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What is This?

Emotional and Adrenocortical Responses of Infants to the Strange Situation: The Differential Function of Emotional Expression

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The aim of the study was to investigate biobehavioural organisation in infants with different qualities of attachment. Quality of attachment (security and disorganisation), emotional expression, and adrenocortical stress reactivity were investigated in a sample of 106 infants observed during Ainsworth's Strange Situation at the age of 12 months. In addition, behavioural inhibition was assessed from maternal reports. As expected, securely attached infants did not show an adrenocortical response. Regarding the traditionally defined insecurely attached groups, adrenocortical activation during the strange situation was found for the ambivalent group, but not for the avoidant one. Previous findings of increased adrenocortical activity in disorganised infants could not be replicated. In line with previous findings, adrenocortical

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activation was most prominent in insecure infants with high behavioural inhibition indicating the function of a secure attachment relationship as a social buffer against less adaptive temperamental dispositions. Additional analyses indicated that adrenocortical reactivity and behavioural distress were not based on common activation processes. Biobehavioural associations within the different attachment groups suggest that biobehavioural processes in securely attached infants may be different from those in insecurely attached and disorganised groups. Whereas a coping model may be applied to describe the biobehavioural organisation of secure infants, an arousal model explanation may be more appropriate for the other groups.

For almost three decades the Strange Situation procedure designed by Ainsworth and Wittig (1969) has been the standard procedure for the assessment of quality of attachment in infancy. By exposing infants to increasing challenges to the attachment system, that is, the presence of a strange person and two short separations from the attachment figure, the strange situation allows the assessment of different attachment behaviour strategies in dealing with the challenge (e.g. Ainsworth, Blehar, Waters, & Wall, 1978).

The majority of the infants behave as expected by attachment theory (Bowlby, 1969). Securely attached infants ("B") can use the caregiver as a secure base for exploration as long as the caregiver is present. When the attachment figure leaves the room they show clear indications of emotional concern or distress by exhibiting attachment behaviours like crying or searching for the attachment figure. At reunion they are able to re-establish their emotional stability by establishing bodily contact with the attachment figure. Other infants show different patterns. Insecure avoidantly attached infants ("A") show a lack of overt attachment behaviour in two ways. They show little or no indication of distress or active attachment behaviour during separation and do not seek contact upon reunion. In contrast, they ignore the attachment figure or actively avoid bodily contact. Insecure-resistant or insecure-ambivalent infants ("C") tend to be wary of the stranger and are intensely distressed by separation. When the caregiver returns, they show ambivalent behaviour by seeking close contact and at the same time showing angry resistance. Most important, they are not able to re-establish emotional stability for a long period. Thus, they cannot use the attachment figure for emotional regulation. The behaviour of infants with attachment disorganisation ("D") is characterised by the absence of a coherent behavioural strategy indicated, for example, by behavioural contradictions, breaks, or confusion. Attachment security and disorganisation are conceptually different dimensions (Main & Solomon, 1990, Spangler & Grossmann, in prep.) and

seem to be of different origin (Ainsworth & Eichberg, 1991; Main & Hesse, 1990; Spangler, Fremmer-Bombik, & Grossmann, 1996).

Although from the beginning attachment research has produced a large body of evidence regarding the developmental precursors of attachment patterns (e.g. Ainsworth et al., 1978; Grossmann, Grossmann, Spangler, Suess, & Unzner, 1985; Spangler et al., 1996; De Wolff & van IJzendoorn, 1997) as well as their developmental consequences (e.g. Main, Kaplan, & Cassidy, 1985; Suess, Grossmann, & Sroufe, 1992), some years ago, psychobiologically oriented attachment researchers have begun to investigate the biobehavioural organisation of the attachment system in infancy. Although some studies considered the cardiac function of infants (e.g. Donovan & Leavitt, 1985; Spangler & Grossmann, 1993), the majority of biobehavioural studies included the function of the hypothalamic-pituitary-adrenocortical axis (HPA) during the strange situation (e.g. Gunnar, Mangelsdorf, Larson, & Hertsgaard, 1989; Spangler & Grossmann, 1993). As the activity of the HPA axis is mainly affected by aversive or stressful situations involving novelty, uncertainty, and/or negative emotions (Levine, 1983), it should be specifically sensitive to separation stress in infants.

Our aim was to investigate individual differences in adrenocortical function between infants with different patterns of attachment. Specifically, the main focus was on the role of secure attachment as a buffer from adrenocortical elevation during separation from the caregiver, and the biobehavioural organisation of insecure ambivalent infants. In addition, we investigated individual differences between different groups of attachment quality in the interplay of emotional expression and adrenocortical function.

Adrenocortical responses can be related to behaviour in different ways. First, from the perspective of an arousal or distress model assuming general activation (Selye, 1952), stressful situations would elicit stress responses both on the behavioural and the physiological level. Thus, adrenocortical activation should go along with negative emotional activation (e.g. Levine, Wiener, Coe, Bayart, & Hayashi, 1987; Spangler & Scheubeck, 1993). Second, from a coping model perspective responses may be observed mainly on the physiological level or mainly on the psychological level depending on the availability of coping strategies, the subject's feeling of control, or other moderating parameters. Thus, adrenocortical activity should be most prominent in situations in which behavioural coping responses are not applicable or available (e.g. Levine et al., 1987; Spangler & Scheubeck, 1993; von Holst, 1986), as restricted ability of behavioural adaptation requires physiological adaptation. In contrast, adrenocortical responses are unnecessary, if stress-reducing behavioural strategies are available for the subject.

