Effects of Menstrual Cycle Phase and Stress on Spatial Navigational Strategy Use

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COMPULSORY DECLARATION

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List of Abbreviations

ANOVA	-	Analysis of variance
BMI	-	Body Mass Index
BP	-	Blood Pressure
CG	-	Computer-Generated
HPA	-	Hypothalamic-Pituitary-Adrenal
HPG	-	Hypothalamic-Pituitary-Gonadal
HR	-	Heart Rate
MWM	-	Morris Water Maze
PASAT	-	Paced Auditory Serial Addition Task
PSS	-	Perceived Stress Scale
RAM	-	Radial Arm Maze
SAM	-	Sympathetic-Adrenal-Medullary
SBSODS	-	Santa Barbara Sense of Direction Scale
SRPP	-	Student Research Participation Programme
STAI	-	State-Trait Anxiety Inventory
UCT	-	University of Cape Town
u-PASAT	-	Unpaced Auditory Serial Addition Task
VE	-	Virtual Environment
VR	-	Virtual Reality

List of Key Terms

Allocentric Strategy:	Navigation strategy relying on one's cognitive map of their environment.
Cue:	Specific object or landmark within the environment.
Early Follicular Phase:	Phase of the menstrual cycle that is categorized by low levels of estrogen and progesterone. Typically, days 1-7 post menstruation.
Egocentric-Cue Strategy:	Navigation strategy relying on the location of a specific cue in the environment in relation to the target location
Egocentric-Response Strategy:	Navigation strategy relying on the direction of one's body turns.
Estrogen:	Primary female sex hormone responsible for regulation of female reproductive system and secondary sex characteristics.
Glucocorticoids:	Stress hormone released during the HPA-axis stress response; commonly known as cortisol.
HPA-axis:	Slow-acting stress-response system regulated by the hypothalamus.
Menstrual Cycle:	The monthly cycle involving fluctuations of female sex hormone levels to prepare the body for pregnancy.
Mid/Late Luteal Phase:	Phase of the menstrual cycle pre menstruation that is categorized by moderate estrogen levels and high levels of progesterone. Typically, days 20-last day of cycle.
Navigation Strategy:	The method an organism uses to navigate within their environment.

Ovulatory Phase:	Phase of the menstrual cycle around ovulation that is categorized by high levels of estrogen and low levels of progesterone. Typically, days 13-17 post menstruation.
Progesterone:	Female sex hormone responsible for the regulation of the menstrual cycle and early stages of pregnancy.
SAM-axis:	Fast-acting stress-response system regulated by the sympathetic nervous system.
Spatial Memory:	Memory system underlying the formation of the cognitive maps need for allocentric navigation.
Stimulus-Response Memory:	Memory system underlying egocentric navigation based on the relationships between specific objects and target locations.
Stress:	Body’s physiological response to a threat.
Stressor:	An event an organism appraises as threatening.

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Abstract

Previous research suggests there are significant effects on spatial navigation of menstrual cycle phase and, independently, of stress exposure. However, no published study explores how these variables might interact to affect human navigational strategies. Such exploration is important because women experience stressful events throughout the menstrual cycle. Hence, the primary aim of the research described here was to investigate the potentially interacting effect of menstrual cycle phase and acute psychosocial stress on allocentric (i.e., relying on a cognitive map of the environment, created using distal cues), egocentric-response (i.e., relying on body-turn directions), and egocentric-cue (i.e., relying on the location of specific proximal cues) navigation. The study tested three hypotheses: (1) menstrual cycle phase will significantly affect navigational strategy (women in the early follicular and ovulatory phases will prefer egocentric strategies, whereas those in the mid/late luteal phase will prefer an allocentric strategy); (2) stress will significantly affect navigational strategy (women exposed to an acute psychosocial stressor will prefer egocentric strategies while those unexposed will prefer an allocentric strategy); and (3) there will be a significant interaction effect on navigational strategies (particularly, women in the mid/late luteal phase will use egocentric strategies when stressed and allocentric strategies when not). Naturally-cycling young adult women (N = 60), each in either the early follicular, ovulatory, or mid/late luteal phase of the cycle, were randomly assigned to either a stress-exposure condition (the Paced Auditory Serial Addition Task [PASAT], which required them to rapidly add pairs of verbally presented digits at increasing between-digit intervals) or an equivalent control condition (the *un*-PASAT, which required them to add pairs of digits at a constant, slower, between-digit speed). Hence, the study groups were: Early Follicular+Stress (n = 11), Early Follicular+Non-Stress (n = 10), Ovulatory+Stress (n = 8), Ovulatory+Non-Stress (n = 8), Mid/Late Luteal+Stress (n = 12), and Mid/Late Luteal+Non-Stress (n = 11). After the manipulation, they completed the Hex Maze, a virtual environment navigation task. The task comprises 10 trial pairs. On the first trial of a pair, participants learn a target platform's location. On the second, they must navigate back to that location. This latter trial can be completed using either an allocentric, egocentric-response, or egocentric-cue strategy. Results suggested that the PASAT successfully induced psychological and physiological stress. A mixed linear model analysis detected, with regard to the navigational strategy outcome variables, no significant main effect of menstrual cycle phase (a result inconsistent with previous research), no significant main effect of stress exposure (a result consistent with some prior research), and no significant interaction effect. Although the interaction was non-

significant, trends within the data suggested the results followed the predicted direction. Hence, this relatively small-scale study may serve as a foundation for future research as it provides valuable hypothesis-generating insight into the possible interaction of menstrual cycle phase and stress, and encourages the further investigation of this topic.

CHAPTER ONE:

GENERAL INTRODUCTION

A small body of research on spatial cognition investigates how female performance may change under different physiological or environmental conditions (e.g., across different phases of the menstrual cycle, or in the presence of stress-inducing stimuli). For instance, some studies show that activational effects of female sex hormones, as they vary across the menstrual cycle, have a distinct influence on spatial cognitive performance (Cashdan & Gaulin, 2016; Hausmann, Slabbekoom, Van Goozen, Cohen-Kettenis, & Guntiirkun, 2000; McCormick & Teillon, 2001). As several independent reviews indicate, however, most of those studies investigate spatial cognitive performance using mental rotation and similar non-navigational tasks (Hampson, 2018; Voyer, Voyer, & Bryden, 1995; Zhu, Kelly, Curry, Lal, & Joseph, 2015). Few previously published studies share the specific focus of this thesis: how women's spatial navigational strategies might vary across the menstrual cycle (i.e., as levels of sex hormones change) and under different conditions of stress. The absence of this research focus is particularly puzzling given that navigating through one's environment is a necessary skill for survival and is fundamental in any organism's attempts to successfully interact with the external world (O'Keefe & Nadel, 1978).

Psychological research has identified two main types of navigational strategies: a spatial *allocentric* type and a non-spatial *egocentric* type (Iaria, Petrides, Dagher, Pike, & Bohbot, 2003). The latter is further divided into two distinct strategies – egocentric-response strategy and egocentric-cue. Women tend to use an egocentric strategy during the early follicular and ovulatory phases of their menstrual cycle, and the allocentric strategy during the mid/late luteal phase (Hussain, Hanafi, Konishi, Brake, & Bohbot, 2016; Scheuringer & Pletzer, 2017). However, studies demonstrating this data pattern do not account for the potential moderating effects of environmental factors on the relationship between menstrual cycle phase and navigational strategy use.

One such environmental factor is the stress being experienced by naturally-cycling women as they make spatial decisions and use their spatial abilities to navigate their immediate environment. Broadly speaking, only a handful of studies investigate the relationship between stress and navigational strategy use. This line of research is marked by contradictory results. Whereas some studies suggest that exposure to stress increases the use of allocentric strategies, others suggest it increases the use of egocentric strategies (Thomas, Laurance, Nadel, & Jacobs, 2010; van Gerven, Ferguson, & Skelton, 2016). A universal

limitation of these studies, however, is that they do not account for the menstrual cycle phase of their female participants and thus fail to provide real-life insight into the interaction between the presence of environmental stress and the cycle phase in influencing choice of navigational strategy.

Additionally, all of the currently published studies in this literature only distinguish between the allocentric strategy and one egocentric strategy (typically, the response strategy). This is because they use measures based on popular rodent tasks (e.g., the Morris Water Maze and the Radial Arm Maze). To date, only two studies has been published using a measure, the Hex Maze, that allows concurrent investigation of all three strategies (Ferguson, Williams, Skelton, & Krigolson, 2019; Spriggs, Kirk, & Skelton, 2018).

Given that variations in levels of female sex hormones across the menstrual cycle do not occur independently of external environmental factors (and, specifically, that women at any phase of their cycle may encounter stress-provoking events), more research should investigate the influence of these factors on the hormone-related differences commonly observed in cognition and behaviour. Hence, the research presented in this thesis investigates the novel question of whether navigational strategy use is affected by an interaction between exposure to acute psychosocial stress and menstrual cycle phase. This chapter briefly introduces the topic and describes the structure of the overall thesis. Chapter Two presents a concise but detailed literature review while Chapter Three presents the rationale and hypotheses for the research project. Chapter Four reports the pilot study conducted in preparation for the major study presented by the thesis. Chapter Five describes the methods used in that major study, and Chapter Six presents its results. Finally, Chapter Seven presents a general discussion and the final conclusions of the thesis.

CHAPTER TWO:
LITERATURE REVIEW

Separate bodies of literature investigate the effects of endocrine factors (including menstrual cycle phase) and psychological stress on spatial navigation performance (e.g., efficiency of navigation, choice of navigational strategy) . However, no published study has investigated the possible *interacting effects* of menstrual cycle phase and psychological stress on human spatial navigation. Because that interaction is the focus of the research, this literature review aims to situate the current investigation within the current body of literature and to demonstrate why it is important to explore the possible synergistic effects of menstrual cycle phase and stress on navigational strategy.

Because spatial navigation in humans and rodents relies on similar memory systems (i.e., broadly speaking, both spatial and response systems) that engage similar brain regions (i.e., the hippocampus and caudate nucleus, respectively; Iaria et al., 2003; White & McDonald, 2002), this literature review discusses relevant rodent studies alongside the pertinent human studies.

The structure of the review is as follows. First, I define and explain the two main types of strategies (viz., egocentric and allocentric) that humans use in their everyday navigation. Then, I review the separate strands of literature that explore the effects of menstrual cycle phase and of stress on the choice and use of navigational strategy. Thereafter, I briefly discuss published studies that have examined interacting effects of menstrual cycle phase and stress on similar cognitive functions (viz., emotional learning and declarative memory). That section serves to highlight the need for spatial navigation research examining the interaction effect. The review concludes with a summary statement of the way in which previous research provides a rationale for conducting the current study.

Egocentric and Allocentric Navigational Strategies

The psychological literature defines the term *navigational strategy* as the methods an organism uses to drive purpose-driven movement within its environment (Iaria et al., 2003; O'Keefe & Nadel, 1978; Roche, Mangaoang, Commins, & O'Mara, 2005). Human navigational strategies are conventionally divided into two main categories: non-spatial *egocentric navigation* and spatial *allocentric navigation.* These are distinct at both cognitive and neural levels (i.e., they use different frames of reference, have different underlying memory systems, and rely on interaction within and between different brain regions).

Egocentric navigation uses the organism’s location as a reference point. Naturally, then, this reference point changes as the organism’s perspective changes. A non-spatial

stimulus-response memory system underlies this type of navigation because, here, directed movement toward a target location relies on distances to a single environmental landmark or on learning a series of body turns (Iaria et al., 2003). Both humans and rodents use this memory system to navigate non-spatially (White & McDonald, 2002).

The broad egocentric category comprises two distinct sub-strategies (Foreman & Gillet, 1997; Hussain et al., 2016; Spriggs et al., 2018). An *egocentric-cue strategy* uses subject-to-object relationships in its reference frames and relies on the locations of specific cues within the external environment. In contrast, an *egocentric-response strategy* uses the direction of body turns to guide navigation within the environment and does not involve the encoding of environmental information. Early studies in the field strongly implicated the caudate nucleus in the cognitive processing underlying egocentric strategies generally (Iaria et al., 2003; Nadel & Hardt, 2004; Packard, Hirsh, & White, 1989). More recent neuroimaging studies suggest, however, that parietal lobe processing is important when the egocentric-cue strategy is used whereas the striatum is more active when the egocentric-response strategy is used (Burgess, 2008; Chersi & Burgess, 2015).

Allocentric (i.e., world-centered frame of reference) spatial navigation relies on cognitive processing involving the formation of stimulus-stimulus associations between notable environmental landmarks. These associations are the foundation upon which both humans and rodents build cognitive maps of the environment (O'Keefe & Nadel, 1978; Sholl, 1996; White & McDonald, 2002). When using an allocentric navigational strategy, the perspective of the navigator is unnecessary for the encoding of environmental information. Hence, the strategy is independent of the person's location within the environment. The hippocampus has been strongly implicated in cognitive processing that underlies the allocentric strategy and its associated spatial memory system (Eichenbaum, 2017; Maguire, Burgess, & O'Keefe, 1999; Nadel & Hardt, 2004).

Many neuroimaging and lesion studies support the notion that the hippocampus is critically involved in the cognitive processes underlying allocentric spatial navigation (see, e.g., Bartsch et al., 2010; Bohbot, Lerch, Thorndycraft, Iaria, & Zijdenbos, 2007; Nedelska et al., 2012; Parslow et al., 2004; Zaehle et al., 2007). Recently, Guderian et al. (2015) investigated performance on an allocentric spatial memory task across patients with severe hippocampal damage (SHD), patients with moderate hippocampal damage (MHD), and healthy controls. Their task was based within a virtual environment (VE) that contained a circular arena and distal cues that could provide orientation details. During trials comprising the learning phase, a circular boundary and a landmark within the arena provided spatial

reference frames while participants navigated toward a target object. This phase consisted of 5 blocks comprising four trials each. The first block of four trials featured a different target object on each trial. Although each target was in a different location, the participant's instruction on each trial was the same: Move from your starting location toward the object and collect it. On subsequent learning trials (i.e., those contained within blocks 2-5), participants first viewed a static display of one of the four previously-seen target objects in front of a grey background. They then entered the arena from varying locations and were instructed to navigate toward the location where the displayed object had been placed during Block 1. There were 16 such trials (four blocks of four, with each block featuring each of the four target objects only once).

The subsequent test phase consisted of four blocks of four trials each. Within each block, recall for two of the four targets was tested. The participants' starting location was different across each trial within a block. Furthermore, each block featured either the circular boundary or the proximal landmark but not both. During each trial of this phase, participants were again instructed to navigate to the target's location and to press the space bar upon reaching it.

Results showed that, compared to the other two groups, SHD patients were markedly impaired in both boundary-based and landmark-based recall of object locations. The performance of MHD patients did not differ significantly from that of controls. Moreover, performance on both types of recall task was strongly related to bilateral hippocampal volume but not to volume of the other brain regions (e.g., thalamus, caudate nucleus). Taken together, these results supported the authors' conclusions that the observed deficits in allocentric spatial recall were best explained by reduced bilateral hippocampal volume in SHD participants.

Measures of Navigational Strategies

Previous research has modified widely-used rodent tasks (e.g., the radial arm maze [RAM; Olton & Samuelson, 1976] or the Morris water maze [MWM; Morris, 1984]) to investigate human navigational strategies. In the rodent literature, a RAM typically features a number of identical arms (six or eight, depending on the task) radiating outwards from a central circle (see Figure 1, panel (a)). Food is placed at the end of at least one of the arms and the animal is tasked with learning, over multiple training trials, where the food is. In the paper describing the development of the RAM, Olton and Samuelson (1976) note that the arms do not serve as distinct navigational cues given their identical appearance and equal dimensions. Instead, the external testing room contains numerous visual distal cues (i.e., a

sink, a table) that allow rats to orient themselves and to learn the location of the arm containing the food. The extent to which the animal remembers which arm contains food is a measure of its spatial memory.

The MWM, in contrast, assesses spatial memory by placing the animal into a circular pool of cool, milky water. It can escape from the pool by locating and then standing on a platform that is submerged beneath the water's surface (see Figure 1, panel (b)). Once the animal stands on the platform it rises above the surface and the animal is released from the pool. With training, it is expected that the animal will, irrespective of its starting location, learn to use distal cues in the surrounding testing room to navigate directly to the escape platform. Hence, the task traditionally involves learning and testing trials. The learning trials allow the rodent to learn the location of the escape platform as it remains visible. During the testing trials, however, the platform is submerged below the surface and the animal is required to navigate back to the location of the now-invisible escape platform. Latency and path length to reach the escape platform are the conventional measures of spatial memory in the MWM.

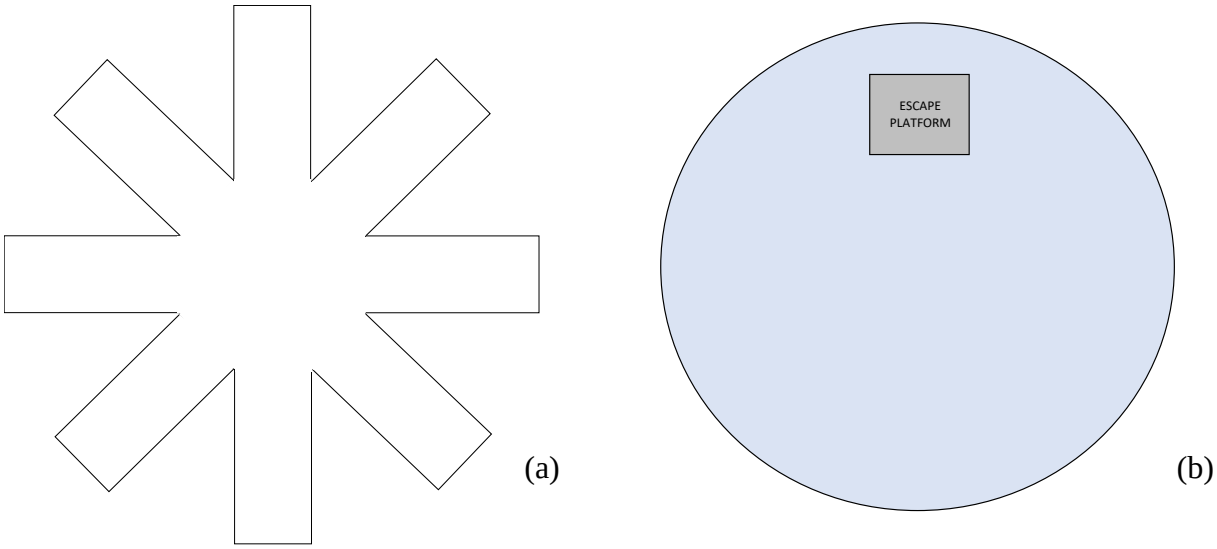


Figure 1. Diagrammatic representations of (a) the Radial Arm Maze (RAM) and (b) the Morris Water Maze (MWM).

These paradigms have been adapted into VE tasks that allow investigation into human spatial navigation. For instance, the Computer-Generated (CG) Arena (Jacobs, Laurance, & Thomas, 1997; Jacobs, Thomas, Laurance, & Nadel, 1998; Thomas, Hsu, Laurance, Nadel, & Jacobs, 2001) is based on the apparatus and procedure of an MWM task. Specifically, the participant completes the task in a VE that features a circular arena housed within a square

room. Each of the four walls in the room is marked by distinct environmental cues. A square target is located in a fixed place on the floor of the circular arena. This target is invisible until the participant walks over its location. Just as in the MWM, the procedure requires the participant to locate the target, learn its location, and return to that location repeatedly over a series of acquisition trials. Immediately after the acquisition trials, participants complete a probe trial that requires them to navigate back to the target location. However, they are not informed that the target has been removed from the arena, and so the probe trial measures their accuracy of target knowledge and persistence of search. In the CG Arena, as in the standard rodent MWM task, strategy use is determined partially by path taken to reach target location during the probe trial. An allocentric strategy would be indicated by a relatively short and direct path to the target location, irrespective of the starting location, whereas an egocentric-response strategy would be marked by a relatively long and indirect path that most likely stops in a different location to the desired target location, depending on the participant's starting location. The CG Arena environment can be modified to include proximal cues along the circular arena wall. In such a modified environment, participants using an egocentric-cue strategy would follow a path marked by a specific proximal cue, regardless of whether its location (relative to the distal cues in the environment) changes across trials. The CG Arena has been used to investigate the effects of acute psychosocial stress, traumatic brain injuries, and mild Alzheimer's disease on various aspects of spatial navigation (Jheng & Pai, 2009; Skelton, Bukach, Laurance, Thomas, & Jacobs, 2000; Thomas et al., 2010).

Virtual radial maze tasks, based on the apparatus and procedure of a standard rodent RAM, have also facilitated investigation into how sex differences and hippocampal damage affect spatial memory and spatial navigation (Andersen, Dahmani, Konishi, & Bohbot, 2012; Astur, Tropp, Sava, Constable, & Markus, 2004; Goodrich-Hunsaker & Hopkins, 2010). Bohbot et al. (2007) used a virtual eight-arm radial maze to investigate correlations between gray matter volumes and the use of allocentric and egocentric-response navigational strategies. The maze was surrounded by an external environment featuring mountains, sky, a sunset, two trees, and a short wall separating the trees and the mountains. Each arm ended at the top of a staircase which led down to a lowered room. Only four arms held target objects (square boxes). These were not visible to the participants from their fixed starting position in the center of the maze. Hence, they had to navigate the arms in order to learn which held the objects and which were empty. The task given to them comprised two parts. The first part allowed access to only the four arms that held objects. The second part allowed access to all

arms, with the previously empty arms now holding the objects. On the second part, participants were told to retrieve the objects from the previously empty arms and to avoid the arms that held objects in part one. Their starting location remained the center of the maze, as in the first part. However, in the second part, the outside landscape was concealed and thus any landmarks that might have been used to facilitate an allocentric strategy were hidden. Participants who used an allocentric strategy to learn the location of the boxes in part one would thus be unable to complete the part two task without making many errors. In contrast, those who used an egocentric-response strategy would be able to complete the part two task with few errors as they would not be affected by the absence of distal cues.

The Hex Maze

Most existing VE tasks distinguish only between allocentric navigation and one type of egocentric navigation (egocentric-response or egocentric-cue) at a time. To enable simultaneous measurement and comparison of the allocentric strategy to *both* egocentric sub-strategies, Spriggs et al. (2018) developed a new VE spatial navigation task, the Hex Maze.

The Hex Maze presents a six-arm RAM that follows the paradigmatic procedures of an MWM task. Thus, there are a fixed number of pathways along which the participant may find the desired target (a circular platform), but there is only one such target. The target remains in the same position across trials, but the starting location of the individual changes so that they start *learning trials* at the end of one arm and *probe trials* at the end of another. The starting locations remain the same, however, for each trial type throughout the task (i.e., participants always start at the end of the same arm for all testing trials and the end of the same arm for all probe trials. Additionally, on learning trials the target location is marked by a colored sphere at the end of an arm while the empty arms are marked by light grey spheres. On subsequent probe trials, which require the participant to navigate back to the target location, the colored sphere cue swaps places with a light grey sphere and thus no longer marks the target location.

