Acknowledgments

In 2014 I joined the research project "Oxidative stress, hormones and relationships", a collaborative project between NTNU and the University of New Mexico (UNM) in Albuquerque, USA. The project introduced me to the exciting field of social neuroendocrinology. I became responsible for the recruitment and data collection of biological samples and questionnaires. Getting to know all the work behind an empirical study has been fascinating and informative, but admittedly also frightening. Being in a position where I had to explain to the customs and border protection why I had sent, what they classified as a possible bio-bomb of Norwegian urine and saliva (dry ice within a closed container is apparently gas under pressure) to USA, was to say the least, scary.

For every challenge that arose, professor and **super**visor Leif Edward Ottesen Kennair, was available to give advice. Your door has always been open for academic support, but also to consolation and continuing enthusiasm boosts. Thank you so much! I also want to thank my friends and colleagues, professor Steven Gangestad, dr. Nicholas Grebe and dr. Trond Viggo Grøntvedt for letting me be a part of your research group and for all your feedback and support. I look forward to a continuing collaboration.

Lastly I want to thank my family and friends for support and proofreading numerous drafts to this paper.

**Title page picture by Grete Wolden (2014). The author of this paper looking for distilled biochemical love.*

Abstract

This study assesses how the neuropeptide oxytocin (OT) is distributed in a Norwegian sample and its relationship with adult romantic attachment orientations. Previous research on associations between OT and adult romantic attachment have been equivocal and theoretical models appear openly conflicting. By combining prior empirical research and a novel hypothesis (Identify and Invest), it's attempted to reconcile diverging data and models to gain a more complete understanding of the oxytocinergic system. OT secretion is here suggested to vary in response to romantic attachment orientations and allocate psychological resources, such as attention, reward sensitivity, or interpretation of cues in the romantic relationship. A sample of 148 romantically involved individuals was analyzed using two different sampling procedures (an OT baseline measure and a controlled lab procedure to measure OT change). Regression analyses suggest that secure romantic attachment orientation is positively associated with OT, and avoidant romantic attachment orientation is negatively associated with OT. This effect was only evident in the total and in the female sample. The OT baseline distributions are positively skewed, as expected from previous OT research on young adults. Surprisingly, the OT baseline and OT change were correlated negatively, which indicate that the oxytocinergic system differs in momentarily reactions and more permanent features. Overall, the results in this paper imply that OT is implicated in regulating important processes of romantic attachment.

Keywords: oxytocin, baseline level, pre-post change, adult romantic attachment, neuroendocrinology

Introduction

Romantic relationships are some of the most influential forces of human behavior and have a profound effect on both culture and civilization (Young & Wang, 2004). The process of developing, maintaining and dissolving romantic bonds within close relationships are associated with a range of physical and mental health outcomes (Braithwaite, Delevi, & Fincham, 2010; Cotten, 1999). The study of these processes also provides valuable insight into different mating and reproduction strategies (Carter, 2014; Crespi, 2016). In the academic literature there is a growing interest in neuromodulatory effects on social behaviors. This has generated a large body of research focusing on what functions hormones might have in close social relationships and attachment representations (Gangestad & Grebe, 2017; Lee, Macbeth, Pagani, & Young, 2009). Discovery of the physiological and anatomical characteristics of the hormone oxytocin (OT) has resulted in an immense interest from different scientific fields with its effect on physiology, emotions, cognitions and behavior (Crespi, 2016; den Hertog, de Groot, & van Dongen, 2001).

Characteristics of the oxytocinergic system

OT is a neuropeptide hormone that is primarily synthesized in the hypothalamus and secreted in both the central nervous system (CNS) and into the peripheral circulation (Carter, 2014; Knobloch & Grinevich, 2014). The name oxytocin derives from Greek root words for “quick birth”, coining its peripheral functions in obstetrics, such as uterine contractions and stimulating the milk letdown reflex during lactation (den Hertog et al., 2001; Lee et al., 2009). OT is transported to the posterior pituitary gland where it is released into the peripheral blood stream to influence several physiological functions concerning smooth muscle contractions for both men and women (Lee et al., 2009). OT is enabled within the central nervous system by nuclei of the hypothalamus that secretes through neural tissue into central dendrites. These molecules project along axons to different brain regions binding to OT receptors (OTR) that influences intracellular cascades (Johnson & Buisman-Pijlman, 2016; Lee et al., 2009; Strathearn, 2011). OT contains distinctive neural properties that allow secretion from neural soma, axons and dendrites, thus broadly affecting the central nervous system (Carter, 2014). From anatomical and pharmacological research, OT has been shown to stimulate brain regions pertaining to

social behaviors and reward related pathways, such as the mesolimbic-dopamine system (Johnson & Buisman-Pijlman, 2016; Strathearn, 2011; Young & Wang, 2004)

In popular science OT has been called the "love" or "bonding" hormone for its neuromodulatory effects (Gangestad & Grebe, 2017; Shen, 2015). The "love/bonding" reputation likely originates from the discovery of OT as a key mediator in mother-infant relationships, such as maternal caregiving and infant attachment, across mammalian species (Galbally, Lewis, Van Ijzendoorn, & Permezel, 2011; Gangestad & Grebe, 2017; Panksepp, Nelson, & Bekkedal, 1997). These neuromodulatory properties of the oxytocinergic system are suggested to have evolved from its ancestral peripheral functions to maternal behavior and then co-opted into different social contexts like social bonds and attachments (Crespi, 2016; Gangestad & Grebe, 2017; Lee et al., 2009). Substantial research across mammalian species has examined OT as a neuroendocrine mediator in caregiving behavior and attachment processes (Carter, 2014; Crockford, Deschner, Ziegler, & Wittig, 2014).

OT and the existing scientific research

Landmark studies on OT from animal research have focused on prairie (*Microtus ochrogaster*) and montane (*Microtus montanus*) voles and their mating systems (Insel & Shapiro, 1992; Young & Wang, 2004). These sister groups from the same genus display remarkable distinction in social organization. The prairie vole display enduring monogamous relationships with their mating partners and are highly involved in parental care; the montane vole, in contrast, exhibits a polygynous mating strategy and little parental care (Insel & Shapiro, 1992; Young & Wang, 2004). Neuroanatomical studies have shown contrasting distributions of OT receptor (OTR) density in brain regions between these voles (Insel & Shapiro, 1992). Supporting OT's function in attachment processes, experimental administration of OT in adult prairie voles accelerated pair bonding and infusion of OT antagonists prevented the formation of pair bonding and selective sexual preferences (Carter, Devries, & Getz, 1995; Williams, Insel, Harbaugh, & Carter, 1994). These findings regarding OT's neuromodulatory effects are supported as well in other animal studies, e.g. mediating maternal behavior in sheep, and influencing huddling and proximity seeking in frequency in black-tufted marmosets (Kendrick et al., 1997; Smith, Agmo, Birnie, & French, 2010). Animal studies have thus strengthened OT's role as a mediator in formation and maintenance in pair bonding and affiliate behavior. Research from

these animal studies suggests additionally a considerable sex difference of the oxytocinergic system (Lee et al., 2009). From landmark studies on rodents, females have in general higher OT secretion and OTR density in the brain than males (Carter, 2014; Young & Wang, 2004). Francis, Young, Meaney, and Insel (2002) furthermore revealed a possible epigenetic sex-difference of OTR expression from variations in maternal care. They reported that females rats (*Long-Evans breed*) which received high levels of maternal care as pups had increased OTR binding as adults, but not in the male rat group that received high maternal care. This difference suggests that variations of social behavior between sexes can affect the development and function of the oxytocinergic system (Carter, 2014; Johnson & Buisman-Pijlman, 2016; Young & Wang, 2004).

Considerable work has also been done on human subjects, both in experimental OT administration and endogenous OT studies. There is some support that OT in human females performs a greater number of functions regarding affiliate behavior than in men (Carter, 2014; Kendrick, 2000; Lee et al., 2009; Weisman, Zagoory-Sharon, Schneiderman, Gordon, & Feldman, 2013). However, these associations are not as conclusive as the animal OT research (Carter, 2014).

