THE UNIVERSITY OF CHICAGO

THE RELATIONSHIP BETWEEN ACUTE PSYCHOSOCIAL STRESS, HORMONES, COGNITION, AND SOCIAL BEHAVIOR IN MEN AND WOMEN

A DISSERTATION SUBMITTED TO

THE FACULTY OF THE DIVISION OF THE SOCIAL SCIENCES

IN CANDIDACY FOR THE DEGREE OF

DOCTOR OF PHILOSOPHY

DEPARTMENT OF COMPARATIVE HUMAN DEVELOPMENT

BY

NORA CATHERINE NICKELS

CHICAGO, ILLINOIS

JUNE 2019

TABLE OF CONTENTS

LIST OF TABLES

LIST OF FIGURES

ACKNOWLEDGEMENTS

To my Ph.D. advisor, Dario Maestripieri.

To my dissertation committee members and mentors: Lindsey Richland, Anna Mueller, and Greg Norman.

To my many invaluable research assistants.

To my University of Chicago family.

To my family.

To Andrew.

ABSTRACT

Human social endocrinology research has shown that hormones are sensitive to social stimuli and may show predictable fluctuations during brief social interactions. I present an investigation of the function of potential hormonal and cognitive changes that occur during a brief social interaction by manipulating both the ecological stimuli and physiological state of male and female participants before a potential courtship opportunity. Chapter 2 examines the relationship between psychosocial stress, social interaction, and hormonal reactivity (testosterone and cortisol) in men and women, and suggests that relationship status and psychosocial stress may be important variables moderating the relationship between ecological cues of courtship and subsequent adaptive physiological responses. Chapter 3 explores how psychosocial stress and social interaction relate to cognitive performance. I found little evidence that a courtship opportunity impairs cognitive performance; instead, signs of increased stress (e.g., subjective anxiety, cortisol reactivity) actually seemed to correlate with improved cognitive performance, indicating that acute stress may increase overall attentional focus. Chapter 4 investigates the relationship between hormonal reactivity, social behavior, and stress. My data did not support the connection between testosterone reactivity and displays of courtship behavior, but did support the idea that psychosocial stress may affect the relationship between personality and courtship behavior. Further, the data support the idea that individuals who experience higher increases in cortisol after psychosocial stress may show less courtship display behavior.

CHAPTER 1

Introduction

Affiliative behavior and courtship between potential mating partners are part of a behavioral system that plays a crucial role in sexuality and reproduction. Behavioral neuroendocrine research in many nonhuman vertebrate species has examined how hormones affect courtship behavior, as well as the reverse relationship of how social and behavioral states modulate endocrine function relative to courtship. Given the phylogenetic conservation of brain mechanisms that regulate these relationships (Roney, 2009), it is likely that human baseline endocrine function and hormonal reactivity are sensitive to similar social stimuli, and that both baseline and reactivity of sex hormones play an equally important role in courtship behavior in humans, as in other vertebrates. However, it is not well understood how sex hormones function in humans to promote behavior or cognition that facilitate courtship directly. Further, given the complexity of humans as a species, many variables have the potential to moderate the relationship between endocrine function and courtship behaviors, such as relationship status (a possible indicator of mating motivation), social cognition, and individual differences in personality (e.g., Maestripieri et al., 2014; Wilson et al., 2015). As such, in this dissertation, I focused on the relationship between neuroendocrine function, courtship behavior and cognition, and on how changes in situational factors and individual differences affect the physiological and behavioral output during human courtship.

As naturally-selected, domain-specific psychological mechanisms are triggered to shift vertebrate species' energy to address mating effort, sex hormones increase to induce courtship behaviors and respond to cues of mating opportunities (Roney, 2009). These behavioral responses include recognizing a mating opportunity, determining attractiveness of a member of the opposite sex, and further, predicting a potential mate's interest and perception of one's own attractiveness. In humans, the function and sensitivity of hormones have been shaped by evolution to affect important domain-specific efforts that are related to courtship, such as competition and mating. However, we also know that in humans mating does not automatically occur; courtship and mating often involve significant time and effort, and courtship is often supported by important psychological elements including motivational states and traits, such as self-confidence, assertiveness, or ambition (Buss, 2003). Although not always consciously occurring, these psychological changes may be triggered in humans upon onset of a courtship opportunity that may affect how one feels or behaves during courtship. It is possible that hormones and cognition play a role to deal with both interactions with a potential mate and interactions with competitors for potential mates, but it is not well understood how these factors interact to mediate the relationship between input (ecological cues of courtship and potential mating opportunities) and output (behavioral output manifested during a mating opportunity).

For nonhuman vertebrates and humans, courtship opportunities involve both sexual and competitive elements. In humans, it is likely that the hormones that play important roles during sexual and competitive social interactions, such as testosterone and cortisol, regulate courtship behavior. Further, since the initial phases of courtship involve social evaluation by a potential partner, psychosocial stress associated with social evaluation threat and hormonal changes triggered by such stress must also be taken into consideration (Ponzi et al. 2015; 2016). Finally, social cognition, motivation, and personality are likely to be important as well. For example, cognition is required during human courtship as individuals must advertise their intelligence, personality, and confidence while simultaneously interpreting the behaviors and mental states of the potential partner. Hormones, cognition, motivation, and personality may reciprocally interact in complex ways. For example, stress may dampen the physiological changes that normally occur to aid courtship behavior (e.g., increase in testosterone). However, extroverted individuals may not experience significant psychosocial stress triggered by social evaluation by a potential mate (Wilson et al. 2015), while introverted individuals may have to deal with this stress by diverting cognitive resources (Ponzi et al. 2015). By manipulating social and physiological variables and assessing cognitive performance and personality traits, we can better understand the function of physiological and cognitive changes that happen during a courtship social interaction and how they are moderated by individual differences. In this project I investigated: 1) how brief social interactions with opposite-sex confederates interact with testosterone and cortisol reactivity, and how psychosocial stress affects this relationship; 2) the interaction between brief social interactions with opposite-sex confederates with cognitive performance, and the effects of psychosocial stress on this interaction or cognitive performance going into a social interaction; and 3) the relationship between hormonal reactivity to social interactions / psychosocial stress and behavioral output that may be adaptive for courtship, as well as how individual differences in relationship status (a potential proxy for motivation; Maestripieri et al. 2014), personality, and gender may matter to behavioral output after these types of social interactions.

CHAPTER 2

Relationship between stress, social interaction, and hormonal reactivity in men and women

Abstract

In humans, both the associations between baseline testosterone and relationship status, motivation, and personality, and the sensitivity of testosterone reactivity to social stimuli have shown strong support for the function of testosterone in courtship and mating effort. Previous work has shown that even subtle cues of courtship induced by brief social interactions with an individual of the opposite sex can trigger increases in cortisol and testosterone. However, less is known about how a situational factor such as psychosocial stress occurring prior to a social interaction courtship opportunity may affect the relationship between an ecological trigger of courtship and adaptive neuroendocrine responses. This study investigated the effects of psychosocial stress on the physiological response to social interaction with an opposite sex confederate. For both males and females, single individuals showed significantly more positive changes in both testosterone and cortisol following a brief social interaction when compared to individuals who were in a relationship. Male and female participants who went through a psychosocial stress treatment prior to the social interaction did not show this difference in terms of singles vs. those in relationships. This study suggests that relationship status and psychosocial stress may be important variables moderating the relationship between an ecological cue of a potential courtship opportunity and subsequent adaptive physiological responses.

Introduction

Behavioral endocrinology of human courtship

For most vertebrate species, cues from potential mates are known to trigger species-specific courtship behaviors (Buss 1989; 2003). Specifically, neuroendocrine mechanisms in males have been shown to regulate species-typical courtship behaviors that are activated by visual, auditory, or chemosensory cues from conspecific females (Andersson, 1994; Zilioli et al., 2016). Across many species, testosterone regulates energy distribution and promotes investment into mating efforts in a variety of ways (Ellison, 2009; Ellison & Gray, 2009; Gleason et al., 2009; Higham et al. 2013). One primary function of testosterone is to increase energy allocation to courtship and mating efforts, signaling trade-offs with other behavioral and physiological priorities (e.g., investment in parenting, immune function, etc.) The role of testosterone as a signal to coordinate behavioral investment in courtship and mate pursuit in many vertebrate species has more recently been applied to the initiation and formation of human romantic relationships (Roney & Gettler, 2015).

In humans, both the associations between baseline testosterone and relationship status, motivation, and personality, and the sensitivity of testosterone reactivity to social stimuli provide strong support for the function of testosterone in courtship and mating effort (Roney & Gettler, 2015; Ellison & Gray, 2009; Maestripieri et al. 2010; 2014). For example, evidence for an overall decrease in baseline testosterone in expectant and new fathers shows that testosterone may serve a variety of functions, one of which being to reduce the likelihood of distraction of courtship and mating and therein reduce distraction from the mother and infant (van Anders & Watson, 2006). Importantly though, for nonhuman vertebrate and humans, courtship opportunities involve both sexual and competitive elements. Therefore, testosterone's function may be multifold in the evolutionary framework of human courtship, by serving to simultaneously confront competition for courtship opportunities, increase approach behaviors to interact with potential mates, overcome stress or apprehension involved with social interaction, and increase self-confidence and self-perceptions of status (see: Boksem et al., 2012).

The leading theories in the social endocrinology of competition and status, the Biosocial Model of Status (Mazur, 1985; Mazur & Booth, 1998) and the Challenge Hypothesis (Wingfield et al., 1990) have sought to explain the biological underpinnings of dominance-related behaviors by suggesting that these behaviors are linked to changing levels of testosterone during human social competition. The Challenge Hypothesis was first developed to explain increases in testosterone during territorial and/or reproductive challenges in nonhuman animals, but has since then been used in the literature to explain changes in testosterone in human males in response to social status competition (Archer, 2006). Expanding upon this idea, the Biosocial Model of Status predicts that testosterone increases in response to status gained through victory and decreases in response to status lost through defeat (Mazur and Booth, 1998). Theoretically, both the Challenge Hypothesis (Wingfield et al. 1990) and the Biosocial Model of Status (Mazur 1985) predict increases in testosterone during social situations, such as courtship, to facilitate successful competition against other potential mates partners.

There is a large empirical literature that has linked acute increases in testosterone to situations involving social competition and social status (status seeking and status maintenance) (e.g., Mazur & Booth, 1998; Archer, 2006; Mehta et al., 2008; Bateup et al., 2002; Zilioli & Watson, 2012; Henry et al., 2017). In terms of the connection to human courtship, both of these social interactions that have been linked to short-term fluctuations in testosterone fit with a functional model wherein increases in testosterone may be an adaptive strategy to encourage successful courtship strategies. For example, one review of recent studies that examined the causal effects of testosterone on social interactions in both animals and human argues that testosterone, in the context of social behavior, is best understood in terms of the maintenance of social status (Eisenegger et al., 2011). If increases in testosterone encourage motivation to achieve and maintain high social status, this may benefit the display of higher status and dominance behaviors relative to potential competitors seeking a similar courtship opportunity. Further, this may also boost self-confidence and motivation to overcome potentially maladaptive psychological processes that are simultaneously occurring (e.g., apprehension) upon interaction with a potential mate. Another example is that of the finding of situational changes in testosterone concentrations predicting future aggressive behavior in men (Carré et al., 2009) and future willingness to engage in a competitive task (Carré & McCormick, 2008). This function of heightening testosterone may serve to induce willingness to interact with a potential mate, even if other competitors are present.

Recently, both field and laboratory studies that have examined human male reactive testosterone responses to social interactions with females have allowed for examination of the role of testosterone in early stages of courtship that could lead to relationship initiation (Flinn et al., 2012; Miller et al., 2012; Murcia et al., 2009; Peters et al., 2016; Ronay & Hippel, 2010; Roney et al., 2003, 2007, 2010; Roney & Gettler, 2015; van der Meij et al., 2008). Roney and colleagues (2003) were some of the first to test for hormonal testosterone reactions of human males to brief social encounters with opposite sex individuals, considered to be potential mating partners in this context. They found that salivary testosterone of young men increased significantly over baseline levels after engaging in a short conversation with a young woman (this effect was only evident, or was most pronounced, in single men than in men in stable romantic relationships; see below) but did not increase significantly over baseline levels after engaging in short conversation with another young man (Roney et al., 2003; 2007). In these studies, the changes in testosterone also correlated with participants' ratings of confederates as potential romantic partners (Roney et al., 2003) and ratings of male participants' extraversion and degree of self-exposure (Roney et al., 2007). Consistent with the notion that dominance may also play a role in fluctuations of testosterone during brief encounters with a potential mate, van der Meij et al. (2008) found that higher salivary testosterone increases after brief interactions with women were most evident in men with aggressive dominant personalities, men who were not involved in committed, romantic relationships, and men who had been sexually inactive for over a month. As such, individual differences in personality, relationship status, and sexual motivation may affect the function of testosterone reactivity. Further research is needed to determine specifically how individual differences in testosterone responses to potential courtship opportunities correlate with individual differences in other personality traits (e.g. extroversion) or behavioral output. This is particularly important, as courtship situations often involve both advertisement of personality and use of personality traits to interact in an adaptively successful way.

Although male testosterone appears to respond positively toward psychological alignment toward mating initiation and mating-related stimuli (Roney, 2016), it remains unclear whether female testosterone reactivity serves a similar purpose. Much research on females has focused on the effects of menstrual cycle phase on female sexual psychology and behavior (Roney, 2015; Goldey & van Anders, 2011; Goldey, 2015). Importantly, however, Lopez et al. (2009) did replicate the findings of Roney and colleagues (2003; 2007), measuring similar endocrine responses in women to brief interaction with male confederates. Females showed significant reactive increases in cortisol after viewing a movie clip of a courtship situation with an attractive man and a young woman (Lopez et al., 2009). Researchers are currently calling for further research on the endocrinology of competition and courtship, and are emphasizing the importance of including females in study designs (Casto & Prasad, 2017; Cobey & Hahn, 2017). Although most previous studies of testosterone and competition have been conducted with men, there is some evidence that similar effects can be observed in women as well (Bateup et al., 2002; Henry et al., 2017). It is possible that female-female competition for potential courtship opportunities may induce a similar testosterone increase in females to aid in self-confidence, to preserve dominance and status, and to emphasize motivation to affiliate with potential mates.

Human courtship and stress reactivity

While studying the adaptive behavioral strategies during courtship in humans, we must consider multiple ecological cues of courtship opportunity that have the ability to influence many factors that may promote or subdue courtship behaviors. One important ecological cue may be psychosocial stress. All social evaluative interactions, including courting and mating, involve stressful components, and courtship in particular may cause significant apprehension both in anticipation of and during courtship opportunities (Wilson et al., 2015; Ponzi et al., 2015; 2016). We know that human courtship strategies did not evolve in a stress-free environment, and that any kind of social evaluation places pressures upon and threatens one's own self perceptions, through dampening self-motivation, self-confidence, or perception of social status. Further, stress has the ability to dampen social or sexual motivation, decreasing willingness to approach or affiliate with potential mates. Therefore, it is important that we consider the effects of stress on courtship behaviors to study how stress may affect courtship behavior in either adaptive or maladaptive ways.

Stress activates the hypothalamic pituitary adrenal (HPA) axis, which involves the secretion of corticotropin-releasing hormone and adrenocorticotropic hormone, stimulating the release of glucocorticoids (Sapolsky et al., 2000). In humans, the primary glucocorticoid is cortisol; cortisol mobilizes energy resources by elevating blood glucose levels and can be measured in blood, urine, and saliva (Kirschbaum & Hellhammer,1994). The peak cortisol response occurs approximately 20-40 minutes after onset of stress. Much research places the focus of cortisol increase as a coping mechanism to aversive physical or psychological conditions, and as an adaptive function to divert energy and suppress unessential processes (Sapolsky, et al., 2000). Importantly, cortisol responses are also elicited when one is faced with a psychological threat to one's self esteem and when psychological well-being is threatened (Dickerson & Kemeny, 2004). In line with the social self-preservation theory, which predicts cortisol reactivity when one is motivated to maintain social status or acceptance, we could potentially expect courtship apprehension to trigger a cortisol increase in addition to a testosterone increase and other psychological or cognitive responses that may prove to alter courtship behaviors.

There is already some indirect evidence for the role of the HPA axis in human courtship, as displayed by cortisol increases that could reflect anxiety or apprehension before or during opportunities for courtship (Roney et al., 2007, 2009; van der Meij et al., 2010). In multiple experiments, Roney et al. (2007) found that changes in cortisol from baseline were significantly greater among male participants who interacted with female confederates relative to males in control conditions. Lower baseline cortisol concentrations have also been shown to predict larger testosterone responses to interactions with potential female mates (Roney et al. 2010). Similarly, van der Meij and colleagues (2010) found that cortisol levels of men increased when they interacted with a female whom they reported as attractive. Beyond a one-on-one social interaction, cortisol has been found to be sensitive to the sex composition of the environment; Chesser (2013) found that human participants' cortisol levels increased significantly in the presence of opposite sex individuals compared to environments of single sex individuals. Jaremka and Collins (2017) utilized a study design where brief social exchanges were only anticipatory. College students watched short videos of an opposite-sex confederate introducing himself or herself, and although cortisol levels increased over baseline in all conditions, the increases were strongest for male participants and for participants who perceived the confederate as a more desirable dating partner (Jaremka & Collins, 2017).

It remains unclear overall how stress during courtship and subsequent increases in cortisol may interfere with or trigger other physiological and psychological responses to courtship. For example, an increase in cortisol may subdue or promote appropriate courtship behaviors based on the profile of testosterone reactivity. If testosterone shows acute increases in response to a courtship opportunity, despite a simultaneous increase in cortisol, courtship behaviors may not be hindered in a maladaptive way. However, if a cortisol increase is accompanied by a testosterone decrease, stress may prove to interfere with subsequent motivation, self-confidence, and affiliative behaviors. It is currently uncertain whether cortisol responses are generally associated with particular courtship behavior patterns, or whether the association is an effect that is strictly mediated by motivation, cognition, or individual differences in personality. Further, anxiety itself may actually drive the physiological responses of testosterone, in addition to its effects on cortisol. The effects of stress on testosterone may depend specifically on the type of social stress induced and may mediate the relationship between individual differences and behavior. For example, Crowley et al. (2018) found that testosterone mediated the relationship between uncertainty and the amount of disclosure between romantic partners.

Aims / Hypotheses

This chapter investigated the function of changes that occur during a courtship opportunity by manipulating both the ecological social stimuli and physiological state of male and female participants before a courtship interaction. Specifically, it focuses on how testosterone fluctuates to facilitate courtship, how stress responsivity and cortisol alter the effects of testosterone, and how the relationship between these endocrine responses is mediated by individual differences (e.g., relationship status). Recent research focusing on both baseline and hormonal reactivity during brief social interactions that can be interpreted as courtship interactions has suggested that the ecological cues of courtship opportunity can influence many factors that may promote or subdue courtship behaviors.

Aim 1: Investigate the effects of social interaction on testosterone and cortisol reactivity

In order to explore the relationship between testosterone reactivity and courtship behavior, I measured how brief social interactions with a potential courtship partner affect testosterone and cortisol responses in both males and females. I expected that we will replicate findings of male testosterone and cortisol increases during brief interactions with females. For females, I expected to observe a similar effect, such that testosterone would increase following a brief social interaction with a potential male courtship partner.

Aim 2: Investigate the effects of psychosocial stress and cortisol increases on testosterone reactivity

We would not expect courtship behavior in humans to evolve independent of ecological stress, and courtship interactions involve a social evaluation that may cause significant psychosocial stress. Therefore, I expected psychosocial stress to potentially dampen self-confidence, motivation, and approach behaviors, as well as affect cognition in a way that may hinder social cognition and impression management during courtship. To measure the effects of psychosocial stress and cortisol increase on testosterone reactivity, I utilized a psychosocial stress treatment to experimentally manipulate an ecologically stressful interaction prior to a potential courtship social interaction. In response to psychosocial stress I expected the following: 1) if cortisol increases dramatically due to psychosocial stress, testosterone increases would be dampened, and 2) if cortisol reactivity to stress is low, this may be associated with higher increases in testosterone. Further, I predicted that these changes will be most apparent in individuals whose motivations are in line with potential courtship behaviors (e.g., single individuals, individuals with higher short-term mating orientation, etc.).

Methods

Study Design / Overview

Participants were randomly assigned prior to arrival to either a control or psychosocial stress condition (Trier Social Stress Test), and those assigned to the stress condition were also semi-randomly assigned to interact with either same-sex or opposite-sex judges during the Trier Social Stress Test (TSST), based on researcher availability. Upon arrival at the lab, participants provided consent, filled out questionnaires (see below), and participated in either the control or TSST condition. Following the assigned condition, all subjects went through a social interaction with an opposite-sex confederate. Finally, participants completed two social decision making tasks and participate in a phased debriefing. Following full debriefing, participants were compensated and the experimental session ended. Saliva samples were collected at various time points throughout the tasks to assess hormone levels via ELISA: once before the control/TSST condition, once after the control/TSST condition, and once after the social interaction. Cognitive tasks were also administered throughout the protocol, in conjunction with the collected saliva samples. The Social Science Institutional Review Board at the University of Chicago had approved all experimental procedures (IRB #12-1251).

Participants

Participants were 156 individuals (age range: 18-35 years) from the greater Chicago area, surrounding the University of Chicago campus. All participants were recruited on the University of Chicago campus through fliers, UChicago Marketplace, and a human subject recruitment website (Sona System). Data was collected continuously throughout fifteen months, from July 2017 through August 2018. All study participants completed a digital prescreen consent form and eligibility questionnaire via Qualtrics software, a digital main protocol consent form and questionnaire via Qualtrics software, and also a written informed consent form at the laboratory before participating in the study. All participants were paid $20 compensation after completion of the procedures: $15 as base pay, and $5 as "winnings" from the decision making tasks. Descriptive statistics of the participant sample can be found in Table 1.

Table 2.1. Descriptive statistics for the participant sample. Note: The sample size was 156.

	%, Median, or Count
N	156
Median Age	21
White, %	32.6
Black, %	14.4
Hispanic, %	9.1
Asian (East Asian), %	27.3
Asian (Indian), %	8.3
Female, %	62.1

Student (Undergraduate and Graduate), %	82.69
Married, %	5.3
In a relationship, %	46.2
Heterosexual, %	100

Table 2.1, continued. Descriptive statistics for the participant sample. Note: The sample size was 156.

Experimental Procedure

All experimental procedures took place at the Behavioral Biology Laboratory, located in the Biopsychological Sciences Building (940 E. 57th Street) at the University of Chicago. Prior to arrival at the lab, participants consented to a prescreen questionnaire administered via Qualtrics software. If eligible, participants consented to the main study via a full study consent process via both Qualtrics software and in person at the laboratory.

All experimental procedures took place between 11:30 AM and 5:30 PM. Participants always interacted with an experimenter, or "greeter", of the same sex throughout the entire experimental session. Upon arrival, participants were taken to the testing room, where they completed questionnaires for 20 minutes. An initial demographic survey asked information about participants' age, ethnicity, sexual orientation, SES, marital or relationship status (single or in a relationship), etc. At the end of this period, they provided a baseline saliva sample and performed a baseline cognitive task. They then either took part in the Trier Social Stress Test or sat in a room doing nothing for a similar period of time as a control condition. Another saliva sample and cognitive task was collected and performed after the TSST or the control condition, approximately fifteen minutes after the start of the TSST or control condition. Approximately ten to fifteen minutes after the TSST or control condition had ended, participants went through a brief social interaction task with a confederate of the opposite-sex. After that, participants completed a single shot version of the Ultimatum Game and an iterated version of the Prisoner's Dilemma game; for each of these social decision making tasks, participants played against hypothetical partners of the opposite sex – one of a older age group and one of a younger age group (counter-balanced). Participants were shown a photo of their partner, were told this was the partner they would be playing against, and were told that it was a real participant whom has been selected at random as their partner. To increase motivation for all decision making tasks, we informed participants that they had a chance of obtaining an actual monetary reward, a commonly used procedure when administering monetary and other decision making tasks. After these social decision making tasks, participants went through a phased debriefing process (see below). Upon completion of all procedures, participants were fully debriefed and given compensation of $15 for participation and $5 in winnings ($20 total).

Questionnaires/Materials

In addition to a demographics survey and a pre-general health survey given before saliva samples were collected, participants completed the following questionnaires:

<u>Prior to lab arrival:</u>

Reduced version of the Morningness-Eveningness Questionnaire (rMEQ; Adan & Almirall, 1991): We assessed chronotype with the reduced version of the Morningness-Eveningness Questionnaire. The rMEQ is a validated, 5-item Likert-type scale obtained from the original 19-item version of the MEQ (Horne & Östberg, 1976). The rMEQ identifies participants' preferences in relation to sleeping and waking time, the time of day when they experience maximal efficiency, their level of tiredness within half an hour of awakening, and their self-perceived chronotype (Adan & Almirall, 1991). Scores for the rMEQ range from 4 to 25; scores below 12 identify participants as evening-types and scores above 17 as morning-types. In previous studies from our laboratory, we have shown that interindividual variation in chronotype is associated with cortisol reactivity to psychosocial stress as well as with socio-sexuality and cognitive function (Maestripieri, 2014; Piffer et al., 2014; Ponzi et al., 2015; Marvel-Coen et al., 2018).

