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Effects of a Postpartum Sleep Schedule on Childless Women's Sleep, Circadian Amplitude, Daytime Sleepiness, Performance, and Mood

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Effects of a Postpartum Sleep Schedule on Childless Women's Sleep, Circadian

Amplitude, Daytime Sleepiness, Performance, and Mood

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ABSTRACT

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OBJECTIVES: A complex process of physiological and environmental changes during the postpartum period confounds our understanding of the discrete impacts of postpartum sleep fragmentation. The aim of this dissertation was to isolate the effects of postpartum sleep fragmentation by manipulating the sleep of childless women in the laboratory to model a postpartum sleep fragmentation schedule. Actigraphically and polysomnographically-recorded sleep, daytime functioning, mood, and melatonin amplitude were quantified.

METHODS: Eleven healthy, childless women (25.4 [SD±2.3] years, 72.7% white, $23,000 [SD±$11,000] household income) contributed continuous wrist actigraphy and daily psychomotor vigilance test (PVT) for one baseline week followed by 3 consecutive nights of overnight polysomnography: an adjustment/sleep disorder screening night, a baseline night, and a night of experimental sleep fragmentation during which they were awakened 3 times for 30-35mins each. During nocturnal awakenings, women engaged in a standardized protocol that included feeding, changing, and rocking a doll in dim light (<3 lux) to model postpartum motor activity and postures. First-morning baseline and fragmentation night voids were collected for 6-sulphatoxymelatonin assays to estimate circadian rhythm amplitude. Baseline and post-fragmentation Multiple Sleep Latency Tests (MSLT) and Profile of Mood States Surveys (POMS) were administered. A final week of at-home actigraphy monitoring, daily PVTs, and POMS captured recovery.

RESULTS: Sleep time did not change between baseline (M=461±28min) and sleep fragmentation nights (M=448±34min; p=.17), while sleep efficiency decreased (M=90.9%±6.1%; M=74.4%±3.9%, respectively; p<.001). Frequency of PVT lapses increased significantly from baseline (M=1.62±1.83) to the week after fragmentation (M=2.72±1.76; p=.01). Mood disturbance increased from baseline (M=-1.00±7.10) to after sleep fragmentation (M=8.55±12.9; p=.037). MSLT scores increased from baseline (M=13.1±3.54) to after sleep fragmentation (M=16.3±3.51; p=.02), indicating decreased daytime sleepiness. No changes in time spent in nocturnal sleep stages, 6-sulphatoxymelatonin concentration and actigraphy-defined total sleep time were found after sleep fragmentation compared to baseline.

CONCLUSIONS: The current study is the first to experimentally examine the effects of a simulated postpartum sleep disturbance schedule on aspects of physiology and behavior. Results suggest no changes in measured physiological components of a single night of simulated postpartum sleep fragmentation, but significant deficits in mood and neurobehavioral performance. Disruption of sleep continuity in the absence of measured physiological changes may be sufficient to cause poorer mood and performance.

Legend

AANAT = arylkylamine *N*-acetyltransferase

AASM = American Academy of Sleep Medicine

ANOVA = Analysis of Variance

aMT6 = 6-sulphatoxymelatonin

EDS = Excessive Daytime Sleepiness

ELISA = Enzyme-linked Immunosorbent Assay

M-E = Morningness-Eveningness

MSLT **=** Multiple Sleep Latency Test

PLMA = Periodic Limb Movements with associated Arousals

PLMS = Periodic Limb Movements in Sleep

PMDD = Premenstrual Dysphoric Disorder

POMS = Profile of Mood States Survey

PSG = Polysomnography

PVT = Psychomotor Vigilance Test

TMD = Total Mood Disturbance

TNF-α = Tumor Necrosis Factor Alpha

TST = Total Sleep Time

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Effects of a Postpartum Sleep Schedule on Childless Women's Sleep, Circadian Amplitude, Daytime Sleepiness, Performance, and Mood

Introduction

The maternal postpartum period is characterized by sleep fragmentation, which is associated with daytime impairment, mental health disturbances, and changes in melatonin levels. While sleep fragmentation can cause each of these outcomes, new mothers vary in the severity to which they experience these outcomes. These individual differences are likely a result of a complex process of individual physiological and environmental differences, so the relative contribution of sleep fragmentation to each of these outcomes is unknown. The purpose of this study was to understand the basic impact of sleep fragmentation, common during the maternal postpartum period, on sleep architecture, nocturnal melatonin, mood, daytime sleepiness, performance, and recovery. This was done by applying a maternal postpartum sleep schedule to healthy childless women, and modeling postpartum nocturnal activities such as light exposure, posture, and movement. The innovative component of the study that is expected to contribute to the advancement of our understanding and interventions was studying childless adult women in the sleep laboratory. This allowed for isolation of the specific effects of a single night of sleep fragmentation from postpartum physiological and environmental changes, while controlling for nocturnal activities, light levels, and postural changes; such stringent control is difficult to logistically and ethically use among a true postpartum population.

The study protocol (diagrammed in *Figure 1* below for a single participant) was used to monitor each of 11 childless women's sleep and performance at home for one week before she came into the sleep laboratory for an assessment of daytime

sleepiness, followed by three consecutive nights in the laboratory. The first night served as both an adjustment night to the sleep laboratory and as a screening for sleep disorders; the second night was used as a measure of baseline sleep; the third night was the experimental sleep fragmentation night. After the baseline and experimental sleep fragmentation nights, morning urine samples were collected to estimate nocturnal melatonin levels. Another assessment of daytime sleepiness occurred the day proceeding the experimental sleep fragmentation night, followed by a week of at-home monitored sleep and daily performance assessments and every-other-day mood surveys to quantify recovery.

Figure 1. Study protocol for a single participant

*Baseline Assessments
^Lab adjustment and sleep disorder screening night for participant (data not used)
Actigraphy=At home sleep with fixed bed times (10pm-12pm) and rise times (6am-8am)
PVT=Psychomotor Vigilance Test (performance assessment)
MSLT=Multiple Sleep Latency Test (in lab daytime sleepiness assessment)
POMS=Profile of Mood States survey
S=In lab sleep periods recorded using polysomnography
M=Urine 6-sulphatoxymelatonin samples (overnight & first morning void)

To provide a context for the current study, the introduction is split into two major sections. The first section provides a background on normative sleep and maternal postpartum sleep fragmentation. The second section focuses on the effects of sleep disturbance on the circadian system of the sleep-wake cycle, sleep architecture, daytime sleepiness, mood, and recovery.

Sleep and Postpartum Sleep Disturbances

Normal Sleep

Sleep occurs in cycles, each cycle lasting about 70-120 minutes (Kryger, Roth, & Dement, 2011). The human sleep cycle begins with three stages of non-Rapid Eye Movement (NREM) sleep (stages N1, N2, and N3), then transitions into REM sleep. If the sleep cycle is interrupted at any point, it must start from the beginning. While a thorough understanding of the function of these components remains largely unknown, particular attention and importance has been placed on REM sleep and stage N3 sleep, the deepest stage of NREM sleep characterized by delta EEG waves. There is support for a role of stage N3 in the restorative aspect of sleep, as specific disruption to stage N3 leads to increased daytime sleep propensity (Dijk, Groeger, Stanley, & Deacon, 2010). REM sleep likely plays a role in cognitive functioning, especially the consolidation of new memories (for review see McCoy & Strecker, 2011).

Sleep Fragmentation

Sleep fragmentation is a type of sleep disturbance in which there is a disruption in sleep continuity. This differs from sleep deprivation (a reduction in total sleep time) in that the quality of sleep is compromised, not the quantity. Existing research indicates

both sleep deprivation and sleep fragmentation yield similar adverse health consequences. Experimentally induced sleep fragmentation among healthy adults causes poorer mood (Bonnet, 1985), impaired daytime functioning (Bonnet, 1985; Durmer & Dinges, 2005; Zaharna & Guilleminault, 2010), hormonal alterations (Zaharna & Guilleminault, 2010), and daytime fatigue (Zaharna & Guilleminault, 2010) comparable to the impact of sleep deprivation. Moreover, as sleep deprivation alters the concentration of nocturnal melatonin (a biomarker of the central circadian clock) (Cajochen, Jewett, & Dijk, 2003; Kavčič et al, 2011), sleep fragmentation may also disrupt the concentration of melatonin. These specific effects of sleep disturbances will be presented in more detail in the following sections. Yet the protocol of experimental sleep fragmentation used in these studies differs from the fragmentation among postpartum mothers, who experience much longer periods of consolidated sleep, but also longer periods of wakefulness. The effects of postpartum sleep fragmentation are not fully understood.

Postpartum Sleep Disturbances

Postpartum mothers experience sleep disturbances, characterized by sleep fragmentation, during the early postpartum period (Gay, Lee, & Lee, 2004; Hunter, Pychnovsky, & Yount, 2009; Montgomery-Downs, Insana, Clegg-Kraynok, & Mancini, 2010). At postpartum week 2, new mothers obtain approximately 7.2 hours of total sleep time, despite spending about 9 hours in bed (Montgomery-Downs et al., 2010). Thus, they experience reduced sleep efficiency and increases in wake after sleep onset (Gay et al., 2004; Matsumoto, Shinkoda, Kang, Seo, 2003).

Mechanisms for Postpartum Sleep Disturbances

Infant Sleep

Postpartum maternal sleep is interrupted by frequent infant nocturnal awakenings (Santiago, Nolledo, Kinzler, & Santiago, 2001). Infants are not born with a sleep-wake rhythm that promotes one consolidated nocturnal sleep period. Instead they exhibit polyphasic sleep, whereby they have multiple sleep periods across a 24-hour period. A stable sleep-wake rhythm in infants develops over time, as sleep periods gradually lengthen so that most infants can "sleep through the night" about three months after birth (de Weerd & van den Bossche, 2003; Kleitman & Engelmann, 1953). These frequent infant nocturnal awakenings have implications for maternal sleep as there are strong correlations between maternal nocturnal awakenings and infant movements indicative of wake (Nishihara, Horiuchi, Eto, & Uchida, 2002; Wulff & Siegmund, 2000).

Postpartum Physiological Changes

New mothers experience physiological changes throughout pregnancy and the postpartum period that may be impacting their sleep and mental health above and beyond the nocturnal infant demands. Steroid hormones such as estrogen, progesterone, and cortisol increase during pregnancy and abruptly decrease after parturition (Bonnar, Franklin, Nott, & McNeilly, 1975; Tulchinsky, Hobel, Yeager, & Marshall, 1972; West & McNeilly, 1979); this abrupt transition has posited to be associated with mood disturbances (Bloch et al., 2000; Bloch et al., 2005; Doornbos et al., 2009). Estrogen and progesterone have sleep-promoting effects (Ross, Murray, & Steiner, 2005). Estrogen decreases sleep onset latency, nocturnal awakenings and

REM sleep, while increasing total sleep time. Progesterone is sedative, decreasing wakefulness and latency to NREM sleep as well as decreasing REM sleep. High cortisol levels are associated with stress and depressive symptoms (Manber & Armitage, 1999).

Mothers who choose to breastfeed may be further altering their sleep and their infant's sleep in unique ways from formula feeding mothers. Breastfeeding may have a soporific effect on infants as a result of the rise of 4 nucleotides present in breast milk that have circadian rhythms (Sánchez et al., 2009). Prolactin levels, which rise throughout pregnancy, return to pregravid levels in a few weeks among non-lactating women. However, breastfeeding induces the release of oxytocin, a hormone that stimulates pituitary lactotrophic cells and preserves high prolactin levels, which in turn is strongly associated with sleep (Sassin, Frantz, Weitzman, & Kapen, 1972). Therefore, hormonal changes postpartum may be further altering sleep and mental health, and perhaps their impact may even be protective of maternal sleep.

Physiological changes during the postpartum period that may impact sleep and mental health are not limited to hormonal changes. Most new mothers complain of at least one physical health symptom during the early postpartum period (Ansara, Cohen, Gallop, Kung, & Schei, 2005). The most commonly reported symptoms include general pain, headaches, fatigue, backaches, abdominal pain, bowel incontinence, and breast soreness (Cheng & Li, 2008; Webb et al., 2008). While there is a lack of literature specifically on postpartum physical pain and sleep, more than half of adults with chronic pain also experience sleep disturbances (Marty et al., 2008; McCracken & Iverson, 2002; Rohrbeck, Jordan, & Croft, 2007). Further, postpartum physical health conditions are associated with emotional well-being and depressive symptoms (Webb et al., 2008),

which is bidirectionally associated with sleep (Dørheim, Bondevik, Eberhard-Gran, & Bjorvatn, 2009; McCoy, 2011; Posmontier, 2008; Ross et al., 2005).

Postpartum Environmental Changes

The postpartum period is also characterized by adjustment to a new parenting role that includes caring for an infant and coping with the resultant emotional and social change (Brage Hudson, Elek, & Fleck, 2001). Child-care responsibilities and lack of knowledge related to parenting can be sources of frustration and fatigue for new mothers (Kanotra et al., 2007). Postpartum fatigue is indirectly associated with stress via depressive symptoms and sleep quality (Groër et al., 2005; Song, Change, Park, Kim, & Nam, 2010). New mothers may vary substantially in their adjustment to the postpartum period as a result of available social support. New mothers report social networks as their primary source of support (Leahy-Warren, 2007), and social support is a recognized buffer for stressful life events and predictor of emotional and physical well-being (Hung, 2004). Thus, the impact of the postpartum period on sleep and mental health is not limited to physiological changes, but also includes the environment.

Effects of Sleep Disturbance

Circadian Rhythm

An established model of sleep postulates that sleep and wake are regulated by two fundamental processes: a homeostatic sleep drive (Process S) and a circadian timing system (Process C) (Borbély, 1982). The homeostatic process regulates sleep propensity on the basis of prior wake time. The circadian system governs the timing of nearly all 24-hour endogenous biological processes within the body, including sleep and

wake. These two processes interact such that as sleep drive accumulates across the waking day, an increasing circadian wake drive compensates for the buildup of sleepiness and results in a relatively steady level of wakefulness across the day. As the need for sleep dissipates in the first few hours of nocturnal sleep, an increase in circadian sleep drive during the second half of the night allows sleep to continue (Dijk & von Schantz, 2005). When these processes are not in harmony, mood, daytime functioning, and physical health may be impaired (Mendlewicz, 2009; Mosendane, Mosendane, & Raal, 2008; Reid & Zee, 2009).

The circadian system is controlled by a central circadian pacemaker in the suprachiasmatic nuclei (SCN) of the hypothalamus (Eastman, Mistleberger, & Rachtschaffen, 1984; Ralph, Foster, Davis, & Menaker, 1990). The SCN are paired nuclei, each containing approximately 10,000 small, highly dense neurons. Individual neurons within the SCN are capable of producing circadian oscillations, which synchronize remarkably to generate coordinated outputs that regulate overt rhythms. These oscillatory rhythms are self-sustaining in conditions absent of time cues (zeitgebers) and are regulated by a network of transcriptional-translational feedback loops (Reppert & Weaver, 2001).

The period of these rhythms is not exactly 24-hours, and under conditions in which there are no time cues, the rhythms will drift slightly each day (Czeisler, et al., 1999; Duffy et al., 2011). Most adults have a period longer than 24 hours - the average circadian period is approximately 24 hours 9 minutes in length. However, about 35% of females and 14% of males have circadian periods shorter than 24 hours (Duffy et al., 2011). Under typical conditions, the circadian rhythm is reset on a daily basis by

zeitgebers, which allows the circadian rhythm to maintain a periodicity that matches the 24-hour solar day (Pittendrigh & Daan, 1976). The circadian rhythm is most vulnerable to time cues when they are not expected to be present, such as light exposure during the subjective night (Roenneberg, Daan, & Merrow, 2003). The restorative benefits of sleep are most pronounced when sleep occurs during the typical sleep hours shaped by an individual's circadian rhythm (Gillette & Abbott, 2005).

Melatonin serves as a biomarker for the circadian rhythm and is measurable in humans in plasma, saliva, and urine through its urinary metabolite, 6-sulphatoxymelatonin (aMT6) (Benloucif et al., 2008). Melatonin is an indole hormone secreted by the pineal gland with a robust circadian rhythm: concentrations of melatonin are stable and very low during the day, with an abrupt rise a couple hours before habitual bedtime, peaking overnight, and decreasing to the low daytime level close to habitual rise time. Melatonin is synthesized from serotonin primarily in the pineal gland. Daytime levels of serotonin are high, but decrease at night in the presence of melatonin formation due to its consumption by this process. Arylkylamine *N*-acetyltransferase (AANAT), a critical enzyme in the conversion of serotonin to melatonin, is considered the rate-limiting factor in the production of melatonin. AANAT follows a circadian rhythm of low activity during the day, limiting daytime production of melatonin (for review see Borjigin, Zhang, & Calinescu, 2012; Ganguly, Coon, & Klein, 2002). The activity of AANAT is regulated by the suprachiasmatic nuclei (SCN) of the hypothalamus, the site of the central circadian pacemaker.

Postpartum women experience a disruption in their circadian rhythm (Matsumoto et al., 2003; Nishihara et al, 2002; Thomas & Burr, 2006; Wulff & Siegmund, 2000).

Thus far, the postpartum circadian rhythm has primarily been characterized by a blunted pattern of nocturnal levels of melatonin compared to nulliparous controls (Thomas & Burr, 2006) and pregnancy levels (Wierrani, Hlawka, Kroiss, Grünberger, 1997). These altered melatonin levels may be associated with postpartum mood disorders such as depression (Anderson, 2010; Parry et al., 2008). However, the cause of the altered postpartum circadian rhythm is not known.

Changes in hormones during the postpartum period may cause disruption of the circadian rhythm. During pregnancy, estradiol and progesterone levels were inversely related to melatonin (Pang et al., 1987). While no human studies relate prolactin, oxytocin, and melatonin postpartum, a study in early postpartum ewes found prolactin levels were inversely related to melatonin (Molik, Misztal, Romanowicz, Zieba, 2010)

In addition, sleep disturbances may influence melatonin levels. The impact of total sleep deprivation on melatonin varies across studies. Total sleep deprivation combined with continuous 50 lux light exposure and sedentary activity (regularly seated) overnight reduced overall nocturnal melatonin production compared to normal sleep nights (Kavčič et al, 2011). Conversely, another study among young adult males found no effect of sleep deprivation on nocturnal melatonin levels (von Truer, Norman, & Armstrong, 1996). These studies did not address sleep fragmentation, which would relate to postpartum women. Yet, similar contradictory evidence exists regarding the impact of sleep fragmentation, as seen in patients with obstructive sleep apnea (OSA), on nocturnal melatonin levels. Wikner et al. (1997) studied males with OSA before and after treatment, and healthy control males. Melatonin did not differ before and after treatment of OSA, nor did the melatonin levels in OSA patients differ from healthy

controls. Conversely, Hernández et al. (2007) found that patients with OSA did not have a nocturnal melatonin peak, but healthy controls did.

The distinction between postpartum sleep disturbance and hormonal changes on melatonin patterns has not been made. The current study manipulated the sleep of childless women using a postpartum-like schedule in order to isolate the impact of postpartum sleep disturbance on a major metabolite of melatonin, aMT6.

Sleep Architecture

Sleep fragmentation can impact the amount of time spent in specific sleep stages. Among healthy adults who were awakened by an audiometer every minute throughout the sleep period (and verbally confirmed each awakening), the percentage of time spent in stages N2, N3, and REM sleep was significantly decreased, while the time spent in stage N1 sleep was increased compared to a baseline night (Bonnet, 1985). Total sleep time on the sleep fragmentation night was only one hour less than the total sleep time on the baseline night. Similarly, another study that experimentally induced sleep fragmentation among healthy adults by playing auditory tones at on average every 2 minutes caused a change in the amount of time spent in specific sleep stages while preserving total sleep time. During the fragmentation night time spent in stage N3 and REM were significantly reduced, and time spent in stages N1 and N2 were significantly increased compared to the screening night (Roerhs et al., 1994).

There is still limited research about time spent in certain sleep stages postpartum. While one study found postpartum women spend significantly more time in stage N3 and less time in REM than during pregnancy (Driver & Shapiro, 1992), another

study suggested no differences in sleep stages between pregnancy and postpartum (Nishihara & Horiuchi, 1998). The most recent study compared postpartum women to childless controls and found postpartum women spent a greater percentage of time in stage N3 than controls (Nishihara et al., 2004). These authors suggest that this increase was more likely to be explained by the release of prolactin secretion promoted by breast-feeding than the rebound effect of sleep disturbance because there was no change in the time spent in stage N3 among women at postpartum weeks 9-13 on uninterrupted nights of sleep versus nights when they awoke for infant caregiving. Therefore, the current study aimed to characterize sleep architecture as a result of the sleep fragmentation experienced by women during the early postpartum period, without the mitigating effects of hormonal changes by using a sample of childless, adult women.

Daytime Sleepiness

Excessive daytime sleepiness (EDS) is a common consequence of sleep disturbance and refers to an increased propensity to fall asleep and suffer sleep attacks when sleep is not desired (Benbadis, 2005). EDS can have devastating consequences including decreased performance, diminished intellectual capacity, and increased risk for vehicular and industrial accidents (Carskadon & Dement, 1987; Philip et al., 2010). Daytime sleepiness has been the primary recognized cause for many high profile disasters, including Three Mile Island, Chernobyl, Challenger explosion, and Exxon Valdez (Walsh, Dement, & Dinges, 2005). Of particular concern, perception of sleepiness is not a reliable indicator of sleep onset in monotonous situations, suggesting people are not reliable judges of their own propensity to fall asleep during inappropriate times (Herrmann et al., 2010).

Sleep disorders commonly associated with EDS include sleep insufficiency, sleep apnea, narcolepsy, idiopathic hypersomnia, and periodic limb movement disorders (Benbadis, 2005). Sleep apnea and periodic limb movement disorders are both disorders characterized by sleep fragmentation, supporting the notion that sleep fragmentation can result in daytime sleepiness. In the laboratory, when experimental sleep fragmentation was produced among healthy adults by the presentation of auditory tones at approximately 2-minute intervals, objective levels of daytime sleepiness were increased (Roehrs et al, 1994).

