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Anxious attachment style predicts an enhanced cortisol response to group psychosocial stress

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Running Head: Stress reactivity and attachment style

Key words: Trier Social Stress Test; TSST-G; HPA axis; stress reactivity; saliva; group stressor; healthy females

Abstract

Insecure attachment style is associated with poor health outcomes. A proposed pathway implicates the hypothalamic-pituitary-adrenal axis (HPA-axis), dysregulation of which is associated with a wide range of mental and physical ill-health. However data on stress reactivity in relation to attachment style is contradictory. This relationship was examined using the novel Trier Social Stress Test for groups (TSST-G): a group-based acute psychosocial stressor. Each participant, in the presence of other group members, individually performed public speaking and mental arithmetic tasks. Seventy-eight healthy young females (20.2 ± 3.2 years), in groups of up to 6 participants completed demographic information and the Vulnerable Attachment Style Questionnaire (VASQ), and were then exposed to the TSST-G. Physiological stress reactivity was assessed using salivary cortisol concentrations, measured on 7 occasions at 10-minute intervals. Vulnerable attachment predicted greater cortisol reactivity independent of age, smoking status, menstrual phase and BMI. Supplementary analysis indicated that insecure anxious attachment style (high scores on the insecurity and proximity seeking sub-scales of the VASQ) showed greater cortisol reactivity than participants with secure attachment style. Avoidant attachment style (high scores for insecurity and low scores for proximity seeking) was not significantly different from the secure attachment style. Attachment style was not associated with **the timing of the cortisol peak** or post-stress recovery in cortisol concentrations. These findings in healthy young females indicate subtle underlying changes in HPA axis function in relation to attachment style and may be important for future mental health and well-being.

Introduction

Attachment style is suggested to be important for regulating threat appraisal, stress response and recovery from stress, although the mechanisms underlying this complex interplay are not well understood (Diamond, 2001). Within the adult attachment literature,

insecure attachment style is generally conceptualised along two dimensions, namely attachment anxiety and avoidance (Brennan et al., 1998). A securely attached individual is considered to be an individual with low levels of both (Brennan et al., 1998). High attachment anxiety is associated with preoccupation with the availability and responsiveness of the other, maximization of negative experiences and hyper-vigilance to potential threat. Attachment avoidance is associated with a tendency to devalue intimacy and dependency and maximize autonomous behaviour strategies when faced with potential threat. Insecure attachment style is known to predict a range of poor physical and mental health outcomes (Bifulco et al., 2002a; Bifulco et al., 2002b; Carr et al., 2013; Jinyao et al., 2012; Puig et al., 2013). The biological underpinnings of these links however are not clear.

One of the proposed pathways implicates the hypothalamic-pituitary-adrenal (HPA) axis (Repetti et al., 2002), dysregulation of which is associated with a wide range of mental and physical ill-health (McEwen, 2000). A flattened diurnal rhythm of cortisol secretion has been reported in anxious attachment style (Oskis et al., 2011; Quirin et al., 2008) however there are mixed findings from studies examining reactivity of the HPA axis in relation to attachment style. For example in one study avoidant (but not anxious) attachment predicted enhanced stress-induced cortisol responding in females (Powers et al., 2006) whilst the opposite was found in another study (Quirin et al., 2008). Other studies have reported insecure dismissing attachment style to predict enhanced cortisol reactivity (Pierrehumbert et al., 2012; Rifkin-Graboi, 2008) whereas secure and dismissive attachment styles have been reported as similar elsewhere (Kidd et al., 2011). Further studies show no relationship between cortisol responding to a stressor and attachment style (e.g. Ditzen et al., 2008; Smeets, 2010).