The Autism-Spectrum Quotient (AQ; Baron-Cohen et al., 2001): The AQ is a 50-item self-report measure of autistic-like traits in adults with normal IQ. It includes items such as "I prefer to do things with others rather than on my own" or "I tend to have very strong interests that I get upset about if I can't pursue". This questionnaire has been widely used and validated in many studies of children, adolescents, and adults. In the AQ, four-point Likert-type responses are dichotomized to yield a binary score (0,1) for each item. The total AQ score can range between 0 (no autistic-like traits) to 50 (maximum autistic-like traits). In previous studies from our laboratory, we have shown that the AQ is associated with cortisol reactivity to psychosocial stress as well as with socio-sexuality (Del Giudice et al., 2014; Ponzi et al., 2016).

Trait portion of State-Trait Anxiety Inventory (STAI-T: Spielberger & Gorsuch, 1983): The State-Trait Anxiety Inventory, or STAI, is a long-standing measure that uses two scales to report two measures of anxiety (state anxiety and trait anxiety). Trait anxiety can be defined as an individual measure of intensity and frequency of experienced anxiety, which involves feelings of apprehension and heightened response of the autonomic nervous system (Spielberger & Gorsuch, 1983). Importantly, trait anxiety is seen as a relatively stable trait, and seeks to measure individual differences in proneness to experiencing anxiety as a personality trait. The STAI trait scale consists of twenty statements that have individuals rate, on a four-point Likert scale, different statements about how they feel *generally* (e.g., "I feel nervous and restless.").

Adverse Childhood Experiences Module (BRFSS-ACE; Bynum et al., 2010): The Adverse Childhood Experience (ACE) Module of the Behavior Risk Factor Surveillance System measures adverse childhood experiences of verbal, physical, or sexual abuse, including experiences of family dysfunction. The use of the study of ACEs is connected to the treatment of stress-related health outcomes and is included in this study to attempt to control for differences in stress reactivity based on prior developmental experience.

Birth weight items: Information regarding birth weight and gestational age, along with the participant's self-reported source of information, was collected to account for differences in extremely low birth weights that may be correlated with self-reported levels of psychopathology. Early life, low birth weight individuals have been shown to have elevated levels of depression, anxiety, and avoidant personality problems (Boyle et al., 2011).

UCLA Loneliness Scale (Russell et al., 1980): The UCLA Loneliness scale is a measurement used in social psychological research focusing on loneliness. Although studies have shown that loneliness is a unique psychological experience, measures of loneliness have also been found to be correlated with measures of social risk taking and negative affect (Russell et al., 1980).

<u>Upon lab arrival:</u>

Big-Five Personality Inventory (BFI; John, Donahue, & Kentle, 1991) Personality was assessed using the Big Five Inventory, a 44-item, frequently used questionnaire measuring personality traits along five dimensions: extraversion, agreeableness, conscientiousness, neuroticism, and openness to experience.

MacArthur Scale of Subjective Social Status (Adler & Stuart, 2007): The MacArthur Scale of Subjective Social Status is a developed measure of subjective social status meant to capture individuals' sense of standing on a social ladder. Based on its wording, it seeks to take into account sense of place on multiple dimensions of social position or status (e.g., socioeconomic, general status among a similar demographic, etc.)

Ten-Item Personality Inventory (TIPI; Gosling et al., 2003): The TIPI is a ten-item, brief measure of personality based on the Big-Five Personality Inventory. It has sufficient levels of convergence with the widely used Big-Five inventory and is usable in situations where short measures are needed. In this study, we use the TIPI to measure pre- and post-stress and social interaction self-reported personality ratings throughout the experimental protocol.

Shortened version of the Math Anxiety Rating Scale (sMARS; Alexander & Martray, 1989): The sMARS is an internally consistent and reliable, abbreviated form of the Mathematics Anxiety Rating Scale (MARS). In this study, we sought to measure math anxiety to capture individual differences in specific tension, apprehension, or fear surrounding math, due to the fact that the Trier Social Stress Test (TSST) involves a math task that is used to induce acute psychosocial stress (see below for more detail about the TSST).

The Multidimensional Sociosexual Inventory (MSOI; Jackson & Kirkpatrick, 2007): The MSOI is a 23-item measure of sociosexual orientation and behavior. It is comprised of two attitudinal scales with 7-point Likert-type items that measure short-term (STMO) and long-term (LTMO) mating orientation, and one scale measuring past sexual experience (PSE). The PSE includes three questions about the lifetime number of sexual partners, the lifetime number of casual sexual partners, and the number of sexual partners in the previous year.

State portion of State-Trait Anxiety Inventory (STAI-S: Spielberger & Gorsuch, 1983): The State-Trait Anxiety Inventory, or STAI, is a long-standing measure that uses two scales to report two measures of anxiety (state anxiety and trait anxiety). Unlike trait anxiety, which seeks to measure individuals differences in proneness to anxiety as a personality trait, state anxiety measures the intensity of anxiety as an acute, emotional state (Spielberger & Gorsuch, 1983). The STAI state scale consists of twenty statements that have individuals rate, on a four-point Likert scale, different statements about the intensity of their anxiety "right now, at this moment".

Trier Social Stress Test

The Trier Social Stress Test (TSST; Kirschbaum et al. 1993) is a broadly used, standardized task that is used to study hormonal responses to mild psychosocial stress in a laboratory setting. In the current study, the experimenter explained to each participant that he or she would be giving a 5-minute presentation about himself or herself for a mock job interview. Each presentation took place in front of a "selection committee" composed of two unfamiliar confederates ("judges") trained to maintain neutral facial expressions and provide no positive feedback to the participant. Each participant was informed that he or she must keep speaking for 5 minutes and that the presentation will be video-recorded for subsequent analyses of content and non-verbal behavior. If the participant ever stopped speaking before the 5 minutes are up, the judges waited in silence for the participant to resume or otherwise prompted him or her to continue. If the participant again stopped speaking, one of the judges asked one of several standardized questions (e.g. "What do you think about teamwork?"). Upon completing the 5-min speech, the judges asked each participant to perform a difficult arithmetic calculation (i.e., serially subtracting the number 17 from 2,023) out loud for another 5 minutes or until he or she reaches zero. Anytime the participant made a mistake, he or she was notified and asked to restart from the beginning. After this task, the confederates thanked the participant and left the room.

Although the "greeter" who interacted with the participant was always the same sex relative to the participant, the sex of the TSST judges alternated related to the participant, and was assigned semi-randomly, based on availability. Therefore, female participants were assigned to interact with either two same-sex TSST judges (a female "talking judge" and a female "timing judge"), or two opposite-sex TSST judges (a male "talking judge" and a male "timing judge"). Likewise, male participants were assigned to interact with either two same-sex TSST judges (a male "talking judge" and a male "timing judge"), or two opposite-sex judges (a female "talking judge" and a female "timing judge"). The semi-random assignment was based on availability of the research assistants, but was also counter-balanced as much as possible.

Participants who were assigned to the control condition and who did not participate in the TSST simply sat by themselves for 10 minutes until their original experimental "greeter" returned to let them know they could continue moving forward in the study. Participants had access to several magazines during this waiting control period.

Social Interaction

Following the control/TSST condition, a second saliva sample, and a second cognitive task, all participants partook in a social interaction task, where they interacted with an opposite-sex confederate whom they had not encountered yet at that point in the session. The social interaction task that was used in this experiment was adapted from brief social interaction tasks that have been used in several research studies in which a social interaction involving a confederate posed as either another participant or experimenter led to physiological and behavioral changes (Roney et al., 2003; Roney et al., 2007). In our study, the experimental "greeter" let the participant know that they needed approximately five or ten minutes to pass before moving on to the next part of the study, and that the participant was free to relax until the experimenter returned. Several minutes after the departure of the "greeter", an opposite-sex confederate entered the room and introduced himself or herself as a research assistant there to collect data off of a digital video camera (earlier in the session, this video camera was used to collect a digital photograph of every participant, as well as used to record the TSST session for participants assigned to the TSST condition). Chairs were arranged in the room such that the participants always sat directly across from the confederate with a small conference table positioned between them. Confederates then attempted to engage in natural, friendly conversation, while simultaneously uploading data from the digital video camera onto a computer or hard drive. The research confederates were free to use whatever means of engaging in conversation seem natural to them. Script or specific prompts were not used to avoid interactions seeming excessively artificial. Conversations lasted seven minutes, at which point the experimenter re-entered the room and interrupted the confederate and participant to seemingly complete the rest of the study protocol. Descriptive statistics for research assistant confederates during the social interaction can be found in Table 2.2.

Table 2.2. Descriptive statistics for the research confederates / social interaction partners.

	% or Count
N	15
Female, %	46.7

White, %	46.7
Black, %	20
Asian, %	33.3

Table 2.2, continued. Descriptive statistics for the research confederates / social interaction partners.

Following the brief social interaction, confederates completed a survey that assessed their impressions of the participants' behaviors during the social interaction. A factor analysis done using this survey in a Roney et al. (2003) study showed that three distinct factors can be parsed out from these items, and these three factor scales were used as composite variables in this survey for the present study. First, a display factor is characterized by items in which the participants are seen as projecting information about himself or herself to the confederate. It can be interpreted as a measure of courtship-like behavior from the participant, and includes the following items: 'tried to impress you', 'eager to talk about himself', and 'showed off to you'. Second, a polite interest factor is characterized by items suggesting attempts by the participant to gather information about the confederate, and includes the items: 'asked questions about you', 'listened carefully', and 'interested in hearing about you'. Third, a general arousal factor includes the following items: 'was not bored', 'was excited', and 'was speaking fast'.

Similar to the confederates' survey, participants also completed surveys in which they rated the impressions of the research confederates and the nature of their conversations with them during a phased debriefing process at the conclusion of the study (see below).

Phased Debriefing

Following the final saliva sample of the procedure, we utilized a phased debriefing method to gather information about the participants' assessment of the confederate and their conversation together, and also to probe for suspicion about the interaction. While alone filling out a final questionnaire via Qualtrics, participants were informed in the survey that the current project was exploring first impressions and were asked to fill out a survey assessing their impression of the confederate whom they interacted with and their conversation together. The survey was adapted from a rating instrument used by Roney, et al. (2003; 2007). Participants answered items assessing their impressions of the confederates' perceived positivity of the conversation (i.e. how exciting, interesting, pleasant, and stressful they found the interaction to be), of the confederate conversation partner's physical attractiveness (i.e. beautiful, sexy, and cute), of how much the confederate conversation partner made eye contact, of how much they believed the confederate conversation partner liked them, and finally of how desirable they found the confederate conversation partner to be as short-term or long-term romantic partners.

Following the phased debriefing survey, participants were probed for suspicion regarding the true purpose of the study and when the true purpose of the study occurred, and were fully debriefed in person upon return of the experimental greeter.

Saliva Sample Collection and Hormonal Assays

All saliva samples were collected between 12:00 PM and 5:00 PM, as previous studies have shown that afternoon hormone levels, although lower than morning levels, are more stable and therefore better suited for studies of social endocrinology (e.g., Gray et al. 2004). Saliva was collected by passive drool into plastic tubes. Saliva samples were stored in a refrigerator at -20°F. Samples were assayed for testosterone and cortisol concentrations using ELISA kits purchased from Salimetrics. Saliva sample concentrations were calculated based on kit standards using a 4-parameter nonlinear regression curve fit. For cortisol, the intra-assay CV based on concentration was 4.85% and the inter-assay CV based on concentration was 7.15%. For testosterone, the intra-assay CV based on concentration was 5.38% and the inter-assay CV based on concentration was 6.63%.

Data Analysis

All statistical analyses were carried out with R and/or SPSS. Whenever hormonal data were not normally distributed, they were log or square root transformed. When sphericity assumptions were violated, Greenhouse-Geisser corrected p-values were reported. Alpha was set at 0.05 and adjusted for multiple comparisons where necessary.

Results

Data Transformations

Three individuals did not complete the full protocol due to researcher error, participant withdrawal from the study, or computer error, and therefore were not included in data analyses. Two more individuals were excluded from all hormonal analyses due to having baseline or delta hormonal concentrations over three standard deviations away from the mean. One participant was excluded from all hormonal analyses due to saliva samples being heavily contaminated with blood. After excluding these six individuals, data were analyzed for a total of 150 individuals, of whom 76 (46 females, 30 males) underwent the TSST stress manipulation and 74 (47 females, 27 males) were in the control condition.

In the overall sample (control and TSST participants combined), baseline cortisol levels were in line with assay protocol salivary cortisol example PM range norms (F: M=0.21, SD=0.17 μg/dL; M: M=0.18, SD=0.12 μg/dL). Baseline cortisol data were positively skewed for both males and females and were therefore log transformed, which resulted in a normal distribution of baseline cortisol. Therefore, the transformed data were used in subsequent analyses. Baseline testosterone levels were in line with assay protocol salivary testosterone example norms (F: M=52.38, SD=19.51 pg/mL; M: M=152.57, SD=59.13 pg/mL). Baseline testosterone data were positively skewed for both males and females. A square root transformation of female testosterone resulted in a normal distribution of baseline testosterone data for females, and a log transformation of male testosterone resulted in a normal distribution of baseline testosterone data for males. Therefore, the transformed data were used in the following analyses. Whenever delta hormonal levels that were not normally distributed were used as dependent variables, generalized linear models were used for statistical analyses. Whenever percentage change scores that were not normally distributed were used as dependent variables, log transformed percentage change scores were used for statistical analyses.

Background variables in TSST and Control Participants

There were no significant differences between control and TSST participants in terms of age, ethnicity, sexual orientation, marital status, income, subjective social status relationship status, and relationship length. There were no significant differences between control and TSST participants in terms of any measured personality variables. There were no significant differences between control and TSST participants in terms of the following variables that may impact hormonal levels, such as: exercise in the last 24 hours, whether they drink alcohol, whether or not they are currently using hormonal medication, the time of day they were tested, menstrual cycle phase (for females), how many hours they had slept the previous night, time they went to sleep the night before the day of testing, time of waking the morning of the day of testing, whether they had eaten or drank in the past hour, whether anything had worried them that day, and whether they had felt sick at all in the past week.

Background variables in Males and Females

There were no significant sex differences in age, ethnicity, sexual orientation, marital status, relationship status, relationship length, income, subjective social status, autistic-like characteristics, loneliness ratings, or trait anxiety scores. There were significant sex differences between male and female participants in BFI-C personality scores [Conscientiousness subscale: $F(149)=7.88$, $p=0.006$] and BFI-N personality scores [Neuroticism subscale; $F(149)=12.96$, $p<0.001$]. Specifically, female participants reported higher score than males in Conscientiousness (F: M=33.32, SD=6.60; M: M=30.23, SD=6.48) and higher scores than males in Neuroticism [F: M=25.46, SD=6.46; M: M=21.60, SD=6.26]. There were significant sex differences in math anxiety ratings [sMARS; $F(149)=11.67$, $p=0.001$], such that females (M=58.84; SD=18.25) reported higher levels of math anxiety than males (M=48.77, SD=16.26). There were also significant sex differences in short-term mating orientation measures [STMO; $F(149)=18.64$, $p<0.001$], such that females (M=35.88; SD=15.77) had lower scores on the short term mating orientation inventory than males (M=48.77; SD=16.26). Therefore, further analyses that combined data from male and female participants controlled for these following scales: BFI-C, BFI-N, sMARS, STMO.

There were no significant sex differences in terms of the following variables that may impact hormonal levels, such as: exercise in the last 24 hours, whether they drink alcohol, whether or not they are currently using hormonal medication, the time of day they were tested, menstrual cycle phase (for females), how many hours they had slept the previous night, whether they had eaten or drank in the past hour, whether anything had worried them that day, and whether they had felt sick at all in the past week. There were significant differences between male and female participants in terms of what time participants went to sleep the night before testing [$F(144)=15.70$, $p<0.001$], such that females went to bed earlier than males. However, there were no significant differences between males and females in terms of hours of sleep the night before or wake-up time the morning of testing. Finally, 37 females reported using some form of hormonal contraception, and 56 females reported using no form of hormonal contraception.

Baseline cortisol and baseline testosterone (T)

No significant differences in baseline cortisol were found between control and TSST participants, $t(148)=-1.17$, $p=0.242$, or between men and women, $t(148)=0.99$, $p=0.325$. Men had significantly higher baseline T than women: $t(147)=-16.41$, $p<0.001$; F: M=52.38, SD=19.51 pg/mL; M: M=152.57, SD=59.13 pg/m. Therefore, all analyses involving testosterone were run for males and females separately. There were no significant differences in baseline testosterone between control and TSST participants in males [$t(54)= 1.11$, $p=0.27$] or in females [$t(91)=0.12$, $p=0.91$]. Because baseline cortisol was significantly positively correlated with baseline testosterone in both males ($r=0.41$, $p=0.002$) and females ($r=0.46$, $p<0.001$), further analyses involving cortisol were also run separately for men and women.

Baseline cortisol in males was significantly related to relationship status [$F(1,57)=3.65$, $p=0.033$], such that males who were in a relationship had significantly lower levels of baseline cortisol than male participants who were single (M=0.14 µg/dL, SD=0.08; M=0.21 µg/dL, SD=0.13, respectively). No other variables that may impact hormone levels were related to baseline levels of cortisol in men. Therefore, for further cortisol analyses with male participants, baseline testosterone and relationship status were controlled for. Across all female participants, baseline cortisol varied significantly in relation to whether or not the participant had eaten or drank anything within the last hour $F(1,92)=4.01$, $p=0.048$). Specifically, female participants who had eaten or drank within the last hour had significantly lower baseline cortisol levels than females who had not (M=0.14 μg/dL, SD=0.08; M=0.22 μg/dL, SD=0.18, respectively). No other variables that may impact hormone levels were related to baseline levels of cortisol in females. Therefore, for further cortisol analyses with female participants, baseline testosterone and whether the individual had eaten or drank anything within the last hour were controlled for.

In males, no other variables that may impact hormone levels were related to baseline levels of T. Therefore, for further T analyses with male participants, only baseline cortisol was controlled for. In females, baseline T was significantly associated with time of waking the morning of testing (r=0.22, p=0.037), and whether or not the participant had eaten or drank anything within the last hour [F(1,92)=5.38, p=0.023]. Specifically, female participants who had eaten or drank within the last hour had significantly lower baseline T levels than females who had not (M=40.39 pg/mL, SD=10.45; M=54.16 pg/mL, SD=19.94, respectively). Baseline T in females was significantly related to contraceptive use [F(1,92)=8.22, p=0.005], such that female participants using contraceptives had significantly lower levels of baseline T than female participants not using contraceptives (M=45.51 pg/mL, SD=15.52; M=56.92 pg/mL, SD=20.64, respectively). No other variables that may impact hormone levels were related to baseline levels of T in women. Therefore, for further T analyses with female participants, baseline cortisol, hormonal contraceptive use, time of waking, and whether or not the participant had eaten or drank anything in the last hour were controlled for.

Stress Manipulation and State Anxiety

Prior to the treatment/control session TSST and control participants did not differ in trait anxiety [STAI-T; t(149)=-0.094, p=0.925] or state anxiety [STAI-S; t(149)=-1.244, p=0.925]. After the manipulation, participants who underwent the TSST (M=45.20, SD=12.67) had significantly higher state anxiety levels than controls (M=31.38, SD=8.30): t(148)=-7.89,

$p<0.001$ (Figure 2.1). To control for baseline state anxiety levels, a change score between anxiety levels before and after the experimental manipulation was created. Participants who underwent the TSST (M=8.45, SD=11.43) had significantly higher state anxiety change scores than controls (M=-3.38, SD=6.99): $t(148)=-7.62$, $p<0.001$. These differences remain significant even when the effect of the experimental manipulation on state anxiety was analyzed separately in males and females.

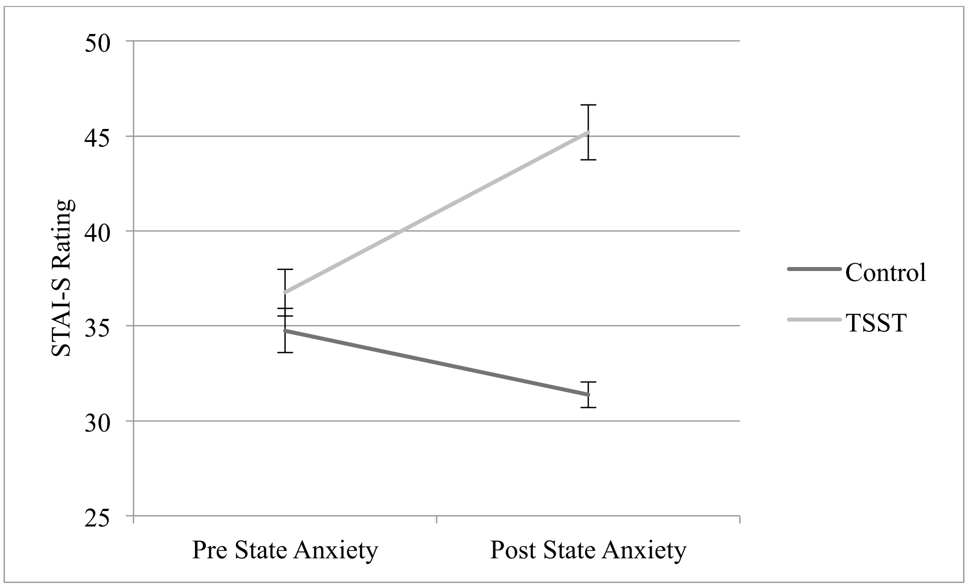

Figure 2.1. Subjective participant state anxiety measures before and after the control/treatment session as measures by the STAI-S. Error bars represent ± SEM.

Cortisol Response to TSST Manipulation

Males: A repeated measures, Greenhouse-Geisser corrected general linear model using cortisol concentration at the three time points (Time) as the within-subjects factor and TSST/control (Treatment) as the between-subjects factor revealed a significant main effect of Treatment [$F(1,52)=11.44$, $p=0.001$] and a significant interactive effect of Time and Treatment

[F(1.63,52)=9.59, p<0.001] (Figure 2.2). No significant main effect of Time was found. Post-hoc analyses showed that a) TSST and control participants did not differ in cortisol at baseline [F(1,55)=0.971, p=0.343], b) TSST males displayed higher cortisol levels when compared with control males at Timepoint two [F(1,55)=18.66, p<0.001], and c) TSST males displayed higher cortisol levels when compared with control males at Timepoint three [F(1,55)=12.58, p=0.001].

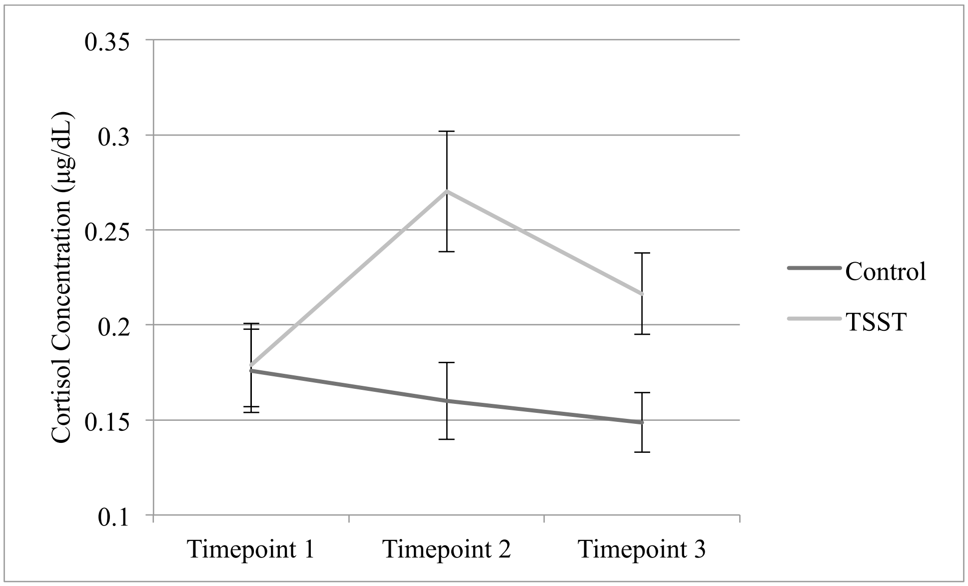

Figure 2.2. Cortisol concentrations (µg/dL) of male control and TSST participants' at three timepoints. Error bars represent ± SEM.

Delta cortisol responses were defined as the difference scores between cortisol levels at Timepoint three and Timepoint one. Mean (untransformed) delta cortisol responses for male control participants and male TSST participants were -0.027 µg/dL (SD=0.065) and 0.038 µg/dL (SD=0.13), respectively. A general linear model showed main effects of TSST [F(1,55)=5.60, p=0.02] (Figure 2.3), such that control participants had significantly smaller delta cortisol responses to the control task than TSST participants. These results indicate that the TSST was effective in inducing an increase in cortisol among male participants, while no significant cortisol change was observed among male controls.