Administration studies in humans have found that intranasal OT can increase positive couple interaction in a conflict compared to a placebo administration (Ditzen et al., 2009), improve memory for faces (Guastella, Mitchell, & Dadds, 2008) and ability to interpret social information (Domes, Heinrichs, Michel, Berger, & Herpertz, 2007). It has also shown to increase empathy and trusting behavior in different experimental paradigms (Baumgartner, Heinrichs, Vonlanthen, Fischbacher, & Fehr, 2008; Kosfeld, Heinrichs, Zak, Fischbacher, & Fehr, 2005), to reduce social anxiety symptoms (Heinrichs, Baumgartner, Kirschbaum, & Ehlert, 2003; Kirsch et al., 2005) and facilitate social support (Cardoso, Valkanas, Serravalle, & Ellenbogen, 2016). Several endogenous OT studies also supports this proposed prosocial view of OT, e.g. low levels of circulating OT have been implicated in perceiving social interactions as less rewarding (Bell, Nicholson, Mulder, Luty, & Joyce, 2006) In addition, research on both in- and outpatient samples have revealed negative correlations with depression and anxiety symptoms (Scantamburlo et al., 2007). Research related to romantic relationships has found that high relationship quality is positively correlated with circulating OT, and new romantic relationships had elevated OT levels compared to singles (Holt-Lunstad, Birmingham, & Light, 2008; Schneiderman, Zagoory-

Sharon, Leckman, & Feldman, 2012). Holt-Lunstad et al. (2008) additionally found that a caring physical touch from a spouse increases OT secretion and further decreases stress hormones and systolic blood pressure. This prosocial view of OT is not absolute and several contradictory findings have been published, termed the *OT paradox* (Bethlehem, Baron-Cohen, van Honk, Auyeung, & Bos, 2014). Such conflicting studies have shown that OT secretion in women increases in response to relational and interpersonal distress (Tabak, McCullough, Szeto, Mendez, & McCabe, 2011; Turner, Altemus, & Enos, 1999). Thus contradicts OTs proposed anxiolytic effects, as stated by Holt-Lunstad et al. (2008). Several results also propose OT to have a "dark" side in both males and females, e.g. in increasing envy and gloating, to motivate in-group favoritism and even promote ethnocentrism (De Dreu, Greer, Van Kleef, Shalvi, & Handgraaf, 2011; Shamay-Tsoory et al., 2009). Panksepp et al. (1997) found in an intriguing experiment in rats (*Long-Evans*) that OTs neuromodulatory effects could be dose-dependent. Low OT administrations to rats increased gregariousness, whereas high OT doses had opposite social outcomes, perhaps foreshadowing later research on the "dark" side of OT in human research. These findings appear to contradict interpretations of OT as a merely prosocial love hormone and give an approach to assess more complex neuromodulatory properties of OT in psychological research.

Social contexts and attachment orientations

By theorizing about which social circumstances trigger OT release and how sex and individual differences may impact the oxytocinergic system, we can achieve a better understanding of how neural resources are utilized (Gangestad & Grebe, 2017). Controlling individual and contextual variations may be of particular interest regarding the unclear literature in the field of social neuroendocrinology. Beliefs and expectations individuals have about themselves and their intimate relationships may accordingly impact the oxytocinergic system (Carter, 1998). This will further affect the allocation of psychological resources into various processes in developing, maintaining or dissolving romantic bonds (Carter, 1998; Crockford et al., 2014; Gangestad & Grebe, 2017). Beliefs and expectations that guide these processes in adult romantic relationships can be studied as adult romantic attachment orientations (Hazan & Shaver, 1987; Simpson, 1990).

The theory of attachment originally targeted maternal-infant bonds that promoted care-eliciting behavior from the infant and caregiving behavior from the parent (Feeney, 2008; Galbally et al., 2011; Simpson, Rholes, & Phillips, 1996). Individuals will according to attachment theory develop expectations about the availability and responsiveness of attachment figures which develop into internal working models that shape relational expectations about themselves and others (Feeney, 2008; Simpson, 1990). Ainsworth, Blehar, Waters, and Wall (1978) identified three principal patterns of attachment in children from these working models. These cognitive, emotional and behavioral dynamics have extended beyond maternal-infant bonds to describe related processes in adult relationships and patterns of romantic attachment. These interpersonal patterns are classified as anxious, avoidant and secure orientations (Hazan & Shaver, 1987; Simpson et al., 1996). This dimensional classification describes the general orientation individuals have toward romantic relationships (Simpson, 1990). Individuals who exhibit an anxious romantic attachment style have a strong desire to form selective social bonds, to spend considerable time nurturing and caring about the relationship, but also display a great deal of ambivalence in their relationships (Simpson, 1990). They may have a tendency to be unreasonably concerned about abandonment, loss and their partner's lack of investment in the relationship (Simpson et al., 1996). Individuals who display an avoidant romantic attachment find it difficult to trust other people completely and are nervous to form close relationships (Simpson et al., 1996). According to Simpson (1990) people with avoidant romantic attachment style score low on trust, reciprocity, commitment and satisfaction in romantic relationships. Persons with a secure romantic attachment style are characterized by a need to form close relationships and display working models that find it easy to depend and be depended on and trusting their romantic partner (Simpson, 1990).

Neuromodulatory function on attachment

Differences in romantic attachment orientations are proposed to be a part of the same biological system as the infant-caregiver bonds, and are thus considered as behavioral homologies continuing to regulate emotions and behavior in close relationships through life (Fraley & Shaver, 2000). However studies have shown that attachment orientations from infancy to adulthood display some flexibility, and Crowell, Fraley, and Shaver (1999) revealed in a systematic review the continuity of

attachment orientations are only moderately correlated. The issue of stability of individual differences may be affected by new close relationships and critical periods in development (Fraley & Shaver, 2000).

Considerable research from animal studies supports OTs neuromodulatory effects on attachment-related processes (Carter, 1998, 2014). OT's effects are displayed in both caregiver-infant and in adult pair bonding, but few studies have directly focused on the relation between OT and adult romantic attachment in humans (Carter, 1998; Insel & Young, 2001). Marazziti et al. (2006) published a study in which they found a positive correlation between OT and anxiety in adult romantic attachment, suggesting OT serves to counteract stress in the relationship in both sexes. Schneiderman et al. (2012) did not find this relationship between romantic attachment style and OT in a sample of young adults. Samuel et al. (2015) published a negative correlation between categorical insecure adult attachment in pregnant women and OT. Further analyzing Samuel et al. (2015) categorical attachment approach, 95 % of the individuals with insecure attachments had higher scores on anxiety dimensions. This suggests that higher OT is associated with attachment anxiety orientations.

Some scholars propose that there is a sex difference between OT and attachment processes (Crespi, 2016). Supporting this view, Weisman et al. (2013) published a positive correlation between attachment anxiety and OT in females. This effect did not show in the male sample. Weisman et al. (2013) suggested however an inverse tendency for males, presenting a non-significant negative correlation between attachment anxiety and OT ($r = -0.15$, $p = .05$) in the male sample. Research from neuroendocrinology has revealed that steroid hormones, such as estradiol and progesterone, can have distinctive effects on the oxytocinergic system (Quinones-Jenab et al., 1997; Rissman, 2008). In normally ovulating women (not using hormonal contraceptives), OT appears to vary in response to the menstrual cycle (Salonia et al., 2005). The follicular and peri-ovulatory phases are characterized with increased estradiol, which seem to have a potentiating effect on OT (Quinones-Jenab et al., 1997) The luteal phase on the other hand have lower levels of estradiol, but increased progesterone (Salonia et al., 2005). This is proposed to have a dampening effect on the oxytocinergic system, although the interaction is not as definite as the OT and estradiol effect (Thomas, Crowley, & Amico, 1995). Most hormonal contraceptives deliver higher dosages of synthetic estradiol than progestin (Grøntvedt, Grebe,

Kennair, & Gangestad, 2017). Consequently, using this type of contraceptive will interfere with the normal hormonal variation through the menstrual cycle with artificial higher estradiol levels. Hormonal contraceptives have shown to influence sexual desire in romantic relationships, thus can be considered to affect processes of development, maintenance and dissolution in these relationships (Grøntvedt et al., 2017).

