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Circadian Rhythmicity of Mood : An Exploratory Study

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Abstract

Human circadian rhythms are widely observed to fluctuate across the 24-hour circadian period, spanning cognitive, behavioral, and physiological domains. Circadian rhythm (CR) systems, particularly the sleep-wake cycle, are widely studied. Dysregulation of the sleep-wake cycle, common in shift work and mood disorders, diminishes mood regulation, resulting in increased negative mood or inappropriate mood responses. Although emotions have been investigated in the context of circadian variability in the sleep-wake cycle, circadian effects on emotional state per se have infrequently been examined. Previous studies suggest an increase in Positive Affect (PA) and decrease in Negative Affect (NA) as the day progresses, while the reverse occurs in the earlier hours of the day. Our study aimed to investigate circadian variation in PA versus NA, and extend these findings to the specific emotional states of Affection and Annoyance. As part of a larger study, thirteen male participants completed affect assessments using the Brief Mood Introspection Scale (BMIS) seven times over a 24-hour period. Primary findings corroborate previous research finding an increase in PA and decrease in NA during the evening, with the reverse occurring in the morning. Future research should include female participants, longitudinal designs, and objective measures of mood, such as cortisol or testosterone levels, in addition to subjective measures. These findings have clinical relevance, particularly for comparing patients' reported mood ratings with expected ratings based on circadian rhythm of mood. Early-morning NA may reflect normal circadian fluctuations, but late-day NA could indicate a severe clinical condition. In summary, this study replicates circadian patterns in PA and NA but finds unique circadian behaviors in Affection and Annoyance, demanding further exploration.

Keywords: circadian rhythm, mood, sleep-wake cycle

MONTCLAIR STATE UNIVERSITY

Circadian Rhythmicity of Mood: An Exploratory Study

By

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CIRCADIAN RHYTHMICITY OF MOOD: AN EXPLORATORY STUDY

A THESIS

Submitted in partial fulfillment of the requirements

For the degree of Master of Arts

by

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Montclair, NJ

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Introduction

Currently, mood disorders represent a substantial threat to both the well-being of individuals and the economic stability of nations. In 2019, the World Health Organization reported a 13% global increase in psychiatric illnesses since 2010, with mood disorders, including depression and anxiety, affecting millions of people worldwide. Mood disorders, such as depression, grew from 193 million to 246 million people affected worldwide, as well as anxiety, which grew from 298 million to 374 million people in 2020 (Arias et al., 2022). Mood disorders are associated with mental health conditions including neurological, mental, and substance use disorders, suicide risk, as well as psychosocial, intellectual, and cognitive disabilities (WHO, 2019). In the United States, severe mental health conditions have been associated with significant economic burden, with mood disorders accounting for a significant portion of hospitalizations among individuals under 45 years old, excluding pregnancy and childbirth (Duszynski-Goodman & Henderson, 2023). Globally, mood disorders such as major depressive disorder are estimated to cost approximately one trillion USD in lost productivity each year, which includes an estimated 193.2 billion in lost revenue within the U.S. alone (WHO, 2019).

Mood disorders are a heterogeneous class of psychiatric illnesses which include depression and bipolar-related disorders (APA, 2013). Mood disorders demonstrate a complex etiology wherein complex interactions among social, environmental, genetic factors generate symptomatology (McClung, 2013; Lau & Eley, 2010). Notably, the environmental component is consistently implicated in etiological theories and research illustrating the importance of external change associated with features of mood disorder symptomatology. The individual is constantly exposed to cycles of environmental change in factors such as temperature or photoperiod. Across

evolution, entrainment to these cycles improved adaptive fitness became a crucial aspect of maintaining internal homeostasis (Albrecht, 2010). These external factors dictate internal physiological, cognitive, and behavioral processes that promote optimal responses to environmental change as a function of time across a 24-hour period, crucial for human survival.

This system represents the human circadian rhythm (CR), often referred to as the internal physiological "clock," which orchestrates the synchronized pattern of endogenous processes corresponding to external temporal cues. Circadian rhythm regulation spans many domains, controlling processes such as sleep-wake cycle, energy level, body temperature, immune function, and digestion (Walker et al., 2020). The most well-known being the sleep-wake cycle, which requires the alignment of physiological function such as sleep, with environmental cues such as the darkness of nighttime, without interruption, in order to function properly (Walker et al., 2020). Proper sleep-wake cycle function is crucial to emotional regulation and disruption has been linked to emotional volatility, inappropriateness, heightened negative emotion, as well as clinical mood disorders (Fishbein, 2021). Multiple studies underscore the significance of CR as a key regulator in the etiology of mood disorders, and a growing body of evidence links alterations in the sleep-wake cycle to severe adverse health outcomes (Cho et al., 2019; Fishbein et al., 2021).

The circadian theory of mood disorders posits that some mood disorders may stem from the dysregulation of the endogenous circadian system, resulting in irregular function which negatively impacts well-being (Srinivisian et al., 2006). Circadian dysregulation is associated with a range of adverse health conditions, including immunologic, neurodegenerative, psychiatric, and cardiometabolic diseases (Fishbein et al., 2021). Various factors can contribute to the desynchronization between the internal circadian clock and the external environment,

including shiftwork, alterations in the sleep-wake cycle, environmental stressors, and more (Schnell et al., 2014).

The significance of a well-regulated circadian rhythm becomes evident in populations that are especially vulnerable to disruptions in their sleep-wake cycle and display elevated rates of mood disorders compared to the general population. In industrialized nations, a significant proportion of the population engages in shiftwork, comprising approximately 15% of the U.S. workforce and 20% in the European Union (Barger et al., 2012; Eurofound, 2012). Moreover, epidemiological reports demonstrate that certain population groups, such as shift workers, are particularly vulnerable to mood disorders such as depression (Wyse et al., 2017). When the sleep wake cycle is misaligned with the environment, which is often the case for the average, modern day shift worker, the endogenous circadian rhythm becomes dysregulated, with negative downstream consequences in subjective well-being.

This is supported by previous empirical evidence which consistently finds an increased risk of depression in shift worker population as well as an association between night shift work and short- and long-term mental illness (Torquati et al., 2019). Furthermore, multiple epidemiological studies have shown that shift workers, especially those on night shifts, face increased risk of experiencing depression, suicidal tendencies, and mood disorders (Wyse et al., 2017; Bara & Arber, 2009; Weaver et al., 2018). This is compounded by further research demonstrating that shift work is also associated with increased risk of workplace accidents, a factor cited by safety regulation authorities in investigations of industrial disasters like the Three Mile Island incident in 1979, the Chernobyl disaster in 1986, and the Exxon Valdez spill in 1989 (Williamson et al., 2011). This prevalence places a substantial portion of the population at risk, particularly of circadian disruption as well as workplace injury and associated health issues,

including mood disorders. This just begins to highlight the potential link between the rising economic burden of increased mood disorder prevalence and the industrial crises commonly attributed to the shift worker population. It also underscores the significance of circadian rhythms in the context of mood disorders. Therefore, delving into the circadian rhythm of mood is of particular interest when addressing the ongoing global mental health crisis.

Given mounting evidence of an association with severe adverse health conditions, circadian dysregulation emerges as a potentially significant threat to the health and finance of the general public. With the proportion of the U.S. population affected by mental illness being over an estimated 32.1% of adults (NAMI, 2023), enhanced prognosis and innovative circadian-related therapeutic techniques will be crucial for managing symptoms and preventing reoccurrence of mood disorders. Although mood disturbances and alterations in the sleep-wake cycle often co-occur, our understanding of the temporal dynamics of these conditions is limited. Despite the existing body of evidence illustrating the connection between CR and mood disorders, there remains a notable gap in research concerning the patterns of circadian variation for all the moods that a human can experience across the 24-hour circadian cycle. With greater understanding of the pattern of circadian variation in mood, clinicians may be able to improve targeted, time-dependent interventions, diagnostic tools, and improved specificity in therapeutics. To address this gap and contribute to the understanding of the circadian rhythm's impact on mood, we aim to replicate previous research demonstrating circadian variation in mood and extend the investigation on the circadian rhythm of mood to additional mood categories. Ultimately, the present study intends to enrich the existing literature and provide novel insights on the circadian rhythm of mood to aid in addressing the ongoing surge in mood disorder prevalence.

Circadian Rhythm and Mood

Circadian Rhythm is defined as the inherent oscillations of physiological and behavioral patterns occurring over a 24-hour circadian period, regulated by the suprachiasmatic nucleus (SCN) (Fishbein et al., 2021). The SCN maintains internal homeostasis through the synchronization of CR processes which regulate pattern of variation in mood, energy level, body temperature, heart rate, sleep cycle, quality, and duration, among many others, with environmental cues (McClung, 2013). This process of coordination of the body's physiological clock with the pattern of environmental cues from light exposure is referred to as entrainment, or the establishment of an individual CR (Schmal et al., 2020). Generally, the body of existing literature demonstrates that all CR related parameters, organized by the SCN, will follow the same pattern of variation across a 24-hour period unless disrupted by environmental factors, illness, stress, or other external influences (Claudio & Andrea, 2022). The pattern of observed change within a circadian parameter, such as heart rate, across the 24-hour circadian period is referred to as its circadian variation. The pattern of variation demonstrated by CR parameters across 24-hours can be described using terms including peak, or the highest observed numerical value, and trough, or nadir which refers to the lowest observed numerical value (Murray et al., 2020). When the observed values of such parameters are atypical, or the pattern of fluctuation is shifted or delayed, the CR regulated process is said to be dysregulated, or not functioning properly. Oftentimes, circadian dysregulation is caused by a circadian disruption, or interruption of the circadian rhythm cycle and function, which results in an atypical pattern of variation within the affected circadian parameter (Fishbein et al., 2021).

Furthermore, the outcome of atypical variation can include alternations to sleep cycle, mood disturbances, delayed motor activity peaks, and decreased subjective energy levels

(Claudio & Andrea, 2022). In particular, the circadian rhythm is closely linked to the sleep-wake cycle, and a regulated sleep-wake cycle with uninterrupted sleep is critical to effective emotional regulation as well as stable emotional state (McClung, 2013). Interruption to the sleep-wake cycle has been linked to negative emotions, inappropriate emotions, emotional volatility, and to clinical mood disorders (Walker et al., 2020). Previous research has found decreased mood regulation, increased negative mood, and inappropriate moods are all associated with shift work as well as with sleep loss generally (Weaver et al., 2018). This is supported further by prior research in which sleep-deprived individuals reported increased sleepiness, negative affect, coldness, and subjective fatigue (Schwarz et al., 2019).

