Research Article

Prenatal Stress and Human Infant Development

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Abstract

Background: There are several studies in rodents, primates but also in humans showing that prenatal stress has a potential impact on cognitive and emotional development in the offspring which may even last until adolescence. Therefore, this study investigated whether maternal stress in pregnancy is associated with impairment of infant cognitive or behavioural development. Method: N=108 pregnant mothers collected salivary cortisol once during each trimester of pregnancy. N=104 babies were tested with the Bayley Scales of Infant Development at an infant’s age of five months. Results: Mothers’ salivary cortisol levels at the end of pregnancy correlated negatively with mental developmental status of the offspring (r=-0.25, p=0.01). Children of mothers in a high stress subgroup of pregnant women’ perform worse in the Mental and Motor Scale of Bayley Scales of Infant Development (p<0.05). Conclusions: Prenatal hormonal stress at the end of pregnancy seems to have a significant impact on the outcome of the offspring. These data strengthen the ‘Fetal Programming Hypothesis’ stating that maternal mental and hormonal state in pregnancy impacts early human development.

Keywords: pregnancy, prenatal stress, cortisol, infant development, child development, Fetal Programming Hypothesis

Introduction

Evidence for an impact of prenatal stress on early behavioural and mental development in human infants has been recently provided [1-3]. Psychological prenatal stress was found to be associated with changes in maternal cortisol levels and lower infant mental and motor scores as well as goal-directedness in later infancy [4]. Psychological measures like the Pregnancy-related Anxiety Questionnaire in mid-pregnancy correlated with mental and motor development of the child at an infants’ age of eight months. Furthermore, early morning value of cortisol at the end of pregnancy revealed an association with impairment of infants’ developmental status at 3 and 8 months [1]. The foci of primary interest in the studies looking at prenatal stress and outcome for the offspring are: infant temperament [5,6], consequences affecting infant growth [7,8], premature birth [9,10], attentional organization [11-14] and natural disaster [15,16]. Correspondingly studies in primates and rodents have described differential impact of prenatal stress on the offspring [17-20].

Efforts to elucidate the potential biological background of this phenomenon have primarily focused on rodents and primates: Infant behavioral alterations linked to maternal stress during pregnancy seem to be associated
with changes in the hypothalamo-pituitary-adrenal response in rats [21,22]. Altered receptor densities in hippocampal sites have been proposed [23,24] as well as altered function of the amygdale [18,25].

Derived from these animal models the 'Fetal Programming Hypothesis' was confirmed assuming that the human fetal hypothalamic-pituitary-adrenal (HPA) axis is programmed during pregnancy. According to this hypothesis, prenatal events and stressors are programming the infant brain altering its normal neurodevelopment and behaviour [26]. Studies looking at the fetal HPA and later development of the children of prenatally stressed mothers are still rare and have a variety of methodological weaknesses [27].

On a clinical level, a recent prospective longitudinal study [28] on antenatal state/trait anxiety has provided evidence for an association between prenatal anxiety and externalizing problems in later childhood: high levels of antenatal anxiety in the second trimester of pregnancy, explained 22% of the variance of ADHD–symptoms at age 8-9 years and 15 % of the variance of externalizing problems. In the AVON Longitudinal Study [29] prenatal anxiety assessed with a short screening questionnaire was found to double the risk for hyperactivity and inattention problems as well as conduct disorder at an age of 47 and 81 months. In the same study, maternal anxiety at 32 weeks of pregnancy was also found to be related to observer report measures of attention at 3 weeks and 12 months of age.

In the light of the actual relevance of these findings, specifically for the understanding of the pathogenesis of ADHD, this study examined in a sample of 104 human infants whether prenatal hormonal stress has impact on infant development.

Methods

Sampling

Data collection took part between November 2007 and August 2009. The sample will be described in more detail below. It consists of over hundred healthy European pregnant women recruited in early pregnancy (week of gestation: 13.6±1.68) through local newspaper, homepages and in obstetricians’ offices in Heidelberg and surrounding area. Exclusion criteria were (a) inability to speak and read German language, (b) twin pregnancy, (c) advanced pregnancy (>19 week of pregnancy), (d) inability to come to the laboratory at an infant’s age of three and five months. The study protocol was approved by the ethic committee of the university Clinic of Heidelberg. All pregnant women were informed about the course and the aim of the study and gave written informed consent.