The mechanism through which sleep fragmentation causes daytime sleepiness, while preserving total sleep time, is not yet understood. Recent lines of evidence suggest roles for the activation of specific cytokines in mediating the relation between sleep fragmentation and daytime sleepiness. Tumor necrosis factor alpha (TNF-α) is involved in the pathogenesis of many chronic inflammatory diseases and immune responses (Popa, Netea, can Riel, can der Meer, Stalenhoef,, 2007; Zhang et al., 2009). Cytokines such as TNF-α also have established roles in the regulation of sleep. Administration of exogenous TNF-α induces sleepiness and produces excessive sleep whereas inhibition of TNF-α decreases the onset of spontaneous sleep (for review see Krueger, 2008). Mice lacking the TNF-α receptor did not show cognitive dysfunction or changes in biomarkers of sleep propensity (Kaushal, Ramesh, & Gozal, 2012; Ramesh et al., 2012) after chronic experimental sleep fragmentation, in contrast to control mice. Total sleep time and sleep architecture was preserved during the sleep fragmentation protocol (Kaushal et al., 2012), suggesting the TNF-α pathway works independently of these variables.

Adenosine, an inhibitory neuromodulator, may also serve as a potential mediator between sleep fragmentation and daytime sleepiness. Experimental systemic and intracerebral injection of adenosine has increased sleep (Benington, Kodali, & Heller, 1995; Portas, Thakkar, Rainnie, Greene, & McCarley, 1997), whereas adenosine antagonists (ex. caffeine) increase arousal (Fredholm, Battig, Holmén, Nehlig, & Zvartau, 1999). Adenosine may facilitate sleep through the inhibition of neurons related to arousal in the basal forebrain (Porkka-Heiskanen et al., 1997). Levels of adenosine have been shown to rise in the basal forebrain during sleep deprivation (Porkka-Heiskanen et al., 1997; Porkka-Heiskanen, Strecker, & McCarley, 2000) as well as during sleep fragmentation (McKenna et al., 2007). However, the role of cytokines and adenosine in the mediating the relation between sleep fragmentation and daytime sleepiness is still unclear. Cytokines and adenosine may be driving the effects of sleep disturbance, providing a compensatory response, or providing an additional unknown function.

Postpartum mothers, who are an otherwise healthy population without pre-existing sleep disorders, experience both a unique form of fragmented sleep that differs from the sleep fragmenting disorders, but appear to experience similar consequences. Postpartum mothers experience impaired neurobehavioral performance throughout the first 12 postpartum weeks compared to childless controls (Insana & Montgomery-Downs, 2013). At 6-weeks postpartum, mothers fall asleep on a Multiple Sleep Latency Test (MSLT) (an objective measure of daytime sleepiness) in an average of 11.8 minutes, a time that is borderline moderate daytime sleepiness that may or may not be at pathological levels (Insana & Montgomery-Downs, 2012). Furthermore, there are

large individual differences in the daytime sleepiness values – the standard deviation for these women was 4.6 minutes with a range of 3.1-18.6 minutes. What is unclear is how much of this variation has to do with the actual sleep disturbances experienced versus physiological (e.g. postpartum physical and hormonal changes) and environmental factors (e.g. life changes associated with caring for an infant). The current study attempted to control the environmental factors that could be influencing sleepiness, and used a non-postpartum sample to control for the physiological changes characterizing this period, to provide a clearer understanding of how sleep fragmentation during the early postpartum period impacts daytime sleepiness and performance.

Mood

Both sleep deprivation and sleep fragmentation can have negative effects on mood. One week of experimental chronic restriction of sleep to 4-5 hours per night resulted in worsened mood as reported on the Profile of Mood States (POMS) total mood disturbance, and subscales of fatigue, vigor, confusion, and tension (Dinges et al, 1997). Similar mood disturbances are seen after experimental sleep fragmentation; two nights of consecutive sleep fragmentation caused by awakenings from an auditory tone every minute throughout the nights degraded mood to levels that were greater than a single night of total sleep deprivation, despite obtaining over 5 hours of total sleep time each night (Bonnet, 1985).

Postpartum mood disturbances are prevalent; 85% of mothers report transient changes in mood during the first postpartum week (Ross et al., 2005), and an estimated 7-17% of women are diagnosed with postpartum depression after the fourth postpartum

week (Figueiredo & Conde, 2011; Horowitz, Murphy, Gregory, Wojcik, 2009; Milgrom et al., 2008; Ross et al., 2005; Pearlstein, Howard, Salisbury, Zlotnick, 2009). The adverse outcomes of postpartum depression affect the entire family unit and include: disruption in the marriage, increased neglect in child caregiving, delayed child development, and a poorer mother-infant relationship (Field, 2010; McCoy, 2011; Righetti-Veltema, Conne-Perreard, Bousquet, & Manzano, 2002). In many cases, a diagnosis of postpartum depression triggers recurring or chronic episodes of depression throughout the woman's life (Cooper & Murray, 1995; Goodman, 2004).

Yet the etiology of postpartum mood disturbances is a complex system that is not clearly understood. While the literature explained above suggests a role for sleep disturbance in causing mood disruptions postpartum, evidence also supports physiological and social causes of mood disruptions. Postpartum women experience marked changes in concentrations of sex steroids after birth, which could provide a possible mechanism for mood disruptions (for review see Russell, Douglas, & Ingram, 2000). Moreover, postpartum mood disruption could be caused by social factors, such as low levels of social support or the recent experience of a stressful event (Robertson, Grace, Wallington, & Stewart, 2004). Postpartum mood disturbances likely do not have a single cause, but rather are a combination of exposure to multiple risk factors. The current study examined the role of a single night of postpartum-like sleep disturbance on mood.

Recovery from Sleep Disturbance

The trajectory of recovery after sleep disturbance is not well elucidated. Neurobehavioral deficits caused by 5 consecutive nights of 4 hours of time in bed improved in a dose-response relation with increasing time in bed on a subsequent recovery night. However, even after 10 hours in bed after the imposed sleep restriction, neurobehavioral deficits remained, suggesting either a longer sleep period or multiple nights are required to return to baseline levels of performance (Banks, Van Dongen, Maislin, & Dinges, 2010).

Moreover, the average postpartum woman experiences chronic sleep fragmentation until their infant has developed a consolidated sleep-wake schedule that allows for sleeping through the night, which occurs around the age of three months (Kleitman & Engelmann, 1953; de Weerd & van den Bossche, 2003). Yet when, or even if, a woman recovers from this sleep disturbance is unknown. Preliminary data from our lab indicate mothers with one child between the ages of 6-30 months old did not differ in objective daytime sleepiness from mothers with one 6-week old child, despite the fact their sleep had normalized to the level of controls. The current study provided basic information concerning the impact of a single night of postpartum-like sleep disturbance on total sleep time, neurocognitive performance, and mood for a week following the sleep disturbance.

Statement of the Problem

The purpose of the current study was to understand the basic effects of a single night of sleep fragmentation, as seen during the maternal early postpartum period, on

nocturnal sleep stages, daytime impairment and recovery, mood, and melatonin levels. Only one night of sleep fragmentation occurred in the current study because of interest in the effects of acute sleep fragmentation, Multiple nights of sleep disturbance would begin to capture a different phenomenon, recovery sleep, whereby any alterations in sleep architecture on the first sleep disturbance night impact sleep architecture differently on subsequent nights (e.g. REM rebound, or increases in REM sleep) (Kilduff, Kushida, & Terao, 2005; Roehrs et al., 2004). Understanding the impact of acute postpartum sleep fragmentation, and whether there are even changes to sleep architecture on just one night, was the goal of the current study; examining multiple nights captures a different phenomenon that was beyond the scope of the current study.

New mothers experience sleep fragmentation that is associated with daytime impairment, mental health disturbances, and blunted levels of melatonin. However, there have also been large individual differences reported on each of these measures among new mothers. These individual differences are likely a result of individual physiological and environmental differences. No study has yet controlled for the physiological and environmental factors that could be impacting some of these outcomes, likely due to the difficulties of studying this population in a laboratory and the logistic and ethical barriers to changing and manipulating their routine. This study attempted to overcome these barriers and provide a basic understanding of the effects of a night of postpartum-like sleep fragmentation by manipulating the sleep of healthy, adult women without children to resemble what is observed during the early postpartum period. This study design allowed for the isolation of the postpartum-like sleep disturbances effects, without the confounding physical, hormonal, or social changes as

a result of giving birth, helping further understand the specific role of the sleep disturbance on specified outcomes.

Further, it is expected that data from the current study will provide a foundation for future studies in which environmental changes such as nocturnal light exposure, posture, and activities performed simultaneously to infant feeding/caregiving, can be systematically manipulated to determine their individual contributions to sleep and related outcomes. It also provides a basis for future studies of the role postpartum hormonal changes have on sleep, daytime sleepiness, performance, and mood. Understanding the relative importance of these factors to sleep-related outcomes is expected to guide postpartum interventions.

Research Questions and Hypotheses

RQ#1: How does a single night of experimentally-simulated postpartum sleep disturbance affect nocturnal melatonin release among healthy nulliparous women?

H1: One night of postpartum-like sleep disturbance will cause a suppression of 6-sulphatoxymelatonin compared to the baseline night.

This hypothesis was based on the blunted nocturnal levels of melatonin that have been described among postpartum women (Thomas & Burr, 2006). Other support for this hypothesis can be found in some studies that showed decreased total nocturnal levels of melatonin after experimental sleep deprivation (Kavčič et al., 2011) and absence of a nocturnal melatonin peak among patients with obstructive sleep apnea, a sleep disorder characterized by sleep fragmentation (Hernandez et al., 2007). Among patients with

Alzheimer's Disease (AD), a decreased amplitude of the melatonin rhythm has been hypothesized to explain the irregular sleep-wake rhythm and fragmented sleep symptoms (for review, see Sack et al., 2007).

RQ#2: How does a single night of experimentally-simulated postpartum sleep disturbance affect sleep architecture among healthy nulliparous women?

> H2a: Percentage of time spent in stage N1 sleep will be increased in the experimental night compared to the baseline night.

> H2b: Percentage of time spent in stage N2 sleep will be increased in the experimental night compared to the baseline night.

> H2c: Percentage of time spent in stage N3 sleep will be reduced in the experimental night compared to the baseline night.

> H2d: Percentage of time spent in REM sleep will be reduced in the experimental night compared to the baseline night.

Each time there is a disruption in the sleep cycle (such as an awakening), the sleep cycle must begin from the beginning - you cannot pick up in the sleep stage you left off in prior to the awakening. Thus, multiple awakenings from sleep fragmentation would theoretically lead to more time spent in stages at the beginning of the sleep cycle (N1 and N2) and less time spent in stages at the end of the sleep cycle (N3 and REM). Experimental sleep fragmentation studies support this, showing significant increases in time spent in stage N1 sleep, and decreases in time spent in stages N3 and REM sleep

(Bonnet, 1985; Roehrs et al., 1994). However, stage N2 sleep was increased in one study (Bonnet, 1985) and decreased in another (Roehrs et al., 1994).

RQ#3: How does a single night of experimentally-simulated postpartum sleep disturbance impact daytime sleepiness among healthy nulliparous women?

> H3: One night of experimental sleep disturbance will cause greater daytime sleepiness on a Multiple Sleep Latency Test than baseline night.

This hypothesis was based on widespread evidence that sleep disturbance results in daytime sleepiness (Benbadis, 2005). Sleep disorders characterized by sleep fragmentation cause excessive daytime sleepiness (Benbadis, 2005); postpartum women, who also experience fragmented sleep, demonstrate daytime sleepiness (Insana & Montgomery-Downs, 2012). Finally, experimental sleep fragmentation causes increased levels of daytime sleepiness (Roehrs et al., 1994).

RQ#4: How does a single night of experimentally-simulated postpartum sleep disturbance impact the next week's sleep, performance, and mood among healthy nulliparous women?

> H4a: Experimental sleep disruption will cause increased actigraphy-measured total sleep time on the proceeding at-home recovery night, compared to baseline at-home total sleep time, and total sleep time will decrease to baseline levels after the first recovery night.

This hypothesis was based on the 10-20% increase in total sleep time the night following partial and total sleep deprivation (Benoit et al., 1980; Webb & Agnew, 1975).

The night directly following experimental sleep fragmentation also had a greater total sleep time; however, this was not significant (Bonnet, 1985).

> H4b: Experimental sleep disruption will cause worse performance on a psychomotor vigilance test (PVT) on the morning after experimental sleep fragmentation compared to baseline condition, and will gradually improve to baseline levels across the recovery week.

This hypothesis was based on the worsening performance seen after sleep disruption, and that after one night of recovery sleep, performance levels are still significantly worse than controls (Banks et al., 2010; Sallinen et al., 2008). This suggests multiple recovery nights are necessary for performance to return to baseline, but the number of nights necessary is not yet clear.

> H4c: Experimental sleep disruption will cause higher POMS total mood disturbance scores the day after experimental sleep fragmentation compared to baseline condition, and will gradually improve to baseline levels across the recovery week.

This hypothesis was based on research that indicates sleep disturbance can impair mood (Bonnet, 2005). One night of sleep deprivation decreased mood, and took 2 recovery nights to return to baseline (Ikegami et al., 2009). Acute experimental sleep fragmentation decreased mood among healthy adults (Bonnet, 1985; Martin, Engleman, Deary, & Douglas, 1996). Among women within the first postpartum week, the amount of sleep lost at night was directly associated with worse mood (Swain, O'Hara, Starr, & Gorman,1997).

Supplemental Analyses: If any of the hypotheses concerning the worsening of mood, PVT performance, or MSLT scores were supported, supplemental analyses comparing these results to existing data from postpartum women in the lab were conducted in order to determine how effects of the single sleep fragmentation night compares to postpartum outcomes on these measures. A previous longitudinal study conducted in the lab continuously monitored sleep and daily PVT among new mothers across the first 12 postpartum weeks; mood surveys were administered biweekly and an MSLT was conducted among 21 of these women at postpartum week six. The number of lapses worsened across the early postpartum period, supporting a cumulative effect of sleep disturbance during this period, and lapses were significantly higher than controls at each week (Insana et al, 2013). At six weeks postpartum, mothers experienced moderate sleepiness on the MSLT (sleep onset latency average = 11.8, SD = 4.6) (Insana, 2010). Mood scores on the POMS were collected after odd-numbered weeks. Data on the mood scores are unpublished, but available for analyses.

Methods

Participants

Target Sample

Participants were healthy, childless women between the ages of 18-40. Only women were included in the current study because of the hormonal differences between men and women and the inadequate power of this small sample-size study to split analyses based on sex. Furthermore, women were chosen over men because mothers are still the primary parent responsible for nocturnal infant caregiving (Insana, Garfield,

& Montgomery-Downs, in press). Participants were excluded based on the following criteria (see *Figure 2*):

- History, current diagnosis, or high risk for major depressive disorder (≥16 score on the Center for Epidemiologic Studies Depression (CES-D) Scale (Radloff, 1977) (Appendix A)

- Symptoms of Premenstrual Dysphoric Disorder (PMDD), in accordance with the DSM-IV (Appendix A)

- Blindness or any type of ocular light perception dysfunction (including color blindness)

- Significant medical illness that put the participant under routine physician care

- Sleep disorder identified by a history of or symptoms (Appendix A)

- Drug or alcohol use (≥3 score on the Drug Use Questionnaire [DAST-10]; ≥3 score on the Short Michigan Alcohol Screening Test [SMAST]) (Appendix A)

- Irregular sleep schedule, travel across more than 1 time zone, or working night shifts within the month before the study

- Extreme sleep periods – habitual bedtimes prior to 9pm or later than 2am, and wake times prior to 6am or later than 11am

- Excessive napping, defined as napping ≥3 times during the baseline week

- Excessive daytime sleepiness, or a score of ≤8 on a baseline Multiple Sleep Latency Test (MSLT)

Figure 2. Study eligibility decision tree.
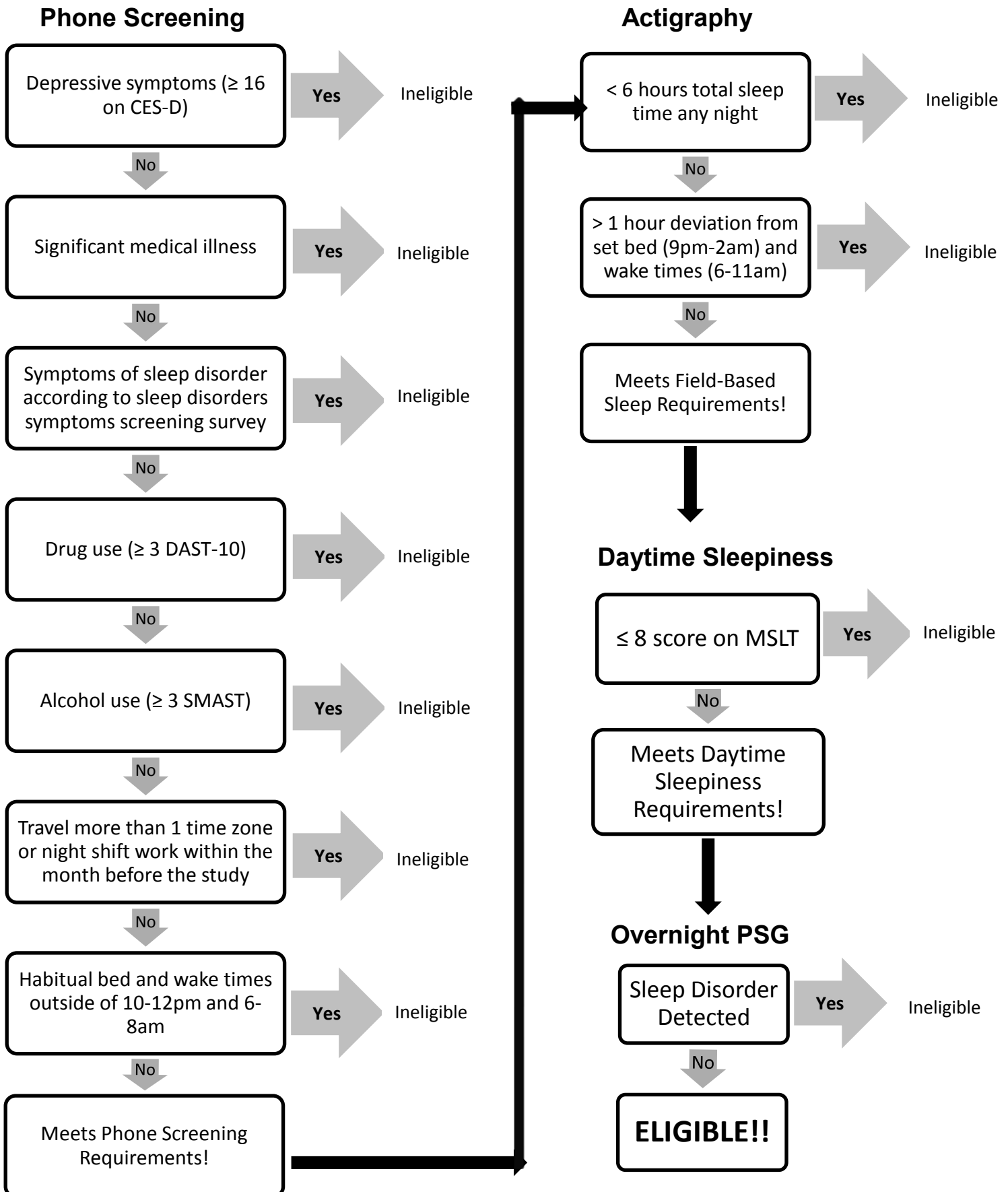
Participants who were deemed ineligible for the study on the basis of high risk for major depressive disorder or symptoms of a sleep disorder were provided with information about resources for evaluation and treatment.

Recruitment occurred via word-of-mouth and community advertisements placed to encourage a large range of socioeconomic status and diverse racial/ethnic participants. The target sample size was n=10, which was derived from effect sizes calculated based on previous studies. G Power Version 3.1 was used to derive required sample sizes (Faul, Erdfelder, Land, & Buchner, 2007). Power = 0.8 and a two-tailed alpha = 0.05 were the criterion in each of the analyses.

Power Analyses

Target power for melatonin analyses:

Based on effect sizes (*d*=2.30 and *d*=2.78) calculated from previous studies that compared melatonin suppression of acute light exposure during a 30 minute nocturnal awakening (Bojkowski et al., 1987; Gooley et al., 2011, respectively) and between constant light or dim light overnight, a sample size of n=4-5 for a within-subjects design was required to achieve necessary power.

Target power for Polysomnography (PSG) analyses:

Previous studies of changes in sleep architecture after one night of experimental sleep fragmentation have used sample sizes of 36 subjects (Roehrs et al., 1994) and 11 subjects (Bonnet, 1985). However, based on the effect sizes from Roehrs et al. (1994) comparing changes in percentage of time spent in SWS (*d* = 2.11) and time spent in

stage 1 sleep (*d* = 1.06) between baseline and sleep fragmentation nights, a sample size of N = 5-10 for a within subjects design was required to achieve necessary power.

Target power for MSLT analyses:

A sample size of approximately 10 per group (n=20) is typically reported in the literature when examining differences in levels of sleepiness between groups using the MSLT (Geisler et al, 2006; Afifi et al 2005). Based on an effect size (*d* = 1.02) calculated using a larger sample of 36 healthy young adults' changes in MSLT scores after a night of experimental sleep fragmentation, a sample size of N = 10 for a within-subjects design was required to achieve necessary power.

Target power for recovery analyses:

Sample sizes of roughly 10-15 subjects are commonly reported in the literature when examining recovery sleep and performance from sleep disturbances (Bonnet, 1985; Axelsson et al, 2008; Sallinen et al, 2008; Wehrens, Hampton, Kerkhofs, & Skene, 2012). Within these studies, large effect sizes (range: *d* = 0.87-2.24) have been reported, suggesting adequate sample sizes to examine recovery from experimental sleep fragmentation.

The above power analyses and previous study success supported the attained sample size of N = 11 for a within subjects design with 80% power at an alpha = 0.05.

Compensation

Each participant received $15 per week of at-home actigraphy based sleep/wake monitoring, plus an additional $20 for each overnight PSG and daytime MSLT, resulting

in $130 for each participant's successful completion of the entire study. Participants who failed to complete the entire study were compensated at half the rate for each successful portion she did complete.

Measures

Urinary 6-sulphatoxymelatonin (aMT6s)

At the end of the baseline and experimental sleep fragmentation nights, participants provided an overnight urine sample to assess 6-sulphatoxymelatonin (aMT6s), the primary metabolite of melatonin, to answer Research Question #1. The hormone melatonin is the gold standard biomarker for the central circadian clock and can be measured in plasma, saliva, or urine (Benloucif, et al., 2008). The main advantage of using urinary aMT6s is sampling can occur overnight without any additional sleep disturbances. Saliva sampling overnight would require participants to be awake at night to provide samples. Levels of melatonin in plasma are about three times greater than that of saliva, allowing for a greater resolution and sensitivity than saliva or urine methods. If sampling occurs at frequent intervals, plasma melatonin provides the strongest signal of the three methods, making it a preferred method. However, plasma sampling is a more expensive and invasive procedure that requires an indwelling catheter inserted into a participant's vein to allow continuous collection of plasma melatonin overnight.