This combination of a RAM and MWM task allows the participant to use any of the three navigation strategies on each probe trial. If using an allocentric strategy, she would follow the landmarks from the outside environment surrounding the arena (i.e., a mountain range, a large body of water, and an island; these are visible through multiple windows of the arena room) to reach the arm in which the platform was originally located (thereby solving the task successfully). If using an egocentric-response strategy, the individual would follow the same direction of body turns as on the learning trials regardless of the variation in starting location (and thus would enter a different arm to the one that originally held the platform).

Finally, if using an egocentric-cue strategy, the individual would follow the colored cue sphere to its new location in an alternate arm to the original (and thus would, again, enter a different arm to the one that originally held the platform).

Spriggs et al. (2018) suggest that the value of the Hex Maze goes beyond its ability to distinguish between these three navigational strategies. They argue that it also allows exploration of numerous aspects of strategy use (i.e., preference, acquisition, stability, and competence), that it does not bias participant strategy use, and that, hence, it is a promising measure for valid investigation of human spatial navigation (see also Ferguson et al., 2019).

Variation in Navigational Strategies across the Menstrual Cycle

The menstrual cycle comprises three main phases, each characterized by particular patterns of hormone levels. The early follicular phase is characterized by low estrogen and low progesterone levels, the ovulatory phase by high estrogen and low progesterone levels, and the mid/late luteal phase by intermediate estrogen levels and high progesterone levels. The rodent equivalent of the menstrual cycle is the estrous cycle, which features similar hormonal trends across its various phases (e.g., the pre-estrus phase is characterized by high estrogen and increasing progesterone levels, similar to those found in the ovulatory and luteal phases of the menstrual cycle; the estrus phase is characterized by low estrogen and decreasing levels of progesterone, similar to those found in the follicular phase of the menstrual cycle). Of particular interest to this thesis is that hippocampal activity and functioning is influenced by the levels of these female sex hormones (Cooke & Woolley, 2005; Protopopescu et al., 2008).

Prior research exploring variation in human female spatial abilities across the menstrual cycle has tended to focus on mental rotation and other non-navigational cognitive constructs (for a review, see Hampson, 2018). Numerous studies in this literature suggest that higher concentrations of estrogen (e.g., as present during the ovulatory and luteal phases) are associated with lower scores on mental rotation tasks (see, e.g., Courvoisier et al., 2013; Zhu et al., 2015). More pertinently for the current thesis, a separate strand of literature has explored the role of sex hormones in rodent spatial navigation (for a review, see Brake & Lacasse, 2018). Generally, those studies indicate that female rats in the pre-estrus phase tend to use spatial memory systems whereas those in the estrus phase tend to use response memory systems.

Only two empirical studies have explored effects of cycle phase on choice and use of navigational strategy in humans. Hussain et al. (2016) asked 45 naturally cycling young women, aged between 23 and 36 years with a cycle of 25-34 days, to complete a VE RAM

navigation task using either an egocentric-response or an allocentric strategy, depending on their preference. Each participant was assigned to either an early follicular, ovulatory, or mid/late luteal group based on self-reported menstrual cycle information and confirmatory serum hormone level measurements. The navigation task comprised two parts. In the first part, four of the maze's eight arms were accessible and held objects at their end. Participants were told to retrieve all four objects and to remember the arms they visited so that they could avoid them on subsequent trials. In the second part, all arms were accessible and the walls surrounding the maze were lifted so as to conceal the previously visible outside landscape. Moreover, and unbeknownst to the participants, each of the eight arms now held an object. The participant was then asked to enter the four previously empty arms and avoid the four arms which had held objects in part one. If an allocentric strategy was used in part two, the participants would have required information from the landmarks in the outside environment to complete the task (i.e., to avoid the four arms that held objects in part one). Given that these landmarks were concealed, the use of an allocentric strategy would result in a higher number of errors (i.e., entering one of the four original arms from part one). On the other hand, for the egocentric-response strategy, the landmarks would be irrelevant and thus no increase in errors would be expected.

Results suggested that those in the mid/late luteal phase preferred using the allocentric strategy, whereas those in the other phases preferred using the egocentric-response strategy. The authors proposed that this data pattern might be explained by the fact that higher progesterone levels (as typically found during the mid/late luteal phase, but not during the other phases) promote hippocampal activity and depress caudate nucleus activity.

In a similar study, Scheuringer and Pletzer (2017) asked 51 men and 49 women to complete a VE map-based navigation task. This task was based in a computerized 2D 10 x 10 matrix containing 10 different symbols that were repeated once in each column and row, with the location of each symbol varying across trials. Before each trial, the participant received short written instructions describing a path within the matrix. These instructions were either focused around cardinal directions (thus encouraging an allocentric strategy) or personal directions (thus encouraging an egocentric-response strategy). The participant's task was then to use the given directions to move an arrow along the described path, using either an allocentric or an egocentric-response strategy. Most pertinent to the concerns of this review is the finding that women in the luteal phase, compared to those in the follicular phase, had significantly better navigational performance (as determined by reaction time and accuracy) when using an allocentric strategy.

Together, these studies suggest that women use different navigational strategies across the main phases of their cycle. Specifically, they indicate that the allocentric strategy is used more during the luteal phase of the cycle whereas the egocentric-response strategy is used more during the follicular phase. This trend is consistent with that observed in rodents. Together, the human and rodent literature suggest that as levels of estrogen and progesterone fluctuate, hippocampal and caudate nucleus activity change and subsequently, navigational strategy use changes too.

Effects of Stress on Choice and Use of Navigational Strategies

Although the studies reviewed above suggest there is a significant relationship between navigational strategy and hormonal variation across cycle phases, existing research has tended to ignore the potential effects of environmental factors on this relationship. One such environmental factor is the stress being experienced by naturally-cycling women as they complete navigational tasks.

In psychological literature, *stress* is defined as a body's physiological response to an (applied or implied) external threat (Kemeny, 2003; Nicolaides, Kyratzi, Lamprokostopoulou, Chrousos, & Charmandari, 2015; Sapolsky, 2015). When the organism appraises an event as threatening, two systems can be activated: the fast-acting sympathetic-adrenal-medullary (SAM) axis, or the slower-acting hypothalamic-pituitary-adrenal (HPA) axis. An activated SAM system increases activity of the sympathetic nervous system, which in turn releases adrenaline that stimulates the release of norepinephrine (NE) in the brain (Joels, Fernandez, & Roozendaal, 2011). The released NE goes on to activate the amygdala and subsequently alters activity in the hippocampus, caudate nucleus, and frontal lobes (Packard, Cahill, & McGaugh, 1994; Wolf, Atsak, de Quervain, Roozendaal, & Wingenfeld, 2016). Activation of the HPA system results in hypothalamic neurons secreting corticotropin-releasing hormone (CRH), which in turn triggers the discharge of adrenocorticotropin (ACTH) from the pituitary gland. ACTH travels through the vascular system to the adrenal glands, where it induces the secretion of two classes of stress hormones: glucocorticoids (of which cortisol is the primary type in humans) and catecholamines (adrenaline and noradrenaline). During physiological stress reactions, glucocorticoids cross the blood-brain barrier and bind to receptors in specific brain regions (Alderson & Novack, 2002; Kemeny, 2003; Nicolaides et al., 2015). Three important brain regions containing glucocorticoid receptors are the hippocampus, the amygdala, and the prefrontal cortex, all of which are involved in learning and memory processes (Arnsten, 2009; Gray, Kogan, Marrocco, & McEwen, 2017; Lupien, Maheu, Tu, Fiocco, & Schramek, 2007). Given the modulating

effect of an activated amygdala on the hippocampus and the affinity of cortisol to bind to hippocampal receptors, and to thus influence spatial memory processes, the physiological stress response may influence the choice and use of navigational strategies in humans.

People frequently make spatial decisions and use their spatial abilities to navigate their immediate environment while experiencing physical and/or psychological stress. Thus, the relationship between stress and spatial navigation has both practical and theoretical importance. Only a few published studies investigate the relationship between stress and spatial navigation, however. In one such study, Duncko, Cornwell, Cui, Merikangas, and Grillon (2007) exposed 14 of their 28 male participants to an acute physical stressor, the Cold Pressor Test (CPT; Hines & Brown, 1932), before administering a virtual MWM task. Their results indicated that those exposed to the stressor made fewer errors and thus navigated more efficiently compared to those unexposed. Contrarily, Richardson and Tomasulo (2011) found that male and female participants exposed to an acute psychosocial stressor, the Star Mirror Trace Task (SMTT; Reynolds & Stacey, 1955), performed more slowly but equally accurately compared to unexposed participants on a VR navigation task. They proposed that this relatively slower performance could be accounted for by participants switching from an allocentric strategy to an egocentric-response strategy when under stress.

Consistent with this proposal are the findings of other research studies suggesting that stress exposure preferentially impairs map-based spatial strategies. For instance, Thomas et al. (2010) exposed 15 of their 29 participants to an acute psychosocial stressor, the Trier Social Stress Test (TSST; Kirschbaum, Pirke, & Hellhammer, 1993), before they completed the CG Arena. Their results showed that TSST exposure impaired allocentric, but not egocentric, spatial navigational performance in young adult women. This finding is consistent with those reported within the rodent literature (see Cazakoff, Johnson, & Howland, 2010) for a review). In one example from that literature, Park, Zoladz, Conrad, Fleshner, and Diamond (2008) exposed rats to a 30-min predator stress condition immediately before testing them in a 6-arm radial arm water maze, a hippocampus-dependent spatial memory task. They found that allocentric navigational performance immediately after stress induction, as well as 24 hours later, was significantly impaired.

van Gerven et al. (2016) reported directly contrasting results, however. They administered the Paced Auditory Serial Addition Task (PASAT), a well-known neuropsychological test that has been shown to induce acute stress in laboratory settings, to a gender-balanced half of their 116 participants. Their results showed that, when stressed, female participants preferred using an allocentric spatial strategy to solve a VR MWM

navigation task. Consistent with this finding, Bohbot, Gupta, Banner, and Dahmani (2011) found a significant difference in basal levels of cortisol between participants who spontaneously used an egocentric-response strategy and those who used an allocentric spatial strategy. Specifically, they reported that lower basal levels of cortisol were associated with use of an egocentric-response strategy to navigate an eight-arm virtual RAM, whereas higher levels were associated with use of an allocentric strategy. Unfortunately, Bohbot and colleagues make no reference to their participants' menstrual cycles, and even while noting that female sex hormones may mediate the relationship between stress and navigational strategies, neither Thomas et al. nor van Gerven et al. specified their female participants' menstrual cycle phases during testing.

In a complete departure from the studies reviewed above, all of which found significant effects of stress on navigation, Klopp, Garcia, Schulman, Ward, and Tartar (2012) found that stress had no significant effect on allocentric navigational performance. They used the TSST to induce stress in 30 undergraduate students who subsequently completed a virtual MWM task. Just as in a standard MWM, the participants were required to learn where a submerged platform was located in a circular pool of water. For the probe trials, they were told their goal was to navigate to the submerged platform within 60 seconds and that their starting location would be different. They were also informed that if they could not find the platform in the given 60 seconds, the platform would rise above the surface of the water, and they would have to then navigate toward it as quickly as possible. Length of path and latency to the target on the probe trials were not significantly different in stressed participants compared to those exposed to non-stressful control condition (n = 20).

In summary, there are inconsistent findings in the research literature investigating the effects of stress on choice and use of navigational strategies. Further exploration of the topic is clearly necessary.

Interaction of Menstrual Cycle and Stress Effects on Cognition

So far, this literature review has explored the independent effects of menstrual cycle and stress on choice and use of spatial navigational strategies. No current study in the human navigation literature investigates the potential interactional effects of menstrual cycle and stress on spatial navigation, however.

It is important to explore this interaction because previous neuroendocrine research has shown that the stress response differs across the phases of the menstrual cycle (see Kajantie & Phillips, 2006; Kirschbaum, Kudielka, Gaab, Schommer, & Hellhammer, 1999; Lustyk, Olson, Gerrish, Holder, & Widman, 2010; Rohleder, Wolf, Piel, & Kirschbaum,

2003). Kudielka and Kirschbaum (2005) noted that estrogen appears to have a strong modulating effect on HPA-axis functioning (i.e., higher levels of estrogen reduce activity in the HPA axis), including responsiveness and sensitivity to the glucocorticoid negative feedback loop. The implication here then, is that higher circulating levels of estrogen can buffer the effects of stress on the organism.

Sharma and colleagues (2014) provided empirical data supporting this line of reasoning. They found that postmenopausal women who had been pretreated with estradiol[1] and who then received an overnight 14-hour intravenous infusion of ketoconazole and cortisol had significantly lower ACTH secretion levels than women had been pretreated with placebo. Here, then, pretreatment with estrogen reduced responsiveness to cortisol infusions (as measured by ACTH secretion levels), indicating that high levels of this sex steroid can inhibit HPA-axis functioning.

More recently, and further in support of this notion, Albert, Pruessner, and Newhouse (2015) compared brain activity and subjective mood in response to a psychosocial stressor (the Montreal Imaging Stress Task [MIST; Dedovic et al., 2005]) in two groups of premenopausal women: high estradiol (specifically during the ovulatory phase) and low estradiol (specifically during the early follicular phase). Their results showed that women in former group had a significantly lower stress response than those in the latter group. Specifically, during MIST exposure they had significantly less deactivation in limbic regions and post-MIST they reported significantly lower levels of subjective distress. Similarly, Maki et al. (2015) found that women at different phases of the cycle experienced differing cortisol responsivity to a psychosocial stressor (the TSST). Specifically, they found that women in the early follicular phase demonstrated a significant cortisol response to the TSST, whereas women in the luteal phase did not.

Together, these studies suggest that higher levels of estrogen (e.g., as are present in the ovulatory and luteal phases relative to the follicular phase) reduce the physiological responses to stressors. Given this varied vulnerability to stress induction procedures across cycle phases, one can posit that effects of stress on cognition will also differ across the phases of the menstrual cycle (i.e., as levels of female sex hormones, especially estrogen, fluctuate).

The notion that effects of stress on cognitive functions vary as levels of sex hormones fluctuate is supported by evidence from a small group of human studies (see, e.g., Andreano, Arjomandi, & Cahill, 2008; Espin et al., 2013). I review that literature below, but not before

[1]The most common form of estrogen.

noting that my discussion is not exhaustive because the cognitive functions that are the focus of those studies extend well beyond the single domain (spatial navigation) that is the focus of this thesis. In any case, this body of literature is small and, moreover, relatively few empirical studies investigating the main effects of stress on various aspects of cognition use a female sample; most such studies use male-only samples (see Het, Ramlow, & Wolf, 2005)).

In a recent review exploring sex differences in stress effects on emotional learning, Merz and Wolf (2017) indicate that exposure to acute stress at the post-encoding phase of emotional learning enhances memory consolidation for women in luteal phase but not for women in other phases of the cycle (see e.g., Andreano et al., 2008). The authors explain that the HPA axis and the hypothalamic-pituitary-gonadal (HPG) axis (the system responsible for the production of estrogen and progesterone) are interconnected and that this interconnectivity is perhaps why the arm of the physiological stress response centered on the HPA axis fluctuates across various levels of female sex hormones (and, hence, across the menstrual cycle). Consequently, this interconnectivity might explain why the main effect of stress on various cognitive functions (and specifically, for them, emotional memory) differs across cycle phases.

In a representative empirical study from this literature, Antov and Stockhorst (2018) compared declarative memory performance (as determined by the 'story recall' subtest from the Rivermead Behavioral Memory Test [RMBT; Wilson et al., 1992]) in participants exposed to pre-learning stress (the TSST). Their female participants were either in the early follicular or ovulatory phases during testing. Their analyses detected that stressed women in the early follicular phase had significantly poorer recall than stressed women in the ovulatory phase. These results, then, support the suggestion that the relationship between elevated cortisol (as seen, for instance, following exposure to an acute psychosocial stressor) and declarative memory performance (which is hippocampal-dependent) can be modulated by levels of female sex hormone levels.

Given the findings that the main effect of stress on emotional learning and, independently, on declarative memory (i.e., cognitive functions that also, to some degree, involve hippocampal activity) varies across the difference phases of the cycle, it is not farfetched to postulate that the same may be true for spatial learning, memory, and navigation. However, no published human study has investigated whether and how menstrual cycle phase may interact with stress exposure to affect choice and use of navigational strategies.

In a recent rodent study, however, ter Horst, Kentrop, de Kloet, and Oitzl (2013) explored the interacting effects of stress and estrous cycle phase on learning strategy in a circular hole board task. Female mice were required to use either a spatial or a stimulus-response strategy to navigate toward an exit hole on a grey round plate. The plate featured 12 equidistant holes, all placed 10cm from the rim of the board. The mice were required to learn the location of the exit hole during six training trials. On these training trials, a transparent plastic bottle was placed next to the open exit hole. Thirty minutes prior to the first training trial, some mice experienced a 10-min immobilization stressor before completing the test-trial. The test-trial was used to determine strategy use. In this trial, the original exit hole remained open, but the bottle was moved next to a newly opened hole. Results showed that stressed mice in the estrus phase tended to use a spatial strategy (i.e., they exited through the original exit hole) significantly more frequently than their non-stressed counterparts in the same phase, who tended to exit through the second hole marked by the plastic bottle. However, this stress-induced difference in strategy use was not observed in the other phases of their cycle, and no main effect of estrous phase was detected in unstressed mice. To explain these findings, ter Horst et al. (2013) looked to the reported stress-induced increase in estrogen during the estrus phase (see Shors, Pickett, Wood, & Paczynzki, 1999). Specifically, they argued that higher levels of estrogen during the estrus phase could have facilitated the use of a spatial strategy over a stimulus-response strategy in their female sample.

In summary, it is clear that these two endocrine variables (menstrual cycle phase and the physiological stress response) can and do interact, and that this interaction can influence cognitive performance on hippocampal-dependent tasks. Hence, it is important to explore whether this interaction effect also influences spatial navigation performance. Although ter Horst et al. (2013) investigated this question in rodents, no published study has done so with human participants.

Overall Summary

This literature review provided a brief overview of the current body of literature investigating the separate, independent effects of menstrual cycle phase and stress on choice and use of navigational strategies. There is a consistent finding that phase of the menstrual cycle has a statistically significant effect on choice and use of navigational strategy. Specifically, women in the follicular phase of their cycle (characterized by low estrogen and low progesterone levels) tend toward using an egocentric strategy, whereas those in the luteal phase (characterized by moderate estrogen and high progesterone levels) tend to favor an allocentric strategy. In contrast, prior research investigating the effect of stress on choice and

use of navigational strategy has delivered inconsistent results. Whereas some studies report significant stress-induced decreases in allocentric strategy use, others report significant stress-induced increases in allocentric strategy use. Still others report a non-significant main effect of stress on spatial navigation.

Although the existing body of research has explored effects of hormonal variation across the menstrual cycle phase and exposure to stress independently, these effects do not occur in isolation from one another, as can be seen by the interconnectivity between the HPA and HPG axes. Hence, the extant spatial navigation literature contains results that, although statistically and practically significant, are missing some key element of ecological validity because they do not consider the interacting effects of hormones and stress. By measuring the effects of this interaction on choice and use of navigational strategy, this thesis attempts to make an important and novel contribution to the current body of literature on this topic.

CHAPTER THREE:
SPECIFIC AIMS AND HYPOTHESES

The primary aim of this thesis was to investigate possible interaction effects of menstrual cycle phase and stress exposure on choice and use of navigational strategies. Secondary aims were to investigate the main effects of (a) menstrual cycle phase (and to thereby systematically replicate a relatively consistent set of findings indicating that navigational strategy varies as levels of sex hormones fluctuate), and (b) stress exposure (and to thereby attempt to resolve some of the inconsistency in the literature investigating that effect). Overall then, this thesis contributes to the growing body of literature around the separate, independent effects of menstrual cycle phase and acute stress exposure on choice and use of navigational strategies. Furthermore, it is novel in its exploration of the potential interactional effects of those factors on navigation.

Another novel contribution of the research presented in this thesis is that only two previously published studies have described the simultaneous investigation of allocentric, egocentric-cue, and egocentric-response strategies (Ferguson et al., 2019; Spriggs et al., 2018). Neither of these studies shared a similar research question as investigated here, however. Hence, this study is the first to examine the interacting and main effects of menstrual cycle phase and acute stress exposure on the use of those three distinct navigational strategies.

The study tested the following specific hypotheses:

1. Menstrual cycle phase will have a significant main effect on choice and use of navigational strategy. Participants in the early follicular and ovulatory phases will tend to use one of the egocentric strategies, whereas those in the mid/late luteal phase will tend to use an allocentric strategy.
2. Laboratory-induced acute psychosocial stress will have a significant main effect on choice and use of navigational strategy. Participants exposed to a stressor, but not those unexposed, will tend to use one of the egocentric strategies, rather than an allocentric strategy.
3. Laboratory-induced acute psychosocial stress and menstrual cycle phase will have a significant interaction effect on choice and use of navigational strategy. This interaction effect will be particularly evident among participants in the mid/late luteal phase. Women in that phase who are exposed to a stressor will

tend to use one of the egocentric strategies, whereas those who are in that phase but not exposed to the stressor will tend to use an allocentric strategy.

Before testing these hypotheses in the major study that forms the centerpiece of this thesis, I conducted a pilot study to determine the feasibility of the research project and to examine the appropriateness of the proposed measures and methods for investigating the research questions at hand.

CHAPTER FOUR:

PILOT STUDY

Broadly speaking, I conducted this pilot study to gain some insight into the feasibility of a research project centered on questions involving interacting effects of menstrual cycle phase and stress on navigational strategy. More specifically, I wished to test whether a set of proposed methods and measures would provide an appropriate foundation for an investigation of the research questions I had. This is, conventionally, one of the main purposes of a pilot study (van Teijlingen & Hundley, 2001). Additionally, given that there are no published studies examining the interacting effects of exposure to acute psychosocial stress and menstrual cycle phase on navigational strategy use, I used the pilot study as a means to assess whether potentially significant findings might arise from a full-sized study.

Because the pilot study was constrained by time and resource limitations, I could not explore variations across the three main phase groups (i.e., early follicular, ovulatory, mid/late luteal), and so I assigned women who were in the first two of those phases into a single Non-Luteal phase group. Additionally, I only compared the two main navigation strategy types (allocentric and egocentric [i.e., egocentric-response and egocentric-cue combined, as in most previous research studies]) to one another.

Hence, the pilot study tested the following hypotheses:

1. Menstrual cycle phase will have a significant main effect on navigational strategy use: Participants in the Non-Luteal phase group will tend to use an egocentric strategy, whereas those in the Luteal phase group will tend to use an allocentric strategy.
2. Laboratory-induced acute psychosocial stress will have a significant main effect on navigational strategy use: Participants exposed to the stressor, but not those unexposed, will tend to use an egocentric rather than an allocentric strategy.
3. Induced stress and menstrual cycle phase will have a significant interactional effect on navigational strategy use, particularly among participants in the Luteal phase group. For instance, participants in that group who are exposed to an acute psychosocial stressor will tend to use an egocentric strategy, whereas those who are in that group but not exposed to the stressor will tend to use an allocentric strategy.