Whether attachment style predicts acute stress responding remains unclear. The disparity in the literature may in part be related to the wide array of methodologies that have been used to investigate this issue. Stressors have ranged from the Trier Social Stress test (TSST) for

individuals (e.g. Ditzen et al., 2008; Smeets, 2010) to experimental conflict negotiation (e.g. Powers et al., 2006) visualization of hypothetical distressing situations (e.g. Rifkin-Graboi, 2008) and behavioural interference tasks (Kidd et al., 2011). The most commonly used tool to assess attachment style in adult stress reactivity studies is the Experiences in Close Relationships Scale (Brennan et al., 1995), which assesses attachment in romantic relationships. The Vulnerable Attachment Style Questionnaire (VASQ; Bifulco et al., 2003) is arguably a more appropriate measure for use in research investigating attachment and HPA axis activity as, rather than romantic attachment, it focuses on how individuals generally relate to others. Furthermore it performs somewhat better in predicting depression than other self-report measures of attachment (Bifulco et al., 2003), as well as predicting negative psychosocial well-being and mental health in university students (Carr et al., 2013). The VASQ, was developed and validated in relation to an in-depth interview procedure (Attachment Style Interview; Bifulco et al., 2002a; Bifulco et al., 2002b) which has been used in previous research examining HPA axis activity and attachment style (Oskis et al., 2014; Oskis et al., 2011).

The Trier Social Stress Test (TSST; Kirschbaum et al., 1993) comprises uncontrollability and socio-evaluative threat known to reliably activate HPA axis function (Dickerson et al., 2004) and has recently been adapted for use in group settings: the TSST-G (von Dawans et al., 2011). A primary motivation was to increase the rate of participant exposure to the TSST but it provides the opportunity to examine the impact of social dynamics on stress reactivity (Häusser et al., 2012). In the present study we adapted the TSST-G to maximise opportunities for group interaction which may attenuate or increase stress reactivity depending on the characteristics of the individual within the group. Given that those with high attachment insecurity easily perceive threats in their environment, frequently experience social interactions as stressful and excessively ruminate about psychologically distressing experiences (Burnette et al., 2009; Shaver et al., 2002) they might find the group version of

the TSST particularly stressful. We chose to investigate an all-female sample since sex is known to moderate the link between attachment style and HPA reactivity (Kiecolt-Glaser et al., 1996; Kirschbaum et al., 1995; Stroud et al., 2002). Young, healthy participants were recruited to explore whether attachment style might be a pre-clinical indicator of vulnerability rather than a consequence of concurrent poor health. Due to discrepancies in the stress reactivity and attachment literature the aim of this study was to examine self-reported attachment and physiological stress responding to a group psychosocial stressor.

Methods

Participants

Eighty-one female undergraduate student participants were recruited. They did not receive financial incentives but did receive course credits. Cortisol data was missing for one participant as the salivary volume was insufficient for assay purposes, and another participant did not complete the attachment questionnaires. A single participant was removed from the data set on the basis that their cortisol data were more than 5 standard deviations above the mean for each sample, and their data remained as outliers following square root transformation. Analyses were performed on 78 participants, age ranging from 18 to 33 (mean \pm SD: 20.1 \pm 3.1) years. Participants were ethnically diverse; of those who disclosed their ethnicity, 26 were Asian (Indian, Chinese, Pakistani, Bangladeshi, Arabic), 31 were white European, 13 were African Caribbean, and 4 were mixed race.

To control for **sex** differences in cortisol reactivity only females were recruited. To reduce the impact of variables known to influence cortisol reactivity exclusion criteria included medication, illness and history of psychiatric illness. As cortisol reactivity is influenced by the menstrual cycle and body mass index (BMI) the number of days since last period was recorded, as was height and weight (Dockray et al., 2009; Smyth et al., 2013). Two participants used oral contraceptives. The majority of participants (86%) were non-smokers.

