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BRIEF REPORT

Oxytocin and cortisol in romantically unattached young adults: Associations with bonding and psychological distress

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Abstract

Despite extensive research on the involvement of oxytocin (OT) in mammalian bonding, less is known about its role in human social affiliation across the life cycle. Forty-five romantically unattached young adults participated. Plasma oxytocin and salivary cortisol were assessed using enzyme immuno-assay, and self-report measures of bonding, attachment, anxiety, and depression were collected. Oxytocin was associated with bonding to own parents and inversely related to psychological distress, particularly depressive symptoms. Cortisol was related to attachment anxiety. Regression analysis indicated that the adult's representations of bonding to parents predicted OT levels above and beyond cortisol, psychological distress, and attachment. Findings are consistent with antistress models of oxytocin and suggest that oxytocin may play a role in bonding-related cognitions across the life span.

Descriptors: Oxytocin, Cortisol, Depression, Anxiety, Bonding

Although extensive research has implicated the nanopeptide oxytocin (OT) in mammalian bonding (Carter, 1998; Insel & Young, 2001), less is known about the specific role of OT in processes of human affiliation. Recent findings indicate that OT is involved in a range of socially related thoughts and behaviors in human adults, including trust (Kosfeld, Heinrichs, Zak, Fischbacher, & Fehr, 2005), partner support (Grewen, Girdler, Amico, & Light, 2005), and the ability to read the mental states of others (Domes, Heinrichs, Michel, Berger, & Herpertz, 2007). Similarly, new studies assessing OT across pregnancy and the postpartum have indicated that maternal oxytocin levels during the first trimester of pregnancy predicted bonding-related thoughts and interactive behaviors to both the fetus during late pregnancy and the newborn infant in the first postpartum month (Feldman, Weller, Zagoory-Sharon, & Levine, 2007; Levine, Zagoory-Sharon, Feldman, & Weller, 2007).

Several authors suggest that oxytocin has an overall antistress effect and that it functions to attenuate the stress response (Carter, 1998; Uvnas-Moberg, 1998; Heinrichs & Gaab, 2007). Support for this hypothesis is derived from animal studies pointing to the overall calming and growth-promoting effects of oxytocin (Uvnas-Moberg, 1998). Studies in humans, however, are less consistent. Few studies showed positive associations between OT and cortisol—a biomarker of the stress response—as well as with measures of stress and anxiety (Marazziti et al., 2006; Taylor et al., 2006), whereas others report negative correlations between OT and physiological and behavioral indicators of stress (Altemus, Deuster, Galliven, Carter, & Gold, 1995; Heinrichs & Gaab, 2007; Meinlschmidt & Heim, 2007). As such, the relationship between OT and cortisol in humans and the effects of psychological distress on the OT system are not yet fully understood.

Studies assessing plasma OT in human adults have typically examined OT in groups of romantically attached individuals or in heterogeneous groups with regards to current romantic relationships (e.g., Tops, van Peer, Korf, Wijers, & Tucker, 2007). As a result, it is not fully clear whether the functioning of the OT system is limited to periods of bond formation or whether it has an anti-stress effect and is associated with bonding-related processes across the life cycle, regardless of current relational status.

In light of the above, the goal of this study was to examine plasma OT levels in a homogenous group of young adults who are not currently in a romantic relationship. OT was examined in relation to cortisol, measures of psychological distress, measures of attachment, and the individual's representations of bonding to his or her own parents. We expected OT to be positively related

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to measures of attachment and bonding and negatively related to psychological distress.

Method

Participants

Forty-eight healthy university students not involved in a romantic relationship participated. Due to technical reasons, the final analysis included 45 participants (21 men and 24 women). Mean age was 24.63 years ($SD = 3.16$ years).