In parallel with this, circadian rhythms are also closely linked to mood through hormonal regulation and arousal state activation, which ultimately affect the sleep-wake cycle (McClung, 2013). For the purpose of the present study, mood can be defined as a valanced state of being similar to those found in emotions or affect, and encompassing subjective experience, behavioral, and physiological response (Stone et al., 2006). Mood is also often characterized by dimensions such as arousal (active vs. calm) and valence (pleasant vs. unpleasant), which represents the spectrum on which all moods exist (Russel, 1980). Related to this, mood states are defined as discrete affective states with different characteristics, including happy, sad, calm, anxious, loving, angry, and more (Thayer, 1989). Mood can be influenced by subjective experiences, time of day, photoperiod, environment, energy levels, and sleep quality (Sikka & Gross, 2014). Mood is also associated with sleep quality, as evidenced by the impact of circadian rhythm processes (Franzen et al., 2008).

The cumulative body of evidence suggests that mood is profoundly shaped by interactions with different components of the circadian system, encompassing external circadian

modulators (e.g., light exposure, food, and drugs), alterations in clock genes, and behavioral patterns (Claudio & Andrea, 2022). Cyclical variations are also evident in mood and emotional regulation across the 24-hour circadian period. Initially, the natural variation in mood was noticed in cases of unipolar depression, characterized by lower mood levels during the day and improved mood in the evening (Riemann et al., 1991). In non-clinical populations, the consensus of evidence demonstrates a different pattern, with pleasantness to be generally highest during the day and lowest at night, following a similar pattern to energetic arousal, although it is unclear why. While unpleasantness is hypothesized to be lowest during the day and highest at night, the reason for this pattern is currently unclear (Emens et al., 2020). Previous studies which extracted information from the periodic mood cycling exhibited and recorded on Twitter, have generally consistently observed good mood to start in the morning and diminish throughout the day (Golder & Macy, 2011; Dzogang et al., 2017). Recent studies have further defined the change in pleasant mood across 24-hours as a diurnal pattern, although the exact timing of each peak varies between studies (Golder & Macy, 2011). While attempts to define the pattern of change in mood over a 24-hour circadian period has been limited to positive affect and negative affect in general, there are still many more mood categories to be investigated.

Moreover, another aspect of interest that reinforces and contributes to scientific understanding of the relationship between CR and mood are chronotypes (Goel et al., 2013). Chronotype represents the inter-individual differences in circadian rhythmicity that results from preference of sleep-wake cycle timing, or the natural inclination to be a “morning person” or “evening person” (Zou et al., 2022). Broadly, there are two recognized profiles: morning chronotypes, who prefer to accomplish most tasks in the first half of the day, and evening chronotypes, who prefer the latter half (Goel et al., 2013). Earlier research indicates the

association between CR and individuals with a morning chronotype, who exhibit an advanced CR, approximately three hours ahead of its evening counterpart (Taillard et al., 2003; Smith et al., 1989). Research suggests that one's chronotype can significantly impact mood state fluctuations, mediated by the timing of peak activity levels on circadian processes, notably affecting the sleep-wake cycle (Goel et al., 2013). Morning-oriented individuals align well with socioenvironmental entrainment, typically experience improved mood stability, increased positivity, and higher energy levels earlier in the day (Goel et al., 2013).

In contrast, evening-oriented individuals feel more active and emotionally balanced later in the day, often experiencing more severe mood fluctuations and negative emotions (Zou et al., 2022). This incongruity between internal rhythm and socioenvironmental cues, such as in the case of working a night shift, can lead to disruptions in mood, productivity, as well as overall well-being (Taillard et al., 2003). Heightened mental or physical activity later in the day is at odds with the body's intrinsic preparation for sleep, disrupting the typical biological rhythm. Previous studies consistently link evening chronotype to heightened self-blame, stress, negative cognitive biases, rumination, and reduced happiness (Zou et al., 2022). Furthermore, consensus among existing research has established a strong association between eveningness and depression, also identifying the chronotype as an independent risk factor (Kivela et al., 2022; Zou et al., 2022; Goel et al., 2013; Taillard et al., 2003). While the exact mechanism of action through which chronotype mediates the relationship between CR and mood is unclear, prior studies affirm the robustness of the association between the three factors. Recognizing and accommodating these differences in chronotypes can significantly contribute to managing mood fluctuation and enhancing mental well-being for individual with an evening chronotype.

In addition, variation in mood is associated with another aspect, sleep quality, for which alterations in CR can have physiological and behavioral manifestations. Somatic and cognitive changes, which have a profound impact on an individual's functional abilities, are directly impacted during impairment of the sleep-wake cycle (Claudio & Andrea, 2022). Notably, the diagnostic criteria for several mood disorders, such as depression, include items related to activity and sleep alterations, emphasizing their strong correlation (Schnell et al., 2014). Previous literature has also demonstrated an association between circadian variation in mood and sleep quality, where sleep deprivation resulted in increased impaired emotional regulation and cognitive inhibition due to circadian misalignment (Franzen et al., 2008). Similarly, Liu et al. (2020) found poor sleep quality to be positively correlated with negative affect, while negatively correlated with positive affect and self-control. Since the time that the relationship between mood and sleep-wake cycle was observed, their relationship has been substantially elucidated. Nevertheless, a comprehensive understanding of the causal significance of circadian rhythm in the context of mood disorders is yet to be established.

Circadian Rhythm and Mood Disorders

In humans, numerous pieces of evidence point to a bidirectional relationship between mood disorders and circadian rhythm (Schnell et al., 2014). Mood disorders are a subset of mental health disorders distinguished by a clinically significant disturbance in how individuals respond emotionally to external stimuli, resulting in behaviors that are considered maladaptive (APA, 2013). Included in this category are seasonal affective disorder (SAD), major depressive disorder (MDD), and bipolar disorder (BD), with further subdivisions based on cause, timing, or duration (APA, 2013). According to the *Diagnostic and Statistical Manual of Mental Disorders (DSM)*, fundamental diagnostic criteria for mood disorders include disruption to sleep and

activity patterns. Despite the frequent co-occurrence of mood disturbances and dysfunctional sleep-wake cycle, the shared psychophysiological correlates of these conditions remain poorly understood (Sikka & Gross, 2014). To this extent, it is crucial to keep in mind that the physiological oscillation between periods of sleep and wakefulness conforms to a circadian pattern that is regulated by the alignment between the external environment and internal biological clock (Franzen et al., 2008). Viewed from this perspective, uncovering the connection between mood state fluctuations and the CR system emerges as a crucial relationship, essential for elucidating deeper insights into the etiology of mood disorders, which can potentially be leveraged into promising treatment innovation (Bechtel, 2015). For this reason, this section will explore the existing body of literature on the bidirectional link of mood disorders with circadian factors.

Mood disorders are characterized by dysfunction in circadian rhythm, associated with mood disorder symptomatology, linked to seasonal variations in circadian parameters, and often require manual alteration to the circadian rhythm pattern to ensure successful treatment (Schnell et al., 2014). Albeit it is unclear in which direction the causality between mood disorders and CR runs. Regularly, individuals with mood disorders such as MDD and BD exhibit irregularities in various circadian parameters including fluctuations in body temperature, blood pressure, as well as monoamine secretion pattern such as in melatonin and cortisol across a 24-hour period (Emens et al., 2009; Bunney & Bunney, 2000; McClung, 2013; Bechtel, 2015; Srinivasan et al., 2006). Individuals suffering from various mood disorders also exhibit daily fluctuations symptom severity, typically experiencing mood improvement in the evening (Wirz-Justice, 2008). However, it is also worth mentioning that some individuals may display a contrary pattern featuring mood diminishment in the evening (Joyce et al., 2005). Prior results have indicated a

higher occurrence of diminished circadian rhythm amplitude linked to depressive disorders (Bechtel, 2015). This connection often results in individuals experiencing mood disorders showing delayed phases in Positive Affect (PA) and Negative Affect (NA), suggesting a potential cause-and-effect link between circadian rhythms and mood disorders (Claudio & Andrea, 2022).

Moreover, individuals with mood disorders often exhibit altered circadian regulation, sleep-wake cycles, as well as a CR phase delay leading to insomnia at night and daytime sleepiness (Srinivisian et al., 2006). Further evidence of the strong association between the two factors is exemplified by the diagnostic criteria for mood disorders such as MDD, BP, post-traumatic stress disorder (PTSD), and GAD, which lists sleep pattern disruption as a hallmark symptom (Bechtel, 2015). In particular, depression has been linked to circadian disruption, with nearly all affected individuals reporting sleep-cycle disturbances as well as displaying altered sleep-wake cycle phases and duration (McClung, 2013; Nechita et al., 2015). Patients with MDD have also been found to exhibit lower levels of expression of established circadian-related genes, directly impacting the entire 24-hour circadian period (Li et al., 2013). Further existing research has observed sleep deprivation to impair emotional regulation and cognitive inhibition as a result of circadian misalignment, further strengthening support for the interconnection between circadian variation in mood and sleep quality (Franzen et al., 2008).

Another aspect of interest that reinforces our understanding on the relationship between mood disorders and CR, is the inclusion of seasonal affective disorders such as SAD, in the DSM. The inclusion of a seasonal clinical mood disorder as a mental health condition underscores the substantial impact of circadian rhythms on mood states, emphasizing the tangible effect that CR has on influencing mood and highlighting its profound significance

(Fishbein et al., 2021). Specifically, the etiology of seasonal affective disorder stems from seasonal changes in environmental cues, which induces clinical symptomatology as well as changes in circadian parameters (Bechtel, 2015). To give an example of the extent of the seasonal influence on mood, SAD features include a longitudinal pattern of elevated depressive symptomatology during the winter season, when circadian function is most negatively impacted, followed by spontaneous improvement in the summer, when circadian function is most positively impacted, in a reoccurring pattern over several years (Fishbein et al., 2021; Schnell et al., 2014). Moreover, individuals diagnosed with SAD will often exhibit irregular or delayed circadian function that is divergent from the immediate environmental cues (Bunney & Bunney, 2000). Research shows that the incidence of SAD varies depending on the latitude of the affected population, with higher rates in regions characterized by shorter photoperiods during the winter (Salgado-Delgado et al., 2011). Interestingly, SAD patients experiencing symptoms at home report temporary symptom relief and improved mood while on vacation in southern regions, resulting from the reduced seasonal effect on CR function (Kronfield-Schor & Einat, 2012).