Methods

Design and Setting

This quasi-experimental study used a 2x2 factorial design. The independent variables were menstrual cycle phase (two levels: Non-Luteal, Luteal) and experimental condition (two levels: Stress, Non-Stress). The dependent variable was the preferred navigational strategy (allocentric or egocentric), as determined by performance on a VR navigation task, the CG Arena.

The study was conducted in a research laboratory housed within the Department of Psychology at the University of Cape Town (UCT). Ethical approval was granted by the Department's Research Ethics Committee (reference number: PSY2018-026), and all study procedures followed guidelines for human research laid down by the Declaration of Helsinki (World Medical Association, 2013).

Participants

I used convenience sampling to recruit 28 regularly-cycling women from the population of female UCT undergraduate students. They were aged 18-28 years (M = 20.84, SD = 2.34), with a menstrual cycle of 25-34 days in duration.

All completed a screening questionnaire prior to enrolment to ensure eligibility. Individuals who reported a history of (a) psychological, endocrinological, or neurological illness, (b) substance abuse, or (c) pregnancy within 2 years of testing were excluded from participation. I also excluded individuals who were currently breastfeeding, who had taken any form of hormonal contraception within 3 months of testing, or who presented with abnormally high blood pressure levels (i.e., 140/90 at baseline). After applying these criteria, I excluded 9 potential participants from the study. Moreover, data from one enrolled participant were excluded due to software error. Hence, the final sample consisted of 18 participants.

Participants who reported being within days 1-7 or days 13-17 of the menstrual cycle on the day of testing were assigned to the Non-Luteal phase group. Those who reported being between day 20 and the end of the cycle were assigned to the Luteal phase group. I then used an online application (www.randomizer.org) to randomly assign those within each phase group to one of the experimental conditions. Hence, each participant was a member of one of four study groups: Non-Luteal+Stress (n = 6), Luteal+Stress (n = 4), Non-Luteal+Non-Stress (n = 5), or Luteal+Non-Stress (n = 3).

Materials and Procedure

Pre-manipulation. The following questionnaires were administered before the participant was exposed to the experimental manipulation.

Screening questionnaire. A self-report questionnaire (Appendix A), similar to that used by Hussain et al. (2016), helped determine study eligibility and gathered information regarding menstrual cycle phase.

Santa Barbara Sense-Of-Direction Scale (SBSOD). This instrument (Appendix B) gathered data on participants' self-perceived spatial ability. SBSOD scores correlate well with various objective measures of performance on spatial cognition tasks, and the scale has high internal (α = .88) and test-retest (r = .91) reliability (Hegarty, Richardson, Montello, Lovelace, & Subbiah, 2002).

State-Trait Anxiety Inventory (STAI). Participants completed the State-Trait Anxiety Inventory (Spielberger, Gorsuch, Lushene, Vagg, & Jacobs, 1983; see Appendix C). This instrument comprises two 20-item Likert-type scales, one measuring state anxiety (i.e., current anxiety level) and the other measuring trait anxiety (i.e., general anxiety level). The STAI has excellent psychometric properties with a reliable factor structure, excellent construct and concurrent validity, and excellent internal consistency (Spielberger & Vagg, 1984).

Experimental manipulation. Each participant was exposed to either an acute psychosocial stressor or to an equivalent non-stressful control condition. Following van Gerven et al. (2016), I used a modified version of the Paced Auditory Serial Addition Task (PASAT), a well-known neuropsychological test, to induce stress (see also Lustyk et al., 2010; Mathias, Stanford, & Houston, 2004). This task requires participants to rapidly add a series of single-digit numbers, presented sequentially and verbally to them on a standard desktop computer monitor via Inquisit (Version 5) software. Participants are instructed to add each number to the one that precedes it (e.g., if the presented sequence is 3-5-9-2, the correct responses are 8-14-11).

In this study, as in van Gerven et al. (2016), the task included a practice block, consisting of 14 digits, and four test blocks, each consisting of 60 digits. During the practice block, digits were presented with an inter-stimulus interval (ISI) of 2.7 s, whereas in the test blocks they were presented with progressively shorter ISIs (2.1 s in the first, 1.7 s in the second, 1.3 s in the third, and 0.9 s in the fourth). If the participant added incorrectly, or failed to select a response within the ISI, the software gave error feedback (i.e., a sharp beeping sound) before presenting the next number.

I administered the '*un*-Paced Serial Addition Task' (*u*-PASAT; based on that used by van Gerven et al., 2016) as a control condition. This task is identical to the PASAT in that single-digit numbers are presented sequentially and verbally to participants, and they are required to add each digit to the one that precedes it and to select the correct response. The difference here, however, is that all digits, across all blocks, are presented with ISIs of 3 s. If the participant added incorrectly, there was no error feedback; instead, they were simply instructed to pick up again from the next presented digit.

Both the PASAT and *u*-PASAT took approximately 12-15 minutes to complete.

Subjective and objective measures of stress. Participant heart rate (HR) was measured using a Polar Electro h10 Heart Rate Sensor (Kempele, Finland). Blood pressure (BP) measurements were taken with a standard blood pressure cuff. Participants also completed the STAI-State several times throughout the testing session as a repeated measure of self-reported state anxiety.

Navigation task. The CG Arena (Jacobs et al., 1997; Jacobs et al., 1998; Thomas et al., 2001) is a non-immersive desktop VE navigation task modeled on the MWM. The participant is presented with a first-person view of a large square room with a low circular wall spanning its inner boundaries. In the Arena, participants shift their viewpoint using the computer keyboard, and can thereby use landmark or distal cues and the spatial relations between them to locate and relocate specific locations on the Arena floor. In the current study, images were posted on the room's four walls (see Figure 2). These images served as distal cues (i.e., cues not connected directly to the target). There were also two cue objects, a red cube and a green cube, placed inside the room, at a height above the circular wall, in the middle of the northwest (NW) and southeast (SE) quadrants respectively. These objects served as landmark cues (i.e., cues used to deduce one's position based on the distances and positions of these within-Arena objects).

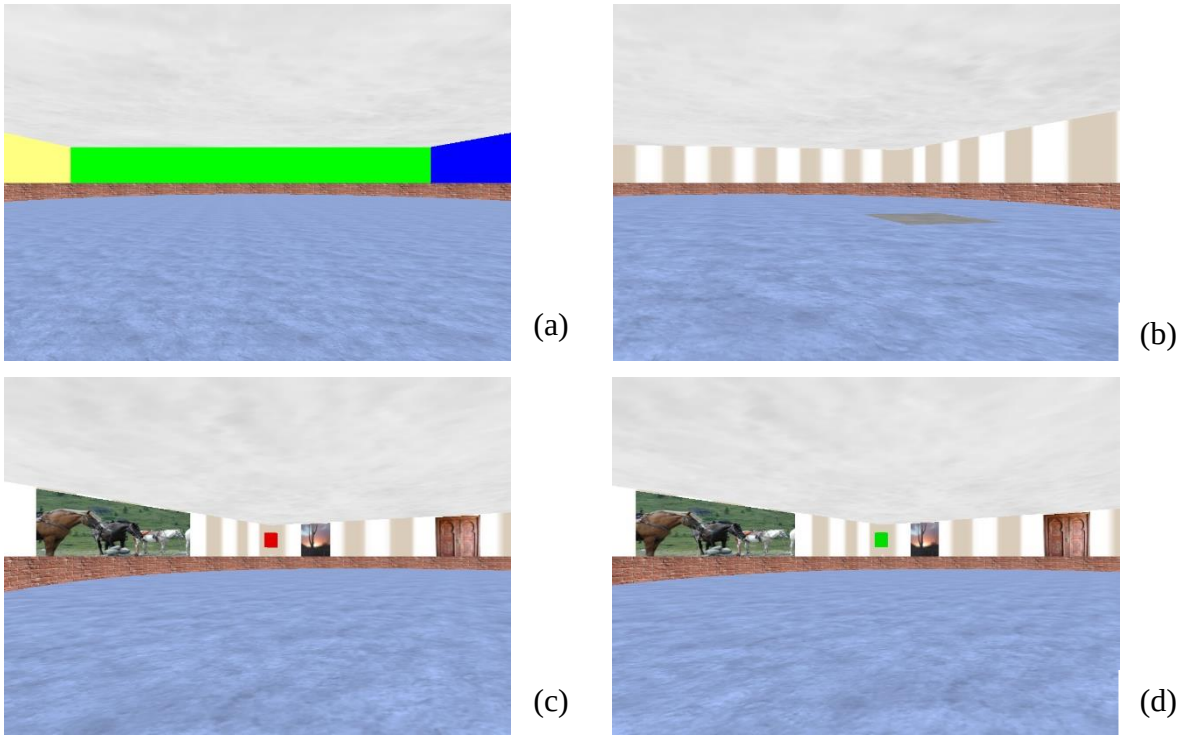


Figure 2. View from within the CG Arena on different trial types. Panel (a) depicts the waiting room. Panel (b) depicts a visible target trial. Panel (c) depicts a Standard trial with a view of northwest (NW) quadrant; the cue object, the red cube, is visible in the corner. Panel (d) depicts an Inter-Trial Strategy Probe trial with a view of the NW quadrant; the cue object has been swapped with the green cube that had been in the opposite quadrant on the previous Standard trial.

I used a CG Arena protocol that systematically replicated the navigation task described by van Gerven et al. (2016). First, the participant entered the *waiting room*, an environment that featured plain-colored walls and that was devoid of all distal and other cues. Exposure to the waiting room allows participants to become comfortable with navigating within the Arena before commencing task trials. Once the participant signaled she was comfortable, she moved on to complete four *visible target trials*. On those trials, she had to navigate, as quickly and directly as possible, to a square plainly visible on the Arena floor. When the participant reached this target, a bell rang, and I pressed the spacebar to end the trial. The visible target trials took place in a room featuring no distal or landmark cues.

After completing the set of visible target trials, the participant was administered 10 Standard Trials (STs) with 10 Inter-Trial Strategy Probe trials (ITSPs) interspersed between them, such that they alternated in pairs (i.e., ST – ITSP – ST – ITSP, and so on).

On each ST, the participant's task was to locate a square that was hidden until she moved over it. This target remained in the same location, in the NW quadrant of the arena, and in line with the red cube. When the participant moved over the target, it became visible,

the bell rang, and the participant became trapped so that she could no longer move around the Arena. I then pressed the spacebar to end the trial. The participant's starting position on each ST varied, although each starting point was close to the circular wall, at one of the Arena's cardinal points. On the first three STs, the participant was encouraged to look around the Arena after finding the target and to try to remember her location.

ITSPs matched the formal structure of the STs. There were some key differences between the types of trials, however: On ITSPs, the target was always hidden, and the cue objects swapped position from where they were on the STs (i.e., the green cue was in the NW quadrant, whereas the red cue was in the SE quadrant). Participants were informed that they should navigate to the location in which they believed the target had been located on the previous ST. They were informed that the target would not appear and that no feedback would be given. However, they were not informed that the cue objects had changed position. On the ITSPs, participant starting positions alternated between the NE and SW cardinal points, near the circular wall. Hence, the two goal locations were equidistant from the starting points. The trial ended when the participant indicated she had reached the target location. If she identified the target location as being in the NW quadrant (i.e., where the target had been located on the ST trial), she was classified as using an allocentric strategy. If she identified the target location as being in the SE quadrant (where the red cue object was now placed), she was classified as using an egocentric strategy. Hence, the ITSPs assessed participant navigational strategy selection on a trial-by-trial basis.

Post-Arena questionnaires. Participants completed a structured questionnaire (see Appendix D) to gather self-report data on how they solved the CG Arena task. Answers to these questions were used to confirm the strategy categorization made based on task performance. A second questionnaire (Appendix E) gathered some background information about prior video game experience. I used this information to confirm there were no significant between-group differences with regard to this variable.

Procedure

Each participant was tested individually in a single 90-min session. Sessions ran between 8am and 4pm depending on participant availability.

The test session began with the completion of informed consent procedures (see Appendix F) and the screening questionnaire. Individuals who did not meet the study's eligibility criteria were dismissed immediately. Those who were eligible then completed the STAI-Trait, STAI-State, and SBSODS. Immediately thereafter I took baseline measures of

HR and BP. Next, each participant was randomly assigned to an experimental condition, completing either the PASAT or *u*-PASAT.

Immediately post-manipulation, I took a second set of HR and BP measures and the participant completed the STAI-State for a second time. After a 10-min rest period during which the participant read a non-evocative magazine, I took a third set of HR and BP measures. The participant then completed the CG Arena task. Immediately thereafter, I took a fourth set of HR and BP measures, and the participant completed the STAI-State for a third time. Then, I administered the two post-Arena questionnaires. Upon completion of those questionnaires, I took a final (fifth) measure of HR and BP, and the participant completed the STAI-State for the final (fourth) time. At the end of these study procedures, I debriefed the participant (see Appendix G), thanked her for her time and involvement, and dismissed her.

Data Management and Statistical Analyses

I conducted all statistical analyses using SPSS (version 25.0) with the threshold for statistical significance set at $\alpha = .05$. Effect size estimates were interpreted using conventional guidelines (small: Cohen's $d = 0.2$, $\eta^2 = .01$, Cramer's $V = .07$-$.21$; medium: Cohen's $d = 0.5$, $\eta^2 = .06$, Cramer's $V = .21$-$.35$; large: Cohen's $d = 0.8$, $\eta^2 = .14$, Cramer's $V > .35$; J. Cohen, 1988).

Prior to conducting inferential analyses, I generated and examined a complete set of descriptive statistics to ensure that all assumptions underlying those analyses were met. In cases where they were not met, I adjusted the analytic plan. Details of these adjustments are given at the appropriate places in the Results section.

Sample characteristics. A series of one-way ANOVAs (for continuous variables) and chi-squared tests (for categorical variables) assessed the magnitude of between-group differences with regard to basic sample characteristics (e.g., age, STAI-Trait scores, SBSOD scores, video game experience, average length of menses).

Manipulation check. Four linear mixed models, each including the fixed effects of Condition (Stress, Non-Stress), Time (Baseline, Post-Stressor, Post-Rest, Post-Arena, Pre-Debrief), and the interaction between the two, assessed magnitude of changes in physiological and self-reported stress over study time for participants in the two conditions (i.e., they investigated the effectiveness of PASAT in inducing stress, and sought to confirm that the *u*-PASAT did not induce stress). A series of independent- and paired-sample *t*-tests followed up the significant results of the mixed models, thus seeking to confirm the precise location of the effects. Finally, because it was ethically important to ensure that Stress-condition participants left the laboratory in the same psychological state as when they

entered, a series of paired-sample *t*-tests investigated the magnitude of differences between the first (Baseline) and last (Pre-Debrief) measures of physiological and self-reported stress within that condition.

Navigation strategies. Following van Gerven et al. (2016), I calculated a single outcome variable that captured the degree to which participants used an allocentric (versus an egocentric) strategy when completing the spatial navigation task. To calculate this variable, *Average Allocentric %*, I divided the number of ITSP trials on which the participant had used an allocentric strategy by the total number of ITSP trials the participant had completed (i.e., Average Allocentric % = [Allocentric trials/Total ITSP trials] x 100). Hence, participants with higher values for this variable tended toward more frequent use of an allocentric strategy when completing the CG Arena task, whereas those with lower values tended toward more frequent use of an egocentric strategy. Initial analyses revealed that the data, even after standard attempts at transformation, were not appropriate to be examined using factorial ANOVA. Thus, to investigate whether the independent variables (stress and menstrual cycle phase) had significant main and/or interaction effects on that outcome, I ran a linear mixed-model analysis that included fixed effects of Cycle Phase, Condition, and the interaction between the two.

Results and Discussion

Sample Characteristics

Analyses detected no significant between-condition (Stress versus Non-Stress) differences on most measured sociodemographic, lifestyle, psychological, and physiological variables (see Table 1). These results suggest that subsequent analyses were not confounded by between-condition differences on any of the measured sociodemographic, lifestyle, psychological, and physiological variables.

Table 1.
Sample Characteristics: Descriptive statistics and between-condition comparisons (N = 18)

	Experimental Condition						
	Stress		Non-Stress				
Measure	Non-Luteal (*n* = 6)	Luteal (*n* = 4)	Non-Luteal (*n* = 5)	Luteal (*n* = 3)	*F* / x^2	*p*	ESE
Age (years)	20.50 (1.76)	20.25 (1.50)	20.60 (0.89)	22 (5.20)	0.36	.782	.07
Range	19-23	18-21	20-22	19-28			
Education (years)	13.67 (1.21)	12.75 (0.50)	14 (0.71)	12.67 (0.58)	2.40	.111	.34
Range	12-15	12-13	13-15	12-13			
STAI-Trait	35 (7.01)	36.75 (6.90)	34 (6.52)	45 (21.28)	0.83	.499	.15
Range	24-44	31-46	23-40	22-64			
SBSOD	4.40 (.98)	4.05 (1.45)	3.80 (1.62)	4.08 (.92)	0.20	.896	.04
Range	3.13-5.60	2.50-5.93	1.93-5.67	3.10-4.93			
Sleep					4.33	.632	.35
4-6	2	1	2	2			
7-8	4	3	2	1			
9-10	0	0	1	0			
Exercise					15.72	.401	.54
> 1	1	2	0	3			
1-2	3	1	1	0			
3-4	1	1	1	0			
5-6	0	0	1	0			
7-8	1	0	1	0			
> 8	0	0	1	0			
Alcohol					5.75	.119	.57
0-2	6	4	3	3			
3-5	0	0	2	0			
Cigarettes					4.19	.651	.34
0	5	3	4	3			
10-20	0	1	1	0			
20-40	1	0	0	0			
Video Games							
0	3	4	5	3	7.2	.616	.37
3-5	1	0	0	0			
5-10	1	0	0	0			
> 10	1	0	0	0			
Age at first period	13.17 (.98)	13.25 (.50)	13.10 (2.41)	11 (2.65)	1.27	.318	.20
Range	12-14	13-14	10-16	8-13			
Menses Duration					11.03	.088	.55
3-5	3	4	2	0			
6-8	3	0	3	2			
> 8	0	0	0	1			

Notes. For continuous variables, means, standard deviations, and ranges are presented; for categorical variables, frequencies with percentages in parentheses. *Sleep* represents average duration per night, in hours, over the last month; *Exercise*, average amount per week, in hours; *Alcohol*, average number of drinks per week; *Cigarettes*, average number smoked per week; *Video Games*, playing experience, in years; *Menses Duration*, average period length, in days. STAI = State-Trait Anxiety Inventory; SBSOD = Santa Barbara Sense of Direction scale; ESE = effect size estimate (η^2 for ANOVAs, and Cramer's *V* for chi-squared tests).

Manipulation Check

Table 2 presents descriptive statistics and results of the mixed-model analyses for the HR, BP, and STAI-State data. Figure 3 is a graphic depiction of the time course of these data. Analyses detected significant main effects of (a) Condition on HR, (b) Condition on STAI-State scores, and (c) Time on STAI-State scores. All other main effects, and all two-way interactions, were non-significant.

Table 2.
Physiological and Self-report Data: Descriptive Statistics and Linear Mixed-Model Analyses (N = 18)

	Experimental Condition					
Measure / Measurement	Stress (n = 10)	Non-Stress (n = 8)	Effect	F	df	p
HR						
Baseline	84.10 (5.59)	79.00 (14.83)	Condition	6.17	1,31	.019*
Post-Stressor	86.90 (12.22)	72.57 (12.18)[a]	Time	0.22		.646
Post-Rest	79.40 (6.80)	72.57 (13.16)[a]	Interaction	1.39		.247
Post-Arena	76.60 (6.29)	71.86 (12.95)[a]				
Pre-Debrief	75.10 (5.61)	73.14 (12.75)[a]				
BP: Systolic						
Baseline	118.60 (12.53)	119.25 (7.40)	Condition	0.12	1,31	.736
Post-Stressor	114.70 (7.69)	116.29 (9.38)[a]	Time	1.09		.304
Post-Rest	114.80 (7.12)	109.86 (6.15)[a]	Interaction	0.02		.888
Post-Arena	114.50 (9.17)	114.43 (8.14)[a]				
Pre-Debrief	112 (7.02)	113.14 (8.11)[a]				
BP: Diastolic						
Baseline	73.60 (6.31)	72 (7.64)	Condition	0.45	1,31	.506
Post-Stressor	71.20 (7.35)	69.71 (4.86)[a]	Time	1.04		.315
Post-Rest	71 (8.30)	70.43 (2.57)[a]	Interaction	0.001		.980
Post-Arena	69.50 (4.40)	72.43 (6.24)[a]				
Pre-Debrief	72 (6.45)	70.14 (7.03)[a]				
STAI-State						
Baseline	32.00 (12.21)	27.88 (9.48)	Condition	4.44	1,32	.043*
Post-Stressor	48.90 (12.83)	36.88 (10.20)	Time	11.41		.002**
Post-Arena	30.60 (7.88)	31.88 (12.82)	Interaction	1.06		.311
Pre-Debrief	27.80 (7.15)	29.25 (12.14)				

Note. Means are presented with standard deviations in parentheses. HR = heart rate (measured in beats per minute [bpm]); BP = blood pressure (measured in mmHG); STAI = State-Trait Anxiety Inventory. [a]n = 7; one dataset lost due to hardware malfunction.
*p < .05. **p < .01. ***p < .001.