Vulnerable Attachment Style Questionnaire (VASQ)

The VASQ (Bifulco et al., 2003) is a brief self-report tool, which assesses general adult attachment. It is designed to assess overall attachment vulnerability as well as two dimensions of attachment: a global dimension of attachment insecurity common to all insecure subtypes (representing a deep-rooted mistrust of others and their motives) and a proximity seeking dimension reflecting the strategy individuals use to manage their insecurity (i.e. some individuals with high insecurity develop excessive neediness and vigilance of others, whilst other individuals develop an aversion to closeness with others). The scale comprises 22 items measured on a five-point scale, ranging from strongly disagree to strongly agree. Low scores on the 12-item insecurity subscale (e.g. "I find it hard to trust others" and "People let me down a lot") represent secure attachment and high scores reflect insecure attachment. The proximity seeking subscale consists of 10 items (e.g. "I get anxious when people close to me are away" and "I look forward to spending time on my own"). Low scores represent propensity for avoidant behaviour and high scores reflect a need for closeness with others. Cronbach's alpha was .81 for the insecurity scale and .74 for the proximity scale. A total attachment vulnerability measure can be derived by summing items on both scales. The VASQ can also be used to categorise participants according to secure, insecure anxious or insecure avoidant attachment styles. The insecure anxious attachment style category is derived from high scores on both insecure and proximity-seeking measures. The insecure avoidant type category is derived from high scores on the insecurity scale and low scores on the proximity scale.

Procedure

The study was approved by the University of Westminster Ethics Committee. Following recruitment, groups of participants were invited to attend a test session at a set time and

place. In line with best practice guidelines (Kirschbaum et al., 1993; Smyth et al., 2013) testing commenced in the afternoon between 13:00 and 15:00 hr, to control for changes in basal cortisol secretion in the morning and following the post-prandial period. Participants were asked to refrain from food, caffeine, alcohol, exercise and smoking 30 minutes prior to the research session. The TSST-G (von Dawans et al., 2011) included 3 main phases: the group preparatory period (30 min); the group stress task period (22 min); and a group resting and debriefing period (40 min). During the preparatory period, groups of up to 6 participants met in Room 1 where they were informally seated around a single table and introduced to the experimenter, they were free to talk to each other at this time.

Following informed written consent, participants completed in silence demographic questions, the date of their last menstruation and the VASQ, if they had not already completed it on-line (it had been available since the beginning of the recruitment period). Each participant then received a large sticker with a number between 1 and 6. They were informed that they would be identified with this number during the task period and that the numbers would be called in a random order. Participants were then introduced to the saliva-sampling method. Following this participants were given 10-minute quiet time to prepare notes for a mock job interview. They were asked to prepare a free 2-minute speech as if applying for a job of their choice and to introduce themselves to the committee. They were asked to convince the committee that they were the most suitable candidates for the position. After this preparatory period the baseline saliva sample was collected immediately prior to leaving Room 1. Participants were taken into Room 2 (a short distance away) and instructed to stand in a straight line in front of the already seated committee, comprising one woman and one man. The committee were wearing white laboratory coats and there were two conspicuous video cameras pointing at the participants. A committee member called the number of each participant in turn in a random order to make a 2-minute speech as if applying for a job. After all participants gave their speech (a total of up to 12 minutes), the

committee asked the participants, in the same order, to serially subtract the number 17 from a given number (e.g. 4878) as fast and accurately as possible for 80 seconds. Each participant received an individual starting number to avoid learning effects. Standard responses from the committee were followed where participants ended their speech before the 2-minute duration (e.g. 'you still have time, please continue') or failed in the subtraction task (e.g. 'you made a mistake please start again from the number ...') (von Dawans et al., 2011). Immediately after all participants had completed the TSST-G, participants were returned to Room 1, where they collected saliva samples every 10 minutes up to 40 minutes following the TSST-G period. During this time they were debriefed.