Procedure

Participants were recruited by ads posted around campus. Participants arrived at the laboratory between 4:00 p.m. and 6:00 p.m. These hours were chosen in light of previous research, which suggests stability in the diurnal cycle of plasma OT in the afternoon and evening hours (Forsling, Montgomery, Halpin, Windle, & Treacher, 1998). Participants' were asked to refrain from eating or drinking (other than water) for 30 min prior to arrival. Blood and saliva samples were collected by a registered nurse and then participants completed a set of self-report measures. A subgroup of 18 participants (10 men and 8 women) from the main group was instructed to wait for 20 min after the first blood sampling in an isolated room with a calm and relaxing atmosphere, after which a second blood sampling was conducted in order to ascertain OT stability in plasma during the experiment's duration. This subgroup comprised of the first 18 participants in the study and did not differ on any demographic or test variables from the following participants. Participants received approximately $13 for their participation. The study was approved by the University's Ethics Committee and all subjects signed an informed consent form.

Self-Report Measures

The State Trait Anxiety Inventory (STAI). The STAI (Speilberger, Gorsuch, & Lushene, 1970) is a well-validated 40-item scale consisting of two separate scales (20 items each) to measure stable individual differences in anxiety proneness (trait) and current (state) anxiety.

The Beck Depression Inventory (BDI). The BDI (Beck, 1978) was used to measure subjects' depressive symptoms. It is a widely used 21-item instrument that measures the level of depressive symptoms on a 3-point scale and demonstrates good reliability (Beck, Steer, & Garbin, 1988).

Adult Attachment styles. The Adult Attachment styles (Brennan, Clark, & Shaver, 1998) defines two dimensions of adult attachment: attachment-related anxiety and attachment-related avoidance. Each dimension is measured with a reliable and valid 18-item Likert scale, in which higher scores imply higher attachment anxiety or attachment avoidance (Shaver & Mikulincer, 2002).

The Parental Bonding Instrument (PBI). The PBI (Parker, Tulping, & Brown, 1979) includes 25 questions for mother and father each. Participants rate their relationships with each parent. Two factors are derived for each parent: parental care and parental overprotection. The instrument has shown acceptable internal consistency and reliability (Wilhelm & Parker, 1990).

Hormone Collection and Analysis

Oxytocin. Blood was sampled between 4:30 p.m. and 6:30 p.m. by a single draw of either 4 ml or 9 ml of blood from antecubital veins. Blood was drawn into chilled vacutainer tubes containing lithium heparin that were supplemented with 400 KIU of Trasylol (Bayer, Germany) per 1 ml blood. OT samples were kept ice-chilled for up to an hour before being centrifuged at 4°C at $1000 \times g$ for 15 min. Supernatants were collected and stored at -70°C until assayed. Determination of OT was performed using a commercial OT ELISA kit (Assay Design, Ann Arbor, MI) as described previously (Carter et al., 2007; Feldman et al., 2007; Levine et al., 2007). Measurements were performed in duplicate and the concentrations of samples were calculated by using MatLab-7 according to relevant standard curves. The intra-assay and interassay coefficient are less than 12.4% and 14.5%, respectively.

Cortisol (free cortisol). Saliva for cortisol analysis was sampled during blood sampling. Participants were asked to place a roll of cotton in their mouths, chew on it for a minute until it became saturated, and place it in a Salivette (Sarstedt, Rommelsdorft, Germany). Salivettes were kept ice-chilled for up to 2 h before being centrifuged at 4°C at $1000 \times g$ for 15 min. The samples were stored at -20°C until assayed. Cortisol levels were assayed using a commercial ELISA kit (Assay Design). Measurements were performed in duplicate, according to the kit's instructions. Cortisol levels were calculated by using MatLab-7 according to relevant standard curves. The intra-assay and interassay coefficients are less than 10.5% and 13.4%, respectively.

Data Reduction

To reduce the number of variables and to derive meaningful constructs that can be used as a priori theoretically relevant predictors of OT, we averaged the following variables.

Psychological distress. Mean BDI ($M = 6.02$, $SD = 4.77$) and Trait anxiety ($M = 40.4$, $SD = 8.89$) scores were all below clinical cutoffs and appropriate for a sample of healthy adults. Depression and anxiety scores were interrelated, $r = .38$, $p < .01$, and the two scores were averaged into a Psychological Distress construct.

