alterations associated with infant abuse may play an important role in the occurrence of maladaptive behavior in adulthood, including the perpetuation of infant abuse across generations.

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1. Introduction

Child maltreatment poses a substantial risk for psychological and behavioral disorders but its neurobiological consequences are only beginning to be investigated [31]. Preliminary research has focused on the physiological systems most likely to be affected by stress and trauma, such as the catecholamine systems and the hypothalamic–pituitary–adrenal (HPA) axis [30]. Earlier studies reported alterations in the circadian patterns of cortisol in physically or sexually abused children with depressive symptoms [9,14]. Recently, it has been reported that prepubertal maltreated children who develop posttraumatic stress disorder (PTSD) have significantly elevated concentrations of urinary cortisol, epinephrine, norepinephrine, and dopamine compared to children who were not maltreated [4]. Furthermore, studies of the neurobiological correlates of PTSD in combat veterans have been consistent with those of childhood trauma in reporting significant elevations of catecholamines and their metabolites in plasma and urine [2,5,15], higher concentrations of corticotropin-releasing-hormone (CRH) in the cerebrospinal fluid (CSF) [1], and lower concentrations of plasma cortisol [30].

Research with animal models can help us disentangle the many confounding variables that make research on child maltreatment difficult to interpret [26]. Naturally occurring infant abuse in nonhuman primates shares a number of characteristics with child maltreatment [19,37]. The investigation of infant abuse in two large captive populations of pigtail macaques (Macaca nemestrina, about 400 individuals) and rhesus macaques (Macaca mulatta, over 3000 individuals) over a period of 5–7 generations has shown that 5–10% of all infants born every year are physically abused by their mothers and that infant abuse is concentrated in some matrilines and among closely related individuals such as mothers and daughters, or sisters [20,27]. Thus, abuse appears to be transmitted across generations along the maternal line, and macaque females that are abused as infants are likely to become abusive mothers themselves.

Infant abuse in macaques generally occurs in the first 2–3 months of infant life and manifests itself as infant dragging, crushing, throwing, stepping or sitting on [18,33]. The consequences of abuse may range from infant distress to serious injury and death. Abusive mothers alternate short bouts of abuse with long periods of appropriate caregiving behavior [18,34]. Abusive mothers are typically consistent in their physical patterns of abuse over time and across infants [25]. Although there may be some variation in the rate with which some abusive mothers abuse their infants from year to year, mothers that have abused their offspring before are very likely to repeat the same patterns of behavior with all of their successive infants [20,25].

Furthermore, these individuals also abuse adopted infants with rates similar to those exhibited with their biological offspring, suggesting that infant characteristics play a minor role in the occurrence of abuse [24]. Instead, abusive behavior appears to be a stable maternal characteristic.

Macaque abusive mothers do not show any gross behavioral abnormalities in their social interactions with their conspecifics [18,33]. However, they can be differentiated from other mothers for their controlling parenting styles, as they score higher than controls on measures of maternal protectiveness and rejection [18,33]. In pigtail macaques there is evidence that abuse is likely to be immediately preceded by stressful social events such as intra-group aggression or infant kidnapping [17,21]. Since abusive mothers are not more likely to find themselves in such stressful situations than other mothers, this suggests that abusive mothers are individuals that are particularly vulnerable to stress or with problems in emotion regulation [36]. Consistent with this hypothesis, studies of infant abuse in Japanese macaques (Macaca fuscata) have shown that abusive mothers have high anxiety and that their abusive behavior is reduced or eliminated by treatment with anxiolytic drugs [35]. The mechanisms by which anxiolytic drugs reduce abusive behavior, however, are not known. The endogenous opioid system does not appear to be involved because treatment with naltrexone, an opioid receptor blocker, had no significant effects on maternal abusive behavior in rhesus macaques [8].

Aside from these pharmacological data, there is little information available on the neuroendocrine correlates of abusive parenting in nonhuman primates. Previous studies have found that rhesus macaque abusive mothers do not differ from controls in plasma estrogen and progesterone concentrations during late pregnancy and early lactation [22,23]. Since these hormones may be involved in the regulation of maternal responsiveness, it does not appear that abusive behavior is the result of a deficit in maternal motivation or alterations in some of its endocrine substrates. The above reviewed behavioral data, however, suggest that macaque abusive mothers might exhibit alterations in the neurobiological substrates of emotion regulation and that these alterations may be the result of early adverse experience (see also [32]). For example, in nonhuman primates, other negative rearing experiences such as early maternal deprivation or exposure to an unpredictable rearing environment have been shown to result in changes similar to those associated with human PTSD, including...
The results of this study confirm and extend the findings of preliminary human research on the neurobiology of child maltreatment and offer new insights into the relation between early abuse and adult behavior.