Saliva Sampling Collection

Cortisol was measured in saliva samples collected using Salivettes (saliva sampling devices, Sarstedt Ltd., Leicester, England) at baseline (immediately before the TSST-G: S1, at 0 min) immediately after the public speaking task (S2, at 12 min), after the mental arithmetic task (S3, at 22 min), and every 10 min up to 60 min (S4, at 32 min S5, at 42 min, S6, at 52 min, and S7, at 62 min). This cortisol profile allowed us to capture the rise in cortisol, the cortisol peak, and the decline of cortisol (i.e. the recovery period) (Dickerson et al., 2004; Smyth et al., 2013). Saliva samples were frozen at -20°C until assayed at the University of Westminster. Samples were thawed and centrifuged for 10 minutes at 3,500 rpm. Cortisol concentrations were determined by enzyme linked immuno-sorbent assay developed by Salimetrics LLC (USA). The standard range in the assay was 0.33–82.77 nmol/l. Intra and inter-assay variations were both below 10%.

Statistical analysis

Cortisol data were moderately skewed and therefore a square root transformation was applied which normalised distributions, although cortisol concentrations shown in figures are representative of original units. Descriptive statistics were explored for each cortisol sample measured throughout the TSST-G procedure, and a one-way within-subjects analysis of variance was conducted to examine differences in cortisol over time. Within subjects contrasts were used to assess the pattern of cortisol secretion.

As participants performed the TSST-G tasks at slightly different time-points, cortisol reactivity was computed for each individual as their peak sample minus baseline. Cortisol recovery was computed as individual peak sample minus sample 7 (recovery). Pearson's correlation coefficients were used to examine relationships between these cortisol indices, VASQ attachment measures and demographics variables. Significant relationships between cortisol and attachment measures were examined in a multiple regression analysis controlling for variables known to affect cortisol stress reactivity. Participants were categorised according to the VASQ attachment style: secure (n=20), insecure anxious (n=37) and insecure avoidant (n=21). A one-way between-subjects analysis of variance explored group differences in cortisol stress reactivity and Bonferroni post hoc tests were applied. Chi-square was used to examine the association between participants' peak cortisol time and attachment style group.

Results

Results indicated that the TSST-G induced an overall cortisol response in this sample ($F_{(6, 462)} = 7.623, p < .001$), illustrated in Figure 1. Within subjects contrasts revealed a significant quadratic effect ($F_{(1, 77)} = 23.807, p < .001$), such that on average cortisol increased from

baseline peaked at the fourth sampling point (10 min after the completion of the TSST-G) and subsequently declined.

Insert Figure 1 here

Relationships between cortisol data and attachment variables measured by the VASQ were examined using focused composite cortisol indices: individual peak sample minus baseline (cortisol reactivity) and individual peak sample minus sample 7 (recovery). Descriptive statistic and intercorrelations for all variables are presented in Table 1.

Insert table 1 here

There was a significant positive relationship between cortisol reactivity and VASQ vulnerability score ($r = .289, p = .010$) in that participants with a higher level of vulnerable attachment exhibited a greater increase in cortisol from baseline to peak value. In other terms, participants who demonstrated an insecure anxious attachment style (those scoring highly on both VASQ subscales) displayed greater cortisol reactivity. With regards to the dimensions of the VASQ, insecurity was significantly positively correlated with cortisol reactivity, whereas proximity was not. There were no relationships between attachment measures and cortisol recovery. There were also no relationships between cortisol measures and pertinent demographic characteristics, apart from age, which was positively related with both cortisol reactivity and recovery. In terms of VASQ attachment measures, vulnerability was unrelated to demographic variables, however, insecurity was positively related with age, and proximity was positively correlated with smoking status.

A multiple regression analysis was conducted to examine whether the relationship between cortisol reactivity and vulnerable attachment remained significant when variables known to

affect cortisol reactivity were included in the model (Table 2). Vulnerable attachment and age remained significant independent predictors of cortisol reactivity.