Urinary aMT6 does not allow for the precise measurement of evening melatonin onset, but allows for assessing overnight concentration of melatonin (Matthews, Guerin, & Wang, 1991). After participants' morning void, urine was stirred in the commode hat

to homogenize and then divided into two approximately 50 mL aliquots. One aliquot was transported to WVU Hospital's Laboratories, where the sample was centrifuged and creatinine was analyzed within 24 hours by the Alkaline Picrate method. aMT6 concentration varies with urine volume, yet in practice, urine volume can be difficult to accurately measure. However, renal clearance can be estimated when urine volume is unknown through measurement of creatinine. Creatinine was collected as part of the current study's protocol to control for urine concentration in order to overcome methodological issues in obtaining exact urine volumes (Klante, Brinschwitz, Secci, Wollnik, & Steinlechner, 1997).

The other aliquot was centrifuged the same day as collection in Dr. Kinsey's laboratory in the Life Science Building for 5 minutes at 2000 x g. A 1 mL aliquot was pipetted into a small test tube to be stored separately and used for the aMT6 analysis. This was to prevent repeated freeze thaw cycles if multiple testing on the urine samples was needed. Both samples were immediately stored in a -20°C freezer. Within the week, these samples were transferred into a -80°C freezer for long-term storage (samples can be stored at ≤-20°C for up to 1 year). Once all samples were collected, batch assay was conducted according to ALPCO's (Salem, New Hampshire) aMT6 enzyme-linked immunosorbent assay (ELISA) kit's protocol.

Overnight Polysomnography (PSG)

The established 10-20 system was used to set up participants for PSG (Klem, Luders, Jasper, & Elger, 1999). Minimum montage used included central referential electroencephalogram C3 and C4 (backup), occipital referential electroencephalogram

O1 and O2 (backup), right and left electro-oculogram leads, 2 submental electromyogram leads on the participant's chin, and electrocardiogram. High resolution video monitoring simultaneously collected video recording and was used to help score sleep. In addition to the minimum montage described above, on the first PSG adjustment/screening night, abdominal and thoracic respiratory belts, an oral/nasal thermistor, a snore sensor, and a pulse oximeter were used to screen for respiratory disorders of sleep. Leg EMG sensors were placed on the anterior tibialis muscles to screen for Periodic Limb Movement Disorder. These extra channels provided the data necessary to detect a sleep disorder, if present. When the first night of screening indicated no sleep disorders, the participant continued in the study and the next two nights (baseline and experimental) used the minimal montage to increase ease of movement overnight and reduce discomfort of excess wires. The overnight PSG protocol allowed for precise examination of time spent in sleep stages (these cannot be measured using actigraphy) that is needed to address Research Question #2.

Multiple Sleep Latency Test (MSLT)

The laboratory-based Multiple Sleep Latency Test (MSLT) was used as an objective measure of daytime sleepiness, operationally defining the degree of sleepiness as sleep latency (Afifi et al., 2005). Daytime sleepiness is a complex system unlikely to be easily measured through just one method. Objective measures of sleepiness, such as the MSLT and Maintenance of Wakefulness Test (MWT), have typically been used in preference over subjective measures, which lack sensitivity and specificity (Coelho, Narayansingh, & Murray, 2011). The MSLT is considered the gold standard measure of sleepiness and has been used to diagnose narcolepsy and sleep

disorders with a secondary presentation of excessive daytime sleepiness (Arand et al., 2005; Mitler et al., 1979; Van den Hoed et al., 1981). The protocol for the proposed study followed standardized, established guidelines for the MSLT (Littner et al., 2005). The MSLT required participants' sleep to be monitored for one to two weeks prior to the test via actigraphy or sleep/wake diaries. Participants were asked to refrain from caffeine and alcohol the day of the MSLT. On the day of the MSLT, participants came into the laboratory and were given four nap opportunities, each spaced at 2 hour intervals from each other, with the first nap beginning 1.5-3 hours after termination of nocturnal sleep. Each nap opportunity was 20 minutes in duration and participants were in a quiet, dark bedroom and instructed to "Please lie quietly, assume a comfortable position, keep your eyes closed and try to fall asleep." The minimum montage described above for the second and third overnight PSG was used. High resolution video monitoring simultaneously collected video recording and was used to help score sleep onset as well as ensure participants remained awake during the breaks between MSLT naps. Sleep latency time was defined as the time it takes from lights out to the first 30-second epoch of any sleep stage using the American Academy of Sleep Medicine's 2008 rules for PSG sleep stage scoring. In any of the nap opportunities, if sleep did not occur, the sleep latency was recorded as 20 minutes. The outcome measure of the test was the average sleep onset latency from the 4 nap opportunities and was used to test the hypothesis for Research Question #3, as well as to screen participants at baseline for eligibility in the study. Interpretation of the MSLT scores typically follows a 'rule of thumb.' Average sleep onset latency scores ≤5mins indicate pathological level of daytime sleepiness and scores between 10 and 20 minutes are considered normal

levels of sleepiness. Scores between 5 and 10 minutes fall into a diagnostic grey area (Afifi et al., 2005). The use of a baseline MSLT score of ≤8 as exclusion criteria in the current study is based on the definition of sleepiness for diagnostic purposes, as identified by the *International Classification of Sleep Disorders* (AASM, 2005).

Actigraphy

Sleep/wake periods were monitored at home using Mini Mitter's Actiwatch-64 (AW-64) actigraphs (Bend, Oregon). The actiwatch looks and is worn like a wrist watch on the participant's non-dominant wrist. It uses an accelerometer sensor to interpret movement; these data on movement intensity were downloaded onto the laboratory computer via a reader. The actiwatch was programmed at the most sensitive 15-epoch setting, which allowed up to 11 days of continuous monitoring. Because actiwatches record motion like an accelerometer, periods of nocturnal sleep and daytime naps were identified using the sleep diary filled out by participants on their personal digital assistant (described below) to corroborate data from actigraphy outputs. The Actigraph software was used to calculate total sleep time and ensure participants maintained a regular sleep schedule during the baseline period, as well as to assess recovery for Research Question #4. Actigraphy has been well-validated for detecting sleep/wake patterns among adults (Benson et al., 2004; Edinger, Means, Stechuchak, & Olsen, 2004; Morgenthaler et al., 2007; Sadeh, 2011). Its advantage over the traditional, gold standard PSG is that it can conveniently record continuously for long periods of time, and it allows for measurement of sleep patterns in the field. It has been correlated with PSG for differentiating sleep from wake, with 91% agreement rates (de Souza et al., 2003).

A Palm Zire 72 handheld personal digital assistant (PDA) was used to collect field-based sleep and actiwatch diaries. These diaries behaviorally corroborated actigraphy data and are essential to help control artifact that could misidentify periods of immobility as sleep (Acebo & LeBourgeois, 2006). Further, an electronic diary was chosen over a paper and pencil diary because of improved adherence to filling out logs when using an electronic diary compared to paper-and-pencil logs (Stone, Shiffman, Schwartz, Broderick, & Hufford, 2002) as well as participants' reported increased preference for the electronic over paper version in recording of behaviors (Lam et al., 2010) and infant sleep (Müller, Hemmi, Wilhelm, Barr, & Schneider, 2011).

Psychomotor Vigilance Test (PVT)

The psychomotor vigilance test (PVT) was self-administered using the PDA each morning within two hours after awakening and prior to the consumption of any caffeine to prevent possible caffeine-related or circadian alerting effects (Lim & Dinges, 2008). Each test lasted 5 minutes, during which stimuli were presented at random inter-stimulus intervals. The use of this 5-minute test is supported by a validation of PVTs less than 10 minutes in duration (Loh, Lamond, Corrian, Roach, & Dawson, 2004). Reaction times were recorded for response to each individual stimulus, and the number of lapses (reaction times ≥500ms) was calculated from the reaction times. The PVT has become the predominant test of vigilant attention used in sleep research because of its high sensitivity to sleep deprivation, predictive ability of performance in simulated driving tasks, and relatively minor learning effects (Basner & Dinges, 2011; Jackson, Croft, Kennedy, Owens, & Howard, 2013; Lim & Dinges, 2008). The frequency of lapses was

used to assess recovery of performance after experimental sleep fragmentation for Research Question #4.

Profile of Mood States (POMS)

The Profile of Mood States (POMS) (Appendix A) was administered in the laboratory prior to and after the experimental sleep fragmentation, and it was given to each subject to self-administer at home each day for the recovery week (automatic text messages were sent from the research team to remind the women to fill this out). The POMS consists of 65 adjectives rated by participants using a 5-point Likert scale. Six subscales have been derived from the POMS: tension-anxiety, depression-dejection, anger-hostility, fatigue-inertia, confusion-bewilderment, and vigor-activity. Subscale ranges vary: tension-anxiety (0–36); depression (0–60); anger-hostility (0–48); fatigue (0–28); confusion bewilderment (0–28); and the reverse-scored vigor-activity subscale has a range from -32 to 0. A POMS Total Mood Disturbance (TMD) score is derived from the addition of the six subscale scores, has a range from -32 to 200, and is an indicator of a global dimension of mood disturbance. POMS TMD scores and all but the vigor-activity subscale are negative dimensions, so high scores represent worse mood. Survey responses reflected the participant's mood during the past day. Internal consistencies of the POMS when administered to adults 17-60 years old ranged from 0.84 (confusion-bewilderment scale) to 0.95 (depression-dejection scale) (McNair, Lorr, & Droppleman, 1992). Scores on the POMS were used to assess recovery of mood after experimental sleep fragmentation for Research Question #4.

Procedure

Once participants were identified, they came into the laboratory where a researcher gave a laboratory tour, described the research protocol in full, provided instructions for using the equipment (actigraph and PDA), described the surveys, collected demographic information, and answered any residual questions from participants. Informed consent and a Health Insurance Portability and Accountability Act (HIPAA) authorization were administered to participants. The procedure for the study is illustrated in *Figure 1*, and is split into 3 phases.

Phase I

Participants began the study with a 7-day field based protocol, which served both to normalize sleep and provide a baseline measurement after which the experimental night was compared. Participants were also asked to keep a consistent, approximately 8-hour sleep period (more if needed) with a bedtime based on habitual sleep periods. Participants wore an actigraph on their non-dominant wrist and filled out on their PDA: when they put on and took off the actigraph, a sleep diary, and completed a PVT each morning within 2 hours after awakening. The nightly sleep time and average daily number of lapses on the PVT were averaged across the week to create stable baseline measures.

Phase II

At the end of the 7-days, participants came into the laboratory for the second phase, a laboratory-based protocol. They came into the laboratory about 1-1.5 hours after awakening on the 7th day so that research personnel could apply the PSG sensors

before the first MSLT nap opportunity (scheduled 1.5-3 hours after awakening that morning). Prior to or immediately after the first nap opportunity, a researcher downloaded the data from the week of actigraphy to ensure participants followed protocol and determine average bedtime, rise time, and time spent in bed (these values were used for the in-laboratory nights). Participants underwent the MSLT, conducted according to standardized protocol (Littner et al., 2005). Between nap opportunities participants remained in the laboratory and engaged in leisure activities such as reading, watching a movie, using the internet, or anything else that involved low levels of activity. They brought a lunch and snacks, as this was not be provided by the researcher team. Participants were continuously monitored with video recording to ensure they did not fall asleep between the nap opportunities. Additionally, they filled out the POMS survey between naps.

At the end of the MSLT day, participants who had average sleep onset latencies equal to or less than 8 minutes were disqualified from the study. Otherwise, they were allowed to go home for a few hours, and then asked to come back about 1.5 hours before their habitual bedtime (determined via actigraphy data from baseline week) to get set up for the first overnight PSG. Participants slept according to their habitual sleep periods determined by baseline actigraphy data. This night served as an adjustment night in order to account for the first-night effect, which is a phenomenon by which participants experience increased awakenings, decreased time spent in REM sleep, longer latencies to SWS and REM, and a greater number of stage changes during the first night spent in a sleep laboratory (Agnew, Wilse, Webb, & Williams, 1966). It also served to ensure none of the participants had a sleep disorder. Data from this night

were not used in subsequent analyses. After the first night, participants had sensors removed, left the laboratory, and were instructed to come back that night. During their day away from the laboratory, participants refrained from napping, and wore their actiwatch.

The second night of the protocol was conducted in the same manner as the first night but was used as a baseline night for subsequent analyses. Urine was also collected overnight for the measurement of aMT6. Participants voided urine within 10 minutes of bedtime. Consistent with other studies collecting overnight aMT6, this pre-bedtime void was not collected for analysis (Davanipour, Poulsen, Weimann, & Sobel, 2009; Roach et al., 2005). From that point on, all urine passed during the night was collected in a commode hat and remained there at room temperature. At the end of the sleep period, participants voided urine into the commode hat. After the pre-bedtime urine void, participants were kept in dim light (12 lux in the direction of gaze one meter away, or more accurately, 3.5 lux from the participant's perspective lying in bed, verified by a light meter) while bio-calibrations were conducted until lights out. The bedroom was completely dark during sleep periods (0 lux). A red light was used in the bathroom if participants awakened in the middle of the night to void. Dim and red light were used in order to minimize the impact of light on 6-sulphatoxymelatonin (aMT6s) secretion. Participants returned home after the second night.

On the third night, participants again came into the laboratory about 1.5 hours before bedtime to get hooked-up for PSG. This night served as the experimental night. Based on data from our lab indicating women report an average of 2.9 awakenings each night during the second postpartum week (Winser, 2012), and the

recommendations from the AAP that breastfed newborns should be fed at intervals between 1.5-3 hours, mothers were awakened 3 times throughout the night. Previous literature indicates feeding time is approximately 13.8-16 minutes (De Carvalho et al., 1982; Thoman et al., 1971), but that mothers spend an additional 12 minutes engaged with the infant in non-feeding activities (Thoman et al., 1971). Based on the combination of these data and literature, it was decided the protocol for the current study would consist of 3 awakenings of about 30-35 minutes each, spaced equally apart throughout the night. Each participant's bedtime and habitual time spent in bed was preserved; scheduled awakenings added into the sleep period resulted in a rise time that was 1 hr 30 mins – 1 hr 45 mins later than typical for each participant.

No published data exist on the activities new mothers engage in at night. The current protocol for the 35 minute awakenings was therefore not based on research. However, it was based on anecdotal information from mothers and was standardized for each participant. Posture and lighting levels (1 lux in the direction of gaze, verified by a light meter) during each awakening were kept consistent between participants because both changes in posture and lighting levels can impact melatonin levels and sleepiness (Cajochen et al., 2005; Deacon & Arendt, 1994; Gooley et al., 2011). Each participant was told that they would be awakened three times throughout the night by an audio clip of a baby crying via an intercom next to the participant's bed. The crying baby was sufficient to wake each participant for each awakening, confirmed by EEG data. They were then given the following standard instructions via the intercom during each of the three awakenings:

1. Pick up the baby doll, change the doll's diaper with a diaper that was in the participant's room (5 minutes - participant was standing during the procedure).

2. Sit in the rocking chair and pretend to feed the baby doll with the bottle provided (15 minutes – participant was seated)

3. Burp the baby (5 minutes – participant was seated)

4. The participant was then asked to stand and put the baby back down on the spare bed, and was allowed to go to the bathroom (if needed) (5 minutes – participant was standing)

5. The researcher entered to fix any PSG sensors that had fallen off, after which the participant will be told to go back to bed.

The researcher entered the room each time the participant was moved from one posture to another to ensure participants did not trip over or drop the equipment in the dimly lit room. In some circumstances, the researcher also entered to provide the participant with an extra blanket. During all interactions with the participant at night, conversation was kept to a minimum. Once participants were awakened in the morning, first morning void was collected (and combined with all samples from any time they used the bathroom overnight). They remained in the laboratory that day for the second MSLT (which began 1.5-3 hours after awakening). The MSLT was conducted as described above during the baseline session.

Phase III

After the laboratory Phase II, participants went home and their sleep was monitored with actigraphy and a sleep diary on the PDA for 7 nights, and they self-

administered the PVT each morning within 2 hours after awakening (same protocol as the first 7 nights of the study, except sleep was ad libitum). The POMS was filled out every other day to capture changes in mood during recovery. At the end of the study, a researcher picked up the equipment from the participant either in the laboratory or at a location convenient for her. After full participation in the study, each participant received $130 in compensation.

Results

Participants

Women were recruited via university-wide and the Department of Psychology listservs, an ad on Craigslist, and word of mouth between March and November 2013. *Figure 3* illustrates how many women expressed initial interest and then declined to begin or continue participation, or were excluded at various points throughout the study. Based on an *a priori* power analyses, the target sample size was n=10. An extra participant was included in case one participant's data were incomplete or an outlier on any measure was found, and had to be excluded post-hoc. As no post-hoc exclusions were necessary, 11 women contributed data to the study. Women who completed the study were all nulliparous with no reported history or current symptoms of depression, as defined by a score of <16 on the CES-D (Radloff, 1977); they did not report symptoms of premenstrual dysphoric disorder (PMDD) and all had regular menstrual cycles (defined as being confident in their ability to predict when their period will start, to the week); they did not work night shifts or travel through >1 time zone during the month prior to participating in the study; these women did not report blindness or visual impairments that would interfere with the production of melatonin; they did not have a

major medical illness for which they received routine physician care; were not smokers;
they consumed no more caffeine per day than the amount in two 6-oz cups of coffee,
consumed on average <3 alcoholic drinks per week, and were told not to have any
alcoholic beverages the day of any overnight polysomnography (PSG).

Based on the PSG screening night, no participant had any respiratory events.
Three of the completed participants had periodic limb movements in sleep (PLMS), with
the highest number of PLMS/hour at 8.3 for one participant (periodic limb movements
with associated arousals [PLMA]/hour=1.0). The other two had PLMS/hours indices of
2.6 (PLMA=0.8) and 1.6 (PLMA=0). The International Classification of Sleep Disorders
(AASM, 2005) requires >15 PLMS/hour along with a clinical sleep disturbance or
complaint of daytime fatigue before it is considered a sleep disorder. As PLMS are
common findings on PSG recordings, and none of these participants had clinically
significant levels of PLMS that were consistent with a disorder, had complaints of
daytime fatigue, or showed signs of excessive daytime sleepiness (based on recorded
MSLT scores), all participants' data were included in the final analyses.

Demographic characteristics of the total sample are in *Table 1*. Between-groups
one-way Analysis of Variances (ANOVA) and chi-square tests were used to analyze
whether the participants who began but did not complete the study differed from those
who completed the study. No differences were found between groups on any
demographic variable or baseline actigraphically-recorded total sleep time.

Figure 3. Flowchart of participants throughout the study.

Women who expressed interest in the study (n=189)

Excluded* (n=162)

Women who completed the phone screening (n=27)

Excluded (n=8)
    Based on ineligibility[#] (n=4)
    Scheduling conflict arose prior to start of study (n=4)

Women who began the study (n=19)

Excluded (n=8)
    Dropped out due to scheduling conflicts (n=2)
    Based on ineligibility^ (n=6)

Women who successfully completed the study (n=11)
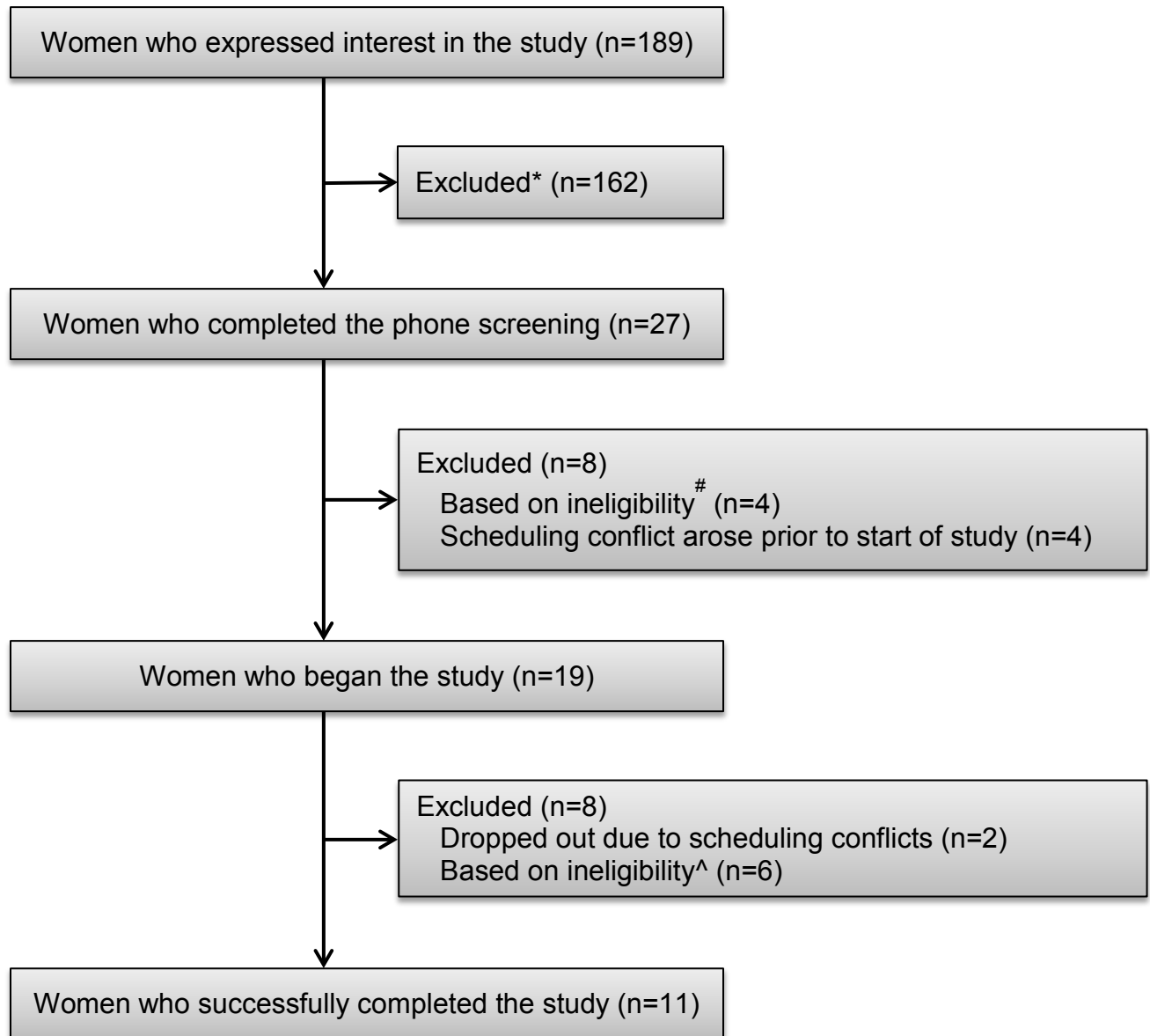
*Did not differentiate between those who were unable to participate due to scheduling difficulties and those who were no longer interested.