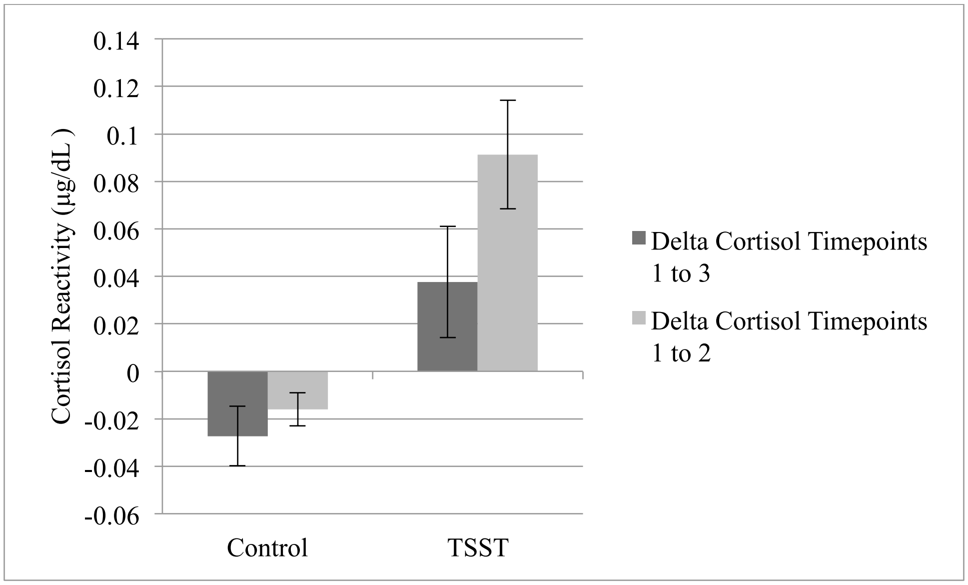

Figure 2.3. Cortisol reactivity (µg/dL) for control and TSST males, for both timepoints 1 to 3 and timepoints 1 to 2. Error bars represent ± SEM.

Females: A similar repeated measures, Greenhouse-Geisser corrected general linear model revealed a significant main effect of Treatment [F(1,89)=8.27, p=0.005] and a significant interactive effect between Time and Treatment [F(1.77,52)=4.69, p=0.014] (Figure 2.4). No significant main effect of Time was found. Post hoc tests showed that a) TSST and control participants did not differ in cortisol at baseline [F(1,92)=2.91, p=0.092], b) TSST females displayed higher cortisol levels when compared with control females at Timepoint two [F(1,92)=11.13, p=0.001], and c) TSST females displayed higher cortisol levels when compared with control females at Timepoint three [F(1,92)=8.88, p=0.004].

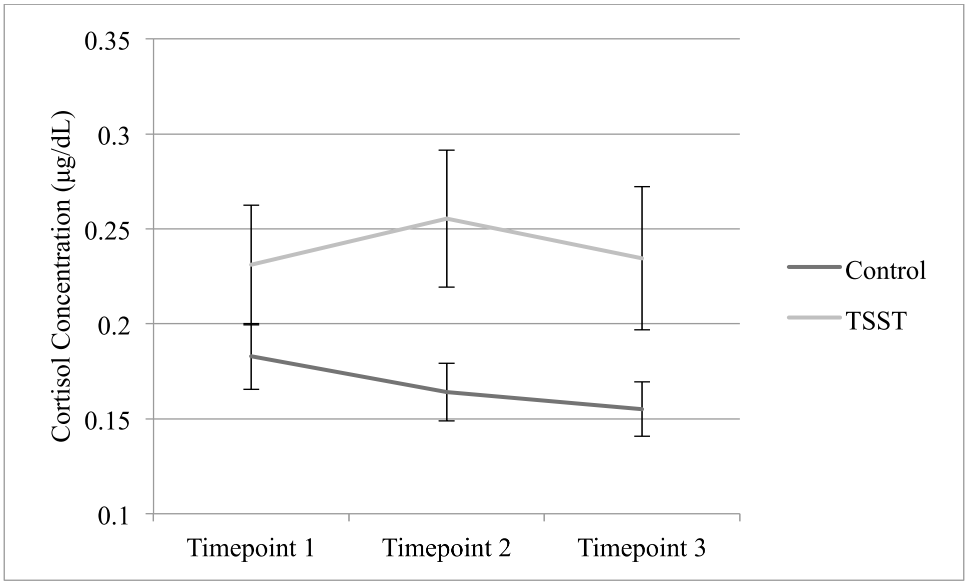

Figure 2.4. Cortisol concentrations (µg/dL) of female control and TSST participants' at three timepoints. Error bars represent ± SEM.

Mean (untransformed) delta cortisol responses for female control participants and female TSST participants were -0.02 µg/dL (SD=0.05) and 0.004 µg/dL (SD=0.12), respectively. A general linear model did not show main effects of TSST [$F(1,92)=2.40$, $p=0.125$], although control participants have smaller delta cortisol responses to the control task than TSST participants. However, mean (untransformed) delta cortisol responses calculated as the difference in cortisol levels from Timepoint two to Timepoint one for female control participants and female TSST participants were -0.03 µg/dL (SD=0.068) and 0.02 µg/dL (SD=0.10), respectively. A general linear model showed main effects of TSST [$F(1,92)=56.53$, $p=0.012$] (Figure 2.5), such that control participants had significantly smaller delta cortisol responses (Timepoint one to Timepoint two) to the control task than TSST participants. These results indicate that the TSST was effective in inducing an increase in cortisol among female participants, while no significant cortisol change was observed among female controls.

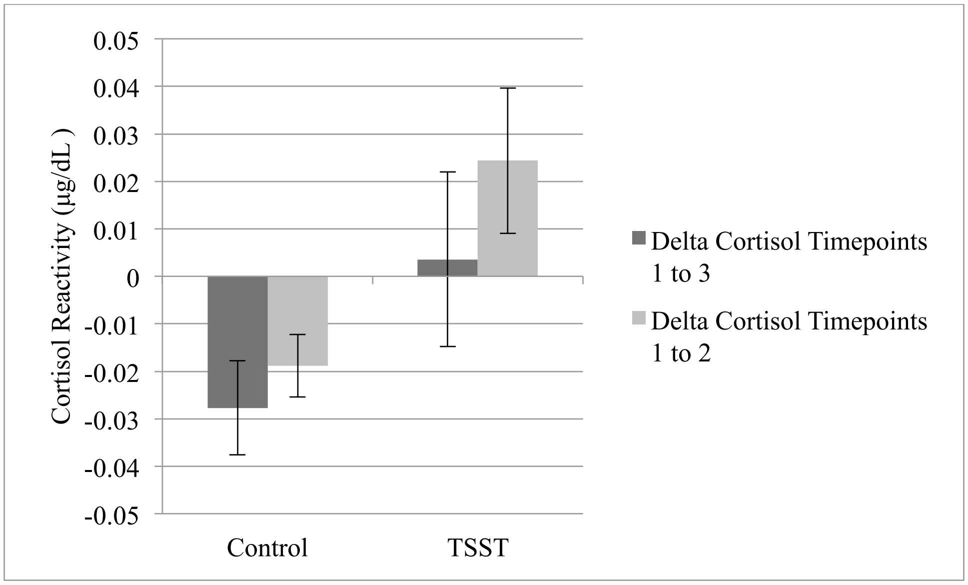

Figure 2.5. Cortisol reactivity (µg/dL) for control and TSST females, for both timepoints 1 to 3 and timepoints 1 to 2. Error bars represent ± SEM.

Testosterone Response to TSST Manipulation

Males: A repeated measures, Greenhouse-Geisser corrected general linear model using T at the first two time points (before and after the TSST or control manipulation) as the within-subjects factor and TSST/control as the between-subjects factor revealed no significant main effects of Time or Treatment in males. The model also revealed no significant interaction of Time and Treatment: $F(1, 53)=1.84$, $p=0.145$ (Figure 2.6). Post-hoc analyses showed that male control and male TSST participants did not differ in T levels at baseline [$F(1,55)=1.84$, $p=0.180$] or at Timepoint two after the stress/control manipulation baseline [$F(1,55)=0.58$, $p=0.450$.]

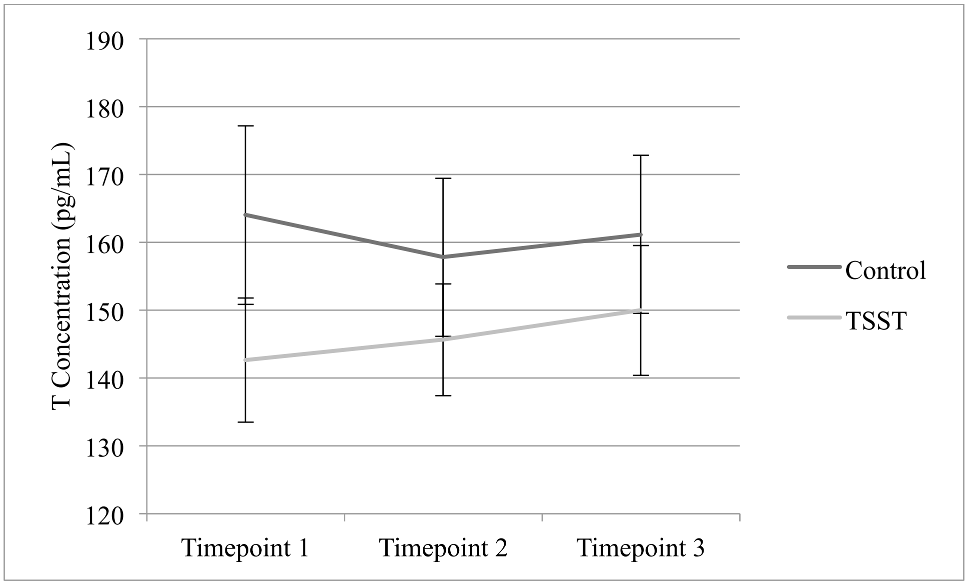

Figure 2.6. Testosterone (T) concentrations (pg/mL) of male control and TSST participants' at three timepoints. Error bars represent ± SEM.

Delta T responses pre and post TSST / control manipulation were defined as the difference scores between testosterone levels at Timepoint two and Timepoint one. Mean (untransformed) delta T responses for male control participants and male TSST participants were -6.23 pg/mL (SD=24.64) and 3.03 pg/mL (SD=20.61), respectively. A generalized linear model showed no main effects of TSST [Wald(1)=2.61, p=0.106], such that control participants did not have significantly smaller delta T responses to the control task than did TSST participants (Figure 2.7). Thus, the TSST did not have a significant effect on T levels in male participants.

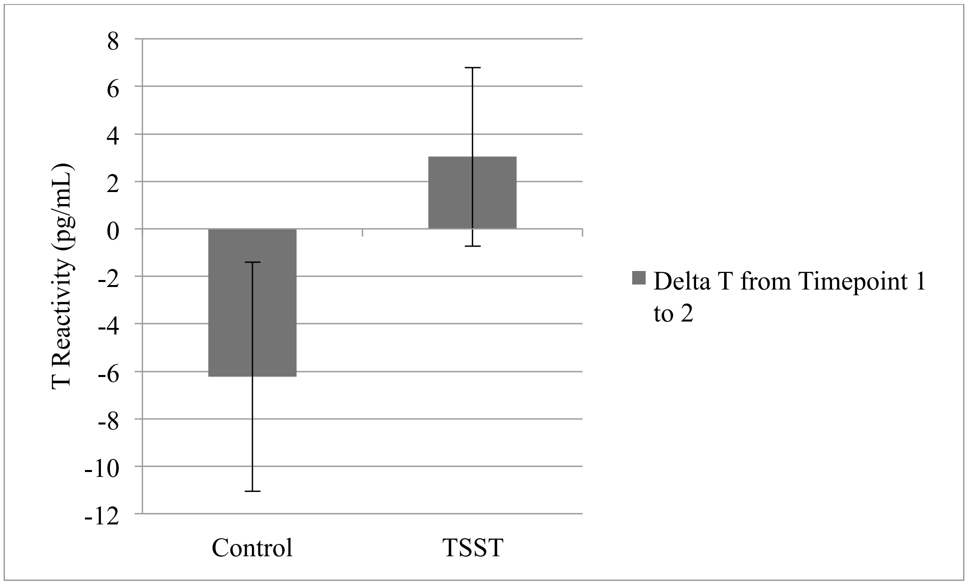

Figure 2.7. T reactivity (pg/mL) for control and TSST males, for timepoint 1 to 2. Error bars represent ± SEM.

Females: A similar analysis revealed no significant main effects of Time or Treatment on T in females, and no significant interaction of Time and Treatment: F(0.28,87)=0.147, p=0.839. Post-hoc analyses showed that female control and female TSST participants did not differ in T levels at baseline [F(1,92)=0.62, p=0.435] or at Timepoint two after the stress/control manipulation baseline [F(1,55)=0.38, p=0.541] (Figure 2.8).

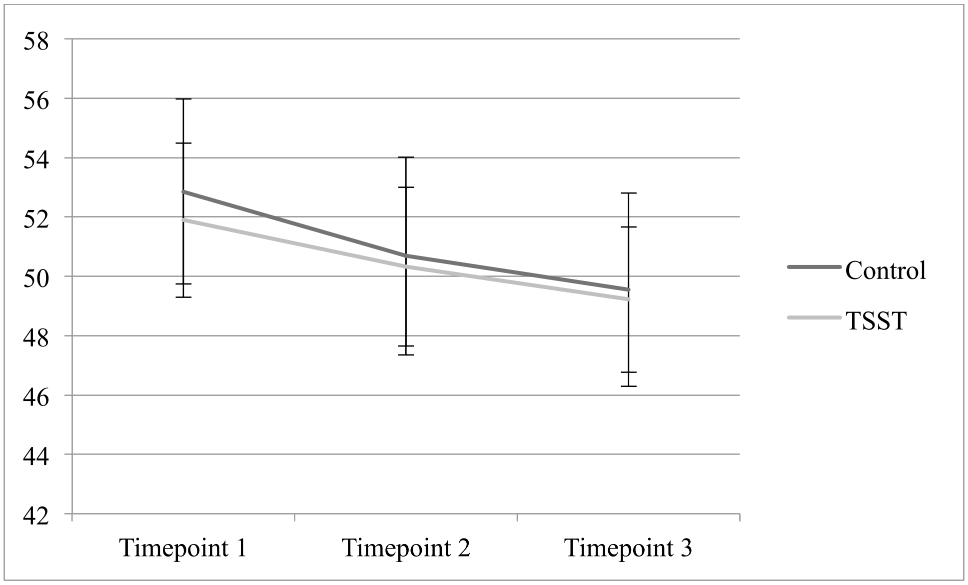

Figure 2.8. Testosterone (T) concentrations (pg/mL) of female control and TSST participants' at three timepoints. Error bars represent ± SEM.

Mean (untransformed) delta T responses for female control participants and female TSST participants were -2.18 pg/mL (SD=7.61) and -1.57 pg/mL (SD=8.43), respectively. A general linear model showed no main effects of TSST [$F(1,92)=0.003$, $p=0.953$], such that control participants did not have significantly smaller delta T responses to the control task than did TSST participants (Figure 2.9). Thus, the TSST did not have a significant effect on T levels in female participants.

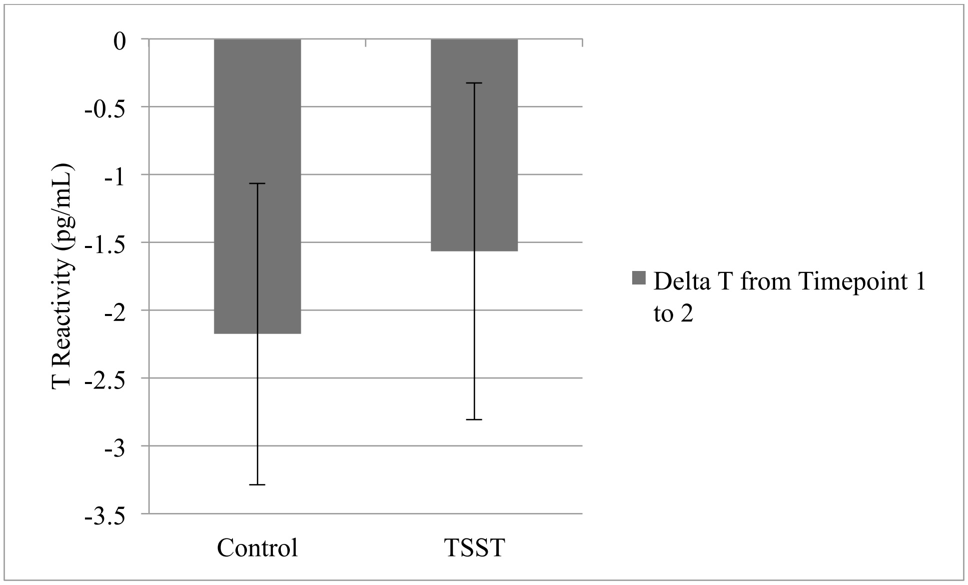

Figure 2.9. T reactivity (pg/mL) for control and TSST females, for timepoint 1 to 2. Error bars represent ± SEM.

Relationship Between Math Anxiety and Hormonal Reactivity to the TSST

To test the relationship between trait math anxiety subjective ratings and subjective anxiety and/or hormonal reactivity for TSST participants, bivariate correlations and multiple linear regression models were run for TSST participants only. For both TSST males and females, math anxiety was not significantly related to change in subjective state anxiety (STAI-S) ratings or cortisol reactivity before and after the TSST. For TSST males, math anxiety was also not significantly related to testosterone reactivity before and after the TSST.

However, for females who underwent the TSST, math anxiety ratings (sMARS) were a significant predictor of testosterone reactivity after the TSST (but before the social interaction). In a multiple regression model that controlled for time since waking, hormonal contraception, whether or not the female participant had eaten that morning, and baseline cortisol (log transformed), math anxiety was a significant predictor of delta T ($\beta = 0.357$, $t = 2.31$, $p = 0.26$).

Specifically, math anxious female participants who underwent the TSST experienced higher increases in testosterone during the TSST than less math anxious females did (Figure 2.10).

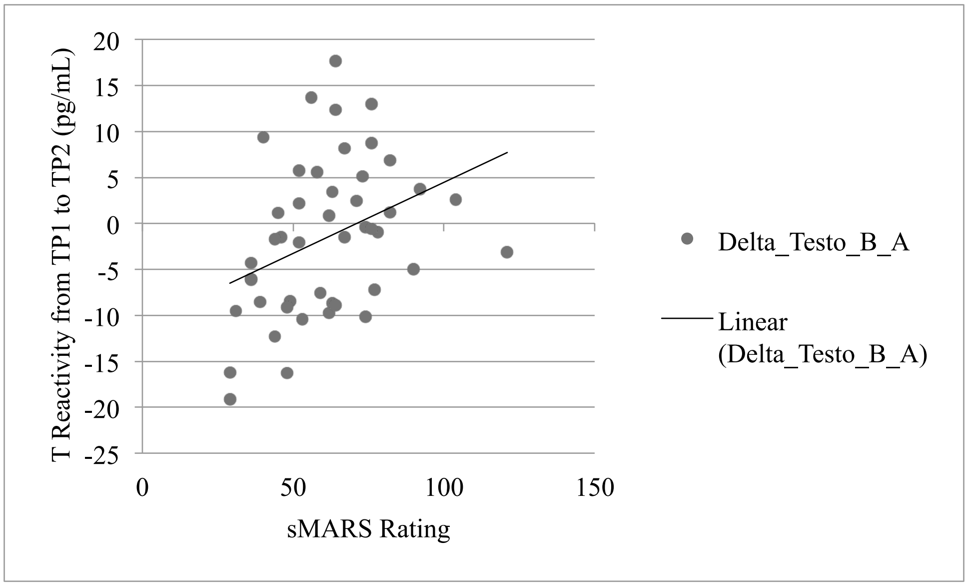

Figure 2.10. Relationship between subjective math anxiety (sMARS) and testosterone (T) reactivity before and after the TSST in TSST females.

Cortisol Response to Social Interaction

Males: Change scores are presented separately within each condition since we hypothesized and found significant differences in cortisol levels at the second timepoint (post TSST manipulation) between control and TSST condition participants (Table 2.3). Average change from the second sample to the third sample (before and after the social interaction) was +2.16% in the control condition [raw baseline (second sample) M=0.160 μg/dL, SD=0.105; third sample M=0.149 μg/dL, SD=0.082], and -10.87% in the TSST condition [raw baseline (second sample) M=0.270 μg/dL, SD=0.174, third sample M=0.216 μg/dL, SD = 0.118]. To check for any observed differences in cortisol levels associated with the social interaction in control participants, paired t tests on the log transformed data revealed no significant increase in cortisol from timepoints two to three in the control condition: t(26)=0.74, p=0.47. Therefore, in all control male participants, there was not an overall significant change in cortisol levels before and after the social interaction.

	% Change from TP2 to TP3	**TP2 Raw Baseline (µg/dL)**	**TP 2 SD**	**TP3 Raw Baseline (µg/dL)**	**TP3 SD**
Control	+2.16%	0.160	0.105	0.149	0.082
TSST	-10.87%	0.270	0.174	0.216	0.118

Table 2.3. Cortisol reactivity of male control and TSST participants before and after the social interaction task.

However, data depicted in Figure 2.11 suggest that female relationship status in control participants may have influenced cortisol reactions to the social interaction. Although relationship status was significantly related to baseline cortisol at timepoint one [F(1,29)=6.67, p=0.015] and timepoint 2 [F(1,29)=5.64, p=0.025] before the social interaction in TSST males, there was no significant relationship between relationship status and baseline cortisol at timepoint one [F(1,26)=1.17, p=0.29] or timepoint two [F(1,29)=1.07, p=0.31] in control males. Mean (untransformed) delta and percent change cortisol responses (timepoint two to timepoint three) for male single participants and male participants who were in a relationship were 0.0018 µg/dL (SD=0.063) / 16.55% (SD=54.66) and -0.028 µg/dL (SD=0.044) / -15.82% (SD=25.40), respectively. A general linear model controlling for cortisol and testosterone at timepoints one and two showed main effects of relationship status [F(1,20)=16.05, p=0.001] (Figure 2.11), such that single male control participants had significantly larger delta cortisol responses (Timepoint two to Timepoint three) to the social interaction task than male control participants who were in a relationship at the time. These results indicate that the social interaction was associated with an increase in cortisol among male single participants, while no such increase was observed in control males who were in a relationship.

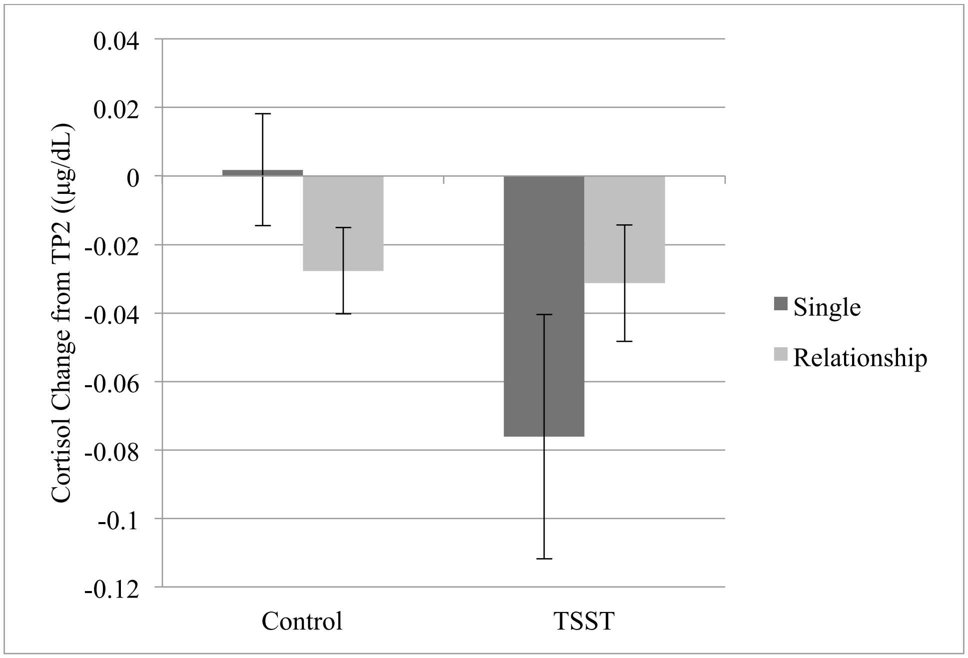

Figure 2.11. Cortisol reactivity (µg/dL) for control and TSST males, for timepoint 2 to 3. Error bars represent ± SEM.