Insert Table 2 here

The VASQ can be used to group participants according to secure, anxious or avoidant attachment styles. The insecure anxious attachment style category is derived from high scores on both insecure and proximity-seeking measures. In a supplementary analysis a one-way between subjects ANOVA was performed to examine the difference in cortisol reactivity between the three groups. There was a significant effect of attachment style group on cortisol reactivity ($F_{(2,75)} = 5.300, p = .007$), see Figure 2. Bonferroni post hoc tests indicated that the insecure anxious group was significantly different from the secure group ($p = .011$). There was no association between when participants peaked and their attachment style group ($X^2 = 16.405, p = .173$).

Insert Figure 2 here

Discussion

Vulnerable attachment, determined by the VASQ, predicted greater cortisol reactivity to a group psychosocial stressor independent of age, smoking status, menstrual phase and BMI. Whilst there were no differences in the timing of the cortisol peak, supplementary analysis revealed that participants with an insecure anxious attachment style (a combination of high scores on the insecurity and proximity seeking sub-scales of the VASQ) showed greater stress-induced cortisol reactivity than participants with secure attachment style. Individuals with avoidant attachment style (high scores for insecurity and low scores for proximity seeking) did not differ from the secure attachment style group in terms of their cortisol

reactivity. Attachment style was not associated with the post-stress recovery in cortisol concentrations. These data provide evidence for an association between attachment style and increased reactivity of the HPA axis in response to a standardised group psychosocial stressor in healthy young female participants.

The results are consistent with the work of Quirin et al. (2008), which also showed that anxious attachment style predicted greater cortisol responding in females with no effect for avoidant attachment. However, the results are in contrast with other work showing that avoidant (but not anxious) attachment predicted enhanced stress-induced cortisol responding in females (Powers et al., 2006). The results are also inconsistent with other studies showing no relationship between attachment style and cortisol reactivity (Ditzen et al., 2008; Smeets, 2010).

These discrepancies in findings may in part be attributable to the choice of stressor. We chose to use an adapted form of the TSST for use with groups. The TSST is a reliable activator of HPA axis function comprising the key elements of uncontrollability and socio-evaluative threat (Dickerson et al., 2004). It is not possible to compare the size of the cortisol response described here to those that would be elicited by the individual TSST; hence whether the group nature of the stressor represented a particularly potent stimulus for this group is undecided. It would be interesting to repeat this study using the individual TSST to explore further this possibility. However, it is noteworthy that the modifications to the TSST-G employed here (i.e. free group interactions in the preparatory phase and open visibility during the stressor) produced a statistically significant cortisol stress reactivity response. This provides opportunities for exploration of other social interventions in stress responding such as reported by Häusser et al. (2012) in terms of group social identity.

Another distinguishing feature of this study was the use of the VASQ for the assessment of attachment style. We chose to use this self-report tool as it has been shown to perform somewhat better in predicting depression than other self-report measures of attachment (Bifulco et al., 2003), as well as predicting negative psychosocial well-being and mental health in university students (Carr et al., 2013). The study has several other strengths in that self-reported menstrual phase, age, smoking and BMI were all accounted for in the modelling of cortisol reactivity. The study also controlled for time of day and collected multiple saliva samples at 10 minute intervals for more than 60 minutes, providing a full neuroendocrine response profile, enabling accurate examination of individual cortisol reactivity and recovery.

The findings may reflect subtle underlying changes in HPA axis function linked to attachment style that are important for future mental health and well-being. For example an enhanced cortisol response to the TSST has been shown to predict depressive symptoms in young adults (Morris et al., 2012) and suicidal ideation in female adolescents with a history of mental health concerns (Giletta et al., 2014). The results are also consistent with evidence showing greater cortisol reactivity to the TSST in older adults subjected to separation from both parents during childhood (Pesonen et al., 2010) and in young adults exposed to severe pre-natal stress (Entringer et al., 2009).