[#]Reasons for ineligibility to begin study: on antidepressant medication (n=1), symptoms of a sleep disorder (n=2), major medical condition (n=1).

^Reasons for ineligibility to continue study: sleep/wake times differed across baseline week by >3 hours (n=1), score <8 on MSLT (n=4), sleep disorder symptoms on screening night (n=1).

Table 1.

Participant demographics

Participant	Age	Income	Years of Education	Race/Ethnicity	BMI	Marital Status	Work Status	Hormonal Birth Control	M-E Score	Baseline TST (hh:mm)
119	29.8	$50,000	18	Asian	16.9	Married/Living Together	Unemployed	No	35	8:17
122	23.2	$20,000	17	Biracial	22.7	Married/Living Together	Full-time Student	Yes	44	7:05
326	26.1	$12,000	19	Asian	20.4	Single	Full-time Student	No	43	8:24
355	23.0	$12,000	17	White	26.0	Single	Full-time Student	Yes	67	7:05
391	26.1	$30,000	18	White	23.2	Single	Full-time Student	No	48	8:05
412	24.4	$15,000	18	White	23.3	Married/Living Together	Full-time Student	No	78	7:32
474	26.2	$13,500	20	White	19.1	Single	Full-time Student	Yes	71	7:52
533	23.9	$25,000	17	White	20.5	Single	Full-time Student	No	61	6:53
696	23.2	$25,000	16	White	23.1	Married/Living Together	Full-time Student	Yes	52	8:22
827	29.0	$32,000	18	White	35.6	Single	Full-time	Yes	54	7:16
886	23.8	$20,000	17	White	25.4	Single	Full-time Student	Yes	42	7:41
Mean/ Percentage	25.4	$23,100	17.7	72.7% White	23.3	63.6% Single	81.8% Full-time Student	54.5% Yes	54.1	7:41
Standard Deviation	2.3	11,300	1.10		4.8				13.6	0:33

M-E=Morningness-Eveningness Questionnaire; TST=Total Sleep Time; BMI calculated from participants' weights and heights in the laboratory during consent visit; Participant numbers generated with online random number generator (http://www.random.org)

Data Management

Data were checked for missing variables prior to conducting analyses. One Psychomotor Vigilance Test (PVT) data point was missing on the first day of the study for a single participant due to participant non-adherence. One night of actigraphy data was missing on the third day of the recovery week due to participant non-compliance (she forgot to put the actigraph back on before she went to sleep). Pairwise deletion was used in these cases. One participant had 40 minutes of unscoreable data during the baseline overnight PSG due to technician error. Percentage of time spent in each sleep stage for this participant was based on the total sleep that was scored. On one of the post-fragmentation MSLT nap opportunities, a participant had an unscoreable recording because of PSG equipment malfunction after the nap opportunity had started. The participant's MSLT score was based on the three usable naps.

SPSS version 21.0 was used for data analysis. For all of the following analyses, a $p<.05$ was considered statistically significant, except in noted cases where a Bonferroni correction was used. Partial eta squared (η^2_p) (small=.01, medium=.06, large=.14), Cohen's d (small=.2, medium=.5, large=.8), and Cramer's V (small=.1, medium=.3, large=.5) were used to calculate effect sizes.

Normality and Outliers

Continuous variables were checked for skewness and kurtosis. PVT lapse frequency on the 7th day of the recovery period was slightly leptokurtic (Kurtosis/SE Kurtosis=3.3). Square root transformation was sufficient to achieve normality, and analyses were run on the transformed and untransformed variable. However, statistical

results did not differ; therefore, results are reported on untransformed values. Percent time spent in REM sleep on the sleep fragmentation night was slightly negatively skewed (Skewness/SE Skewness=-3.2) and leptokurtic (Kurtosis/SE Kurtosis=3.9). Square root and logarithmic transformations were insufficient to achieve normality, so non-parametric statistics were used when analyzing this variable. All other overnight PSG, actigraphic, MSLT, and POMS TMD data were normally distributed with no outliers.

Results for Research Question #1

RQ#1: How does a single night of experimentally-simulated postpartum sleep disturbance affect nocturnal melatonin release among healthy nulliparous women?

The intra-assay coefficient of variation of the ELISA was 4.4%. The results did not support the hypothesis, that *one night of postpartum-like sleep disturbance will cause a suppression of 6-sulphatoxymelatonin compared to the baseline night*. A paired samples t-test was run to test for differences in 6-sulphatoxymelatonin (aMT6) concentration between baseline and sleep fragmentation nights. There was not a significant difference between aMT6 concentrations following the baseline (*M*=23.3, *SD*=13.5) and sleep fragmentation nights (*M*=25.0, *SD*=12.2; *t*[10]=.69, *p*=.51, *d*=.09) (see **Figure 4**).

Figure 4. aMT6 concentrations during baseline and sleep fragmentation nights for individual participants.
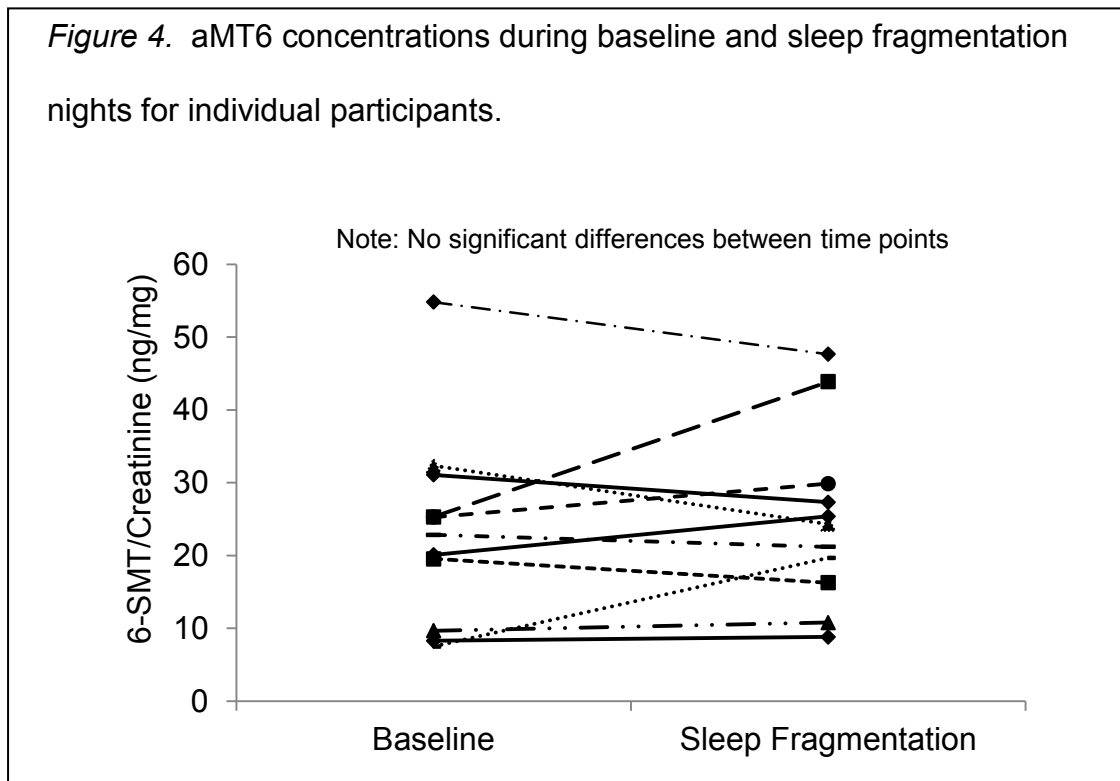
Research Question #1: Supplemental Analyses

Because ***Figure 4*** indicates individual differences in changes in aMT6 concentration, participants change score (baseline aMT6 concentration - sleep fragmentation aMT6 concentration) was compared to selected demographics that either had sufficient variation within the sample to test for differences (income and baseline TST) or were of particular interest to circadian outcomes (use of oral contraception and M-E scores) to determine whether certain individual differences were driving differences in aMT6 changes. Correlations for continuous measures (income, M-E scores, baseline TST) and a t-test (for the categorical measure use of oral contraception) revealed no relation between any of these demographic variables and the change in aMT6 concentration.

Results for Research Question #2

RQ#2: How does a single night of experimentally-simulated postpartum sleep

disturbance affect sleep architecture among healthy nulliparous women?

Following the American Academy of Sleep Medicine's (AASM) standard scoring

criteria, all overnight PSG studies were scored by two independent analysts (HMD and

ALM), one who was blind to the participants' identity (HMD). Blinding to the condition

(whether it was the initial adjustment night, baseline, or sleep fragmentation night) was

not possible because the recording montage differed between the adjustment and

subsequent nights, and because the forced awakenings on the experimental night could

be easily differentiated from the baseline night. However, the order of scoring of both

participant and condition were randomized for HMD so that she did not score either

participants or their PSG records chronologically. Minor discrepancies between HMD

and ALM were settled by consensus so that the final agreement on the overnight stage,

arousal, and event scoring was 100%.

To determine whether the protocol was successful at simulating postpartum

sleep disturbance (maintaining total sleep time and decreasing sleep efficiency during

the experimental night compared to the baseline night), two paired samples t-tests were

conducted to test the difference in total sleep time and sleep efficiency between the

baseline and sleep fragmentation nights. There was not a significant difference in total

sleep time between baseline (*M*=461min, *SD*=28min) and experimental nights

(*M*=448min, *SD*=34min; *t*[10]=1.49, *p*=.17, *d*=.42). There was a significant decrease in

sleep efficiency from baseline (*M*=90.9%, *SD*=6.1%) to experimental nights (*M*=74.4%,

SD=3.9%; *t*[10]=9.31, *p*<.001, *d*=3.20). The protocol was also successful in awakening

each participant three times equally spaced throughout the night of approximately the

same duration for each awakening (see *Table 2*). In sum, these data support that the

sleep fragmentation protocol was successful in simulating postpartum sleep

disturbance.

Table 2.

Descriptive information on sleep fragmentation night

	Mean (min)	SD (min)	Range (min)
Duration of Awakenings			
Awakening 1	32.9	3.2	29.8 - 38.7
Awakening 2	31.8	1.8	29.6 - 34.7
Awakening 3	32.4	2.5	29.9 - 37.1
	Mean (hh:mm)	SD (hh:mm)	Range (hh:mm)
Duration Between:			
Lights Out & Awakening 1	2:06	0:09	1:48-2:21
Awakenings 1 & 2	2:06	0:07	1:59-2:20
Awakenings 2 & 3	2:06	0:07	2:00-2:19
Awakening 3 & Lights On	2:06	0:07	1:56-2:18

Hypothesis 2a, that the *percentage of time spent in stage N1 sleep will be*

increased in the experimental night compared to the baseline night, was not supported.

A paired samples t-test using a Bonferroni correction (p=.012) was run to test for

differences in time spent in stage N1 sleep between baseline and sleep fragmentation

nights. There was not a significant increase in the proportion of time spent in stage N1 during the sleep fragmentation night (*M*=6.7%, *SD*=2.5%) compared to baseline (*M*=4.3%, *SD*=2.4%; *t*[10]=2.77, *p*=.02, *d*=.97) (see **Figure 5**).

Hypothesis 2b, that the *percentage of time spent in stage N2 sleep will be increased in the experimental night compared to the baseline night*, was not supported. A paired samples t-test using a Bonferroni correction (*p*=.012) was run to test for differences in time spent in stage N2 sleep between baseline and sleep fragmentation nights. There was not a significant increase in stage N2 sleep on the sleep fragmentation (*M*=39.7%, *SD*=5.4%) night compared to baseline (*M*=43.5%, *SD*=8.3%; *t*[10]=1.83, *p*=.10, *d*=.55) (see **Figure 5**).

Hypothesis 2c, that the *percentage of time spent in stage N3 sleep will be reduced in the experimental night compared to the baseline night*, was not supported. A paired samples t-test using a Bonferroni correction (*p*=.012) was run to test for differences in time spent in stage N3 sleep between baseline and sleep fragmentation nights. There was not a significant decrease in stage N3 sleep on the sleep fragmentation night (*M*=30.2%, *SD*=4.8%) compared to baseline (*M*=28.8%, *SD*=6.3%; *t*[10]=.64, *p*=.54, *d*=.24) (see **Figure 5**).

Hypothesis 2d, that the *percentage of time spent in REM sleep will be reduced in the experimental night compared to the baseline night,* was not supported. With a Bonferroni correction (*p*=.012), there was not a significant decrease in REM sleep on the sleep fragmentation (*Mdn*=25.9%) night compared to baseline (*Mdn* =22.9%) with the Wilcoxon's signed-rank test, *Z*=.44, *p*=.66, *r*=.13 (see **Figure 5**).

Figure 5. Time spent in each sleep stage between baseline and sleep fragmentation nights (Error bars represent SE).
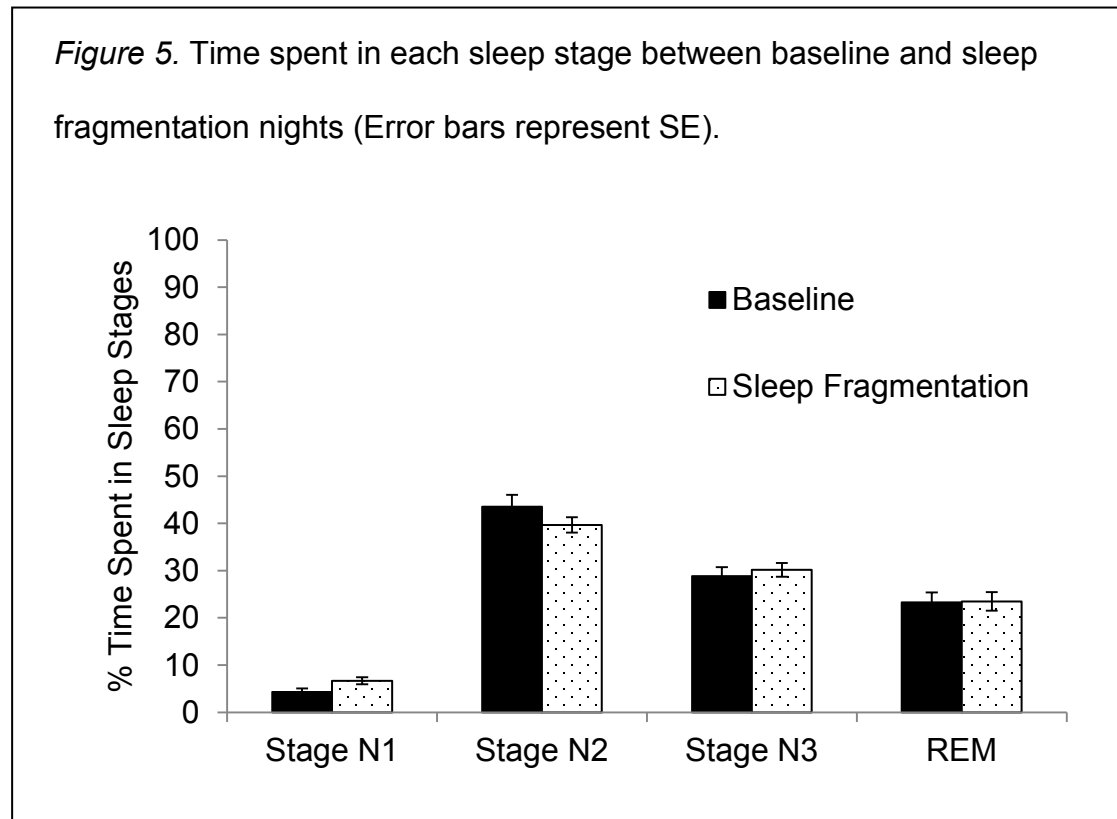
Research Question #2 Supplemental Analyses

Because interesting patterns emerged through visual analysis of the data, comparisons were made to determine whether sleep onset latency following each of the three forced awakenings changed across the sleep fragmentation night. A repeated-measures ANOVA was used to test for a polynomial shape across time. There was a significantly linear trend of decreasing sleep latencies across the night (F[2]=11.2, p=.007, η^2_p =.53) (see **Figure 6**).

Figure 6. Sleep onset latency after each successive forced awakening on the sleep fragmentation night (Error bars represent SE).
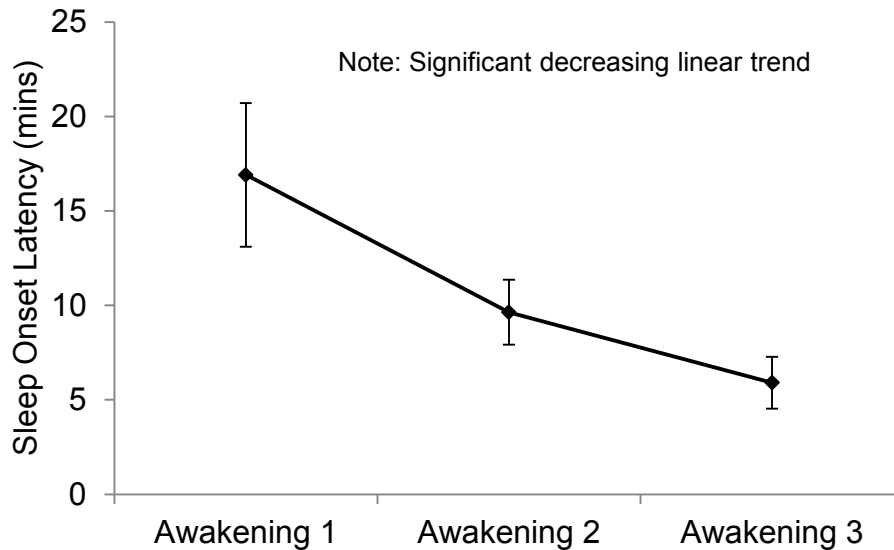
Results for Research Question #3

RQ#3: How does a single night of experimentally-simulated postpartum sleep disturbance impact daytime sleepiness among healthy nulliparous women?

The Multiple Sleep Latency Test (MSLT) was scored in real-time per standard scoring criteria (Littner et al., 2005) and was then also examined 3 months after data collection was completed independently by both ALM and HMD. Both were blind to whether the test was pre or post sleep fragmentation and their order was randomized to prevent an order effect of systematic changes across participants. HMD had administered 2% and ALM administered 56% of the MSLT nap opportunities. The retrospective scores were then compared to each other and to the original real-time scores. There was a 47.7% agreement rate on each nap opportunity between HMD retrospective scoring and the original scoring. Systematic discrepancies were observed

between the original and retrospective scores: when the original MSLT was administered by a research assistant, there was greater discrepancy between the original and retrospective scores. ALM administered 86% of the baseline MSLT nap opportunities; research assistants administered 89% of the post-fragmentation MSLT naps opportunities. To avoid errors in the planned comparisons due to systematic differences in scoring, MSLT scores were retrospectively changed according to rules described in Appendix A.

In accordance with standard criteria, the baseline week of actigraphy simultaneously served as monitoring to ensure sufficient sleep prior to the MSLT, specifically that the night prior to the MSLT was representative of the participants' normal sleep (Littner et al., 2005). A t-test performed comparing the average total sleep time of the first six days of the baseline week (*M*=462, *SD*=33) to the seventh day (the night before the MSLT) (*M*=454, *SD*=53) confirmed there was no significant change in total sleep time (*t*[10]=.65, *p*=.53, *d*=.19).

The hypothesis that *sleep fragmentation would cause greater daytime sleepiness on an MSLT compared to baseline* was not supported. A paired samples t-test was run to test for differences between baseline and post-sleep fragmentation MSLT scores. There was a significant <u>decrease</u> in objective sleepiness (an increase in the MSLT score) between the day following baseline (*M*=13.1, *SD*=3.54) and the day following sleep fragmentation nights (*M*=16.3, *SD*=3.51; *t*[10]=2.83, *p*=.02, *d*=.92) (see ***Figure 7***).

Figure 7. MSLT average sleep onset latencies between baseline and sleep fragmentation nights for each individual participant (Error bars represent SD).
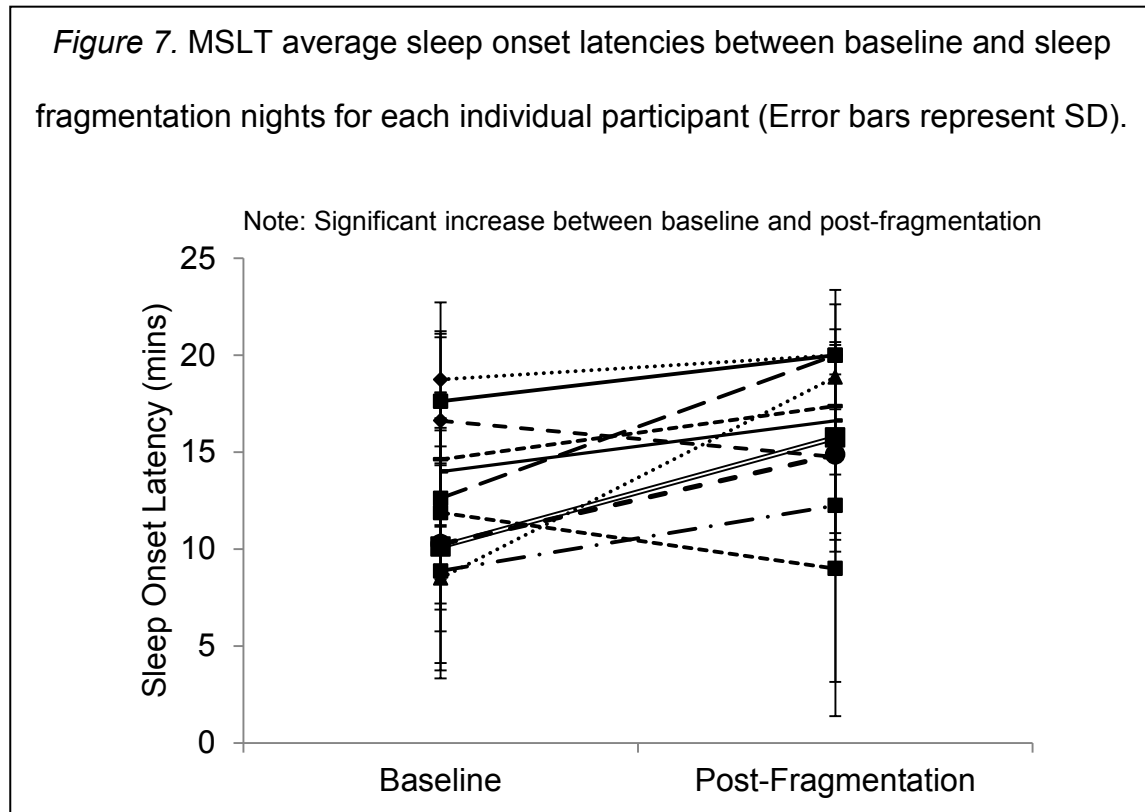
Research Question #3: Supplemental Analyses

In order to determine whether sleepiness at certain times of the day was driving the overall improvement in sleepiness, supplemental analyses were made comparing the MSLT scores of the four individual naps from baseline to post-sleep fragmentation. A 2 (condition: baseline vs. post-fragmentation) x 4 (each individual nap) repeated-measures ANOVA was conducted. A significant main effect of condition was qualified by a significant condition x nap interaction, $F(3)=5.65$, $p=.004$, $\eta^2_p =.39$. Post-hoc dependent t-tests using a Bonferroni correction ($p=.012$) determined that there was a significantly shorter sleep onset latency during the baseline MSLT for the fourth nap during baseline compared to post-fragmentation, $t(10)=3.43$, $p=.006$, $d=1.56$ (see **Figure 8**).