Bonding. Mother Care and Father Care scores from the PBI, reflecting the caring internal representation of the adult of his or her own parents, were interrelated, $r = .42$, $p < .01$, and the two scores were averaged into a Parental Bonding construct.

Results

Hormones

Mean OT level was 258.76 pg/ml ($SEM = 38.41$), median OT level was 180 pg/ml, minimum OT level was 114.1 pg/ml, and maximum level was 1255.03 pg/ml. These levels are similar to earlier reports of OT levels in adults analyzed by ELISA (Feldman et al., 2007; Levine et al., 2007). The 18 participants whose blood was drawn twice during the experiment showed a high level of individual stability in OT levels. Pearson's product-moment correlation between the first OT level and the second was $r = .93$, $p < .001$. This affirms stability of OT in plasma during the experiment's time window. A similar intrasubject stability in plasma OT has been demonstrated in the past in nonpulsatile states (Levine et al., 2007). OT levels were not associated with gender,

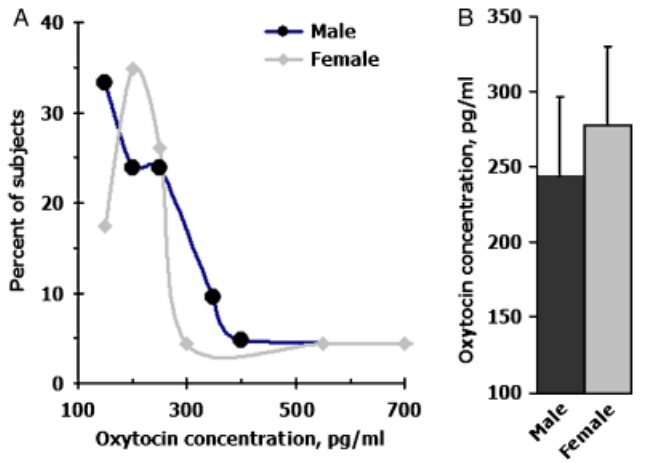

Figure 1. a: The x axis shows groups for every 50 pg/ml range, presented as the highest value of each range. The y axis shows the number of subjects in each range group, represented as the percentage of total participants (female $N = 23$, male $N = 21$). The highest OT values were excluded (male OT = 1255, female OT = 1192). b: OT values averaged according to gender, the error represented as *SEM*. Gender differences were not found to be significant.

body weight, or height. Mean cortisol level was 2.61 pg/ml ($SEM = 0.27$). A one-way ANOVA revealed that cortisol was significantly higher in men ($M = 3.32$, $SEM = 0.28$) than in women ($M = 2$, $SEM = 0.26$), $F(1,43) = 9.05$, $p < .01$. Nine participants of the sample indicated they had engaged in sexual activity during the week prior to the experiment, but this was not found to be associated with OT or cortisol levels. Of the 24 women participating in the experiment, 4 indicated that they were taking some sort of contraceptives. Levels of OT and cortisol in these individuals did not differ from women not taking birth control medication. Figure 1 presents scatter plots for OT in men and women. OT values were log-transformed prior to data analysis.

Pearson Correlation Analyses

These revealed that plasma OT was negatively correlated with depression, $r = -.38$, $p < .01$, but not with anxiety, $r = .09$, $p > .10$. OT was also negatively associated with the construct of Psychological Distress, $r = -.35$, $p < .05$. OT was positively related to Parental Bonding, $r = .39$, $p < .01$ (Mother Care, $r = .42$, $p < .01$; Father Care, $r = .36$, $p < .05$). Cortisol levels were related to Attachment Anxiety, $r = .37$, $p < .05$, but not to Attachment Avoidance, $r = .11$, $p > .10$. Psychological distress was negatively related to Bonding, $r = -.38$, $p < .01$. The correlation between cortisol and OT was $r = -.22$, $p > .10$.