Applying the arousal model to the infant strange situation behaviour one would expect adrenocortical responses in infants exhibiting behavioural stress in terms of crying or negative expression. This would apply for the C-babies and two of the secure attachment subgroups (B3/B4: cf. Ainsworth et al., 1978) both characterised by a high proneness to distress being expressed in a high amount of crying or negative emotional expression during separation (Belsky & Rovine, 1987), whereas adrenocortical responses should not be observed in the A-infants and the other two of the secure attachment subgroups (B1, B2) both characterised by low proneness to distress. Applying the coping model, an increase in cortisol during the strange situation would be expected in insecure infants as compared to secure ones. Although the traditional insecure patterns (A and C) represent coherent and organised patterns (Main & Solomon, 1990), they cannot be seen as adequate strategies to rapidly establish proximity with the attachment figure. Regarding the A pattern as a defence strategy (Ainsworth et al., 1978) which helps to reduce behavioural arousal, it cannot be considered effective if it does not reduce tonic physiological arousal (Ursin, Baade, & Levine, 1978). Although C-babies show heightened attachment behaviour, which may be regarded as adaptive, their strategy is not wholly satisfactory, because they cannot use the attachment figure for behavioural regulation (Cassidy & Berlin, 1994). As the behavioural pattern of the D-babies indicates definite episodes of behavioural disorganisation, the D-status denotes a failure to establish an organised or coherent behavioural attachment pattern (Main & Solomon, 1990). Thus, cortisol increases may be expected in the insecurely attached infants who do not have adequate or effective strategies (A, C) or the disorganised infants (D) who do not have any coherent strategy at all.

Hitherto, there have been five studies investigating adrenocortical function in the strange situation. The first study was conducted by Gunnar et al. (1989). Using the classification of attachment security (A, B, C) they did not find differences in adrenocortical response between the attachment groups. The findings of the second study (Spangler & Grossmann, 1993) were in line with theoretical expectations from the coping model. Adrenocortical activation was found in a group of insecure infants (A, C) as well as in the disorganised infants, but not in secure infants. High cortisol values after the strange situation in disorganised infants were also reported in a third study by Hertsgaard, Gunnar, Erickson, and Nachmias (1995). They did not find, however, heightened cortisol in avoidant infants. Finally, there were two studies (Gunnar, Broderson, Nachmias, Buss, & Rigatuso, 1996; Nachmias, Gunnar, Mangelsdorf, Parritz, & Buss, 1996) finding cortisol increases in insecure infants during the strange situation and also during a different challenging situation, but each time only in those infants who additionally were characterised by behavioural inhibi-

tion. This indicates an interplay between temperamental characteristics and quality of attachment relationships for adrenocortical function.

Taken together, these findings are partly in line with theoretical expectations. The most consistent finding is an adrenocortical activation has not been observed for the group of securely attached infants. In addition, both studies including analysis of attachment disorganisation (Hertsgaard et al., 1995; Spangler & Grossmann, 1993) reported adrenocortical activation for the disorganised infants. Regarding the traditionally insecure groups, however, findings were not equivocal, though not contradicting. There may be methodological as well as psychological explanations for this inconsistency. Methodological issues refer to the time schedule of cortisol assessment or the limited number of subjects per attachment group. Regarding assessment time, interpretation of some findings may be difficult due to missing baseline values (Hertsgaard et al., 1995) or due to early post-assessment of cortisol (immediately after the strange situation, Gunnar et al., 1989) eventually leaving not enough time for a response of the slowly reacting adrenocortical system. Regarding group size, as in normal samples the majority of infants show a secure attachment pattern, the number of subjects of the traditional insecure groups (A, C) often was too low for statistical comparisons. Thus two groups were combined leading to significant effects in some studies (Gunnar et al. 1996; Nachmias et al., 1996; Spangler & Grossmann, 1993), but not in the Hertsgaard et al. (1995) study. Combining the groups may mask existing effects in one of the groups, and it leaves open the question whether a given effect may be due to only one or both of the groups.

A psychological explanation for the inconsistency refers to the additional consideration of processes related to attachment disorganisation or behavioural inhibition. First, three of the studies (Gunnar et al., 1989, 1996; Nachmias et al., 1996) did not include the analysis of attachment disorganisation. Inclusion of disorganised infants with an underlying secure pattern in the group of "forced" securely attached infants may obscure potential differences between securely and insecurely attached infants. This argument may be specifically important given the finding that in both studies including disorganisation D-infants had higher cortisol responses. Second, the findings of Nachmias et al. (1996) and Gunnar et al. (1996) about the influence of attachment security on adrenocortical function only in infants with high behavioural inhibition suggest a conceptualisation of attachment security as a social buffer in infants with restricted behavioural dispositions. That is, the influence of attachment on adrenocortical function may depend on the availability of additional behavioural competencies contributing to behavioural regulations in stressful situations, and, thus, may not be reliably assessed in studies only including the attachment security dimension. For example, adrenocortical responses in

avoidant infants may only hardly be identified (e.g. Hertsgaard et al., 1995), because a majority of these infants may be characterised by low behavioural inhibition.

The first main objective of our study was to replicate findings about adrenocortical responses of insecurely attached or disorganised infants by avoiding methodological issues of existing studies. Data assessment should include both baseline levels and response levels of cortisol, and the sample should be large enough to allow for statistical comparisons between all attachment groups. Moreover, behavioural analyses should consider measures of both attachment security and disorganisation as well as of behavioural inhibition. This would allow us to investigate the specific role of a secure attachment as a social buffer in stressful situations, particularly in infants with restricted abilities for behavioural regulation.

Second, our aim was to study the biobehavioural organisation of insecure-ambivalent infants, for which up to now systematic data have not been available. Their behavioural pattern is characterised by a high level or intensity of negative expression during separation, indicating stress and emotional activation, and by its ineffectivity regarding behavioural regulation, indicating a lack of behavioural coping ability. This complicates the interpretation of biobehavioural organisation in these infants, because both the arousal and the coping model offer an explanation for an adrenocortical activation during the strange situation in them. From an arousal model perspective, we would expect high cortisol responses because they exhibit marked behavioural distress. From a coping model perspective, we would expect high cortisol values because of the inappropriateness of their attachment strategy. Moreover, within attachment theory, negative expression is seen as an attachment behaviour, and as such is regarded as an appropriate coping response after separation. Whereas negative expression may indicate stress in both secure and insecure infants, its adaptive function in secure infants may help them to regulate their behaviour before it comes to an adrenocortical activation. In consequence, crying *per se* would not necessarily predict adrenocortical activation. In contrast, aiming to explain high cortisol values in ambivalent infants within a coping model, we have to present indications for the dysfunctionality of negative expression in these infants.