Figure 8. MSLT sleep onset latencies for individual nap opportunities between baseline and the day after experimental sleep fragmentation (Error bars represent SE).
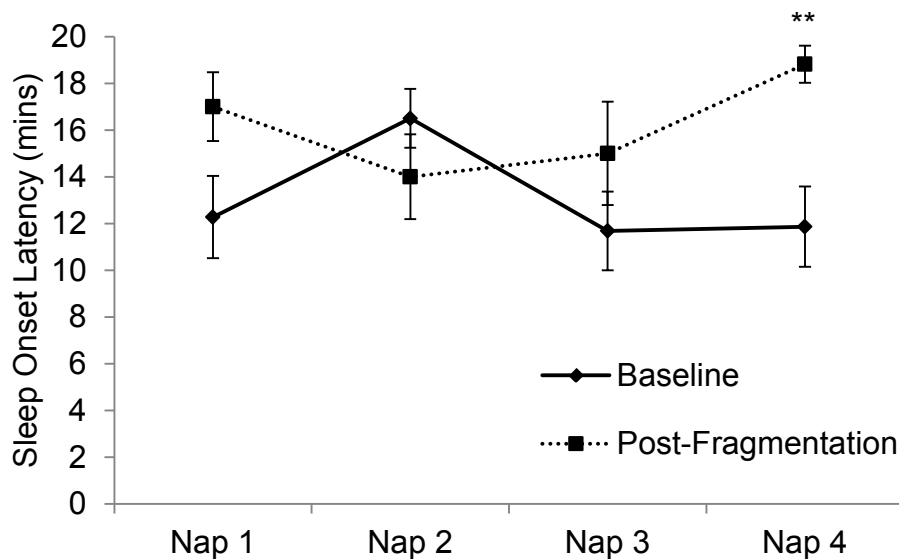
**=p<.01

Because **Figure 7** indicates some participants had increases and others had decreases in MSLT scores, participants change score (baseline MSLT – post-sleep fragmentation MSLT) was compared to selected demographics (income, birth control pill use, M-E questionnaire, and baseline TST) to determine whether certain individual differences were driving differences in MSLT changes. Correlations for continuous measures (income, M-E scores, baseline TST) and a t-test (for the categorical measure use of oral contraception) revealed no relation between any of these demographic variables and the change in MSLT scores.

Results for Research Question #4

RQ#4: How does a single night of experimentally-simulated postpartum sleep disturbance impact the next week's sleep, performance, and mood among healthy nulliparous women?

Actigraphy Total Sleep Time

Six participants took daytime naps during the study; for these participants, 24-hour sleep time was used. Specifically, one participant napped on two days during the baseline week, two participants napped on two days during the recovery week, and three participants napped on one day during the recovery week.

In order to test the hypotheses that *experimental sleep disruption will cause increased actigraphy-measured total sleep time on the proceeding at-home recovery night, compared to baseline at-home total sleep time, and total sleep time will decrease to baseline levels after the first recovery night*, first a repeated-measures ANOVA with total sleep time as the dependent variable and time since beginning of the study until the first in-lab day (baseline) was conducted to determine whether total sleep time changed across the baseline week. There was no significant change in total sleep time across the baseline week, $F[6]=.55$, $p=.77$, $\eta^2_p =.05$ (see **Figure 9**). These data were averaged and a paired samples t-test was used to test for differences in the average total sleep time between baseline and the total sleep time the night after the sleep fragmentation protocol. The results did not support the hypothesis; there was not a statistically significant change in total sleep time between the baseline average ($M=461$, $SD=33$) and the night after sleep fragmentation ($M=463$, $SD=83$; $t[10]=.12$, $p=.91$,

d=.03). Given the null findings, the second part of the hypothesis that *total sleep time will decrease to baseline levels after the first recovery night*, was not tested.

Figure 9. Total sleep time across baseline and post-fragmentation weeks (Error bars represent SE).
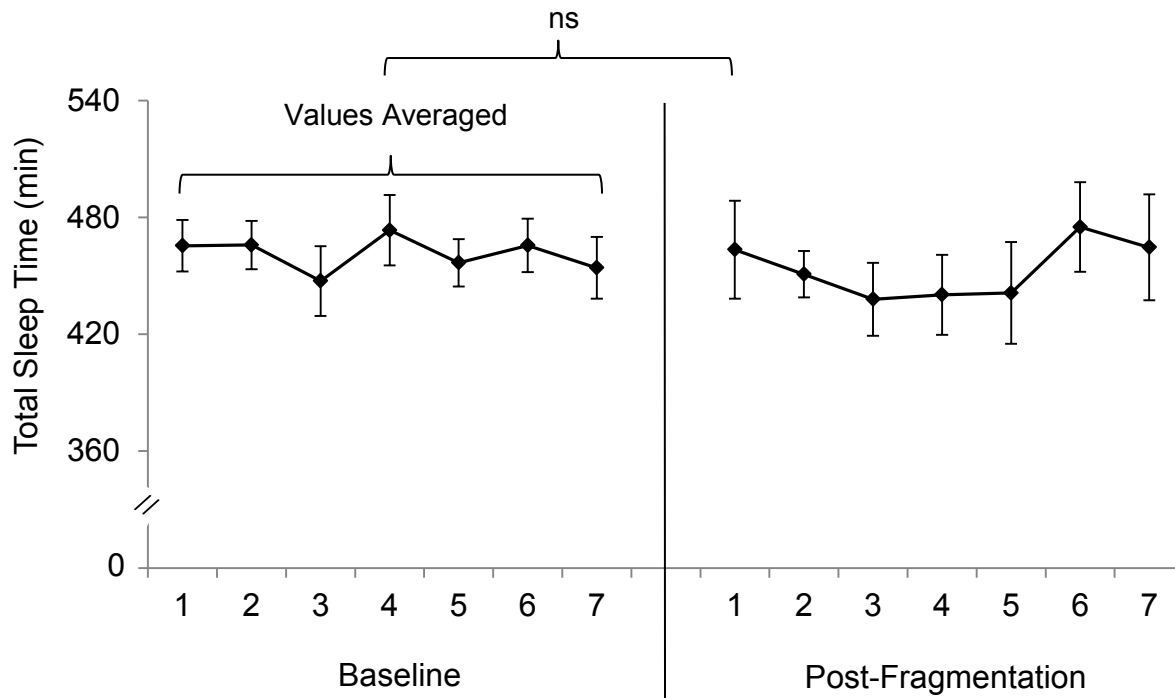
Performance

In order to test the hypotheses that *experimental sleep disruption will cause worse performance on a psychomotor vigilance test (PVT) on the morning after experimental sleep fragmentation compared to baseline condition, and will gradually improve to baseline levels across the recovery week*, first two repeated-measures ANOVAs with frequency of PVT lapses as the dependent variable and either time from beginning of the study until the first in-lab day (baseline) or time since sleep disruption as the independent variable were conducted to determine whether frequency of lapses

changed across baseline and recovery weeks. There was no significant change in frequency of PVT lapses across the baseline (F[6]=2.72, p=.08, η^2_p =.23) or recovery weeks (F[7]=1.31, p=.26, η^2_p =.12) (see **Figure 10**).

Given the lack of change in frequency of PVT lapses within each of the baseline and recovery weeks, the data were averaged and a paired samples t-test was used to test for differences in the average frequency of PVT lapses between baseline and recovery weeks. By using the averages of values across baseline and recovery weeks, the comparisons are more robust to daily performance variations within a week. The results supported a reduction in performance after sleep fragmentation; there was a statistically significant increase in frequency of PVT lapses (*M*=2.72, *SD*=1.76) compared to the baseline average (*M*=1.62, *SD*=1.83; *t*[10]=2.98, *p*=.01, *d*=.61).

Figure 10. Frequency of PVT lapses across baseline and post-fragmentation weeks (Error bars represent SE).
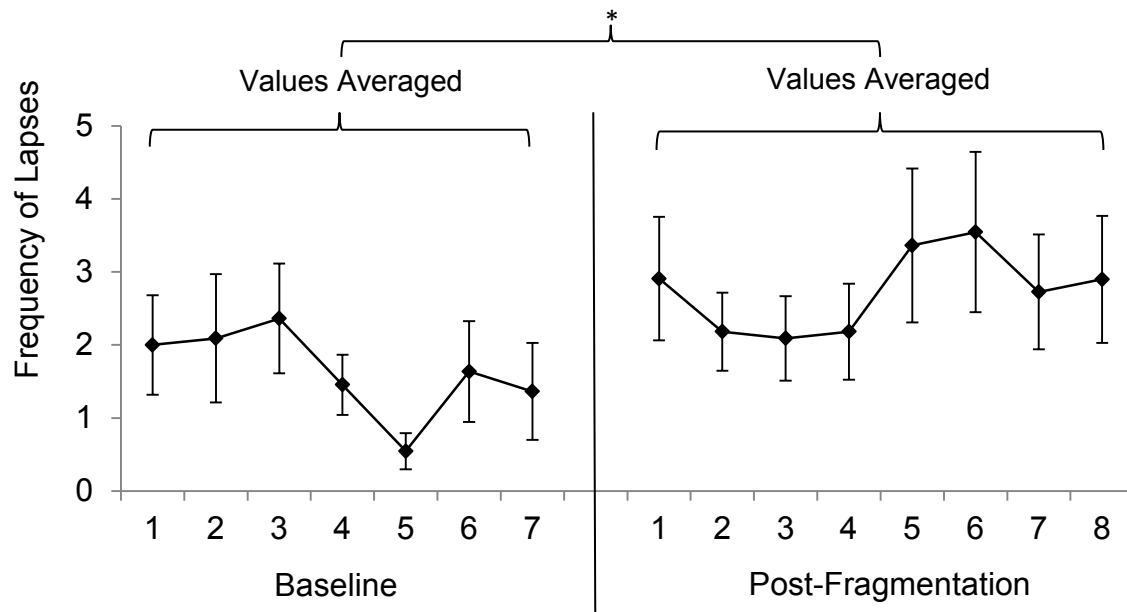
Mood

In order to test the hypotheses that e*xperimental sleep disruption will cause higher POMS total mood disturbance scores the day after experimental sleep fragmentation compared to baseline condition, and will gradually improve to baseline levels across the recovery week,* first a repeated-measures ANOVA with POMS TMD as the dependent variable and time since sleep disruption as the independent variable was conducted to determine whether POMS TMD scores changed across the post-fragmentation week. As only one baseline measure was taken for this survey, tests for a change in baseline score were not performed. After a Greenhouse-Geisser correction because the assumption of sphericity was not met, there was not a significant change in

POMS TMD scores across the post-fragmentation week (F[3]=.37, *p*=.65, η^2_p =.04) (see ***Figure 11***).

Given the lack of significant change in POMS TMD scores across the post-fragmentation weeks, those data were averaged and a paired samples t-test was used to test for differences in between baseline POMS TMD score and the average POMS TMD score across the post-fragmentation week. There was a statistically significant increase from baseline POMS TMD scores (*M*=-1.00, *SD*=7.10) compared to the post-fragmentation week average (*M*=8.55, *SD*=12.9; *t*[10]=2.40, *p*=.037, *d*=.92) (see ***Figure 11***).

Figure 11. POMS TMD between baseline and after experimental sleep fragmentation (Error bars represent SE).
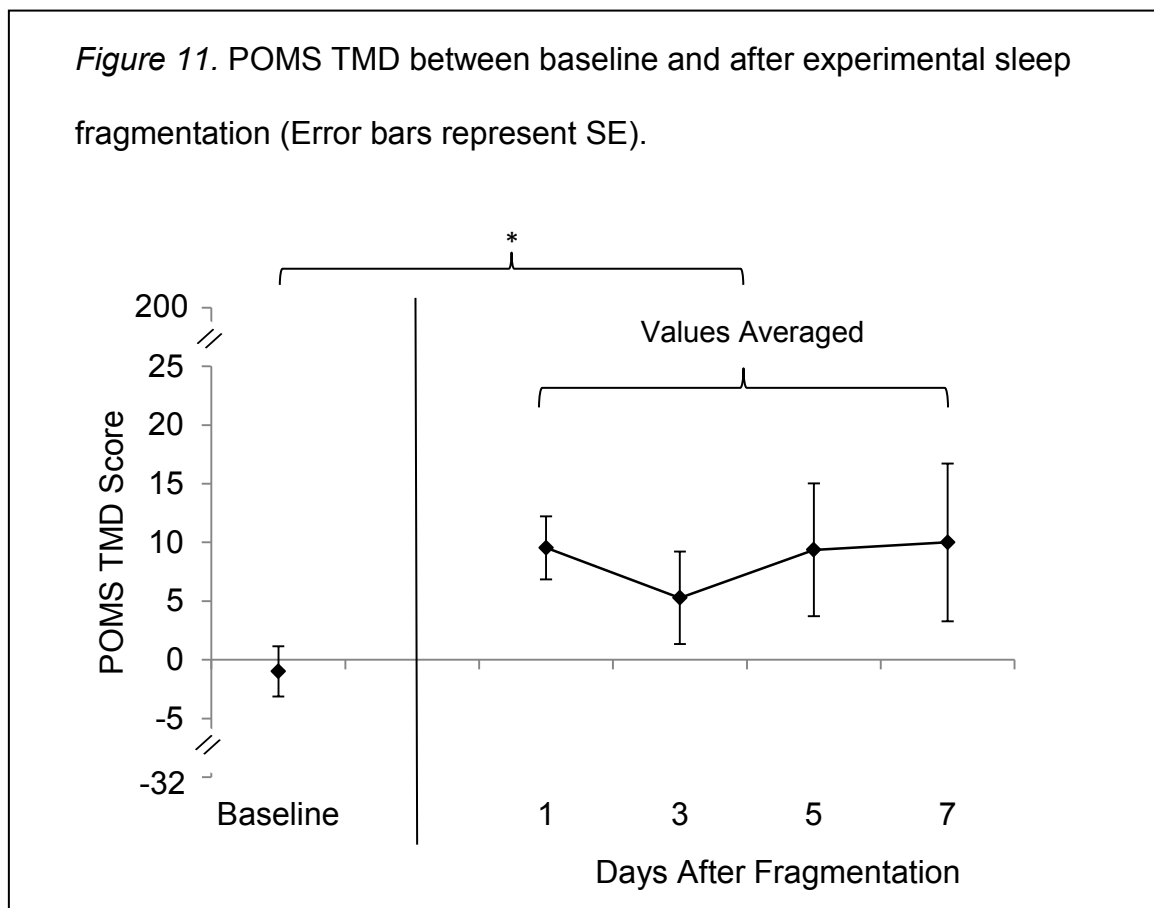
Research Question #4: Supplemental Analyses

While analyzing the actigraphy sleep recovery data, there was a trend observed in napping behavior, with more frequent naps observed after sleep fragmentation. Therefore, the prevalence of napping before and after sleep fragmentation was analyzed statistically. A McNemar's test was conducted to determine whether there was a change in number of participants who napped before and after experimental sleep fragmentation. During the baseline week, 9.1% of participants napped compared to 45.5% of participants who napped during the recovery week, however, this was not a statistically significant difference, $p=.22$, Cramer's V=.29. Of the naps taken during the recovery week, 42.9% were taken the day after the recovery night from the MSLT.

After finding significant differences on the POMS total mood disturbance scale between baseline and the average of POMS scores post-fragmentation, paired samples t-tests were conducted on each of the seven POMS subscales to understand specifically which components of mood were affected by the sleep fragmentation. There was a significant increase in the fatigue subscale ($t[10]=2.38$, $p=.04$, $d=.73$) and decrease in the vigor subscale ($t[10]=4.26$, $p=.002$, $d=1.59$) after sleep fragmentation compared to baseline. There was no significant change on the tension, depression, anxiety, or confusion subscales (see **Figure 12**).

Figure 12. POMS subscale scores at baseline and the average of subscales after sleep fragmentation (Error bars represent SE).
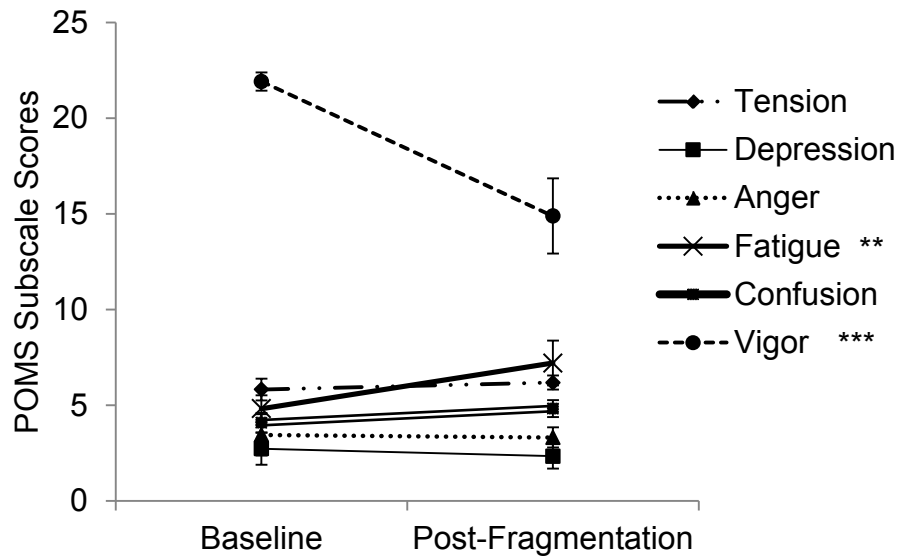
p<.05; **p*<.01

Planned Supplemental Analyses

Comparison to Postpartum Data

There were changes to PVT performance and mood in the current study, so these data were compared to existing postpartum PVT performance and mood at the earliest recording period.

PVT Performance

To determine whether there were any performance differences between early postpartum women and nulliparous women in the current study, an existing database with frequency of PVT lapses averaged across postpartum week 2 among primiparous women were compared to the average lapse frequency post-experimentally induced

sleep fragmentation in the current study's sample. The lapse frequency for primiparous women was positively skewed, so was square root transformed. Levene's test was significant, so equal variances were not assumed. An independent t-test indicated the lapse frequency was significantly greater for postpartum women (*M*=5.80, *SD*=4.69) than nulliparous women in the current study (*M*=2.72, *SD*=1.76; *t*[25.2]= 3.32, *p*=.003, *d*=.87).

Mood

To determine whether there were any mood differences between early postpartum women and nulliparous women in the current study, existing data of POMS TMD scores at postpartum week 3 (the earliest time POMS was administered) among primiparous women were compared to the average POMS TMD scores post-experimentally induced sleep fragmentation in the current study's sample. The POMS TMD scores for primiparous women were positively skewed, so these data were square root transformed. POMS TMD scores were not significantly different between postpartum women (*M*=21.4, *SD*=24.4) and nulliparous women in the current study across the recovery week (*M*=8.55, *SD*=12.9; *t*[77]= 1.71, *p*=.09, *d*=.66).

Discussion

The primary goal of this study was to understand the discrete impacts of postpartum sleep disturbance on sleep architecture, mood, circadian rhythm amplitude, daytime sleepiness, and recovery. The average age of the current sample (25.4 years) was consistent with the average age of first time mothers in the U.S. (25.0 years) (Laughlin, 2011). However, there were fewer women who were married or cohabitating

(36.4%) compared to first time mothers in the U.S. (76.3%) (Martinez, Daniels, & Chandra, 2012). Furthermore, this sample was highly educated, with 100% of participants earning at least a Bachelor's degree, compared to 24% of all mothers in the U.S. (Laughlin, 2011).

A functional model of postpartum sleep fragmentation was established; women had significantly lower sleep efficiency on the sleep fragmentation night compared to baseline. The current sample's PSG-recorded average sleep efficiency (74.4%) was slightly lower than the actigraphically-recorded sleep efficiency of women in the second week postpartum (79.7%) (Montgomery-Downs et al., 2010). In an attempt to isolate the impacts of sleep fragmentation from total sleep time, the time in bed of each participant from baseline to sleep fragmentation night was preserved. This resulted in a total sleep time that was only slightly higher than that previously reported among postpartum women (M=7.46hr±0.56, M=7.2hr±0.95, respectively, Cohen's d=0.33).

Results indicated no changes in 6-sulphatoxymelatonin (aMT6), time spent in nocturnal sleep stages, and total sleep time after sleep fragmentation compared to baseline. There was a significant decrease in daytime sleepiness after fragmentation compared to baseline, specifically on the fourth and final MSLT nap opportunity. Performance and mood decrements were seen the week after sleep fragmentation compared to baseline. Interpretations of these findings are described in respective sections below.

Melatonin

The averages of aMT6 concentration among the current sample are consistent with overnight aMT6 concentrations reported in previous studies. The current sample had pre- and post- concentrations of 23.3 ng/ml (*SD*=13.5) and 25 ng/mg (*SD*=12.2), respectively, which compares well to creatinine adjusted 6-SMT concentrations found in premenopausal women (*M*=28.5 ng/mg, *SD*=19) (Schernhammer et al., 2004).