Study design

A prospective longitudinal study was assessed collecting salivary cortisol in each trimester of pregnancy on three following days in a defined time interval under controlled conditions. At five month of age, infants’ mental and motor development were tested by the Bayley Scales of Infant Development [30]. Mothers brought their infants to the laboratory at a day-time when their infant was alert, fed and rested.

Measures

Prenatal stress measures: Stress was detected by the physiological hormonal response of the pregnant subjects in salivary cortisol. Basal salivary cortisol levels were collected using salivette collection devices (Sarstedt, Germany). The subjects were asked to suck on cotton rolls for two to three minutes on three following days between eleven and one o’clock in the morning. Subjects were instructed to collect saliva in a quiet, non-stressed situation before lunch time. They had to store the salivettes in the refrigerator (–20°C) and sent them back in a covered envelope after they collected three cortisol probes. The salivettes were centrifuged at 3000rpm for 5 minutes and salivary-free cortisol concentrations were analyzed in the pharmacological laboratory of the University of Heidelberg.

Postnatal measures: 1) Development of the child: Child Development was assessed via the Bayley Scales of Infant Development II according to [30] at an infants’ age of five months. The Bayley Scales of Infant Development
include three subscales: Motor Scale, Mental Scale and Behaviour Rating Scale. The Mental and Motor Scales measure the infants' present stage of cognitive, language, personal-social, and fine and gross motor development. The Motor Scale measures the control of the gross and fine muscle groups and their functions: rolling, crawling and creeping, sitting, standing, walking, running and jumping; and supplementary fine motor handling involved in prehension, adaptive use of writing implements and the imitation of hand activities. The Mental and Motor Scale are nominal Dichotomy; The Behaviour Rating Scale is an ordinal scale and allows to assess the child’s development on a 5-point- Likert scale, ranging from 1 (=worst coding) to 5 (=best coding). Two researchers trained for reliability independently observed the videotaped sessions for the Behaviour Rating Scale; Cohen’s kappa was 0.88. Mental and Motor Scale were coded during the testing and was recoded by a second trained coder via the videotaped sessions. Cohen’s kappa was 0.95.

Statistical analysis

Interrater-reliability for the development-ratings of the child was examined by intraclass correlation. Kolmogorov-Smirnov-Test was applied to test for normal distribution. Analyzing the alteration of prenatal cortisol over the course of pregnancy paired student’s t-test was assessed. For each trimester of pregnancy correlation between prenatal stress and postnatal developmental status of the child was assessed by Pearson product-moment correlation for normally distributed data. Spearman’s rho correlation was used for non-normally distributed data. Mann-Whitney U-test was assessed comparing group differences between women classified as ‘low-stressed’ and ‘high-stressed’ by median-split of the cortisol level in each trimester of pregnancy. Two sub- groups of ‘extreme low-stress’ and an ‘extreme high-stress’ were assessed by using the 25% and 75%-percentile of the cortisol level. Group differences for the infant development was tested by Mann Whitney U-Test. A p-value of 0.05 was regarded as significant. All calculations were done with the computer software SPSS 17.0.

Results

Sampling

Out of 121 women contacted, 111 women were included in the first study wave during early pregnancy (gestational week 13.6 ± 1.68). Ten women declined to take part at the study because of early abortion (3, 30%), lack of interest (6, 60%), or illness of the mother (1, 10%). Out of 111 women included in the first study wave, N=104 women sent back their second data set in mid-pregnancy (gestational week 22.0 ± 2). Drop-outs occurred due to abortion (1, 14.5%), loss of data during mailing (1, 14.5%), and forgetting to send back the data set in time (5, 71%). N=106 women were included in the last study wave (gestational week 32.13 ± 2) during pregnancy. Drop-outs in the last study wave can be explained because of handicap of the fetus (1, 33%) and forgetting to send back the data set in time (2, 67%). N=104 mother-infant dyads came to the laboratory at an infant’s age of five (M=5.1 months, SD=0.3) months. The final sample consists of 104 women ranging from 17 and 42 years of age (M=31.04, SD=5.23) at the beginning of their pregnancy. Seventy-two per cent of our sample finished the highest normal school track in Germany and received their “Abitur” (equivalent to first year college).