As a consequence, our third main objective was to study the differential function of negative expression in infants with different patterns of attachment. Exhibiting the prototypical behavioural pattern expected by attachment theory, secure infants show crying or negative expression during separation from the mother, which leads to contact with her after reunion, which in turn enables the children to re-establish their emotional state and to return to exploration. Thus, emotional expression fulfils a relational function as an emotional signal (Campos, Campos, & Barrett,

1989), and its function as an attachment behaviour is obvious. Therefore, high amounts of crying should not lead to adrenocortical activation in these infants. In contrast, crying may have lost its social or relational function and may be restricted to emotional expression in insecure infants. Avoidant infants show little emotional expression during separation. They are not able to use this type of attachment strategy. Ambivalent infants show marked distress during separation, sometimes even very extreme crying, but their emotional expression seems not to be functional with respect to emotional regulation. Despite exhibiting attachment behaviour, they are not able to reorganise their emotional state with the help of the caregiver. In conclusion, regarding relations between negative emotional expression and adrenocortical activity the coping model may be appropriate for secure infants but not for insecure ones, for which the arousal model may be a more useful description of biobehavioural organisation. As pointed out by Pawlik (1995), inter-individual differences not only occur in the number or intensity of specific psychological characteristics, but also in different ways of associations between different psychological characteristics. Thus, our aim was not only to compare emotional behaviour and adrenocortical processes between the different attachment groups, but also to assess biobehavioural associations within each of the attachment groups.

METHOD

Subjects

The sample consisted of 106 healthy German white infants and their mothers (53 girls/53 boys). The children's families represent a fairly wide range of socioeconomic status, including 42% upper-middle class, 24% middle class, and 27% lower class, as assessed by the father's education and occupation and the total family income. In all but one case, the mother was the infant's primary caretaker. Sixteen mothers (15%) were working outside the family in part-time jobs at the end of the first year.¹ During mothers' working time these infants were cared for by the father or the grandmother. For one infant, the father was the primary caretaker throughout the first year, while the mother was doing a full-time job. In this case the father took part in the study (and was treated as a "mother" in this paper). None of the infants was in day care.

¹ The low number of mothers working outside the family is typical at this age period. In Germany, mothers get financial help from the government during the first two years. Thus, most of the mothers would remain at home during this period.

Procedure

At the age of 12 months ($M = 12.4$; range: 11.7–13.7) the infants were observed during the strange situation. The strange situation is a 20-minute situation involving a sequence of episodes which progressively activates the attachment system: Entrance into an unfamiliar environment, the arrival of a stranger, two brief separations from the mother, and two subsequent reunions with her (Ainsworth et al., 1978). Immediately after the strange situation there was a 30 minute free-play situation, during which the mother filled out a German version of the Toddler Temperament Scale (Fullard, McDevitt, & Carey, 1984), a 97-item caretaker questionnaire designed to assess the nine temperamental dimensions as described by Thomas, Chess, and Birch (1968). With regard to the circadian rhythm in cortisol the data assessment was scheduled for a limited time range in the morning (cf. Gunnar et al., 1989; Nachmias et al., 1996; Spangler & Grossmann, 1993), midway between two feedings, that is, about 90min after the preceding feeding. Most of the assessments (86%, $n = 91$) started between 8.00a.m. and 10.00a.m. in 12 cases (11%) the beginning was between 10.00a.m. and 10.45a.m. The morning assessment was not possible in three cases and took place in the afternoon. The mother-infant pairs were collected from home by a taxi about 15 to 20 minutes before the scheduled time point (i.e. about 30 minutes before the beginning of the strange situation).

Behavioural Analyses

The quality of infant-mother attachment was analysed from the videotapes by trained observers. For the traditional ABC-classification according to procedures described by Ainsworth et al. (1978) the training was accomplished using strange situation videotapes from the Bielefeld longitudinal sample and were cross-validated by Mary Main (Grossmann, Grossmann, Huber, & Wartner, 1981). Reliability for quality to attachment was 90% ($\kappa = .87$). D-classification was accomplished according to Main and Solomon (1990). For the D-classification two different observers were trained with tapes analysed by an expert judge (Erik Hesse). The observers rated the infants' behaviour on the 9-point scale for disorganisation. Reliability for the two observers for the D-status (5 or above) was 80% and 82% ($\kappa = .63$). In case of disagreement in attachment classification, conference scores were used.

In addition, to assess behavioural measures of the infants' negative emotional state during the strange situation instances of the expression of negative emotions were assessed for every 1-second interval during the strange situation. Every occurrence of crying, fussing, or sad face was coded as a negative emotional expression. This was done by an additional

observer blind to the attachment classifications. Inter-observer reliability was 94% ($\kappa = .83$).

Assessment of Adrenocortical Activity

The cortisol response was assessed from saliva. To collect saliva, the infant's mouth was swabbed with a small sterile cotton dental roll. It took about 1–5min to obtain the saliva sample. Saliva samples were collected immediately before the strange situation, as well as 15 and 30min after its end. In addition, the mothers had been asked to collect saliva samples in the morning before the first feeding. We used two post-assessments of cortisol, because in the Spangler and Grossmann (1993) study attachment differences were most obvious 30min after the strange situation. All saliva samples were frozen until being assayed to -20°C after the end of the assessment procedure. To get the infants' saliva out of the roll, the samples were centrifuged (5000rpm). The assessment of cortisol levels was done by radioimmunoassay (RIA). All samples from one individual were analysed in one assay to minimise the variability of the results. To guarantee validity of analysis, duplicate assays were performed whenever possible. Some samples with a volume of less than 0.1ml were too small for duplicate assay. In these cases single assays were accomplished. Each sample was analysed by two different laboratories: the Department of Animal Physiology, University of Bayreuth/Germany (Professor Dr D. von Holst); and the Research Centre for Psychobiology and Psychosomatic, University of Trier/Germany (Dr Clemens Kirschbaum). The use of two different labs was necessary because a change of the antiserum after analysis of the first half of the sample in the former lab resulted in an overall decrease of values. Therefore, the samples were once again analysed in a different laboratory. The comparison of the values of the different labs enabled us to identify and reanalyse invalid cortisol values. Inter-lab correlation for the twice-analysed first half of the sample was $r = .85$. The cortisol values used for the present analyses were calculated as the mean value over the two labs, if valid determinations were present. Alternatively, and in cases in which outliers or extreme values were identified in one of the lab samples, only the value of the other lab was used. The occurrence of small samples and extreme values was similar for the different attachment groups.