The concentration of aMT6 among the current sample did not change between baseline and sleep fragmentation nights, a finding that did not support the hypothesized decrease in aMT6 concentration on the sleep fragmentation night compared to baseline. However, the hypothesis was based on differences in melatonin levels among postpartum women (Thomas & Burr, 2006) and other populations who experienced fragmented sleep (Hernandez et al., 2007; Sack et al., 2007) in naturalistic settings, not in a highly controlled laboratory. Kavčič and colleagues (2011) did measure the response of melatonin in the laboratory, but they used an experimental sleep _deprivation_ protocol, not sleep _fragmentation,_ and their subjects were exposed to light at night, whereas the current protocol was careful to only use a very dimly lit environment in order to parse out the effects of the fragmentation from the light exposure.

Conversely, results were consistent with recent literature that fragmented the sleep of healthy _men_ for one night, with preserved total sleep time, and found no changes in plasma melatonin concentration (Gonnissen, Hursel, Rutters, Martens, & Westerterp-Plantenga, 2013). This study also utilized a sleep fragmentation protocol that had longer periods of consolidated sleep (90 mins of sleep before participants were

awakened); however, the awakenings were very brief (about 2 minutes – enough time for the participant to confirm awakening by shutting off an alarm). There were no changes in posture or light exposure during these awakenings.

Based on the existing literature, there are two distinct but possible conclusions that could be drawn to interpret the findings. The first is that the associations previously found between fragmented sleep and reduction in concentration of melatonin is largely due to environment influences, such as light at night, or postpartum hormonal changes. The second is that multiple nights of sleep fragmentation are necessary in order to observe the physiological changes in melatonin. It is likely that a combination of these factors account for some of the suppression of melatonin found among sleep fragmented populations.

There are only a couple of studies that have examined how sleep patterns change during the postpartum period, and findings are inconclusive. The lack of existing evidence is likely due to the difficulties in the longitudinal data collection from pre-pregnancy, pregnancy, and the postpartum period that would adequately address this question; such a longitudinal study has never been done. Wolfson, Crowley, Anwer, & Bassett (2003) show no changes in bedtime, but later rise times by an average of 52 minutes between the end of pregnancy and 2-4 weeks postpartum. This supports the current study's protocol of keeping bedtimes constant, but extending rise times. Recently, using a small sample (n=12) of women who had a history of major depression disorder, Sharkey, Pearlstein, & Carskadon (2013) characterized changes in circadian phase from the third trimester of pregnancy to the 6th postpartum week. There was an average phase delay of 42 minutes from pregnancy to postpartum; six women had

phase delays >1 hour. However, there were no changes in actigraphically-recorded bedtimes and wake times, in contrast to the findings of Wolfson and colleagues. Further research in the change of sleep periods and circadian phase during the postpartum period is warranted.

Sleep Architecture

The current study's sleep fragmentation protocol resulted in no changes in any of the nocturnal sleep stages. This finding counters the literature on experimental sleep fragmentation among healthy subjects. Previous studies have reliably demonstrated sleep fragmentation increases in stage N1 and decreases in stage N3 and/or REM (Bonnet, 1985; Roehrs et al.,1994). However, these studies have used sleep fragmentation protocols that simulate sleep-fragmenting disorders such as obstructive sleep apnea. In these protocols, participants are awakened every couple of minutes for only brief moments of time, throughout the entire night. At the time of the proposal, the current study was the first that was known to experimentally simulate longer periods of consolidated sleep, with longer periods of wake. However, recently one other group created a sleep fragmentation protocol that allowed for longer periods of consolidated sleep. Gonnissen et al. (2013) interrupted the sleep of healthy males about every 90 minutes throughout the night (5 awakenings in total). However, the awakenings were brief; as long as it took participants to turn off an alarm (about 2 min). The results showed a decrease in stage REM sleep at the expense of an increase in stage N2 sleep. This protocol was the most similar to that used in the current study, but still took advantage of more frequent awakenings. It is possible current results did not show

changes in time spent in stage N3 or REM sleep because there were too few

awakenings.

While working as the night technician for the sleep fragmentation night, ALM

noticed a trend in real-time of decreasing sleep onset latencies after each of the three

scheduled awakenings. This observation provided the impetus for analyzing the

supplemental data of the changes in sleep onset latencies from each awakening across

the night. The statistical analyses supported this observation, indicating subjects have a

higher sleep propensity after each progressive awakening. This effect of time of night on

sleep propensity has been previously reported in sleep fragmentation protocols where

participants were awakened every two hours (Helmus et al., 1996; Nykamp et al.,

1999). Furthermore, in their seminal study, Dijk and Czeisler (1995) used a forced

desynchrony protocol of a 28 hour rest-activity cycle in an environment free of time cues

to study the circadian regulation of sleep propensity independent of the homeostatic

sleep drive. Sleep propensity was heightened during 2200 and 0600 hours, regardless

of when the sleep period occurred. The current findings provide further support for a

circadian modulation (Process C) of sleep propensity in the context of the two-process

model of sleep regulation. As the latency to sleep onset was at a time of night when the

most hours of sleep were accrued, it is not reasonable to assume that only the

homeostatic sleep drive (Process S: the regulation of sleep propensity on the basis of

prior wake time) regulates sleep propensity. The interaction between the two processes,

the circadian timing and homeostatic sleep drive, explains the consolidated nocturnal

sleep period. The homeostatic sleep drive is prominent in the first few hours of nocturnal

sleep, weakening across the night as more sleep is accrued, at which point the

circadian sleep drive predominantly takes over, allowing sleep to continue (Dijk & von Schantz, 2005).

Daytime Sleepiness

Results did not support an increase in objective daytime sleepiness after sleep fragmentation. Conversely, women had a decrease in daytime sleepiness, and while this decrease was not significant, it was supported by a large effect size (Cohen's *d*=0.92). However, both the average baseline and post-fragmentation MSLT scores were >10, a score that is considered normative among a healthy population (Afifi et al., 2005). So while the scores non-significantly increased, the meaningfulness of the change in scores from a clinical perspective in diagnosing daytime sleepiness is negligible.

The result of no difference in objective sleepiness before and after sleep fragmentation was unexpected when taken in the context of the literature on the impact of sleep fragmentation with daytime sleepiness. Sleep disorders that are characterized by sleep fragmentation cause daytime sleepiness (Benbadis, 2005). Furthermore, experimental sleep fragmentation among healthy populations in a study design that carefully extended sleep periods in accordance with increased nocturnal wakefulness to ensure total sleep time did not differ, caused significantly decreased MSLT scores by 4.5 mins after a single night of fragmentation (Roehrs, 1994). However, this study fragmented sleep with an auditory tone every 2 minutes, a procedure that altered sleep architecture; participants spent significantly more time in the lighter stages of sleep

(stages N1 and N2) and less time in stage N3 and REM sleep. This change in sleep architecture is something the current procedure did not achieve.

One potential explanation for the inconsistency between current results and previous literature of the impact of sleep fragmentation on daytime sleepiness is there were no changes in stage N3 sleep in the current study. The reduction in percentage of the sleep period spent in stage N3 in previous studies may explain the associated increase in daytime sleepiness in the aforementioned studies. There were no changes in total sleep time, but decreases in stage N3 sleep and MSLT scores when sleep was experimentally fragmented to cause a transient increase in blood pressure or heart rate but no visible EEG arousal (Martin, Wraith, Deary, & Douglas, 1997). When gaboxadol, a stage N3-enhancing drug, was administered prior to each night of a 4-night sleep restriction protocol (5 hours of sleep each night), total sleep time did not differ between the placebo and gaboxadol groups, but stage N3 was significantly enhanced in the gaboxadol group on each night. While both placebo and gaboxadol groups had decreased MSLT scores from baseline to after the 4-night sleep restriction protocol, the decrease was significantly less for the gaboxadol compared to placebo (Walsh et al., 2008). Taken in context, given the current findings of minimal changes in sleep architecture, it is likely not surprising there were no effects on MSLT scores.

However, there is also support for the idea that the effect of sleep fragmentation on daytime sleepiness is mediated by the disturbance of sleep continuity as opposed to changes in sleep stages. The results of a clustered experimental sleep fragmentation (disturbances every 30 sec for 30 min, followed by 60 min uninterrupted sleep) protocol indicated greater portions of time spent in stage N3 sleep compared to subjects whose

sleep was fragmented every 90 sec throughout the night. However, there were no differences in MSLT scores between the clustered or regular sleep fragmentation, despite the differences in sleep architecture (Martin, Brander, Deary, & Douglas, 1999). Similarly, another study used a sleep fragmentation protocol in which participants' sleep was fragmented for two nights approximately 70 times each night. Results showed no difference in sleep architecture between the baseline and experimental nights, however, there was a significant decrease in MSLT scores (Stepanski, Lamphere, Roehrs, Zorick, & Roth, 1987). Interpretation of the current results lend support to either the importance of alterations to sleep stages in causing daytime sleepiness or the necessity of more frequent nocturnal awakenings to cause sufficient interruption of sleep continuity to reduce daytime sleepiness.

After spending 3 consecutive nights in the laboratory, the post-fragmentation MSLT was the final laboratory-based procedure in the protocol. It cannot be discounted that participants may have been less likely to fall asleep because they were excited they were nearing the end of the study. On the fourth and final nap of the post-fragmentation MSLT, 81.8% of participants did not fall asleep. This compares to 27.3% who did not fall asleep on the final nap of the baseline MSLT. While this likely does not explain the lack of decrease in MSLT scores as a result of sleep fragmentation, it may explain the significant increase in MSLT scores after sleep fragmentation.

Post-Fragmentation Week

Total Sleep Time

Contrary to the hypothesis, nocturnal total sleep time monitored for one week post-sleep fragmentation showed no difference in total sleep time the night immediately after sleep fragmentation compared to baseline. There was also no change in total sleep time across the post-fragmentation week. Supplemental data analysis on naps found no statistically significant difference in naps between baseline and recovery weeks, and the majority of the naps taken occurred the day after the first recovery night from sleep fragmentation. It's possible that the lack of change in sleep stems from the participants' instructions to sleep ad libitum during the recovery week, compared to the consistent sleep schedule with at least 8 hours in spent in bed each night attempting to sleep during baseline. This was not required during the recovery week, so it is unknown whether participants merely did not physiologically need to extend their sleep after sleep fragmentation, or they chose not to extend their sleep (for potentially social or work reasons).

Performance

Neurocognitive performance, assessed each morning of the study protocol, showed a significant difference from pre- to post-fragmentation, lending support to the hypothesis of a decrease in neurocognitive performance after sleep fragmentation despite no difference in total sleep time. These results provide causal evidence that postpartum-like sleep fragmentation has a negative impact on performance independent of total sleep time. There was no trajectory of recovery among these participants;

statistically, the PVT values did not change across the post-fragmentation week. As Banks et al. (2010) reported, a 10 hour time in bed protocol the night after chronic sleep restriction was insufficient to return performance to baseline values. If participants in the current study did not allow for enough time devoted to sleep after the sleep fragmentation night, their performance data may be a reflection of a buildup of sleep debt that has not been adequately repaid.

Supplemental analyses compared performance of the current sample of nulliparous women with primiparous women in the second postpartum week and found postpartum women had significantly worse performance than nulliparous women. This difference is most likely an effect of the impact of cumulative sleep disturbance on performance among postpartum women (Insana, Williams, & Montgomery-Downs, 2013), as the postpartum women were already in their second postpartum week. Furthermore, women experience sleep disturbances during the third trimester (Wilson et al., 2011; Wolfson et al., 2003), which means they are already entering the postpartum period with sleep debt, as opposed to the current sample who were well rested prior to their sleep fragmentation night.

Mood

Results indicate that mood, as scored through the POMS total mood disturbance, worsens after experimental sleep fragmentation. At baseline, participants had an average score of -1.00 (lower scores are indicative of better mood), which rose to an average of 8.55 across the post-fragmentation week. Supplemental analyses comparing these data to POMS scores of healthy postpartum women 2 weeks after birth found no

significant difference between groups. However, the p-value was approaching

significance and there was a moderate effect size (Cohen's d=0.66) trending towards

postpartum women reporting worse mood. Yet, these differences may not be a function

of differences between postpartum women and non-postpartum controls, as another

validation study of the POMS on a healthy population of 208 adult women found

averages POMS scores of 20.3 (SD=33.1) (Nyenhuis, Yamamoto, Luchetta, Terrien, &

Parmentier, 1999).

The substantially lower baseline mood scores of the women in the current study

compared to the healthy sample may be an effect of age; in the Nyenhuis et al. (1999)

study, the women were on average 44 years (range 18-94) compared to the current

study's 25 years (range 23-30). Previous literature using the POMS to measure mood

report age-related declines in mood states (Gibson, 1997). Alternatively (or additionally),

the lower baseline scores may be an effect of sleep, as chronic sleep restriction can

cause mood disturbances (Dinges et al., 1997). The women in the current sample were

screened to be healthy sleepers, and they averaged 7 hours 41minutes of total sleep

time during baseline, approximately 20 minutes more than that obtained by 25-year olds

(Ohayon, Carskadon, Guilleminault, & Vitiello, 2004). They also obtained consistent

sleep periods during the baseline week, another aspect of sleep that has been found to

improve mood (Takasu, Takenaka, Fujiwara, & Toichi, 2012).

The supplemental analyses suggest it may not be the obvious dimensions of

mood (e.g. depression, anxiety) that are impacted. When subscales were analyzed,

there were significant changes in fatigue (increased) and vigor (decreased) post-

fragmentation. However, no significant changes were noted in tension, depression,

anxiety, or confusion. This is consistent with previous work that found chronic sleep restriction impacts POMS total mood disturbance scores, and specifically subscales of fatigue, vigor, confusion, and tension (Dinges et al., 1997). However, it is inconsistent with the postpartum depression literature; based on the occurrence of postpartum depression, there would be an expected increase in the depression subscale after one night of sleep fragmentation. As this was not the case, it is speculated the relation between sleep and postpartum depression is a result of the longer-term sleep fragmentation postpartum women experience (that was not modeled in the current study), potentially combined with sleep quality during pregnancy (Wilson et al., 2011; Wolfson et al., 2003). Environmental factors, such as low levels of social support or a recent stressful event (Robertson et al., 2004) or changes in concentrations of sex steroids after parturition (Russell et al., 2000) may also be contributing factors of postpartum mood disturbances and warrant further research.

Limitations

The study had a number of limitations, including the small sample size. Although the sample size was consistent with previous literature that have similar experimental sleep fragmentation protocols, it limited analyzing whether individuals who did have changes in dependent variables from baseline to sleep fragmentation nights differed from those who did not have changes in dependent variables on demographic measures, for instance. Individual differences such as these may give insight into why some women were more affected by the sleep fragmentation protocol than others.

Another limitation may have been the rigorous selection criteria. While these criteria were put in place to reducing confounding factors on the outcome variables of interest, there were a large proportion of women who were excluded from continued participation because they did not meet minimum scores on the MSLT (n=4). However, their baseline actigraphy sleep appeared healthy. Participants who were more resistant to the effects of sleep disturbances may have been unintentionally selected. Those who were excluded may be more sensitive to the effects of sleep disturbances, and may have been impacted more by the sleep fragmentation.

Finally, the awakenings may not have been long enough to model the early postpartum period. While the protocol followed the available literature in guiding the number and duration of awakenings, recent unpublished findings from our lab found mothers with very young infants (<1 month) self-reported an average of 3.6 awakenings each night, and each awakening was an average duration of 55 minutes. This is over 20 minutes longer than the protocol used in the current study. These unpublished data also found that as the infant ages, the duration of the awakenings shortens. However, it supports early postpartum longer nocturnal awakenings.

Future Directions

Future research should expand upon this sleep fragmentation protocol to determine what factors may contribute to the impairment that was generally not seen in this study. Some additions to the protocol could include multiple nights of sleep fragmentation in an attempt to assess the effects of a chronic postpartum sleep fragmentation as opposed to an acute focus.

Furthermore, variations in nocturnal activities and lighting levels could be assessed to determine their impact on physiological measures and performance outcomes. The current protocol carefully kept subjects out of light levels that could disrupt their melatonin levels and the focus of the awakenings was solely on infant caregiving. This was in order to isolate the impact of the sleep fragmentation. Future studies should work to understand how the differences in activities and behaviors postpartum women engage in may be negatively impacting their sleep. Unpublished data from our lab indicate 81.5% of women reported using an electronic device (television, computer, cell phone, backlit tablet, or a combination of these) during nocturnal awakenings and 89% of women are using at least one extra light source during caregiving, but their light sources vary widely in intensity. Examining the impact of these behaviors, both in field-based and laboratory settings, are important next steps to understanding maternal sleep disturbances.

Finally, more research should be directed towards the impact of postpartum changes in physiology on sleep and circadian rhythms. Early evidence suggests hormonal changes in postpartum women may be contributing to changes in circadian rhythms and sleep architecture. Feeding method may be moderating some of the hormonal changes, as breast-feeding promotes the release of prolactin, a hormone strongly associated with sleep. However, much of the impact of postpartum physiological changes on sleep remains unclear.

Conclusion

A complex process of physiological and environmental changes during the postpartum period confounds our understanding of the discrete impacts of postpartum sleep fragmentation. The present study sought to isolate the effects of postpartum sleep fragmentation on sleep architecture, nocturnal melatonin, mood, daytime sleepiness, performance, and recovery by manipulating the sleep of childless women in the laboratory to model postpartum sleep fragmentation. The postpartum reduction in sleep efficiency with no change to total sleep time was successfully modeled. No differences were found in nocturnal melatonin concentration or time spent in sleep stages between baseline and sleep fragmentation nights. A significant decrease in daytime sleepiness after fragmentation compared to baseline was observed, specifically on the fourth and final MSLT nap opportunity. There was no change in total sleep time the night after sleep fragmentation. Sleep fragmentation did cause performance and mood decrements across the week post-sleep fragmentation compared to baseline. Results suggest no physiological changes of a single night of simulated postpartum sleep fragmentation, but significant deficits in mood and neurobehavioral performance. These findings suggest that the mere disruption of sleep continuity in the absence of the measured physiological changes is sufficient to cause poorer mood and performance, stressing the importance of examining both subjective and objective measures in sleep fragmented populations.

This study expands the literature by providing a basic understanding of the acute effects of *postpartum* sleep fragmentation. It was also the first to model this sleep disturbance using childless women, establishing the feasibility of using childless women

in future studies to assess chronic sleep fragmentation or alterations in the nocturnal

environment, such as the impact of light and electronic use at night.

References

Acebo, C., & LeBourgeois, M. K. (2006). Actigraphy. *Respiratory Care Clinics of North America, 12, 23*-30.

Afifi, L., Kushida, C.A., Carskadon, M.A. (2005). Multiple sleep latency test. In C.A. Kushida (p 11-24). Sleep deprivation: clinical issues, pharmacology, and sleep loss.

Agnew, H.W., Webb, W.B., & Williams, R.L. (1966). The first night effect: an EEG study of sleep. *Psychophysiology, 2,* 263-266.

American Academy of Sleep Medicine (2005). The International Classification of Sleep Disorders. Westchester, IL: Author.

Anderson, G. (2010). The role of melatonin in post-partum psychosis and depression associated with bipolar disorder. *Journal of Perinatal Medicine, 38,* 585-587.

Ansara, D., Choen, M.M., Gallop, R., Kung, R., & Schei, B. (2005). Predictors of women's physical health problems after childbirth. *Journal of Psychosomatic Obstetrics & Gynecology, 26,* 115-125.

Arand, D., Bonnet, M., Hurwitz, T., Mitler, M., Rosa, R., & Sangal, B. (2005). The clinical use of the MSLT and MWT. *Sleep, 28,* 123-144.

Axelsson, J., Kecklund, G., Åkerstedt, T., Donofrio, P., Lekander, M., & Ingre, M. (2008). Sleepiness and performance in response to repeated sleep restriction and subsequent recovery during semi-laboratory conditions. *Chronobiology International, 25,* 297-308.

Banks, S., Van Dongen, H.P.A., Maislin, G., & Dinges, D.F. (2010). Neurobehavioral dynamics following chronic sleep restriction: Dose-response effects of one night for recovery. *Sleep, 33,* 1013-1026.

Basner, M. & Dinges, D.F. (2011). Maximizing sensitivity of the psychomotor vigilance test (PVT) to sleep loss. *Sleep, 34,* 581–91.

Benbadis, S. R. (2005). Questionnaires and rating scales. In. C. A. Kushida (Ed.) *Lung biology in health and disease*: Vol. 193, pp. 1-9, *Sleep deprivation: Clinical Issues, Pharmacology, and Sleep Loss Effects*. New York, NY: Marcel Dekker.

Benington, J.H., Kodali, S.K., & Heller, H.C. (1995). Stimulation of A_1 adenosine receptors mimics the electroencephalographic effects of sleep deprivation. *Brain Research, 692,* 79-85.

Benloucif, S., Burgess, H.J., Klerman, E.B., Lewy, A.J., Middleton, B., & Murphy, P.J. (2008). Measuring melatonin in humans. *Journal of Clinical Sleep Medicine, 4,* 66-69.

Benoit, O., Foret, J., Bouard, G., Merle, B., & Landau, J. (1980). Habitual sleep length and patterns of recovery sleep after 24 hour and 36 hour sleep deprivation. *Electroencephalography and Clinical Neurophysiology, 50,* 477-485.

Benson, K., Friedman, L., Noda, A., Wicks, D., Wakabayashi, E., & Yesavage, J. (2004). The measurement of sleep by actigraphy: Direct comparison of 2 commercially available actigraphs in a nonclinical population. *Sleep, 27,* 986-989.

Bloch, M., Rubinow, D.R., Schmidt, P.J., Lotskias, A., Chrousos, G.P., & Cizza, G.

(2005). Cortisol response to ovine corticotrophin-releasing hormone in a model of

pregnancy and parturition in euthymic women with and without a history of

postpartum depression. *The Journal of Clinical Endocrinology & Metabolism, 90,*

695-699.

Bloch, M., Schmidt, P.J., Danaceau, M., Murphy, J., Nieman, L., & Rubinow, D.R.

(2000). Effects of gonadal steroids in women with a history of postpartum

depression. *American Journal of Psychiatry, 157,* 924-930.

Bojkowski, C.J., Aldhous, M.E., English, J., Franey, C., Poulton, A.L., Skene, D.J., &

Arendt, J. (1987). Suppression of nocturnal plasma melatonin and 6-

sulphatoxymelatonin by bright and dim light in man. *Hormone and Metabololic*

Research, 19, 437-440.

Bonnar, J., Franklin, M., Nott, P.N., & McNeilly, A.S. (1975). Effect of breast-feeding on

pituitary-ovarian function after childbirth. *British Medical Journal*, *4*, 82-84.