Females: Average change from the second sample to the third sample (before and after the social interaction) for females was +0.98% in the control condition [raw baseline (second sample) M=0.164 µg/dL, SD=0.104; third sample M=0.155 µg/dL, SD=0.098], and -6.15% in the TSST condition [raw baseline (second sample) M=0.255 µg/dL, SD=0.245, third sample M=0.235 µg/dL, SD = 0.256] (Table 2.4). To check for any observed differences in cortisol levels associated with the social interaction in female control participants, paired t tests on the log transformed data revealed no significant increase in cortisol from timepoints two to three in the control condition: $t(46)=0.95$, $p=0.35$. Therefore, in female control participants, there was not a significant change in cortisol levels before and after the social interaction.

	% Change from TP2 to TP3	**TP2 Raw Baseline (µg/dL)**	**TP 2 SD**	**TP3 Raw Baseline (µg/dL)**	**TP3 SD**
Control	+0.98%	0.164	0.104	0.155	0.098
TSST	-6.15%	0.255	0.245	0.235	0.256

Table 2.4. Cortisol reactivity of female control and TSST participants before and after the social interaction task.

However, likewise as with control males, data depicted in Figure 2.12 suggest that female relationship status in control participants may have influenced cortisol reactions to the social interaction. Mean (untransformed) delta cortisol responses (timepoint two to timepoint three) for female single participants and female participants who were in a relationship were 0.0045 µg/dL (SD=0.062) / 8.44% (SD=36.65) and -0.027 µg/dL (SD=0.040) / -9.09% (SD=36.56), respectively. A general linear model controlling for cortisol and testosterone at timepoints one and two showed main effects of relationship status [$F(1,40)=5.60$, $p=0.02$] (Figure 2.12), such that single female control participants had significantly larger percent change cortisol responses (Timepoint two to Timepoint three) to the social interaction task than female control participants who were in a relationship at the time. These results indicate that the social interaction was associated with an increase in cortisol among female single participants, while no such increase was observed in control females who were in a relationship.

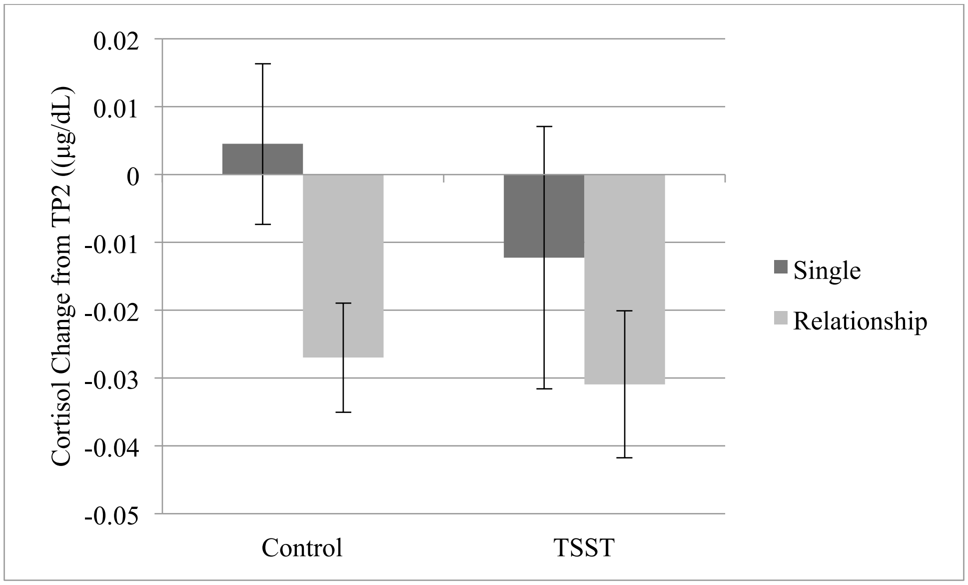

Figure 2.12. Cortisol reactivity (µg/dL) for control and TSST females, for timepoint 2 to 3. Error bars represent ± SEM.

Testosterone Response to Social Interaction

Males: Change scores are presented separately within each condition since we hypothesized an increase in T levels in the control condition (i.e., individuals who did not go through the stress manipulation before interacting with an opposite sex confederate) but were agnostic about possible reactive changes in the TSST group (Table 2.5). Average change in males from the second sample to the third sample (before and after the social interaction) was +2.68% in the control condition [raw baseline (second sample) M=157.12 pg/mL, SD=58.38; third sample M=161.17 pg/mL, SD=59.33], and +2.50% in the TSST condition [raw baseline (second sample) M=145.66 pg/mL, SD=45.23, third sample M=149.98 pg/mL, SD = 52.48]. To check for any observed differences in T levels associated with the social interaction in control participants, paired t tests on the log transformed data revealed no significant increase in T from timepoints two to three in the control condition: $t(25)=-1.21$, $p=0.24$. Likewise, for male TSST participants, paired t tests on the log transformed data revealed no significant increase in T from timepoints two to three: t(29)=-0.70, p=0.49. Therefore, in all control male participants, there was not an overall significant change in T levels before and after the social interaction, and in all TSST participants, there was not an overall significant change in T levels before and after the social interaction.

	% Change from TP2 to TP3	**TP2 Raw Baseline (pg/mL)**	**TP 2 SD**	**TP3 Raw Baseline (pg/mL)**	**TP3 SD**
Control	+2.68%	157.12	55.38	161.17	59.33
TSST	+2.50%	145.66	45.23	149.98	52.48

Table 2.5. Testosterone (T) reactivity of male control and TSST participants before and after the social interaction task.

A general linear model using change in T from timepoint two to timepoint three as the dependent variable and relationship status was used to analyze the relationship between relationship status and testosterone reactivity during the social interaction in control males. Controlling for T at timepoints one and two, the model showed main effects of relationship status [F(1,25)=5.58, p=0.03], such that control male participants who were single had significantly larger delta T responses to the social interaction task (M=9.21 pg/mL, SD=14.05) than control male participants who were in a relationship (M=-3.46 pg/mL, SD=11.44; Figure 2.13). This relationship between relationship status and change in T was not significant in males who had gone through the TSST [F(1,29)=1.63, p=0.21]. Thus, relationship status may have influenced T reactivity during the social interaction, such that single males show an increase in T while males in a relationship do not, but this relationship only exists in male participants who did not go through the stress manipulation.

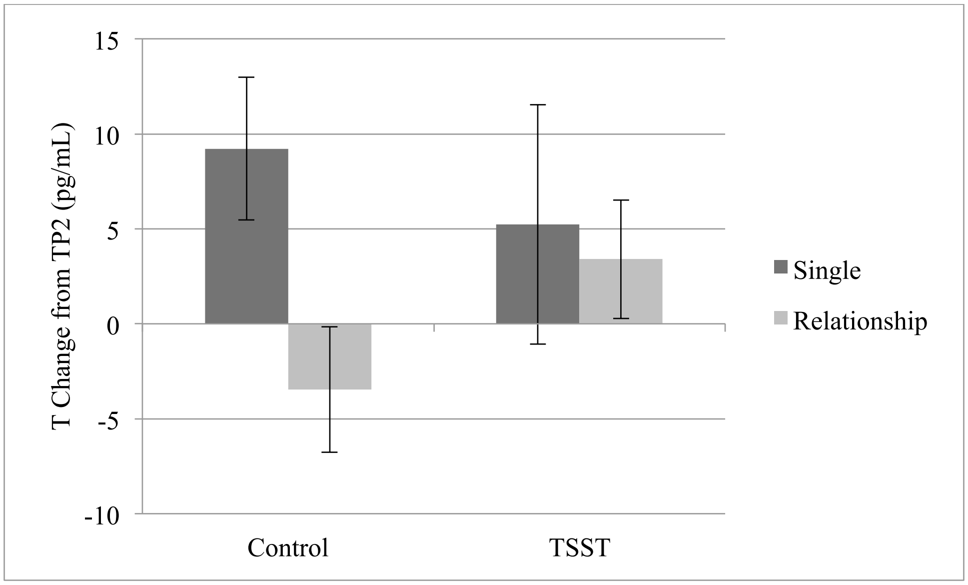

Figure 2.13. Testosterone (T) reactivity (pg/mL) for control and TSST males, for timepoint 2 to 3. Error bars represent ± SEM.

<u>Females:</u> Again, change scores in females are presented separately within each condition since we hypothesized an increase in T levels in the control condition (i.e., individuals who did not go through the stress manipulation before interacting with an opposite sex confederate) but were agnostic about possible reactive changes in the TSST group (Table 2.6). Average change in females from the second sample to the third sample (before and after the social interaction) was +2.66% in the control condition [raw baseline (second sample) M=50.68 pg/mL, SD=22.84; third sample M=49.55 pg/mL, SD=22.38], and -0.53% in the TSST condition [raw baseline (second sample) M=50.33 pg/mL, SD=18.16, third sample M=49.22 pg/mL, SD = 16.66]. To check for any observed differences in T levels associated with the social interaction in control participants, paired t tests data revealed no significant increase in T from timepoints two to three in the control condition: $t(46)=1.21$, $p=0.27$. Likewise, for female TSST participants, paired t tests revealed no significant increase in T from timepoints two to three: $t(45)=0.89$, $p=0.38$. Therefore, in all control female participants, there was not an overall significant change in T

levels before and after the social interaction, and in all TSST participants, there was not an overall significant change in T levels before and after the social interaction.

	% Change from TP2 to TP3	**TP2 Raw Baseline**	**TP 2 SD**	**TP3 Raw Baseline**	**TP3 SD**
Control	+2.66%	50.68	22.84	49.55	22.38
TSST	-0.53%	50.33	18.16	49.22	16.66

Table 2.6. Testosterone (T) reactivity of female control and TSST participants before and after the social interaction task.

A general linear model using change in T from timepoint two to timepoint three as the dependent variable and relationship status was used to analyze the relationship between relationship status and testosterone reactivity during the social interaction in control females. Controlling for T at timepoints one and two, the model showed main effects of relationship status [F(1,46)=6.60, p=0.01], such that control female participants who were single had significantly larger delta T responses to the social interaction task (M=0.78 pg/mL, SD=7.43) than control female participants who were in a relationship (M=-3.71 pg/mL, SD=6.98; Figure 2.14). This relationship between relationship status and change in T was not significant in females who had gone through the TSST [F(1,45)=0.56, p=0.46]. Thus, relationship status may have influenced T reactivity during the social interaction such that single females show an increase in T while females in a relationship do not, but this relationship only exists in female participants who did not go through the stress manipulation.

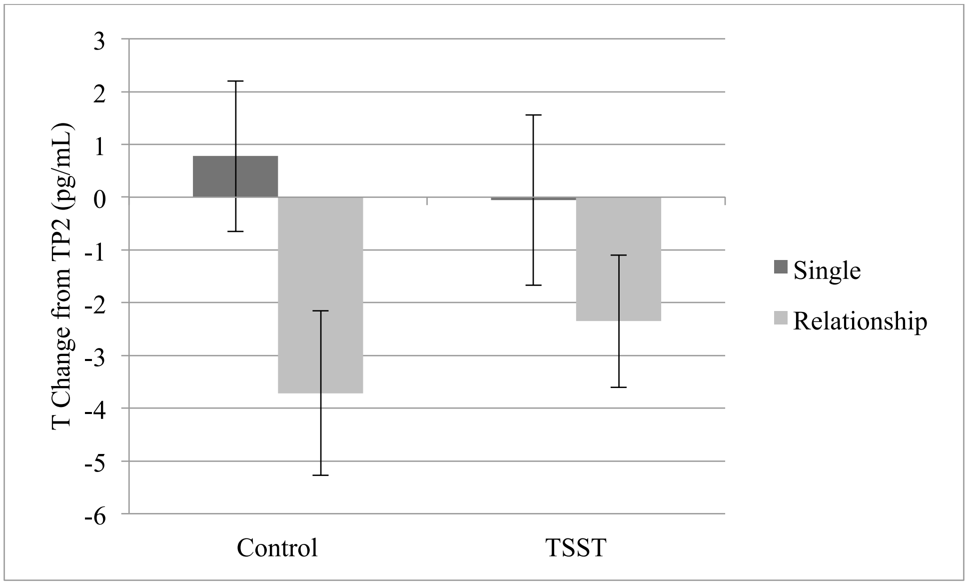

Figure 2.14. Testosterone (T) reactivity (pg/mL) for control and TSST females, for timepoint 2 to 3. Error bars represent ± SEM.

Discussion

Among all control male and female participants, we did not find significant increases in testosterone after taking part in a brief conversation with a confederate of the opposite sex. However, we did find a significant difference in testosterone reactivity between single individuals and individuals who are in a relationship. Specifically, single individuals had significantly more positive testosterone changes after the social interaction compared to individuals who were in a relationship. This relationship was found in both male and female participants, though single males had more positive changes in testosterone after interacting with a female confederate. These results are consistent with those of previous studies showing that in single men, verbal interactions with women (of less than five or ten minutes) can induce increases in testosterone (Roney et al. 2003; van der Meij et al. 2008).

We found the same relationship in terms of cortisol reactivity. First, control males and females did not show significant differences in cortisol levels before and after the social interaction test. However, similar to testosterone reactivity, there is a difference between individuals who are single and individuals who are in a relationship in terms of cortisol reactivity during the social interaction task. In particular, singles show significantly more positive increases in cortisol during the social interaction when compared to individuals who are in a relationship. These results are consistent with those of previous studies in suggesting that cortisol increases could reflect anxiety or apprehension before or during opportunities for courtship (Roney et al., 2007, 2009; van der Meij et al., 2010). Roney et al. (2007) found that changes in cortisol from baseline were significantly greater among male participants who interacted with female confederates relative to males in control conditions. Similar to our study, Roney et al. (2007) and van der Meij et al. (2010) found that singles showed greater hormonal reactivity when compared to individuals in relationship.

In this study, the TSST was effective in increasing both subjective state anxiety and cortisol concentrations in male and female participants. The data presented here suggest that, although the TSST may not affect testosterone concentration directly, the TSST may have a psychological effect that may later affect physiological sensitivity to a social interaction. Unlike controls, in individuals who underwent the TSST and had significantly higher cortisol baseline concentrations at the beginning of the social interaction, relationship status had no effect on testosterone reactivity. The same phenomenon holds true for cortisol reactivity during the social interaction. That is, for individuals assigned to the TSST, the significant difference between singles and those in a relationship in cortisol reactivity during the social interaction no longer exists.

These results are consistent with the conclusions of a recent review article by Zilioli and Bird (2017). After reviewing literature on testosterone reactivity (primarily in human males), the authors conclude that both motivational factors and situational factors have the ability to influence the relationship between evolutionarily salient social contexts, such as an interaction with a potential mate, and physiological reactivity to these contexts (Zilioli & Bird, 2017). Differences in relationship status could, at least in part, reflect differences in socio-sexual motivation. A situational factor, which the authors define as an external factor outside of the control of the individual, also has the ability to influence the relationship between ecological cue and physiological response (Zilioli & Bird, 2017). In terms of the data here, psychosocial stress may have interfered with the physiological response to social stimuli, which explains why the increase of testosterone and cortisol in single individuals no longer differs when compared to individuals in a relationship for TSST participants.

Finally, one particularly interesting finding in terms of individual difference effects on hormonal reactivity was found in females who were assigned to the TSST stress condition. Specifically, there was a significant relationship between female self-reported math anxiety and testosterone reactivity during the TSST. Females who were math anxious had more positive changes in testosterone reactivity from timepoint one to timepoint two (during the TSST) than females who were less math anxious. This relationship was not found in male participants who underwent the TSST. This finding may have important implications for the use of the TSST. As slight modifications in the timing or durations of different components of or in the confederates involved in a psychosocial stress-inducing procedure have been shown to generate significant variation in the cortisol and/or testosterone response (Andrews et al. 2007; Wadiwalla et al., 2010; Duchesne et al., 2012; Liu et al., 2017; Kim et al., 2018), it is possible that slight variations in trait or math anxiety in participants may affect cortisol or testosterone response as well.

Overall, this study confirms the importance of individual differences in relationship status (a proxy for motivation) and situational variables as factors in moderating hormonal reactivity to a potential courtship interaction. Further, it appears that psychosocial stress, while not affecting testosterone reactivity during a social interaction directly, plays a role in altering how single males and females react to a social interaction interpreted as a potential courtship opportunity. While the exact mechanism remains unclear, it is possible that psychosocial stress may subdue the adaptive physiological response (increase in testosterone) in individuals who are engaged in a courtship. However, this relationship is only relevant in individuals who are single, supporting the idea that motivational factors of relevant relationship status act as a moderating factor in testosterone's role in potentially inducing an adaptive courtship behavioral response. Future studies should continue to focus on the potential moderating motivational factors, whether biological, sociological, or psychological in nature, as there is growing evidence that these factors have the potential to influence the relationship between evolutionarily salient social contexts (e.g., interaction with mates) and adaptive neuroendocrine fluctuations that follow.

CHAPTER 3

Relationship between stress, social interaction, and cognition in men and women

Abstract

Cognitive resources are necessary for impression management, for advertisement of psychological traits that signal strengths as a potential courtship partner (e.g. intelligence, personality, humor), and for use of social cognition skills that allow one to read the interpretations and behaviors of a partner. All of these may have implications for how cognition can affect social behavior during a social interaction of potential courtship. Further, as stress and apprehension have the ability to affect cognition, it is also possible that under evaluative stress, cognitive resources are being diverted in a way that is specifically adaptive or specifically maladaptive during courtship. This study investigated the effects of a brief social interaction that could be potentially viewed as a potential courtship opportunity on cognitive performance in men and women, as well as the effect of psychosocial stress on cognitive performance after psychosocial stress and potential cognitive changes that occur during a courtship opportunity. Results did not replicate previous findings that cognitive performance decreased following a brief social interaction with an opposite sex confederate. Further, psychosocial stress was not found to affect cognitive performance. Instead, higher subjective anxiety changes and higher increases in cortisol following acute stress led to improved cognitive performance. Further research focusing on the specific context of stress is needed to clarify how and if stress affects cognition by improving or decreasing attention, particularly in terms of social situations that induce either ecological cues of courtship and/or cues of psychosocial stress and evaluative threat.

Introduction

Human courtship and cognition

One potential mediator of the relationship between cues of courtship and the output of resulting courtship behavior is cognition. A model of courtship cognition would involve specifying the relationship between input cues and psychological cognitive changes related to attraction. Social interactions may affect an individual's cognitive functioning based on whom the interaction partner is and what psychological processes take place during the interaction. During courtship interactions, it is possible that cognitive resources could be diverted in a way that is specific; whether this potential diversion is adaptive or maladaptive may depend on individual differences in cognitive performance or susceptibility to courtship apprehension. An interesting question is that of the potential ability of courtship cues to prime activation of psychological constructs for courtship and mating, and what cognitive psychological changes mediate the relationship courtship opportunities presented, endocrine function, and behavioral responses during courtship.

Cognitive resources are necessary for impression management, for advertisement of psychological traits that signal strengths as a potential courtship partner (e.g. intelligence, personality, humor), and to read the interpretations and behaviors of a partner. Some recent studies that have explored the effects of opposite sex social interactions on cognition; these studies have primarily been done with male participants or have only found effects in male participants (Roney, 2003; Karremans et al., 2009; Nauts et al., 2012). However, these studies do provide evidence of the importance of cognition in ways that fit with the framework of the need for cognitive resources for both impression management and social cognition.

Social cognition may be involved in that ecological cues relevant to courtship opportunities may prime courtship-related changes in motivation to interpret others' behavior (e.g. empathy, read the mind of others') or changes in self perception. For example, there is some evidence that individuals alter how they present their own attitude to more closely mirror attitudes or preferences of attractive members of the opposite sex (Morier & Seroy, 1994; Zanna & Pack, 1975). In two experiments, Roney (2003) found that visual exposure to women has the ability to prime changes in attitude, mood, and personality trait descriptions in male participants. The fact that these effects were found without participant awareness suggest a model in which input cues from potential courtship can prime psychological representations and alter courtship behaviors subconsciously (Roney, 2003).

Impression management may also be important in courtship. During social interactions, individuals need to exert cognitive effort to strategically control their behavior and monitor the impression they make to other individuals (Vohs et al., 2005); therefore, courtship situations where individuals are strategically working to impress potential mates can certainly become an interaction that is cognitively taxing. Some studies have shown effects of social interactions on cognition in specific directions in terms of cognitive impairment. Karremans et al. (2009) demonstrate that men's, but not women's cognitive performance declined following opposite sex social interactions, and the effect occurred more strongly when men reported their interaction partners as more attractive. Nauts et al. (2012) suggest that actual interaction is not even a necessary requirement for cognitive impairment to occur; they found that men's (but not women's) cognitive performance declined after they were led to believe that they would interact with an opposite sex confederate via a computer.

There are potential moderating factors that may affect the relationship between cognitive function and courtship behavior. Cognitive impairment effects during or after social interactions may be particularly strong if one's self-presentational concerns are higher, both in terms of personality and in terms of one's impression of the partner (e.g., if one perceives the partner as more attractive). As men engage less in impression management if a woman is perceived as having a low mate value (Wilson & Daly, 2004), it is possible that cognitive resources are more quickly drained during courtship situations that are more ecologically valid or involve higher stakes in terms of perceived partner mate value. Another possible moderating role is that of romantic relationship status. Romantically involved men or women would engage in less cognitive effort toward self-presentation due to lowered motivation to seek a potential mate (see: Karremans & Verwijmeren, 2008; Simpson, et al., 1990). However, relationship status did not predict change in cognitive functioning following a social interaction (Karremans et al., 2009). It is possible that men may be interested in short-term mates regardless of their relationship status. Finally, ecologically valid courtship opportunities may cause stronger effects in men than in women, and lead to strong effects of cognitive impairment in men, based on research that has suggested that men are more likely than women to consider mixed-sex interaction in terms of a mating game. As men have been shown to be more likely to look for sexual interest in the behavior of opposite-sex individuals and to overestimate sexual interest of women (Abbey, 1982; Shotland & Craig, 1988), men might be overall more eager to engage in cognitively demanding efforts to impress a potential mate.

Human courtship, stress reactivity, and cognition

It is possible that cognition may play an important mediating role in terms of adaptive behavioral strategies during courtship. If stress and apprehension have the ability to affect cognition, we can ask the question: are cognitive resources being diverted by stress in a way that is specifically adaptive or specifically maladaptive during courtship? If individuals differ in stress responsivity, it is possible that they may differ in terms of cognitive performance as well.

Human and nonhuman animal studies have well documented the phenomenon of chronic exposure to stress having an impact on brain structures involved in cognitive function (Lupien et al., 2009; McEwen & Sapolsky, 1995; Sandi, 2013). A 2007 review assessing the effects of stress and stress hormones on human cognitive performance discusses specifically the effects of an exposure to an acute stressful situation and subsequent endogenous hormonal changes on cognition (Lupien et al. 2007). Similar to exogenous administration of glucocorticoids, exposure to psychosocial stress and an endogenous increase of glucocorticoids will induce cognitive impairments, particularly influencing learning, memory, and cognitive functions sustained by the frontal lobes and the hippocampus, the two brain regions which contain the highest concentrations of glucocorticoid receptors (Lupien et al., 2007). Although corticosteroids can be essential for cognitive performance in that they have the ability to protect the brain against adverse events, they also have shown the ability to damage and disrupt memory and other cognitive functions (de Kloet et al., 1999).

Age and gender have been found to be influential factors in terms of the individual differences in the impact of stress on cognition (Helbig & Backhaus, 2017; Sandi, 2013). Smeets et al. (2009) tested the differential effects of stress in men and women explicitly looking at the effects on social cognition, as humans often have to perform complex social decision making while under stress (e.g., social conflicts). They found differential effects among men and women for the Movie for the Assessment of Social Cognition (MASC), but not for the Reading the Mind in the Eyes Test (RMET); high cortisol-responding men had elevated MASC scores, while only low-cortisol responding women had elevated MASC scores. As social cognition may be important during courtship interactions, this is important evidence concerning the relationship between stress, cognition, and courtship behavior.

Many of the studies that have examined the effects of psychosocial stress on cognition have utilized the Trier Social Stress Test (TSST; Kirschbaum et al. 1993). Kuhlmann et al. (2005) reported that the TSST led to impaired memory retrieval in male participants. More recent research suggests that the TSST may interact with specific task cognitive load and stimulus type, as opposed to uniformly affecting memory in a negative way (Guez et al., 2016). These TSST studies have also reported gender and baseline behavioral differences in response to the TSST and have broad implications for differences in emotional and physiological responses to the TSST (Abelson et al., 2014; Kelly et al., 2008; Abelson et al., 2014; Kirschbaum et al., 1996; Wolf et al., 2001). These studies also have implications for cognitive stress management training. Gaab et al. (2003) found that group-based cognitive-behavioral stress management training reduced neuroendocrine responses to the TSST, implying that this type of training may prove useful to prevent the negative effects of acute psychosocial stress.