Theoretical models

The juxtaposition of findings regarding OT has primarily been using two theoretical models that can support either model. The model "**Tend and Befriend**" predicts a positive correlation between relationship quality and OT secretion (Carter, 1998; Uvnas-Moberg, Handlin, & Petersson, 2015). The second model, "**Calm and Connect**" predicts a negative correlation between OT and relationship quality (Taylor, 2006). Where "Tend and Befriend" theorizes that OT is a modulator that facilitates gregariousness when stress or anxiety in the relationship occur, "Calm and Connect" proposes that heightened OT secretion modulates positive qualities in the relationship due to anxiolytic effects and nurturing behaviors (Carter, 1998; Taylor, 2006). These models are not necessarily mutually exclusive and may be dependent on different assessment measures of OT (e.g. baseline vs. reactivity change or natural vs. fabricated settings) and individual differences. Grebe et al. (2017) propose a new model that attempts to reconcile the diverging findings in neuroendocrine literature on OT. This model is titled "**Identify and Invest**" and argues that OT secretion is tagged to principal persons in relationships, and is contingent of the representations individuals may have about themselves and their partner (Grebe et al., 2017). These representations concern social cues of vulnerability and emotional engagement that are "identified", and then leading to "investment" in caring features of the romantic relationship (Grebe et al., 2017). OT is consequently secreted to attend special attention to the reactions and needs of a specific romantic partner. Individual differences in perceiving these features will then orient psychological resources to respond in a seeming adaptive manner to protect the relationship (Grebe et al., 2017).

Aims and hypotheses

The aims of this study are to describe the distributions of baseline circulating OT in a Norwegian sample and address previous findings between OT and romantic

attachment orientations. Despite OT's acknowledged importance, it is still not well understood how endogenous OT is related to individual differences in romantic attachment, sex, relationship length and social circumstances that naturally occur. By using two different OT sampling procedures in this study, we may resolve some of the paradoxical empirical patterns describing this field. The first procedure is a baseline saliva sample provided by two saliva samples approximately one week apart. The second procedure is a controlled lab procedure to selectively evoke an OT secretion response by thinking about the romantic relationship. The lab procedure offers an opportunity to examine natural changes of OT secretion in a specific condition, thus adding the important factor that hormonal effects are connected to the circumstances that release these molecules into the blood stream (Gangestad & Grebe, 2017). It is assumed that the OT baseline and OT change levels will covary. However, the relationship between baseline measures and change in secretion is not well assessed in the scientific literature (Crockford et al., 2014). The direction of this correlation is thus exploratory. Results from these measures may nevertheless elucidate properties regarding OT secretion and how the oxytonergic system affects romantic attachment.

In previous human research, most studies have used non-directional hypotheses to predict correlations between OT and romantic attachment orientations. This study uses Grebe et al's (2017) "Identify and Invest" hypothesis, which proposes that perceptions of involvement and vulnerability of the romantic relationship will motivate to engage in different aspects in the relationship. These motivational states are suggested to be regulated by the oxytocinergic system, which allocates bonding resources into the romantic relationship. This framework permits to form directional predictions on how OT varies in relation to adult romantic attachment orientations. In light of previous findings of the oxytocinergic system, it's expected that several factors can moderate the relationship between OT secretion orientations and attachment. Schneiderman et al. (2012) found that new romantic partners had increased OT levels. This can be explained as a social circumstance where the relationship requires special attention to the needs and reactions from the new romantic partner. From the "Identify and Invest" model this high OT levels will decrease as the relationship continues and stabilize (Grebe et al., 2017). From research on sex difference in the oxytocinergic system, a large body of research underscores that neuromodulatory effects exists in both sexes (Carter, 2014; Lee et al., 2009). However, several studies have implicated a more essential role for OT in

females concerning behavioral responses and physiological characteristics as higher OTR density and OT peripheral circulation (Carter, 2014; Keverne & Kendrick, 1992; Lee et al., 2009; Weisman et al., 2013; Young & Wang, 2004). In addition, this study will assess how hormonal contraceptives may influence the oxytocinergic system based on research on OT change within the menstrual cycle and administration studies on progesterone and estradiol (Quinones-Jenab et al., 1997; Rissman, 2008; Salonia et al., 2005).

From the mentioned findings and theoretical conceptualizations, three main predictions are formulated: **1a**) Anxious romantic attachment is positively associated with OT baseline and OT response secretion. The anxiety orientation, characterized as a continuing suspicion concerning rejection and abandonment in the relationship, is comparable to the perceptions of emotional engagement and vulnerability, as described in the "Identify and Invest" framework. **1b**) The association between females' attachment anxiety and OT will be stronger than in males. The "Identify and Invest" framework does not predict any sex differences in social function (Grebe, 2016). Nevertheless, some studies have shown a sex specific function for females and OT, such as stronger correlations between OT and relational distress measures (Feldman, 2012; Taylor, 2006; Weisman et al., 2013).

2a) Avoidant romantic attachment is negatively associated with baseline OT and OT change secretion. Individuals scoring high on the avoidance dimension in attachment find it uncomfortable being close to and trust other people. The "Identify and Invest" framework does not explicit formulate how OT influences this attachment orientation. This prediction is primarily supported from animal studies where endogenous OT is blocked and affiliative behavior clearly reduces, and from human studies regarding experience of pleasure in social interactions (Bell et al., 2006; Insel & Shapiro, 1992; Lee et al., 2009; Smith et al., 2010; Williams et al., 1994). It appears fitting as an extension of the "Identify and Invest" model that a dampening of the oxytocinergic system may accordingly regulate behavior and motivational states that lead away from proximity and dependence in the relationship. **2b)** Despite the absence of studies on sex differences in the association between attachment avoidance and OT, this study will explore if there exists a difference in OT change and baseline between males and females.

3a) Secure romantic attachment is associated with OT baseline and OT change secretion. The secure dimension is characterized by positive orientation regarding

thoughts and behavior on trust, reciprocal dependence and proximity. According to Simpson et al. (1996), secure individuals tend to score low on both the anxiety and avoidance dimensions. The nature of this prediction is exploratory as there is not been, as I know of, published theoretical conceptualizations or empirical data that can call for a specific direction on this prospective relationship. **3b)** As follows, this study will explore if OT change and baseline deviates between males and females.

In the light of the mentioned research on factors that may influence the oxytocinergic system, this study proposes that the relationship between OT and the specified attachment orientations will be moderated by hormonal contraceptives and relationship length. I assume that women using hormonal contraceptives in these samples are using the most common contraceptive, thus having relative higher levels of synthetic estradiol, which is implied to have a potentiating effect on OT (Grøntvedt et al., 2017; Quinones-Jenab et al., 1997). These women are accordingly predicted to have higher levels of OT. Regarding relationship length, it's predicted that participants in new romantic relationships will have higher OT levels than participants in longer lasting relationships. OT secretion across this dynamic period is previous been published by Schneiderman et al. (2012) and can represent a period in the relationship that demands extra attention to social cues of emotional engagement and vulnerability (Grebe et al., 2017).

Methods

Participants

A total of 148 individuals participated in this study (116 women; 33 men; mean age = 23; *SD* = 2.84). Recruitment was conducted through posters and advertising in lectures at the university campuses in Trondheim and through social media channels like faculty blogs and Facebook. Additionally, the major city newspaper in Trondheim wrote a story about the study with recruitment info. All participants reported to be in an exclusive heterosexual romantic relationship with their partner lasting at least one month (mean relationship length = 28 months, *SD* = 2.86). One participant reported relationship length to be 375 months (>31 years) at the age of 21. This impossible value was changed to the mean relationship length in the sample.

From the recruitment material, the participants were given information that they could not partake simultaneously with their romantic partner in the study or engage in any activity that could increase the pulse considerable 15 minutes before arriving the study location. This included eating, caffeine drinks and use of nicotine or dental products. Pregnant females were excluded because different stages in the pregnancy have shown to affect OT secretion and may bias the OT outcome measure (Gimpl & Fahrenholz, 2001). A proportion of 79 % of the female participants reported use of hormonal contraceptives at the time of the study.