Bonnet, M.H. (1985). Effect of sleep disruption on sleep, performance, and mood.

Sleep, 8, 11-19.

Borbély, A.A. (1982). Two process model of sleep regulation. *Human Neurobiology, 1,*

195-204.

Borjigin, J., Zhang, L.S., Calinescu, A-A. (2012). Circadian regulation of pineal gland

rhythmicity. *Molecular and Cellular Endocrinology, 349,* 13-19.

Brage Hudson, D., Elek, S.M., Fleck, M.O. (2001). First-time mothers' and fathers' transition to parenthood: Infant care self-efficacy, parenting satisfaction, and infant sex. *Issues in Comprehensive Pediatric Nursing, 24,* 31-43.

Cajochen, C., Jewett, M.E., & Dijk, D-J. (2003). Human circadian melatonin rhythm phase delay during a fixed sleep-wake schedule interspersed with nights of sleep deprivation. *Journal of Pineal Research, 35,* 149-157.

Cajochen, C., Münch, M., Kobialka, S., Kräuchi, K., Steiner, R., Oelhafen, P., . . . Wirz-Justice, A. (2005). High sensitivity of human melatonin, alertness, thermoregulation, and heart rate to short wavelength light. *The Journal of Clinical Endocrinology & Metabolism, 90,* 1311-1316.

Carskadon, M.A. & Dement, W.C. Daytime sleepiness: Quantification of a behavioral state. *Neuroscience & Biobehavioral Reviews, 11,* 307-317.

Cheng, C.Y. & Li, Q. (2008) Integrative review of research on general health status and prevalence of common physical health conditions of women after childbirth. *Women's Health Issues, 18,* 267-280.

Coelho, F.M.S., Narayansingh, M., & Murray, B.J. (2011). Testing sleepiness and vigilance in the sleep laboratory. *Current Opinion in Pulmonary Medicine, 17,* 406-411.

Cooper, P.J. & Murray, L. (1995). Course and recurrence of postnatal depression. Evidence for the specificity of the diagnostic concept. *The British Journal of Psychiatry, 166,* 191-195.

Czeisler, C.A., Duffy, J.F., Shanahan, T.L., Brown, E.N., Mitchell, J.F., Rimmer, D.W., et al. (1999). Stability, precision, and near-24-hour period of the human circadian pacemaker. *Science, 284,* 2177-2181.

Davanipour, Z., Poulsen, H.E., Weimann, A., & Sobel, E. (2009). Endogenous melatonin and oxidatively damaged guanine in DNA. *BMC Endocrine Disorders, 9(22)* Retrieved from: http://www.biomedcentral.com/1472-6823/9/22.

De Carvalho, M., Klaus, M.H., & Merkatz, R.B. (1982). Frequency of breast-feeding and serum bilirubin concentration. *American Journal of Diseases of Children, 136,* 737-738.

de Souza, L., Benedito-Silva, A. A., Pires, M. L., Poyares, D., Tufik, S., & Calil, H. M. (2003). Further validation of actigraphy for sleep studies. *Sleep, 26*, 81-85.

de Weerd A.W. & van den Bossche, R.A.S. (2003). The development of sleep during the first months of life. *Sleep Medicine Reviews, 7,* 179-191.

Deacon, S. & Arendt, J. (1994). Posture influences melatonin concentrations in plasma and saliva in humans. *Neuroscience Letters, 167,* 191-194.

Dijk, D.J., Groeger, J.A., Stanley, N., & Deacon, S. (2010). Age-related reduction in daytime sleep propensity and nocturnal slow wave sleep. *Sleep, 33,* 211-223.

Dinges, D. F., Pack, F., Williams, K., Gillen, K. A., Powell, J. W., Ott, G. E., . . . Pack, A.I. (1997). Cumulative sleepiness, mood disturbance, and psychomotor vigilance performance decrements during a week of sleep restricted to 4-5 hours per night. *Sleep 20,* 267-277.

Dijk, D-J. & Czeisler, C.A. (1995). Contribution of the circadian pacemaker and the sleep homeostat to sleep propensity, sleep structure, electroencephalographic slow waves, and sleep spindle activity in humans. *The Journal of Neurosciences, 15(5)*, 3526-3538.

Dijk, D-J. & von Schantz, M. (2005). Timing and consolidation of human sleep, wakefulness, and performance by a symphony of oscillators. *Journal of Biological Rhythms, 20,* 279-290.

Doornbos, B., Fokkema, D.S., Molhoek, M., Tanke, M.A.C., Postema, F., & Korf, J. (2009). Abrupt rather than gradual hormonal changes induce postpartum blues-like behavior in rats. *Life Sciences, 84,* 69-74.

Dørheim, S.K., Bondevik, G.T., Eberhard-Gran, M., & Bjorvatn, B. (2009). Sleep and depression in postpartum women: a population-based study. *Sleep, 32,* 847–55.

Driver, H.S. & Shapiro, C.M. (1992). A longitudinal study of sleep stages in young women during pregnancy and postpartum. *Sleep, 15,* 449-453.

Duffy, J.F., Cain, S.W., Chang, A.M., Phillips, A.J.K., Münch, M.Y., Gronfier, C., . . . Czeisler, C.A. (2011). Sex difference in the near-24-hour intrinsic period of the human circadian timing system. *Proceedings of the National Academy of Sciences, 108,* 15602-15608.

Durmer, J.S. & Dinges, D.F. (2005). Neurocognitive consequences of sleep deprivation. *Seminars in Neurology, 25,* 117-129.

Eastman CI, Mistberger RE, Rechtschaffen A. Suprachiasmatic nuclei lesions eliminate circadian temperature and sleep rhythms in the rat. Physiol Behav 1984;32:357-368.

Edinger, J.D., Means, M.K., Stechuchak, K.M., & Olsen, M.K. (2004). A pilot study of inexpensive sleep-assessment devices. *Behavioral Sleep Medicine, 2,* 41-49.

Faul, F., Erdfelder, E., Lang, A.G., & Buchner, A. (2007). G*Power 3: A flexible statistical power analysis program for the social, behavioral, and biomedical sciences. *Behavior Research Methods, 39,* 175-191.

Field, T. (2010). Postpartum depression effects on early interactions, parenting, and safety practices: A review. *Infant Behavior and Development, 33,* doi:10.1016/j.infbeh.2009.10.005.

Figueiredo, B. & Conde, A. (2011). Anxiety and depression in women and men from early pregnancy to 3-months postpartum. *Archives of Women's Mental Health, 14,* 247-255.

Fredholm, B.B., Bättig, K., Holmén, J., Nehlig, A., & Zvarau, E.E. (1999). Actions of caffeine in the brain with special reference to factors that contribute to its widespread use. *Pharmacological Reviews, 51,* 83-133.

Ganguly, S., Coon, S.L., & Klein, D.C. (2002). Control of melatonin synthesis in the mammalian pineal gland: the critical role of serotonin acetylation. *Cell and Tissue Research, 309*, 127-137.

Gay, C.L., Lee, K.A., & Lee, S. (2004). Sleep patterns and fatigue in new mothers and fathers. *Biological Research for Nursing, 5,* 311-318.

Geisler, P., Tracik, F., Crönlein, T., Fulda, S., Wichniak, A., Popp, R., . . . Hajak, G. (2006). The influence of age and sex on sleep latency in the MSLT-30 – A normative study. *Sleep, 29,* 687-692.

Gibson, S.J. (1997). The measurement of mood states in older adults. *Journal of Gerontology: Psychological Sciences, 52B(4),* 167-174.

Gillette, M.U. & Abbott, S.M. (2005). Fundamentals of the circadian system in Basics of Sleep Guide, M. Opp (ed.), Sleep Research Society, 131-138.

Gonnissen, H.K.J., Hursel, R., Rutters, F., Martens, E.A.P., & Westerterp-Plantenga, M.S. (2013). Effects of sleep fragmentation on appetite and related hormone concentrations over 24 h in healthy men. *British Journal of Nutrition, 109,* 748-756.

Goodman, J.H. (2004). Postpartum depression beyond the early postpartum period. *Journal of Obstetric, Gynecologic, & Neonatal Nursing, 33,* 410-420.

Gooley, J.J., Chamberlain, K., Smith, K.A., Khalsa, S.B.S., Rajaratnam, S.M.W., Van Reen, E., . . . Lockley, S.W. (2011). Exposure to room light before bedtime suppresses melatonin onset and shortens melatonin duration in humans. *The Journal of Clinical Endocrinology & Metabolism, 96,* E463-E472.

Groër, M., Davis, M., Casey, K., Short, B., Smith, K. & Groër, S. (2005). Neuroendocrine & immune relationships in postpartum fatigue. *The American Journal of Maternal/Child Nursing, 30,* 133-138.

Helmus, T., Rosenthal, L., Bishop, C., Roehrs, T., Krsevska, S., & Roth, T. (1996). Nocturnal sleep latencies among alert, alert-deprived and sleepy subjects. *Electroencephalography and Clinical Neurophysiology, 99,* 10-15.

Hernández, C., Abreu, J., Abreu, P., Castro, A., & Jiménez, A. (2007). Nocturnal melatonin plasma levels in patients with OSAS: the effect of CPAP. *European Respiratory Journal, 30,* 496-500.

Herrmann, U.S., Hes, C.W., Guggisberg, A.G., Roth, C., Gugger, M., & Mathis, J. (2010). Sleepiness is not always perceived before falling asleep in healthy, sleep-deprived subjects. *Sleep Medicine, 11,* 747-751.

Horowitz, J.A., Murphy, C.A., Gregory, K.E., Wojcik, J. (2009). Best practices: community-based postpartum depression screening: results from the CARE study. *Psychiatric Services, 60,* 1432-1434.

Hung, C.H. (2004). Predictors of postpartum women's health status. *Journal of Nursing Scholarship, 36,* 345-351.

Hunter, L.P., Rychnovsky, J.D., & Yount, S.M. (2009). A selective review of maternal sleep characteristics in the postpartum period. *Journal of Obstetric, Gynecologic, & Neonatal Nursing, 38*, 60-68.

Ikegami, K., Ogyu, S., Arakomo, Y., Suzuki, K., Mafune, K., Hiro, H., & Nagata, S. (2009). Recovery of cognitive performance and fatigue after one night of sleep deprivation. *Journal of Occupational Health, 51,* 412-422.

Insana, S.P. (2010). Sleep and sleepiness among first-time postpartum parents. West Virginia University. ATT: 3420901.

Insana, S.P. & Montgomery-Downs, H.E. (2012). Sleep and sleepiness among first-time postpartum parents: A field- and laboratory-based multimethod assessment. *Developmental Psychobiology,* DOI 10.1002/dev.21040.

Insana, S.P., Williams, K.B., & Montgomery-Downs, H.E. (2013). Sleep disturbance and neurobehavioral performance among postpartum women. *Sleep, 36,* 73-81.

Insana, S.P., Garfield, C.F., & Montgomery-Downs, H.E. (in press). A mixed-method examination of maternal and paternal nocturnal caregiving. *Journal of Pediatric Health Care.* Retrieved from: http://dx.doi.org/10.1016/j.pedhc.2013.07.016

Jackson, M.L., Croft, R.J., Kennedy, G.A., Owens, K., & Howard, M.E. (2013). Cognitive components of simulated driving performance: Sleep loss effects and predictors. *Accident Analysis & Prevention, 50,* 438-444.

Kanotra, S., D'Angelo, D., Phares, T.M., Morrow, B., Barfield, W.D., & Lansky, A. (2007). Challenges faced by new mothers in the early postpartum period: An analysis of comment data from the 2000 pregnancy risk assessment monitoring system (PRAMS) survey. *Maternal and Child Health Journal, 11,* 549-558.

Kaushal, N., Ramesh, V., Gozal, D. (2012). TNF-α and temporal changes in sleep architecture in mice exposed to sleep fragmentation. *PLOS One, 7,* e45610.

Kavčič, P., Rojc, B., Dolenc-Groselj, L., Claustrat, B., Fujs, K., & Poljak, M. (2011). The

impact of sleep deprivation and nighttime light exposure on clock gene expression

in humans. Croatian Medical Journal 2011;52:594-603.

Kilduff, T.S., Kushida, C.A., & Terao, A. (2005). Recovery from Sleep Deprivation. In

C.A. Kushida (p 485-98). Sleep deprivation: clinical issues, pharmacology, and

sleep loss.

Klante, G., Brinschwitz, T., Secci, K., Wollnik, F., & Steinlechner, S. (1997). Creatinine

is an appropriate reference for urinary sulphatoxymelatonin of laboratory animals

and humans. *Journal of Pineal Research, 23,* 191-197.

Kleitman, N. & Engelmann, T.G. (1953). Sleep characteristics of infants. *Journal of

Applied Physiology, 6,* 269-282.

Klem, G.H., Luders, H.O., Jasper, H.H., & Elger, C. (1999). The ten-twenty electrode

system of the International Federation. The International Federation of Clinical

Neurophysiology. *Electroencephalography and Clinical Neurophysiology

Supplement, 52,* 3-6.

Krueger, J.M. (2008). The role of cytokines in sleep regulation. *Current Pharmaceutical

Design, 14,* 3408-3416.

Kryger, M. H., Roth, T., & Dement, W. C. (2011). Normal human sleep: An overview. In

Principles and practice of sleep medicine (5th ed., pp. 16-18). St. Louis: Elsevier

Saunders.

Lam, J., Barr, R.G., Catherine, N., Tsui, H., Hahnhaussen, C.L., Pauwels, J., . . . Brant, R. (2010). Electronic and paper diary recording of infant and caregiver behaviors. *Journal of Developmental & Behavioral Pediatrics, 31,* 685-693.

Laughlin, L. (2011). Maternity leave and employment patterns: 2006-2008. *Current Population Report,* P70-128. U.S. Census Bureau, Washington, DC.

Leahy-Warren, P. (2007). Social support for first-time mothers: An Irish study. *The American Journal of Maternal/Child Nursing, 32,* 368-374.

Lim, J. & Dinges, D.F. (2008). Sleep deprivation and vigilant attention. *Annals of the New York Academy of Sciences, 1129,* 305–22.

Littner, M.R., Kushida, C., Wise, M., Davila, D.G., Morgenthaler, T., Lee-Chiong, T., . . . Kramer, M. (2005). Practice parameters for clinical use of the multiple sleep latency test and the maintenance of wakefulness test. *Sleep, 28,* 113-121.

Loh, S., Lamond, N., Dorrian, J., Roach, G., & Dawson, D. (2004). The validity of psychomotor vigilance tasks of less than 10-minute duration. *Behavior Research Methods Instruments & Computers, 36,* 339–46.

Manber, R. & Armitage, R. (1999). Sex, Steroids, and Sleep: A Review. *Sleep, 22,* 540-555.

Martin, S.E., Brander, P.E., Deary, I.J., & Douglas, N.J. (1999). The effect of clustered versus regular sleep fragmentation on daytime function. *Journal of Sleep Research, 8,* 305-311.

Martin, S.E., Engleman, H.M., Deary, I.J., & Douglas, N.J. (1996). The effect of sleep

fragmentation on daytime function. *American Journal of Respiratory and Critical

Care Medicine, 153,* 1328-1332.

Martin, S.E., Wraith, P.K., Deary, I.J., & Douglas, N.J. (1997). The effect of nonvisible

sleep fragmentation on daytime function. *American Journal of Respiratory and

Critical Care Medicine, 155,* 1596-1601.

Martinez, G., Daniels, K., & Chandra, A. (2012). Fertility of men and women aged 15-44

years in the United States: National survey of family growth, 2006-2010. *National

Health Statistics Reports, 51,* Hyattsville, MD: National Center for Health Statistics.

Marty, M., Rozenberg, S., Duplan, B., Thomas, P., Duquesnoy, B., & Allaert, F. (2008).

Quality of sleep in patients with chronic low back pain: a case-control study.

European Spine Journal, 17, 839-844.

Matthews, C.D., Guerin, M.V., & Wang, X. (1991). Human plasma melatonin and urinary

6-sulphatoxymelatonin: studies in natural annual photoperiod and in extended

darkness. *Clinical Endocrinology, 35,* 21-27.

Matsumoto, K., Shinkoda, H., Kang, M.J., & Seo, Y.J. (2003) Longitudinal study of

mothers' sleep-wake behaviors and circadian time patterns from late pregnancy to

postpartum. *Biological Rhythm Research, 34,* 265-278.

McCoy, S.J.B. (2011). Postpartum depression: An essential overview for the

practitioner. *Southern Medical Journal, 104,* 128-132.

McCoy, J.G. & Strecker, R.E. (2011). The cognitive cost of sleep lost. *Neurobiology of Learning and Memory, 96,* 564-582.

McCracken, L.M. & Iverson, G.L. (2002). Disrupted sleep patterns and daily functioning in patients with chronic pain. *Pain Research & Management, 7,* 75-79.

McKenna, J.T., Tartar, J.L., Ward, C.P., Thakkar, M.M., Cordeira, J.W., McCarley, R.W. et al. (2007). Sleep fragmentation elevates behavioral, electrographic and neurochemical measures of sleepiness. *Neuroscience, 146,* 1462-1473.

McNair, D.M., Lorr, M., & Droppleman, L.F. (1992). *Manual: Profile of Mood States.* San Diego, CA: Educational and Industrial Testing Services.

Mendlewicz, J. (2009). Disruptions of the circadian timing systems: Molecular mechanisms in mood disorders. *CNS Drugs, 23,* 15-26

Milgrom, J., Gemmill, A.W., Bilszta, J.L., Hayes, B., Barnett, B., Brooks, J., . . . Buist, A. (2008). Antenatal risk factors for postnatal depression: a large prospective study. *Journal of Affective Disorders, 108,* 147–157.

Mitler, M.M., van den Hoed, J., Carskadon, M.A., Richardson, G., Park, R., Guilleminault, C., & Dement, W.C. (1979). REM sleep episodes during the multiple sleep latency test in narcoleptic patients. *Electroencephalography and Clinical Neurophysiology, 46,* 479-481.

Molik, E., Misztal, T., Romanowicz, K., & Zieba, D.A. (2010). The effects of melatonin on prolactin and growth hormone secretion in ewes under different photoperiods, during the early post partum period. *Small Ruminant Research, 94,* 137-141.

Montgomery-Downs, H.E., Insana, S.P., Clegg-Kraynok, M.M., & Mancini, L.M. (2010). Normative longitudinal maternal sleep: the first 4 postpartum months. *American Journal of Obstetrics & Gynecology, 203,* 465e1-7.

Morgenthaler, T., Alessi, C., Friedman, L., Owens, J., Kapur, V., Boehlecke, B., . . . Swick, T.J. (2007). Practice parameters for the use of actigraphy in the assessment of sleep and sleep disorders: an update for 2007. *Sleep, 30,* 519–29.

Mosendane, T., Mosendane, T., Raal, F.J. (2008). Shift work and its effects on the cardiovascular system. *Cardiovascular Journal of Africa, 19,* 210-215.

Müller, S., Hemmi, M.H., Wilhelm, F.H., Barr, R.G., & Schneider, S. (2011). Parental report of infant sleep behavior by electronic versus paper-and-pencil diaries, and their relationship to actigraphic sleep measurement. *Journal of Sleep Research, 20,* 598-605.

Nishihara, K. & Horiuchi, S. (1998). Changes in sleep patterns of young women from late pregnancy to postpartum: Relationships to their infants' movements. *Perceptual & Motor Skills, 87,* 1043-1056.

Nishihara, K., Horiuchi, S., Eto, H., & Uchida, S. (2002). The development of infants' circadian rest-activity rhythm and mothers' rhythm. *Physiology & Behavior, 77,* 91-98.

Nishihara, K., Horiuchi, S., Eto, H., Uchida, S., & Honda, M. (2004). Delta and theta power spectra of night sleep EEG are higher in breast-feeding mothers than in non-pregnant women. *Neuroscience Letters, 268,* 216-220.

Nyenhuis, D.L., Yamamoto, C., Luchetta, T., Terrien, A., & Parmentier, A. (1999). Adult
and geriatric normative data and validation of the profile of mood states. *Journal of
Clinical Psychology, 55,* 79-86.

Nykamp, K., Rosenthal, L., Helmus, T., Gerhardstein, R., Day, R., Roehrs, . . . & Roth,
T. (1999). Repeated nocturnal sleep latencies in narcoleptic, sleepy and alert
subjects. *Clinical Neurophysiology, 110,* 1531-1534.

Ohayon, M.M., Carskadon, M.A., Guilleminault, C., & Vitiello, M.V. (2004). Meta-
analysis of quantitative sleep parameters from childhood to old age in healthy
individuals: Developing normative sleep values across the human lifespan. *Sleep,
27(7)*, 1255-1273.

Pang, S.F., Tang, P.L., Tang, G.W., Yam, A.W., & Ng, K.W. (1987). Plasma levels of
immunoreactive melatonin, estradiol, progesterone, follicle stimulating hormone,
and beta-human chorionic gonadotropin during pregnancy and shortly after
partutition in humans. *Journal of Pineal Research, 4,* 21-31.

Parry, B.L., Meliska, C.J., Sorenson, D.L., Lopez, A.M., Martinez, L.F., Nowakowski, S.,
. . . Kripke, D.F. (2008). Plasma melatonin circadian rhythm disturbances during
pregnancy and postpartum in depressed women and women with personal or family
histories of depression. *American Journal of Psychiatry, 165,* 1551-1558.

Pearlstein, T., Howard, M., Salisbury, A., & Zlotnick, C. (2009). Postpartum depression.
American Journal of Obstetrics & Gynecology, 200, 357-364.

Philip, P., Sagaspe, P., Lagarde, E., Leger, D., Ohayon, M.M., Bioulac, B., . . . Taillard, J. (2010). Sleep disorders and accidental risk in a large group of regular registered highway drivers. *Sleep Medicine, 11,* 973-979.

Pittendrigh, C.S. & Daan, S. (1976). A functional analysis of circadian pacemakers in nocturnal rodents IV Entrainment: Pacemaker as Clock. *Journal of Comparative Physiology, 106,* 291-331.

Popa, C., Netea, M.G., van Riel, P.L.C.M., van der Meer, J.W.M., & Stalenhoef, A.F.H. (2007). The role of TNF-α in chronic inflammatory conditions, intermediary metabolism, and cardiovascular risk. *Journal of Lipid Research, 48,* 751-762.

Porkka-Heiskanen, T., Strecker, R.E., & McCarley, R.W. (2000). Brain site-specificity of extracellular adenosine concentration changes during sleep deprivation and spontaneous sleep: An in vivo microdialysis study. *Neuroscience, 99,* 507-517.