The assays were highly specific for cortisol. The sensitivity of the assay used in Trier was 0.43nmol/l. The intra-assay variation was 6.7% for 2.2nmol/l, 4.7% for 5.5nmol/l and 4.0% for 13.2nmol/l. The respective values for inter-assay variation were 9.0%, 8.6%, and 7.1% (Dressendoerfer, Kirschbaum, Rohde, Stahl, & Strasburger, 1992). The sensitivity of the assay used in Bayreuth was 2.6ng per tube. The intra-assay

variation was 5.9% for 0.1ng, 4.1% for 0.2ng, and 3.8% for 0.5ng. The inter-assay variation was 9.2% for 0.1ng, 6.0% for 0.2ng, and 5.8% for 0.5ng (for further details see Fenske, 1987)

Measures and Statistical Analyses

The following measures were derived from the behavioural analyses: (1) security of attachment (traditional ABC-classification); (2) attachment disorganisation (D, nonD); and (3) duration of negative emotional expression (%) separately for each episode of the strange situation. The scale approach versus withdrawal of the Toddler Temperament Scale was used as a measure for behavioural inhibition. The following cortisol measures were used: morning cortisol, cortisol before, and 15min and 30min after the strange situation. To control for extreme scores the cortisol values were natural-log-transformed for statistical analyses. To assess statistical significance of changes in behavioural and physiological measures analyses of variance with repeated measures were conducted. To control for heterogeneity of variance, Greenhouse–Geisser corrections for degrees of freedom were used.

Preliminary Analyses Regarding Assessment Time of Cortisol

To control for effects of assessment time on cortisol due to circadian rhythm, preliminary analyses were conducted. Correlation between cortisol and assessment time was: $r = -.24$, $-.05$, and $-.13$ for the cortisol before and 15min and 30min after the strange situation, respectively. Only the correlation for the base level (before) reached statistical significance ($P < .05$). There were no differences between the various attachment groups with respect to assessment time. Mean assessment time before the strange situation was 09.16, 09.23, and 09.05a.m. for secure, insecure-avoidant, and insecure-resistant infants, respectively, and 09.18 and 09.07 for nonD and D infants. Excluding the three afternoon assessments, mean assessment time for the various assessment groups ranged within 5min from 09.07 to 09.12a.m. To control for the relations between assessment time and cortisol values, assessment time was used as a covariate for statistical analyses.

RESULTS

Quality of Attachment

Regarding the traditional (forced) ABC-classification, the distribution of securely and insecurely attached infants was comparable to previous findings (van IJzendoorn & Kroonenberg, 1988). Sixty-six out of 106

(62%) infants were classified as securely attached (B). There were 35 B1/B2 and 31 B3/B4. Twenty-one infants (20%) were classified as insecure-avoidant (A) and 16 infants (15%) were rated as insecure-ambivalent (C). Three infants were unclassifiable. In addition, 23 (22%) infants (15 B, 1 A, 4 C, and 3 unclassifiable) were classified as disorganised (D). The proportion of the D-pattern was also comparable to previous findings (Ainsworth & Eichberg, 1991; Main & Hesse, 1990; van IJzendoorn, 1995). Thus, including the D-status, the analysis of the attachment patterns yielded the following distribution: 51 (48%) B; 20 (19%) A; 12 (11%) C; and 23 (22%) D. Both dimensions of attachment classification were not related to infants' sex and social class.

As some cortisol values were missing for technical reasons (e.g. too little saliva, failure in RIA), the sample sizes for the following analyses differed. Thus, for the cortisol analyses the final sample sizes for the attachment security groups A, B, C were 17, 54, and 12, respectively, and for attachment disorganisation D and nonD were 19 and 66, respectively. As argued earlier, security and disorganisation are regarded as different constructs and as such they should be included as two separate independent factors for the following ANOVAS. However, due to the low frequencies of D-infants with underlying A ($n = 1$) and C ($n = 4$) this was not possible. Therefore each time, separate analyses were conducted for the two dimensions.

Emotional Expression during the Strange Situation

Preliminary analyses were conducted to control for sex and social class effects for emotional expression. There were no sex effects. Correlations between the emotional expression scores for strange situation episodes 2 to 8 and social class revealed only one significant correlation. High emotional expression during episode 4 was related to low social class ($r = -.22$, $P < .05$). To compare the behavioural changes across the strange situation episodes of infants with different attachment quality two-way MANOVAS with one repeated measures factor (episodes 2–8) and one independent factor for attachment group were conducted for negative expression, separately for attachment security (A, B, C) and attachment disorganisation (D, nonD). As expected, there were marked behavioural differences between the different attachment security groups. There was a main effect for attachment [$F(2,99) = 44.78$, $P < .001$] and for episode [$F(3.5,345.5) = 76.65$, $P < .001$], and an interaction between episode and attachment [$F(7.0,345.5) = 11.87$, $P < .001$]. Simple main effect ANOVAS revealed that the mean negative expression was significantly above zero for the B-infants [$F(1,101) = 179.1$, $P < .001$] and C-infants [$F(1,101) = 194.7$,

$P < .001$], but not for the A-infants [$F(1,101) = 2.55$, n.s.]. A significant change in negative expression across episodes could be observed in all groups [$F_s(3.5,345.5) = 5.5, 93.9$, and 42.7 for the A-, B-, and C-infants, respectively, $P_s < .001$]. There were, however, different change patterns for the groups (see Fig. 1). Whereas in A-babies there was only a slight increase during episode 6 (when the child was alone), B-babies showed a slight increase during the first and a marked increase during the second separation. In C-babies there was a marked increase already during the first separation with a further increase during the second one. Duncan *post-hoc* comparisons were conducted to test for between-group differences in the different episodes. Although there were no differences between the groups during the second episode, C-infants had slightly higher values than the A- and B-babies in episode 3 ($P < .05$). During episodes 4-8 the A-babies had the lowest values, whereas the C-babies had the highest values with the B-babies lying in between. All between groups in these episodes difference were significant ($P < .05$) with the exception that B- and C-infants did not differ during episode 6.

Regarding attachment disorganisation the two-way MANOVA revealed only a significant effect for episode [$F(3.5,358.4) = 61.39$, $P < .001$]. There were no effects regarding disorganisation. Thus, expression of negative emotion seems not to be dependent on attachment disorganisation.