Attentional resources, attentional biases, and similar core executive functions have been shown to be affected by psychosocial stress, in addition to memory (Chajut & Algom, 2003; Jamieson et al., 2012; Pilgrim et al., 2014; Sato et al., 2012; Shields et al., 2016). Previous research, however, has revealed a number of inconsistencies in terms of how specifically acute stress influences core executive functions and selective attention. Because selective attention has the potential to be improved by increased perceptual load due to the exhaustion of cognitive resources (leaving less resources available for distractor processing), it is still unclear whether acute stress can have a similar effect as perceptual load, leading to cognitive benefits (Sato et al., 2012). Sato and colleagues (2012) found that perceptual load interacted with a stress manipulation, such that a high perceptual load removed cognitive interference for control participants, whereas high perceptual load and increased anxiety interacted to increase cognitive interference in stressed participants. Others have shown that stress may improve selectivity of attention or help to remove attentional bias (Chajut & Algom, 2003; Jamieson et al., 2012). Clearly, further research, particularly focusing on the specific context of stress, is needed to clarify how and if stress affects cognition by improving or decreasing attention and altering consequent decision making.

Overall, as individual differences in cognitive function, personality, and stress responsivity may lead to different strategies that are utilized during a courtship opportunity, triggered psychosocial apprehension and stress may affect cognition and also dampen the physiological changes that normally occur to aid courtship behavior (e.g., increase in testosterone). One potential example is that extroverted individuals may not experience psychosocial apprehension that is triggered by evaluation by a potential mate, while introverted individuals may have to deal with this psychosocial apprehension by diverting cognitive resources (Wilson et al., 2015; Ponzi et al., 2016). Another potential example is that of individual differences in social status. As subjective social status has been found to moderate cortisol responses to social evaluative threat (Gruenewald et al., 2006), it is possible that individuals who have high self confidence or consider themselves to have high social status may not experience a suppression in cognitive performance caused by anxiety. By measuring cognitive abilities before and after a potential courtship interaction , both with and without a previous stressful manipulation, we may be able to better understand what differing behavioral strategies exist and how they are used differently across personalities, gender, and abilities.

Aims / Hypotheses

Aim 1: Assess the relationship between cognitive performance and courtship behavior

In order to measure how cognition might mediate the relationship between cues of courtship and resulting courtship behavior, I used a cognitive task to measure cognitive performance prior to and after a brief social interaction. Because cognitive resources may be exhausted during courtship interactions in order to result in positive courtship behaviors, I expected that both male and female cognitive performance will be impaired following a courtship interaction. As impression management and social cognition required for courtship behavior may create a cognitive load, I expected cognitive impairment to be highest in individuals who both rate their interaction partner as more attractive and in individuals who are rated to show higher courtship behaviors (e.g. increased eye contact, increased conversation orientation, etc.).

Aim 2: Investigate the effects of psychosocial stress on cognitive performance before and after a social interaction

We would not expect courtship behavior in humans to evolve independent of ecological stress, and courtship interactions involve a social evaluative threat that may cause significant psychosocial stress. Therefore, we may expect psychosocial stress to potentially dampen self-confidence, motivation, and approach behaviors, as well as affect cognition in a way that may hinder social cognition and impression management during courtship. I expected that: 1) psychosocial stress will result in decreased cognitive performance after stress as compared to individuals who do not take part in a stress treatment; 2) cognition may be impaired most dramatically following stress for individuals with high responding cortisol overall, therefore leading to less courtship behavior due to impairment of social cognition and impression management. However, for individuals who experience a lesser increase in cortisol following stress, I predicted that stress may improve selectivity of attention, and this in turn may facilitate the expression of courtship behavior.

Methods

Study Design / Overview

Participants were randomly assigned prior to arrival to either a control or psychosocial stress condition (Trier Social Stress Test), and those assigned to the stress condition were also semi-randomly assigned to interact with either same-sex or opposite-sex judges during the Trier Social Stress Test (TSST), based on researcher availability. Upon arrival at the lab, participants provided consent, filled out questionnaires (see below), and participated in either the control or TSST condition. Following the assigned condition, all subjects went through a social interaction with an opposite-sex confederate. Finally, participants engaged in a phased debriefing process. Following full debriefing, participants were compensated and the experimental session ended. Saliva samples were collected at various time points throughout the tasks to assess hormone levels via ELISA: once before the control/TSST condition, once after the control/TSST condition, and once after the social interaction. Cognitive tasks were also administered throughout the protocol, in conjunction with the collected saliva samples. The Social Science Institutional Review Board at the University of Chicago had approved all experimental procedures (IRB #12-1251).

Participants

Participants were approximately 120-140 heterosexual individuals (age range: 18-35 years) from the greater Chicago area, surrounding the University of Chicago campus. All participants were recruited through fliers, UChicago Marketplace, and/or a human subject recruitment website (Sona System). Data was collected continuously throughout fourteen montsh, from July 2017 through August 2018. All study participants completed a digital prescreen consent form and eligibility questionnaire via Qualtrics software, a digital main protocol consent form and questionnaire via Qualtrics software, and also a written informed consent form at the laboratory before participating in the study. All participants were paid $20 compensation after completion of the procedures: $15 as base pay, and $5 as "winnings" from the decision making tasks.

Experimental Procedure

All experimental procedures took place at the Behavioral Biology Laboratory, located in the Biopsychological Sciences Building (940 E. 57th Street) at the University of Chicago. Prior to arrival at the lab, participants consented to a prescreen questionnaire administered via Qualtrics software. If eligible, participants consented to the main study via a full study consent process via both Qualtrics software and in person at the laboratory.

All experimental procedures took place between 11:30 AM and 5:30 PM. Participants always interacted with an experimenter, or "greeter", of the same sex throughout the entire experimental session. Upon arrival, participants were taken to the testing room, where they completed questionnaires for 20 minutes. An initial demographic survey asked information about participants' age, ethnicity, sexual orientation, SES, marital or relationship status (single or in a relationship), etc. At the end of this period, they provided a baseline saliva sample and performed a baseline cognitive task. They then either took part in the Trier Social Stress Test or sat in a room doing nothing for a similar period of time as a control condition. Another saliva sample and cognitive task were collected after the TSST or the control condition, approximately fifteen minutes after the start of the TSST or control condition. Approximately ten to fifteen minutes after the TSST or control condition had ended, participants went through a brief social interaction task with a confederate of the opposite-sex. After that, participants completed a single shot version of the Ultimatum Game and an iterated version of the Prisoner's Dilemma game; for each of these social decision making tasks, participants played against hypothetical partners of the opposite sex – one of a older age group and one of a younger age group (counter-balanced). Participants were shown a photo of their partner, were told this was the partner they would be playing against, and were told that it was a real participant whom had been selected at random as their partner. To increase motivation for all decision making tasks, we informed participants that they had a chance of obtaining an actual monetary reward, a commonly used procedure when administering monetary and other decision making tasks. After these social decision making tasks, participants went through a phased debriefing process (see below). Upon completion of all procedures, subjects were debriefed and given compensation of $15 for participation and $5 in winnings ($20 total).

Cognitive Task

In this study, cognitive performance was measured as a variable of attentional control via an arrow-based flanker task (Eriksen & Eriksen, 1974). Attentional control is defined as an executive function that involves the ability to select for goal-relevant information (Engle, 2002). The flanker task we used in this study was presented using E-Prime software (Psychology Tools, Inc.). The type and duration of specific flanker tasks used has varied extensively, making it at times difficult to separate what specific functions of attention are measured based on measured response times (RTs). To attempt to overcome this, we will use two separate RT measures to index attentional control. The first measure planned was a comparison of RTs on trials with interfering information (incongruent trials) to RTs on trials without interfering information (congruent trials; i.e. the Flanker Effect; Sanders & Lamers, 2002). Because RT difference scores can be unreliable (Lord, 1963), we also planned to analyze flanker RTs across all trials (congruent trials + incongruent trials) as a second measure of attentional control. Overall lower RTs are meant to indicate a generally increased ability to sustain attention to the task throughout the course of the entire flanker task.

Trier Social Stress Test

The Trier Social Stress Test (TSST; Kirschbaum et al. 1993) is a broadly used, standardized task that is used to study hormonal responses to mild psychosocial stress in a laboratory setting. In the current study, the experimenter explained to each participant that he or she would be giving a 5-minute presentation about himself or herself for a mock job interview. Each presentation took place in front of a "selection committee" composed of two unfamiliar confederates ("judges") trained to maintain neutral facial expressions and provide no positive feedback to the participant. Each participant was informed that he or she must keep speaking for 5 minutes and that the presentation would be video-recorded for subsequent analyses of content and non-verbal behavior. If the participant ever stopped speaking before the 5 minutes are up, the judges waited in silence for the participant to resume or otherwise prompt him or her to continue. If the participant again stopped speaking, one of the judges asked one of several standardized questions (e.g. "What do you think about teamwork?"). Upon completing the 5-min speech, the judges asked each participant to perform a difficult arithmetic calculation (i.e., serially subtracting the number 17 from 2,023) out loud for another 5 minutes or until he or she reached zero. Anytime the participant made a mistake, he or she was notified and asked to restart from the beginning. After this task, the confederates thanked the participant and left the room.

Although the “greeter” who interacted with the participant was always the same sex relative to the participant, the sex of the TSST judges alternated related to the participant, and was assigned semi-randomly, based on availability. Therefore, female participants were assigned to interact with either two same-sex TSST judges (a female “talking judge” and a female “timing judge”), or two opposite-sex TSST judges (a male “talking judge” and a male “timing judge”). Likewise, male participants were assigned to interact with either two same-sex TSST judges (a male “talking judge” and a male “timing judge”), or two opposite-sex judges (a female “talking judge” and a female “timing judge”). The semi-random assignment was based on availability of the research assistants, but also counter-balanced as much as possible.

Participant who were assigned to the control condition and who did not participant in the TSST simply sat by themselves for 10 minutes until their original experimental “greeter” returned to let them know they can continue moving forward in the study. Participants had access to several magazines during this waiting control period.

Social Interaction

Following the control/TSST condition, a second saliva sample, and a second cognitive task, all participants took part in a social interaction task, where they interacted with an opposite-sex confederate whom they had not encountered yet at that point in the session. The social interaction task that was used in this experiment was adapted from brief social interaction tasks that have been used in several research studies in which a social interaction involving a confederate posed as either an another participant or experimenter has led to physiological and behavioral changes (Roney et al., 2003; Roney et al., 2007). In our study, the experimental “greeter” let the participant know that they needed approximately five or ten minutes to pass before moving on to the next part of the study, and that the participant was free to relax until the experimenter returned. Several minutes after the departure of the “greeter”, an opposite-sex confederate entered the room and introduces himself or herself as a research assistant there to collect data off of a digital video camera (earlier in the session, this video camera was used to collect a digital photograph of every participant, as well as used to record the TSST session for participants assigned to the TSST condition). Chairs were arranged in the room such that the participants always sat directly across from the confederate with a small conference table positioned between them. Confederates then attempted to engage in natural, friendly conversation, while simultaneously uploading data from the digital video camera onto a computer or hard drive. The research confederates were free to use whatever means of engaging in conversation seem natural to them. Script or specific prompts were not used to avoid interactions seeming excessively artificial. Conversations lasted seven minutes, at which point the experimenter re-entered the room and interrupted the confederate and participant to seemingly complete the rest of the study protocol.

Following the brief social interaction, confederates completed a survey that assessed their impressions of the participants’ behaviors during the social interaction. A factor analysis done using this survey in a Roney et al. (2003) study showed that three distinct factors can be parsed out from these items, and these three factor scales are used as composite variables in this survey for the present study. First, a display factor is characterized by items in which the participants are seen as projecting information about himself or herself to the confederate. It can be interpreted as a measure of courtship-like behavior from the participant, and includes the following items: ‘tried to impress you’, ‘eager to talk about himself’, and ‘showed off to you’. Second, a polite interest factor is characterized by items suggesting attempts by the participant to gather information about the confederate, and includes the items: ‘asked questions about you’, ‘listened carefully’, and ‘interested in hearing about you’. Third, a general arousal factor includes the following items: ‘was not bored’, ‘was excited’, and ‘was speaking fast’.

Similar to the confederates’ survey, participants also completed surveys in which they rated the impressions of the research confederates and the nature of their conversations with them during a phased debriefing process at the conclusion of the study (see below).

Phased Debriefing

Following the final saliva sample of the procedure, we utilized a phased debriefing method to gather information about the participants’ assessment of the confederate and their conversation together, and also to probe for suspicion about the interaction. While alone filling out a final questionnaire via Qualtrics, participants were informed in the survey that the current project was exploring first impressions and were asked to fill out a survey assessing their impression of the confederate whom they interacted with and their conversation together. The survey was adapted from a rating instrument used by Roney, et al. (2003; 2007). Participants answered items assessing their impressions of the confederates’ perceived positivity of the conversation (i.e. how exciting, interesting, pleasant, and stressful they found the interaction to be), of the confederate conversation partner’s physical attractiveness (i.e. beautiful, sexy, and cute), of how much the confederate conversation partner made eye contact, of how much they believed the confederate conversation partner liked them, and finally of how desirable they found the confederate conversation partner to be as short-term or long-term romantic partners.

Following the phased debriefing survey, participants were probed for suspicion regarding the true purpose of the study and when the true purpose of the study occurred, and were fully debriefed in person upon return of the experimental greeter.

Saliva Sample Collection and Hormonal Assays

All saliva samples were collected between 12:00 PM and 5:00 PM, as previous studies have shown that afternoon hormone levels, although lower than morning levels, are more stable and therefore better suited for studies of social endocrinology (e.g., Gray et al. 2004). Saliva was collected by passive drool into plastic tubes. Saliva samples were stored in a refrigerator at -20°F. Samples were assayed for testosterone and cortisol concentrations using ELISA kits purchased from Salimetrics. Saliva sample concentrations were calculated based on kit standards using a 4-parameter nonlinear regression curve fit. For cortisol, the intra-assay CV based on concentration was 4.85% and the inter-assay CV based on concentration was 7.15%. For testosterone, the intra-assay CV based on concentration was 5.38% and the inter-assay CV based on concentration was 6.63%.

Data Analysis

All statistical analyses were carried out with R and/or SPSS. Whenever outcome data were not normally distributed, they were log or square root transformed. When sphericity assumptions were violated, Greenhouse-Geisser corrected p-values were reported. Alpha was set at 0.05 and adjusted for multiple comparisons where necessary.

Results

Data Transformation and Exclusion Criteria

Exclusions from the data occurred if participants exhibited flanker performance 3SDs outside the mean or below 50% for congruent trials. Three individuals did not complete the full protocol due to researcher error, participant withdrawal from the study, or computer error, and therefore were not included in data analyses. Two more individuals were excluded from all hormonal analyses due to having baseline or delta hormonal concentrations over three standard deviations away from the mean. One participant was excluded from all hormonal analyses due to saliva samples being heavily contaminated with blood. After excluding these six individuals in addition to excluding individuals who did not fit the cognitive task exclusion criteria, data were analyzed for a total of 132 individuals, of whom 69 (40 females, 29 males) underwent the TSST stress manipulation and 63 (42 females, 21 males) were in the control condition.

TSST Manipulation Check: STAI

Prior to the treatment/control session TSST and control participants did not differ in trait anxiety [STAI-T; $t(149)=-0.094$, $p=0.925$] or state anxiety [STAI-S; $t(149)=-1.244$, $p=0.925$]. After the manipulation, participants who underwent the TSST ($M=45.20$, $SD=12.67$) had significantly higher state anxiety levels than controls ($M=31.38$, $SD=8.30$): $t(148)=-7.89$, $p<0.001$. To control for baseline state anxiety levels, a change score between anxiety levels before and after the experimental manipulation was created. Participants who underwent the TSST ($M=8.45$, $SD=11.43$) had significantly higher state anxiety change scores than controls ($M=-3.38$, $SD=6.99$): $t(148)=-7.62$, $p<0.001$. Correlation analyses showed no relationship of pre-anxiety, post-anxiety, or anxiety difference score (post-STAI minus pre-STAI) with either index of attentional control as measured by flanker RT (all correlations at $p>.05$).

Flanker Performance

A complete table of means and standard deviations for baseline flanker accuracy, response times (RTs), and overall measures are reported in Table 3.1.

Table 3.1. Correlation Matrix with Descriptive Statistics of Baseline Attentional Control.

	Mean	**SD**	***1***	***2***	***3***	***4***	***5***	***6***

[1]Flanker Acc (C)	0.971	0.050	1					
[2]Flanker Acc (I)	0.895	0.150	.466**	1				
[3]Flanker RT (C)	472.52	85.71	-.351**	.099	1			
[4]Flanker RT (I)	529.62	83.69	-.210*	.148	.925**	1		
[5]Flanker Difference Score	56.67	33.06	.380**	.119	-.252**	.134	1	
[6]Overall Flanker RTs	501.07	83.10	-.287**	.126	.982**	.981**	-.063	1

Note: * denotes p<.05, ** denotes p<.001. (C) denotes congruent trials, (I) denotes incongruent trials.

Table 3.1, continued. Correlation Matrix with Descriptive Statistics of Baseline Attentional Control.

Indices of Attentional Control from the Flanker Task

The first index of attentional control was a difference score that subtracted the RTs on congruent trials from RTs on incongruent trials. This measure was highly skewed at all three timepoints throughout the protocol, and multiple transformations were unable to normalize this measure. Further, models that included this measure as the outcome did not significantly account for variance in cognitive performance and were not analyzed further. Therefore, I focused on our second index of attentional control, which was simply comprised of flanker RTs across all trials (congruent + incongruent). I reasoned that if participants are matched on flanker accuracy, then lower overall RTs indicate a relatively increased ability to sustain attention to the task at hand throughout the course of the flanker task. Overall flanker RTs at baseline did not significantly relate to gender [$F(1, 130)=1.18$, $p=0.28$) or treatment [$F(1,130=0.053$, $p=0.82$).

Social Interaction and Attentional Control

The first aim of this study was to test whether individuals experience decreases in cognitive performance after a brief social interaction with an opposite-sex confederate. Paired t tests on the log transformed data revealed a significant increase in cognitive performance (as displayed by a decrease in overall RTs) from timepoints two to three in the control condition: $t(62)=2.403$, $p=0.019$. Therefore, in all control participants, there was an overall significant change before and after the social interaction, but in a positive direction, such that cognitive performance improved from timepoint two to timepoint three (Figure 3.1).

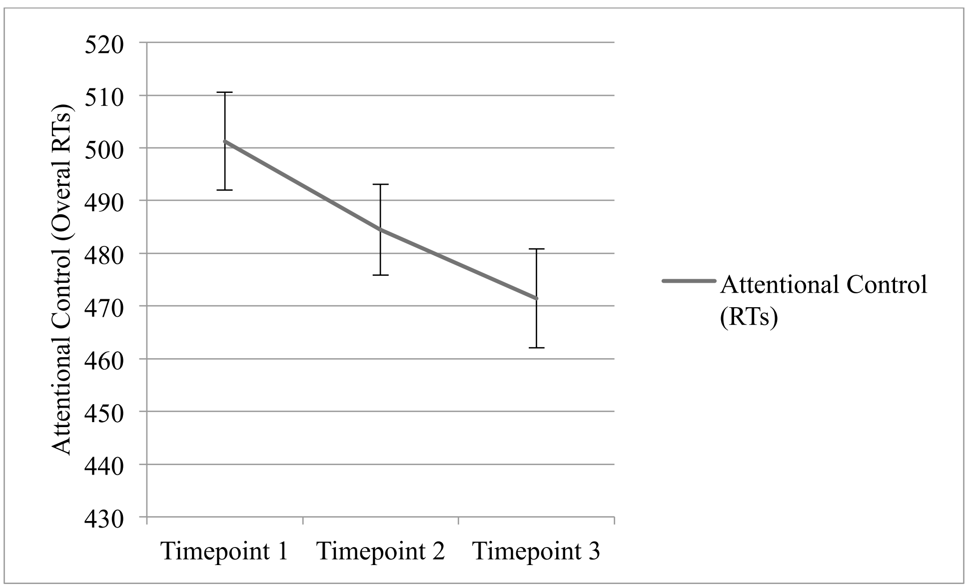

Figure 3.1. Control participants attentional control throughout the protocol as measured by overall flanker RTs. Error bars represent ± SEM.

As impression management and social cognition required for courtship behavior may increase cognitive load, we expected cognitive impairment to be higher both in individuals who rated their interaction partner as more attractive and in individuals who were rated to show higher courtship behaviors (e.g. increased eye contact, increased conversation orientation, etc.). However, after controlling for flanker performance at timepoints one and two, there were no relationships between behavior as rated by the confederate or interest as rated by the participants with cognitive performance after the social interaction task.

Stress, Social Interaction, and Attentional Control

The second aim of this study was to assess the effects of psychosocial stress on cognitive performance, as well as on the change in cognitive performance that occurs before and after a brief social interaction. A repeated measures, Greenhouse-Geisser corrected general linear model using flanker performance at the three time points (Time) as the within-subjects factor and TSST/control (Treatment) as the between-subjects factor revealed no significant main effect of Treatment [$F(1,128)=0.40$, $p=0.53$] and no significant interactive effect of Time and Treatment [$F(1.65,128)=0.49$, $p=0.58$] (Figure 3.2). A significant main effect of Time was found [$F(1.65,128)=34.84$, $p<0.001$] (Figure 3.2). Post-hoc analyses showed that a) controlling for treatment, total RT flanker performance significantly differed at timepoint two compared to timepoint one [$F(1,129)=426.3$, $p<0.001$], such that participants had improved attentional control (as measured by lower total RTs) at timepoint two (M=479.33, SD=67.32) than at timepoint one (M=501.19, SD=83.41); and b) controlling for treatment and flanker performance at timepoint one, total RT flanker performance significantly differed at timepoint three compared to timepoint two [$F(1,129)=45.60$, $p<0.001$], such that participants had improved attentional control (as measured by lower total RTs) at timepoint three (M=466.48, SD=73.53) than at timepoint one (M=479.33, SD=67.32). Overall, there was no difference in cognitive performance between control and treatment individuals, and all participants showed significant increases in cognitive performance throughout the protocol.

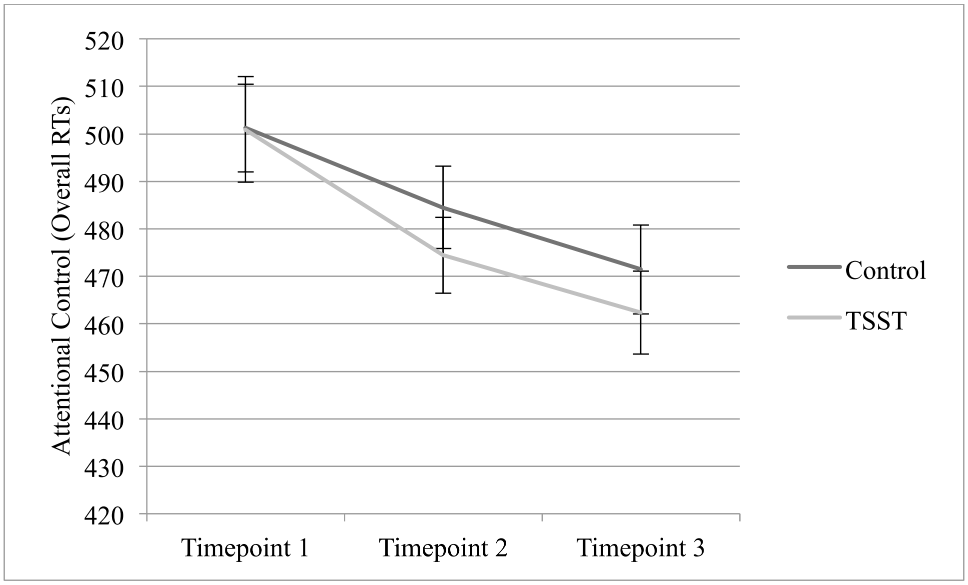

Figure 3.2. Control vs. treatment participants' attentional control throughout the protocol as measured by overall flanker RTs. Error bars represent ± SEM.

Cognitive Performance, Subjective Anxiety, and Cortisol

Although there was no significant effect of treatment on cognitive performance, I predicted that individuals who had greater changes in stress responses (higher increases in cortisol and higher increases in subjective anxiety) would be particularly affected in terms of cognitive performance. To understand the relationship between cognitive performance and both subjective and physiological anxiety, I examined the relationship between change in attentional control performance (difference values from total RTs of flanker performance from timepoints three to two, three to one, and two to one), change in cortisol reactivity, and change in subjective state anxiety (STAI-S) measures.

Correlation analyses revealed no significant relationships between state anxiety or cortisol reactivity and cognitive performance in control participants. However, for TSST participants, correlation analyses did reveal a significant relationship between the change in cognitive performance from timepoint three to timepoint one and change in subjective state anxiety before and after the TSST (r=-.324, p=0.007), such that individuals who had decreased in cognitive performance throughout the protocol (increased in total RTs) showed lower increases in subjective anxiety after the TSST (Figure 3.3A). Further, for treatment participants, correlation analyses revealed a significant relationship between change in cognitive performance from timepoint three to timepoint two and change in cortisol concentration from timepoint three to timepoint one (r=-.276, p=0.023), such that individuals who had decreased in cognitive performance during the social interaction (increased in total RTs) had lower increases in cortisol over the course of the TSST and social interaction (Figure 3.3B). Overall, controlling for cortisol reactivity, TSST individuals who had higher subjective anxiety after the TSST increased in cognitive performance throughout both the TSST and social interaction. Controlling for subjective anxiety changes, TSST individuals who had higher increases in cortisol throughout both the TSST and social interaction increased in cognitive performance during the social interaction task.