Target sample size to this study was 200 participants to detect a moderate correlation of .30 at 85 % power. Data collection was terminated before reaching the target sample size due to the study time limit permitted by the Regional Committee for Medical and Health Research Ethics (REC) West.

All participants were compensated for the participation with a movie ticket. The procedures in the study were administered in accordance with guidelines for ethical research with human subjects and approved by REC West.

Procedure

Testing was conducted between 0800 and 1900 at Dragvoll Campus in Trondheim, Norway. The participants arrived to the study location where they were greeted by an experimenter and guided to a laboratory containing 4 semi-private cubicles. This set up allowed to administer up to 4 participants at the same time. After

a short reception, the participants were given an informed consent form and asked if they had any questions about the study procedure. When the consent form was provided to the experimenter, the participants were handed the first series of questionnaires and materials to submit the first saliva sample. After completing these tasks, participants were given a thought writing task about romantic acceptance and connectedness toward their partner. The participants were informed that they had approximately 10 minutes to fulfill this task. Following this procedure, participants were handed a second series of questionnaires and after 15 minutes into this set, asked to submit a second saliva sample. Lastly the participants returned to the study site approximately one week later to deliver a follow up morning saliva sample. For a schematic outline of the procedures with time expectations for each task, see figure 1.

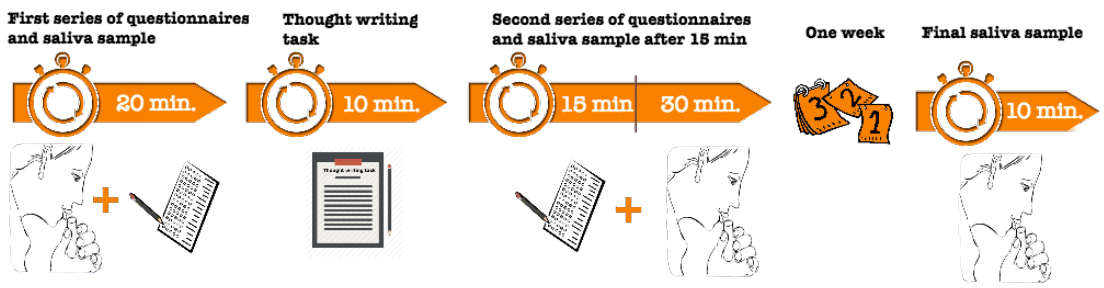

Figure 1. Schematic of laboratory procedure

Questionnaires

All forms and questionnaires were administered in Norwegian. Besides the standardized measurements in Norwegian, all questionnaires were translated. In order to preserve the precise properties of the English questionnaires, two native Norwegian speakers translated the measures to Norwegian, and then back-translated to English.

From the first series of questionnaires, participants provided basic demographic information on sex, age, and relationship length. Women answered in addition a measure providing information on hormonal contraceptives.

After completing the first set of questionnaires, the participants were given a thought-writing task to elicit OT secretion by priming thoughts about trust and connection with their romantic partner. The instructions were given on a paper with additional space to execute the task.

"Please spend a few minutes thinking about your relationship with your partner. Then write about ways that your partner responds to you in ways that show that your partner truly accepts and connects with you, or how you wish your partner would respond to you in ways that show that your partner truly accepts and connects with you. In total, you'll have about 10 min for this task. So you have a few minutes to gather your thoughts before writing."

This design is meant to evoke an OT response by thinking about his or hers relationship, comparable to study designs with tasks related OT change tasks such as "warm touch" intervention from a spouse (Holt-Lunstad et al., 2008) and speech reactivity task from individuals that had experienced relational harm (Tabak et al., 2011).

After the thought writing task, participants completed a collection of measures targeting romantic relationships and individual differences in the second questionnaire set. This packet included Simpson et al. (1996) Adult Attachment Questionnaire (AAQ), a dimensional measure of adult romantic attachment orientations. The AAQ is a seventeen items questionnaire that rates how individuals generally relate to romantic partners, thus it does not measure specifically their current relationship. All items were responded on a 7-point likert scale, ranging from strongly disagree (1) to strongly agree. The AAQ contains the following items: (1) "I find it relatively easy to get close to others"; (2) "I'm not very comfortable having to depend on other people"; (3) "I'm comfortable having others depend on me"; (4) "I rarely worry about being abandoned by others"; (5) "I don't like people getting too close to me"; (6) "I'm somewhat uncomfortable being too close to others"; (7) "I find it difficult to trust others completely"; (8) "I'm nervous whenever anyone gets too close to me"; (9) "Others often want me to be more intimate than I feel comfortable being"; (10) Others often are reluctant to get as close as I would like"; (11) "I often worry that my partner(s) don't really love me"; (12) "I rarely worry about my partner(s) leaving me"; (13) "I often want to merge completely with others, and this desire sometimes scares them away"; (14) "I'm confident others would never hurt me by suddenly ending our relationship"; (15) "I usually want more closeness and intimacy than others do"; (16) "The thought of being left by others rarely enters my mind"; and (17) "I'm confident that my partner(s) love me just as much as I love them".

The AAQ measure consists of two orthogonal dimensions that measure romantic attachments orientations. Simpson et al. (1996) presented these dimensions

by a principal axis factor analysis and a varimax rotation of the 17 items. The first dimension taps high to low levels of avoidance, and the second dimension measures high to low levels of anxiety. These dimensions are labeled respectively romantic attachment orientation avoidance and anxiety. Low levels in both dimensions consequently results in secure attachment orientation. The AAQ have shown good psychometric properties and been verified trough previous research (Fraley & Shaver, 2000; Ravitz, Maunder, Hunter, Sthankiya, & Lancee, 2010; Simpson et al., 1996).

According to Simpson et al. (1996), the AAQ can be keyed to calculate scores to construct the avoidance, anxiety, and secure romantic attachment orientations. Higher scores on item 1 to 3 and 5 to 9 reflect avoidance, while higher scores on item 4, 9 to 17 indicate anxiety. Items 1, 3, 4, 12, 14, 16 and 17 must be reversed-coded to before constructing the anxiety and avoidance attachment orientations. Low scores on both dimensions represent secure romantic attachment orientation. Secure attachment has been coded from the total sample by reverse-coding all items except 1, 3, 4, 12, 14, 16 and 17. Assessing internal consistency for each attachment orientation, all presented high reliabilities. Cronbachs alpha were respectively .826 for avoidance, .817 for anxiety and .851 for secure.

Hormonal assays

In all, the study collected saliva samples for OT extraction at three collection points. Each saliva sample consisted of two test tubes that could contain 5 mL of passive drool. All samples were frozen at - 20ºC after they were provided. The samples were shipped in a styrofoam box with dry ice from NTNU Dragvoll to UNM Hominoid Reproductive Ecology Laboratory to be maintained frozen until the time of assay. The saliva samples were processed prior the assay in a 6:1 concentration. This was achieved through defrosting, followed by mixing the solutions with a vortex mixer and 15 minutes of centrifuging the solution. This preparation removes redundant solvents from the saliva samples. Using a vacuum concentrator controlled at 4 ºC, 1.5 ml portion of the supernatant saliva was dried down to a waterless state. The saliva was restored with an assay buffer of 250 µL. All concentrations were measured in duplicate using an ELISA kit from Enzo Life Sciences (ADI-901-153A; Farmington, NY). Circulating OT from saliva samples has shown reliable alterations after intranasal OT administration (Daughters et al., 2015; van IJzendoorn, Bhandari, van der Veen, Grewen, & Bakermans-Kranenburg, 2012). The experimenter initiated

the second saliva sample 25 minutes after starting the thought writing task. This time range was calculated to capture changes of endogenous peripheral OT secretion (e.g. White-Traut et al., 2009). Enzo reports a 15 pg/mL sensitivity for this assay and a mean OT recovery of 90 % in samples. Mean intra assay coefficient of variations (CV) was 7.1 % and mean inter assay CV was 14.7 %.

Statistical analyses

This study used SPSS version 24.0 (2016) in all primary analyses. The statistical program R version 3.4.0 (2017) was conducted in making OT density plots for males and females. The selection of statistical methods is based on analyses applied in hormonal and attachment studies to better investigate prior findings and act in accordance with the scientific methods in neuroendocrinology (Marazziti et al., 2006; Schneiderman et al., 2012; Tabak et al., 2011; Weisman et al., 2013).