Hierarchical Regression Analysis

Hierarchical regression analysis was preformed to predict plasma OT levels. Variables were entered in five blocks. In the first block cortisol was entered, in the second block, Psychological Distress (average of depression and anxiety), in the third, Attachment Anxiety was entered, and in the fourth, Attachment Avoidance. Parental Bonding (average of Mother Care and Father Care), was entered in the final block. Results (Table 1) showed that the model as a whole was significant. Unique predictors of OT were Psychological Distress (negatively), and Bonding (positively). Overall, the variables used here explained 29% of the variance in plasma OT.

Discussion

Results of this study demonstrate associations between the OT system and measures of bonding and psychological distress in adults who are not currently in romantic relationships. These data point to relationships between the functioning of the OT system and bonding-related cognitions, regardless of the individual's present attachment status. As such, the links between OT and measures of both psychological distress and bonding to one's own parents may suggest that one mechanism by which OT buffers stress and depression across the lifespan is through its association with the attachment system.

The associations found here between OT and bonding are consistent with the few existing studies on the relations of OT with the parent–infant bond assessed both concurrently and retrospectively. Meinlschmidt and Heim (2007) demonstrated that intranasal administration of oxytocin in adult men with early parental separation did not decrease the level of cortisol as compared to controls. Levine and colleagues (2007) found associations between an increase in maternal plasma oxytocin from the first to the third trimester of pregnancy and the mother's bonding to her fetus, and Feldman et al. (2007) showed links between oxytocin across pregnancy and the postpartum with the mother's bonding-related thoughts and behaviors toward her infant. The present findings may suggest that the adult's representations of the experience of being cared for as a child, which are likely shaped in the first years of life, are related to the OT system in adulthood. Because the OT system functions as a feedback loop (Uvnas-Moberg, 1998), bonding-related representations and OT may function in a mutually influencing manner throughout development, with more positive representations of parental care related to increased oxytocin levels. However, due to the fact that our data are correlational, it is not possible to infer causality but only to demonstrate associations between OT levels and bonding-related representations.

Table 1. *Hierarchical Multiple Regression Predicting Plasma OT levels*

Predictors	Beta	*R*	R^2 Change	*F* Change	*df*
Cortisol	– .22	.15	.02	1.04	1,41
Psychological Distress[a]	– .29*	.41	.14	6.88*	2,40
Attachment–Anxiety	.09	.41	.00	.20	3,39
Attachment–Avoidance	.00	.42	.01	.30	4,38
Parental Bonding[b]	.38*	.54	.12	5.84*	5,37

Note: R^2 total = .29: $F(5,37) = 3.03$, $p < .05$.
[a]Average of depression (BDI) and trait anxiety (STAI) scores.
[b]Average of Mother Care and Father Care (PBI).
*$p < .05$.

The negative correlation between OT and the averaged measure of anxiety and depression is consistent with theories on the role of OT as an anti-stress hormone. According to this approach, OT functions mainly by increasing calm states, reducing stress and negative mood, and facilitating approach behaviors that are critical to bond formation. The findings that OT is specifically related to depression are consistent with previous models and require further research in human adults (Carter, 1998; Uvnas-Moberg, 1998).

Bonding and distress were independently related to OT, suggesting that these measures possibly tap two separate roles of the OT system: the anti-stress function and attachment-building function. This view is consistent with Tops and colleagues (2007), who showed independent contribution of stress and attachment variables to the prediction of OT in adults. The two separate functions of OT in relation to stress and affiliation may provide a partial explanation to the inconsistent findings on the links between OT and measures of stress.

A possible limitation of this study is the fact that OT was sampled in the periphery and not centrally. Although the literature on the relations between central and peripheral OT is incomplete, animal research point to concordance between OT levels in the brain and those in the periphery (Carter et al., 2007; Wotjak et al., 1998), and human research, including the present study, indicates that peripheral OT is associated with bonding-related representations (Levine et al., 2007; Tops et al., 2007).

Future research is required to assess the relations of oxytocin and a range of behavioral measures and physiological systems and to specify the brain systems implicated in the regulation of OT. Further study of the links between oxytocin, measures of bonding, and various indices of distress may provide a more comprehensive understanding of the oxytocin system and its role in the formation and maintenance of social bonds across the lifespan.

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