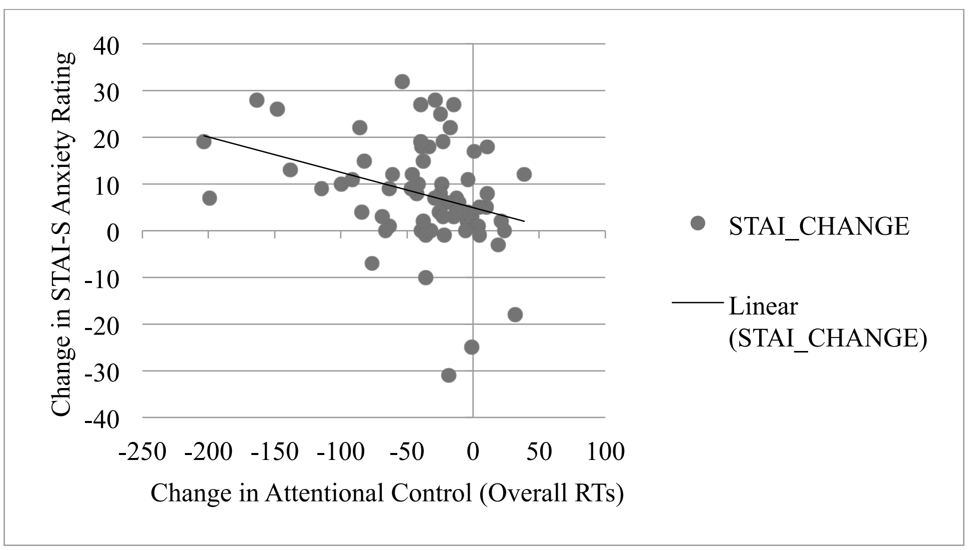

Figure 3.3A. Relationship between change in subjective anxiety (STAI-S) rating during the TSST session and change in attentional control (as measured by overall RTs) for TSST participants throughout the entire protocol.

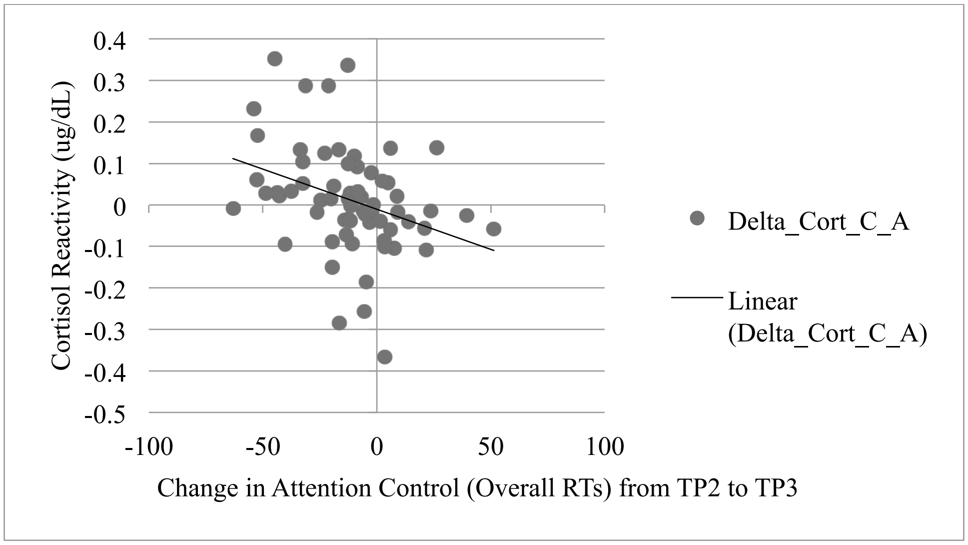

Figure 3.3B. Relationship between cortisol reactivity through the entire protocol and change in attentional control (as measured by overall RTs) during the social interaction for TSST participants.

Discussion

Control participants did not experience decreases in cognitive performance after a brief social interaction with a confederate of the opposite sex. Instead, the data presented here show a significant increase in cognitive performance throughout the protocol. It is possible that individuals did not reach "peak" performance during practice trials, and instead simply continued to improve in task performance as experience was gained throughout the protocol.

There was no difference in cognitive performance between individuals who participated in a psychosocial stress treatment and control individuals. That is, stress treatment did not affect cognitive performance, and stress treatment also did not affect cognitive performance changes that occur during a brief social interaction. Both treatment and control individuals improved in cognitive performance from timepoint one to timepoint two, and again from timepoint two to timepoint three.

Cognitive performance was not significantly related to individual differences in gender, personality, or relationship status. Further, changes in cognitive performance were not related to behavior as rated by the participant (e.g., how attractive the participant found the opposite-sex confederate during a social interaction) or to behavior as rated by the confederate (e.g., how engaged the participant was towards conversation with the opposite-sex confederate).

Change in cognitive performance was not related to cortisol reactivity or subjective anxiety in control participants. However, there were significant relationships between cortisol reactivity and changes in subjective anxiety and cognitive performance in individuals who experienced a psychosocial stress treatment. Specifically, for individuals who had higher increases in subjective anxiety throughout the TSST, cognitive performance increased more drastically throughout the entire protocol. For individuals who had higher increases in cortisol throughout the TSST and the social interaction, cognitive performance increased more drastically throughout the social interaction. Therefore, although cognitive performance increased in most individuals throughout the protocol, it was particularly improved for individuals who experienced higher changes in subjective and physiological anxiety. In past research, attentional resources and similar core executive functions have been shown to be affected by psychosocial stress, but in inconsistent ways (Chajut & Algom, 2003; Jamieson et al., 2012; Pilgrim et al., 2014; Sato et al., 2012; Shields et al., 2016). A few of these studies have shown that stress may improve selectivity of attention or help to remove attentional bias (Chajut & Algom, 2003; Jamieson et al., 2012), which is what the data presented here find as well.

Overall, little of what is presented here supports the idea that ecological cues of courtship opportunities, at least as presented in this protocol, have effects on cognition. Although most individuals improved in cognitive performance throughout the protocol, it is likely that this happened because individuals gained experience with the task throughout the study, as no differences were found between treatment groups or individual differences related to courtship. If anything, signs of increased stress (e.g., subjective anxiety, cortisol reactivity) actually seemed to correlate with improved cognitive performance, indicating that acute stress may increase overall attentional focus.

CHAPTER 4

Exploring the role of hormonal reactivity, social interaction, and stress in measured behavioral outcomes

Abstract

This study investigated the effects of a brief social interaction on courtship-like behavioral output, as well as the effect of psychosocial stress on behavior. Results did not replicate previous findings that testosterone reactivity is correlated with potentially adaptive display behavior in men, nor displayed evidence for this in women. Instead, lower cortisol reactivity, as well as certain personality traits in males, were found to be significantly related to display behaviors that may aid in courtship. Further, the relationships between stress and personality may differ for males and females. Further research focusing on more salient inputs of courtship opportunity is needed to clarify how and if hormonal reactivity is related to courtship behavior, particularly in terms of social situations that may induce psychosocial stress and social evaluative threat in individuals who may differ in personality.

Introduction

Previous research has reported some effects of stress on decision making in humans. However, the findings are mixed. One study suggests that stressed individuals in a social context engage in greater trusting behavior than individuals who are not socially stressed (von Dawans et al., 2012). However, another recent study found that while participants who were not under stress showed no differences in decision making in a nonsocial gambling and a social trust context, stressed subjects gambled more money in a nonsocial context and entrusted less money in a social context (FeldmanHall et al., 2015). In a previous study, I showed that psychosocial stress affects both social and financial decision-making, but some of the effects are different for men and women (Nickels et al., 2017). As courtship is a social situation in which individuals make potentially unconscious decisions about their social behavior, it is possible that stress has the ability to effect courtship behaviors in terms of their willingness to trust and interact with a potential mate in a positive way.

Many studies that focus on the effects of stress on cortisol and testosterone include gender as a potential moderator. Gender has been found to moderate the effects of stress on decision making and risk taking (van den Bos et al. 2009; Lighthall et al., 2009; Mather & Lighthall, 2012; Nickels et al., 2017). Taylor et al. (2000) suggested that females' stress response could be characterized as "tend and befriend" rather than as "flight or fight". Taylor argued that women are born with a more invested role in the care of offspring, and responses to stress or threat that would have been successfully passed on are those that protected both the offspring and the self. "Tending" may be an alternative behavioral response that has evolved in females, as the male-typical fight and flight response may place offspring in jeopardy (Taylor et al., 2000). There is some evidence in support of Taylor et al.'s theory (Lighthall et al., 2009; Nickels et al. 2017), but more research is needed with study designs that include both males and females participants when measuring responses to acute psychosocial stress interactions.

In addition to the potential moderating effects of gender on stress and courtship behavior, we should also consider the potential moderating effects of the gender make-up of the surrounding social environment and ecological cues. For example, several studies using the TSST have shown that in addition to the sex of the participant, the sex of the judges, and slight modifications in the timing or durations of different components of the a psychosocial stress-inducing procedure can generate significant variation in the cortisol response (Andrews et al., 2007; Wadiwalla et al., 2010; Duchesne et al., 2012; Liu et al., 2017; Kim et al., 2018). In a recent laboratory study, we found that differences in the sex of the talking judge in the TSST were associated with a significant increase in cortisol across male and female participants, and also with an increase in testosterone among male, but not female, participants (Kim et al., 2018). One likely explanation for the effects of male talking judges on participants' testosterone levels is that interacting with these individuals stimulated competitiveness in the participants. Overall, more research is necessary to test the hypothesis that variation in the sex of individuals presently inducing psychosocial stress may generate significant variation in hormone concentrations and subsequent behavior. For laboratory studies in particular, confederate gender is a potentially important variable that can influence psychological processes related to romantic/sexual attraction or competition (e.g., for competition for status, either with same-sex or with opposite-sex individuals) in the participants.

Although gender effects have not been found in all studies, the moderating effects of gender that have been found in some decision making situations have been attributed to the interaction between different behavioral and endocrine stress responses and different behavioral and neural mechanisms involved in decision making in men and women (Starcke & Brand, 2012). A recent review of studies examining gender differences in decision making tasks proposed that differences in men and women may exist in decision making under ambiguity. This review concluded that these gender differences are generally more related to differences in activation of neuronal circuits dealing with emotional and cognitive control over emotional events. If emotional and cognitive control during social interactions is related to differences in behavior between men and women, it is possible that differences in output of courtship behavior between men and women may be due to behavioral biological mechanisms in how men and women respond to psychosocial stress. As endocrine function and cognitive control in addition to gender may have the ability to affect stress response, hormones and cognition may be required to function together to produce adaptive responses when confronted with a potential courtship opportunity.

Aims / Hypotheses:

Aim 1: Investigate the relationship between hormonal responses and behavior

In order to explore the relationship between hormonal reactivity and courtship behavior, I will measure how brief social interactions with a potential courtship partner affect testosterone responses in both males and females and how this testosterone reactivity correlates with courtship behavior. I expect that, for control participants, higher testosterone increases will correlate with positive courtship behavior (e.g. increased eye contact, increased conversation orientation, etc.). I expect these correlations to also be strongest in individuals who rank their interaction partner as more attractive (physically, as a potential relationship partner, etc.).

Aim 2: Investigate the effects of psychosocial stress and cortisol increases on courtship behavior

Courtship interactions involve a social evaluation that may cause significant psychosocial stress. Therefore, we may expect psychosocial stress to potentially dampen self-confidence, motivation, and approach behaviors in a way that may hinder social cognition and impression management during courtship. To measure the effects of psychosocial stress and cortisol increase on behavior, I experimentally manipulate psychosocial stress prior to a potential courtship social interaction. I expect that: 1) if cortisol increases are high and testosterone increases are dampened, individuals who experiences psychosocial stress will display less courtship behaviors due to lowered motivation to maintain higher social status, dominance, and approach behaviors;

2) if baseline cortisol in stressed individuals is lower, this may predict higher increases in testosterone (see Maestripieri et al., 2010), and in turn I would expect increases in courtship behaviors.

Aim 3: Investigate how gender and individual differences in personality and motivation affect courtship behavior

Finally, in order to measure how gender and individual differences in personality, relationship status, and baseline cognition moderate or mediate the relationship between endocrine behavior and courtship behavior, I will measure individuals differences using multiple scales that will measure personality traits (e.g., introversion, extroversion), trait anxiety, autistic-like characteristics, and other individual characteristics that may moderate responses to courtship and stress. I expect that: 1) individuals who are more extroverted, have lower trait-anxiety, less autistic-like characteristics, or who are not in a relationship will have higher amounts of courtship-oriented behavioral outcomes; and 2) gender may moderate the relationship between stress and courtship behavior, such that higher testosterone increases in males after stress may facilitate courtship behaviors for males, while lower testosterone increase in females after stress may be an alternative strategy for females. For example, if stress renders females more cooperative as opposed to more competitive, females may show lower testosterone increases, but still exhibit courtship behaviors that are cooperative and positive in terms of social cognition, leading to overall courtship facilitation.

Methods

Study Design / Overview

Participants were randomly assigned prior to arrival to either a control or psychosocial stress condition (Trier Social Stress Test), and those assigned to the stress condition were also semi-randomly assigned to interact with either same-sex or opposite-sex judges during the Trier Social Stress Test (TSST), based on researcher availability. Upon arrival at the lab, participants provided consent, filled out questionnaires (see below), and participated in either the control or TSST condition. Following the assigned condition, all subjects went through a social interaction with an opposite-sex confederate. Finally, participants completed two social decision making tasks and participate in a phased debriefing. Following full debriefing, participants were compensated and the experimental session ended. Saliva samples were collected at various time points throughout the tasks to assess hormone levels via ELISA: once before the control/TSST condition, once after the control/TSST condition, and once after the social interaction. Cognitive tasks were also administered throughout the protocol, in conjunction with the collected saliva samples. The Social Science Institutional Review Board at the University of Chicago had approved all experimental procedures (IRB #12-1251).

Participants

Participants were 156 individuals (age range: 18-35 years) from the greater Chicago area, surrounding the University of Chicago campus. All participants were recruited on the University of Chicago campus through fliers, UChicago Marketplace, and a human subject recruitment website (Sona System). Data was collected continuously throughout fifteen months, from July 2017 through August 2018. All study participants completed a digital prescreen consent form and eligibility questionnaire via Qualtrics software, a digital main protocol consent form and questionnaire via Qualtrics software, and also a written informed consent form at the laboratory before participating in the study. All participants were paid $20 compensation after completion of the procedures: $15 as base pay, and $5 as "winnings" from the decision making tasks.

Experimental Procedure

All experimental procedures took place at the Behavioral Biology Laboratory, located in the Biopsychological Sciences Building (940 E. 57th Street) at the University of Chicago. Prior to arrival at the lab, participants consented to a prescreen questionnaire administered via Qualtrics software. If eligible, participants consented to the main study via a full study consent process via both Qualtrics software and in person at the laboratory.

All experimental procedures took place between 11:30 AM and 5:30 PM. Participants always interacted with an experimenter, or "greeter", of the same sex throughout the entire experimental session. Upon arrival, participants were taken to the testing room, where they completed questionnaires for 20 minutes. An initial demographic survey asked information about participants' age, ethnicity, sexual orientation, SES, marital or relationship status (single or in a relationship), etc. At the end of this period, they provided a baseline saliva sample and performed a baseline cognitive task. They then either took part in the Trier Social Stress Test or sat in a room doing nothing for a similar period of time as a control condition. Another saliva sample and cognitive task was collected and performed after the TSST or the control condition, approximately fifteen minutes after the start of the TSST or control condition. Approximately ten to fifteen minutes after the TSST or control condition had ended, participants went through a brief social interaction task with a confederate of the opposite-sex. After that, participants completed a single shot version of the Ultimatum Game and an iterated version of the Prisoner's Dilemma game; for each of these social decision making tasks, participants played against hypothetical partners of the opposite sex – one of a older age group and one of a younger age group (counter-balanced). Participants were shown a photo of their partner, were told this was the partner they would be playing against, and were told that it was a real participant whom has been selected at random as their partner. To increase motivation for all decision making tasks, we informed participants that they had a chance of obtaining an actual monetary reward, a commonly used procedure when administering monetary and other decision making tasks. After these social decision making tasks, participants went through a phased debriefing process (see below). Upon completion of all procedures, participants were fully debriefed and given compensation of $15 for participation and $5 in winnings ($20 total).

Questionnaires/Materials

In addition to a demographics survey and a pre-general health survey given before saliva samples were collected, participants completed the following questionnaires:

<u>Prior to lab arrival:</u>

Reduced version of the Morningness-Eveningness Questionnaire (rMEQ; Adan & Almirall, 1991): We assessed chronotype with the reduced version of the Morningness-Eveningness Questionnaire. The rMEQ is a validated, 5-item Likert-type scale obtained from the original 19-item version of the MEQ (Horne & Östberg, 1976). The rMEQ identifies participants' preferences in relation to sleeping and waking time, the time of day when they experience maximal efficiency, their level of tiredness within half an hour of awakening, and their self-perceived chronotype (Adan & Almirall, 1991). Scores for the rMEQ range from 4 to 25; scores below 12 identify participants as evening-types and scores above 17 as morning-types.

The Autism-Spectrum Quotient (AQ; Baron-Cohen et al. 2001): The AQ is a 50-item self-report measure of autistic-like traits in adults with normal IQ. It includes items such as "I prefer to do things with others rather than on my own" or "I tend to have very strong interests that I get upset about if I can't pursue". This questionnaire has been widely used and validated in many studies of children, adolescents, and adults. In the AQ, four-point Likert-type responses are dichotomized to yield a binary score (0,1) for each item. The total AQ score can range between 0 (no autistic-like traits) to 50 (maximum autistic-like traits).

Trait portion of State-Trait Anxiety Inventory (STAI-T: Spielberger & Gorsuch, 1983): The State-Trait Anxiety Inventory, or STAI, is a long-standing measure that uses two scales to report two measures of anxiety (state anxiety and trait anxiety). Trait anxiety can be defined as an individual measure of intensity and frequency of experienced anxiety, which involves feelings of apprehension and heightened response of the autonomic nervous system (Spielberger & Gorsuch, 1983). Importantly, trait anxiety is seen as a relatively stable trait, and seeks to measure individual differences in proneness to experiencing anxiety as a personality trait. The STAI trait scale consists of twenty statements that have individuals rate, on a four-point Likert scale, different statements about how they feel *generally* (e.g., "I feel nervous and restless.").

Adverse Childhood Experiences Module (BRFSS-ACE; Bynum et al., 2010): The Adverse Childhood Experience (ACE) Module of the Behavior Risk Factor Surveillance System measures adverse childhood experiences of verbal, physical, or sexual abuse, including experiences of family dysfunction. The use of the study of ACEs is connected to the treatment of stress-related health outcomes and is included in this study to attempt to control for differences in stress reactivity based on prior developmental experience.

Birth weight items: Information regarding birth weight and gestational age, along with the participant's self-reported source of information, was collected to account for differences in extremely low birth weights that may be correlated with self-reported levels of psychopathology.

Early life, low birth weight individuals have been shown to have elevated levels of depression, anxiety, and avoidant personality problems (Boyle et al., 2011).

UCLA Loneliness Scale (Russell et al., 1980): The UCLA Loneliness scale is a measurement used in social psychological research focusing on loneliness. Although studies have shown that loneliness is a unique psychological experience, measures of loneliness have also been found to be correlated with measures of social risk taking and negative affect (Russell et al., 1980).

Upon lab arrival:

Big-Five Personality Inventory (BFI; John, Donahue, & Kentle, 1991) Personality was assessed using the Big Five Inventory, a 44-item, frequently used questionnaire measuring personality traits along five dimensions: extraversion, agreeableness, conscientiousness, neuroticism, and openness to experience.

MacArthur Scale of Subjective Social Status (Adler & Stuart, 2007): The MacArthur Scale of Subjective Social Status is a developed measure of subjective social status meant to capture individuals' sense of standing on a social ladder. Based on its wording, it seeks to take into account sense of place on multiple dimensions of social position or status (e.g., socioeconomic, general status among a similar demographic, etc.)

Ten-Item Personality Inventory (TIPI; Gosling et al., 2003): The TIPI is a ten-item, brief measure of personality based on the Big-Five Personality Inventory. It has sufficient levels of convergence with the widely used Big-Five inventory and is usable in situations where short measures are needed. In this study, we use the TIPI to measure pre- and post-stress and social interaction self-reported personality ratings throughout the experimental protocol.

Shortened version of the Math Anxiety Rating Scale (sMARS; Alexander & Martray, 1989): The sMARS is an internally consistent and reliable, abbreviated form of the Mathematics Anxiety Rating Scale (MARS). In this study, we sought to measure math anxiety to capture individual differences in specific tension, apprehension, or fear surrounding math, due to the fact that the Trier Social Stress Test (TSST) involves a math task that is used to induce acute psychosocial stress (see below for more detail about the TSST).

The Multidimensional Sociosexual Inventory (MSOI; Jackson & Kirkpatrick, 2007): The MSOI is a 23-item measure of sociosexual orientation and behavior. It is comprised of two attitudinal scales with 7-point Likert-type items that measure short-term (STMO) and long-term (LTMO) mating orientation, and one scale measuring past sexual experience (PSE). The PSE includes three questions about the lifetime number of sexual partners, the lifetime number of casual sexual partners, and the number of sexual partners in the previous year.

State portion of State-Trait Anxiety Inventory (STAI-S: Spielberger & Gorsuch, 1983): The State-Trait Anxiety Inventory, or STAI, is a long-standing measure that uses two scales to report two measures of anxiety (state anxiety and trait anxiety). Unlike trait anxiety, which seeks to measure individuals differences in proneness to anxiety as a personality trait, state anxiety measures the intensity of anxiety as an acute, emotional state (Spielberger & Gorsuch, 1983). The STAI state scale consists of twenty statements that have individuals rate, on a four-point Likert scale, different statements about the intensity of their anxiety “right now, at this moment”.

Trier Social Stress Test

The Trier Social Stress Test (TSST; Kirschbaum et al. 1993) is a broadly used, standardized task that is used to study hormonal responses to mild psychosocial stress in a laboratory setting. In the current study, the experimenter explained to each participant that he or she would be giving a 5-minute presentation about himself or herself for a mock job interview. Each presentation took place in front of a “selection committee” composed of two unfamiliar confederates (“judges”) trained to maintain neutral facial expressions and provide no positive feedback to the participant. Each participant was informed that he or she must keep speaking for 5 minutes and that the presentation will be video-recorded for subsequent analyses of content and non-verbal behavior. If the participant ever stopped speaking before the 5 minutes are up, the judges waited in silence for the participant to resume or otherwise prompted him or her to continue. If the participant again stopped speaking, one of the judges asked one of several standardized questions (e.g. “What do you think about teamwork?”). Upon completing the 5-min speech, the judges asked each participant to perform a difficult arithmetic calculation (i.e., serially subtracting the number 17 from 2,023) out loud for another 5 minutes or until he or she reaches zero. Anytime the participant made a mistake, he or she was notified and asked to restart from the beginning. After this task, the confederates thanked the participant and left the room.

Although the “greeter” who interacted with the participant was always the same sex relative to the participant, the sex of the TSST judges alternated related to the participant, and was assigned semi-randomly, based on availability. Therefore, female participants were assigned to interact with either two same-sex TSST judges (a female “talking judge” and a female “timing judge”), or two opposite-sex TSST judges (a male “talking judge” and a male “timing judge”). Likewise, male participants were assigned to interact with either two same-sex TSST judges (a male “talking judge” and a male “timing judge”), or two opposite-sex judges (a female “talking judge” and a female “timing judge”). The semi-random assignment was based on availability of the research assistants, but was also counter-balanced as much as possible.

Participants who were assigned to the control condition and who did not participate in the TSST simply sat by themselves for 10 minutes until their original experimental "greeter" returned to let them know they could continue moving forward in the study. Participants had access to several magazines during this waiting control period.