Results

OT baseline distributions

The OT baseline represents the mean from the first and third saliva sample collected approximately 7 days apart. The third saliva sample was missing for 16 participants. These missing samples are likely to represent a drop out from the morning follow up procedure 7 days later. These participants can accordingly not be used to estimate the average OT score and were excluded from all baseline analyses. For the available valid sample (n = 132), mean OT values for males and females are 45.25 pg/ml (SD = 22.19). The OT baseline distribution is positively skewed (1.41, SE = .22) with a long right tale (kurtosis = 3.1, SE .42). When computing males and females exclusively, the female OT baseline (n = 106) was 46.62 pg/ml (SD = 23.07). This distribution is similar to the total sample, with data clustered around the low end of the scale (skew = 1.42, SE = .24) and leptokurtic spreading (kurtosis = 2.96, SE = .47). When computing the male sample (n = 26), the mean OT baseline is 39.57 pg/ml (SD = 17.41). The male OT distributions cluster somewhat different, with a less positive skew (.76, SE = .46) and kurtosis (.34, SE = .89). The distribution of male and female baseline OT values is presented in a density estimate in figure 2.

The OT baseline distributions regarding skewness and kurtosis indicate a departure from a normal distribution in all samples. Previous research using OT as an outcome variable recommends using data transformations in these distributions (Schneiderman et al., 2012; Weisman et al., 2013) To spot if the OT baseline distribution deviates from normality, Z-scores were calculated to the total sample for skewness (6.41) and kurtosis (12.92). This states that the distribution is significantly different from a normal distribution, $p < .001$ (Field, 2013; Kim, 2013). When applying the normality tests Saphiro-Wilk and Kolmogorov-Smirnov, this further support a deviation from normality, $D(132) = .12$, $p < .001$, $D(132) = .91$, $p < .001$. To better the positive asymmetry and peakedness of the distribution, all baseline OT values were log transformed (natural log). The transformed OT baseline values are used in all statistical analyses if not specifically mentioned in the analysis.

To investigate individual baseline stability, a paired t-test and Pearson product-moment correlation analysis were conducted between the first and third OT saliva sample. The t-test presented no significant difference between the two samples, $t(132) = .58$, $p = .561$. The correlation analysis showed that the OT values are

significant stable over time, though modest in size, r = .20, p = .020.

To compare the male and female distributions, an independent t-test with OT baseline as the predictor variable was executed. The two distributions presented a non-significant difference, $t(130) = 1.37$, $p = .172$. In addition, when computing the untransformed data with an independent Mann-Whitney U-test, the difference was still not significant, U = 1.15, $p = .182$.

A Levenes test was computed to examine homogeneity of variance for males and females. OT baseline values were approximate equal and indicated no violation of homoscedasticity, $F(1, 129) = .812$, $p = .319$.

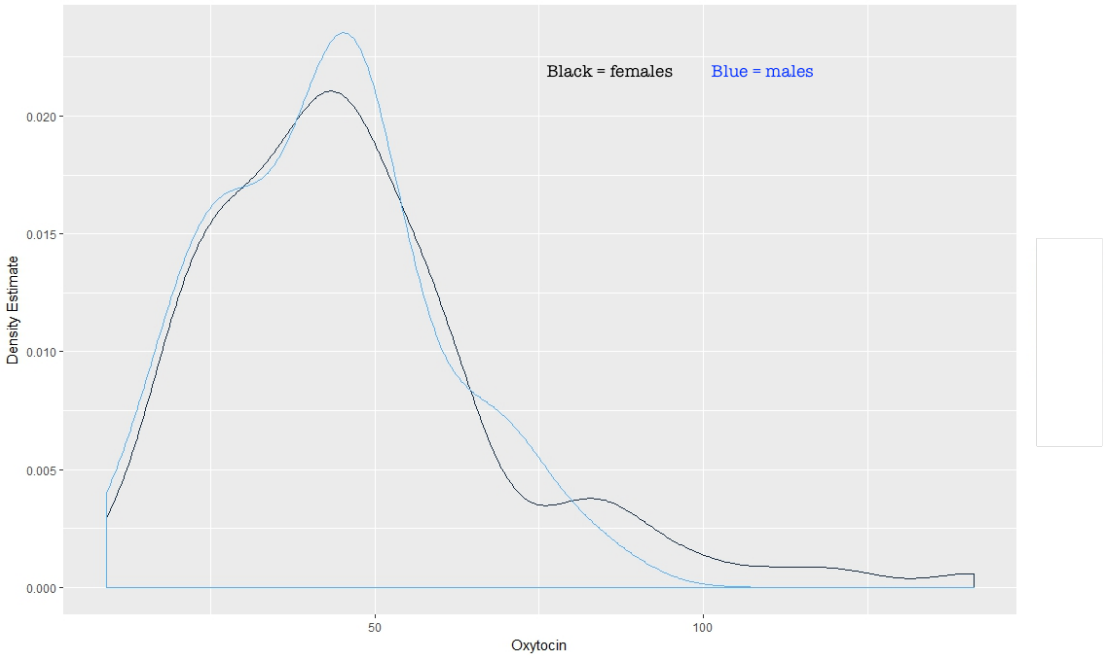

Figure 2. Untransformed baseline density plot with males (blue) and females (black).

OT change distributions

The OT change values represent the difference between the OT extraction from the first saliva sample and second (post thought-writing task) saliva sample. One participant was missing the second saliva sample. This lacking sample may be a result of incomprehensible marking or ruined sample rather than drop out, since the participant delivered the third sample a week later. In addition, one participant had an OT change score of 5 SD above the mean (113 pg/ml). In comparison, the second

biggest outlier was 68 pg/ml. This extreme outlier is probably a result of an erroneous assay or recording of the OT sample. This score is consequently considered not to be valid and thus excluded in all OT change analyses. This participant was also excluded in the OT baseline values for missing the third saliva sample. The remaining OT change values for males and females (n = 146) are 2.53 pg/ml (SD = 19.10). Distributions of OT change clustered around the center (kurtosis 1.59, SE = .40) with a small positive skew (.39, SE = .20). Computing OT change exclusively in females was 2.82 pg/ml (SD = 19.99). Distributions clustered, like the total sample, around the center (kurtosis = 1.57, SE = 2.81) with a positive skew (.34, SE = .23). The male OT change was 1.54 pg/ml (SD = 16.16) with a similar positive skew in the distribution (.60, SE = .41.), but less leptokurtic (kurtosis = 1.06, SE = .80).

The OT change values indicate that the values are distributed asymmetric. Z-scores for skewness (1.93) and kurtosis (3.98) imply non-normality in medium sized samples ($50 < n < 300$) (Kim, 2013). To further evaluate if the OT change values display substantial departure from normal distribution, Saphiro-Wilk and Kolmogorov-Smirnov are used. These tests support that the values deviate significantly from a normal distribution, respectively D(147) = .10, p = .003; D(146) = .97, p = .002. Previous research on OT change, such as Holt-Lunstad et al. (2008) and Tabak et al. (2011) have used data transformation in these OT distributions. All values regarding OT change, were thus log-transformed prior conducting statistical analysis to better approximate normal distributions and follow the data practice in the field of neuroendocrinology.

In order to investigate if OT from sample 2 is significant different from sample 1, a paired t-test was conducted. This statistic displayed that there is a significant change between these samples, $t(145) = 17.84$, $p < .001$. These results was similar when computed separately for females ($t(112) = 14.61$, $p < .001$) and males ($t(32) = 12.14$, $p < .001$).

To compare the male and female OT change distributions, an independent t-test was executed with average OT change as the predictor variable. The two distributions presented a non-significant difference, $t(144) = -.123$, p = .902. By assessing the untransformed data as an extension, an independent Mann-Whitney U-test was conducted. This nonparametric test presented no significant difference, U = 1.787, p = .717. A Levene's test indicated no differences in variances between males and females, $F(1, 144) = .93$, p = .337. The assumption of homoscedasticity was

consequently not violated.