Lastly, the success of circadian focused therapies in treating mood disorders represents another facet lending support to the relationship between mood disorders and CR (Walker et al., 2020). Multiple previous studies have observed that interventions targeting the circadian rhythm system, such as through methods including bright light therapy, sleep deprivation, lithium stimulants and melatonin depressants, have demonstrated positive effects in relieving depressive symptoms (Schnell et al., 2014; Bechtel, 2015; Claudio & Andrea, 2022; McClung, 2013). To exemplify, clinical trials of the drug Agomelatine, a melatonin agonist, demonstrated rapid and long-term relief from depressive symptoms in individuals with SAD, MDD, and BD (Schnell et al., 2014), with supporting evidence from animal trials (Rainier et al., 2012; Bourin et al., 2004).

In this case, Agomelatine advanced the circadian phase in rodents suffering from a circadian phase delay which prevented the onset of sleep until later in the circadian night (McClung, 2013). The melatonin derived drug was successful in resolving depressive symptomatology through the normalization of time of sleep onset as well as maintain quality across the circadian night, ensuring emotional refreshment in the morning (Claudio & Andrea, 2022). Consequently, such efficacy highlights the therapeutic potential of circadian targeted interventions aimed at enhancing mood through adjustments to the circadian sleep-wake cycle (Schnell et al., 2014).

Moreover, the combined treatment of a selective serotonin reuptake inhibitors (SSRIs) pharmaceutical, alongside circadian-focused light therapy, has shown improved efficacy in alleviating symptoms of MDD when compared to light therapy alone (Lam et al., 2016; Burns, 2021). Conversely, previous evidence from animal models demonstrates that after injected with SSRIs, rodents exhibit an advance in their circadian rhythm phase and alterations in circadian-related gene expression (Cuesta et al., 2008; Mendoza et al., 2007). Additionally, the deliberate interruption of the sleep-wake cycle by exposing rats to dim light during the circadian night was observed to result in an increased number of anxiety-like responses the following day (Walker et al., 2020). These findings suggest that the deliberate interruption of CR also has the potential to negatively influence mood as well as increase the possibility of mood disorder symptom presentation.

Ultimately, there is substantial evidence to indicate that mood disorders are linked to a disturbance in the fundamental biological processes also responsible for regulating circadian rhythms (Schnell et al., 2014). The bidirectional nature of this connection suggests that mood disorders can both influence and be influenced by circadian dysregulation in the sleep-wake cycle (McClung, 2013). Previous research indicates a strong association between mood disorders

and circadian dysfunction, and many individuals with psychiatric illnesses exhibit symptoms of circadian dysregulation (Salgado-Delgado et al., 2011; Bechtel, 2015; Fishbein et al., 2021).

Additionally, the shared symptomatology of mood disorders, including alterations in sleep patterns and features, highlights a similarity in the biological correlate between the entire class of conditions, indicating support for the potential of CR targeted modification in mood disorder management (. In the case of mood disorders, symptomatology is primarily manifested through the exhibition of persistent disturbances to mood. Despite the evident importance of the relationship around CR and mood state regulation, little is known about the intricate pattern of variation for individual mood states associated with the emergence or treatment of mood disorders. Given the importance of circadian rhythm in addressing mood disorders, exploring the intricate and dynamic correlations among prevailing mood states becomes pivotal for comprehending the origins of this category of mental health conditions. Furthermore, while some emotional states, such as happiness, sadness, depression, and anxiety, have been examined for a diurnal pattern, there are still many additional mood states to explore in this area.

Positive and Negative Affect

Positive Affect (PA) refers to an individual's experience of pleasant emotions such as joy, enthusiasm, and alertness (Hasler et al., 2012). PA is often characterized by feelings of happiness and contentment (Russel, 1980). Typically, PA exhibits a quadratic diurnal variation with a peak occurring between mid-day and mid-afternoon and the lowest values in the early morning and late evening (Murray et al., 2002). Perceived energy levels follow a similar pattern, increasing in the early morning and peaking around mid-day before declining in the evening, supported by objective measures of cortisol blood serum concentrations and heart rate (Bechtel, 2015).

Previous studies investigating variation in individual mood state adjectives have also established

a strong correlation between circadian pattern in PA and perceived energy levels (Wood et al., 1992; Stone et al., 2006).

Empirical evidence from studies on the diurnal patterns of mood states indicate that the happiest mood is experienced during the circadian day, while the saddest mood occurs during the circadian night, as assessed through Visual Analog Scales for happiness and sadness (VAS) (Gift, 1989; Boivin et al., 1997). Through the distinct assessment of pleasant and unpleasant feelings by measures such as the Positive and Negative Affect Scale (PANAS), as well as Negative Affect (PANAS-NA) and Positive Affect (PANAS-PA) subscales (Watson et al., 1988), only PA was found to exhibit a significant circadian rhythm (Murray et al., 2009). Positive Affect, or feelings of happiness, were observed to peak around noon, with a trough in the late afternoon (Stone et al., 2006), although this pattern may be influenced by the sampling window, which typically spans less than 24 hours (Emens et al., 2020).

Collectively, the body of current research on the relationship between Positive Affect and circadian rhythm suggests a persistent trend wherein Positive Affect tends to peak later in the daytime, and reach a nadir in the early morning hours (Murray et al., 2002; Murray et al., 2009; Golder and Macy, 2011; Hasler et al., 2012; Miller et al., 2015; Emens et al., 2020). This variation in Positive Affect appears to be opposite to that of Negative Affect (NA) across the 24-hour circadian period, with the highest PA levels coinciding with the lowest NA, and the highest NA occurring at the same time as the lowest PA (Stone et al., 2006; Miller et al., 2015; Murray et al., 2002).

However, it is worth noting that much of the research demonstrating a strong circadian effect in PA utilized composite scores including potentially extraneous mood state adjectives related to multiple mood dimensions (Sikka & Gross, 2014). As a result, it unclear whether the

observed pattern of circadian variation reflects changes in perception of energy level, from the arousal dimension of mood, or pleasantness, the valence dimension of mood, as defined by Thayer's (1998) model of arousal-valance. To address this, adjectives related to arousal would be removed from the composite score, leaving only those adjectives which fall in the middle of the arousal spectrum in order to examine a singular valence dimension of PA (Emens et al., 2020; Thayer, 1998; Russel, 1980). This approach aims to investigate whether the established circadian pattern of Positive Affect will replicate when terms related to arousal are excluded, shedding light on the CR of specific mood states encompassed by PA. Past literature has often employed tools like the VAS or PANAS to assess mood, both of which measure positive and negative mood as part of the same spectrum (Sikka & Gross, 2014; Murray et al., 2020). Given the vital role of diminished Positive Affect in the context of mood disorder symptomatology, particularly in contrast to the predominant expression of Negative Affect, comprehensive insights into the timing profile, patterns of PA, and associated factors throughout the 24-hour circadian period are crucial. Determining the exact pattern of variation in PA will be imperative to defining a profile of CR of mood for individuals affected by mood disorders, for use in diagnosis, treatment interventions, and more (Rusting & Larsen, 1998; Kivela et al., 2022).

Another mood with evidence of circadian variation is Negative Affect, which refers to an individual's experience of unpleasant or distressed emotions, including feelings of anger, fear, and sadness (Hasler et al., 2012; Sikka & Gross, 2014). When investigating the circadian effect on NA, it is important to note that the mood state encompasses a broad range of emotions and is not consistently associated with any singular mood category in the existing body of literature (Kivela et al., 2022; Sikka & Gross, 2014). Frequently employed NA assessment measures such as the PANAS often characterize NA using a range of adjectives including distressed, upset,

guilty, scared, irritable, nervous, and more (Watson et al., 1988). Moreover, results from existing evidence that report a diurnal pattern of NA support previous research on circadian PA pattern, and demonstrate an inverse relationship with Positive Affect, where the highest values for one affect coincide with the lowest values for the other (Stone et al., 2006; Murray et al., 2002; Miller et al., 2015). Specifically, the numerical highest and lowest self-reported values for the 24-hours period suggest that NA may be highest, and PA lowest during the circadian night, around 3:00 am, with the opposite pattern occurring later in the afternoon, typically between 3:00 pm and 5:00 pm (Emens et al., 2020; Stone et al., 2006).

Historically, there has been a lack of consensus in previous research regarding a clear, significant pattern of circadian variation in NA (Emens et al., 2020). Some studies have found no significant diurnal variation in NA, while others found significant variation, but with different peak and trough times in each study. These discrepancies include a peak around 3:00 am and a trough in the afternoon (Emens et al., 2020), peak around 10:00 am and 5:00 pm with a trough around 12:00 pm (Stone et al., 2006), and a peak around 12:00 am with a trough around 12:00 pm (Miller et al., 2015; Murray et al., 2002; Boivin et al., 1997). Despite these variations, observational evidence supports the idea that NA fluctuates across the 24-hour circadian period (Stone et al., 2006; Miller et al., 2015; Porto et al., 2006). In comparison, in individuals affected by depressive mood disorders, NA has been observed to peak later in the day, which is notably different from peak time observed in non-clinical populations (Emens et al., 2020). This is supported by previous research has demonstrated an association between circadian variation in mood and symptom severity in MDD, meaning, when the body's internal clock is set later than environmental cues, mood disturbances become more pronounced (Emens et al. in 2009; Hasler et al., 2010). Yet the reasons for this relationship remain unelucidated, and the existing body of

evidence is weak in presenting a clear consensus of evidence to support of a clear, circadian pattern in NA (Emens et al., 2020; Miller et al., 2015).

The existing body of literature on the relationship between Negative Affect and CR can be summarized as a circadian pattern in which NA tends to be highest during the circadian night, followed by a trough approximately 12 hours later (Emens et al., 2020; Murray et al., 2002). In contrast to PA, there is a lack of consensus in previous research on whether NA exhibits diurnal properties of variation, which has led to speculation by some researchers that the expression of Negative Affect primarily occurs during the circadian night, creating difficulty in assessing the mood state accurately (Emens et al., 2020). The exact timing of NA peaks and troughs may vary between studies, possibly reflecting the influence of extraneous mood states or the manifestation of affective disorders resulting from altered circadian variation in mood or energy levels (Stone et al., 2006; Emens et al., 2020). One interpretation of this issue is that highly variable changes in negative emotions are a consequence of high load work and lunch breaks that provide a buffer against the demands of the work environment, and randomly diminishing or exacerbating negative affect (Stone et al., 2006).