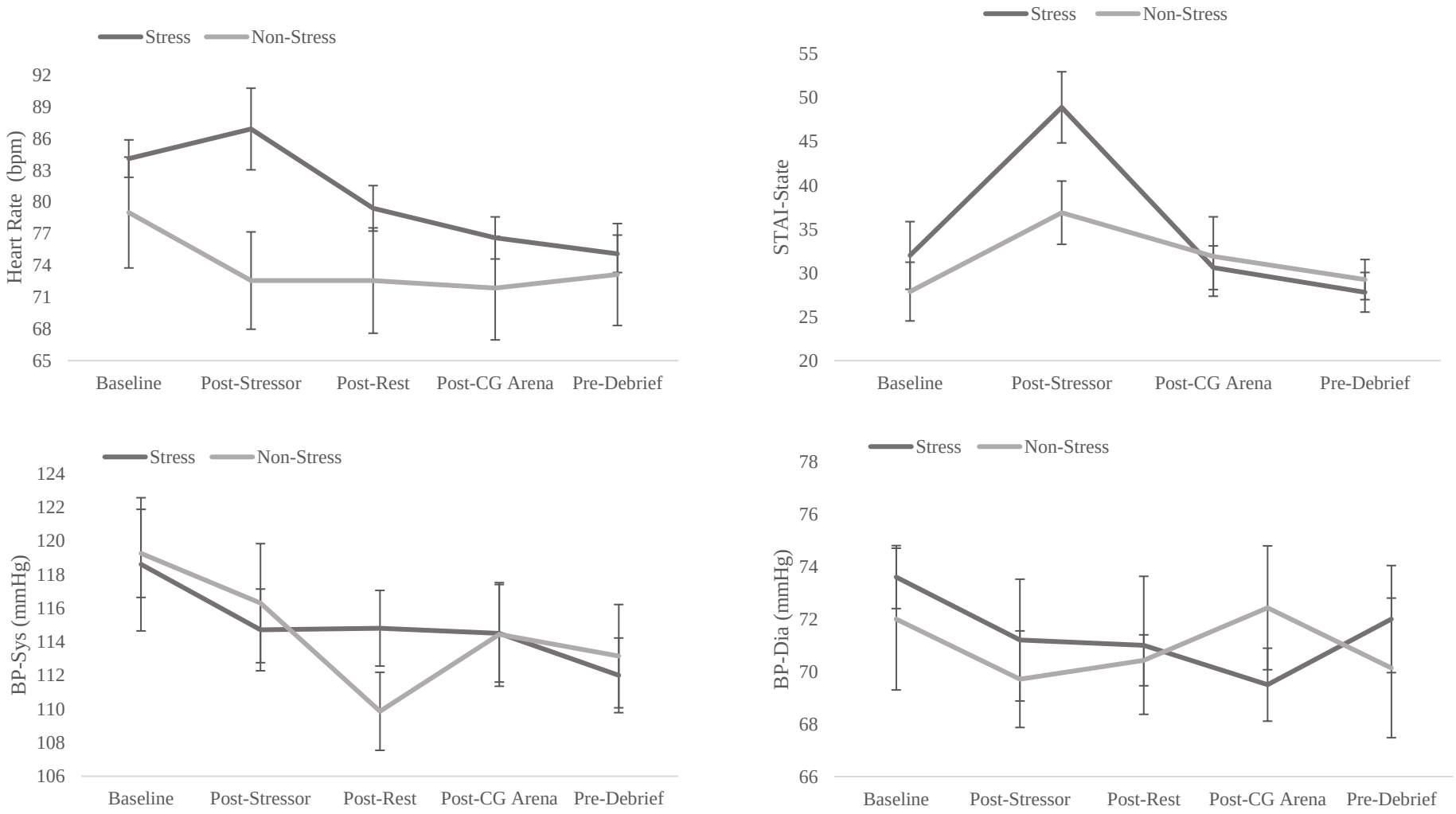


Figure 3. Physiological and self-report measures of stress across the experiment; Stress condition *n* =10, Non-Stress condition *n* = 8. Error bars represent the standard error of the mean for each measurement point. STAI = State-Trait Anxiety Inventory; BP-Sys = systolic blood pressure ; BP-Dia = diastolic blood pressure.

Further regarding the significant main effects of Condition, independent-sample *t*-tests compared, for participants in the Stress and Non-Stress conditions, changes in HR and in STAI-State scores from baseline to the immediate post-stressor measure. These analyses both detected significant between-condition differences associated with large effect sizes. The order of means suggested that the magnitude of changes in HR and STAI-State scores for participants in the Stress condition exceeded those for participants in the Non-Stress condition (see Table 3).

Table 3.
Changes in Heart Rate and STAI-State Scores from Baseline to Post-stressor (N = 18)

	Experimental Condition				
	Stress	Non-Stress			
Variable	(*n* = 10)	(*n* = 8)	*t*	*p*	ESE
ΔHR	2.80 (11.08)	-5.57 (5.13)[a]	-2.09	.028*	0.91
ΔSTAI-State	16.90 (9.31)	9.00 (9.13)	-1.80	.045*	0.86

Note. Means are presented with standard deviations in parentheses. HR = heart rate (measured in beats per minute [bpm]); STAI = State-Trait Anxiety Inventory; ESE = effect size estimate (Cohen's *d*). [a]*n* = 7; one dataset lost due to hardware malfunction.
*$p < .05$. All *p*-values reported are one-tailed.

Paired-samples *t*-tests examined the same data, but this time compared changes in HR and STAI-State scores from baseline to the immediate post-stressor measure within each of the Stress and Non-Stress conditions separately (see Table 2 for the relevant descriptive statistics). For participants assigned to the Stress condition, these analyses detected (a) a non-significant increase in HR, $t(9) = -0.80$, $p = .223$, Cohen's $d = 0.29$, and (b) a significant increase in STAI-State scores, $t(9) = 5.74$, $p < .001$, $d = 1.35$. For participants assigned to the Non-Stress condition, these analyses detected (a) a significant decrease in HR, $t(6) = 2.96$, $p = .013$, $d = 0.37$, and (b) a significant increase in STAI-State scores, $t(7) = 2.79$, $p = .014$, $d = 0.91$.

Finally, for data from the Stress condition only, four separate paired-sample *t*-tests compared baseline HR, BP, and STAI-State values to those at the pre-debrief measurement point. Analyses detected only two significant between-measure differences: At pre-debrief, participants' HR and systolic BP readings were significantly lower than at baseline (see Table 4).

Table 4.
Changes in Stress Measures from Baseline to Pre-Debrief Measurement: Stress-group participants (N = 10)

	Measurement Point				
Variable	Baseline	Pre-Debrief	*t*	*p*	ESE
HR	84.10 (5.59)	75.10 (5.61)	5.73	< .001***	1.61
BP					
Systolic	118.60 (12.53)	112 (7.02)	2.04	.036*	0.65
Diastolic	73.60 (6.31)	72 (6.45)	0.88	.427	0.16
STAI-State	32.00 (12.21)	27.80 (7.15)	0.92	.384	0.42

Note. Means are presented, with standard deviations in parentheses. HR = heart rate (measured in beats per minute [bpm]); BP = blood pressure (measured in mmHG); STAI = State-Trait Anxiety Inventory; ESE = effect size estimate (Cohen's *d*).
*$p < .05$. **$p < .01$. ***$p < .005$. All *p*-values reported are one-tailed.

Taken together, these results suggest that exposure to the PASAT was associated with mild increases in HR levels and substantial increases in STAI-State scores. In contrast, exposure to the *u*-PASAT was associated with substantial decreases in HR levels and relatively minor increases in STAI-State scores. Hence, at both physiological and subjective levels, participants in the two experimental conditions were distinguishable from one another at the immediate post-manipulation measurement point. However, within the Stress condition, both physiological and self-report measures of stress were not significantly increased over baseline when the participants left the laboratory, indicating no chronic effects of the experimental manipulation.

Effects of Cycle Phase and Stress on Navigation Strategy

Table 5 presents the cell means and standard deviations for the primary outcome variable, Average Allocentric %. Analyses of these data did not detect a significant main effect of Cycle Phase, $F(1,14) = 1.82$, $p = .198$. As Figure 4 shows, participants in both the Luteal and Non-Luteal groups showed a preference for using an allocentric strategy, although that preference was slightly more marked in the former than in the latter. Hence, Hypothesis 1 was not confirmed. However, because the observed direction of means was in the predicted direction the results of this pilot study are promising, and one might speculate that a full investigation, powered by a large enough sample, would confirm the hypothesis.

Table 5.
Navigation Strategy (Average Allocentric %) Across Study Groups (N = 18)

	Experimental Condition	
	Stress	Non-Stress
Cycle Phase Group	(*n* = 10)	(*n* = 8)
Non-Luteal (*n* = 11)	46.25 (31.45)	81.00 (13.06)
Luteal (*n* = 7)	58.25 (26.70)	100.00 (0.00)

Note. Mean percentages are presented, with standard deviations in parentheses.

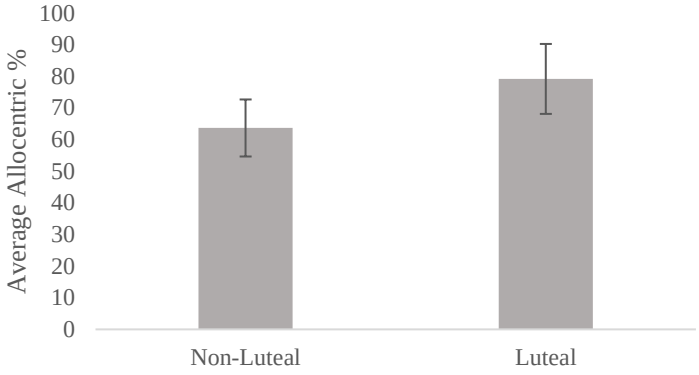


Figure 4. Effect of menstrual cycle phase on navigational strategy. Participants in the Luteal phase tended to use an allocentric strategy slightly more frequently than did those in the Non-Luteal phase, although both preferred allocentric over egocentric strategies (Average Allocentric % *M* = 79.13±9.00 versus *M* = 63.63±7.13). Error bars represent the standard error of each respective group mean.

The analysis did, however, detect a significant main effect of Condition, $F(1,14) = 11.10$, $p = .005$. As Figure 5 shows, participants in the Stress condition tended toward using an egocentric strategy much more frequently than did those in the Non-Stress condition. This result confirms Hypothesis 2.

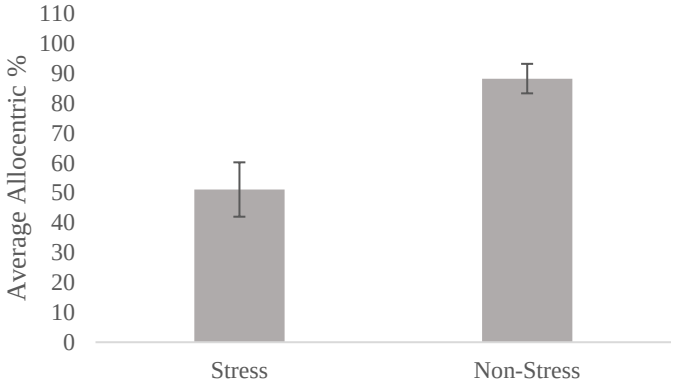


Figure 5. Effect of experimental condition on navigational strategy. Participants in the Stress condition tended to use an egocentric strategy more frequently than did those in the Non-Stress condition, who tended strongly toward using an allocentric strategy (Average Allocentric % *M* = 52.25±7.60 versus *M* = 90.50±8.61). Error bars represent the standard error of each respective group mean.

These findings regarding the main effect of stress on navigational strategy use differ from those of van Gerven et al. (2016) which is particularly interesting given that this pilot study's stress induction and navigation tasks represented systematic replications of those used in their study. One possible reason for the difference in findings is that, although the navigation task itself was a systematic replication of the task used in their study, the virtual environments differed. Additionally, I excluded women taking hormonal birth control, whereas van Gerven and colleagues did not. Because ingestion of such medication alters natural estrogen and progesterone levels, thus affecting the neural action of these hormones, it is possible that strategy use of their participants was confounded. Moreover, van Gerven and colleagues included participants currently taking mood-stabilizing medication, whereas my pilot study excluded such individuals. The PASAT may not have been effective in inducing stress for participants taking such medication given their ability to buffer the psychological and neurological effects of stress, and thus they may have preferred using allocentric spatial strategies. Unfortunately, van Gerven et al. (2016) did not analyze the effects of these factors on their outcomes, nor did they control for them within their analysis. Without information from such analyses, it is impossible to say whether, and to what degree, their results may have been affected by those factors.

Finally, the analysis did not detect a significant Condition x Cycle Phase effect, $F(1, 14) = 0.093$, $p = .765$. Hence, Hypothesis 3 was not confirmed. However, the data shown in Table 5 and Figure 6 suggest that the effect trended toward the predicted direction. Specifically, participants in the Luteal+Non-Stress group had an average Allocentric % that was 41.75% higher than that of those in the Stress condition (i.e., those in the Luteal+Stress group had a stronger preference for an egocentric strategy than those in the Luteal+Non-Stress group, as predicted). Of note here too is that only participants in the Non-Luteal+Stress group showed a preference for an egocentric over an allocentric strategy (i.e., Average Allocentric % < 50). Overall, then, the results are promising and suggest that a larger-scale investigation of this novel question may find a significant interaction.

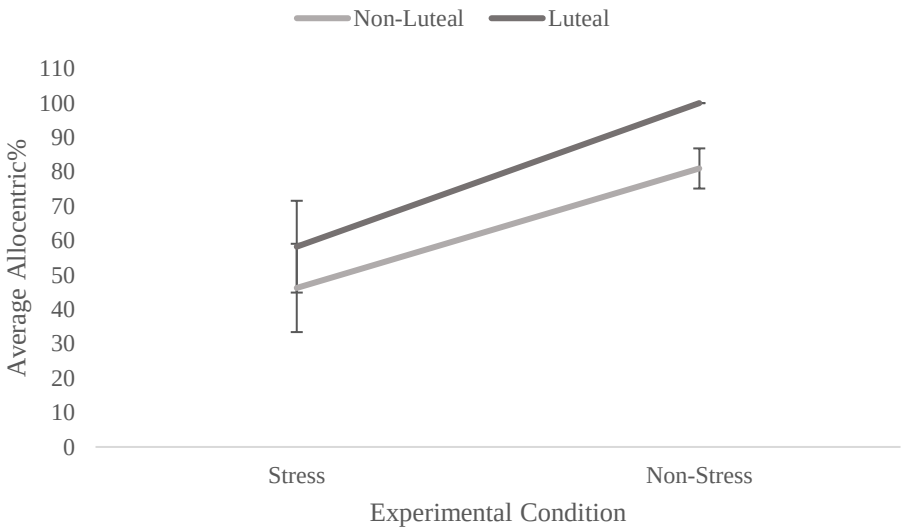


Figure 6. Non-significant interaction effect (Cycle Phase x Condition) on navigational strategy. Only participants in the Non-Luteal phase and the Stress condition showed a preference for an egocentric strategy over an allocentric strategy (i.e., Average Allocentric % < 50; descriptive statistics reported in Table 5). Error bars represent standard error of means for each group.

Conclusion

Overall, the results of this pilot study provide valuable insight for the main study. In particular, the analyses confirmed the hypothesis that stress impairs female spatial navigation. Furthermore, although the main effect of menstrual cycle phase and the interaction effect of menstrual cycle phase and stress were non-significant, descriptive statistics indicated that the directions of the effects were almost as predicted. Thus, these results are promising, and the data patterns encourage the larger-scale (and better-powered) exploration of the research question at hand.

Additionally, the pilot study allowed me to examine a proposed study methodology thoroughly. This examination permitted three broad conclusions. First, all the background and self-report questionnaires completed pre-manipulation appear to be good measures for comparing groups on demographic, lifestyle, psychological, and physiological factors that may influence navigational performance and response to the stress manipulation. One shortcoming of the current set of measures, however, was that none of them captured information about the current life climate of the participant (e.g., how much stress the individual had experienced over the month prior to reporting). Although the STAI gathers information about both general and in-the-moment anxiety levels, it does not ask whether the participant is experiencing high levels of stress in her daily life. Hence, I decided to add to

the main study's materials a standardized measure of stress perception and current levels of experienced stress, the Perceived Stress Scale (PSS; S. Cohen & Williamson, 1988), to ensure no group of participants is experiencing any more daily stress than another.

Second, the pilot study's results encouraged the use of the PASAT as the stress manipulation task and the use of the *u*-PASAT as the control task. In preparation for the main study, however, it should be noted that participants completing *u*-PASAT may still experience a slight rise in subjective stress given the mathematical nature of the task: Almost all pilot participants in the Non-Stress condition noted that, for them, mathematical tasks provoke anxiety. Nonetheless, I decided to continue to use the PASAT in the main study given that (a) it is time- and resource-efficient, (b) the two conditions were distinguishable at the post-stressor measurement point (with participants in the Stress condition reporting higher subjective anxiety and higher heart rates), and (c) there appear to be no long-lasting negative effects of completing this task.

Finally, the pilot study's navigation task, the CG Arena, was useful as it allowed me to compare allocentric and egocentric navigation strategies in much the same way as prior research had done (see, e.g., Hussain et al., 2016; Scheuringer & Pletzer, 2017). The results were promising and indicated differences where I had predicted them, supporting the continuation of this investigation with a main study. However, to expand the scope of this pilot study and the current body of literature, I thought it necessary to use a navigation task that would allow the investigation of these effects on three distinct navigational strategies (i.e., egocentric-response, egocentric-cue, allocentric). Hence, I decided to use the Hex Maze (Spriggs et al., 2018) in the main study. This recently published measure is the first VR navigation task to allow the concurrent measurement of all three strategies in humans.

CHAPTER FIVE:
METHODS

Design and Setting

This quasi-experimental study used a 3 x 2 factorial design. The independent variables were menstrual cycle phase (three levels: Early Follicular, Ovulatory, Mid/Late Luteal) and experimental condition (two levels: Stress, Non-Stress). The dependent variable was the preferred navigational strategy (allocentric, egocentric-cue, or egocentric-response), as determined by performance on the Hex Maze task.

Participants were assigned to a phase group based on the day of the cycle they were in when administered the study procedures. After being assigned to a phase group, the participant was randomly assigned to either the Stress or Non-Stress condition. Hence, I created six distinct groups of participants (see Table 6).

The study was conducted in a research laboratory housed within the University of Cape Town's Department of Psychology. Ethical approval was granted by the Department's Research Ethics Committee (reference number: PSY2019-004), and all study procedures followed the guidelines for human research laid down by the Declaration of Helsinki (World Medical Association, 2013)

Participants

Recruitment. Using convenience sampling, I recruited 84 female undergraduate students. Some recruitment was orchestrated via the UCT Department of Psychology's Student Research Participation Program (SRPP). Potential participants recruited via that route were notified of the study and the relevant eligibility criteria on the SRPP Vula site (see Appendix H). Other recruitment was orchestrated via a UCT Department of Student Affairs research invitation email (see Appendix I).

Eligibility criteria. I included only regularly-cycling women (i.e., with a menstrual cycle of between 25 and 34 days), aged 18-30 years, in the early follicular, ovulatory, or mid/late luteal phases of their menstrual cycles. Furthermore, and consistent with similar studies, potential participants were excluded if they (a) had taken, within 3 months of testing, any hormonal contraceptive medication; (b) had a prior history of neurological illness or substance abuse; (c) were experiencing any mood, anxiety, or endocrine disorder; (d) were taking a mood- or anxiety-stabilizing psychiatric medication; (e) had been pregnant within the past 2 years; or (f) were currently breastfeeding. All these factors can significantly affect

the natural cycle and associated hormone levels (De Leo, Musacchio, Cappelli, Piomboni, & Morgante, 2016; Liu, Gold, Lasley, & Johnson, 2004).

After applying these criteria, I excluded 22 of the original 84 participants. Two other participants were excluded because they had blood pressure readings that were unusually high (i.e., above 140/90) upon presentation at the laboratory.[2] Hence, the final sample consisted of 60 participants (see Table 1), with an average age of 19.45 years (*SD* = 2.10, range = 18-29) and an average educational level of 12.58 years (*SD* = 0.85, range = 12-15).

Table 6.
Sample Size for Each of the Six Study Groups (N = 60)

	Menstrual Cycle Phase Group		
Experimental Condition	Early Follicular	Ovulatory	Mid/Late Luteal
Stress	11	8	12
Non-Stress	10	8	11

Power analysis. I conducted this analysis using G*Power software (Faul, Erdfelder, Lang, & Buchner, 2007). With type of analysis set at ANOVA (fixed effects, special, main effects and interactions), and statistical parameters set at power (1 - β) = .80, α = .05, Cohen's *f* = .09 (a small effect size, as reported in Van Gerven et al., 2016) and groups = 6, the software indicated that an *N* of 1590 would be needed to adequately power the study. When Cohen's *f* was set to .25 (a medium effect size), the software indicated that an *N* of 211 would be needed to adequately power the study. When Cohen's *f* was set to .40 (a large effect size) the software indicated that *N* = 86 would be needed to adequately power the study.

Although it might be argued that the effect size the study seeks to detect will be of smaller magnitude than .40, an *N* of 60 is consistent with other studies in this field (Hausmann et al., 2000; Hussain et al., 2016; Iaria et al., 2003; Spriggs et al., 2018).

Materials and Procedure

Pre-manipulation. After scheduling individual appointments, I met each potential participant at the agreed-upon time in the laboratory (between 9am and 5pm) and administered consent procedures (see Appendix F) and the screening questionnaire. The

[2]Both participants were rescheduled for a second testing session, but their blood pressure readings were equally high at that session and so I excluded them from participation and encouraged them to consult with a medical professional.

screening questionnaire (see Appendix J) was similar to that used in the pilot study. This one, however, included a more specific time frame for responses (i.e., over the last month) and included more response options regarding alcohol and cigarette consumption.

Individuals who did not meet the eligibility criteria were dismissed immediately. Those who met the criteria were formally enrolled in the study and assigned to one of the three menstrual cycle phase groups. Specifically, if the participant reported she was within days 1-7 of the onset of her period, she was assigned to the Early Follicular group. If she reported being within days 13-17 of her cycle, she was assigned to the Ovulatory group. If she reported being between day 20 and the end of her cycle, she was assigned to the Mid/Late Luteal group.

I then measured the participant's height and weight and asked her to complete the STAI-Trait, STAI-State, and SBSOD questionnaires (for complete descriptions of these instruments, see Chapter 3). Participants also completed the Perceived Stress Scale (PSS; S. Cohen & Williamson, 1988). This 10-item scale (Appendix K) measured participants' self-reported perceptions of everyday stress and the degree to which they perceived life situations as stressful. Here, this scale was used to ensure no individual or group of participants was experiencing significantly more daily life stress than any other individual or group. Such confirmation was especially important because data collection ran over a 4-month period, starting early in the semester and ending just prior to exam season, when baseline stress levels may have been higher. The scale has solid psychometric properties, with internal reliability scores ranging from r = .74 to .91, and test-retest scores ranging from r = .73 to .90 over 2-week intervals (Lee, 2012).

Next, I strapped the HR sensor to the participant's chest and the BP cuff to her arm, and recorded baseline measurements as I did in the pilot study. The participant was then randomly assigned to either the Stress or Non-Stress experimental condition using an online randomization application (www.randomizer.org).

Stress manipulation. The experimental manipulation was almost identical to that used in the pilot study. Those assigned to the Stress condition were administered the PASAT. To increase their experience of socioevaluative threat, I told the participant that the task tests 'thinking speed' and that most people of her age do well. Those assigned to Non-Stress condition were administered the *u*-PASAT and were told nothing regarding what the test assesses. Immediately post-manipulation, I took a second measure of HR and BP, and the participant completed the STAI-State questionnaire for a second time.

Post-manipulation. A 30-min rest period followed the manipulation. During that time, the participant read a non-evocative magazine. After this rest period, I took a third set of HR and BP measurements. Thereafter, the participant completed the VR navigation task.

Navigation task: The Hex Maze. This piece of software, developed and first described by Spriggs et al. (2018), presents a six-arm RAM that follows the paradigmatic procedures of an MWM task. Hence, there are a fixed number of pathways along which the participant may find the desired target (a circular platform), but there is only one such target. Across a set of learning trials, that platform remains in a fixed position. In contrast, the participant's starting location changes from trial to trial. The software runs on a standard desktop computer, and participants move within the maze using the arrow keys on a standard keyboard.

Figure 7 (provided by and with permission from the developers) provides an aerial view of the maze, which is contained by a low stone wall and located within a large circular arena. This arena is held within a larger round room that has four windows. Two large windows are placed in the randomly labelled north and south quadrants. The north window shows a mountain range peaking, while the south shows a large body of water surrounding an island. There are also two narrower but equally high windows in the east and west quadrants that show views of the mountain ranges coming down to meet the large body of water. At the end of each of the six arms is a light grey sphere floating above the stone wall (Spriggs et al., 2018).