2. Methods

2.1. Subjects

Subjects of this study were 20 adult female rhesus macaques living in four social groups at the Field Station of the Yerkes National Primate Research Center in Lawrenceville, Georgia. The adult females ranged in age from 6 to 15 years and were all multiparous having delivered at least one offspring prior to this study. The social groups were housed in 38 × 38 m outdoor compounds with attached indoor areas and consisted of 2–5 adult males and 30–35 adult females with their immature offspring. All monkeys were fed standard monkey chow twice daily (at 0900 and 1500 h) and fruit once daily (1500 h). Water was available ad libitum. The dominance ranks of all adult females were assessed with data on aggression, submission, and displacements collected prior to onset of the study. Each adult female was classified as high, middle, or low ranking depending on whether her dominance rank fell into the upper, middle, or bottom third of her group’s hierarchy.

2.2. Procedure

Ten subjects and 10 controls were selected according to whether or not they had a history of abusive parenting with their previous offspring. The 10 abusive mothers selected for this study had been observed in previous years and their abusive behavior had been documented (Maestripieri, 1998), whereas none of the 10 controls had ever been observed to abuse their offspring. For 6 of the 10 abusive mothers, we had direct evidence that these individuals had themselves been abused as infants because early abuse had been observed by research or caretaker personnel and recorded in the animals’ files. For four abusive mothers, abuse during infancy was not directly witnessed but inferred from the fact that the subjects were born to abusive mothers that repeatedly abused their previous and subsequent offspring. None of the 10 controls of this study (or their siblings) had been abused as infants.

Two CSF samples were collected from each individual, one 4–8 weeks prior to parturition and the other 3–4 weeks after parturition. Date of parturition was estimated with ultrasonography tests and later confirmed retrospectively. All samples were obtained between 1000 and 1200 h. Prior to sample collection, all animals had been trained to run into an indoor capture area, where they were transferred via a transfer box into a standard squeeze cage. CSF samples were obtained as soon as possible following anesthesia induction (with ketamine hydrochloride IM, 15 mg/kg) and time to obtain the sample was recorded for each subject. One 2–3 ml CSF sample was collected from the cisterna magna using a 5 ml syringe with a 1 in., 22-gauge, bevel-tipped needle [12,13]. CSF samples were analyzed using liquid chromatography with electrochemical detection [29] and assayed for concentrations of the serotonin metabolite, 5-hydroxyindoleacetic acid (5-HIAA), the noradrenaline metabolite, 3-methoxy-4-hydroxyphenylglycol (MHPG), and the dopamine metabolite, homovanillic acid (HVA). CSF samples were also assayed for concentrations of CRH with radioimmunoassay. CRH data were only available for samples collected during pregnancy. All inter- and intra-assay variabilities were less than 10%.

Focal behavioral observations (1 h per week) of subjects and controls began 8 weeks prior to the estimated date of parturition and continued through the fourth postpartum week. Observation sessions were randomly distributed between 0800 and 1900 h. During late pregnancy, behavioral data collection focused on social interactions between the focal females and all of the other group members including contact, grooming, and aggression. Following birth, behavioral data collection also included mother–infant interactions such as contact, grooming, restraining, and rejection [18]. Incidents of infant abuse were recorded as: (1) dragging: the mother drags her infant by its tail or leg while walking or running; (2) crushing: the mother pushes her infant on the ground with both hands; (3) throwing: the mother throws her infant a short distance with one hand while standing or walking; (4) hitting: the mother violently slaps her infant with one hand or arm; (5) biting: common definition; (6) stepping or sitting on: the mother steps on her infant with one foot or both feet, or sits on her infant; (7) rough grooming: the mother pulls her infant’s hair or otherwise roughly grooms it causing distress calls. All behavioral measures were recorded in terms of frequency of occurrence.

Correlations between variables were assessed with the Spearman’s correlation coefficient. Comparisons between subjects and their matched controls were made with the Wilcoxon matched-pair signed-test. Other nonparametric tests used in data analyses included the Kruskal–Wallis
analysis of variance, the Mann–Whitney test, and the chi square test. All tests were two-tailed. Probabilities < 0.05 were considered statistically significant.

