1

Introduction to Circadian Rhythms

Laura K. Fonken and Randy J. Nelson

1.1 Introduction

For the past 3–4 billion years, life on Earth evolved under the predictable pattern of solar days; that is, exposure to light during the day and dark at night. Temporal constraints are obvious when considering the "rules of life." That is, individuals cannot do everything all the time. For instance, energetic requirements are somewhat continuous, whereas energy production or consumption is somewhat sporadic. All organisms partition temporal energetic activities. Indeed, temporal partitioning of photosynthesis, metabolism, gene expression, reproduction, defense, growth, activity, and inactivity is universal among plants and animals. During the evolution of life, organisms internalized the temporal rhythm of Earth's rotation and eventually developed self-sustaining biological clocks. These internal rhythms with periods of approximately 24 hours are called circadian rhythms, and the structures that generate them are called circadian clocks. A human's primary circadian biological clock is a paired cluster of about 20,000 nerve cells in the hypothalamus at the base of the brain, called the suprachiasmatic nucleus (SCN). The period of a circadian clock is approximately 24 hours, but daily light exposure sets it to precisely 24 hours. Having our clocks set closer to our environment's light–dark rhythms optimizes how our bodies function and how we behave.

Circadian clocks are a nearly universal feature of life on this planet, yet over the past century and a half we have managed to manipulate the amount of light in the environment so much that we are disrupting them. As we will learn throughout this book, either too much light exposure at night or too little light exposure during the day can disrupt central and peripheral timing mechanisms, how internal rhythms are entrained to the external environment, and the typical and optimal 24-hour physiological and behavioral functioning of individuals.

1.1.1 Central Pacemaker in the Suprachiasmatic Nucleus

Circadian rhythms in mammals are ubiquitously expressed throughout the body and are regulated by a hierarchy of independent self-sustaining molecular and cellular

https://doi.org/10.1017/9781009057646.002 Published online by Cambridge University Press

clocks. This hierarchy is entrained by external Zeitgebers ("time givers") including light (primary), food, exercise, and even social cues. Rhythms throughout the body are subsequently maintained in a synchronized manner via intermediary neural and humoral cues. But where are these signals initiated? The primary pacemakers in mammals are the paired suprachiasmatic nuclei (SCN) that govern rhythms throughout the brain and body. The SCN are located directly above the optic chiasm in the anterior hypothalamus and contain a diverse cellular make-up. SCN neurons produce the inhibitory neurotransmitter gamma aminobutyric acid (GABA) and various neuropeptides including arginine vasopressin (AVP), cholecystokinin (CCK), gastrin-releasing peptide (GRP), prokineticin 2 (Prok2), and vasoactive intestinal polypeptide (VIP) (reviewed in Moore et al., 2002; Patton & Hastings, 2018). The SCN comprise two distinct regions with unique neuropeptide expression: the ventrolateral "core" contains neurons that express VIP and GRP, whereas the dorsal shell contains neurons that express AVP and CCK.

SCN neuron firing is tightly synchronized in "core" and "shell" regions through neural connections and timed release of these key neuropeptides (Patton & Hastings, 2018). VIP is an important synchronizer of neuronal networks in the SCN (Abrahamson & Moore, 2001); mice lacking VIP or VIP receptor 2 (VPAC2) exhibit attenuated behavioral rhythms and desynchronized circadian rhythms in cultured neurons from the SCN (Aton et al., 2005; Colwell et al., 2003; Harmar et al., 2002). Interestingly, in SCN neurons with the VIP or VPAC2 genes knocked out, circadian rhythms are restored by co-culture with neurons from a wild-type SCN, suggesting that other molecules such as AVP also synchronize rhythms in the SCN (Maywood et al., 2011). Indeed, an AVP receptor antagonist prevents restoration of rhythms in VPAC2 knockout SCN neurons (Maywood et al., 2011).

Rhythms in the SCN are primarily entrained by light information that is communicated directly from the retina through the retinohypothalamic tract to the SCN (Beier et al., 2021; Hattar et al., 2006; Moore & Qavi, 1971). In addition to retinal input, the SCN core receives input from the thalamus and raphe nucleus and the shell receives input from the hypothalamus, neocortex, and brainstem (Fernandez et al., 2016; Leak & Moore, 2001).