Attachment measures

The romantic attachment orientation distributions varied to some extent from each other. The secure attachment distribution was negative skewed (- .61, *SE* = .20) with a small kurtosis (-.13, *SE* = .40). The avoidance distribution presented a positive skew (.81, *SE* = .20) and similar degree of kurtosis the as secure attachment data (.79, *SE* = .40). The anxiety distribution display a positive skew (.74, *SE* = .20) and a slightly more leptokurtic spread to the left (kurtosis = .79, *SE* = .40). According to Dunlap, Chen and Greer (1994) skew less than +/- 3 does not substantially affect test-retest reliability in the data set, and can be conducted in statistical analysis. These data are thus considered valid and can be used in the following correlation analysis and as predictor variables in the regression analyses.

Correlation analyses

Pearsons correlations in table 1 reveal that secure romantic attachment have significant negative correlations with the orientations anxious (r = - .85, p < .001) and avoidant (r = - .85, p < .001) romantic attachment. Anxious and avoidant romantic attachment orientations are additionally positively correlated with each other, r = .38, p < .001. Sex (coded with males as the higher value) is significantly positive correlated with secure romantic attachment orientation (r = .19, p = .020) and negatively correlated with the romantic attachment orientation avoidance (r = .19, p = .020).

OT baseline has a significant positive correlation with secure romantic attachment (r = .24, p = .006) and relationship length (r = .18, p = .040), and is negative correlated with avoidant romantic attachment (r = - .23, p = .007) and OT change (r = - .26, p = .002).

Table 1

Correlations between secure romantic attachment (Sec), anxious romantic attachment (Anx), avoidant romantic attachment (Avo), sex, relationship length (RL), OT change (OTC) and OT baseline (OTB) in the total sample

	1	2	3	4	5	6	7
1. Sec	-						
2. Anx	-. 810**	-					
3. Avo	- .852**	.383**	-				
4. Sex	.191*	- .155	- .162*	-			
5. RL	.081	- .063	- .072	.112	-		
6. OTC	- .730	.057	.066	.010	- .022	-	
7. OTB	.237**	- .168	- .233**	- .120	.179*	- .264**	-

** p < .01, * p < .05

Distinct correlation analyses were conducted for males and females to investigate differences between the sexes, and additionally to include the variable hormonal contraceptives for the female sample. The romantic attachment orientations in females, displayed in table 2, show highly related correlations like the total sample. Secure attachment is negatively correlated with anxious (r = - .85, p < .001) and avoidant (r = - .80, p < .001) attachment orientation. Anxious and avoidant romantic attachment have a significant positive correlation similar to the total sample, r = .36, p < .001.

OT baseline is significantly positive correlated with secure romantic attachment (r = .24, p = .006) and relationship length (r = .18, p = .040). OT baseline is also negative correlated with avoidant romantic attachment (r = - .23, p = .007) and OT change (r = - .26, p = .002).

Table 2

*Correlations in the **female sample** between secure romantic attachment (Sec), anxious romantic attachment (Anx), avoidant romantic attachment (Avo), hormonal contraceptives (HC), relationship length (RL), OT change (OTC) and OT baseline (OTB)*

	1	2	3	4	5	6	7
1. Sec	-						
2. Anx	- .849**	-					
3. Avo	- .800**	.362**	-				
4. HC	- .071	.074	- .041	-			
5. RL	.103	- .032	- .144	- .177	-		
6. OTC	- .081	.079	.054	- .047	- .036	-	
7. OTB	.255**	- .174	- .259**	.017	.208*	- .259**	-

** p < .01, * p < .05

The male correlation matrix in table 3, display similar correlations between the romantic attachment orientations in both the total and the female sample. Secure attachment is negatively correlated with anxious (r = - .84, $p < .001$) and avoidant (r = - .84, $p < .001$) attachment orientation. Anxious and avoidant romantic attachment have a significant positive correlation that is somewhat higher than the total and female samples, r = .41, $p < .001$.

Table 3

*Correlations in the **male sample** between secure romantic attachment (Sec), anxious romantic attachment (Anx), avoidant romantic attachment (Avo), relationship length (RL), OT change (OTC) and OT baseline (OTB).*

	1	2	3	4	5	6
1. Sec	-					
2. Anx	-. 843**	-				
3. Avo	- .835**	.407*	-			
4. RL	- .079	- .122	.248	-		
5. OTC	- .410	- .094	.166	.017	-	
6. OTB	.310	- .291	- .228	.181	- .301	-

** p < .01, * p < .05

Regression analyses

A multiple regression analysis was conducted to predict OT baseline values from romantic attachment orientations, relationship length, sex and hormonal contraceptives. Predictor variables were inserted in two steps using a blockwise entry. First the chosen romantic attachment orientation was entered and then sex and relationship length was added. The romantic attachment orientations were inserted first in all the analyses as they are expected from both adult attachment theory and past research to explain more variation than the other predictor variables. Separate regression analyses were additionally conducted for males and females. Only the female sample included hormonal contraceptives as a variable in these analyses.

The correlation matrix presents no substantial correlations between the predictor variables that are used in each regression model, and thus indicates no problems concerning multicollinearity within these values. This assumption is supported by collinearity statistics displaying VIF and corresponding Tolerance values close to 1 in all models. The Durbin-Watson presents values approximate to 2 in all analyses, which signal that the assumption of independent errors is acceptable. By conducting scatterplots of the standardized residuals versus the standardized

predicted values in all the regression models, they display no apparent violations of homogeneity of variance or linearity.

Table 4

Multiple regression model OT baseline and OT change in the total sample

	OT baseline			*OT change*		
	B	R^2	F change	*B*	R^2	F change
Step 1 Avoidance	- .109**	.047	7.459*	.024	- .003	.623
Step 2		.087	3.881*		- .016	.050
Avoidance	- .112**			.024		
Rel. length	.003*			.020		
Sex	- .211*			< .001		
Step 1 Anxious	- .076	.021	3.779	.020	- .004	.463
Step 2		.066	4.123*		- .017	.060
Anxious	- .085*			.021		
Rel. length	.004*			< .001		
Sex	- .204			.020		
Step 1 Secure	.130**	.049	7.222**	- .032	- .002	.768
Step 2		.095	4.301*		.006	.065
Secure	.140**			- .033		
Rel. length	.003*			< .001		
Sex	- .222*			.024		

* p <.05, ** p <.01 **, R^2 is adjusted R

Results from the regression analyses, as presented in table 4, revealed that the avoidant and secure attachment styles significantly predict OT baseline values in the first stepwise entry, respectively b = - .109, $t(130)$ = -2.73, p = .007 for avoidance, and b = -.130, $t(130)$ = 2.80, p = .006 for secure. Avoidant romantic attachment explained a significant part of the variance in OT baseline, R^2 = .047, $F(1,130)$ = 7.46, p = .007. The second step of the avoidant model increased the explained variance in the model, R^2 = .087, $F(3,128)$ = 5.81, p = .002. All variables in the second step in the avoidance model significantly contributed to predict OT baseline values. Secure romantic attachment explained a significant part of the variance in OT baseline, R^2 =

.049, $F(1,130) = 7.72$, $p = .006$. The variables added in the second step of the model increased the explained variance, $R^2 = .095$, $F(3,128) = 5.57$, $p = .001$.

The regression model using anxious romantic attachment became significant in the second entry of variables, $t(130) = -2.73$, $p = .007$. Here, the predictor variables anxious and relationship length were significant, and the second step of the model explained significant variance in OT baseline, $R^2 = .066$, $F(3,128) = 4.07$, $p = .008$.