Another interpretation implies that the composite scores used to represent NA are not necessarily reliable, potentially due to the inclusion of mood item adjectives that reference extraneous mood states such as anger, irritability, or worry (Emens et al., 2020). Studies that succeeded in finding significant circadian effect for NA did not employ composite scores with mood items related to multiple mood states (Hogarth et al., 2014; Emens et al., 2020; Woods et al., 1992), such as anger, frustration, energy level, anxiety, and sadness. While previous studies that did utilize a combination of mood items related to multiple mood states in the NA composite score, often failed to find circadian variation (Emens et al., 2020; Stone et al., 2006). In

accordance with proposed future directions from previous studies, the present study will address these complexities by separating out mood items related to specific mood states within Negative Affect, such as “nervous” or “angry,” from the composite score in order to examine circadian variation in NA more effectively (Emens et al., 2020; Stone et al., 2006). Additionally, focusing on a specific mood aspect of NA will help to mitigate the influence of moods that are more subject to relativity, providing a clearer understanding of the circadian effect on this aspect of emotional experience (Sikka & Gross, 2014). Recognizing the significance of both NA and PA within the context of mood disorder symptomatology, where excessive levels are prevalent, it is essential to understand the timing, profiles, and associated factors across the entire 24-hour circadian period. Similarly, determining the exact pattern of variation in NA remains salient in defining an associated CR profile which can be used to diagnose or treat various mood disorders with greater accuracy.

Affection and Annoyance

Another mood that has not specifically been investigated for a pattern of variation across the circadian period is the mood state of Affection. Affection is often characterized by feelings of warmheartedness, fondness, tenderness, and caring, playing a significant role in human emotional experience (APA, 2023). Generally, Affection represents a low-arousal, high-pleasantness mood state (Hartmann et al., 2023). Circadian rhythms, which regulate the concentration of various hormones related to the sleep-wake cycle and sexual function, have a substantial influence on Affection (Koop et al., 2022). Moreover, previous evidence indicates a connection between emotions related to romantic love and sleep disturbances in teenagers, wherein heightened feelings of romantic love are associated with shorter sleep duration (Bode & Kushnick, 2021). Related to this is the circadian pattern of hormone secretion, particularly in

testosterone, which is characterized by elevated levels during the early morning and late evening periods (Bode & Kushnick, 2021). Beyond an established association with feelings of aggression, testosterone has also been suggested to potentially enhance feelings of Affection in certain situations, as demonstrated by heightened prosocial behavior in non-human mammals during peak levels (Kelly et al., 2022). Taken together and based on evidence of an existing association with circadian function, is it plausible that Affection also exhibits a pattern of circadian variation.

Of the existing research on the circadian rhythm of mood, there is limited evidence for a pattern of circadian variation in Affection. Oftentimes, Affection is not included as an individual mood state in studies assessing diurnal mood patterns, although it is unclear why. In some instances, mood items related to Affection are included in a PA composite score as one of the many mood states broadly encompassed by the overarching emotional experience (Sikka & Gross, 2014). Some previous studies have included mood items such as "warm" or "loving," within PA composite scores, and the results demonstrated a circadian rhythm effect (Stone et al., 2006). For instance, Stone et al. (2006) included "warm" as an item and found two peaks around 12:00 pm and 8:00 pm, with a wide trough between 2:00 and 5:00 pm. However, it is possible that this pattern of variation was influenced by the sampling window employed, between 7:00 am and 9:00 pm, rather than across a full 24-hour circadian period (Stone et al., 2006; Hogarth et al., 2014; Miller et al., 2015).

Ultimately, there is presently no known research specifically focused on examining the circadian effect on Affection. Instead, much of the research related to the circadian effect on mood states related to Affection is based on findings which include referential mood items in PA composite scores (Kivela et al., 2022). This makes it challenging to isolate the impact of diurnal

mood fluctuations on Affection and determine whether the strong circadian variation exhibited in PA extends to additional mood states (Rusting & Larsen, 1988; Murray et al., 2002; Sikka & Gross, 2014). To address these challenges, this study aims to isolate mood adjectives related to Affection to explore their potential circadian variation, which may offer new insights into this aspect of emotional experience (Emens et al., 2020; Thayer, 1989). By examining Affection as a distinct mood state, researchers hope to uncover a unique circadian pattern and demonstrate the possibility of additional patterns in additional mood states. In mood disorders, the expression of pleasant mood is diminished, and understanding the dynamic association between CR and Affection may hold the potential to inform improved, tailored therapies (Walker et al., 2020; Fishbein et al., 2021). Moreover, this knowledge may enable healthcare professionals to better align treatment strategies with individualized circadian rhythms, potentially improving and enhancing the expression of emotional experiences related to Affection.

Lastly, another mood with a potential pattern of circadian variation is Annoyance, which is often characterized by feelings of frustration, irritability, anger, and disdain, represents an internal emotional state that is typically unpleasant (Barata et al., 2016). Irritability, which is a mood state closely related to Annoyance, is listed as a feature diagnostic criterion for many mood disorders, including the presentation of a persistent or excessive state of irritability, anger, or frustration (Hartmann et al., 2023). It is possible for feelings of Annoyance to manifest as a symptom of an existing mood disorder, but also as a result of co-morbidity with an attention deficit disorder, generalized anxiety disorder, and more, complicating the diagnostic picture (APA, 2013; Sikka & Gross, 2014; Kivela et al., 2022). Moreover, previous research demonstrates an association between heightened feelings of Annoyance and impaired sleep quality (Vandekerckhove & Wang, 2017), which is often related to circadian dysfunction. Also

regulated by CR, is testosterone, a hormone often associated with traits like aggression and dominance, which can contribute to increased Annoyance in some individuals (Kelly et al., 2022; Bribeiskas & Hill, 2010). Circadian variation in testosterone levels, with spikes in the morning and evening, may influence mood state fluctuations, including Annoyance (Kelly et al., 2022). However, the possible circadian effect in Annoyance is a topic that has not been extensively explored in previous research, with little known previous research focused on the specific circadian effect.

The most relevant previous evidence supporting circadian variation in Annoyance are from studies that utilized assessments of individual mood item adjectives in their examination of the rhythmicity of the unpleasant mood state (Stone et al., 2006; Emens et al., 2020). For example, Stone et al. (2006) included several mood item adjectives related to Annoyance, such as "Angry," "Frustrated," and "Impatient" in the assessment of circadian mood patterns. Researchers found that subjective ratings for "Frustrated" peaked around 10:00 am, with a trough around 12:00 pm; while "Angry" peaked around 2:00 pm, with a trough around 12:00 pm; and "Impatient" peaked around 4:00 pm, with a trough around 12:00 pm (Stone et al., 2006). Notably, results demonstrated sharp peaks at different times of the day for the different mood items related to Annoyance, suggesting a complex pattern of variation. In response, some researchers have speculated that Annoyance may be tied to factors such as fatigue state or recent experience (Stone et al., 2006; Emens et al., 2020) which varies daily as the result of transient obstacles which manifests as an unstable daily pattern (Stone et al., 2006; Emens et al., 2020).

Another facet which contributes to our understanding of the relationship between CR and Annoyance is support from prior anecdotal evidence on the written or verbal expression of the mood state through social media posts across a 24-hour period (Dzogang et al., 2017). Prior

research on the pattern of variation in anger expressed through Twitter posts demonstrated local circadian troughs in the morning around 7:00 am and 10:00 am, which gradually reach a peak during late night, around 2:00 am and lasts until early next morning (Dzogang et al., 2017). In follow-up study, anger expressed through tweets, or posts, from users on the Twitter platform, reached a peak around 2:00 am, with a sharp trough around 8:00 am (Dzogang et al., 2018). However, to our knowledge, no known study has yet to confirm such speculation by assessing circadian variation for the specific affect.

Overall, there is uncertainty regarding the circadian variation of Annoyance, as past research has included Annoyance-related terms in NA composite scores, making it challenging to discern the specific effect of a circadian pattern for Annoyance (ten Thijj et al., 2020; Wood et al., 1992). To address this, some researchers have recommended that future studies consider a separate score for the Annoyance-specific mood state (Emens et al., 2020). Therefore, the current study aims to use a composite score with mood items more specifically related to the mood state of Annoyance, to explore potential circadian variation and provide insights into this distinct aspect of the emotional experience (Thayer, 1989). By isolating Annoyance as a separate mood state, we hope to uncover a unique circadian pattern less influenced by extraneous mood states, which can offer a novel perspective on the Annoyance and CR relationship. Previous research indicates Annoyance is interconnected with mood disorder symptoms, diagnostic criteria, and treatment considerations, making it an important aspect to consider in the evaluation and management of mood disorders (Emens et al., 2020). By comprehending the circadian patterns of Annoyance, clinicians may be able to tailor interventions to address specific mood-related challenges during vulnerable times of the day, potentially leading to better outcomes and improved quality of life for individuals suffering from mood disorders.

Present Study

In this research, we propose several design improvements and recommendations for a study focused on understanding circadian rhythms and emotional states. First, we suggest unmasking the CR effect by implementing a 24-hour long sampling period with consistent time series measurements taken every 4 hours (Goel et al., 2013). Previous studies that do not employ an unmasking protocol with frequent, even sampling time series, report varying patterns of circadian variation with delayed or accelerated temporal peaks, deviating from that of the research consensus by one to two hours (Stone et al., 2006; Hogarth et al., 2014; Murray et al., 2002). Additionally, measuring affect across a broad 24-hour time window which covers active and resting states, should help compensate for the potential manifestation of affective disorder symptomatology in participants as well as strengthen potential patterns of variation in mood weakened by inadequate sampling procedures (Sikka & Gross, 2014). While our study employs a slightly different unmasking method, we anticipate to observe a circadian variation in Positive Affect which peaks during the circadian day, as well as in Negative Affect, which peaks during the circadian night, in line with previous findings (Stone et al., 2006; Miller et al., 2015; Sikka & Gross, 2014).

In addition, whereas most previous research employed the VAS or PANAS self-report measures to assess subjective mood, the present study will employ the Brief Mood Inventory Scale (Watson et al., 1988). Previous researchers have suggested future research employ a different, more diverse measure of affect which features multiple mood categories and dimensions, in an effort to examine potential improved efficacy at differentiating between mood states and producing stronger patterns of variation (Goel et al., 2013; Kivela et al., 2022). The present study also proposes to control for the arousal dimension of mood in Positive Affect and

Affection mood composite scores, through the exclusion of mood inventory items classified as a high or low arousal state by the arousal-valence model of emotion (Thayer, 1989).