Changes in Adrenocortical Activity during the Strange Situation

Preliminary analyses revealed no effects of sex and social class for the cortisol values. To test the hypothesis about individual differences in the adrenocortical response between the traditional attachment groups, a two-way MANOVA with one repeated measure for time (before, after 15min, after 30min) and one independent factor for attachment security (A, B, C) and with assessment time as a covariate was conducted. There was a main effect for attachment security [$F(2,79) = 4.69$, $P < .05$]. There was no main effect for time and no interaction between attachment security and time. Duncan *post-hoc* tests revealed higher cortisol values for the insecure-ambivalently attached infants ($P < .05$). Figure 2 indicates that the cortisol values of the C-babies were highest 15min after the strange situation preceded by a slight cortisol increase during the strange situation. As the C-infants had heightened cortisol values already before the strange situation, this increase was tested by a separate one-way ANOVA for the 15min post-cortisol with attachment security (A, B, C) as the independent factor and pre-assessment cortisol as a covariate. Individual differences between the attachment groups approached significance [$F(2,79) = 2.46$, $P < .10$]. *Post-hoc* Duncan comparisons indicated a higher

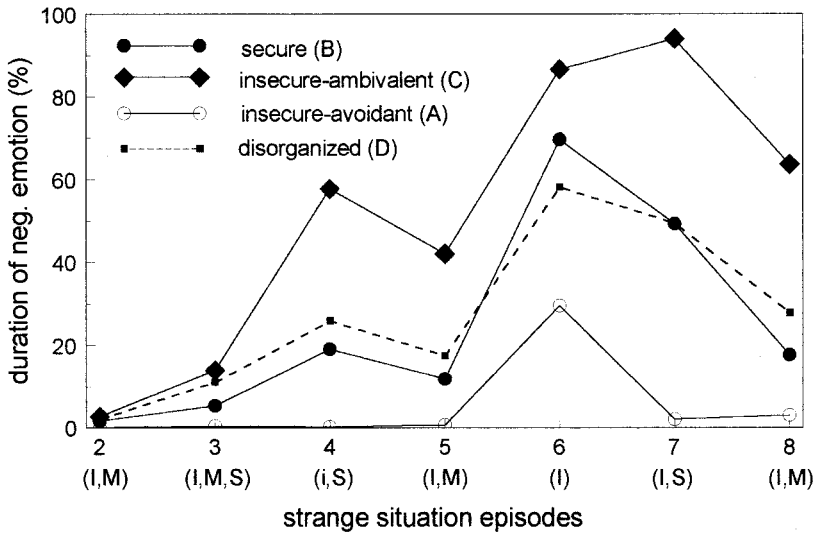


FIG. 1. Changes in negative emotional expression during the strange situation for different attachment groups (M, Mother; I, Infant; S, Stranger.)

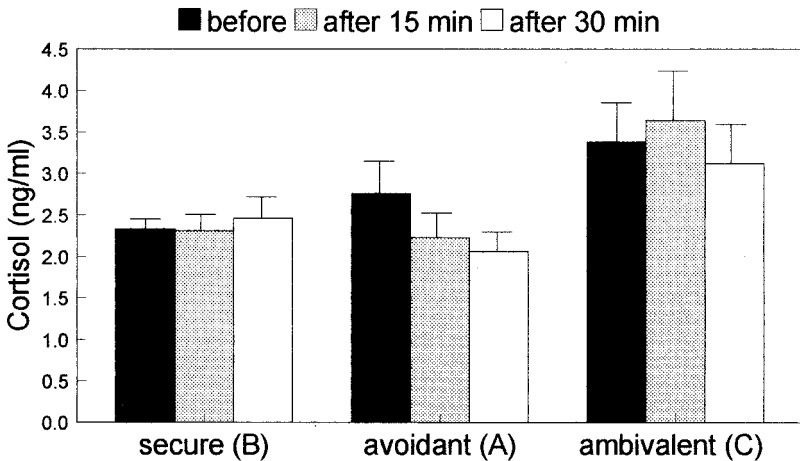


FIG. 2. Cortisol values before, and after the strange situation for the traditional attachment security groups (secure, insecure-avoidant, insecure-ambivalent). (Error bars indicate 1 SE.)

increase for the C-babies as compared to both the B- and the A-infants ($P < .10$). As the baseline values before the strange situation could be the response to the challenge brought about by the preparation (e.g. going to the university), the attachment groups were compared with respect to their morning cortisol values. The morning values were 6.51, 5.05, and 3.55 for the A-, B-, and C-infants, respectively, indicating low morning values for the C-infants. Analysis of variance, however, revealed no significant difference [$F(2,70) = 2.01$, n.s.]. Thus, insecure-ambivalent infants seem to respond to the preparation procedure for data assessment rather than having tonically high cortisol values.

Regarding attachment disorganisation, a two-way MANOVA with one repeated measure for time (before, after 15min, after 30min) and one independent factor for attachment disorganisation (D, nonD) revealed no significant effect.

Interaction between Attachment and Behavioural Inhibition

Behavioural inhibition as assessed by mother questionnaire was related to infant behaviour during the strange situation. There was a significant correlation between behavioural inhibition and the mean negative emotional expression during the strange situation ($r = .54$, $P < .001$). To assess the interplay between behavioural inhibition and attachment for the adrenocortical response during the strange situation a three-way MANOVA with one repeated measure for time (before, after 15min and 30min), two independent factors for attachment security (B vs. A/C) and behavioural inhibition (low, high), and assessment time as a covariate was conducted. There was a main effect for behavioural inhibition [$F(1,78) = 4.60$, $P < .05$] qualified by an interaction between behavioural inhibition and attachment security [$F(1,78) = 5.50$, $P < .05$]. As can be seen from Fig. 3, insecurely attached infants with high behavioural inhibition had higher overall cortisol values than each of the three other groups (Duncan, $P < .05$). The interaction effect remains significant in a separate two-way ANOVA for the 15min post-cortisol using pre-cortisol as a covariate to control for the baseline differences before the strange situation. There were no significant differences between the four groups regarding morning cortisol values [$F < 1.0$, n.s.]. The pattern of finding remains basically the same, when the disorganised infants were included within the insecure group.