Table 5

Multiple regression models for OT baseline in females and males

	Females OT baseline			*Males OT baseline*		
	B	R^2	F change	*B*	R^2	F change
Step 1 Avoidance	- .121**	.060	7.580**	- .122	.012	1.313
Step 2		.073	1.693		.043	.068
Avoidance	- .106*			- .165		
Rel. length	.004			.003		
Hormonal cont.	.071					
Step 1 Anxious	- .075	.021	3.168	- .181	.047	2.220
Step 2		.054	2.791			
Anxious	- .078			- .182		
Rel. length	.005*			.002		
Hormonal cont.	.094					
Step 1 Secure	.137**	.057	7.223**	.215	.059	2.556
Step 2		.080	2.250			
Secure	.129*			.247		
Rel. length	.004*			.003		
Hormonal cont.	.089					

* $p < .05$, ** $p < .01$, R^2 is adjusted R

All regression models using OT change as an outcome variable found no significant predictor variables and could not account for any significant variance in the OT change values. When conducting the same OT change analyses separately in females and males, no significant predictions or explained variance emerged. This included hormonal contraceptives in the female regression model. The following table

(table 5) contains OT baseline for females and males, but do not display the non-significant results with OT change as the outcome variable.

The female OT baseline results showed approximate equal model predictions and explained variances compared to the total sample in the first step. However, the second step entering relationship length and hormonal contraceptives fell insignificant in improving the following regression models.

The romantic attachment orientations avoidance (b = - 1.21, $t(102) = -2.75$, p = .007) and secure (b = .137 , $t(102) = 2.69$, p = .008) in females are significantly predicting OT baseline values in step 1 in the regression models. Avoidant romantic attachment is significant in accounting for 6 % of the variance in OT baseline, R^2 = .060, $F(1,102) = 7.580$, p = .007. The second step of the model did not significantly improve the prediction. Avoidance was the only predictor variable in step 2 that made a significant contribution in predicting OT baseline.

The regression model using the romantic attachment orientation secure, explained a significant part of the variance in OT baseline, R^2 = .057, $F(1,102) = 7.22$, p = .008. The variables added in the second step of the model did not significantly explain more variance. All predictor variables except hormonal contraceptives made a significant contribution in step 2 in this regression model.

Anxious romantic attachment orientation does not explain a significant proportion of variance in OT baseline in step 1 or step 2. Only relationship length is significant as a predictor variable to this regression model.

No significant results emerged from the OT baseline regression models that were conducted in the separate male sample.

Discussion

Investigating romantic attachment orientations from a neuroendocrine perspective provides a unique opportunity to better understand how OT may interact and regulate motivational states and behaviors in romantic relationships. The results suggest that OT baseline values are associated with romantic attachment orientations. The current findings offer accordingly partial support for the main predictions in this study. In the total and female sample, a positive relationship between secure romantic attachment and OT baseline was found, and an opposite relationship was found between avoidant attachment and OT baseline. Consistent with the prognostics from the correlation matrix, both avoidance and secure attachment orientations predicted OT baseline values. The explained variances in these orientations reflect value sizes as seen in prior OT research (e.g. Marazziti et al., 2006; Schneiderman et al., 2012; Tabak et al., 2011). The negative relationship between avoidant attachment and OT baseline indicates that a dampening of the oxytocinergic system may affect central psychological processes that are considered important in romantic relationship (Carter, 1998; Braithwaite et al., 2010). The leveled secretion of OT baseline can thus be implicated in reward sensitivity that initiate and maintain states that may account for low satisfaction in a romantic relationship (Bethlehem et al., 2014). The dampened oxytocinergic system can furthermore be considered to influence interpretation toward romantic partners behavior as uncomfortable close and consequently develop emotional distance. The positive association between secure attachment orientation and OT suggests correspondingly that higher levels of OT baseline can be involved in states that promote trust, satisfaction and emotionally closeness toward a romantic partner. OT may thus stimulate psychological resources to motivate different social behaviors or potentiate certain physiological states in romantic interactions. From the negative and positive associations between OT and romantic attachment orientations that emerged, OT can have a neuromodulatory function to regulate reward sensitivity. This can be supported by OTs central secretion to the mesolimbic dopamine system, which is known for its effect on the reward related effects (Johnson & Buisman-Pijlman, 2016; Strathearn, 2011). From the "Identify and Invest" hypothesis OT secretion is expected to orient psychological resources as a response to threats or signals that the individual perceive in the relationship. This is suggested as OT baseline regulates properties of attention and appraisal to cues and signals in a

romantic relationship. These characteristics may enable or inhibit development and maintenance of stable and supportive relationships. Simpson (1990) claimed that individuals with an avoidant or anxious romantic attachment may often desire stable and supportive relationships, but their mental models prevent them to achieve this type of romantic bond. The romantic attachment orientations can thus be understood in an "Identify and Invest" framework to demand different psychological resources toward the perceived cues in the relationship.

This study additionally proposed that relationship length, sex and hormonal contraceptives would cause a moderation effect between OT and the romantic attachment orientations. When including relationship length and sex as predictor variables in the regression models with avoidant and secure attachment orientations, the explained variance in OT baseline increased significantly. This increase indicates that these variables improve the prediction in OT baseline. OT change values however, showed no significant associations with any of the predictor variables in any analyses. This null result in OT change is noteworthy to recognize to understand how neuroendocrinological changes can be tied to specific circumstances and individual differences, but not others. From the result section, OT change increased significantly when the participants were primed, in the thought-writing task, to think about their romantic partner. This suggests that a neuroendocrine effect took place when engaging in this mental activity. However, attachment orientations, relationship length, sex and hormonal contraceptives did not detect this effect and can thus represent a lack of fit regarding conditions that triggers immediate OT secretion. Prior neuroendocrinological research have shown that immediate OT changes occur in response to individual differences such as perceived discrepancy between self and partner involvement and in a range of specific conditions (Grebe et al. 2017; Gordon et al., 2008; Holt-Lunstad et al., 2008; Tabak et al., 2011). Further research should address if the associations between the oxytocinergic system and romantic attachment orientations is restricted to a more stable OT secretion pattern than an OT changeable response secretion

An extension of the main attachment predictions was whether OT secretion differs between males and females. The female regression models were significant at the same attachment orientations as the total sample, and explained approximate the same levels of variance in OT baseline. The female sample additionally included hormonal contraceptives to examine if higher levels of synthetic estradiol would have

a potentiating effect on OT secretion. This effect was not found in either OT measure or in any attachment orientation. One possible explanation to this null finding on hormonal contraceptives is that the female sample is biased by that the females participating in this study were using atypical contraceptives, such as progestin-only contraceptives (mini pill) or combination contraceptives with higher levels of progestin. If the sample is highly skewed to these types of hormonal contraceptives, an alternative effect should have emerged in the opposite direction of what I predicted. While estradiol may have a positive effect on the oxytocinergic system, progesterone (progestin) binds to OT receptors in the body, thus antagonizing their function (Grazzini, Guillon, Mouillac, & Zingg, 1998). It is also possible that the female sample have zeroed out an effect by possessing approximate equal groups of both typical and atypical hormonal contraceptives. It is regardless no empirical indication that hormonal contraceptives are associated with OT in this current study.

Despite previous research presenting OTs neuromodulatory effect on romantic attachment orientations, no effect appeared between OT and any predictor variables in the male sample (Marazziti et al., 2006; Schneiderman et al., 2012; Opacka-Juffry & Mohiyeddini, 2012). This absence of significant relationships and predictions represent a striking difference from the female sample. In animal research, OT has in general shown a marked sex difference regarding affiliate behavior (Francis, Young, Maeney & Insel, 2002; Young & Wang, 2004). Human neuroendocrine research has however not been as conclusive in social behavior as the animal research. There may be a range of different reasons behind this, such as less control in human studies for extraneous factors, fewer empirical studies and variation between species in the oxytocinergic system (Carter 2014). It is essential to mention that interactions between other neurotransmitter systems exist (Gangestad & Grebe, 2017; Brodal, 2013). OT does not act exclusively in the central nervous system, but is involved in a comprehensive and complex interplay with other neurotransmitter systems that we are only starting to identify (Gimpl & Fahrenholz, 2001; Gangestad & Grebe, 2017). The interactions can potentiate or inhibit synthesizing and secretion in other hormones (Carter, 2014). The neuropeptide arginine vasopressin (AVP) is closely related to OT in both structure and its social function, and these have shown to influence each other by binding to each other's receptors (Carter, 2014). AVP has however been proposed to be more important than OT in regulating affiliate behavior in males than females in animal studies (Carter, 2014; Young & Wang, 2004). The endogenous secretion of

AVP is located from the same neuroendocrine regions as OT, but is secreted differently (Carter, 2014). Where OT have been reported to be released into the bloodstream deviates pulses, the AVP does not follow this secretion pattern (Theodosis, 2002). Future work would benefit to include AVP as a measure to assess if this neuropeptide serves equal functions in males as OT in females.