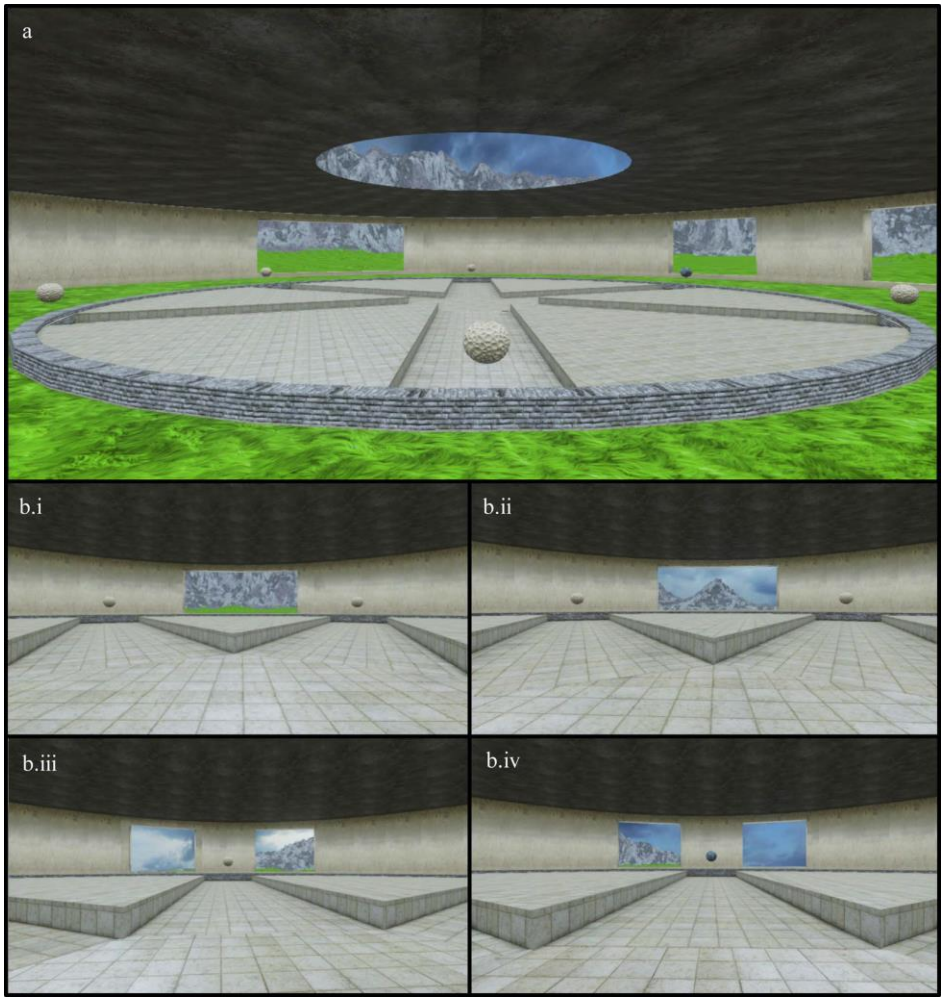


Figure 7. The Hex Maze. (a) A view of the virtual environment from above. Note that participants never see the maze from this angle. (b) Four views of the virtual environment looking (i) north, (ii) south, (iii) east, and (iv) west from the center of the maze.

The structure of the actual navigation task presented to participants mirrored that used by Spriggs et al. (2018).

Phase 1: Training. This phase of the protocol comprised three distinct kind of trials: (a) exploration, (b) visible platform, and (c) explicit probe. The *exploration trial* aimed to familiarize the participant with the environment and how to move within it. At the beginning of this trial the participant was teleported to the grassy area in the far eastern area of the room. She was instructed to explore the room, look outside the windows, and let the experimenter know when she was satisfied with her knowledge of the environment and the keyboard.

Thereafter, the participant completed four *visible platform trials*. This set of trials aimed to familiarize the participant with the platform-finding procedure. She began at the end of one arm and was instructed to move as quickly and directly as possible to a platform (easily visible at all times) located within the maze, on the floor of the arena. To prevent any

learning of the environment during these trials, the inner wall was raised so that it masked the outside landscape (i.e., the windows were blocked out) and the spheres were removed from the end of the arms. The location of the visible platform was different on each trial. When the participant reached the platform, she became trapped within that space and a beep sounded. She was instructed to look around the arena until she stated she was ready to move on, at which point I began the next trial.

After completing the set of visible trials, the participant completed a single *explicit probe* trial. She was told this was the first "Show-Me" trial. I explained that I would like to see how good her "intuition" is and instructed her to go to the end of the arm where she thought the invisible platform might be placed in the upcoming set of trials. The participant started in the center of the maze, facing east, and was not informed that one sphere was colored dark grey while the others were colored light grey. The aim of this trial was to determine whether the participant guessed that the platform would be hidden in the arm with the dark grey sphere, and/or if her navigation was unintentionally biased by something else in the environment.

Phase 2: Testing. This phase of the Hex Maze protocol followed immediately after the training phase and was comprised of 10 pairs of trials. Each pair consisted of a learning ("Find-It") trial followed by an explicit probe ("Show-Me") trial. The 10 trial pairs were divided equally over two environmental configurations in which the starting and platform locations changed to reduce the effects of overtraining. Before starting the testing session, I informed the participant that she would complete two types of trials that would be presented alternately. I then briefly described the structure of each trial type. On the "Find-It" trials, participants were asked to find an "invisible" platform (i.e., one hidden below the floor of the room) at the end of an arm. The end of each arm held a floating sphere; all were a light grey color except the arm which held the platform, which was marked by a dark grey sphere (first 5 trial pairs) or a blue sphere (second 5 trial pairs). When the participant walked across the space occupied by the platform, it rose below her, and a beep sounded. As in the visible platform trials, the participant was unable to move off the platform but was able to look around the environment as much as she liked before informing me she was ready to move on.

On the "Show-Me" trials, the platform was removed from the arena and the participant was asked to navigate back to the place it was located on the immediately preceding "Find-It" trial. When the participant reached the end of her chosen arm, a beep sounded, and the participant was prevented from leaving the arm.

Participants were also told that the platform would always be in the same location and that they would be informed which type of trial they would be completing. However, they were not informed of any changes in setting between the two trial types. That is to say, they were not told that (a) on each Show-Me trial, the different colored sphere (i.e., dark grey in the first environment, blue in the second environment) changed location and therefore no longer marked the arm in which the platform had been located, and (b) the starting location would differ from trial to trial. These changes in trial setting allowed me to distinguish between the use of three different navigational strategies by observing which arm the participant chose during the Show-Me trials. If the participant followed the windows in the arena and headed directly for the arm in which the platform had originally been located, she was scored as using an allocentric strategy. If she chose the arm that now housed the colored sphere, she was scored as using an egocentric-cue strategy. If she followed the direction of the body turns (e.g., entered the second left arm from the starting point), she was scored as using an egocentric-response strategy, as her navigation corresponded to the original turning direction.

Post-navigation. Immediately after the participant had completed the Hex Maze task, I took a fourth measure of HR and BP and she completed the STAI-State questionnaire a third time. I then administered a post-maze questionnaire (see Appendix L), as done in the pilot study, to gather information on how the participants completed the Hex Maze. The same video game experience questionnaire as in the pilot study was also administered. Upon completion of those questionnaires, I took a final (fifth) measure of HR and BP, and the participant completed the STAI-State questionnaire for the final (fourth) time. At the end of these procedures, I debriefed the participant completely (see Appendix G).

Figure 8 is a diagrammatic depiction of the entire study procedure.

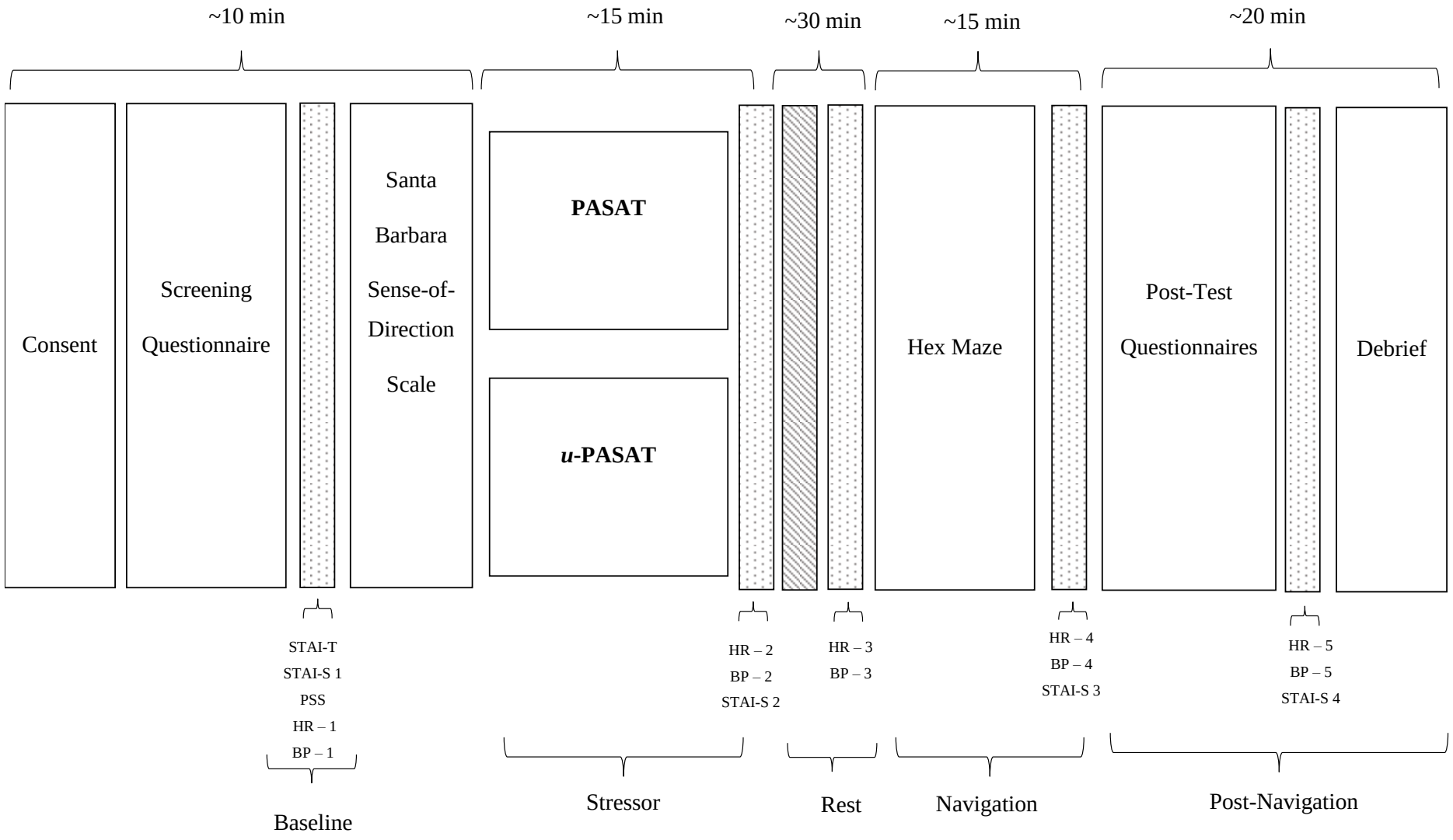


Figure 8. The study procedure. PASAT = Paced Auditory Serial Addition Task; *u*-PASAT – *un*-Paced Auditory Serial Addition Task; HR = heart rate; BP = blood pressure; STAI = State-Trait Anxiety Inventory (T = trait form, S = state form); PSS = Perceived Stress Scale.

Data Management and Statistical Analyses

I completed all statistical analyses using SPSS (version 25.0), with the threshold for statistical significance set at $\alpha = .05$. Effect size estimates were interpreted using conventional guidelines (small: Cohen's $d = 0.2$, $\eta^2 = .01$, Cramer's $V = .07$ - $.21$; medium: Cohen's $d = 0.5$, $\eta^2 = .06$, Cramer's $V = .21$ - $.35$; large: Cohen's $d = 0.8$, $\eta^2 = .14$, Cramer's $V > .35$; J. Cohen, 1988).

Prior to conducting inferential analyses, I generated and examined a complete set of descriptive statistics to ensure that all assumptions underlying those analyses were met. In cases where they were not met, I adjusted the analytic plan. Details of these adjustments are given at the appropriate places in the Results section.

Sample characteristics. A series of one-way ANOVAs (for continuous variables) and chi-squared tests (for categorical variables) assessed the magnitude of between-condition (Stress versus Non-Stress) differences with regard to basic sample characteristics (e.g., age, STAI-Trait scores, SBSOD scores, video game experience, average length of menses).

Manipulation check.[3] A series of independent-sample *t*-tests examined between-condition (Stress versus Non-Stress) differences in the magnitude of change from baseline to the immediate post-stressor measure of physiological and self-reported stress. Then, a series of paired-sample *t*-tests examined within-group differences between baseline and immediate post-stressor measures. A separate series of independent-sample *t*-tests investigated between-condition differences in physiological and self-reported stress at each measurement point (baseline, post-stressor, post-rest, post-Hex Maze, and pre-debrief). Together, these analyses investigated the effectiveness of PASAT in inducing stress and sought to confirm that the *u*-PASAT did not induce stress.

Finally, because it was ethically important to ensure that Stress-group participants left the laboratory in the same psychological state as when they entered, a series of paired-sample *t*-tests investigated the magnitude of differences between the first (baseline) and last (pre-debrief) measures of physiological and self-reported stress within that group.

Navigation strategies. Recall that Hypothesis 1 stated that menstrual cycle phase will have a significant main effect on navigational strategy use (i.e., participants in the early follicular and ovulatory phases will tend to use an egocentric strategy, whereas those in the mid/late luteal phase will tend to use an allocentric strategy), that Hypothesis 2 stated that

[3]For the purposes of these analyses, the participant's menstrual cycle phase was not relevant and therefore was not included as a factor.

stress will have a significant main effect on navigational strategy use (i.e., participants exposed to an acute psychosocial stressor, but not those unexposed, will tend to use one of the egocentric strategies, rather than an allocentric strategy), and that Hypothesis 3 stated that stress and menstrual cycle phase will have a significant interactional effect on navigational strategy use, particularly among participants in the mid/late luteal phase. For instance, participants in that phase who are exposed to an acute psychosocial stressor will tend to use one of the egocentric strategies, whereas those who are in that phase but not exposed to the stressor will tend to use an allocentric strategy. Additionally, those not in that phase will tend towards one of the egocentric strategies, regardless of exposure to the stressor

The primary analysis here, which tested all the major hypotheses simultaneously, was a series of four linear mixed models that included fixed effects of menstrual cycle phase, stress condition, and the interaction between the two. The outcome variables were the *number of Show-Me trials* on which the participant used (a) an egocentric-response strategy, (b) an egocentric-cue strategy, (c) any egocentric strategy (this model facilitated comparison of the current data with those from previous papers that did not use the Hex Maze), and (d) an allocentric strategy. The linear mixed-model analysis extends the capabilities of the standard general linear model (i.e., factorial ANOVA) as it allows for unequal variances (West, Welch, & Galecki, 2014).

The next analytic step used a different outcome variable to capture navigation in the Hex Maze: *preferred strategy over the set of Show-Me trials*. To determine the value of this variable for each participant, I counted, across the 10 Show-Me trials, the number of times she had used each of the three navigational strategies. Whichever was used the most was deemed her preferred strategy. If she showed no preference (e.g., used each of the egocentric strategies five times), her data were excluded from the analysis, which was a 6 (Group: Early Follicular+Stress, Early Follicular+Non-Stress, Ovulatory+Stress, Ovulatory+Non-Stress, Mid/Late Luteal+Stress, Mid/Late Luteal+Non-Stress) x 3 (Preferred Strategy: Egocentric-Response, Egocentric-Cue, Allocentric) chi-squared test of independence.

I then conducted a set of secondary analyses to explore the main effects of menstrual cycle phase and stress separately. These allowed me to report on aspects of Hypotheses 1 and 2 in more depth than the linear mixed models allowed, and to facilitate comparison with previous studies more readily. Hence, the first of these analyses was a 3 (menstrual cycle phase: Early Follicular, Ovulatory, Mid/Late Luteal) x 3 (Preferred Strategy: Egocentric-Response, Egocentric-Cue, Allocentric) chi-square test of association, using data from the Non-Stress group only. This analysis, then, explored the effect of menstrual cycle phase alone

on navigational strategy preference (i.e., it tested Hypothesis 1 directly). The second of these analyses was a 2 (stress condition: Stress, Non-Stress) x 3 (Preferred Strategy: Egocentric-Response, Egocentric-Cue, Allocentric) chi-square test of association, using data from all participants. This analysis, then, explored the effect of stress alone on navigational strategy preference (i.e., it tested Hypothesis 2 directly).

CHAPTER SIX:
RESULTS

Sample Characteristics

The measured data for all the continuous variables (Age, Education, BMI, STAI-Trait scores, SBSOD scores, PSS scores, Age of First Period, and Average Length of Menses) were not normally distributed. However, for all these variables except Age of First Period, the assumptions of independence and homogeneity of variance were upheld, and so I proceeded with the planned analyses because ANOVA is robust against deviations of normality (Blanca, Alarcon, Arnau, Bono, & Bendayan, 2017).

For the variable Age of First Period, the homogeneity of variance assumption was violated, Levene's test $F(1,58) = 4.93$, $p = .030$. In this case, then, I used a Brown-Forsythe test.

The measured data for the categorical variables of average hours of sleep over the past month and average number of exercise hours per week over the past month upheld the assumptions of independence and expected cell counts above 5. Thus, for these variables, I continued with the planned chi-square analyses.

For the variables average number of alcoholic drinks per week over the past month, average number of cigarettes smoked per week over the past month, and years of video game experience, the expected cell counts above 5 assumption was not upheld. Thus, for these variables, the Freeman-Halton extension of Fisher's Exact Test was run to complete the analysis (see Table 7 for relevant *p* and Cramer's V values).

Analyses detected no significant between-condition differences for most of the measured sociodemographic, lifestyle, psychological, and physiological variables (see Table 7). The one exception here was SBSODS score, where participants in the Non-Stress condition had a significantly higher average score than those in the Stress condition. Close investigation of the SBSODS data revealed one possible outlier in the Non-Stress condition, with a score of 6.60 (more than 2 *SD*s higher than the group mean). When that data point was removed, the analyses detected no significant between-condition differences on the SBSODS. I ran all of the analyses described below with and without the data provided by the individual with the outlying SBSODS score; there were no notable differences in the pattern of results, or the trends identified. Hence, I decided to report the analyses that retain that person's data.

Overall, these findings imply that the current results were not confounded by between-group differences on any of the measured sociodemographic, lifestyle, psychological, and physiological variables.

Table 7.
Sample Characteristics: Descriptive statistics and between-group comparisons (*N = 60)*

	Group				
Measure	Stress (*n* = 31)	Non-Stress (*n* = 29)	*F* / x^2	*p*	ESE
Age (years)	19.58 (2.62)	19.31 (1.37)	0.25	.622	.<.01
Range	18-29	18-22			
Education (years)	12.52 (0.72)	12.66 (0.97)	0.40	.531	<.01
Range	12-14	12-15			
BMI	23.49 (3.83)	25.17 (5.09)	2.11	.152	.03
Range	16.51-36.39	18.11-39.02			
STAI-Trait	43.26 (10.75)	42.59 (8.30)	0.73	.788	<.01
Range	24-63	28-59			
SBSOD	3.85 (1.10)	4.40 (0.99)	4.16	.046*	.07
Range	1.60-5.73	2.40-6.60			
PSS	20.42 (5.97)	20.45 (6.12)	< 0.001	.985	<.01
Range	8-31	12-36			
Sleep (hours)			0.67	.414	.11
4-6	15 (48.40)	11 (37.90)			
7-10	16 (51.60)	18 (62.10)			
Exercise (hours)			0.21	.900	.06
> 1	9 (29.00)	8 (27.60)			
1-2	9 (29.00)	10 (34.50)			
3-7	13 (41.90)	11 (37.90)			
Alcohol (drinks)			N/A	.538[b]	.23
0	19 (61.30)	13 (44.80)			
1-2	9 (29)	13 (44.80)			
3-5	1 (3.20)	2 (6.90)			
6-8	1 (3.20)	1 (3.40)			
9-11	1 (3.20)	0			
Cigarettes			N/A	.228[b]	.22
0	29 (93.50)	28 (96.60)			
< 10	2 (6.50)	0			
10-20	0	1 (3.40)			
Video Games (years)			N/A	.621[b]	.17
< 1	28 (90.30)	25 (86.20)			
3+	1 (3.20)	2 (6.90)			
Age at first period	12.45 (0.81)	12.07 (1.33)	1.77[a]	.190	.03
Range	11-14	9-15			
Menses	5.74 (1.75)	5.34 (1.34)	0.96	.331	.02
Range	3-10	3-9			

Note. For continuous variables, means, standard deviations, and ranges are presented; for categorical variables, frequencies with percentages in parentheses. *Sleep* represents average duration per night, in hours, over the last month; *Exercise*, average amount per week, in hours; *Alcohol*, average number of drinks per week; *Cigarettes*, average number smoked per week; *Video Games*, playing experience, in years; *Menses*, average period length, in days. STAI = State-Trait Anxiety Inventory; SBSOD = Santa Barbara Sense of Direction scale; PSS = Perceived Stress Scale; BMI = Body Mass Index; ESE = effect size estimate (η^2 for ANOVAs, and Cramer's *V* for chi-squared tests).
[a]Brown-Forsythe Statistic [b] Freeman-Halton extension of Fisher's Exact Test *$p < .05$.

Manipulation Check

For both HR and STAI-State data, analyses detected significant between-condition differences in the magnitude of change from baseline to immediate post-stressor (see Table 8 and Figure 9, panel (a) and panel (b)). For both those variables, the average change value was much higher for participants in the Stress condition than for those in the Non-Stress condition. Analyses of the analogous BP data detected no significant between-condition differences (see Figure 9, panel (c) and panel (d)).

The next set of analyses compared, within each group separately, indices of stress at baseline versus the immediate post-stressor measurement point. For participants assigned to the Stress condition, the analyses detected:

(a) a significant increase in HR, $t(30) = -6.04$, $p < .001$, Cohen's $d = -0.82$;

(b) a significant decrease in Systolic BP, $t(30) = 2.22$, $p = .034$, $d = 0.35$;

(c) a non-significant decrease in Diastolic BP, $t(30) = .343$, $p = .734$, $d = 0.05$; and

(d) a significant increase in STAI-State scores, $t(30) = -8.02$, $p < .001$, $d = -1.63$.

For participants assigned to the Non-Stress condition, these analyses detected:

(a) a significant decrease in heart rate, $t(28) = 2.05$, $p = .05$, Cohen's $d = 0.36$;

(b) a non-significant decrease in Systolic BP, $t(28) = .57$, $p = .575$, $d = 0.10$;

(c) a significant decrease in Diastolic BP, $t(28) = 2.13$, $p = .042$, $d = 0.28$; and

(d) a significant increase in STAI-State scores, $t(28) = -2.87$, $p = .008$, $d = -0.52$.