Descriptive analyses of physiological prenatal stress measures

The mean basal salivary cortisol level (in ng/ml) increases significantly over the course of pregnancy (t(102)=6.39, p<0.01). From early pregnancy (M=2.12; SD=1.37) to mid-pregnancy (M=3.18; SD=1.69) cortisol levels increase significantly (t(103)=5.75, p<0.01). From second to last trimester of pregnancy (M=3.45; SD=1.68) cortisol levels show further increase but this increase fails to reach level of significance (t(102)=1.62, p=0.11). The increase of cortisol over the course of pregnancy is in accordance with findings reported by Ruiz and colleagues [8]. It thus provides evidence suggesting that our measurement of physiological stress is valid.
Descriptive analyses of developmental status of the children

Mean mental developmental status of our sample is M=95.57, SD=9.12. Mean motor developmental status is M=87.95, SD=4.38. Mean Behavior Rating is M=80.66, SD=9.47. Comparing our results with norm data from Bayley [30], we can conclude that our sample is somewhat but not significantly below the average mean mental and motor developmental status.

Correlation between prenatal stress and Infant development

Prenatal stress in the beginning of pregnancy is not correlated with Mental (r=-.001, p=0.99), Motor (r=-0.02, p=0.86) or Behavior Rating Scale (r=0.06, p=0.53). Prenatal stress in mid-pregnancy is somewhat more associated with Mental (r=-0.13, p=0.24), Motor (r=-0.12, p=0.24) and Behavior Rating Scale (r=-0.09, p=0.41), but not significantly. Prenatal stress at the end of pregnancy is significantly correlated with Mental developmental status of the child (r=-0.25, p=0.01), but not significantly correlated with Motor Development (r=-0.16, p=0.13) and Behavior Rating Scale (r=-0.06, p=0.57).

Group comparison by median-split of the prenatal maternal cortisol level

We use the median-split of the cortisol level for each trimester of pregnancy to compare the group classified as 'high-stressed' and 'low-stressed' for the outcome of the developmental status of the children. Median cortisol for the first trimester of pregnancy is Median=1.76. In the group of 'low-stressed' women, mean cortisol level is M=1.14, SD=0.41, in the 'high-stressed' group Mean cortisol level is M=3.14, SD=1.31. We compare outcome of the child for these two groups. Motor developmental status of the 'low-stressed' group (N=46) is marginally significant higher (M=97.22, SD=8.21) than in the 'high-stressed' group (N=51; M=93.73, SD=9.82, U (94)=1.84, p=0.07). Mental developmental status in the 'low-stressed-group' is also higher (M=88.52, SD=4.44) than in the 'low-stressed' group (M=87.27, SD=4.36). But this difference does not reach a level of significance (U (94)=148, p=0.14). For the Behavior Rating Scale, we can see the same pattern of results with higher mean values in the 'low-stressed' group (M=80.85, SD=10.41) in comparison to the 'high-stressed' group (M=79.78, SD=8.59), but not to a significant level (U (94)=0.99, p=0.32).

In mid-pregnancy, median cortisol is 2.85 with mean cortisol level in the 'low-stressed' group of M=1.85, SD=0.59 (N=47) and M=4.55, SD=1.29 in the 'high-stressed-group' (N=48). The Motor Scale is higher in the group of the 'low-stressed' group (M=96.09, SD=10.40) than in the 'high-stressed' group (M=94.98, SD=8.10), but the difference does not reach a level of significance (U (94)=1.16, p=0.25). The difference in both groups looking at the Mental Scale does not reach a significant level (U (94)=1.33, p=0.18), so does the Behavior Rating Scale (U (94)=-0.69, p=0.49).

At the end of pregnancy, median cortisol level reaches a value of Median=3.3. Cortisol level in the group of 'low-stressed' (N=45) mothers is M=2.21, SD=0.69 and for 'high-stressed' (N=49) mothers M=4.77, SD=1.37. Differences in both groups classified as 'low- vs. high-stressed' comparing Mental (U (93)=1.49, p=0.14), Motor (U (93)=1.06, p=0.29), and Behavior Rating Scales (U (93)=0.61, p=0.54) does not reach a level of significance.