As most of the insecure-ambivalent infants (15 out of 16) were part of the group of infants with high inhibition, it may be assumed that the interaction effect was due to the high cortisol values of the C-babies.

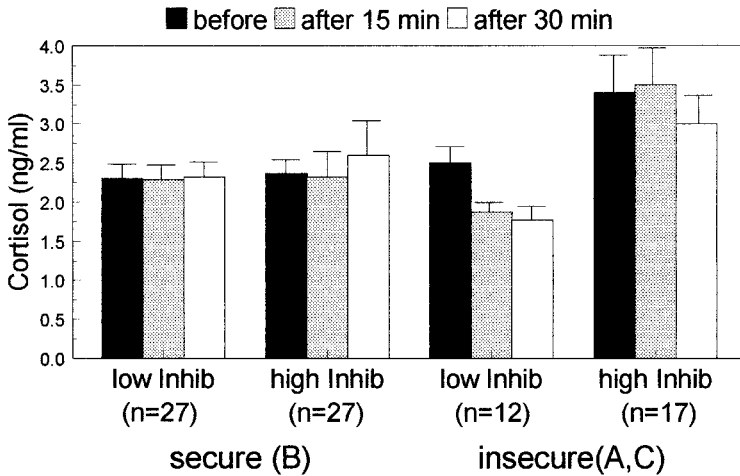


FIG. 3. Cortisol values before, and after the strange situation for securely (B) and insecurely (A, C) attached infants with low and high behavioural inhibition, respectively. (Error bars indicate 1 SE.)

However, this seems not to be the case, because among the highly inhibited infants the cortisol values of the avoidantly attached infants ($M = 2.99$) does not significantly differ from that of the ambivalently attached infants ($M = 3.45$).

An additional test for the interplay between attachment and temperamental factors was accomplished by using the Belsky split within the secure group. According to Belsky and Rovine (1987) the secure group is comprised of infants who are emotionally low reactive (B1/B2) and high reactive (B3/B4). Accordingly, among the insecure group avoidant infants are defined as low reactive and ambivalent infants as high reactive. A two-way MANOVA with one repeated measure for time (before, after 15min and 30min) and one independent factor for attachment security (A, B1/B2, B3/B4, C) and with assessment time as a covariate revealed a main effect for attachment security [$F(3,78) = 3.11, P < .05$]. There was no main effect for time and no interaction between attachment security and time. Duncan *post-hoc* tests ($P < .05$) revealed higher cortisol values for the insecure-ambivalently attached infants ($M = 3.21$) as compared to the other three groups (A, $M = 2.34$; B1/B2, $M = 2.49$; B3/B4, $M = 2.47$). Thus, among the highly reactive infants only the insecure ones (C) had higher cortisol values. There were no significant differences between the four groups regarding morning cortisol values [$F(3,69) = 1.32, n.s.$].

Attachment, Emotional Expression, and Adrenocortical Activity

To assess relations between negative emotional expression during the strange situation and cortisol, correlational analyses were conducted. As can be seen from Table 1, the baseline cortisol before the strange situation was not related to emotional expression during the strange situation. In contrast, moderate but systematic correlations were found for the cortisol values after the strange situation indicating heightened adrenocortical activity in infants with more negative emotional expression. The lowest correlations occurred for episodes 2, 6, and 7. For episode 2 this may be due to the low variance because of low occurrence of negative emotional expression (near zero, see Fig. 1). The nonsignificant correlations during the most stressful second separation when the infant was left alone (episode 6) and when the stranger returned (episode 7) points to the different function of negative expression during separation.

Assuming an arousal model, one should expect that securely attached infants and ambivalent infants should exhibit high cortisol values when given a high level of negative emotional expression. Thus, also within the group of securely attached infants, negative emotions should be associated with heightened adrenocortical activity. To test this hypothesis the infants were divided into three groups with respect to negative emotional expression during the separation episodes, in infants with low, medium, and high values, respectively. Regarding attachment categories the distribution of the infants on these groups was as expected (see Table 2). Whereas there was no avoidant infant within the group of infants with high negative emotional expression, almost all of the ambivalent infants were in that group. Securely attached infants were found in each group. The six groups containing more than one child (avoidant infants with low and

TABLE 1
Correlations between Negative Expression and Cortisol during the Strange Situation

<i>Negative Expression</i>	<i>Cortisol</i>		
	<i>Before</i>	<i>After 15min</i>	<i>After 30min</i>
Episode 2 (I,M)	.10	.09	.12
Episode 3 (I,M,S)	-.01	.23*	.25**
Episode 4 (I,S)	.04	.23*	.32**
Episode 5 (I,M)	.13	.37**	.27**
Episode 6 (I)	.10	.02	.11
Episode 7 (I,S)	.15	.17	.21*
Episode 8 (I,M)	.22*	.26**	.25**
Total	.14	.28**	.29**

* $P \leq .05$; ** $P \leq .01$ (two-tailed).
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medium values, ambivalent infants with high values, and secure infants with low, medium, and high values) were selected for comparison. A two-way MANOVA with one repeated measure factor for time (before, after 15min and 30min), one independent factor for the six groups and assessment time as a covariate revealed a main effect for the groups [$F(5,75) = 2.76, P < .05$]. Duncan *post-hoc* tests ($P < .05$) showed that the cortisol values of the C-infants with high negative expression were significantly higher than the values of any other group (see Fig. 4). In particular, it was also higher than the values of the secure infants with high negative expression.

These findings suggest that negative emotional expression may have developed different functions for infants with secure and insecure attachment. To test for different biobehavioural associations within the different attachment groups, negative emotion during separation and reunion were correlated with the cortisol values after the strange situation separately for each group. To control for baseline cortisol values (before strange situation) partial correlations were calculated. As can be seen from Table 3, there are different patterns of associations. Although there were no systematic relations between emotional expression and cortisol in the group of secure children, all other groups show indication of a positive association between emotional arousal and adrenocortical activity. These correlations were highly significant for emotional expression during reunion. In addition, these significant correlations were considerably different from the correlation of the secure group.