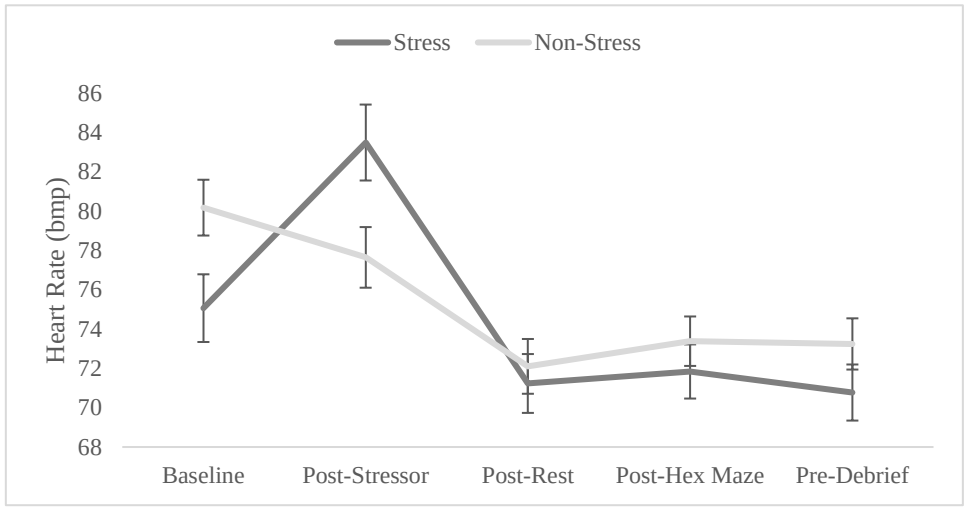
Analyses of data from each measurement point separately detected only three significant between-condition differences: (a) at baseline, participants in the Non-Stress condition had significantly higher average HR than those in the Stress condition; (b) at post-stressor, participants in the Stress condition had significantly higher average HR than those in the Non-Stress condition; and (c) at post-stressor, participants in the Stress condition had significantly higher average STAI-State scores than those in the Non-Stress condition,

The final set of analyses, which focused on the participants in the Stress condition only, detected significant decreases in HR and Systolic BP (but no significant change in Diastolic BP and STAI-State scores) from the first (baseline) to the last (pre-debrief) measurement point (see Table 9).

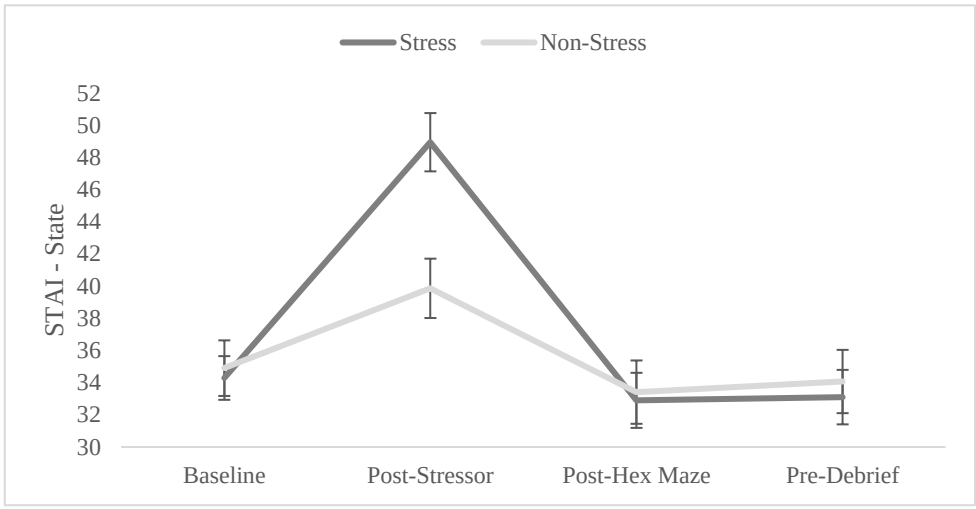
Table 8.
Physiological and Self-reported Stress: Descriptive statistics and between-condition comparisons (N = 60)

	Experimental Condition				
	Stress	Non-Stress			
Measure / Measurement Point	(*n* = 31)	(*n* = 29)	*t*	*p*	ESE
HR					
Baseline (1)	75.06 (9.58)	80.17 (7.64)	-2.27	.027*	-0.59
Post-Stressor (2)	83.48 (10.75)	77.34 (8.30)	2.46	.017*	0.64
Δ2 - 1	8.42 (7.76)	-2.83 (7.45)	5.72	*p* < .001***	1.48
Post-Rest (3)	71.23 (8.33)	72.10 (7.50)	-0.43	.670	-0.11
Post-Hex Maze (4)	71.84 (7.66)	73.38 (6.77)	-0.82	.414	-0.21
Pre-Debrief (5)	70.77 (7.94)	73.24 (7.01)	-1.27	.208	-0.33
BP: Systolic					
Baseline (1)	115.32 (8.86)	116.55 (8.35)	-0.55	.583	-0.14
Post-Stressor (2)	111.61 (11.64)	115.62 (9.90)	-1.43	.157	-0.37
Δ2 - 1	-3.71 (9.31)	-0.93 (8.83)	-1.19	.241	-0.31
Post-Rest (3)	111.26 (8.86)	113.10 (9.03)	-0.80	.428	-0.21
Post- Hex Maze (4)	109.35 (9.23)	112.38 (10.03)	-1.22	.229	-0.32
Pre-Debrief (5)	110.42 (9.11)	110.83 (9.44)	-0.17	.865	-0.04
BP: Diastolic					
Baseline (1)	69.58 (7.30)	72.52 (6.15)	-1.68	.098	-0.43
Post-Stressor (2)	69.26 (7.09)	70.59 (7.45)	-0.71	.482	-0.18
Δ2 - 1	-0.32 (5.23)	-1.93 (4.88)	1.23	.224	0.32
Post-Rest (3)	66.77 (7.62)	69.31 (6.92)	-1.35	.183	-0.35
Post- Hex Maze (4)	69.87 (7.77)	69.21 (7.84)	0.33	.743	0.09
Pre-Debrief (5)	69.77 (7.52)	69.17 (7.20)	0.32	.753	0.32
STAI-State					
Baseline (1)	34.29 (7.56)	34.90 (9.30)	-0.28	.782	-0.07
Post-Stressor (2)	48.94 (10.08)	39.86 (9.93)	3.51	.001**	0.91
Δ2 - 1	14.65 (10.16)	4.97 (9.31)	3.84	< .001***	0.99
Post- Hex Maze (3)	32.90 (9.53)	33.41 (10.60)	-0.20	.845	-0.05
Pre-Debrief (4)	33.10 (9.43)	34.07 (10.59)	-0.38	.708	-0.10

Note. Means are presented with standard deviations in parentheses. HR = heart rate (measured in beats per minute [bpm]); BP = blood pressure (measured in mmHG); STAI = State-Trait Anxiety Inventory; ESE = effect size estimate (Cohen's *d*). Degrees of freedom = (1, 58) for all comparisons.
*$p < .05$. **$p < .01$. ***$p < .001$.



(a)



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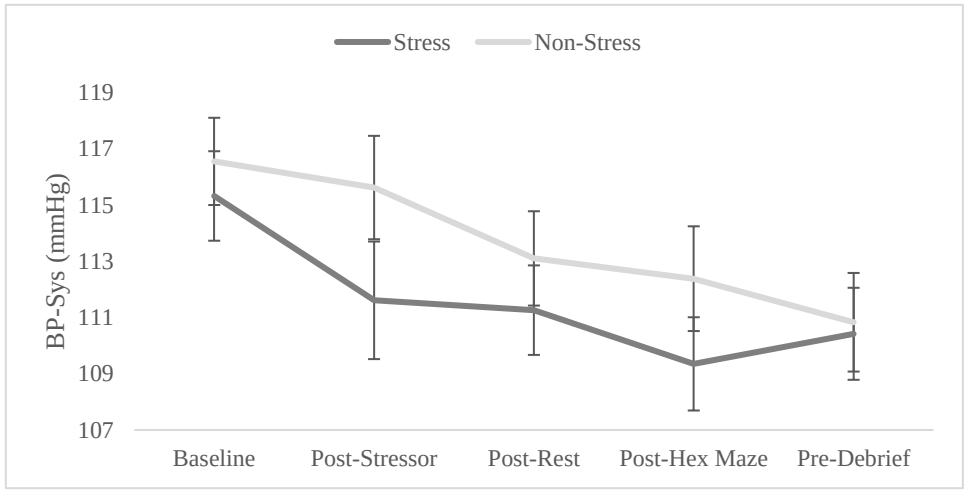

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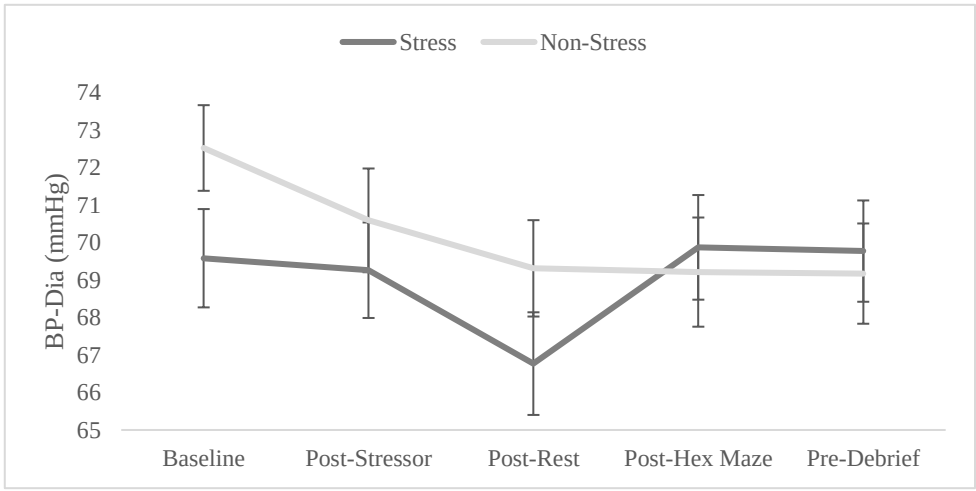

(d)

Figure 9. Physiological and self-report measures of stress across the study. Stress condition *n* =31, Non-Stress condition *n* = 29. Error bars represent the standard error of the mean for each measurement point.

Table 9.
Change in Stress Measures from Baseline to Pre-Debrief: Stress-group participants (N = 31)

	Measurement Point				
Variable	Baseline	Pre-Debrief	*t*	*p*	ESE
HR	75.06 (9.58)	70.77 (7.94)	5.52	< .001***	0.46
BP					
Systolic	115.32 (8.86)	110.42 (9.11)	3.39	.002**	0.55
Diastolic	69.58 (7.30)	69.77 (7.52)	-0.13	.899	-0.03
STAI-State	34.29 (7.56)	33.10 (9.43)	0.76	.453	0.14

Note. Means are presented, with standard deviations in parentheses. HR = heart rate (measured in beats per minute [bpm]); BP = blood pressure (measured in mmHG); STAI = State–Trait Anxiety Inventory; ESE = effect size estimate (Cohen's *d*).
*$p < .05$. ** $p < .005$. ***$p < .001$.

Interim summary. Taken together, these results suggest that exposure to the PASAT was associated with substantial increases in physiological stress (as indexed by HR levels) and in self-reported stress (as indexed by STAI-State scores). In contrast, exposure to the *u*-PASAT was associated with substantial decreases in HR levels and relatively minor increases in STAI-State scores. Hence, at both physiological and subjective levels, participants in the two experimental conditions were distinguishable from one another at the immediate post-manipulation measurement point. However, within the Stress group, both physiological and self-report measures of stress were not significantly increased over baseline when the participants left the laboratory, indicating no chronic effects of exposure to the experimental manipulation.

Effects of Menstrual Cycle Phase and Stress on Navigation Strategy

Primary analyses. The linear mixed-model analyses, which tested the three hypotheses simultaneously, detected no significant main or interaction effects (see Table 10). Of note here is that the standard deviations related to the number of times the participant used each strategy were quite high, indicating substantial interindividual variability.

For the follow-up 6 x 3 chi-square analysis, the expected count of most cells in the contingency table did not meet the required minimum of 5 (see Table 11). Hence, I used the Freeman-Halton extension of Fisher's Exact Test to complete the analysis. Results replicated those of the linear mixed-model analyses, indicating no significant between-group differences in preferred strategy, $p = .309$, Cramer's $V = .28$.

To facilitate comparison with previous papers that compared only egocentric to allocentric strategies (i.e., did not use the Hex Maze and did not split egocentric-response

from egocentric-cue strategies), I also conducted a 6 x 2 chi-square analysis, this time collapsing the two egocentric strategies into one. Again, the expected count of most cells in the contingency table did not meet the required minimum of 5 (see Table 11). Again, I used the Freeman-Halton extension of Fisher's Exact test to complete the analysis and, again, the analysis detected no significant between-group differences in preferred strategy, $p = .237$, Cramer's $V = .28$.

Table 10.
Effects of Menstrual Cycle Phase and Stress on Navigation Strategies: Descriptive statistics and linear mixed-model analyses (N = 60)

	Group									
	Stress			Non-Stress						
	EF	O	MLL	EF	O	MLL				
Strategy Type	(*n* = 11)	(*n* = 8)	(*n* = 12)	(*n* = 10)	(*n* = 8)	(*n* = 11)	Effect	*F*	*df*	*p*
Egocentric										
Response	2.64 (2.25)	1.88 (3.72)	2.75 (4.09)	4.20 (5.57)	3.38 (3.78)	2.09 (2.12)	Menstrual Cycle Phase	0.48	2,54	.311
							Stress Condition	0.76	1,54	.193
							Interaction	0.69	2,54	.252
Cue	2.27 (2.53)	3.00 (3.67)	2.25 (3.82)	3.10 (4.58)	1.50 (2.62)	1.73 (2.41)	Menstrual Cycle Phase	0.24	2,54	.395
							Stress Condition	0.20	1,54	.327
							Interaction	0.56	2,54	.288
Any	4.91 (3.81)	4.88 (4.64)	5.00 (4.67)	7.30 (4.06)	4.88 (4.58)	3.82 (2.56)	Menstrual Cycle Phase	0.99	2,54	.187
							Stress Condition	0.14	1,54	.353
							Interaction	1.08	2,54	.174
Allocentric	4.64 (4.15)	5.00 (4.63)	4.25 (4.62)	2.50 (4.14)	4.63 (4.60)	5.91 (3.02)	Menstrual Cycle Phase	0.78	2,54	.232
							Stress Condition	0.07	1,54	.399
							Interaction	1.13	2,54	.166

Note. In columns 2-6, means (for number of Show-Me trials the participants in the given group used the particular strategy; possible range is 0-10) are presented with standard deviations in parentheses. EF = Early Follicular; O = Ovulatory; MLL = Mid/Late Luteal. All *p*-values reported are one-tailed because the hypotheses were directional.

Table 11.
Frequencies of Preferred Strategy across Study Groups (N = 57)

	Group					
	Stress			Non-Stress		
	EF	O	MLL	EF	O	MLL
Strategy Type	(*n* = 11)	(*n* = 6)	(*n* = 12)	(*n* = 10)	(*n* = 8)	(*n* = 10)
Egocentric						
Response	2.00 (13.30)	1.00 (6.70)	2.00 (13.30)	5.00 (33.30)	3.00 (20.00)	2.00 (13.30)
Cue	3.00 (23.10)	1.00 (7.70)	4.00 (30.80)	3.00 (23.10)	1.00 (7.70)	1.00 (7.70)
Any	5.00 (16.70)	3.00 (10.00)[a]	6.00 (20.00)	8.00 (26.70)	4.00 (13.30)	4.00 (13.30)[b]
Allocentric	6.00 (20.70)	4.00 (13.80)	6.00 (20.70)	2.00 (6.90)	4.00 (13.80)	7.00 (24.10)

Note. Frequencies are presented with percentages in parentheses. Of the original *N* of 60, three participants (two from the Ovulatory Stress group and one from the Mid/Late Luteal Stress group) did not display a preferred strategy and so their data were excluded from the analyses.
[a]*n* = 7; [b] *n* = 11.

Secondary analyses. Figure 10 shows, across the menstrual cycle phase groups, the number of participants (Non-Stress condition only) who preferred each of the three navigation strategies on the set of 10 Show-Me trials. The sample size here is $n = 28$ because one participant had equal counts of the allocentric strategy and the egocentric-response strategy (i.e., did not evince a particular preference), and thus her data were removed from this analysis. Analysis of the contingency table revealed all cells had expected frequencies below 5, with some smaller than 1, indicating that a standard chi-square analysis was not appropriate for the data. Thus, I used the Freeman-Halton extension of Fisher's Exact Test to complete the analysis. Results again replicated those of the previous analyses, indicating no significant between-group differences in preferred strategy, $p = .142$ (one-tailed), Cramer's $V = .31$.

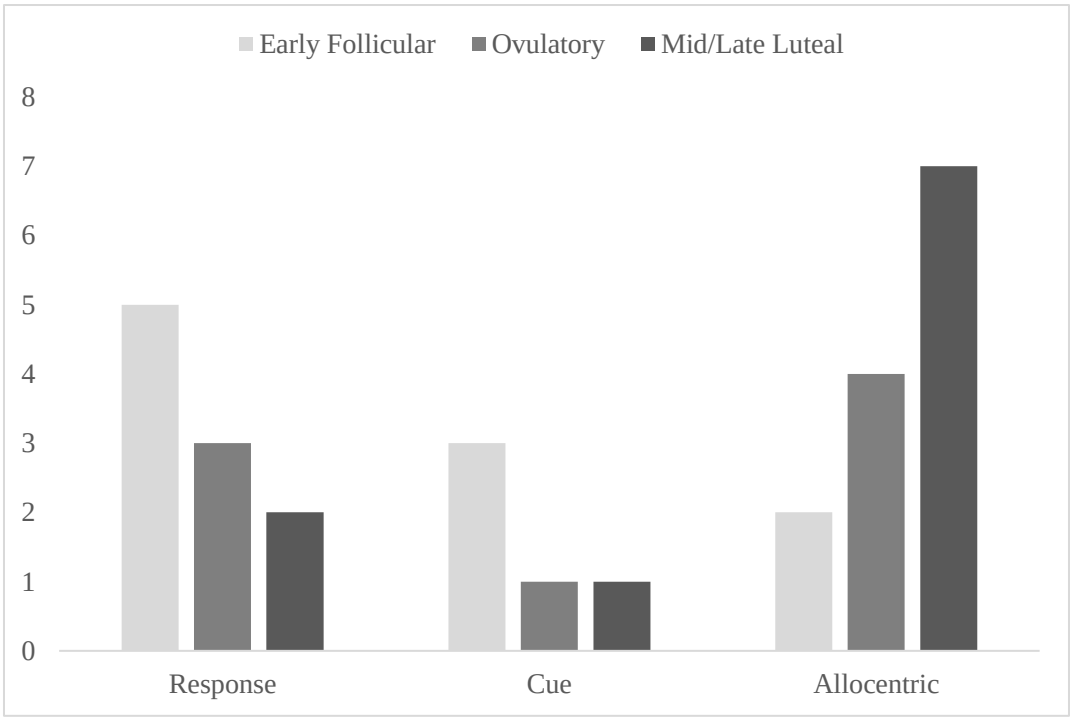

Figure 10. Preferred navigation strategy across the 10 Show-Me trials for participants in different menstrual cycle phases (Non-Stress condition only, $n = 28$).

To facilitate comparison with previous papers that compared only egocentric to allocentric strategies (i.e., did not use the Hex Maze and did not split egocentric-response from egocentric-cue strategies), I also conducted a 3 x 2 chi-square analysis, this time collapsing the two egocentric strategies into one. Again, the expected count of some cells in the contingency table did not meet the required minimum of 5 (see Figure 11). Again, I used

the Freeman-Halton extension of Fisher's Exact test to complete the analysis and, again, the analysis detected no significant between-group differences in preferred strategy, $p = .068$ (one-tailed), Cramer's $V = .38$.

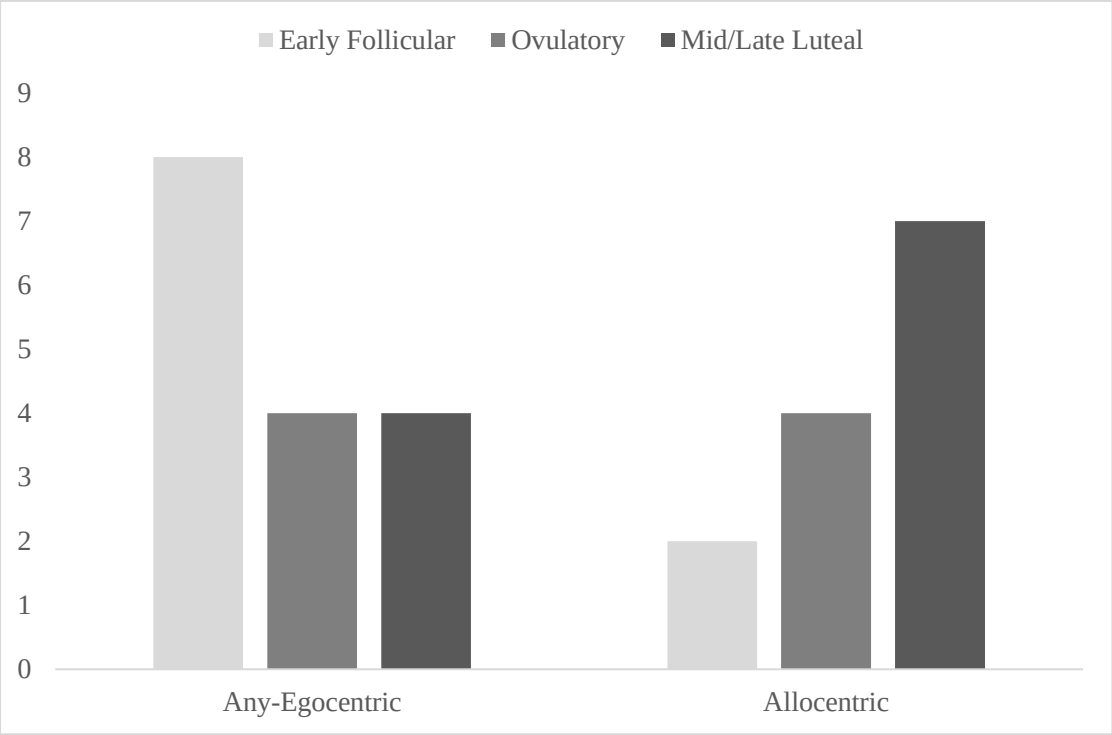

Figure 11. Preferred strategy (this time comparing only egocentric versus allocentric, as in previous research) across the 10 Show-Me trials for participants in different menstrual cycle phases (Non-Stress condition only, $n = 29$).