We can conclude that the only marginally significant differences between the ‘high-and low-stressed’ group exists when we compare the motor developmental status with prenatal cortisol in the first trimester of pregnancy. All other differences do not reach a level of significance. However, we can see that all differences tend in the expected direction. Mothers with high cortisol levels have children with retarded motor, mental and emotional development. Comparing the developmental status dividing the group via the median-split of the cortisol level does not give significant results. We assumed that medium stress levels do not have significant impact on infant development. For this reason, the next methodological step was to divide the group of subjects in a sub-group of ‘extremely low-
stressed' and 'extremely high-stressed' mothers by using the 25%- and 75% percentile of cortisol levels for each trimester of pregnancy. In this calculation, all subjects with medium cortisol levels will be excluded.

**Extreme-group comparison**

In first trimester of pregnancy, cortisol level of the 25%- percentile is 1.15, 50%-percentile=1.76 and 75%-percentile=2.83. We divided the group in an 'extreme high-stressed' group (N=26) which mean cortisol level is M=3.98, SD=1.38 and an 'extreme low-stressed' group (N=21) with a mean cortisol level of M=0.79, SD=0.28. The difference of these both extreme groups for prenatal stress at the beginning of pregnancy in the Mental (U (47)=0.833, p=0.41), Motor (U (46)=1.297, p=0.19) and Behavior Rating Scale (U (46)=0.568, p=0.57) does not reach a level of significance.

In mid-pregnancy, 25% percentile of cortisol is 1.85, 50% percentile is 2.85 and 75% percentile is 4.37. The group of 'extreme low-stressed mothers' (N=24) has a mean cortisol level of Mean=1.36, SD=0.37, whereas the 'extreme high-stressed' group (N=25) has a mean cortisol level of M=5.58, SD=1.09. In the Mental (U (48)=1.30, p=0.20), Motor (U (48)=1.14, p=0.25) and Behaviour Rating Scale (U (48)=0.78, p=0.44), children of highly prenatal stressed mothers performing worse than children from prenatal non-stressed mothers. Anyhow, the group difference does not reach a level of significance.

At the end of pregnancy, 25% percentile of cortisol is 2.27, 50% percentile is 3.30 and 75% percentile is 4.47. The group of 'extreme low-stressed mothers' (N=24) has a mean cortisol level of M=1.70, SD=0.55, whereas the 'extreme high-stressed' group (N=25) has a mean cortisol level of M=5.75, SD=1.34. Children of mothers with a high cortisol level at the end of pregnancy perform significantly worse (M=93.96, SD=8.25) in the Motor Scale of Bayley Scales than children of mothers with a very low cortisol level (M=97.63, SD=12.04). This difference reach the level of significance (U (47)=1.94, p=0.05). Also, children from mothers with high cortisol level at the end of pregnancy perform significantly worse (M=86.67, SD=4.52) in the Mental Scale of Bayley Scale than children from mothers with very low cortisol levels (M=89.67, SD=3.76, U (47)=2.144, p=0.032). In the Behavior Rating Scale, differences in the extreme groups does not reach a significant level (U (47)=0.88, p=0.38), even though we can see that children of highly stressed mothers have lower Behaviour Rating scores (M=80.29, SD=8.26) than children from non-stressed mothers (M=81.17, SD=10.26). All descriptive data of Mental, Motor and Behavior Rating Scale in the groups of ‘extreme highly stressed’ and ‘extreme non-stressed’ mothers over the course of pregnancy can be seen in Table 1-3.

**Table 1:** Motor, Mental and Behavior Rating Scale of children five months of age of mothers classified prenatally as ‘extreme low-stressed’ and ‘extreme high-stressed’ in the beginning of pregnancy (T1)