Social Interaction

Following the control/TSST condition, a second saliva sample, and a second cognitive task, all participants partook in a social interaction task, where they interacted with an opposite-sex confederate whom they had not encountered yet at that point in the session. The social interaction task that was used in this experiment was adapted from brief social interaction tasks that have been used in several research studies in which a social interaction involving a confederate posed as either another participant or experimenter led to physiological and behavioral changes (Roney et al., 2003; Roney et al., 2007). In our study, the experimental "greeter" let the participant know that they needed approximately five or ten minutes to pass before moving on to the next part of the study, and that the participant was free to relax until the experimenter returned. Several minutes after the departure of the "greeter", an opposite-sex confederate entered the room and introduced himself or herself as a research assistant there to collect data off of a digital video camera (earlier in the session, this video camera was used to collect a digital photograph of every participant, as well as used to record the TSST session for participants assigned to the TSST condition). Chairs were arranged in the room such that the participants always sat directly across from the confederate with a small conference table positioned between them. Confederates then attempted to engage in natural, friendly conversation, while simultaneously uploading data from the digital video camera onto a computer or hard drive. The research confederates were free to use whatever means of engaging in conversation seem natural to them. Script or specific prompts were not used to avoid interactions seeming excessively artificial. Conversations lasted seven minutes, at which point the experimenter re-entered the room and interrupted the confederate and participant to seemingly complete the rest of the study protocol.

Following the brief social interaction, confederates completed a survey that assessed their impressions of the participants' behaviors during the social interaction. A factor analysis done using this survey in a Roney et al. (2003) study showed that three distinct factors can be parsed out from these items, and these three factor scales were used as composite variables in this survey for the present study. First, a display factor is characterized by items in which the participants are seen as projecting information about himself or herself to the confederate. It can be interpreted as a measure of courtship-like behavior from the participant, and includes the following items: 'tried to impress you', 'eager to talk about himself', and 'showed off to you'. Second, a polite interest factor is characterized by items suggesting attempts by the participant to gather information about the confederate, and includes the items: 'asked questions about you', 'listened carefully', and 'interested in hearing about you'. Third, a general arousal factor includes the following items: 'was not bored', 'was excited', and 'was speaking fast'.

Similar to the confederates' survey, participants also completed surveys in which they rated the impressions of the research confederates and the nature of their conversations with them during a phased debriefing process at the conclusion of the study (see below).

Phased Debriefing

Following the final saliva sample of the procedure, we utilized a phased debriefing method to gather information about the participants' assessment of the confederate and their conversation together, and also to probe for suspicion about the interaction. While alone filling out a final questionnaire via Qualtrics, participants were informed in the survey that the current project was exploring first impressions and were asked to fill out a survey assessing their impression of the confederate whom they interacted with and their conversation together. The survey was adapted from a rating instrument used by Roney, et al. (2003; 2007). Participants answered items assessing their impressions of the confederates' perceived positivity of the conversation (i.e. how exciting, interesting, pleasant, and stressful they found the interaction to be), of the confederate conversation partner's physical attractiveness (i.e. beautiful, sexy, and cute), of how much the confederate conversation partner made eye contact, of how much they believed the confederate conversation partner liked them, and finally of how desirable they found the confederate conversation partner to be as short-term or long-term romantic partners.

Following the phased debriefing survey, participants were probed for suspicion regarding the true purpose of the study and when the true purpose of the study occurred, and were fully debriefed in person upon return of the experimental greeter.

Saliva Sample Collection and Hormonal Assays

All saliva samples were collected between 12:00 PM and 5:00 PM, as previous studies have shown that afternoon hormone levels, although lower than morning levels, are more stable and therefore better suited for studies of social endocrinology (e.g., Gray et al. 2004). Saliva was collected by passive drool into plastic tubes. Saliva samples were stored in a refrigerator at -20°F. Samples were assayed for testosterone and cortisol concentrations using ELISA kits purchased from Salimetrics. Saliva sample concentrations were calculated based on kit standards using a 4-parameter nonlinear regression curve fit. For cortisol, the intra-assay CV based on concentration was 4.85% and the inter-assay CV based on concentration was 7.15%. For testosterone, the intra-assay CV based on concentration was 5.38% and the inter-assay CV based on concentration was 6.63%.

Data Analysis

All statistical analyses were carried out with R and/or SPSS. Whenever hormonal data were not normally distributed, they were log or square root transformed. When sphericity assumptions were violated, Greenhouse-Geisser corrected p-values were reported. Alpha was set at 0.05 and adjusted for multiple comparisons where necessary.

Results

Behavioral Measures

<u>Participant Surveys:</u> Participants on average rated the opposite-sex confederates slightly above or slightly below the midpoints of the seven-point scales that assessed perception of physical attractiveness (M=4.33, SD=1.60), desirability as a short term romantic partner (M=3.32, SD=1.78), and desirability as a long term romantic partner (M=3.34, SD=1.70). Such ratings suggest that the participants did not have particularly strong interest in opposite sex confederates, but did rate individuals as above average in terms of attractiveness.

All items employed 7-point Likert scales. Three items assessed participants' impressions of confederates' physical attractiveness: beautiful, sexy, and cute ($\alpha = 0.90$). Four items assessed their impressions of the confederates' perceived positivity of the conversation, in terms of how exciting, interesting, pleasant, and stressful they found the interaction to be ($\alpha = 0.90$). Three items assessed their impressions of how desirable they found the confederate conversation partner to be as a friend partner, a short-term romantic partner, or a long-term romantic partner

($\alpha = 0.90$). Single items also measured participants' impressions of how much the confederate conversation partner made eye contact (M=4.78, SD=1.30), of how much they believed the confederate conversation partner liked them (M=4.09, SD=1.19), and of how attractive they found their partner to be (M=4.33, SD=1.60).

Confederate Surveys: As discussed above, confederates completed a survey that assessed their impressions of the participants' behaviors during the social interaction. This questionnaire was factor analyzed via principal components analysis with varimax rotation. The resulting factors, item loadings, and variance accounted for appear in Table 4.1. The first factor has been labeled as the most general measure of excitement or arousal. The second factor has been labeled "polite interest", since the highest loading items relate to gathering information about the conversation partner (e.g., Roney et al., 2003). The third factor has been labeled as the "display" factor, as it is comprised of items that indicate outward projections of information about oneself in an attempt to impress the conversation partner. This factor indexes what would commonly be thought of as courtship-like behavior (e.g., Roney et al., 2003).

Table 4.1. Factor scales for ratings of participants' behavior during the conversation. *denotes item was reversed coded.

Factor	
Polite Interest (28%)	
Was quiet*	.34
Was talkative	.41
Eager to talk about oneself	.33
Was excited	.42
Revealed details about oneself	.49
Was interested	.83
Made eye contact	.80
Asked questions	.73
Listened carefully	.71
Was bored*	.64

Arousal (32%)	
Was quiet*	.80
Was talkative	.80
Eager to talk about oneself	.76
Was excited	.75
Was speaking fast	.70
Revealed details about oneself	.63
Was interested	.39
Listened carefully	.50
Was bored*	.50
Display (18%)	
Showed off to you	.93
Tried to impress you	.92
Eager to talk about oneself	.32
Was speaking fast	.34
Revealed detail about oneself	.34
Asked questions	.39

Table 4.1, continued. Factor scales for ratings of participants' behavior during the conversation. *denotes item was reversed coded.

Factor scales were computed as average scores across items within the respective factors. The participants' ratings of the opposite sex confederates desirability as partners (short term romantic, long term romantic, friend) predicted confederates' ratings of participants' display behaviors ($r=.271$, $p<0.001$) and arousal ($r=.165$, $p=.043$). These correlations support the validity of the behavioral rating scales by demonstrating that confederates were able to accurately detect behaviors that were associated with participants' interest in them.

Relationship between hormones and behavioral measures

The relationship between testosterone reactivity during the social interaction and behavioral ratings were measured in control participants. For control participants, change in T levels were not significantly related to control participants' ratings of the confederates' physical attractiveness (M: $r=.26$, $p=.20$; F: $r=.02$, $p=.90$) or desirability as a romantic partner (M: $r=.25$, p=.22; F: r=.04, p=.81). Further, change in T levels was not significantly related to control participant's behavior as rated by confederates, both in terms of measured display behaviors (M: r=.22, p=.28; F: r=-.03, p=.86) and arousal behaviors (M: r=.11, p=.60; F: r=-.04, p=.80). Therefore, testosterone reactivity during a brief social interaction with an opposite sex confederate was not related to courtship behavior in men or women.

Effects of psychosocial stress on behavioral measures

To test for the overall differences in courtship behavior between control and treatment individuals, both confederate and participant ratings were compared between control and TSST individuals. There were no significant differences between control participants and TSST participants in participant display behavior [F(1,49)=.23, p=.63] or arousal behavior [F(1,49)=.02, p=.88] as rated by confederates. There were also no significant differences between control and TSST participants in participant interest in confederates as romantic partners [F(1,49)=.36, p=.55]. However, there was a significant difference in participant rating of confederate attractiveness [F(1,49)=4.11, p=.04; Figure 4.1], such that participants who underwent the TSST rated confederates as more physically attractive than control participants (TSST: M=4.59, SD=1.63; Control: M=4.07, SD=1.53).

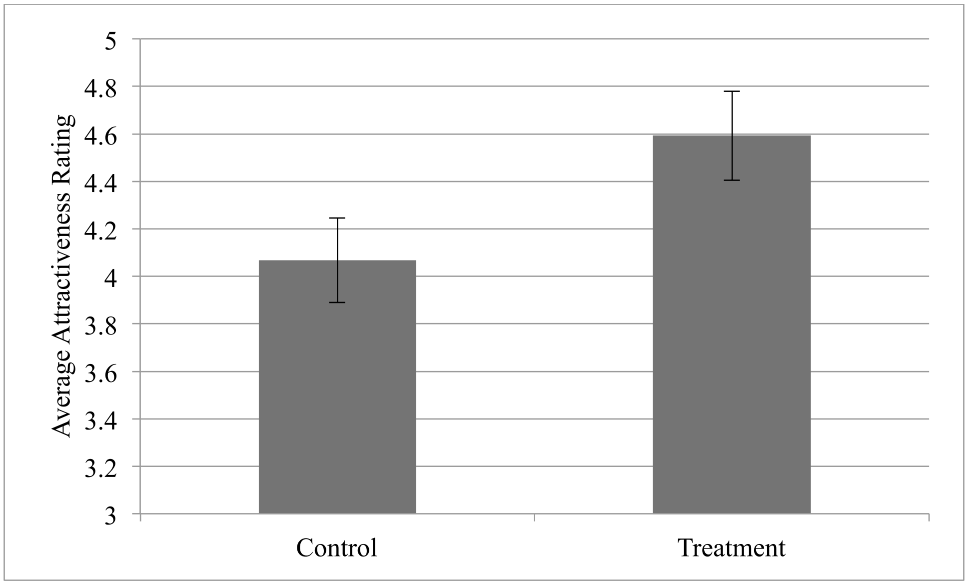

Figure 4.1. Participant ratings of confederate attractiveness for control and TSST individuals. Error bars represent ± SEM.

In control participants, there were no significant relationships between cortisol reactivity during the social interaction and participant courtship behavior (as rated by the confederate nor as rated by interest of the participant). However, in TSST participants, the correlation between changes in cortisol levels and confederate ratings of participants' display behaviors was significant ($r=-.27$, $p=.02$; Figure 4.2), as was the correlation between changes in cortisol and confederate ratings of participants' general arousal behaviors ($r=-.29$, $p=.01$). These results suggest that confederates who interacted with TSST participants detected more courtship-like behaviors directed towards them from participants who showed less positive changes in cortisol levels from before to after the conversation. This effects was restricted to courtship-like behaviors indexed by the display factor and the arousal factor, as change in cortisol was not significant correlated with the polite interest factor or with interest as rated by participants themselves.

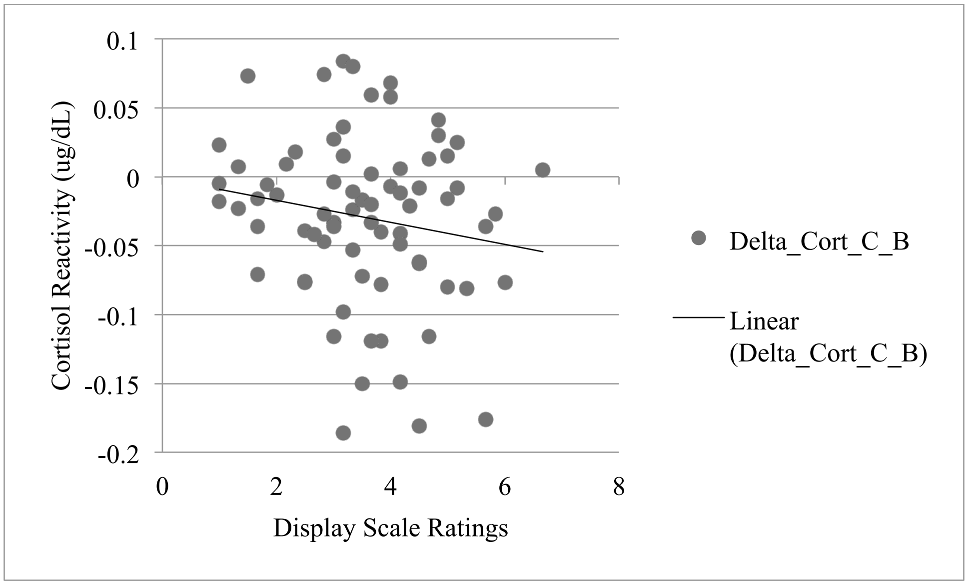

Figure 4.2. The degree to which conversation partners rated the participants as exhibiting display behaviors plotted against the participants change in cortisol levels.

Individual differences and behavioral measures

To assess how individual differences in gender, relationship status, and personality were related to measured ratings of potential courtship behavior, I compared behavior as rated by confederates and as rated by participant interest between groups of participants. There were no significant differences in behavior between singles and individuals in a relationship, or between men and women, for both behavioral measures as rated by confederates and interest and attractiveness ratings as determined by participants.

Males: For control males, there were significant relationships between personality measures and courtship behavior. Specifically, male participants who had higher ratings of extroversion (BFI-E) displayed higher amounts of display behaviors as rated by participants (r=.49, p=.001), and male participants who had higher ratings of agreeableness (BFI-A) rated their conversation partners as more attractive (r=.40, p=.03), rated their partners more highly as individuals who they would be interested in as romantic partners ($r=.58$, $p=.001$), and rated the conversation as more positive overall ($r=.54$, $p=.004$). These relationships were not significant for male participants who went through the TSST before interacting with opposite sex confederates.

Females: For control females, there were also significant relationships between personality measures and courtship behaviors. Specifically, female participants who did not go through the TSST who had higher ratings of extroversion (BFI-E) rated their conversation partners as less attractive ($r=-.391$, $p=.007$). This relationship was not significant for females who were assigned to the TSST treatment condition.

Discussion

In control participants who interacted briefly with an opposite sex confederate, there was no relationship between hormonal reactivity and measured behavioral outcomes (display behaviors as rated by confederates and interest behavior as self-rated by participants). Participants who underwent the TSST rated their confederate partners as physically more attractive when compared to control participants. However, no measures of interest or display behaviors differed between control and TSST participants. Thus, the significant relationship between the stress and ratings of attraction could be related to individuals becoming more cooperative under stress as opposed to becoming more oriented towards displays of courtship-specific behaviors.

Although cortisol reactivity in control participants was not related to courtship-related behaviors, there was a significant relationship between cortisol reactivity and display behaviors as rated by confederates in participants who underwent the TSST. Specifically, confederate partners rated participants who had stronger decreases in cortisol during the social interaction as more expressive of display behaviors. It is also possible that this relationship was only significant in TSST individuals due to these individuals starting at higher cortisol concentrations going into the social interaction task (see Maestripieri et al. 2013, for a discussion of baseline cortisol levels). This result may support that idea that, although psychosocial stress in general may not be related to higher or lower displays of courtship behavior, it is possible that cortisol reactivity may; if individuals are able to show stronger decreases in cortisol responses to psychosocial stress, they are able to display adaptive courtship behaviors more easily.

Finally, this study also assessed potential differences in courtship behaviors among participants based on individual differences that may affect social motivation, such as personality, gender, and relationship status. There were no relationships between gender and courtship behavior overall or between relationship status and courtship behavior. However, some relationships between personality and behavior existed in control participants that were not observed in TSST participants. For male control participants, males who were more extroverted were rated by their conversation partners as expressing higher amounts of display behaviors. Further, males who were more agreeable rated their conversations as more positive and their partners as more attractive, both physically and as potential romantic partners. These relationships did not exist in male TSST participants. For female participants, control females who were more extroverted rated their conversation partners as less attractive; again, this relationship was not found in TSST females.

The correlations found between personality and courtship behaviors were not the same for males and females. It is possible that these differences are related to gender differences in response to stress, as both relationships did not exist in stressed participants. Control females rated participants as more attractive if the participants themselves were more extroverted; this may not be connected to courtship display behaviors, but to agreeableness in general. If females became more socially agreeable under stress, this relationship would disappear. For males, more extroverted and agreeable participants who were not under stress expressed more display behaviors or rated their partners and interactions as more positive overall. It is possible that stress masks the interpretation and behavioral output of potential courtship encounters, albeit differently in males and females.

Overall, though the data presented here may not support the connection between testosterone reactivity and displays of courtship behavior, they do support the idea that psychosocial stress may affect the relationships between personality and courtship behavior. Further, the data support the idea that individuals who experience higher increases in cortisol after psychosocial stress may show less courtship display behavior (see also Wilson et al., 2015, and Ponzi et al., 2016, for similar conclusions). This may have important consequences for individuals who do experience higher psychosocial stress before courtship.

CHAPTER 5

General Discussion

Human social endocrinology research has shown that hormones are sensitive to social stimuli and may show predictable fluctuations in situations in which individuals briefly interact with others. However, we know that during human social interactions, physiology is also interacting simultaneously with cognition and stress responsivity to produce behavioral outputs that function in an adaptively beneficial way. Additionally, individual differences in personality traits may also be interconnected with the physiological and cognitive processes at play during social interactions. In our lab, past studies have shown that by manipulating the physiological and social context during a human social interaction, we can measure how brief social interactions affect endocrine function and behavior in male and female participants. In particular, one social interaction in which endocrine function, cognition, and individual differences may all be interconnected in an important way is a brief social interaction with an opposite sex individual. Specifically, research in our lab has shown that this type of social interaction can elicit an endocrine and behavioral courtship response in male participants (e.g. Roney et al., 2003). Little is still known about how courtship behavior evolved in conjunction with motivation, personality, and social cognition, how psychosocial stress responsivity may alter the effects of hormones and cognition on social interaction behavior, and how these elements have the ability to promote or dampen behaviors or cognitive abilities that facilitate courtship directly.

This study investigated the function of physiological and cognitive changes that may occur during a brief social interaction by manipulating both the ecological stimuli and physiological state of male and female participants. Specifically, this study addressed three central questions: 1) how do brief social interactions with opposite-sex confederates relate to testosterone and cortisol reactivity, and how does psychosocial stress affect this relationship? 2) is there an interaction between brief social interactions with opposite-sex confederates and cognitive performance, and does psychosocial stress affect this interaction or prior cognitive performance going into a social interaction; and 3) what is the relationship between hormonal reactivity to social interactions / psychosocial stress and behavioral output that may be adaptive for courtship, and do individual differences in relationship status, personality, and gender matter to behavioral output after these types of social interactions?

To first address how brief social interactions and psychosocial stress relate to hormonal reactivity, I manipulated both the ecological situation and physiological state of male and female participants before a potential courtship interaction. Specifically, I focused on how testosterone fluctuates to potentially facilitate courtship, how stress responsivity and cortisol may alter the effects of testosterone, and how the relationship between these endocrine responses is moderated by individual differences. Recent research focusing on both baseline and hormonal reactivity during brief social interactions has suggested that the ecological cues of courtship opportunity can influence physiological factors that may promote or subdue courtship behaviors. However, as Chapter 2 illustrates, increases in testosterone mainly occur in single men, not in men in relationships or in women. This effect, however, did not occur in participants who underwent psychosocial stress prior to engaging in a social interaction. Therefore, it appears that psychosocial stress, while not affecting testosterone directly, may play a role in altering later hormonal reactivity to social stimuli. While the exact mechanism remains unclear, it is possible that psychosocial stress may subdue an adaptive physiological response (increase in testosterone) in individuals who are engaged in courtship. However, this relationship is only relevant in individuals who are single, supporting the idea that motivational factors of relevant relationship status may act as a moderating factor in testosterone's role in potentially inducing an adaptive courtship behavioral response. Future studies should continue to focus on the potential moderating factors, whether biological, sociological, or psychological in nature, as there is growing evidence that these factors have the potential to influence the relationship between evolutionarily salient social contexts (e.g., interaction with mates) and adaptive neuroendocrine fluctuations that follow.

To address how brief social interactions and psychosocial stress relate to cognitive performance, in Chapter 3 I described how I further tested males and females on how attentional performance altered before and after brief social interactions, as well as before and after psychosocial stress. Cognitive resources are necessary for impression management, for advertisement of psychological traits that signal strengths as a potential courtship partner (e.g. intelligence, personality, humor), and for use of social cognition skills that allow one to read the interpretations and behaviors of a partner. All of these may have implications for how cognition can affect social behavior. Further, as stress and apprehension have the ability to affect cognition, it is also possible that under evaluative stress, cognitive resources are being diverted in a way that is specifically adaptive or specifically maladaptive during courtship. Although I predicted that both stress and brief social interactions would affect cognitive performance, the data presented in Chapter 3 do not show evidence of changes in cognitive performance before and after a brief social interaction, nor do data show evidence of differences in cognitive reactivity to social interactions in control and stressed participants. Instead, higher subjective anxiety changes and higher increases in cortisol following acute stress in treatment participants led to *improved* cognitive changes. Therefore, although may data may support that idea that subjective and physiological outcomes from acute stress can actually improve attentional resources, brief social interactions do not appear to interact with cognitive performance. Further research focusing on the specific context of stress is needed to clarify how and if stress affects cognition by improving or decreasing attention, particularly in terms of social interactions that induce either ecological cues of courtship and/or cues of psychosocial stress and evaluative threat.

Finally, to address how hormonal reactivity relates to behavioral output that may prove adaptive for successful social relationships initiation and/or courtship, participant behavior was rated by experimenters, and participants were also questioned about their interest in their social interaction partners. As Chapter 4 presents, data from this study did not replicate or find significant relationships between testosterone reactivity and display behavior that may be adaptive to successful courtship in male or female participants. Further, behavioral output that may be specific to courtship (i.e., display and interest) was not generally affected overall by a psychosocial stress treatment. However, the data presented do support the idea that psychosocial stress may present situational changes that affect the relationships between personality and behavior. Specifically, females who were not under psychosocial stress rated participants as more attractive, if the participants themselves were more extroverted; this may not be connected to courtship display behaviors, but to agreeableness in general. For males, more extroverted and agreeable participants who were not under stress expressed more display behaviors or rated their partners and interactions as more positive overall. Further, the data support the idea that individuals who experience higher increases in cortisol after psychosocial stress may show less courtship display behavior. This may have important consequences for individuals who do experience higher psychosocial stress before social interactions that are interpreted as potential courtship opportunities, as well as for key gender differences that exist in the effects of stress on behavioral decision making. Further research could explore ecologically valid social interactions outside the lab and collect more realistic measures of display behavior in humans.

Overall, although this study did not replicate some of the findings of previous research, the data presented here suggest that psychosocial stress and relationship status influence the relationships between social stimuli, hormonal reactivity, and behavior. Crucially, differences in motivation may trigger important, subconscious trade-offs in physiological and behavioral responses to social stimuli. For example, single males and single females who are more motivated to initiate social relationships may experience increases in testosterone to encourage subsequent affiliative or display behavior, while individuals in relationships do not experience this same hormonal reactivity. However, after experiencing psychosocial stress associated with social evaluation threat, the influence of relationship status is masked in such a way as to subdue hormonal increases in response to social stimuli. Therefore, and perhaps most importantly, these data highlight the necessity of considering these individual differences in motivational factors and personality when exploring endogenous changes in hormonal reactivity and behavior within laboratory settings.

Limitations and Future Directions

Although these methods have generated intriguing results, this study could be improved upon in many ways. First, previous laboratory studies of human courtship have used highly attractive confederates, who engaged in friendlier, more deliberately flirtatious conversation. As this study chose to keep the social interaction more neutral, this may have resulted in fewer participants interpreting the encounter as a potential courtship opportunity. This difference may in part account for the discrepancy between my results and those of previous studies. Further, I did not use a control condition in which participants either engaged in conversation with a same sex confederate or simply did not participate in conversation at all.

Second, to assess potential changes in cognitive performance, this study used a fairly simple Flanker task to measure attentional control, and participants were fairly matched on Flanker accuracy, leading to less variation to compare overall. As data showed general increases in task performance throughout the protocol, future studies should use more practice trials to bring participants up to steady performance at baseline. Another option for future research is to use a task that may prove to be more ecologically valid in terms of the connection between cognition and behavioral output. For example, a task that captures social cognition performance, such as measures of primed attitudes or measures of altering of representations of attitudes to match others, may better represent what social cognitive resources are crucial for successful behavioral outputs.