Figure 12 shows, across the stress conditions, the number of participants who preferred each of the three navigation strategies on the set of 10 Show-Me trials. The sample size here is $N = 57$ because three participants did not show a strategy preference and thus their data were removed from this analysis. Analysis of the contingency table revealed all cells had expected frequencies of 5 or higher, thus upholding the assumption for the chi-square analysis. Results again replicated those of the previous analyses, indicating no significant between-group differences in preferred strategy, x^2 (2, $N = 57$) = 2.65, $p = .133$ (one-tailed), Cramer's $V = .22$.

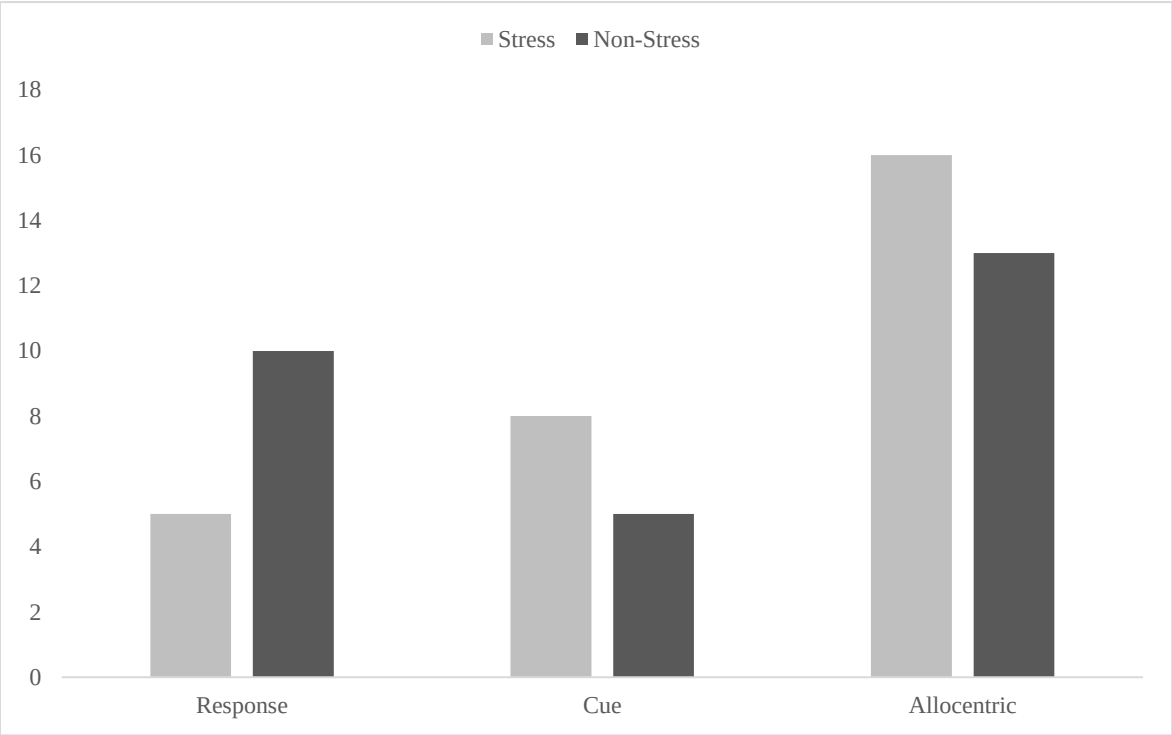

Figure 12. Preferred navigation strategy across the 10 Show-Me trials for participants in the different stress conditions (N = 57).

Again, to facilitate comparison with previous papers that compared only egocentric to allocentric strategies, I also conducted a 2 x 2 chi-square analysis, this time collapsing the two egocentric strategies into one (see Figure 13). Analysis of the contingency table revealed all cells had expected frequencies of 5 or higher, thus upholding the assumption for the chi-square analysis. Results again replicated those of the previous analyses, indicating no significant between-group differences in preferred strategy, x^2 (1, N = 59) = .427, p = .257 (one-tailed), Cramer's V = .09.

Figure 13. Preferred strategy (this time comparing only egocentric versus allocentric, as in previous research) across the 10 Show-Me trials for participants in the different stress conditions (N = 59).

Note that because the results of the linear mixed models were non-significant there was no need for further investigation of the other outcome variable (number of Show-Me trials on which each strategy was used).

CHAPTER SEVEN:
GENERAL DISCUSSION

Previous research has explored the effects of two endocrine-related variables (phase of the menstrual cycle and exposure to acute psychosocial stress) on choice and use of navigational strategy in humans. However, all previously published studies in this literature have investigated each of these variables independently of the other. To this point there has been no study exploring the ecologically relevant question of how the interaction between menstrual cycle phase and stress exposure might affect spatial navigation. Additionally, prior studies have used tasks that measure (alongside allocentric navigation) only one form of egocentric navigation, despite the fact that egocentric-cue and egocentric-response strategies are distinguishable at both neural and behavioral levels. Hence, the research presented in this thesis is novel in that it sought to expand the current body of literature by exploring both the main and interaction effects of menstrual cycle phase and stress exposure on choice and use of allocentric, egocentric-response, and egocentric-cue navigational strategies.

In this chapter, I discuss the results related to each of the study's three hypotheses and explore how these findings fit with those from previous empirical studies and align to existing theoretical frameworks. The chapter, and the thesis, concludes with consideration of the study's limitations and some recommendations for future research.

Effects of Menstrual Cycle Phase on Choice and Use of Navigational Strategy

Hypothesis 1 stated that menstrual cycle phase will have a statistically significant effect on navigation strategy such that (unstressed) participants in the early follicular and ovulatory phases will tend to use an egocentric strategy (egocentric-cue and/or egocentric response), whereas those in the mid/late luteal phase will tend to use the allocentric strategy. This hypothesis was not confirmed.

Analyses detected no significant between-phase differences in strategy use (the frequency of egocentric-response, egocentric-cue, egocentric-combined, and allocentric strategy use) or in overall strategy preference (i.e., the strategy used most frequently) across the 10 Show-Me trials of the Hex Maze task. These non-significant findings remained even when I analyzed only egocentric (response and cue, combined) versus allocentric strategy use, as has been done in all previous studies in this literature. However, perusal of the descriptive statistics related to allocentric versus egocentric (response and cue, combined) strategy preference revealed that the observed means for the Early Follicular and Mid/Late Luteal phase groups followed the predicted direction (i.e., women in the early follicular phase tended toward egocentric strategy use whereas those in the mid/late luteal phase tended

toward allocentric strategy use). Additionally, subsequent inferential analyses came closer to detecting a significant main effect (p = .068) when comparing egocentric (response and cue, combined) to allocentric strategy use than when I compared egocentric-cue versus egocentric-response versus allocentric strategy use (p = .142).

Despite these promising trends, the non-significant current results are inconsistent with findings reported in the two recent studies investigating this effect. Both those studies reported a significant main effect of menstrual cycle phase on navigational strategy. Specifically, Scheuringer and Pletzer (2017) found that participants in the luteal phase, who preferred an allocentric strategy, performed significantly better on a computerized 2D-matrix navigation task than those in the early follicular phase, who preferred an egocentric strategy. Similarly, Hussain et al. (2016) found that women in the early follicular and ovulatory phases preferred using egocentric strategies when solving a VR navigation task, whereas those in the luteal phase tended towards using an allocentric strategy.

The inconsistency in results between the current study and those studies might be attributable to across-study differences in navigation tasks and measures. Scheuringer and Pletzer did not use a 3D navigation task at all. Hussain et al. (2016) used a traditional VE RAM. In the first part of this task, four of the eight arms were accessible and held objects at their end. Participants were instructed to retrieve all the objects and to learn which arms they visited so that they could avoid them in part two of the task. In the second part, all arms became accessible and the walls surrounding the maze were lifted to conceal the surrounding landscape that was visible during part one. Participants were instructed to enter the four previously empty arms and avoid the four arms that previously held the objects. Participant strategy use was then determined by errors made in part two (i.e., the number of times the participant entered one of the arms that had previously held an object). In the Hex Maze, however, on the learning trials all arms are open, and participants must learn the location of one target platform at the end of an arm. On the subsequent probe trials, the landscape remains available for participants to use as a reference, and strategy use is determined by end location rather than errors made. Additionally, and in contrast to the RAM used by Hussain et al. (2016), the Hex Maze contained a proximal cue (the colored sphere) which allowed the use of the egocentric-cue strategy. Hence, the lack of consistency in results could be due to my participants having access to the use of an additional strategy and to differences in determining strategy use.

The results of those two previously published studies support the inference that the use of allocentric (rather than egocentric) navigational strategies is not necessarily associated

with fluctuating levels of estrogen but is instead associated with higher levels of progesterone. That is to say, women in the mid/late luteal phase, when progesterone levels are highest, prefer allocentric strategy use whereas women in the other phases, when progesterone levels are low, prefer egocentric strategy use. As Hussain et al. (2016) note, one neurobiological mechanism that supports this suggestion is that higher progesterone levels promote use of the hippocampus (the brain structure central to allocentric strategies) while depressing use of the caudate nucleus (the brain structure central to the egocentric-response strategy).

Stress and Navigational Strategy

Stress induction manipulation check. Exposure to the PASAT and *u*-PASAT manipulations had notably different effects on objective and subjective indices of stress. For participants in the Stress condition, both heart rate (HR) and STAI-State values increased from baseline to the immediate post-manipulation measurement point. For those in the Non-Stress condition, HR and diastolic BP values decreased over the same period while STAI-State values increased only slightly. These findings are, broadly speaking, consistent with those reported in previous studies that used the same laboratory-based stress induction protocol (see e.g., Mathias et al., 2004; Tanosoto et al., 2015; van Gerven et al., 2016).

Although the current findings replicate those of previous PASAT stress studies with regard to HR and self-reported levels of stress, there is a lack of consistency in BP results. In this study, participants in the Stress condition experienced, on average, decreases in both systolic and diastolic BP from baseline to the immediate post-stressor measurement point. These findings are inconsistent with prior PASAT stress research reporting significant increases in both systolic and diastolic BP from baseline to post-stressor measurement points (Mathias et al., 2004; van Gerven et al., 2016). In contrast to those studies, however, the current sample consisted of women only. This difference is relevant because prior research has shown that men and women react differently to different types of stressors (see, e.g., Kudielka, Hellhammer, & Wust, 2009; Uhart, Chong, Oswald, Lin, & Wand, 2006), with women appearing to have a larger physiological response to social rejection challenges than to achievement challenges such as those inherent in the PASAT (Stroud, Salovey, & Epel, 2002). Hence, the across-study difference in BP response could be due, in part, to the PASAT being a somewhat milder stressor to my female-only sample compared to previous studies that have analyzed the effectiveness of PASAT on male-only or mixed-sex samples. Moreover, this relatively mild stressor may have been relatively short-lived so that by the time the task was over (after approximately 12 minutes) and I took the immediate post-

stressor measurement, participant BP could have returned to its pre-stress level, and then decreased slightly as the task ended.

Researchers in the stress literature have, in recent years, become increasingly concerned about the control conditions that accompany their laboratory-based stress induction protocols (see, e.g., von Dawans, Kirschbaum, & Heinrichs, 2011). The best no-stress control conditions are ones that ensure participants complete tasks that formally match those encountered by stress-group participants, with the only difference being the presence of stress-inducing elements. This structure allows researchers to assume that any performance differences in outcome variables are actually a consequence of exposure to those stress-inducing elements and not due to, for instance, the difference in activities completed during the manipulation. So, for instance, whereas the original presentation of the TSST did not include a control condition, more recently-developed stress induction protocols such as a the Maastricht Acute Stress Test (MAST; Smeets et al., 2012) and Fear Factor Stress Test (FFST; du Plooy, Thomas, Henry, Human, & Jacobs, 2014) have been accompanied by carefully-designed control procedures that match the formal elements of stress induction (e.g., submerging hands in water; public speaking) but lack its stress-inducing components (e.g., submerging hands in warm water rather than cold water; reading aloud from a magazine while alone in a room rather than presenting a self-written speech in front of a panel while being video-recorded).

Hence, although we know that the *u*-PASAT (a) matches the formal and core task elements of the PASAT) but (b) does not include stress-inducing components, it is important to document that it is actually an adequate control task. The current results appear to provide such documentation. In the current sample, the *u*-PASAT had a significantly calming effect on HR and induced a significant decrease in diastolic BP, a non-significant decrease in systolic BP, and a small (but significant) increase in STAI-State scores. This latter increase might be attributed to the mathematical nature of the task; most participants in the Non-Stress condition noted in their debriefing session that they struggled with mathematics. To address this, the *u*-PASAT could be adjusted to require only the addition of numbers where the sum is equal to or less than 10. This will allow the task to still be mathematical in nature (and thus, still a matched control task) while being easier for participants who may feel anxiety adding larger numbers. Nonetheless, the magnitude of change in STAI-State scores from baseline to the immediate post-stressor measurement point were significantly lower in the Non-Stress condition than in the Stress condition. Taken together, this pattern of data suggests that the *u*-PASAT has effects that are non-stressful and distinct from those of the PASAT, and that

therefore it is an adequate control condition for that laboratory-based stress induction protocol.

Effects of stress on choice and use of navigational strategy. Hypothesis 2 stated that exposure to an acute psychosocial stressor will have a significant effect on choice and use of navigational strategy use such that exposed participants, but not those unexposed, will tend to use one of the egocentric strategies rather than an allocentric strategy. This hypothesis was not confirmed.

Analyses detected no significant between-condition (Stress versus Non-Stress) differences in either strategy use or overall strategy preference across the 10 Show-Me trials. This pattern of results held no matter whether navigational strategy was divided into three (egocentric-cue, egocentric-response, allocentric) or two (egocentric [response and cue, combined], allocentric) categories.

These findings suggesting no statistically significant effects of stress exposure on choice and use of navigational strategy are inconsistent with those presented in some previously published studies. For instance, Thomas et al. (2010) found that exposure to acute psychosocial stress impaired female allocentric, but not egocentric-cue, navigation. The age range of women in that study was similar to that of the women studied here.

Thomas et al. (2010) suggest that one reason why stress might impair map-based spatial strategies is that the physiological stress response has direct consequences for hippocampal function (Alderson & Novack, 2002; Kemeny, 2003; McEwen & Sapolsky, 1995). These consequences are due, in part, to the hippocampus containing many glucocorticoid receptors. When the organism experiences a stressor, glucocorticoids cross the blood-brain barrier and bind to these receptors. Occupation of these receptors is part of the signaling that implicates the hippocampus in ending stress responses via glucocorticoid-mediated negative feedback to the HPA axis. However, when stress is severe or unchecked these receptors are too heavily occupied and, consequently, spatial cognitive processing is impaired (Gjerstad, Lightman, & Spiga, 2018).

The current findings (and, as mentioned in Chapter 3, those of the pilot study) also stand in contrast to those of van Gerven et al. (2016). They found that stress biased their male and female participants toward using allocentric rather than egocentric strategies when completing a VR spatial navigation task. As mentioned earlier, van Gerven and colleagues' sample consisted of men as well as women using hormonal birth control and mood-stabilizing medications, which this study did not. Hence, the difference in results between the current study and that one might be at least partly attributable to differences in participants'

hormonal profiles. Specifically, as mentioned earlier there is research indicating that (a) men and women respond to psychosocial stress differently due to differing sex hormones, and (b) oral contraceptives and mood-stabilizing medications dampen the female physiological stress response (see, e.g., Espin et al., 2013; Kajantie & Phillips, 2006; Kirschbaum et al., 1999; Schloesser, Martinowich, & Manji, 2012). Thus, the impact of their stressor (the PASAT) on their sample could have been different to the impact of the same stressor on my sample, which consisted of women taking neither oral contraceptives nor mood-stabilizing medication. Another difference between this study and van Gerven et al. (2016) is that my methods included a 30-minute rest period whereas their study had a 10-minute rest period followed by 10 minutes of non-navigational cognitive tasks. Given that the SAM-axis response is typically shorter in duration, it is possible that the difference in results could be partly due to the differing time periods between the administration of the PASAT and the navigation task, resulting in potentially different stress response states during testing.

The current negative results are, however, consistent with those reported by Klopp et al. (2012). They found that TSST-induced psychosocial stress did not impair allocentric navigation performance on a standard virtual MWM task performed 30-minutes post stress induction. Specifically, their results indicated no significant between-group differences (TSST-exposed versus non-exposed participants) in terms of either path length or time taken to reach a previously learned target location.

One way to account for these inconsistencies in stress effects on navigational strategy is to consider the concept of the inverted-U relationship between the concentration of corticosteroids and numerous neurobiological endpoints (as described by the Yerkes-Dodson Law; Yerkes & Dodson, 1908). Sapolsky (2015) explains that the length and severity of stress exposure (and the subsequent release of cortisol) determines whether there are beneficial and stimulatory or detrimental and over-stimulatory effects on cognition, behaviour, and other biological outcomes. Specifically, stress exposure of mild-to-moderate severity and/or relatively shorter duration will lead to SAM-axis activation and consequent neural stimulation in brain regions (e.g., the hippocampus) where glucocorticoid receptors proliferate. In contrast, stress exposure that is more severe and/or of longer duration will lead to HPA-axis activation and consequent neural overstimulation and inhibitory effects on the same brain regions. This is the classic inverted-U curve.

One must also, however, consider that individual differences in stress responsiveness and vulnerability influence the width, height, and symmetry of each person's inverted-U curve. For instance, it is possible that some participants in laboratory-based stress

experiments are relatively less responsive to the acute psychosocial stressor, and that they thus experienced stimulatory, rather than inhibitory, effects of cortisol on the hippocampus. In the context of this study, that would mean a greater preference for and use of an allocentric strategy. In contrast, other participants may have been relatively more responsive to the PASAT and consequently they would have experienced inhibitory effects of cortisol and, therefore, a greater preference for and use of an egocentric strategy. The presence of such individual variability and consequent opposing effects might account for both (a) the current study's lack of significant findings with regard to the effect of stress on choice and use and navigational strategy, and (b) the inconsistency in findings between this study and others in this literature. Neither this study nor the studies mentioned above considered differences in cortisol responsivity post-stress induction or compared strategy use across low and high responders. Thus, it is difficult to truly understand the effect of stress on navigational strategy use and to compare findings across studies as we cannot be sure the participants match in terms of stress responses. Hence, as noted below, it is important future studies account (either by design or in data analyses) for individual differences in stress responsivity and vulnerability.

Menstrual Cycle Phase x Stress Exposure Interaction Effect on Choice and Use of Navigational Strategy

Hypothesis 3 stated that interactions between exposure to a laboratory-based acute psychosocial stressor and menstrual cycle phase would have a significant effect on navigational strategy use, particularly among participants in the mid/late luteal phase. I tested the specific prediction that stress-exposed women in that phase will tend to use an egocentric strategy, whereas unexposed women in that phase will tend to use an allocentric strategy. This hypothesis was not confirmed. Analyses detected no significant interaction effects on any of the outcome variables tested in the linear mixed-model analyses (viz., average number of times each strategy [i.e., egocentric-response, egocentric-cue, egocentric-combined, and allocentric] were used across the 10 Show-Me trials).

Close inspection of the descriptive statistics proved useful and informative, however. That inspection suggested that participants in Mid/Late Luteal+Non-Stress group not only used the allocentric strategy more heavily than participants in any of the other five study groups, but also showed a clear preference toward the allocentric strategy (i.e., they used it, on average, on more than half of the 10 Show-Me trials). Furthermore, the descriptive statistics showed that participants in the Mid/Late Luteal+Stress group used a combined-egocentric strategy (with both egocentric-response and egocentric-cue strategies being used a

similar number of times) more than the allocentric strategy. Thus, the direction of the observed means follows the predicted direction.

Of interest, naturally, are the neurobiological mechanisms that might drive the interaction effect under consideration. One such mechanism might involve estrogen. There is evidence suggesting that this hormone modulates the physiological effect of stress on women, depressing the magnitude of cortisol responses to stressors and thereby protecting the hippocampus from the deactivational effects of stress exposure (Albert et al., 2015; Foy, Baudry, Foy, & Thompson, 2008). Because estrogen levels are relatively low during the mid/late luteal phase, it is plausible that cortisol responsivity would be quite high during that time and that, consequently, the hippocampus would be more vulnerable to stressors than at other phases of the cycle (Kajantie & Phillips, 2006; Montero-Lopez et al., 2018). Hence, women who experience stress during the mid/late luteal phase may have to rely on extra-hippocampal mechanisms (and, consequently, egocentric-response and/or egocentric-cue strategies) to solve spatial navigational problems.

Limitations and Directions for Future Research

Inferences that might be drawn from this study are constrained by some important methodological limitations. Fortunately, many of these can be addressed quite easily by future research.

First, the study may have been statistically underpowered to detect the effects under consideration. Although a sample size of 60 is slightly higher than some prior research into the effects of either menstrual cycle phase or stress on navigational strategy (see, e.g., Hussain et al., 2016; Klopp et al., 2012), a study seeking to detect an interaction effect and therefore using a larger number of groups should aim to recruit at least 120 participants. Within that larger group of participants should be adequate and, ideally, equal numbers of women within each menstrual cycle phase group. Unfortunately, the time and resource constraints typical of a Master's thesis did not allow me to meet these requirements.

A second limitation is that participants were not all tested at the same time of day (testing took place in a single 90-min session, with the earliest beginning at 9am and the latest at 4pm; test sessions were scheduled at the participant's convenience). This is a limitation because time-of-day effects have been reported in stress research – specifically, because baseline cortisol levels are higher in the morning than in the afternoon, it is more likely one will see greater cortisol responsivity later in the day (see, e.g., Liening, Stanton, Saini, & Schultheiss, 2010). Hence, differences in participant performance might have been further influenced by this time-of-day effect. The relatively small sample size alongside the

relatively large number of study groups, as well as the fact that I did not collect cortisol measures, meant I could not assess the significance of that effect. Future research should aim to test all participants at the same time of day (preferably in the afternoon) so as to avoid these questions.

A third limitation is that no endocrine indices of stress were taken. Doing so would have allowed analyses to control for the individual differences in stress responsiveness (e.g., to compare effects on navigational strategy in women with larger versus smaller magnitudes of cortisol elevations). Thus, future investigations of the interaction effect under consideration should, for instance, collect measures of salivary cortisol and alpha amylase to assess both arms of the physiological stress response (that centered on the sympathetic nervous system, which was measured here via HR and BP but that can be effectively indexed by alpha amylase, and that centered on the HPA axis, which is typically measured via cortisol).

Additionally, and to further contribute to literature on the effect of menstrual cycle phase on navigational strategy choice and use, future studies could build on the methods used here by taking direct sex hormone measurements and then comparing navigational strategy use across the different phases. This design will allow additional insight into the hormonal conditions encouraging allocentric navigational strategy use, and could thereby further investigate the suggestion that allocentric navigation is associated with progesterone rather than estrogen.

Furthermore, future research may wish to further explore the differences in egocentric navigation across the two distinct strategies (i.e., egocentric-response vs. egocentric cue). Although the current study did not directly investigate these differences, trends in the results showing that the egocentric strategies were not always favored equally suggest this may be an interesting topic for future exploration.