<table>
<thead>
<tr>
<th>T1</th>
<th>Group</th>
<th>n</th>
<th>p</th>
<th>M</th>
<th>SD</th>
<th>df</th>
</tr>
</thead>
<tbody>
<tr>
<td>Motor Scale</td>
<td>Extreme low-stress</td>
<td>21</td>
<td>0.2</td>
<td>97.5</td>
<td>7.05</td>
<td>46</td>
</tr>
<tr>
<td></td>
<td>Extreme High-stress</td>
<td>26</td>
<td></td>
<td>94.7</td>
<td>8.63</td>
<td></td>
</tr>
<tr>
<td>Mental Scale</td>
<td>Extreme Low-stress</td>
<td>21</td>
<td>0.4</td>
<td>88.7</td>
<td>3.7</td>
<td>46</td>
</tr>
<tr>
<td></td>
<td>Extreme High-stress</td>
<td>26</td>
<td></td>
<td>87.6</td>
<td>4.9</td>
<td></td>
</tr>
<tr>
<td>Behaviour Rating Scale</td>
<td>Extreme Low-stress</td>
<td>26</td>
<td>0.6</td>
<td>76.6</td>
<td>11.2</td>
<td>46</td>
</tr>
<tr>
<td></td>
<td>Extreme High-stress</td>
<td>21</td>
<td></td>
<td>79.1</td>
<td>9.21</td>
<td></td>
</tr>
</tbody>
</table>

T1: Group Classification by 25%- 75% percentile of prenatal cortisol level of the mothers at the beginning of pregnancy; Motor Scale: Motor Scale of Bayley Scales of Infant Development; Mental Scale: Mental Scale of Bayley Scales of Infant Development; Behaviour Rating Scale: Behaviour Rating Scale of Bayley Scales of Infant Development.
Table 2: Motor, Mental and Behavior Rating Scale of children five months of age of mothers classified prenatally as ‘extreme low-stressed’ and ‘extreme high-stressed’ in mid-pregnancy (T2)

<table>
<thead>
<tr>
<th>T2</th>
<th>Group</th>
<th>n</th>
<th>p</th>
<th>M</th>
<th>SD</th>
<th>df</th>
</tr>
</thead>
<tbody>
<tr>
<td>Motor Scale</td>
<td>Extreme low-stress</td>
<td>24</td>
<td>0.25</td>
<td>96.17</td>
<td>12.18</td>
<td>48</td>
</tr>
<tr>
<td></td>
<td>Extreme High-stress</td>
<td>25</td>
<td></td>
<td>94.96</td>
<td>7.18</td>
<td></td>
</tr>
<tr>
<td>Mental Scale</td>
<td>Extreme Low-stress</td>
<td>24</td>
<td>0.2</td>
<td>88.92</td>
<td>4.06</td>
<td>48</td>
</tr>
<tr>
<td></td>
<td>Extreme High-stress</td>
<td>25</td>
<td></td>
<td>87.32</td>
<td>3.82</td>
<td></td>
</tr>
<tr>
<td>Behaviour Rating Scale</td>
<td>Extreme Low-stress</td>
<td>24</td>
<td>0.46</td>
<td>81.83</td>
<td>11.18</td>
<td>48</td>
</tr>
<tr>
<td></td>
<td>Extreme High-stress</td>
<td>25</td>
<td></td>
<td>80.36</td>
<td>8.44</td>
<td></td>
</tr>
</tbody>
</table>

T2: Group Classification by 25%- 75% percentile of prenatal cortisol level of the mothers in mid-pregnancy;
Motor Scale: Motor Scale of Bayley Scales of Infant Development;
Mental Scale: Mental Scale of Bayley Scales of Infant Development;
Behaviour Rating Scale: Behaviour Rating Scale of Bayley Scales of Infant Development.

Table 3: Motor, Mental and Behavior Rating Scale of children five months of age of mothers classified prenatally as ‘extreme low-stressed’ and ‘extreme high-stressed’ at the end of pregnancy (T3)

<table>
<thead>
<tr>
<th>T3</th>
<th>Group</th>
<th>n</th>
<th>p</th>
<th>M</th>
<th>SD</th>
<th>df</th>
</tr>
</thead>
<tbody>
<tr>
<td>Motor Scale</td>
<td>Extreme low-stress</td>
<td>24</td>
<td>0.05</td>
<td>97.63</td>
<td>12.04</td>
<td>47</td>
</tr>
<tr>
<td></td>
<td>Extreme High-stress</td>
<td>24</td>
<td></td>
<td>93.96</td>
<td>8.25</td>
<td></td>
</tr>
<tr>
<td>Mental Scale</td>
<td>Extreme Low-stress</td>
<td>24</td>
<td>0.03</td>
<td>89.67</td>
<td>3.76</td>
<td>47</td>
</tr>
<tr>
<td></td>
<td>Extreme High-stress</td>
<td>24</td>
<td></td>
<td>86.67</td>
<td>4.52</td>
<td></td>
</tr>
<tr>
<td>Behaviour Rating Scale</td>
<td>Extreme Low-stress</td>
<td>24</td>
<td>0.38</td>
<td>81.17</td>
<td>10.26</td>
<td>47</td>
</tr>
<tr>
<td></td>
<td>Extreme High-stress</td>
<td>24</td>
<td></td>
<td>80.29</td>
<td>8.26</td>
<td></td>
</tr>
</tbody>
</table>