Additionally, the present study emphasizes the importance of conducting research in non-clinical populations to better understand the variation in unpleasant mood across a 24-hour period (Goel et al., 2013). While clinical populations have exhibited a stable effect on the fluctuation of unpleasant emotions, the nature of the relationship remains unclear for naturalistic settings (Franzen et al., 2008). To address this, the present study limited participants to male, undergraduate students, from a population less affected by the variability of work commutes, and to allow for a more controlled investigation of circadian rhythms in mood.

Therefore, the present study aims to build upon existing knowledge by demonstrating the replicability of circadian variation in mood, which has been insufficiently explored in previous research. Although interruption to the sleep-wake cycle has been linked to negative emotions, inappropriate emotions, to emotional volatility, as well as to clinical mood disorders, circadian effects on emotional state *per se* have infrequently been examined (Franzen et al., 2008; Emens et al., 2020). Moreover, to our knowledge, previous research has focused on mood states such as happiness, sadness, depression, and anxiety, highlighting a need for additional research on singular mood states to gain better understanding of CR-related population health (Walker et al., 2020). Circadian rhythm in hormone production, specifically testosterone, is described as having spikes in concentration in the morning and evening, as well as also increase feelings of Affection in some contexts (Kelly et al., 2022). Therefore, we also seek to extend our understanding by including additional affects such as Affection and Annoyance, which have received limited attention in circadian research.

In summary, the current study therefore attempted to replicate previous work demonstrating circadian variation in Positive vs Negative Affect, as well as extend findings to the specific emotional states of Affection vs Annoyance. We predict that results will demonstrate the replication of circadian patterns in Positive and Negative Affect established by previous research. Gaining a deeper understanding of the intricate interplay between mood and biological rhythms holds the promise of uncovering innovative markers for mood disorders, paving the way for time-targeted interventions or other circadian-focused therapies. Such insights have the capacity to transform the approach to mood disorder management, providing fresh perspectives on therapeutic approaches and underscoring the significance of maintaining a consistent circadian environment for mental health and well-being.

Method

Participants

As part of a larger study, 13 Montclair State University undergraduate students with an average age of 19 years ($SD = 2.5$). Participants were required to be 18 years of age or older, male, right-hand dominant, have normal to normal-corrected vision, and no self-reported physical symptoms of illness. Participants completed a five-item demographic questionnaire on personal characteristics and general health which took approximately three minutes to complete. Demographic information for the sample is provided in Table 1.

Participants were compensated for their time through the award of six SONA research credits as well as the option to be entered into a raffle to win a \$150 Amazon gift card. Prior to participation, all participants were provided an online consent form approved by the Institutional Review Board at Montclair State University for the present study. The form was read aloud by the research assistant conducting the screening session while participants read along, and upon

conclusion gave the option to provide consent or non-consent, by selecting the corresponding option on the form. The present study design and procedure is compliant with guidelines for human subject's research and approved by the Institutional Review Board at Montclair State University (IRB-FY#: 22-23-2786).

Materials

Research Equipment

To facilitate remote data collection across a period of 24 hours, participants were provided with research equipment which included the Amazfit GTR 2e smartwatch, used to alert and prompt participants to complete online survey every four hours, as well as an iPad 12 mini tablet device. Participants used the Safari application on the iPad 12 mini to access the internet and complete an online survey through the Qualtrics platform. Prior to the start of participation, participants also utilized the FaceTime application on the iPad 12 mini to complete orientation instructions. Survey links corresponding to a scheduled time of completion as well as a PDF copy of the instructional packet were also provided through the Messages application.

Participant Demographic Questionnaire

The participant demographic questionnaire consisted of five items which asked participants to provide information on various personal characteristics. This questionnaire was modified from the existing, stock questionnaire XM Qualtrics Certified: Demographics. Three items to assess age, gender, and race was retained from the stock questionnaire, and the rest removed. Two items to assess the symptomatology and recent diagnosis of COVID-19 were added from an existing Montclair State University Hawk Check survey frequently completed by all university students. This demographics questionnaire was administered to ensure that the sample group is diverse and representative of the general population. Questions related to health

status served the dual purpose of maintaining the cleanliness of equipment, ensuring the well-being of fellow participants, and preventing the spread of COVID-19. The questionnaire was additionally employed for participant screening, ensuring that potential participants met the predefined eligibility criteria.

Mood Assessment: Brief Mood Inventory Scale

The Brief Mood Introspection Scale (BMIS) consists of 16 mood item adjectives developed by Gashke and Mayer (1988). This self-report measure assesses the current subjective mood of the participants across two mood dimensions, arousal, and valence. The scale features eight mood categories in which every mood state is defined by two mood adjectives related to the specific mood category. This includes: (1) happy (happy, lively), (2) loving (loving, caring), (3) calm (calm, content) (4) energetic (active, peppy), (5) anxious (jittery, nervous), (6) angry (grouchy, fed up), (7) tired (tired, drowsy), and (8) sad (gloomy, fed up). Measurement guidelines provide instructions for the calculation of pleasant mood (happy + loving + calm + energetic) and unpleasant mood (anxious + angry + tired + sad). The BMIS employs a four-point Meddis Style scale (XX = *definitely do not feel*; X = *do not feel*; V = *slightly feel*; VV = *definitely feel*). This scale is then transformed into a numerical value based on the standard four-point Likert scale (1 = *definitely do not feel*; 2 = *do not feel*; 3 = *slightly feel*; 4 = *definitely feel*), in order to calculate a score for each individual mood. Mood scores can be calculated using reverse scoring as well as subtractive scoring. The BMIS is considered a factor-valid instrument suitable for integration with the mood circumplex, (Mayer and Gashke, 1988). Additionally, the Mood Introspection Scale (Mayer et al., 1988) and the Russell Adjective Scale (Russell, 1979) have also been confirmed to be compatible with the BMIS.

Mood was assessed for variation over a 24-hour period using the sixteen mood item adjectives provided to create individual composite scores for the PA, NA, Affection, and Annoyance mood states. Each mood composite score was comprised of two mood items which corresponded to a mood state. Mood composite scores included Positive Affect (Happy + Content), Negative Affect (Sad + Gloomy), Affection (Loving + Caring), and Annoyance (Fed Up + Grouchy). First, all Meddis style scale responses for each mood item were transformed into Likert-scale numerical values using the BMIS guidelines and without additional transformation. For each participant at each Timepoint, the numerical Likert scale value reported for each mood item was summed to calculate a mood composite score.

Study Design

As part of a larger, Montclair State University internally funded grant research project, participants were recruited through Montclair State University's online SONA student research participation system. Students who were interested in participating, volunteered through the SONA website by selecting one of the available participant screening timeslots listed, to videoconference with a research assistant. During the approximately 20-minute screening videoconference over Zoom, participants were described the present study, read the online consent form, provided online consent, and completed an online demographics questionnaire. Following review, participants who met inclusion criteria were scheduled via email for a participation date, FaceTime orientation, and research equipment pick up, about three to four weeks in the future. On the day of participation, students retrieved the research equipment from a research assistant in the Cerebral Lateralization Laboratory on the Montclair State University campus sometime in the morning and completed participant orientation over FaceTime prior to the 12:00 pm start of data collection. Participant orientation provided explanation of instructions

on participation, the use and care of research equipment, location of resource information, and steps to return the research equipment.

Following this, across 24-hours, participants remotely accessed the online BMIS survey using the link corresponding to the Timepoint provided in the Messages application of the iPad remotely, every four hours for a total of seven times. The survey took approximately three minutes to complete, at 12:00 pm on Day 1, 4:00 pm, 8:00 pm, 12:00 am, 4:00 am, 8:00 am, and 12:00 pm on Day 2. Participants were instructed to follow their typical sleep-wake schedule and were woken by alarms on the equipment if the participant was asleep. Participants that accidentally missed a scheduled time to complete the online survey were instructed to complete the survey as soon as possible. Upon the conclusion of data collection at 12:00 pm on Day 2, participants returned all research equipment to a research assistant in the Cerebral Lateralization Laboratory on the Montclair State University campus. Following review of survey responses and inspection of research equipment, students were awarded compensation of SONA credits, if eligible, as well as offered the option to be entered into a raffle to win an Amazon gift card upon the conclusion of the study. Upon return of the research equipment, all surfaces were disinfected using alcohol wipes and all existing data for all iPad 12 mini applications used, erased from the device, in preparation for use by the next participant.

Data Analysis

As a result of so few subjects, the use of traditional inferential statistical tests for data analysis, specifically a repeated measures analysis of variance, was deemed not appropriate¹. In its place, descriptive data was analyzed for a pattern of circadian variation through a combination of visual and numerical comparisons of average peak and trough value for each affect (Murray et al., 2002). At each Timepoint, for each mood state, the mean average mood rating was calculated

using participant mood rating scores. For each mood state, the highest average mood rating was used to identify the peak Timepoint, and the lowest average mood rating was used to identify the trough Timepoint (Murray et al., 2002). The highest and lowest average mood ratings were utilized to provide a clearer conception of the change in intensity over across the of 24-hours circadian period (Refinetti, 2020). For each mood state, average mood rating was used to compare the Timepoint with the highest and lowest average numerical values as well as the 8:00 am and 8:00 pm Timepoints.

In addition, Cohen's d effect sizes were calculated for the average highest and lowest numerical values as well as for 8:00 am and 8:00 pm, for mood state, in order to assess the magnitude of the observed differences in mood ratings. For all analyses, differences were considered significant at $p < 0.05$. All descriptive analyses were performed using IBM SPSS version 28 for Mac computers (2021). Cohen's d effect sizes were computed specifically, using number four online effect size calculator for repeated measures (Lehan & Lehan, 2016). Descriptive data including mean, standard deviation, and correlation coefficient were entered into the online calculator such that the Timepoint with the greater numerical average was input as group one, while the lesser numerical average was group two. All reported Cohen's d effect size values were non-pooled (d_{RM}) and interpreted using Cohen's (1988) guidelines in which a value of 0.2 or lower is considered small, a value between 0.2 and 0.5 is considered medium, and a value between 0.5 and 0.8 or higher is considered large. Statistical significance was not provided by the online calculator or otherwise computed for any of the Cohen's d effect sizes reported, due to small sample size increasing the chance of a type two error occurring.