Conclusion

The initiation of human social relationships, whether friendships or romantic relationships, involve some degree of psychosocial stress resulting from social evaluation threat. The impact of such stress may be different on different individuals due to their personality traits, relationship status, and overall motivation to engage in friendly interactions or courtship. Hormonal and cognitive factors, along with situational-environmental variables, are likely to play an important role in the initiation of relationships, as they are related to stress reactivity, motivation, and behavior. The use of a laboratory procedure to induce psychosocial stress prior to a social interaction test provides an opportunity to experimentally manipulate some of these variables and contribute to our understanding of the dynamics of human relationship formation and the underlying biological and psychological mechanisms.

References

Abbey, A. (1982). Sex differences in attributions for friendly behavior: Do males misperceive females' friendliness? *Journal of Personality and Social Psychology*, *42*(5), 830.

Abelson, J. L., Erickson, T. M., Mayer, S. E., Crocker, J., Briggs, H., Lopez-Duran, N. L., & Liberzon, I. (2014). Brief cognitive intervention can modulate neuroendocrine stress responses to the Trier Social Stress Test: Buffering effects of a compassionate goal orientation. *Psychoneuroendocrinology*, *44*, 60-70.

Adan, A., & Almirall, H. (1991). Horne and Östberg morningness-eveningness questionnaire: A reduced scale. *Personality and Individual Differences,* 12, 241-253.

Adler, N., & Stewart, J. (2007). The MacArthur scale of subjective social status. *MacArthur Research Network on SES & Health.* http:// www.macses.ucsf.edu.

Alexander, L. & Martray, C. (1989). The development of an abbreviated version of the Mathematics Anxiety Rating Scale. *Measurement & Evaluation in Counseling & Development*, 22, 143-150.

Andersson, M. B. (1994). *Sexual selection*. Princeton University Press.

Andrews, J., Wadiwalla, M., Juster, R. P., Lord, C., Lupien, S. J., & Pruessner, J. C. (2007). Effects of manipulating the amount of social-evaluative threat on the cortisol stress response in young healthy men. *Behavioural Neuroscience, 121(5),* 871–876.

Archer, J. (2006). Testosterone and human aggression: An evaluation of the challenge hypothesis. *Neuroscience And Biobehavioral Reviews*, *30*(3), 319-345.

Baron-Cohen, S., Wheelwright, S., Skinner, R., Martin, J., & Clubley, E. (2001). The autism-spectrum quotient (AQ): Evidence from asperger syndrome/high-functioning autism, males and females, scientists and mathematicians. *Journal of autism and developmental disorders*, 31(1), 5-17.

Bateup, H. S., Booth, A., Shirtcliff, E. A., & Granger, D. A. (2002). Testosterone, cortisol, and women's competition. *Evolution and Human Behavior*, *23*(3), 181-192.

Boksem, M. S., Kostermans, E., Milivojevic, B., & Cremer, D. D. (2012). Social status determines how we monitor and evaluate our performance. *Social Cognitive and Affective Neuroscience*, *7*(3), 304-313.

Boyle, M. H., Miskovic, V., Van Lieshout, R., Duncan, L., Schmidt, L. A., Hoult, L., ... & Saigal, S. (2011). Psychopathology in young adults born at extremely low birth weight. *Psychological medicine*, *41*(8), 1763-1774.

Buss, D. M. (1989) Sex differences in human mate preferences: Evolutionary hypotheses tested in 37 cultures. Behavioral and Brain Sciences 12:1–49.

Buss, D. M. (2003) The evolution of desire: Strategies of human mating, 4th edition. Basic Books.

Bynum, L., Griffin, T., Riding, D. L., Wynkoop, K. S., Anda, R. F., Edwards, V. J., ... & Croft, J. B. (2010). Adverse childhood experiences reported by adults-five states, 2009. *Morbidity and Mortality Weekly Report*, *59*(49), 1609-1613.

Carré, J. M., & McCormick, C. M. (2008). Aggressive behavior and change in salivary testosterone concentrations predict willingness to engage in a competitive task. *Hormones and Behavior*, *54*(3), 403-409.

Carré, J. M., Putnam, S. K., & McCormick, C. M. (2009). Testosterone responses to competition predict future aggressive behaviour at a cost to reward in men. *Psychoneuroendocrinology*, *34*(4), 561-570.

Casto, K. V., & Prasad, S. (2017). Recommendations for the study of women in hormones and competition research. *Hormones and behavior*, *92*, 190-194.

Chajut, E., & Algom, D. (2003). Selective attention improves under stress: implications for theories of social cognition. *Journal of personality and social psychology*, *85*(2), 231.

Chesser, S. (2013). Cortisol response to the presence of same and opposite sex individuals in various social environments.

Cobey, K., & Hahn, A. (2017). 21 The Endocrinology of Female Competition. *The Oxford Handbook of Women and Competition*, 371.

Crowley, J. P., Denes, A., Makos, S., & Whitt, J. (2018). Threats to Courtship and the Physiological Response: Testosterone Mediates the Association Between Relational uncertainty and Disclosure for Dating Partner Recipients of Relational Transgressions. *Adaptive Human Behavior and Physiology*, *4*(3), 264-282.

de Kloet, E. R., Oitzl, M. S., & Joëls, M. (1999). Stress and cognition: are corticosteroids good or bad guys?. *Trends in neurosciences*, *22*(10), 422-426.

Del Giudice, M., Klimczuk, A., Traficonte, D., & Maestripieri, D. (2014). Autistic-like and schizotypal personality traits in a life history perspective: diametrical associations with impulsivity, sensation-seeking, and sociosexual behavior. *Evolution and Human Behavior, 35*, 415-424.

Dickerson, S. S., & Kemeny, M. E. (2004). Acute stressors and cortisol responses: a theoretical integration and synthesis of laboratory research. *Psychological bulletin*, *130*(3), 355.

Duchesne, A., Tessera, E., Dedovic, K., Engert, V., & Pressier, J. C. (2012). Effects of panel sex composition on the physiological stress responses to psychosocial stress in healthy young men and women. *Biological Psychology, 89*, 99-106.

Eisenegger, C., Haushofer, J., & Fehr, E. (2011). The role of testosterone in social interaction. *Trends in cognitive sciences*, *15*(6), 263-271.

Ellison, P. T. (2009). *On fertile ground: A natural history of human reproduction*. Harvard University Press.

Ellison, P. T., & Gray, P. B., eds. (2009). *Endocrinology of Social Relationships*. Cambridge, MA: Harvard University Press.

Engle, R. W. (2002). Working memory capacity as executive attention. *Current directions in psychological science*, *11*(1), 19-23.

Eriksen, B. A., & Eriksen, C. W. (1974). Effects of noise letters upon the identification of a target letter in a nonsearch task. *Perception & psychophysics*, *16*(1), 143-149.

FeldmanHall, O., Raio, C. M., Kubota, J. T., Seiler, M. G., & Phelps, E. A. (2015). The effects of social context and acute stress on decision making under uncertainty. *Psychological science*, 0956797615605807.

Flinn, M. V., Ponzi, D., & Muehlenbein, M. P. (2012). Hormonal mechanisms for regulation of aggression in human coalitions. *Human nature*, *23*(1), 68-88.

Gaab, J., Blättler, N., Menzi, T., Pabst, B., Stoyer, S., & Ehlert, U. (2003). Randomized controlled evaluation of the effects of cognitive–behavioral stress management on cortisol responses to acute stress in healthy subjects. *Psychoneuroendocrinology*, *28*(6), 767-779.

Gleason, E. D., Fuxjager, M. J., Oyegbile, T. O., & Marler, C. A. (2009). Testosterone release and social context: when it occurs and why. *Frontiers in neuroendocrinology*, *30*(4), 460-469.

Gosling, S. D., Rentfrow, P. J., & Swann Jr, W. B. (2003). A very brief measure of the Big-Five personality domains. *Journal of Research in personality*, *37*(6), 504-528.

Gray, P. B., Chapman, J. F., Burnham, T. C., McIntyre, M. H., Lipson, S. F., & Ellison, P. T. (2004). Human male pair bonding and testosterone. *Human Nature*, *15*(2), 119-131.

Goldey, K. L. (2015). Multi-Method Approaches to Understanding Bidirectional Links between Sexuality and Testosterone in Women.

Goldey, K. L., & van Anders, S. M. (2011). Sexy thoughts: Effects of sexual cognitions on testosterone, cortisol, and arousal in women. *Hormones and Behavior*, *59*(5), 754-764.

Gruenewald, T. L., Kemeny, M. E., & Aziz, N. (2006). Subjective social status moderates cortisol responses to social threat. *Brain, Behavior, And Immunity*, *20*(4), 410-419.

Guez, J., Saar-Ashkenazy, R., Keha, E., & Tiferet-Dweck, C. (2016). The effect of Trier Social Stress Test (TSST) on item and associative recognition of words and pictures in healthy participants. *Frontiers in psychology*, *7*, 507.

Helbig, S., & Backhaus, J. (2017). Sex differences in a real academic stressor, cognitive appraisal and the cortisol response. *Physiology & behavior*, *179*, 67-74.

Henry, A., Sattizhan, J. R., Norman, G. J., Beilock, S. L., & Maestripieri, D. (2017). Performance during competition and competition outcome in relation to testosterone and cortisol among women. *Hormones and Behavior, 92*, 82-92.

Higham, J. P., Heistermann, M., & Maestripieri, D. (2013). The endocrinology of male rhesus macaque social and reproductive status: a test of the challenge and social stress hypotheses. *Behavioral Ecology and Sociobiology, 67*, 19-30.

Horne, J. A., & Östberg, O. (1976). A self-assessment questionnaire to determine morningness-eveningness in human circadian rhythms. *International Journal of Chronobiology,* 4, 97-110.

Jackson, J. J., & Kirkpatrick, L. A. (2007). The structure and measurement of human mating strategies: toward a multidimensional model of sociosexuality. *Evolution and Human Behavior*, *28*, 382-391.

Jamieson, J. P., Nock, M. K., & Mendes, W. B. (2012). Mind over matter: Reappraising arousal improves cardiovascular and cognitive responses to stress. *Journal of Experimental Psychology: General*, *141*(3), 417.

Jaremka, L. M., & Collins, N. L. (2017). Cortisol increases in response to brief social exchanges with opposite sex partners. *Biological psychology*, *124*, 39-46.

John, O. P., Donahue, E. M., & Kentle, R. L. (1991). *The Big Five Inventory-Versions 4a and 54.* Berkeley, CA: University of California, Berkeley, Institute of Personality and Social Research.

Karremans, J. C., & Verwijmeren, T. (2008). Mimicking attractive opposite-sex others: The role of romantic relationship status. *Personality and Social Psychology Bulletin*, *34*(7), 939-950.

Karremans, J. C., Verwijmeren, T., Pronk, T. M., & Reitsma, M. (2009). Interacting with women can impair men's cognitive functioning. *Journal of Experimental Social Psychology*, *45*(4), 1041-1044.

Kelly, M. M., Tyrka, A. R., Anderson, G. M., Price, L. H., & Carpenter, L. L. (2008). Sex differences in emotional and physiological responses to the Trier Social Stress Test. *Journal of behavior therapy and experimental psychiatry*, *39*(1), 87-98.

Kim, E., Nickels, N. & Maestripieri, D. (2018). Effects of brief interactions with male experimenters shortly before and during the trier social stress test on study participants' testosterone salivary concentrations. *Adaptive Human Behavior and Physiology*, *4*, 329-343.

Kirschbaum, C., Pirke, K. M., & Hellhammer, D. H. (1993). The 'Trier Social Stress Test'–a tool for investigating psychobiological stress responses in a laboratory setting. *Neuropsychobiology*, *28*(1-2), 76-81.

Kirschbaum, C., & Hellhammer, D. H. (1994). Salivary cortisol in psychoneuroendocrine research: recent developments and applications. *Psychoneuroendocrinology*, *19*(4), 313-333.

Kirschbaum, C., Wolf, O. T., May, M., Wippich, W., & Hellhammer, D. H. (1996). Stress-and treatment-induced elevations of cortisol levels associated with impaired declarative memory in healthy adults. *Life sciences*, *58*(17), 1475-1483.

Kuhlmann, S., Piel, M., & Wolf, O. T. (2005). Impaired memory retrieval after psychosocial stress in healthy young men. *Journal of Neuroscience*, *25*(11), 2977-2982.

Lighthall, N. R., Mather, M., & Gorlick, M. A. (2009). Acute stress increases sex differences in risk seeking in the balloon analogue risk task. *PLoS One*,*4*(7), e6002.

Liu, J. J. W., Ein, N., Peck, K., Huang, V., Pruessner, J. C., & Vickers, K. (2017). Sex differences in salivary cortisol reactivity to the Trier Social Stress Test (TSST): A meta-analysis. *Psychoneuroendocrinology*, *82*, 26-37.

López, H. H., Hay, A. C., & Conklin, P. H. (2009). Attractive men induce testosterone and cortisol release in women. *Hormones and Behavior*, *56*(1), 84-92.

Lord, F. M. (1963). Elementary models for measuring change. *Problems in measuring change*, 21-38.

Lupien, S. J., Maheu, F., Tu, M., Fiocco, A., & Schramek, T. E. (2007). The effects of stress and stress hormones on human cognition: Implications for the field of brain and cognition. *Brain and cognition*, *65*(3), 209-237.

Lupien, S. J., McEwen, B. S., Gunnar, M. R., & Heim, C. (2009). Effects of stress throughout the lifespan on the brain, behaviour and cognition. *Nature reviews neuroscience*, *10*(6), 434.

Maestripieri, D. (2014). Night owl women are similar to men in their relationship orientation, risk-taking propensities, and cortisol levels: implications for the adaptive significance and evolution of eveningness. *Evolutionary Psychology, 12*, 130-147.

Maestripieri, D., Baran, N. M., Sapienza, P., & Zingales, L. (2010). Between- and within-sex variation in hormonal responses to psychological stress in a large sample of college students. *Stress, 13*, 413-424.

Maestripieri, D., Klimczuk, A., Seneczko, M., Traficonte, D., & Wilson, M. C. (2013). Relationship status and relationship instability, but not dominance, predict individual differences in baseline cortisol levels. *PLoS One, 8(12):* e84003.

Maestripieri, D., Klimczuk, A., Traficonte, D., & Wilson, M. C. (2014). Ethnicity-related variation in sexual promiscuity, relationship status and testosterone levels in men. *Evolutionary Behavioral Sciences, 8*, 96-108.

Marvel-Coen, J., Nickels, N., & Maestripieri, D. (2018). The relationship between morningness-eveningness, psychosocial variables, and cortisol reactivity to stress from a life history perspective. *Evolutionary Behavioral Sciences, 12*, 71-86.

Mather, M., & Lighthall, N. R. (2012). Risk and reward are processed differently in decisions made under stress. *Current directions in psychological science*, *21*(1), 36-41.

Mazur, A. (1985). A biosocial model of status in face-to-face primate groups. *Social Forces*, *64*(2), 377-402.

Mazur, A. & Booth, A. (1998). Testosterone and dominance in men. *Behavioral And Brain Sciences*, *21*(3), 353-397.

McEwen, B. S., & Sapolsky, R. M. (1995). Stress and cognitive function. *Current opinion in neurobiology*, *5*(2), 205-216.

Mehta, P. H., Jones, A. C., & Josephs, R. A. (2008). The social endocrinology of dominance: Basal testosterone predicts cortisol changes and behavior following victory and defeat. *Journal of Personality and Social Psychology, 94*, 1078–1093.

Miller, S. L., Maner, J. K., & McNulty, J. K. (2012). Adaptive attunement to the sex of individuals at a competition: the ratio of opposite-to same-sex individuals correlates with changes in competitors' testosterone levels. *Evolution and Human Behavior*, *33*(1), 57-63.

Morier, D., & Seroy, C. (1994). The effect of interpersonal expectancies on men's self-presentation of gender role attitudes to women. *Sex Roles*, *31*(7-8), 493-504.

Murcia, C. Q., Bongard, S., & Kreutz, G. (2009). Emotional and neurohumoral responses to dancing tango argentino the effects of music and partner. *Music and Medicine*, *1*(1), 14-21.

Nauts, S., Metzmacher, M., Verwijmeren, T., Rommeswinkel, V., & Karremans, J. C. (2012). The mere anticipation of an interaction with a woman can impair men's cognitive performance. *Archives of Sexual Behavior*, *41*(4), 1051-1056.

Nickels, N., Kubicki, K., & Maestripieri, D. (2017). Sex differences in the effects of psychosocial stress on cooperative and prosocial behavior: evidence for 'flight-or-fight' in males and 'tend-and-befriend' in females. *Adaptive Human Behavior and Physiology, 3*, 171-183.

Peters, B. J., Hammond, M. D., Reis, H. T., & Jamieson, J. P. (2016). The consequences of having a dominant romantic partner on testosterone responses during a social interaction. *Psychoneuroendocrinology*, *74*, 308-315.

Piffer, D., Ponzi, D., Sapienza, P., Zingales, L., & Maestripieri, D. (2014). Morningness-eveningness and intelligence among high-achieving US students: night owls have higher GMAT scores than early morning types in a top-ranked MBA program. *Intelligence, 47,* 107-112.

Pilgrim, K., Ellenbogen, M. A., & Paquin, K. (2014). The impact of attentional training on the salivary cortisol and alpha amylase response to psychosocial stress: Importance of attentional control. *Psychoneuroendocrinology*, *44*, 88-99.

Ponzi, D., Henry, A., Kubicki, K., Nickels, N., Wilson, M. C., & Maestripieri, D. (2015). The slow and fast life histories of early birds and night owls: Their future- or present-orientation accounts for their sexually monogamous or promiscuous tendencies. *Evolution and Human Behavior, 36*, 117-122.

Ponzi, D., Henry, A., Kubicki, K., Nickels, N., Wilson, M. C., & Maestripieri, D. (2016). Autistic-like traits, socio-sexuality, and hormonal responses to socially stressful and sexually arousing stimuli in male students. *Adaptive Human Behavior and Physiology, 2*, 150-165.

Ronay, R., & Hippel, W. V. (2010). The presence of an attractive woman elevates testosterone and physical risk taking in young men. *Social Psychological and Personality Science*, *1*(1), 57-64.

Roney, J. R. (2003). Effects of visual exposure to the opposite sex: Cognitive aspects of mate attraction in human males. *Personality and Social Psychology Bulletin*, *29*(3), 393-404.

Roney, J. R. (2009). The role of sex hormones in the initiation of human mating relationships. *The endocrinology of social relationships*, 246-269.

Roney, J. R. (2015). Evolutionary psychology and endocrinology. *The Handbook of Evolutionary Psychology*.

Roney, J. R. (2016). Theoretical frameworks for human behavioral endocrinology. *Hormones and behavior*, *84*, 97-110.

Roney, J. R., & Gettler, L. T. (2015). The role of testosterone in human romantic relationships. *Current Opinion in Psychology*, *1*, 81-86.

Roney, J. R., Mahler, S. V., & Maestripieri, D. (2003). Behavioral and hormonal responses of men to brief interactions with women. *Evolution and human Behavior*, *24*(6), 365-375.

Roney, J. R., Lukaszewski, A. W., & Simmons, Z. L. (2007). Rapid endocrine responses of young men to social interactions with young women. *Hormones and Behavior*, *52*(3), 326-333.

Roney, J. R., Simmons, Z. L., & Lukaszewski, A. W. (2010). Androgen receptor gene sequence and basal cortisol concentrations predict men's hormonal responses to potential mates. *Proceedings of the Royal Society of London B: Biological Sciences*, *277*(1678), 57-63.

Russell, D., Peplau, L. A., & Cutrona, C. E. (1980). The revised UCLA Loneliness Scale: Concurrent and discriminant validity evidence. *Journal of personality and social psychology*, *39*(3), 472.

Sanders, A. F., & Lamers, J. M. (2002). The Eriksen flanker effect revisited. *Acta Psychologica*, *109*(1), 41-56.

Sandi, C. (2013). Stress and cognition. *Wiley Interdisciplinary Reviews: Cognitive Science*, *4*(3), 245-261.

Sapolsky, R. M. (2002). 11 Endocrinology of the Stress-Response. *Behavioral endocrinology*, 409.

Sato, H., Takenaka, I., & Kawahara, J. I. (2012). The effects of acute stress and perceptual load on distractor interference. *Quarterly journal of experimental psychology*, *65*(4), 617-623.

Shields, G. S., Sazma, M. A., & Yonelinas, A. P. (2016). The effects of acute stress on core executive functions: a meta-analysis and comparison with cortisol. *Neuroscience & Biobehavioral Reviews*, *68*, 651-668.

Simpson, J. A., Gangestad, S. W., & Lerma, M. (1990). Perception of physical attractiveness: Mechanisms involved in the maintenance of romantic relationships. *Journal of Personality and Social Psychology*, *59*(6), 1192.

Shotland, R. L., & Craig, J. M. (1988). Can men and women differentiate between friendly and sexually interested behavior?. *Social Psychology Quarterly*, 66-73.

Smeets, T., Dziobek, I., & Wolf, O. T. (2009). Social cognition under stress: differential effects of stress-induced cortisol elevations in healthy young men and women. *Hormones and Behavior*, *55*(4), 507-513.

Spielberger, C. D., & Gorsuch, R. L. (1983). *State-Trait Anxiety Inventory for Adults: Manual and Sample: Manual, Instrument and Scoring Guide*. Consulting Psychologists Press.

Starcke, K., & Brand, M. (2012). Decision making under stress: a selective review. *Neuroscience & Biobehavioral Reviews*, *36*(4), 1228-1248.

Taylor, S. E., Klein, L. C., Lewis, B. P., Gruenewald, T. L., Gurung, R. A., & Updegraff, J. A. (2000). Biobehavioral responses to stress in females: tend-and-befriend, not fight-or-flight. *Psychological review*, *107*(3), 411.

van Anders, S., & Watson, N. V. (2006). Social neuroendocrinology. *Human Nature, 17(2*), 212-237.

van den Bos, R., Harteveld, M., & Stoop, H. (2009). Stress and decision-making in humans: performance is related to cortisol reactivity, albeit differently in men and women. *Psychoneuroendocrinology*, *34*(10), 1449-1458.

van der Meij, L., Buunk, A. P., van de Sande, J. P., & Salvador, A. (2008). The presence of a woman increases testosterone in aggressive dominant men. *Hormones and Behavior*, *54*(5), 640-644.

van der Meij, L., Buunk, A. P., & Salvador, A. (2010). Contact with attractive women affects the release of cortisol in men. *Hormones and Behavior*, *58*(3), 501-505.

Vohs, K. D., Baumeister, R. F., & Ciarocco, N. J. (2005). Self-regulation and self-presentation: regulatory resource depletion impairs impression management and effortful self-presentation depletes regulatory resources. *Journal of personality and social psychology*, *88*(4), 632.

Von Dawans, B., Fischbacher, U., Kirschbaum, C., Fehr, E., & Heinrichs, M. (2012). The social dimension of stress reactivity: acute stress increases prosocial behavior in humans. *Psychological science*, *23*(6), 651-660.

Wadiwalla, M., Andrews, J., Lai, B., Buss, C., Lupien, S. J., & Pruessner, J. C. (2010). Effects of manipulating the amount of social-evaluative threat on the cortisol stress response in young healthy women. *Stress, 13(3),* 214–220.

Wilson, M., & Daly, M. (2004). Do pretty women inspire men to discount the future?. *Proceedings of the Royal Society of London B: Biological Sciences*, *271*(Suppl 4), S177-S179.

Wilson, M. C., Zilioli, S., Ponzi, D., Henry, A., Kubicki, K., Nickels, N., & Maestripieri, D. (2015). Cortisol reactivity to psychosocial stress mediates the relationship between extraversion and unrestricted sociosexuality. *Personality and Individual Differences, 86*, 427-431.

Wingfield, J. C., Hegner, R. E., Dufty Jr, A. M., & Ball, G. F. (1990). The" challenge hypothesis": theoretical implications for patterns of testosterone secretion, mating systems, and breeding strategies. *The American Naturalist*, *136*(6), 829-846.

Wolf, O. T., Schommer, N. C., Hellhammer, D. H., McEwen, B. S., & Kirschbaum, C. (2001). The relationship between stress induced cortisol levels and memory differs between men and women. *Psychoneuroendocrinology*, *26*(7), 711-720

Zanna, M. P., & Pack, S. J. (1975). On the self-fulfilling nature of apparent sex differences in behavior. *Journal of Experimental Social Psychology*, xxx

Zilioli, S., & Bird, B. M. (2017). Functional significance of men's testosterone reactivity to social stimuli. *Frontiers in neuroendocrinology*, *47*, 1-18.

Zilioli, S., Ponzi, D., Henry, A., Kubicki, K., Nickels, N., Wilson, M. C., & Maestripieri, D. (2016). Interest in babies negatively predicts testosterone responses to sexual visual stimuli among heterosexual young men. *Psychological Science, 27*, 114-118.

Zilioli, S., & Watson, N. V. (2012). The hidden dimensions of the competition effect: Basal cortisol and basal testosterone jointly predict changes in salivary testosterone after social victory in men. *Psychoneuroendocrinology, 37,* 1855–1865.