3. Results

CSF concentrations of 5-HIAA, MHPG, and HVA during pregnancy and lactation were highly correlated (N = 20, 5-HIAA: r = 0.60, p < 0.01; MHPG: r = 0.59, p = 0.01; HVA: r = 0.60, p < 0.01) and not significantly different between these two periods (N = 20, 5-HIAA: z = 0.89, p = 0.37; MHPG: z = 1.24, p = 0.22; HVA: z = 1.17, p = 0.24). Thus, individual differences in monoamine metabolites were relatively stable before and after parturition. To avoid redundancy, only pregnancy data were used in subsequent data analyses. Pregnancy data were also preferred because CRH data were not different in relation to the age of the mother (r = 0.20, 5-HIAA: z = 0.42; HVA: r = 0.24, p = 0.08; MHPG: r = 0.22, p = 0.34) and the dopamine metabolites. In contrast, CSF concentrations of CRH were not significantly correlated with any of the three monoamine metabolites (N = 20, 5-HIAA, r = 0.20, p = 0.38; MHPG, r = 0.28, p = 0.21; HVA, r = 0.36, p = 0.11).

CSF CRH and monoamine metabolite concentrations were not different in relation to the age of the mother (N = 20, CRH: r = 0.40, p = 0.08; 5-HIAA: r = 0.40, p = 0.08; MHPG: r = 0.22, p = 0.34; HVA: r = 0.24, p = 0.29), number of previous offspring (N = 20, CRH: r = 0.13, p = 0.57; 5-HIAA: r = 0.22, p = 0.34; MHPG: r = 0.24, p = 0.29; HVA: r = 0.27, p = 0.24), rank of the mother (CRH: H = 2.99, df = 2, p = 0.22; 5-HIAA: H = 1.44, p = 0.48; MHPG: H = 0.51, p = 0.77; HVA: r = 0.16, p = 0.92), or sex of the infant (N = 20, CRH: z = 0.42, p = 0.67; 5-HIAA: z = 1.48, p = 0.14; MHPG: z = 0.04, p = 0.97; HVA: r = 0.65, p = 0.52). Abusive and control mothers did not differ significantly in any of these variables (N = 20, age, z = 0.38, p = 0.70; number of offspring, z = 0.08, p = 0.94; rank of the mother, chi square = 1.07, df = 2, p = 0.70; sex of the infant, chi square = 0.20, df = 1, p = 1).

All abusive mothers abused their newborn infants, as expected on the basis of their previous parenting and rearing history, and five infants were removed from their mothers shortly after birth. No control mothers exhibited any abusive behavior. CRH and 5-HIAA concentrations were significantly higher in abusive mothers than in their matched controls (CRH: z = −2.40, p = 0.01; 5-HIAA: z = −2.09, p < 0.05; Fig. 1). The differences in MHPG and HVA concentrations between abusive mothers and controls were in the same direction but not significantly different (MHPG, z = −1.78, p = 0.07; HVA, z = −1.48, p = 0.14).

CRH concentrations were positively correlated with the frequency with which abusive and nonabusive mothers attacked other individuals during pregnancy (N = 20, r = 0.44, p = 0.05; Fig. 2a). MHPG concentrations were negatively correlated with the frequency with which other group members made contact with abusive and nonabusive mothers during pregnancy (N = 20, r = −0.71, p = 0.001; Fig. 2b). Aggression and contact received were not significantly correlated (N = 20, r = −0.20, p = 0.38). 5-HIAA concentrations were positively correlated with maternal rejections during the postpartum period (N = 15, 5-HIAA, r = 0.53, p < 0.05, Fig. 2c). There was no significant correlation between neurochemical variables and rates of infant abuse among the five abusive mothers that kept their infants (N = 5, CRH, r = −0.10, p = 0.84; 5-HIAA, r = 0.30, p = 0.54; MHPG, r = 0.40, p = 0.42; HVA, r = −0.10, p = 0.84) and no significant differences between the five abusive mothers that kept their infants and those that lost them (CRH, z = −0.73, p = 0.46; 5-HIAA, z = −1.36, p = 0.17; MHPG, z = −1.40, p = 0.15; HVA, z = −0.94, p = 0.34).
4. Discussion

The CSF concentrations of serotonin, noradrenalin, and dopamine metabolites measured in 20 adult rhesus macaque females 4–8 weeks before parturition and 3–4 weeks after parturition were relatively similar and highly correlated between the two periods, suggesting that these variables do not vary significantly in relation to female hormonal condition or parturition. The CSF concentrations of monoamine metabolites were also significantly interrelated and independent of the individuals' age, number of previous offspring, dominance rank, or sex of their offspring.