Results

Descriptive Data

Descriptive data including mean, standard deviation, and range for Positive Affect, Negative Affect, Affection, and Annoyance are reported in Table 2. The results obtained in the present study generally replicate previous research findings concerning the circadian variation of mood and extends findings of circadian variation to Affection and Annoyance mood states. To this aim, a line graph providing a visual depiction in the change in average mood state ratings over time is presented in Figure 1. We first demonstrate that the intensity of Positive Affect and Negative Affect oscillates oppositely over the course of a day, with PA appearing to reach its highest visual point around noon and NA hitting its lowest visual point around midnight. Further analysis of descriptive data reveals PA reached numerical peak around 4:00 pm ($M = 6.4$, $SD = 1.4$) with a trough around 4:00 am ($M = 4.3$, $SD = 1.5$), similar to prior studies (Stone et al., 2006; Emens et al., 2020; Miller et al., 2015).

Conversely, NA reached highest numerical value around 12:00 am ($M = 3.8$, $SD = .98$), while the lowest numerical point occurred around 8:00 pm ($M = 2.8$, $SD = 1.1$), similar to prior research reporting an inverse circadian pattern between NA and PA (Stone et al., 2006; Murray et al., 2002; Porto et al., 2006). Circadian variation in NA demonstrated a peak during the circadian night, around midnight (Stone et al., 2006; Emens et al., 2020; Murray et al., 2002) and trough during the latter part of the circadian day, towards the evening. Meanwhile, Affection exhibited a distinctive pattern with three apparent peaks around 12:00 pm-A on Day 1, 12:00 am, and 12:00 pm-B on Day 2, as well as two troughs around 8:00 pm and 4:00 am. Descriptive data revealed the highest numerical point occurred at 12:00 pm-A on Day 1 ($M = 5.9$, $SD = 1.2$) and the lowest numerical point occurred at 4:00 am ($M = 4.2$, $SD = 1.9$). Notably, both PA and Affection exhibited discrete diurnal patterns, yet shared a numerical lowest point at 4:00 am. In addition, the highest level of Affection occurred at 12:00 pm on Day 1, compared to a

commensurate average level recorded at the same time on Day 2 ($M = 5.5$, $SD = 1.2$). Further analysis also revealed two troughs with almost identical values at 8:00 pm ($M = 4.7$, $SD = 2$) and 8:00 am ($M = 4.8$, $SD = 1.7$).

Lastly, Annoyance displayed a discrete pattern which visually appeared to follow a normal distribution, with values peaking around midnight. Descriptive data revealed the highest numerical point occurred around 12:00 am ($M = 4.5$, $SD = 1.1$), and the lowest numerical point occurred around 12:00 pm-B on Day2, ($M = 3.5$, $SD = 1.4$). Annoyance was not observed to follow a parallel pattern to NA, with Annoyance experiencing a trough earlier in the day around 12:00 pm, followed by increasing values into the night, while NA displayed a trough later in the day around 8:00 pm, with values increasing into the afternoon. Moreover, Negative Affect and Annoyance shared a temporal peak at 12:00 am.

Lastly, the present study did not identify any obvious outliers in the scatterplots for each affect. However, it is important to note that our ability to judge influential data cases was limited due to the small sample size and absence of previously established values.

Effect Size

A Cohen's d effect size was calculated between the highest average PA recorded at 4:00 pm ($M = 6.38$), and the lowest levels at 4:00 am ($M = 4.31$), revealed a large effect (Cohen's $d = 0.843$). At 8:00 pm ($M = 6.2$, $SD = 1.6$), PA exhibited higher levels compared to 8:00 am ($M = 4.9$, $SD = 1$, Cohen's $d = .928$). While NA exhibited the opposite pattern, with 8:00 am ($M = 3.7$, $SD = 1.5$) levels being higher than at those at 8:00 pm ($M = 2.9$, $SD = 1.1$, Cohen's $d = .996$), resulting in a congruently large effect size. Moreover, comparison between the highest recorded Negative Affect levels at 12:00 am ($M = 3.9$, $SD = .98$) around mid-night, and lowest at 8:00 pm ($M = 2.9$, $SD = 1.1$, Cohen's $d = .833$) in the evening, parallels the same pattern of variation.

The highest recorded Affection levels occurred at 12:00 pm on Day 1 ($M = 5.9$, $SD = 1.2$), and the lowest levels at 4:00 am ($M = 4.15$, $SD = 1.86$, Cohen's $d = 1.20$). Additionally, Affection displayed nominally higher levels at 8:00 am ($M = 4.9$, $SD = 1.7$) compared to 8:00 pm ($M = 4.7$, $SD = 2$, Cohen's $d = .095$) resulting in the smallest effect size obtained for any comparison calculated. The converse pattern was observed for Annoyance, with lower levels observed at 8:00 am ($M = 3.8$, $SD = 1.5$) and higher levels at 8:00 pm ($M = 4.5$, $SD = 1.8$, Cohen's $d = 0.436$), also comparable to levels of NA at the same time. Lastly, the highest recorded average Annoyance was reported at 12:00 am ($M = 4.6$, $SD = 1.1$), with the lowest levels at 12:00 pm on Day 2 ($M = 3.5$, $SD = 1.4$, Cohen's $d = 0.802$).

Discussion

The present study hoped to replicate previous findings on Positive vs Negative Affect as well as extend findings to additional, novel affect states of Affection vs. Annoyance, in hopes of adding to knowledge on temporal variation of mood and aid future research on the existence of circadian effects for additional mood categories. Through building on the existing body of literature, we also hoped to encourage the innovation of circadian-focused therapeutics and temporally dependent clinical diagnosis or treatment intervention. As a consequence of an inaccurate measurement tool, a larger set of our collected data was ineligible for use, resulting in the majority of data analysis and interpretation completed post-hoc. As a result of the exploratory nature of the data analyses in this study, the results should be considered as preliminary evidence for the relationships and must be replicated by future studies. Being a study conducted in natural settings, the current evidence is correlational and unable to differentiate the effect on mood between the various internal and environmental factors at work. However, similar previous

research on circadian variation in mood conducted in closed, laboratory settings provide support for the reliability of results obtained by the present study.

To summarize results, the present study was able to identify patterns of Positive Affect and Negative Affect that align with previous research (Stone et al., 2006; Emens et al., 2020; Murray et al., 2002), supporting the presence of a circadian rhythm for each mood state. PA exhibited its highest ratings during the circadian day and lowest during the night, while NA displayed an inverse pattern, with highest ratings occurring during the circadian night. These findings reflect previous research (Sikka & Gross, 2014), which aids in substantiating the reliability of the present results as well as supports the efficacy of the unmasking protocol employed in assessing circadian mood fluctuations through the confirmation of similar findings (Goel et al., 2013). Our findings support our hypothesis that research on circadian variation in mood could be extended to additional, specific mood states including Affection and Annoyance, although this could not be statistically confirmed. Findings demonstrating individual diurnal patterns of variation in Affection and Annoyance indicate the potential existence of circadian rhythmicity in additional mood states. Affection exhibited a distinct bimodal pattern that differed from that of PA, while Annoyance displayed consistently elevated values throughout the circadian night, and dissipating in the early morning before rising again in the evening. The precursory results suggest that while circadian rhythm does have a strong influence on mood, some moods exhibited more variability than others.

The present study attempted to replicate previous findings of circadian Positive Affect and control for arousal state by isolating the representative composite score to a single mood dimension, using only valence items (Kivela et al., 2022). Results displayed a peak around 4:00 pm and a trough around 4:00 am, aligning with earlier studies that reported PA peaks between

2:00 and 5:00 pm, with a trough around 4:00 am (Ferris et al., 2014; Stone et al., 2006). Notably, these findings align despite our use of composite scores that did not include arousal dimension items. This temporal pattern closely adheres to our hypothesis, successfully replicating the well-established circadian PA pattern which peaks during the day and reaches its lowest point at night (Stone et al., 2006; Hogarth et al., 2014; Sikka & Gross, 2014; Murray et al., 2002). The observation of higher PA values later in the day, around 8:00 pm, compared to lower values earlier in the day, around 8:00 am, is consistent with previous findings demonstrating a bimodal, diurnal PA pattern, although the exact reasons for this pattern remain unclear. In direct contrast, results for circadian variation in NA show an inverse pattern of lower NA levels observed later in the day, around 8:00 pm, compared to higher NA levels earlier in the day, around 8:00 am, showing the opposing oscillation between the two moods. Results correspond to the inverse relationship demonstrated by previous research (Stone et al., 2006; Emens et al., 2020), providing support for the proposed model of circadian variation in PA and NA. Additionally, indicating a successful replication in the present study, apart from some local variation in study parameters such as time zone, location, sampling intervals, and sample population (Goel et al., 2013; Hogarth et al., 2014).

Furthermore, the present study also added to the existing body of research on the circadian rhythm in NA, demonstrating variation in Negative Affect across a 24-hour period. The pattern of Negative Affect observed provides support for the prevailing theory that suggests negative moods are primarily experienced during the circadian night (Emens et al., 2020; Dzogang et al., 2017; Walker et al., 2020). In addition, results demonstrated the highest average reported value at 12:00 am, and lowest at 8:00 pm, in a temporal pattern consistent with our hypothesis as well as previous research reporting the existence of a circadian effect on NA

(Emens et al., 2020; Boivin et al., 1997; Miller et al., 2015). Notably, the comparisons of Cohen's d effect sizes between different timepoints were found to be nonsignificant, suggesting that measuring NA during nighttime may pose certain challenges (Emens et al., 2020). Findings on the NA diurnal pattern were also able shed light on the established link between sleep quality and mood (Golder & Macy, 2011; Goel et al., 2013; Walker et al., 2020). This can be seen in results which demonstrated NA to be at its lowest in the morning hours, gradually rising into the afternoon, and reaching a peak at 12:00 am. It is possible that this pattern indicates support for the postulation proposed by previous research which proposes sleep also serves as a form of emotional refreshment, leading to NA lower levels upon waking (Golder & Macy, 2011). Understanding how circadian rhythms affect mood fluctuations and how these, in turn, relate to sleep quality may offer insights into the etiology of mood disorders and guide interventions for improving sleep patterns in individuals with mood disorders (Schnell et al., 2014).