T3: Group Classification by 25%- 75% percentile of prenatal cortisol level of the mothers at the end of pregnancy;
Motor Scale: Motor Scale of Bayley Scales of Infant Development;
Mental Scale: Mental Scale of Bayley Scales of Infant Development;
Behaviour Rating Scale: Behaviour Rating Scale of Bayley Scales of Infant Development.

Discussion

In summary, we found that (1) prenatal maternal cortisol in the last trimester of pregnancy correlates negatively with Mental Development of the child, but not at an earlier stage of pregnancy. It seems that the influence of cortisol on the developmental status of the child increases over the course of pregnancy indicating that higher cortisol levels of the mother cause retarded motor and mental development of the child. (2) Group differences for median-split of the prenatal hormonal data do not reveal any significant differences concerning the outcome of the child. (3) Looking at extreme sub-groups using women who have highest and lowest cortisol levels (25%- 75%-percentile) during pregnancy, the comparison at the end of pregnancy but not earlier in pregnancy shows significant differences. According to our hypothesis, our data show that mothers with high prenatal cortisol levels have children who perform worse in the mental and motor development test at a postnatal age of five months than children from prenatal non-stressed mothers. These data are in line with the findings of the literature described above [1,11,4,31]. However, what this study adds is a detailed knowledge on the time course of the association between maternal cortisol in pregnancy and infant development. As the studies cited above did not assess cortisol in each trimester of pregnancy or assessed cortisol only twice in pregnancy, they therefore cannot provide a differential analysis of the significance of time of stress onset in pregnancy. The latter seems to play a crucial role going in line with the findings from Huizink [11] and Buitelaar [1]. From these data we can conclude that prenatal hormonal stress of the mother at
the end of pregnancy has a crucial impact on the mental and motor developmental status of the child at an infant’s age of five months. It seems that looking at medium-stressed mothers no correlation between cortisol and child development can be found. This is predominantly present in a comparison of the extreme sub-groups.

These findings can be explained potentially by transmission of cortisol via the placenta and affecting infants’ brain. This is supported in the literature by the following findings: Davis and co-workers [32] found that infants of mothers who had received prenatal cortisol treatment showed a blunted cortisol response to novel and stressful stimuli. Furthermore, it was shown that postnatal behavioural alterations induced by prenatal stress were depending on cortisol response and were not present to the same degree in the offspring of mothers who did not secrete cortisol in response to exogenous stressors because it was experimentally blocked [17].

Limitations of the validity of the present findings result from the fact that prenatal stress was only measured by salivary cortisol on three following days. We know that cortisol has a diurnal rhythm with highest levels in the morning and decreasing over the day. As we measured salivary cortisol between 11am and 1pm, we do not have a cortisol profile but only mean cortisol levels for each trimester of pregnancy. Furthermore, we only looked at prenatal cortisol responses of the mothers and its impact on child development. The correspondence of hormonal responses with subjective feelings of being stressed or whether emotional prenatal stress is influencing children’s development is not looked at.

Furthermore, our data do not contain information about extreme emotional conditions: the sample studied here was a community sample of mentally healthy mothers. The effects of cortisol levels on children’s developmental status may be even stronger when comparing clinical with community samples. In a clinical sample (f. e. migrants, people living in poverty, abused mothers); the range of hormonal and emotional stress may be higher than in the present sample. We can speculate that in these samples, prenatal stress might have even stronger effects on the development of the child and even from the beginning of pregnancy. For these reasons, our results can only be postulated for a ‘normal sample’ and might be even stronger in a clinical sample.

In the light of previous work and these data, further research on the impact of prenatal stress and human infant development seems to be of particular importance in order to identify specific targets for prevention of psychopathology, specifically ADHD.

References