A surprising null result that seems to contradict former OT and attachment studies is between anxious romantic orientation and OT baseline (Opacka-Juffry & Mohiyeddini, 2012; Taylor, 2006; Weisman et al., 2013). However, when including relationship length and sex in the model, they jointly explain significant variance in response to OT. Additionally, the romantic attachment orientation anxious and relationship length became statistical significant predictors to OT baseline. An explanation for the unexpected statistical significance can originate from a shared variance in the predictor variables that have been masked when checking collinearity plots and statistics. This collienearity might then increase the power of statistical contribution to OT baseline and creating significant predictors. Simpson et al. (1996) argue that individuals scoring high on anxious romantic attachment may want to develop stable relationships, but is inhibited by a sustained suspicion and insecurity about the quality of the relationship. This implies that anxious individuals may have shorter relationships. Relationship length in this regression model may consequently share residual variability with anxiety and should be considered with caution.

Contrary to Schneiderman et al. (2012) findings for elevated OT secretion in the initial stages of the romantic relationship, no associations were found between relationship length and OT in the current study. Schneiderman et al. (2012) used two samples to assess OT values; one group consisting of young adults in the beginning of a new romantic relationship, and the other group with young single adults. The use of a group of single adults, may explain why Schneiderman et al. (2012) was not reproduced in this current study. Using an exclusive group of singles may represent a sample with substantially lower circulating OT than individuals in an exclusive relationship. Thus the potentiating effect by being in a new relationship may be zeroed out or be too small alteration to detect a significant effect in the correlation analyses when assessing only individuals in a relationship.

A noteworthy finding of the present study was the negative correlation between OT change and OT baseline. The direction of this covariance was explorative to achieve a better understanding of how the oxytocinergic function in secretion over

a longer period versus a specific situation to eliciting an OT response. This negative relationship is, as far as I know, the first detected association in two secretion measures in a psychology study. This discovery indicates that OT secretion is versatile and may affect different psychological functions depending on momentarily reactions (e.g. states) and more permanent features (e.g. traits). This relationship was however not found in the male sample, in any direction. This raises an interesting question why the male sample, having a significant increase of OT after the thought-writing task, did not show the same neuroendocrine OT pattern as the female sample.

OT distributions

The OT baseline values in this study presented a non-normal distribution that was positively skewed. This asymmetry indicates that higher baseline scores are more deviant from the median than the lower OT scores. This distribution replicates one of the largest published OT distributions as presented in Weisman et al. (2013). Weisman et al. (2013) analyzed baseline measures on 473 adults (mean age 27) associated with an Israeli University over 5 years. The Israeli sample highly resembles the Norwegian sample that was used in this study. The main differences between these samples are that OT values in Weisman et al. (2013) were collected from plasma instead from saliva, the exclusion criteria was less strict (included both single participants and pregnant females), and the Israeli sample achieved a more balanced sex ratio (females = 58,5 %). Even so, the right skewed leptokurtic distribution demonstrates that the OT baseline values are not an artifact spreading and thus assumed to represent a reliable measure. This indicates that OT baseline values in the general population of young adults, sampled from two nations, follow a non normal distribution with a skew to the right. Weisman et al. (2013) also reports a significant sex difference using non-transformed data. Conducting both non-parametric statistics to the untransformed values and parametric statistics to the transformed values, no significant difference emerged in the Norwegian sample.

Comparing the distribution of the OT change in this study to other OT change studies are not useful as this task related change is tied to a specific procedure that, as I know of, is the first of its kind.

Limitations

The sex ratio in this current study is uneven, with 22 % males and 78 % females. This sample skew can lead to methodological problems regarding statistical power in the male sample. To interpret this skew we have to understand the population where the sample was drawn. The recruitment in the current study was mainly carried out at the two largest university campuses in Trondheim focusing on a student population. Recruitment information additionally reached adults who are not affiliated with the university through the municipal newspaper, Adressa. However, all testing was executed in a laboratory located at Dragvoll Campus, Psychology department, which is considerable distance from the center of Trondheim and has a higher female ratio than the other NTNU campuses. Based on numbers from The Norwegian Universities and Colleges Admission Service (Samordna Opptak), 75 % of students studying psychology are female (Samordna Opptak, 2017). We can assume that easier access to the testing location site may be of an important factor to participate in the study, and thus the sex ratio is representative for an undergraduate psychology population. It should also be noted that even though this current study have a lower rate of participating males, the male frequency (n = 33) is nearly the same number other OT studies have used, such as Marazziti et al. (2006) with 45 participants (males = 12) or Tabak et al. (2011) with 49 participants (all females). The male frequency can thus separately be comparable to other published OT research. The male sample should still be considered with caution, as the margin of error is bigger in the male sample than the larger female sample. This can skew the male results to miss a potential genuine effect in the data. Consequently, future research should aim at recruiting larger male samples to increase power.

In the female sample, no associations between hormonal contraceptives and OT were found. That does not mean that hormonal contraceptive effects do not exist. As potencies and dosages of hormonal contraceptives differ between different brands, the predicted potentiating effect on OT from synthetic estradiol in the female sample could be lost in the data (Norsk Legemiddelhåndbok, 2016).

A methodological consideration is the use of Simpson et al. (1996) adult attachment questionnaire, to assess romantic attachment orientations. It exists a range of different attachment instruments focused on romantic attachment (Ravitz et al., 2009). This variety of scales can cause a problem when comparing neuroendocrine studies with each other, but are using different scales. Prior research on associations

between OT values and the romantic attachment orientation anxiety did not reproduce in this study (Marazziti et al., 2006; Weisman et al., 2012). However, these studies used the questionnaire, Experiences in Close relationship (ECR), instead of AAQ, which may not hold equal constructs.

The psychometric properties in the AAQ displayed that secure romantic attachment orientation have strong negative correlations with anxious and avoidant romantic attachment orientations. This is to be expected as secure attachment is reflected in low scores on the avoidant and anxious orientations, thus exhibit less problems that are associated with high scores on avoidant and anxious orientations. A small positive correlation is exposed between the attachment orientations avoidant and anxious, which indicate that they share some variance. Some caution is, consequently, warranted when interpreting these attachment orientations with the other variables.

A general concern regarding neuroendocrine research conducted in the peripheral system, such as this study, is that the coordination between central and peripheral secretion is not fully understood (Theodosis, 2002; Carter, 2014). Because of OTs molecular peptide structure, only a small portion of the secreted hormones in the peripheral circulatory system can pass through the blood brain barrier into the central system, thus restricting permeability for OT between these systems (Brodal, 2013; Gangestad & Grebe, 2017). Although several scholars in neuroendocrinology point that the peripheral OT values reflect central values, these studies are almost exclusively conducted on animals (Burri, Heinrichs, Schedlowski, & Kruger, 2008; Kendrick, Keverne, Chapman & Baldwin, 1988). To be certain of corresponding central secretion, this current study would have needed to collect cerebral spinal fluid to extract levels of OT with a lumbar puncture. This procedure is definitely more likely to cause discomfort than a saliva sample, and demands more advanced and higher priced extractions methods used to analyze than from peripheral samples.

Conclusion

OT has been of keen interest within the field endocrinology, though much research has focused on animal studies or human administration studies. These studies may lack ecological validity under which circumstances they secrete in response to and not controlling for individual differences, such as romantic attachment

orientations. This study lend support to the hypothesis that naturally secreted OT values have a central role in regulating psychological processes in both secure and avoidant romantic attachment. OT may thus be implicated to stimulate differences in sensitivity to reward and how we appraise a romantic relationship. However, this effect was only evident in the total and female sample. An intriguing discovery was that OT baseline values and OT change values in the total and female sample were correlated negatively, which indicate that the oxytocinergic system differs in momentarily reactions and more permanent features. The distribution of OT baseline values in this study additionally supports prior findings that OT baseline distributions are positively skewed in young adults.

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