The findings may also inform basal HPA axis function as it has been shown that the cortisol response to laboratory stress is positively associated with average cortisol concentrations over the day (Kidd et al., 2014). This may provide an eventual route to allostatic overload and negative physical and mental health outcomes (McEwen, 2000; Morris et al., 2012). It may also underpin observed aberrant diurnal profiles of cortisol secretion in anxious attachment style (Oskis et al., 2011; Quirin et al., 2008). The study also provided supportive evidence that avoidant insecure attachment style is somewhat similar to secure attachment

in terms of neuroendocrine function, consistent with evidence concerning the cortisol awakening response (Oskis et al., 2011), as well as the findings that there are lower health risks in insecure avoidant individuals compared to those anxiously attached (Bifulco et al., 2002a; Fraley et al., 2004; Sbarra et al., 2013).

The current results are limited to healthy young females so it would be interesting to repeat the study in healthy young males and with different age ranges. Another limitation is the reliance upon self-reported menstrual phase, not hormonal assessment. Also the cross-sectional design, with no long-term follow-up in relation to health outcomes, means we are unable to draw any conclusions about whether the observed results are implicated in future health outcomes.

Conclusions

In conclusion the study used an adapted version of the newly developed TSST-G to explore the impact of attachment style on acute stress responding within a group setting. Healthy young females with anxious attachment style showed a more marked cortisol response to the stressor. Results obtained were not related to age, self-reported menstrual phase, smoking status or BMI. The results indicate that differences in HPA axis activation may provide a pre-clinical indication of ill-health vulnerability.

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Declaration of Interest

The authors report no conflicts of interest

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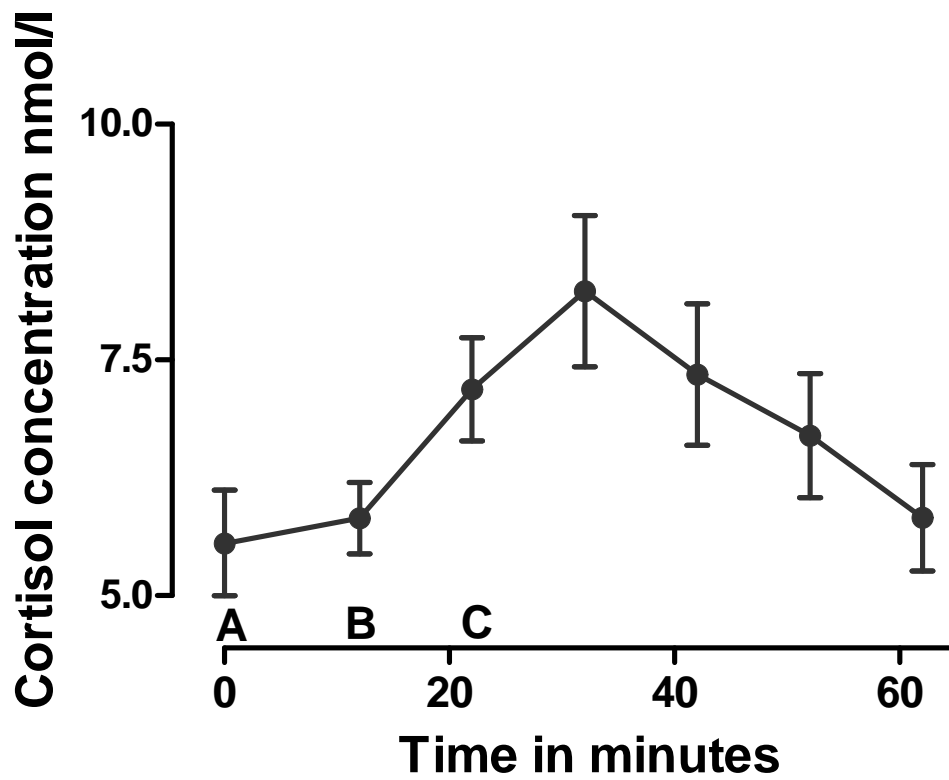


Figure 1 Mean (\pm S.E.M.) salivary free cortisol concentrations (nmol/l) for all participants (N = 78). A: immediately before onset of the TSST-G; B mid-way through the TSST-G; C immediately after the end of the TSST-G.