Summary and Conclusion

This study adds to the growing body of literature on the effects of stress and of menstrual cycle phase on spatial cognition, and specifically, spatial navigational strategies. My specific aims were to (a) systematically replicate previous investigations into the main effects of menstrual cycle phase and stress on females' choice and use of navigational strategy, and (b) launch a novel investigation into the possible interactional effect of those variables on navigation. My results revealed a non-significant main effect for menstrual cycle phase, which is inconsistent with prior research. Analyses also did not detect a significant main effect of stress on navigational strategy use, a result consistent with the findings of

some previous studies but not others. This negative result may be accounted for, at least in part, by individual differences in stress responsiveness and vulnerability leading to different neurological reactions to the PASAT. Finally, the analyses detected a non-significant interaction effect. Some trends in cell means were, however, in the predicted direction; this encourages further, larger-scale, exploration of this research question.

In conclusion, this study adds real-life value to the body of literature examining internal and external effects on the strategies women use to navigate their environments. Proceeding from the assumption that sex hormones and stress hormones do not act independently of each other in natural settings and under natural conditions, I designed a study that, although it produced no statistically significant results, might serve as a foundation upon which to build larger-scale research projects testing hypotheses about how the interaction of menstrual cycle phase and stress exposure might drive choice and use of navigational strategy.

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Appendix A – Pilot Study Screening Questionnaire

Instructions: Please circle your answer from the given choices or write it in the space provided.

Basic Demographic Information

How old are you? _____

Is this your first undergraduate degree? | Yes | | No |

Which education year have you most recently completed?

Matric / Grade 12	1st Year	2nd Year	3rd Year

Other (please specify): _____

Sleep and Exercise

On average, how many hours do you sleep per night?

Less than 4 hours	4-6 hours	7-8 hours	9-10 hours	11+ hours

How many hours did you sleep last night?

_____

On average, how many hours do you exercise per week?

Less than 1 hour	1-2 hours	3-4 hours	5-6 hours	7-8 hours

Other (please specify): _____

Did you exercise in the last 24 hours? If so, for how long?

_____

Alcohol, Smoking & Other Substances

On average, how many alcoholic drinks do you consume per week?

0-2	3-5	6-8	9-11	12-14	More than 14

Did you drink alcohol in the last 24 hours? If so, how many drinks?

_____

Do you smoke cigarettes? Yes No

On average, how many do you smoke per week?

Less than 10 | 10-20 | 20-40 | 40- 60 | 60-80 | 80-100

Other (please specify): _____

How many have you smoked within the last 24 hours?

_____

Do you have a history of substance addiction? Yes No

Are you currently addicted to any substance (other than nicotine)? Yes No

Psychological & Neurological Factors

Are you currently experiencing any psychological disorder (such as depression or anxiety)?

Yes No

Are you currently on any mood/anxiety stabilizing medications? Yes No

Do you have a history of neurological illness (e.g., epilepsy, traumatic brain injury)?

Yes No

Birth Control, Pregnancy and Breastfeeding

Are you currently using any hormonal birth control methods? (i.e. the pill, the implant, injections, IUD, contraceptive patch, vaginal ring) Yes No

If no, but have used previously, when did you stop using said birth control method(s)?

_____

Have you used any emergency contraceptive medication within the last 3 months? (e.g., Plan B) Yes No

Have you been pregnant within the last two years (i.e., since January 2016)? Yes No

Are you currently breastfeeding? Yes No

Menstrual Cycle (the most NB section! Please take your time and answer as accurately as possible)

At what age did you have your first period? _____

On average, how many days is your menstrual cycle (from the day of menses onset to the day prior to next menses onset)?

Less than 20 days	20-24 days	25-34 days	More than 34 days

On average, how long do your periods last (from day of menses onset to day bleeding stops)?

Less than 3 days	3-5 days	6-8 days	More than 8 days

On what date did your second most recent period start? _____

On what date did your most recent period start? _____

On what day of your cycle are you on today? _____

Do you have any hormonal disorders (e.g., endometriosis or polycystic ovarian syndrome)?

Yes	No

Appendix B - SBSOD

This questionnaire consists of several statements about your spatial and navigational abilities, preferences, and experiences.

After each statement, you should circle a number to indicate your level of agreement with the statement.

Circle "1" if you strongly agree that the statement applies to you. Circle "7" if you strongly disagree, or some number in-between if your agreement is intermediate. Circle "4" if you neither agree nor disagree.

1. I am very good at giving directions.

strongly agree 1 2 3 4 5 6 7 strongly disagree

2. I have a poor memory for where I left things.

strongly agree 1 2 3 4 5 6 7 strongly disagree

3. I am very good at judging distances.

strongly agree 1 2 3 4 5 6 7 strongly disagree

4. My "sense of direction" is very good.

strongly agree 1 2 3 4 5 6 7 strongly disagree

5. I tend to think of my environment in terms of cardinal directions (N, S, E, W).

strongly agree 1 2 3 4 5 6 7 strongly disagree

6. I very easily get lost in a new city.

strongly agree 1 2 3 4 5 6 7 strongly disagree

7. I enjoy reading maps.

strongly agree 1 2 3 4 5 6 7 strongly disagree

8. I have trouble understanding directions.

strongly agree 1 2 3 4 5 6 7 strongly disagree

9. I am very good at reading maps.

strongly agree 1 2 3 4 5 6 7 strongly disagree

10. I don't remember routes very well while riding as a passenger in a car.

strongly agree 1 2 3 4 5 6 7 strongly disagree

11. I don't enjoy giving directions.

strongly agree 1 2 3 4 5 6 7 strongly disagree

12. It's not important to me to know where I am.

strongly agree 1 2 3 4 5 6 7 strongly disagree

13. I usually let someone else do the navigational planning for long trips.

strongly agree 1 2 3 4 5 6 7 strongly disagree

14. I can usually remember a new route after I have travelled it only once.

strongly agree 1 2 3 4 5 6 7 strongly disagree

15. I don't have a very good "mental map" of my environment.

strongly agree 1 2 3 4 5 6 7 strongly disagree

Appendix C – STAI

DIRECTIONS:

A number of statements which people have used to describe themselves are given below. Read each statement and then circle the appropriate number to the right of the statement to indicate how you feel *right* now, that is, *at this moment*. There are no right or wrong answers. Do not spend too much time on any one statement but give the answer which seems to describe your present feelings best.

	NOT AT ALL	SOMEWHAT	MODERATELY SO	VERY MUCH SO
1. I feel calm	1	2	3	4
2. I feel secure	1	2	3	4
3. I am tense	1	2	3	4
4. I feel strained	1	2	3	4
5. I feel at ease	1	2	3	4
6. I feel upset	1	2	3	4
7. I am presently worrying over possible misfortunes	1	2	3	4
8. I feel satisfied	1	2	3	4
9. I feel frightened	1	2	3	4
10. I feel comfortable	1	2	3	4
11. I feel self-confident	1	2	3	4
12. I feel nervous	1	2	3	4
13. I am jittery	1	2	3	4
14. I feel indecisive	1	2	3	4
15. I am relaxed	1	2	3	4
16. I feel content	1	2	3	4
17. I am worried	1	2	3	4
18. I feel confused	1	2	3	4
19. I feel steady	1	2	3	4
20. I feel pleasant	1	2	3	4

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STAIP-AD Test Form Y
www.mindgarden.com

DIRECTIONS

A number of statements which people have used to describe themselves are given below. Read each statement and then circle the appropriate number to the right of the statement to indicate how you *generally* feel. There are no right or wrong answers. Do not spend too much time on any one statement but give the answer which seems to describe how you generally feel.

	ALMOST NEVER	SOMETIMES	OFTEN	ALMOST ALWAYS
21. I feel pleasant	1	2	3	4
22. I feel nervous and restless	1	2	3	4
23. I feel satisfied with myself	1	2	3	4
24. I wish I could be as happy as others seem to be	1	2	3	4
25. I feel like a failure	1	2	3	4
26. I feel rested	1	2	3	4
27. I am "calm, cool, and collected"	1	2	3	4
28. I feel that difficulties are piling up so that I cannot overcome them	1	2	3	4
29. I worry too much over something that really doesn't matter	1	2	3	4
30. I am happy	1	2	3	4
31. I have disturbing thoughts	1	2	3	4
32. I lack self-confidence	1	2	3	4
33. I feel secure	1	2	3	4
34. I make decisions easily	1	2	3	4
35. I feel inadequate	1	2	3	4
36. I am content	1	2	3	4
37. Some unimportant thought runs through my mind and bothers me	1	2	3	4
38. I take disappointments so keenly that I can't put them out of my mind	1	2	3	4
39. I am a steady person	1	2	3	4
40. I get in a state of tension or turmoil as I think over my recent concerns and interests	1	2	3	4

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Appendix D – Post CG Arena Questionnaire

Instructions: Please circle your answer from the given choices or write it in the space provided.

Did you know where the target was? Yes No

If yes, where was the target?

_____

_____

How did you find the target?

_____

_____

_____

_____

Did you use any environmental cues to help you find the target? Yes No

If yes, which cues?

_____

_____

_____

Did you follow the same general direction in each trial? Yes No

If yes, in which direction did you go?

_____

Did you follow the same path to reach the target in each trial? Yes No

If so, please describe the path you took.

_____

_____

_____

_____

_____

Appendix E – Video Game Experience Questionnaire

Instructions: Please circle your answer from the given choices or write it in the space provided.

1) Have you ever played video games? Yes No

2) Do you currently play video games? Yes No

If you answered 'Yes' to question (2), please answer the following questions.

3) How long have you been playing video games?

1-6 months | 6-12 months | 1-2 years | 3-5 years | 5-10 years | More than 10 years

4) How often do you currently play video games?

Daily | Weekly | Once a month | Once in 6 months | Once a year | Less than once a year

5) How good do you feel you are at playing video games?

Very good | Good | Not very skilled | No skill

Appendix F – Consent Form

University of Cape Town

Effects of Menstrual Cycle on Spatial Navigational Strategies.

Purpose

I am a Psychology student investigating the effect of the menstrual cycle on female spatial cognition. The purpose of this study is to determine whether menstrual cycle phase affects the use of navigational strategies in women.

Procedure

If you decide to participate in this study, you will first be asked to complete a screening questionnaire. This questionnaire will ensure you meet the eligibility criteria of the study. Should you meet the criteria, you will be enrolled into the full study and asked to complete a self-report spatial ability questionnaire. You will then have to complete a computerized cognition task, and then rest for 30 minutes. Following this, you will be asked to complete a virtual arena task on the computer, and then a post-arena questionnaire. After this, you will be debriefed. Throughout the experimental procedure, measurements of your heart rate and blood pressure will be taken and you will be asked to complete a self-report anxiety questionnaire (the State-Trait Anxiety Inventory). The whole study, including the screening questionnaire, should take approximately 1h30mins.

Possible Risks

There are no foreseeable long-term risks that you may incur.

Possible Benefits

SRPP participant: If you complete the screening questionnaire, you will receive 1 SRPP point. If you complete the full study, you will receive another 2 SRPP points. Furthermore, you will gain insight into psychological research by being a research participant.

DSA participant: After completing the full study, you will be entered into a raffle to win one of four cavendish vouchers, valued at: R750; R500; R300; R250.

Voluntary Participation

Participation in this study is <u>completely voluntary</u> and you have the right to withdraw participation at any time of the study without facing any consequences. No reasons need to be given should you decide to withdraw participation. However, if you withdraw from the study, you will not receive the 2 full-study participation SRPP points / you will not be entered into the raffle.

Confidentiality

Any information about you obtained through this study will remain completely confidential. All data collected with remain in a locked cabinet and/or on a password-locked computer. Only myself, the principal researcher, and my supervisor will have access to the data collected. Additionally, all data collected will not have any identifying factors that can link it back to you.

The various data measurements will be linked by the same number label however this number will not be linked back to any documents containing your name or other identifying details. This consent form will be the only document on which your name appears. The raw data collected in this study will not be published or released to the public at any stage in time.

Questions

If you have any study-related questions, problems or emergencies you can contact me on:

Alexandra McHale menstrualcyclestress@gmail.com

If you have questions about your rights as a study participant, or any comments or complaints about the study, please contact:
Rosalind Adams at the UCT Department of Psychology.
Phone:021 650 3417
Email: rosalind.adams@uct.ac.za.

I have read the above and am satisfied with my understanding of the study and its possible benefits and risks. My questions about the study have been answered. I hereby voluntarily consent to participation in the research study as described.

_____
Name of Participant

_____ _____
Signature of Participant Date

Appendix G – Debrief Form

Effects of Menstrual Cycle phase and Stress on Spatial Navigational Strategies:

Debriefing Form

Thank you for participating in the research study.

This form provides you with information about the study in which you have just participated, and explains in full the methods of collection of data for this research study. The Principle Investigator (the person in charge of this research) will also explain this study to you in full and answer your questions.

Title of Research Study

Effects of Menstrual Cycle phase and Stress on Spatial Navigational Strategies

Principal Investigator and other contact details

Alexandra McHale
Department of Psychology
University of Cape Town

UCT Student Wellness Service
021 650 1017

Kevin G. F. Thomas, Ph.D.
Supervisor
Department of Psychology
University of Cape Town
021 650 4608

What is the purpose of this research study?

The purpose of this research study is to investigate the effects, and possible interacting effect, of menstrual cycle phase and stress on women's navigational strategy use.

What was done during this research study?

During this study, you were required to complete a screening questionnaire, you were then secretly randomly assigned to either the Stress or Non-Stress condition, you completed either the PASAT or U-PASAT based on this assignment, you completed a virtual maze task, and you completed a post-virtual-maze questionnaire. Throughout the experimental period, your heart rate and blood pressure were measured 5 times, and you were required to complete the STAI 4 times.

Was there any deception used in this research study?

There was deception used in the study in that you were not informed of the stress induction task. However, this task only induces a very acute amount of stress that usually subsides within an hour of the task, and it is no more than you would experience on a daily basis. Thus, there are no long-term negative effects from this task, nor the deception.

Is there anything further required of you?

Please do not disclose anything that happened during these research sessions to anyone else, as this may bias future participants and their performance. If you are still feeling stressed after the completion of the study session, please inform us so that we can provide you with the contact details of a clinical psychologist who can provide you with post-session counselling. Additionally, please email me on the day you start your next period so that I can confirm your initial group placement.

Signatures

I have been informed, in detail, about this study's purpose, procedures, and deceptions. I have been given the opportunity to ask questions before I sign. By signing this form, I am not waiving any of my legal rights.

_____ _____

Participant Signature Authorising Date

Appendix H - SRPP Announcement

Dear Students,

I am running a study that tests female spatial navigation across menstrual cycle phases. I am offering **3 SRPP** points for your participation (*1 SRPP* for completing the screening questionnaire [done in the testing session]; *2 SRPP* points for completion of full study).

To participate in the study, you need to meet the following criteria:

- Female aged between 18-30
- Have no history of neurological illness (i.e. epilepsy or traumatic brain injury) or substance abuse (i.e. taking illegal substances or abusing alcohol/prescription medication)
- Not currently taking, or have taken within the last three months, any hormonal contraceptive medication (including but not limited to the birth control pill, the IUD, the injection, the implant, the vaginal ring, the contraceptive patch, Plan B).
- Have no currently mood/anxiety disorder diagnosis, and/or currently taking mood/anxiety stabilizing medication.
- Have not been pregnant within the last two years and are not currently breastfeeding.
- Participants will need to be in certain phases of their menstrual cycle *on the day of testing* (specifically, you will need to be on any of the following days of your cycle: **1-7, 13-17, or 20-end of cycle**). Please keep this in mind when arranging a testing time. Feel free to email me if you need help determining what day of your cycle you are currently on, and subsequently which days you can be tested on.

The study will take approximately 1h30mins of your time and will take place in one session. You will first have to complete a screening questionnaire, and should you meet the eligibility criteria you will be enrolled into the full study. Completing the questionnaire **does not** guarantee you a place in the full study.

If you are interested in participating in this study, please sign up, with your *STUDENT NUMBER*, for a testing session that suits you best here: **SIGN UP (https://doodle.com/poll/ytxihn4ddf9gcg56)**

If you have any further questions, please send me an email at **menstrualcyclestress@gmail.com**.

Regards,

Alexandra McHale

Appendix I - DSA invitation

Dear Students,

My name is Alexandra McHale and I am a Master's student in the Psychology Department. I am running a study that tests female cognition across menstrual cycle phases. Testing sessions will take place from **9 – 17 May 2019**. If you participate in my study, you will be entered into a raffle for a chance to win one of four Cavendish gift-card prizes valued at: R750, R500, R300, R250 respectively.

To participate in the study, you ***need*** to meet the following criteria:

- Female aged between **18-30**
- Participants need to be on *certain days* of their cycle, specifically **days 13-17**, on the day of testing. To work out what day of your cycle you are currently on, take the first day of your most recent period as day 1.
- Each participant should have **25-34 days** in between the start of each period (i.e. a full cycle of 25-34 days long).
- Have **no history** of **neurological illness** (i.e. epilepsy or traumatic brain injury) or **substance abuse** (i.e. taking illegal substances or abusing alcohol/prescription medication)
- **Not currently taking**, or have taken within the last three months, any **hormonal contraceptive medication** (including but not limited to the birth control pill, the IUD, the injection, the implant, the vaginal ring, the contraceptive patch, Plan B).
- Have **no current mood/anxiety disorder diagnosis**, and/or **currently taking mood/anxiety stabilizing medication**.
- Have **not been pregnant** within the *last two years* and are **not currently breastfeeding**.

The study will take *approximately 1h30 minutes* of your time and will take place in one session. You will first have to complete a screening questionnaire, and should you meet the eligibility criteria you will be enrolled into the full study. Completing the questionnaire **does not** guarantee you a place in the full study.

You are invited to participate in this research and your participation is voluntary. All information will be treated in a confidential manner and used exclusively for the purpose of this study. No individual names will be recorded or published. You will not be requested to supply any identifiable information, ensuring anonymity of your responses. You can choose

to withdraw from the research at any time for whatever reason, in accordance with ethical research requirements.

If you are interested in participating in this study or if you have any further questions, please send me an email at **menstrualcyclestress@gmail.com.**

Regards,

Alexandra McHale
Principal Researcher
menstrualcyclestress@gmail.com

Appendix J – Main Study Screening Questionnaire

Instructions: Please circle your answer from the given choices or write it in the space provided.

Basic Demographic Information

How old are you? _____

Is this your first undergraduate degree? | Yes | | No |

Which education year have you most recently completed?

Matric / Grade 12	1st Year	2nd Year	3rd Year

Other (please specify): _____

Sleep and Exercise

On average, over the last month, how many hours do you sleep per night?

Less than 4 hours	4-6 hours	7-8 hours	9-10 hours	11+ hours

How many hours did you sleep last night?

_____

On average, over the last month, how many hours do you exercise per week?

Less than 1 hour	1-2 hours	3-4 hours	5-6 hours	7-8 hours

Other (please specify): _____

Did you exercise in the last 24 hours? If so, for how long?

_____

Alcohol, Smoking & Other Substances

On average, over the last month, how many alcoholic drinks do you consume per week?

0	1-2	3-5	6-8	9-11	12-14	More than 14

Did you drink alcohol in the last 24 hours? If so, how many drinks?

_____

Do you smoke cigarettes? Yes No

On average, over the last month, how many cigarettes did you smoke per week?

0 | Less than 10 | 10-20 | 20-40 | 40- 60 | 60-80 | 80-100

Other (please specify): _____

How many have you smoked within the last 24 hours?

_____

Do you have a history of substance addiction? Yes No

Are you currently addicted to any substance (other than nicotine)? Yes No

Psychological & Neurological Factors

Are you currently experiencing any psychological disorder (such as depression or anxiety)?

Yes No

Are you currently on any mood/anxiety stabilizing medications? Yes No

Do you have a history of neurological illness (e.g., epilepsy, traumatic brain injury)?

Yes No

Birth Control, Pregnancy and Breastfeeding

Are you currently using any hormonal birth control methods? (i.e. the pill, the implant, injections, IUD, contraceptive patch, vaginal ring) Yes No

If no, but have used previously, when did you stop using said birth control method(s)?

_____

Have you used any emergency contraceptive medication within the last 3 months? (e.g., Plan B) Yes No

Have you been pregnant within the last two years (i.e., since January 2016)? Yes No

Are you currently breastfeeding? Yes No

Menstrual Cycle (the most NB section! Please take your time and answer as accurately as possible)

At what age did you have your first period? _____

On average, how many days is your menstrual cycle (from the day of menses onset to the day prior to next menses onset)?

Less than 20 days	20-24 days	25-34 days	More than 34 days

On average, how long do your periods last (from day of menses onset to day bleeding stops)?

_____

On what date did your second most recent period start? _____

On what date did your most recent period start? _____

On what day of your cycle are you on today? _____

Do you have any hormonal disorders (e.g., endometriosis or polycystic ovarian syndrome)?

Yes	No

Appendix K – PSS

The questions in this scale ask you about your feelings and thoughts during the last month. In each case, you will be asked to indicate by circling *how often* you felt or thought a certain way.

0 = Never 1 = Almost Never 2 = Sometimes 3 = Fairly Often 4 = Very Often

1. In the last month, how often have you been upset because of something that happened unexpectedly?	0	1	2	3	4
2. In the last month, how often have you felt that you were unable to control the important things in your life?	0	1	2	3	4
3. In the last month, how often have you felt nervous and "stressed"?	0	1	2	3	4
4. In the last month, how often have you felt confident about your ability to handle your personal problems?	0	1	2	3	4
5. In the last month, how often have you felt that things were going your way?	0	1	2	3	4
6. In the last month, how often have you found that you could not cope with all the things that you had to do?	0	1	2	3	4
7. In the last month, how often have you been able to control irritations in your life?	0	1	2	3	4
8. In the last month, how often have you felt that you were on top of things?	0	1	2	3	4
9. In the last month, how often have you been angered because of things that were outside of your control?	0	1	2	3	4
10. In the last month, how often have you felt difficulties were piling up so high that you could not overcome them?	0	1	2	3	4

Appendix L – Post Hex Maze Questionnaire

Instructions: Please select your answer from the given choices or write it in the space provided.

The following questions are specifically about the *first environment* (i.e., the first 10 trials).

1. Did you know where the target was? Yes | No

2. If yes, where was the target?

_____

_____

_____

3. How did you find the target in the "Find-It" trials?

_____

_____

_____

4. Did you use any cues to help you find the target location on the "Show-Me" trials?

Yes | No

5. If yes, which cues did you use? Windows | Dark Grey Sphere

Or, other (please specify): _____

6. Did you follow the same directions in the "Show-Me" trials as in the "Find-It" trials to reach the trial location? Yes | No

7. If yes, which directions did you follow?

_____

_____

_____

The following questions are specifically about the *second environment* (i.e., the last 10 trials).

8. Did you know where the target was? Yes No

9. If yes, where was the target?

_____

_____

_____

10. How did you find the target in the "Find-It" trials?

_____

_____

_____

11. Did you use any cues to help you find the target location on the "Show-Me" trials?

Yes No

12. If yes, which cues did you use? Windows Blue Sphere

Or, other (please specify): _____

13. Did you follow the same directions in the "Show-Me" trials as in the "Find-It" trials to reach the trial location? Yes No

14. If yes, which directions did you follow?

_____

_____

_____