DISCUSSION

The main objective of this study was the biobehavioural organisation of infants with different quality of attachment. The replication of earlier findings regarding adrenocortical responses during the strange situation could only be partly accomplished. Heightened adrenocortical activity during the strange situation was only found in the group of insecure-ambivalently attached infants. In line with theoretical assumptions outlined above and in line with the findings of all earlier studies (Gunnar et al., 1989; Spangler & Grossmann, 1993; Hertsgaard et al., 1995;

TABLE 2
Negative Expression during Separation (Reunion) and Security of Attachment^a

<i>Negative Expression</i>	<i>Secure (B)</i>	<i>Avoidant (A)</i>	<i>Ambivalent (C)</i>
Low	15 (20)	17 (15)	0 (0)
Medium	31 (27)	4 (6)	1 (1)
High	20 (19)	0 (0)	15 (15)

^a Number of subjects. Downloaded from jbd.sagepub.com at LMU Muenchen on May 16, 2013

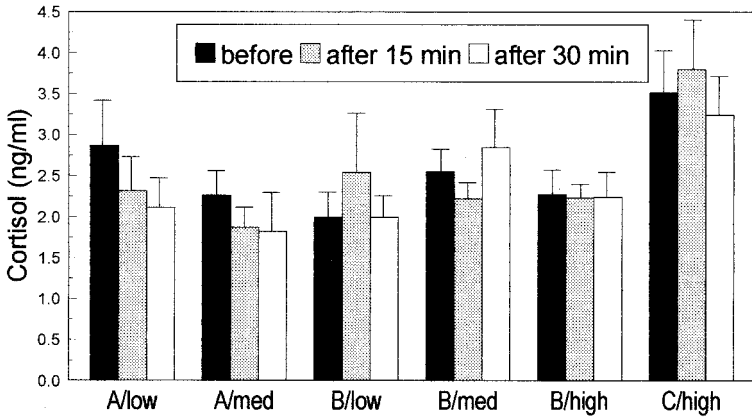


FIG. 4. Cortisol values before, and after the strange situation for secure (B), insecure-avoidant (A) and insecure-ambivalent (C) infants with low, medium, and high negative expression during the separation episodes (episodes 4, 6, 7) of the strange situation. (Error bars indicate 1 SE.)

Nachmias et al., 1996) heightened adrenocortical responses were not observed in securely attached infants. These infants exhibit appropriate behavioural strategies in dealing with the separation from the caregiver. According to Bowlby (1973, p. 150) a physiological adaptation in the “inner ring of life maintaining systems” is not necessary in these infants because regulation can be established on a behavioural level in the “outer ring” of homeostasis.

In contrast to predictions from the coping model, adrenocortical activation was not found for insecure-avoidant subjects. Hitherto, only Spangler and Grossmann (1993) found heightened cortisol values in avoidant infants during the strange situation. However, this was not replicated by Hertsgaard et al. (1995) who found no differences in the cortisol level between avoidant and secure infants. This second failure to replicate is specifically crucial given the fact that the Spangler and Grossmann (1993) study demonstrating adrenocortical activation in avoidant infants included only a small group of avoidant infants, which may raise doubts on the reliability of the effect. The missing adrenocortical activation contradicts interpretations of the avoidant pattern as inappropriate. It may be that the avoidant pattern due to its coherent and organised structure (Main & Solomon, 1990) enables the infant to maintain behavioural organisation at least to a certain degree. According to Main (1981) it can be viewed as a “second best strategy” suitable for minimising the risk of being rejected when contact is needed and, thus,

TABLE 3
 Within-group Correlations between Negative Emotional Expression during Separation. Episodes 4, 6, 7, and reunion (Episodes 5 and 8) and cortisol for the different attachment groups^a

Cortisol	Secure (B) (n = 54-57)		Insecure-avoid (A) (n = 17)		Insecure-ambiv. (C) (n = 12-13)		Disorganised (D) (n = 20-21)	
	Separation	Reunion	Separation	Reunion	Separation	Reunion	Separation	Reunion
After 15min	-.08	-.10	-.07	.00	.23	.88**	.22	.71**
After 30min	.16	-.04	.13	.69**	.14	.89*	.23	.51**

^a Partial correlations controlling for baseline cortisol before strange situation.

The A, B, C groups were selected according to forced classification (attachment security). Thus, there is some overlap with the D-group. * $P \leq .05$; ** $P \leq .01$ (two-tailed).

may serve a specific function. According to Hinde and Stevenson-Hinde (1990) avoidance may, under certain cultural conditions, be seen as adaptive or even more adaptive than the secure strategy. From this perspective any functional behavioural strategies may be appropriate during the strange situation and not only strategies functional from an attachment point of view.

As disorganised infants, by definition, do not have any coherent strategies, behavioural regulation is restricted or even not possible at all. Thus, a coping model would predict heightened cortisol values during the strange situation in this group. As this was indeed found in two previous studies investigating adrenocortical function in disorganised infants (Hertsgaard et al., 1995; Spangler & Grossmann, 1993), the failure to prove adrenocortical activation in disorganised infants in this study is unexpected. Although missing effects may always be due to methodological issues, such as error of measurement, a possible explanation could be that a large proportion of the disorganised infants in this study had an underlying secure attachment pattern. Confidence in the availability of the mother during the strange situation—though incoherently organised on the behavioural level—may work as a social buffer against negative effects of disorganisation, and thus may help to maintain a certain level of behavioural regulation. This explanation was supported by the finding that the disorganised infants with an underlying ambivalent pattern (a group too small for statistical comparisons) were among the subjects with the highest cortisol values. It should be noted, however, that most of the disorganised infants of the Spangler and Grossmann (1993) study also had an underlying secure pattern.

For the first time, this study could demonstrate adrenocortical activation during the strange situation for insecure ambivalent infants. Although these infants try to regulate their labile emotional state by seeking proximity, they are not able to actually use the mother as a source of comfort and emotional regulation. As their behavioural strategy seems to be extremely dysfunctional, heightened adrenocortical activity would be expected from a coping model perspective. Interestingly, the statistical analysis including both pre- and post-assessments of cortisol revealed a main effect for attachment. The cortisol values of the ambivalent group were already higher just before the strange situation, although the findings indicated an additional cortisol increase during the strange situation. This is in line with the behavioural pattern of these infants who, already during the initial less stressful episodes of the strange situation, tend to be anxious and show behaviour indicating high activation of the attachment system being not able to make use effectively of the attachment figure (see also Cassidy & Berlin, 1994). As they did not have heightened morning cortisol this seems to be a definite cortisol response which, however, may be a

response to the whole assessment procedure including the preparatory events rather than the separation alone.