Porkka-Heiskanen, T., Strecker, R.E., Thakkar, M., Bjørkum, A.A., Greene, R.W., & McCarley, R.W. (1997). Adenosine: A mediator of the sleep-inducing effects of prolonged wakefulness. *Science, 276,* 1265-1268.

Portas, C.M., Thakkar, M., Rainnie, D.G., Greene, R.W., & McCarley, R.W. (2007). Role of adenosine in behavioral state modulation: A microdialysis study in the freely moving cat. *Neuroscience, 79,* 225-235.

Posmontier, B. (2008). Sleep quality in women with and without postpartum depression. *Journal of Obstetric, Gynecologic, and Neonatal Nursing, 37,* 722–35.

Radloff, L.S. (1977). Center for Epidemiological studies depression scale. *Applied Psychological Measurement,* 1, 385-401.

Ralph, M.R., Foster, R.G., Davis, F.C., & Menaker, M. (1990). Transplanted suprachiasmatic nucleus determines circadian period. *Science, 247*, 975-978.

Ramesh, V., Nair, D., Zhang, S.X.L., Hakim, F., Kaushal, N., Kayali, F., . . . Gozal, D. (2012). Disrupted sleep without sleep curtailment induces sleepiness and cognitive dysfunction via the tumor necrosis factor-α pathway. *Journal of Neuroinflammation, 9,* 91.

Reid, K.J. & Zee, P.C. (2009). Circadian rhythm disorders. *Seminars in Neurology, 29,* 393-405.

Reppert, S.M. & Weaver, D.R. (2001). Molecular analysis of mammalian circadian rhythms. *Annual Review of Physiology, 63*, 647-676.

Righetti-Veltema, M., Conne-Perreard, E., Bousquet, A., & Manzano, J. (2002). Postpartum depression and mother-infant relationship at 3 months old. *Journal of Affective Disorders, 20,* 291-306.

Roach, G.D., Lamond, N., Dorrian, J., Burgess, H., Holmes, A., Fletcher, A., . . . Dawson, D. (2005). Changes in the concentration of urinary 6-sulphatoxymelatonin during a week of simulated night work. *Industrial Health, 43,* 193-196.

Robertson, A., Grace, S., Wallington, T., & Stewart, D.E. (2004). Antenatal risk factors for postpartum depression: a synthesis of recent literature. *General Hospital Psychiatry, 26,* 289-295.

Roehrs, T., Merlotti, L., Petrucelli, N., Stepanski, E., & Roth, T. (1994). Experimental sleep fragmentation. *Sleep, 17,* 438-443.

Roenneberg, T., Daan, S., & Merrow, M. (2003). The art of entrainment. *Journal of Biological Rhythms, 18,* 183-194.

Rohrbeck, J., Jordan, K., & Croft, P. (2007). The frequency and characteristics of chornic widespread pain in general practice: a case-control study. *British Journal of General Practice, 57,* 109-115.

Ross, L.E., Murray, B.J., Steiner, M. (2005). Sleep and perinatal mood disorders: a critical review. *Journal of Psychiatry and Neuroscience, 30,* 247-256.

Russell, J.A., Douglas, A.J., & Ingram, C.D. (2000). Brain preparations for maternity – adaptive changes in behavioral and neuroendocrine systems during pregnancy and lactation. An overview. *Progress in Brain Research, 133,* 1-38.

Sack, R.L., Auckley, D., Auger, R.R., Carskadon, M.A., Wright, K.P., Vitiello, M.V., & Zhdanova, I.V. (2007). Circadian rhythm sleep disorders: Part II, advanced sleep phase disorder, delayed sleep phase disorder, free-running disorder, and irregular sleep-wake rhythm. *Sleep, 30(11),* 1484-1501.

Sadeh, A. (2011). The role and validity of actigraphy in sleep medicine: an update. *Sleep Medicine Reviews, 15,* 259–267.

Sallinen, M., Holm, A., Hiltunen, J., Hirvonen, K., Härmä, M., Koskelo, J., . . . Müller, K. (2008). Recovery of cognitive performance from sleep debt: Do a short rest pause and a single recovery night help? *Chronobiology International, 25,* 279-296.

Sánchez, C.L., Cubero, J., Sánchez, J., Chanclón, B., Rivero, M., Rodríguez, A.B., & Barriga, C. (2009). The possible role of human milk nucleotides as sleep inducers. *Nutritional Neuroscience, 12,* 2– 8.

Sassin, J.F., Frantz, A.G., Weitzman, E.D., & Kapen, S. (1972). Human prolactin: 24-hour pattern with increased release during sleep. *Science, 177,* 1205-1207.

Santiago, J. R., Nolledo, M. S., Kinzler, W., Santiago, T. V. (2001). Sleep and sleep disorders in pregnancy. *Annals of Internal Medicine, 134,* 396-408.

Schernhammer, E.S., Rosner, B., Willett, W.C., Laden, F., Colditz, G.A., & Hankinson, S.E. (2004). Epidemiology of urinary melatonin in women and its relation to other hormones and night work. *Cancer Epidemiology, Biomarkers & Prevention, 13(6),* 936-943.

Sharkey, K.M., Pearlstein, T.B., & Carskadon, M.A. (2013). Circadian phase shifts and mood across the perinatal period in women with a history of major depressive disorder: A preliminary communication. *Journal of Affective Disorders, 150,* 1103-1108.

Song, J.E., Chang, S.B., Park, S.M., Kim, S., & Nam, C.M. (2010). Empirical test of an explanatory theory of postpartum fatigue in Korea. *Journal of Advances Nursing, 66,* 2627-2639.

Stepanski, E., Lamphere, J., Roehrs, T., Zorick, F., & Roth, T. (1987). Experimental sleep fragmentation in normal subjects. *International Journal of Neuroscience, 33,* 207-214.

Stone, A.A., Shiffman, S., Schwartz, J.E., Broderick, J.E., & Hufford, M.R. (2002). Patient non-compliance with paper diaries. *British Medical Journal, 324,* 1193-1194.

Swain, A. M., O'Hara, M. W., Starr, K. R., & Gorman, L. L. (1997). A prospective study of sleep, mood, and cognitive function in postpartum and nonpostpartum women. *Obstetrics and Gynecology,* 90, 381-386.

Takasu, N.N., Takenaka, Y., Fujiwara, M., & Toichi, M. (2012). Effects of regularizing sleep-wake schedules on daytime autonomic function and psychological states in healthy university students with irregular sleep-wake habits. *Sleep and Biological Rhythms, 10,* 84-93.

Thoman, E.B., Barnett, C.R., & Leiderman, P.H. (1971). Feeding behaviors of newborn infants as a function of parity of the mother. *Child Development, 42,* 1471-1483.

Thomas, K.A. & Burr, R.L. (2006). Melatonin level and pattern in postpartum versus nonpregnant nulliparous women. *Journal of Obstetric, Gynecologic, & Neonatal Nursing,* 608-615.

Tulchinsky, D., Hobel, C.J., Yeager, E., & Marshall, J.R. (1972). Plasma estrone, estriol, progesterone, and 17-hydroxyprogesterone in human pregnancy. *American Journal of Obstetrics & Gynelcology, 112,* 1095-1100.

Van den Hoed, J., Kraemer, H., Guilleminault, C., Zarcone, V.P., Miles, L.E., Dement, W.C., & Mitler, M.M. (1981). Disorders of excessive daytime somnolence: Polygraphic and clinical data for 100 patients. *Sleep, 4,* 23-37.

von Truer, K., Norman, T.R., Armstrong, S.M. (1996). Overnight human plasma melatonin, cortisol, prolactin, TSH, under conditions of normal sleep, sleep deprivation, and sleep recovery. *Journal of Pineal Research, 20,* 7-14.

Walsh, J. K., Dement, W. C., & Dinges, D. F. (2005). Sleep medicine, pulbic policy, and

  public health. In M. H. Kryger, T. Roth, & W. C. Dement (Eds.), *Principles and*

  Practice of Sleep Medicine (4th ed., pp. 648-656). Philadelphia, PA: Elsevier

  Saunders.

Walsh, J.K., Snyder, E., Hall, J., Randazzo, A.C., Griffin, K., Groeger, J., . . .

  Schweitzer, P.K. (2008). Slow wave sleep enhancement with gaboxadol reduces

  daytime sleepiness during sleep restriction. *Sleep, 31(5),* 659-672.

Webb, W.B. & Agnew, H.W. (1975). The effects on subsequent sleep of an acute

  restriction of sleep length. *The Society for Psychophysiological Research, 12,* 367-

  370.

Webb, D.A., Block, J.R., Coyne, J.C., Chung, E.K., Bennett, I.M., Culhane, J.F. (2008).

  Postpartum physical symptoms in new mothers: Their relationship to functional

  limitations and emotional well-being. *Birth, 35,* 179-187.

Wehrens, S.M.T., Hampton, S.M., Kerkhofs, M., & Skene, D.J. (2010). Mood, alertness,

  and performance in response to sleep deprivation and recovery sleep in experienced

  shiftworkers versus non-shiftworkers. *Chronobiology International, 29,* 537-548.

West, C.P. & McNeilly, A.S. (1979). Hormonal profiles in lactating and non-lactating

  women immediately after delivery and their relationship to breast engorgement.

  British Journal of Obstetrics & Gynaecology, 86, 501-506.

Wierrani, W., Grin, W., Hlawka, B., Kroiss, A., & Grünberger, W. (1997). Elevated serum

  melatonin levels during human late pregnancy and labour. *Journal of Obstetrics and*

  Gynaecology, 17, 449-451.

Wikner, J., Svanborg, E., Wetterberg, L., & Röjdmark, S. (1997). Melatonin secretion and excretion in patients with obstructive sleep apnea syndrome. *Sleep, 20*, 1002-1007.

Wilson, D.L., Barnes, M., Ellett, L., Permezel, M., Jackson, M., & Crowe, S.F. (2011). Decreased sleep efficiency, increased wake after sleep onset and increased cortical arousals in late pregnancy. *Australian and New Zealand Journal of Obstetrics and Gynaecology, 51,* 38-46.

Winser, M.A. (2012). Early postpartum awakenings, neurocognitive performance, and feeding methods. West Virginia University. ATT: 1520825.

Wolfson, A.R., Crowley, S.J., Anwer, U., & Bassett, J.L. (2003). Changes in sleep patterns and depressive symptoms in first-time mothers: Last trimester to 1-year postpartum. *Behavioral Sleep Medicine, 1(1),* 54-67.

Wulff, K. & Siegmund, R. (2000). Circadian and ultradian time patterns in human behavior: Part 1: Activity monitoring of families from prepartum to postpartum. *Biological Rhythm Research, 31,* 581-602.

Zaharna, M. & Guilleminault, C. (2010). Sleep, noise and health: Review. *Noise & Health, 12,* 64-69.

Zhang, H., Park, Y., Wu, J., Chen, X.P., Lee, S., Yang, J., . . . Zhang, C. (2009). Role of TNF-α in vascular dysfunction. *Clinical Science, 116,* 219-230.

APPENDIX A.

Sleep Disorders Screening Chart

If you are routinely robbed of a good night's rest you may have a sleep disorder. This chart lists symptoms associated with several common sleep problems. For each symptom you have, decide how severely or how frequently it affects you, on a 10 point scale. Then check the chart to see whether you should seek treatment. If you experience two or more symptoms, consider moving up to the next recommendation level.

Symptoms   very mild 1 2 3 4 5 6 7 8 9 10 severe

Symptom										
Gasp, Choke or stop breathing during sleep.										
Snore loudly, have high blood pressure; overweight.										
Feel creepy crawling sensations in legs while lying down.										
Feel tired and sleepy while driving.										
Arms and legs jerk and twitch during sleep.										
Wake up at night; feel fatigued during the day.										
Fall asleep in front of the TV or while reading during the day or early evening.										
Wake up tired and lethargic in the morning.										
Have occasional sleeplessness at home or during trips.										
Experience disturbing nightmares.										

Treatment Needed?   Not necessarily: ◼ (purple)   Recommended: ◼ (orange)   Definitely: ◼ (teal)

Center for Epidemiologic Studies Depression Scale (CES-D)

Center for Epidemiologic Studies Depression Scale (CES-D), NIMH

Below is a list of the ways you might have felt or behaved. Please tell me how often you have felt this way during the past week.

	During the Past Week			
	Rarely or none of the time (less than 1 day)	Some or a little of the time (1-2 days)	Occasionally or a moderate amount of time (3-4 days)	Most or all of the time (5-7 days)
1. I was bothered by things that usually don't bother me.	☐	☐	☐	☐
2. I did not feel like eating; my appetite was poor.	☐	☐	☐	☐
3. I felt that I could not shake off the blues even with help from my family or friends.	☐	☐	☐	☐
4. I felt I was just as good as other people.	☐	☐	☐	☐
5. I had trouble keeping my mind on what I was doing.	☐	☐	☐	☐
6. I felt depressed.	☐	☐	☐	☐
7. I felt that everything I did was an effort.	☐	☐	☐	☐
8. I felt hopeful about the future.	☐	☐	☐	☐
9. I thought my life had been a failure.	☐	☐	☐	☐
10. I felt fearful.	☐	☐	☐	☐
11. My sleep was restless.	☐	☐	☐	☐
12. I was happy.	☐	☐	☐	☐
13. I talked less than usual.	☐	☐	☐	☐
14. I felt lonely.	☐	☐	☐	☐
15. People were unfriendly.	☐	☐	☐	☐
16. I enjoyed life.	☐	☐	☐	☐
17. I had crying spells.	☐	☐	☐	☐
18. I felt sad.	☐	☐	☐	☐
19. I felt that people dislike me.	☐	☐	☐	☐
20. I could not get "going."	☐	☐	☐	☐

SCORING: zero for answers in the first column, 1 for answers in the second column, 2 for answers in the third column, 3 for answers in the fourth column. The scoring of positive items is reversed. Possible range of scores is zero to 60, with the higher scores indicating the presence of more symptomatology.

DRUG USE QUESTIONNAIRE (DAST -10)

NAME: _____   Date:_____

The following questions concern information about your potential involvement with drugs excluding alcohol and tobacco during the past 12 months. Carefully read each countyment and decide if your answer is "YES" or "NO". Then, check the appropriate box beside the question.

When the words "drug abuse" are used, they mean the use of prescribed or over-the-counter medications used in excess of the directions and any non-medical use of any drugs. The various classes of drugs may include but are not limited to: cannabis (e.g., marijuana, hash), solvents (e.g., gas, paints etc…), tranquilizers (e.g., Valium), barbiturates, cocaine, and stimulants (e.g., speed), hallucinogens (e.g., LSD) or narcotics (e.g., Heroin). Remember that the questions do not include alcohol or tobacco.

Please answer every question. If you have difficulty with a countyment, then choose the response that is mostly right.

These questions refer to the past 12 months only.   YES   NO

1. Have you used drugs other than those required for medical reasons?.....

2. Do you abuse more than one drug at a time?..

3. Are you always able to stop using drugs when you want to?.................

4. Have you had "blackouts" or "flashbacks" as a result of drug use?........

5. Do you ever feel bad or guilty about your drug use?..............................

6. Does your spouse (or parent) ever complain about your involvement
    with drugs?..

7. Have you neglected your family because of your use of drugs?.............

8. Have you engaged in illegal activities in order to obtain drugs?.............

9. Have you ever experienced withdrawal symptoms (felt sick) when
    you stopped taking drugs?...

10. Have you had medical problems as a result of your drug use
    (e.g., memory loss, hepatitis, convulsions, bleeding etc…)?..................

* DAST Score…………………………………
* See scoring instructions for correct scoring procedu

SHORT MICHIGAN ALCOHOL SCREENING TEST (SMAST)

NAME: _____   Date:_____

The following questions concern information about your involvement with alcohol during the past 12 months. Carefully read each countyment and decide if your answer is "YES" or "NO". Then, check the appropriate box beside the question.

Please answer every question. If you have difficulty with a countyment, then choose the response that is mostly right.
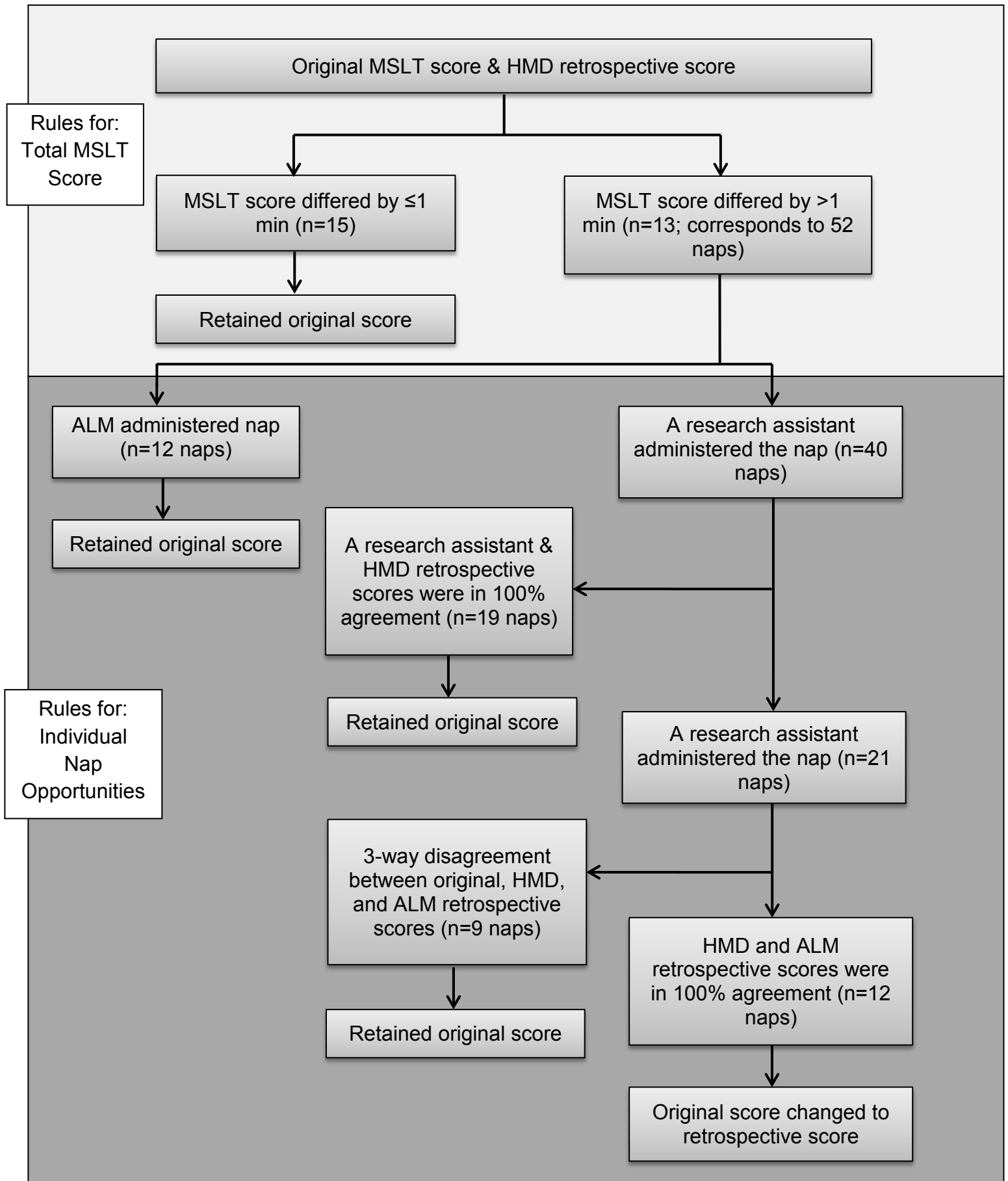
These questions refer to the past 12 months only.	YES	NO
1. Do you feel that you are a normal drinker? (by normal we mean do you drink less than or as much as most other people.)…………………		
2. Does your wife, husband, a parent, or other near relative ever worry or complain about your drinking?...		
3. Do you ever feel guilty about your drinking?................................		
4. Do friends or relatives think you are a normal drinker?..................		
5. Are you able to stop drinking when you want to?...........................		
6. Have you ever attended a meeting of Alcoholics Anonymous (AA)?..............		
7. Has your drinking ever created problems between you and your wife, husband, a parent or other near relative?..............................		
8. Have you ever gotten into trouble at work because of your drinking?..............		
9. Have you ever neglected your obligations, your family, or your work for two or more days in a row because you were drinking?...................		
10. Have you ever gone to anyone for help about your drinking?.........................		
11. Have you ever been in a hospital because of drinking?.................................		
12. Have you ever been arrested for drunken driving, driving while intoxicated, or driving under the influence of alcoholic beverages?................		
13. Have you ever been arrested, even for a few hours, because of other drunken behaviors?..		

* SMAST Score……………………………

* See scoring instructions for correct scoring procedures.

Rules for revisions of MSLT scores

Rules for:
Total MSLT
Score

Original MSLT score & HMD retrospective score

MSLT score differed by ≤1 min (n=15)

MSLT score differed by >1 min (n=13; corresponds to 52 naps)

Retained original score

ALM administered nap (n=12 naps)

A research assistant administered the nap (n=40 naps)

Retained original score

A research assistant & HMD retrospective scores were in 100% agreement (n=19 naps)

Rules for:
Individual
Nap
Opportunities

Retained original score

A research assistant administered the nap (n=21 naps)

3-way disagreement between original, HMD, and ALM retrospective scores (n=9 naps)

HMD and ALM retrospective scores were in 100% agreement (n=12 naps)

Retained original score

Original score changed to retrospective score

MCBEAN'S SPAGHETTI SAUCE

2¼ lbs lean ground beef

2 cans (10 oz.) tomato soup

1 can (14 oz.) Hunt's tomato sauce

2 Tbsps. Lea & Perrins Worcestershire sauce

3-5 drops Tobasco sauce

½ tsp. black pepper

2 tsp. oregano

1 tsp. onion salt

3 cloves garlic, pressed

4 oz. button mushrooms, minimum

3 Tbsps. extra virgin olive oil

1.   In a large frying pan, brown the lean ground beef (at medium to medium high) until there are next to no juices on the bottom of the frying pan.

2.   Combine the tomato soup, the tomato sauce, the Worcestershire sauce, the Tobasco sauce, pepper, oregano, onion salt and garlic into the browned beef.

3.   In a separate smaller frying pan, and usually at the same time or slightly behind the browning of the lean ground beef, sauté the mushrooms (over low heat) in the olive oil. Just brown the mushrooms, do not overcook! Add the mushrooms to the sauce, when ready.

4.   Simmer for about 1 hour. If you want to simmer longer, add small quantities of water when necessary.

Makes 8 servings.