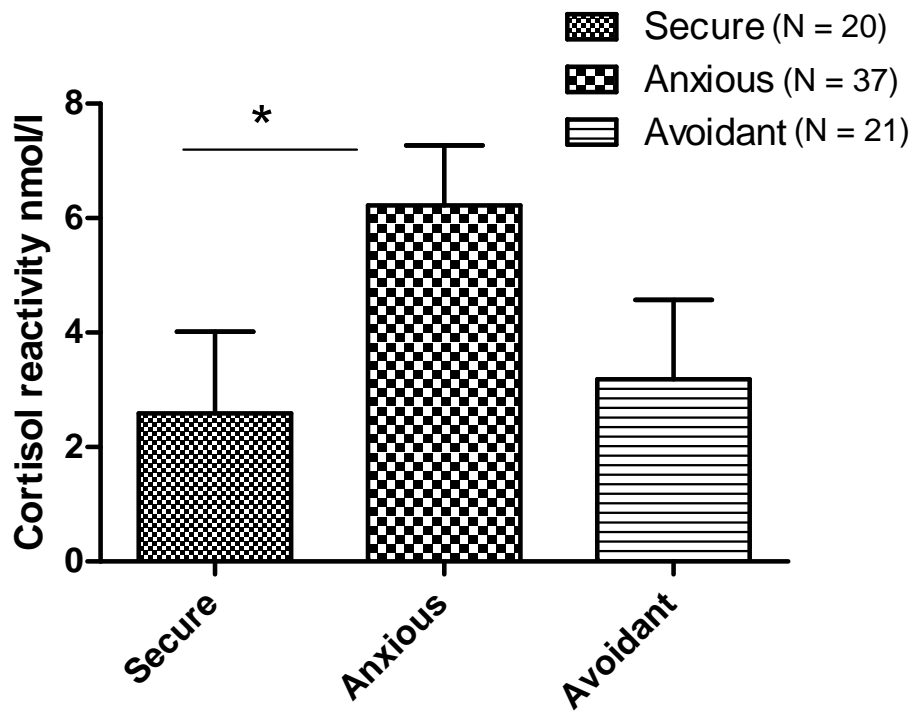


Figure 2 Attachment style differences in cortisol reactivity. Reactivity was significantly greater for the insecure anxious attachment style group in comparison to the securely attached group ($p = 0.011$).

Table 1 Descriptive statistics and intercorrelations between cortisol, VASQ attachment measures and demographic data (N=78)

Variables	Descriptives	Correlations							
		Cortisol recovery	Vulnerability	Insecurity	Proximity	Age	Menstrual cycle phase	Smoking status	BMI
Cortisol reactivity M (SD)	4.47 (6.50)	.185	.289*	.269*	.177	.349**	-.141	-.103	.011
Cortisol recovery M (SD)	4.20 (4.50)		-.020	-.047	.016	.276*	-.176	.015	.017
VASQ Vulnerability M (SD)	60.98 (9.81)			.775**	.770**	.209	-.037	.163	-.119
VASQ Insecurity M (SD)	33.37 (6.39)				.193	.247*	-.051	.001	-.035
VASQ Proximity M (SD)	27.61 (6.32)					-.075	-.006	.254*	-.150
Age M (SD)	20.22 (3.21)						-.054	.099	.218
Menstrual cycle phase % luteal	38.5							-.017	-.007
Smoking status % non-smoker	85.9								-.111
BMI M (SD)	21.46 (3.76)								

* $p < .05$, ** $p < .001$

Table 2 Prediction of cortisol reactivity

Predictors	beta	<i>t</i>	<i>p</i>
VASQ Vulnerability	.241	2.205	.031
Age	.322	2.902	.005
Menstrual cycle phase	-.118	-1.132	.261
Smoking status	-.182	-1.706	.092
BMI	-.051	-.468	.641
<i>R</i> ²	.162 (<i>p</i> = 0.003)		