The current study's discovery of elevated average NA values during the circadian night aligns well with previous research associating heightened NA with depressive and ruminative social media posts (ten Thij et al., 2020; Dzogang et al., 2017), as well as increased rates of suicide in the early morning hours. This underscores the importance of circadian rhythms in comprehending the intricacies of negative emotions and their impact on mental well-being (Walker et al., 2020; Dzognag et al., 2018). Like PA, there's observed diversity in pinpointing the specific hour with the highest NA values, influenced by diverse study parameters. Moreover, PA and NA seem to fluctuate independently, with NA displaying less variability than PA. This indicates that PA and NA are distinct entities rather than opposing poles on a single mood spectrum, as hypothesized by some researchers (Watson et al., 1998; Emens et al., 2020). Additionally, neither slope mirrors the other nor follows a parallel course, implying that PA and

NA function through distinct neural systems, potentially requiring different assessment frameworks (Emens et al., 2020; Walker et al., 2020). Consistent with prior research, the absence of significant diurnal fluctuations in NA suggests a persistent factor contributing to the lack of significant evidence for circadian variation in NA is still active in the present study (Goel et al., 2013). These findings challenge the prevailing notion regarding circadian mood variation and emphasizing the viable detectability of circadian variation in NA even with a limited sample size (Murray et al., 2020). Given the akin pattern of NA variation reported by Stone et al. (2006), the integration of an objective measure into the present study's research protocol would have proven helpful in providing validation of results (Kivela et al., 2022). Ultimately, the present results were able to enhance the existing body of knowledge on the circadian effect in NA and contribute to a more comprehensive understanding of mood state fluctuations.

The present study was also able to extend previous research on the circadian effect on mood by including Affection as a distinct mood state. Results demonstrated circadian variation in the observed level of subjective Affection across a 24-hour period, wherein the highest recorded Affection levels were observed at 12:00 pm on Day 1, and lowest levels at 4:00 am, compared to variation observed in PA, which is highest at 4:00 pm, and lowest around 8:00 pm. Notably, levels of Affection at 12:00 pm on Day 1 ($M = 5.92$) and 12:00 pm on Day 2 ($M = 5.46$) are remarkably similar, potentially indicating the effect of day to day variability on diurnal mood fluctuations as well as the influence of individual factors speculated by prior studies (Stone et al., 2006; Emens et al., 2020). Exploring these factors, such as daily routines, social interactions, or sleep quality, may help uncover the cause of the fluctuations exhibited by Affection and provide insights into mood regulation (Walker et al., 2020; Schnell et al., 2014; Claudio & Andrea, 2022). Data analysis also revealed two congruent, unexplained troughs at 8:00 pm and 8:00 am,

potentially indicating the temporal activity of a shared physiological mechanism underlying the drop in subjective Affection (McClung, 2013). Previous researchers have speculated that the shared physiological mechanism could potentially be testosterone, which exhibits peaks in concentrations around such similar times (Kelly et al., 2022; Bode & Kushnick, 2021). However, the pattern of circadian variation observed for Affection did not resemble that of testosterone, which exhibits higher levels in the morning after waking as well as following the onset of sleep, at which time the lowest levels are observed (Bribiescas & Hill, 2010; Bode & Kushnick, 2021; Kelly et al., 2022).

While the congruent troughs cannot be attributed to parallel fluctuations in testosterone, it remains crucial to identify potential internal or external factors contributing to the similar temporal activity (Sikka & Gross, 2014). Comprehensive understanding of the interactions between circadian mood related elements highly salient to expanding contemporary understanding of mood fluctuations in individuals with mood disorders. Results demonstrate another divergence in the circadian variation of Affection and PA, where Affection displayed higher numerical values at 8:00 pm and lower values at 8:00 am, while PA exhibited the opposite trend with higher values at 8:00 am and lower values at 8:00 pm. Moreover, the slope of change or pattern of circadian variation observed for Affection did not run parallel to that of PA, possibly indicating differential underlying physiological activity as well as cognitive concepts, which necessitate further investigation. However, it is worth noting that PA and Affection were observed to share a temporal lowest average mood rating at 4:00 am, although it is unclear why. The observed circadian patterns in affective states may have implications for understanding different subtypes of mood disorders (Claudio & Andrea, 2022). To illustrate this point, it is possible that individuals with bipolar disorder, characterized by extreme mood swings (APA,

2013), may experience manic episodes more frequently during the circadian day when PA and Affection are elevated, while depressive episodes might be more common during the night when NA is higher (Walker et al., 2020). This observation is also supported by clinical observations (Schnell et al., 2014) of temporal variation in manic episodes and underscores the potential relevance of circadian patterns in mood disorder subtyping and diagnostic accuracy.

Lastly, the present study extended previous research on the circadian effect on mood states and demonstrated circadian variation in the observed level of subjective Annoyance across a 24-hour period. Results revealed that generally, Annoyance did not follow the same pattern as NA despite being considered a substate of the broad NA mood state (Rusting & Larson, 1998), in that Annoyance was lower earlier relative to later in the day, while NA was higher earlier relative to later in the day. While, notably, the observed level of Annoyance was highest at 12:00 am, at the same time as the highest NA levels recorded, the time of lowest recorded levels and overall fluctuation in mood were incongruent. Findings of a discrete pattern of variation in Annoyance support prior speculation from researchers that the inclusion of related mood items within an NA composite score may be impacting ability to measure circadian variation in NA (Emens et al., 2020; Stone et al., 2006). Moreover, this divergence may indicate the differential reliance of Annoyance and NA mood states on separate systems of neural or hormonal pathways (Bechtel, 2015). This discrepancy may also suggest a cognitive, conceptual difference between the two mood states which necessitates separate analysis, as NA was represented in the present study by “sad” and “gloomy” feelings (Goel et al., 2013; Murray et al., 2020). Understanding the reasons behind this incongruity and its potential implications for mood disorders, particularly in relation to emotional dysregulation, warrants further investigation. While there is no strong evidence from pre-existing studies for circadian variation in mood states outside of depression,

anxiety, happiness, and sadness, the present study provides initial evidence for circadian variation in additional mood states.

In summary, the results offer intriguing insights into the temporal dynamics of affective states, day-to-day variability, potential relationships with sleep quality, and the relevance of circadian patterns to different subtypes of mood disorders. Results confirm that mood states, such as Positive Affect (PA) and Negative Affect (NA), exhibit circadian patterns with distinct peaks and troughs at different times of the day. The observed effect of circadian variation on affective states suggests that circadian rhythms play a significant role in regulating mood (Walker et al., 2020). Moreover, these findings affirm the established perspective that mood states vary in accordance with the biological clock, strengthening the link between CR and mood fluctuations (Claudio & Andrea, 2022; Kivela et al., 2022). This is consistent with the notion that mood disorders are associated with disruptions in the sleep-wake cycle, a key component of circadian rhythms (Franzen et al., 2008; Schnell et al., 2014; McClung, 2013; Grierson et al., 2016). The present results begin to shed light on the fluctuations in mood seen in mood disorders by demonstrating circadian patterns in affective states and their potential relevance to mood disorder etiology. Addressing these observations in future research may contribute to a deeper understanding of mood disorders and guide the development of targeted interventions and personalized treatment approaches (Schnell et al., 2014).

Limitations and Future Direction

In this study, we acknowledge both the strengths and limitations of our research. While the study had several strengths, including the use of an effective unmasking protocol, it also faced certain limitations. The most significant of these limitations was the small sample size, which had repercussions on the statistical power, the likelihood of type two errors, our ability to

identify outliers, and the overall variability within the sample. Consequently, the outcome of all statistical analyses became less robust, making it challenging to determine the significance of the differences between timepoints in support of circadian rhythm patterns (Grierson et al., 2016).

Furthermore, given the exploratory nature of our study, we were unable to pinpoint outlier data points, thereby hindering our capacity to mitigate any undue influence or errors originating from individual data points. This is evident from the fact that our data exhibited limited circadian variability for all moods, with only a small proportion of individual mood variances reaching significance. However, it is worth noting the observation of small to medium Cohen's d effect sizes between average mood state ratings and a temporal change in slope for Positive Affect and Negative Affect which aligned with previous findings (Sikka & Gross, 2014). Whether these results were influenced by our sampling method or small sample size remains uncertain.

Additionally, our study did not include an objective, biophysiological measure of mood to complement the subjective measure employed, which resulted in the inability to provide further evidence for the reliability of composite mood scores (Grierson et al., 2016). Nonetheless, previous research has established well-documented circadian effects on other physiological variables, such as body temperature across the same 24-hour circadian period investigated in the present study (Schnell et al., 2014; McClung, 2013). Future research should explore causality, employ longitudinal study designs, and investigate potential reasons behind our findings by incorporating cortisol and testosterone measures in addition to subjective mood reports. Such longitudinal studies may also aim to establish which circadian mood patterns are linked to an increased vulnerability to developing mood disorders.

Our research expands upon previous studies which emphasized the importance of repeated measures and an even, frequent, sampling series over a 24-hour period in order to allow for the observation of increased variation to more effectively unmask circadian patterns (Goel et al., 2013; Grierson et al., 2016). By utilizing a relatively homogeneous sample of Montclair State University undergraduates, we aimed to reduce participant variability and enhance our ability to detect circadian variation in Negative Affect (Stone et al., 2006; Kivela et al., 2022). Our circadian 24-hour unmasking protocol appeared successful in assessing mood variation patterns, with results closely resembling those of previous studies (Goel et al., 2014). Future research endeavors should include multiple 24-hour sampling windows with varying start and stop times, different sampling times, and varying sampling frequencies to assess the impact of sampling methods on circadian variation patterns.

Moreover, the present study does not assume a significant limitation related to participant health background, unlike other research studies where physical or mental health conditions can affect results (Hogarth et al., 2014). The potential risk of psychiatric illness influencing the results is assumed to be mitigated by the comparison of average mood rating values at each Timepoint to a baseline value assessed prior to the specific Timepoint value being examined. These comparisons ensure that any change in mood ratings is interpreted relative to a baseline value, thereby normalizing values that might otherwise exert undue influence on results compared to fluctuations observed in non-clinical populations (Hogarth et al., 2014). However, future work should aim to replicate these findings with more diverse populations, including women, who may exhibit different hormonal regulation patterns and emotional states throughout the day (Stone et al., 2006).

The present study is additionally limited by individual differences as a result of the exclusion of an evaluation of individual chronotype, a factor known for its inter-individual variability and established impact on CR (Rusting & Larson, 1988; Miller et al., 2015). A lack of consideration for the influence of chronotype on individual circadian fluctuation in mood makes it challenging to distinguish whether the results observed stem from individual differences or a genuine pattern of circadian variation (Taillard et al., 2003; Schmal et al., 2020). To address this, future research should ensure to incorporate a measure of morningness-eveningness (Smith et al., 1989) in participant screening criteria. This inclusion would ensure a balanced representation of individual differences across participants and facilitate categorization based on chronotype during analysis, thereby eliminating its potential influence on the study outcomes (Smith et al., 1989). With the additional data provided by chronotype assessment, research could also delve into the relationship between CR patterns and vulnerability to mood disorders depending on chronotype profile (Walker et al., 2020; Bechtel, 2015).