When observing high cortisol values in ambivalently attached infants characterised by marked behavioural distress this could also be interpreted in terms of an arousal model assuming associations between behavioural and physiological activation during stress. This would mean that the adrenocortical activation is part of a general stress response (Selye, 1952), rather than a consequence of an inappropriate attachment behaviour pattern. To test this hypothesis temperamental dispositions in stress reactivity as well as the actual emotional behaviour have to be considered.

First, regarding temperamental dispositions the findings suggest an interplay between attachment and temperamental factors. A comparison of the insecure-ambivalent infants with the secure subcategories B3 and B4 similarly characterised by high proneness to distress (Belsky & Rovine, 1987) revealed that adrenocortical activation was only observed in insecure infants with high proneness to distress (insecure-ambivalent infants) but not in secure ones. Similarly, including behavioural inhibition as a temperamental dimension (e.g. Kagan, 1997) we were able to compare behaviourally inhibited infants with an insecure attachment pattern with those exhibiting a secure attachment pattern. Once again, although infants with low behavioural inhibition (those both securely and insecurely attached) did not exhibit adrenocortical responses, adrenocortical activation was observed in behaviourally inhibited infants with an insecure attachment relationship, but not in those with a secure relationship. Although most of the insecure-ambivalent infants belonged to the group of inhibited infants (cf. Calkins & Fox, 1992), the findings indicated that within the group of infants characterised by behavioural inhibition and insecure attachment not only did the ambivalent infants have high cortisol values but also the avoidant infants did. These interactions between attachment security and temperamental disposition regarding adrenocortical activity is in line with findings of Nachmias et al. (1996) and Gunnar et al. (1989), who both observed cortisol responses during the strange situation as well as other challenging situations in only behaviourally inhibited infants with an insecure attachment relationship. They concluded that a secure attachment relationship may function as a social buffer against less adaptive temperamental dispositions.

Second, regarding emotional behaviour during the strange situation, one would expect positive relations between negative emotional expression and adrenocortical activation within an arousal model perspective. Using a coping perspective this relation would not be expected in the background of attachment theory. As an attachment behaviour, negative expression fulfils a social function by signalling the need for proximity after separation from the caregiver and thus is not restricted to the individual function of

emotional expression. The findings of our study clearly demonstrate that this social function is effective in securely attached infants not only on the behavioural level but also on the physiological level. Even those exhibiting high emotional distress during separation did not show adrenocortical activation. In contrast, the ambivalent infants, also characterised by marked negative emotionality, show high cortisol values during the strange situation. Thus, the biobehavioural association between emotional and physiological processes seems not to be due to a common cause of activation (i.e. high adrenocortical activation is not necessarily combined with a high level of emotional distress). This contradicts an interpretation of the findings in terms of an arousal model.

Going beyond group comparisons of mean differences we analysed biobehavioural relations within the different attachment groups. The biobehavioural correlations within the groups suggest different forms of biobehavioural organisation in infants with different qualities of attachment. Whereas emotional distress and adrenocortical activity were unrelated within the group of secure infants, high behavioural distress was associated with adrenocortical activation, not only for the ambivalent infants but also for the avoidant and disorganised infants. Thus, in securely attached infants, crying or negative expression is not related to physiological stress. It may be used by some or many of them as a means for coping with separation, but other infants may exhibit different attachment behaviours such as searching for or calling the mother. Whether used or not does not seem to be relevant for physiological stress in these infants.

In contrast, the restriction of the function of emotion to expression may be essential in all groups of insecurely attached infants. As emotional expression does not fulfil its social function as an attachment behaviour it may only be an epiphenomenon of experienced stress. The inability to use emotional expression for emotional regulation with the help of the caregiver may be due to different reasons in the different groups. Emotional expression would actually be used by ambivalent infants, but cannot be effectively used for regulation (Cassidy & Berlin, 1994). In avoidant infants, experiences with an unavailable caregiver may lead to fear of being rejected (e.g. Ainsworth et al., 1978). The consequence is a suppression of communication of emotional distress in attachment-activating situations, which means that these infants do not even try to use attachment behaviour for effective emotional regulation. The significant correlation for avoidant infants may be somewhat surprising given the low occurrence of negative expression during separation in these infants ($M = 10.6\%$). With values ranging from 0% through 35% of negative emotion ($SD = 12.7$) there was, however, sufficient variance for a valid correlation determination. Finally, the social adaptability of

emotional expression may be impaired in disorganised infants because it cannot be incorporated within a coherent behavioural strategy. In sum, expression of negative emotions cannot be seen as an adaptive coping behaviour in insecure infants, because either this type of attachment behaviour is not part of their behavioural strategy or, if available, cannot be used effectively for behavioural regulation. Thus, the function of emotional expression may still be given on an individual level, however, without its social or relational function as a means for communication (Campos et al., 1989). This is specifically supported by the fact that the correlations were most prominent for negative emotional expression during the reunion episodes of the strange situation.

In conclusion, the biobehavioural organisations of securely and insecurely attached infants seems to differ and may be described by different models of interplay between the behavioural and the adrenocortical system. A coping model may be most appropriate to describe the biobehavioural organisation of securely attached infants, whereas biobehavioural organisation in insecure or disorganised infants may fit assumptions of an arousal model. Securely attached infants may have a variety of attachment behaviours at their disposal, from which they can select the most appropriate one depending on the demands of the situation (goal-corrected behaviour). As crying is not the only possible response, we do not find a negative correlation between emotional distress and adrenocortical activation within the secure group. If secure infants actually do cry, this primarily seems not to be a consequence of a general activation process. It is not related to physiological arousal, because it fulfils the coping function. In contrast, insecurely attached infants only have a restricted set of coping responses which for the most part may be inappropriate for behavioural regulation. As crying cannot be used as an attachment behaviour (as in avoidant infants) or is dysfunctional (as in ambivalent infants) or cannot be organised within a coherent behavioural strategy (as in disorganised infants) it may be a fixed part of a general stress response pattern. The application of different biobehavioural models to the specific attachment patterns should be seen as an assumption based on our findings. Further evidence is needed to prove the validity of this assumption.

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