The 10 females that abused their infants after parturition and had a history of abusive parenting with their previous offspring had higher concentrations of CRH and 5-HIAA in the CSF than the nonabusive mothers. The differences in MHPG and HVA concentrations between abusive mothers and controls were in the same direction but did not reach statistical significance. Furthermore, among both subjects and controls, high concentrations of CRH, 5-HIAA, and MHPG were associated with antisocial behavior patterns including low amount of contact and high frequencies of aggression and infant rejection. Previous studies have shown that social isolation from other group members and high levels of infant rejection are characteristic of monkey mothers that abuse their offspring [18,34]. Variation among abusive mothers in contact or support received from other individuals, however, may not necessarily be associated with variation in the frequency of infant abuse. In this study, there was no significant correlation between neurochemical variables and the frequency of infant abuse among the abusive mothers, a result that may in part be due to the small sample size for this analysis.

Since we had some evidence that abusive mothers had themselves been abused as infants, one possible interpretation of the results of this study is that the trauma of infant abuse is associated with chronic activation of the serotonergic system as well as CRH neuronal hypersecretion. Unfortunately, the lack of accurate quantitative information on the experiences of abuse occurred in our subjects' infancy prevented us from assessing a possible relation between the severity of the physical trauma (i.e. the frequency, intensity, and duration of abuse) and the magnitude of the neurobiological alterations observed in adulthood.

Primate and human studies generally show impaired CNS serotonin functioning or low CSF 5-HIAA concentrations in impulsive and aggressive individuals [11]. There are, however, different forms of violent behavior and it is clear that infant abuse cannot be simply considered a form of impulsive social aggression [33]. For example, abusive mothers tend to treat their infants as inanimate objects and do not show the signs of arousal (e.g. piloerection, vocalizations) that typically accompany aggressive behavior [33]. Therefore, the neurobiological correlates of abusive parenting may not necessarily be similar to those of aggressive behavior. On the other hand, the findings of this study are generally consistent with those of human research showing that individuals who experienced child maltreatment or other forms of trauma exhibit signs of chronic activation of stress-sensitive neurobiological systems [1,7,15,30].

Fig. 2. (a) Correlation between CSF concentrations of CRH and number of aggressive acts displayed by abusive and nonabusive mothers toward other individuals during pregnancy (n=20). (b) Correlation between CSF concentrations of MHPG and number of contacts from other individuals received by abusive and nonabusive mothers during pregnancy (n=20). (c) Correlation between CSF concentrations of 5-HIAA and number of maternal rejections displayed by abusive and nonabusive mothers during the postpartum period (n=15).
CSF CRH concentrations primarily reflect the activity of extrahypothalamic CRH neurons [7] and such neurons modulate the noradrenergic neurotransmission in the locus coeruleus, amygdala and hippocampus, thus playing an important role in stress reactivity and emotion regulation. Our findings suggest that the hyperactivity of CRH neurons following traumatic or chronic stress may be one of the neurobiological mechanisms mediating the behavioral and emotional characteristics of monkey mothers who abuse their offspring [17,35]. Consistent with this hypothesis, research with rodents has shown that early stress and parental care regulate CRH gene expression, thus providing a mechanism for the transmission of individual differences in CRH-mediated behavioral systems across generations [6,16]. Moreover, a recent study of depressed women provides evidence that hypersecretion of CRH may mediate the relationship between early childhood abuse and the development of mood and anxiety disorders [10]. Because CRH can activate ascending noradrenergic and serotonergic systems [28], early dysregulation of catecholamine systems following traumatic or chronic stress can also contribute to the problems in anxiety, modulation of affect, and impulse control that are likely to be observed among abusive mothers in monkeys or individuals with affective disorders or PTSD in humans. Thus, the neurobiological correlates of infant abuse reported in this study of nonhuman primates could be an important mechanism mediating the effects of early trauma on adult maladaptive behavior in both primates and humans.

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