Furthermore, it's plausible that our results were influenced by environmentally dependent factors, such as the time of year or season, which can affect endogenous circadian rhythm patterns (Golder & Macy, 2011). While one participant was tested in February, a time when reduced sunlight exposure asserts a marked impact on CR, the remaining twelve participants were tested after the spring equinox on March 20, 2023, marking the start of increased photoperiod. Despite the assumption of equal distribution of differences due to change in photoperiod, future studies should strive to collect data within each seasonal photoperiod to better understand the degree of such influences (Albrecht, 2010). Related to this, previous research has established the influence of entrainment on CR by factors including the light-dark cycle, ambient temperature, and food availability (Renfiniti, 2020). Consequently, future studies

should examine meal timing in relation to participant responses and data collection that could account for possible effect of circadian-modulated starvation on results observed.

Generally, to enhance the robustness of results, future replications should involve larger sample sizes, the determination of outlier cutoffs, and the exploration of individual differences in subjective emotional awareness, allowing for the confirmation of current results. Moreover, future research should focus on defining parameters for circadian misalignment based on gender and age group to investigate patterns of circadian variation in mood associated with an increased incidence of psychiatric illnesses (McClung, 2013; Walker et al., 2020). While there is limited research on the many suspected distinct profiles of circadian variation across time, there is even less information on the specific variables and values associated with circadian variation in properly aligned, disrupted, or misaligned states (Schnell et al., 2014; Walker et al., 2020). Therefore, future research should aim to develop a set unambiguous terminology and quantification of variables related to the description of CR features observed in dysregulated, disrupted, and misaligned circadian states. The definition of such features represents a priority in the ability to improve diagnostic accuracy and generate time-dependent or circadian-focused therapies paramount to tackling the globally soaring rates of mood disorders (Chellapa et al., 2020; WHO, 2019; Salgado-Delgado et al., 2011; Riemann et al., 1991).

In conclusion, findings from the present study generally replicate results reported by previous research (Stone et al., 2006; Emens et al., 2020; Sikka & Gross, 2014) demonstrating the influence of CR on both Positive and Negative Affect, thus illustrating the influence of circadian rhythms on mood. Additionally, the current results suggest that circadian effects may extend to further mood states including but not limited to, Affection and Annoyance, which warrants further exploration. The results underscore several gaps in our understanding of the

relationships between circadian rhythms, mood, and mood disorders. While the presence of circadian patterns is evident, the precise mechanisms and causal relationships are not yet fully elucidated (Walker et al., 2020). These findings call for further research to investigate the complex interplay between CR, sleep-wake cycle, and mood, with the goal of shedding additional light on novel therapeutic strategies as well as insights into the etiology of mood disorders (Salgado-Delgado et al., 2011; Murray et al., 2020; Liu et al., 2020). Researchers should also consider building on the current study's findings by using a longitudinal study design with a large, diverse sample population, frequent sampling intervals, and extended sampling windows for a comprehensive assessment of circadian effect on mood. Furthermore, future studies should continue use of the present study design regarding the remote and real-time nature of data collection, which enhanced ecological validity as well as reduced potential data contamination from recall bias (Goel et al., 2013).

From a clinical perspective, these results have implications for temporal considerations within the context of mood disorder diagnostic accuracy (Schnell et al., 2014). Our results indicate the importance of considering time-of-day factors in diagnosing mood disorders, which may help differentiate normal circadian rhythmicity from pathological states. For instance, individuals exhibiting feelings of NA, such as sadness, early in the day may not necessarily be indicative of a pathological state, as it may align with the individual, circadian rhythmicity (Walker et al., 2020). While conversely, the presentation of elevated levels of NA later in the day could signify a more severe clinical condition. Future research should confirm which features of mood CR represents a factor of mental health vulnerability to the development of clinical mood disorders, which could provide innovative mechanisms and novel insights into the efficacy of circadian-focused therapies. Our research contributes to the limited body of recognition on the

significance of circadian dysregulation as a biological risk factor in mood dysregulation, specifically among vulnerable populations such as shift workers (Cho et al., 2019; Chellapa et al., 2020; Schwarz et al., 2019). By uncovering the impact of circadian dysfunction on emotional wellbeing, we can pave the way for the development of evidence-based interventions with improved efficacy, focused on sleep and circadian regulation, such as proper light exposure and sleep hygiene (Walker et al., 2020; Claudio & Andrea, 2022; Kivela et al., 2022; Sikka & Gross, 2014). These interventions would aim to enhance the quality of life, a vital component for promoting optimal mood and safety among individuals working in high-risk settings (Torquati et al., 2019).

Ultimately, optimizing circadian rhythm to align subjective mood with environmental factors could improve the efficacy of mood disorder interventions, reduce the public health burden, promote population equality, mitigate shift-work accidents, and decrease the risk of premature mortality associated with the increasing prevalence of psychiatric illnesses (WHO, 2019; Schnell et al., 2014; Walker et al., 2020; Bechtel, 2015). The knowledge contributed by the present research signifies a promising step in paving the way for enhanced, evidence-driven personalized mental healthcare. The active monitoring and precise modification of dysregulated circadian rhythms sets the stage for a more tailored and effective approach (Schnell et al., 2014). Integrating detailed, longitudinally informed recommendations and time-targeted therapies presents a powerful clinical tool to tackle the increasing prevalence of mood disorders (Sirignano et al., 2022; Goel et al., 2013; Fishbein et al., 2021). This strategy not only allows for customized treatment but also opens doors to refining intervention schedules and temporally dependent diagnostic criteria (Claudio & Andrea, 2022). Ultimately, the methodology and findings

presented by the present study represents a beacon of hope for advancing evidence-based, individualized medicine within the field of mental health.

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Footnotes

Footnote 1: Because there were so few subjects, a repeated measures analysis was not appropriate (as per discussion with Dr. Askew).

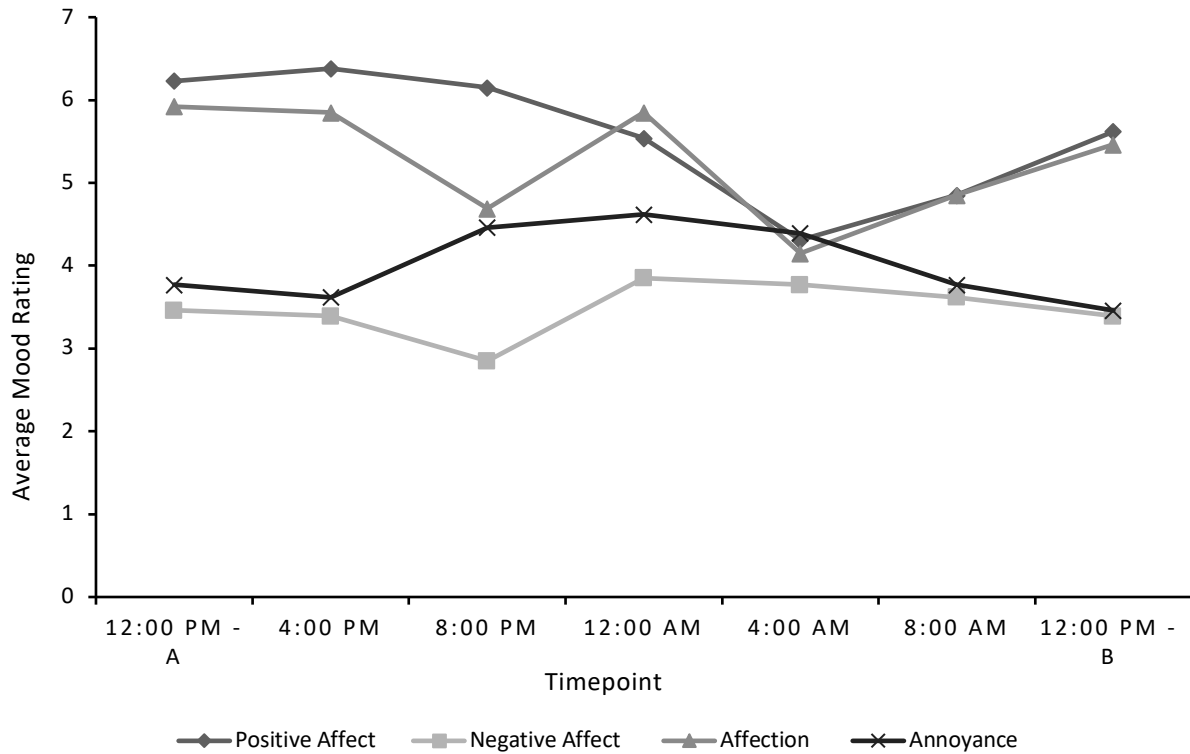
Table 1*Sociodemographic Characteristics of Participants*

Characteristic	<i>n</i>	<i>M</i>	<i>SD</i>	Minimum	Maximum	Range
Age	13	19.7	2.5	18	26	8

Table 2*Descriptive Statistics of Mood States Across 24 Hours*

Mood State	Timepoint						
	12:00 PM - A	4:00 PM	8:00 PM	12:00 AM	4:00 AM	8:00 AM	12:00 PM - B
Positive Affect	6.23 (1.24)	6.38 (1.39)	6.15 (1.57)	5.54 (0.88)	4.31 (1.55)	4.85 (1.00)	5.62 (1.19)
Negative Affect	3.46 (1.20)	3.39 (0.96)	2.85 (1.07)	3.85 (0.98)	3.77 (1.42)	3.62 (1.50)	3.39 (1.33)
Affection	5.92 (1.19)	5.85 (1.28)	4.69 (2.02)	5.85 (1.14)	4.15 (1.86)	4.85 (1.68)	5.46 (1.20)
Annoyance	3.77 (1.48)	3.62 (0.77)	4.46 (1.76)	4.62 (1.12)	4.39 (1.85)	3.77 (1.48)	3.46 (1.39)

Note. Values represent means and standard deviations; *SD* are presented in parentheses. "A" and "B" refer to different days of participation, where mood state data was recorded for the same timepoint. "A" refers to day 1 of participation, while "B" refers to day 2.

Figure 1*Change in Average Mood Rating Over 24-Hour Period*

Note. This figure displays variation in average participant mood rating for each of the four investigated mood states, over a 24-hour period. A visual representation of the fluctuations in each mood state is provided, where all four mood states were assessed seven times at equal intervals, offering insight into diurnal patterns of variation. "A" and "B" refer to different days of participation, where mood state data was recorded for the same timepoint. "A" refers to day 1 of participation, while "B" refers to day 2.