The SCN have unique circadian-focused properties that define them as the primary pacemaker: they receive direct retinal light input; neurons in the SCN have topographically organized coupling mechanisms, which allow them to remain synchronized in the absence of light input (Aton & Herzog, 2005); The SCN are protected from feedback by systemic clock-modifying factors such as glucocorticoids or feeding (Schibler et al., 2015); SCN lesions abolish circadian rhythms throughout the body (Moore & Eichler, 1972; Stephan & Zucker, 1972; Weaver, 1998); electrical and chemical stimulation of the SCN induce phase shifts (Albers et al., 1984; Rusak & Groos, 1982); and transplanting an SCN into an SCN-ablated animal restores circadian activity (Silver et al., 1996). Furthermore, cultured SCN tissue will maintain

long-term (>1 month) oscillations in the absence of external stimulation (Welsh et al., 1995; Yamazaki et al., 2000; Yoo et al., 2004). Thus, the SCN features direct retinal input, synchronized output, and few peripheral feedback mechanisms, thereby optimizing this brain region to act as the primary circadian oscillator. Additional details about the central clock dynamics are provided in Chapter 2.

1.2 Molecular Mechanisms of the Circadian Clock

At the molecular level, cellular circadian rhythms are formed from interlocking transcriptional–translational feedback loops (TTFL) that drive spontaneous oscillations of gene and protein expression with an approximately 24 hour period. Remarkably, the molecular clock components are expressed rhythmically in nearly every cell of the body and are entrained by signals from the primary clock. The core components of this loop involve the induction of Period (*Per1*, *Per2*, and *Per3*) and Cryptochrome (*Cry1* and *Cry2*) gene expression through E-box enhancers by the transcriptional activators circadian locomotor output cycles kaput (*CLOCK*) and brain and muscle arnt-like protein 1 (*BMAL1*) (Gekakis et al., 1998; Hogenesch et al., 1998; Jin et al., 1999). Per and Cry proteins accumulate in the cytoplasm and then form large multimeric complexes which translocate back to the nucleus to interact with CLOCK and BMAL1 and repress their own transcription (Griffin et al., 1999; Kume et al., 1999; Lee et al., 2001). Progressive degradation of the existing inhibitory complexes then occurs, ultimately leading to renewed transcription of *Per* and *Cry*. This feedback loop takes approximately 24 hours to complete a cycle. There are additional feedback loops interlocked with the CLOCK-BMAL1/Per-Cry loop. A prominent loop involves the activation of the retinoic acid receptor-related orphan receptor (ROR) and REV-ERB by the CLOCK-BMAL1 complex which feeds back on BMAL1 to stabilize rhythms (Preitner et al., 2002). Deletion of "core clock genes" (or, in some cases, 2+ paralogs of clock genes) reveals their requirement for rhythms in activity (Bunger et al., 2000; Cox & Takahashi, 2019; Vitaterna et al., 1994).

1.2.1 The Circadian System Is Synchronized to the Environment

Circadian rhythms oscillate at approximately but not exactly 24 hours. Variations in rhythm period occur at the whole organism level down to the individual cellular level (Czeisler et al., 1999). For example, humans display different "chronotypes," meaning some people are early risers or "larks" and others prefer to stay up later and are known as "night owls." These variations in preferred sleep and wake time are associated with distinct endogenous circadian periods: morning larks tend to have circadian rhythms that are shorter than night owls (Roenneberg et al., 2003). A number of genetic factors are associated with distinct chronotypes that include genes related to circadian regulation, as well as glutamate and insulin signaling

https://doi.org/10.1017/9781009057646.002 Published online by Cambridge University Press

pathways (Jones et al., 2019). Input to the circadian system is essential for maintaining everyone on the same 24 hour schedule.

1.2.2 Light Is the Dominant Entrainment Factor for the SCN

Light is the primary entrainment factor for synchronizing circadian rhythms to the 24 hour day. The major neural route of light entrainment occurs via activation of specialized cells in the retina called intrinsically photosensitive retinal ganglion cells (ipRGCs) (Berson et al., 2002; Provencio et al., 2002). ipRGCs are a population of non-vision forming cells that are critical in transducing light information via the retinohypothalamic tract to the SCN (Beier et al., 2021; Hattar et al., 2006; Moore & Qavi, 1971). Prior to the discovery of ipRGCs approximately 20 years ago, the existence of a non-vision forming cell was suspected as some individuals that lacked visual awareness maintained circadian rhythmicity and melatonin responses to light (Czeisler et al., 1995). Moreover, responses to light in animals were poorly explained by the properties of rods and cones (e.g., Brainard et al., 2001; Mrosovsky et al., 2001; Takahashi et al., 1984).

ipRGCs have extensive arbors and are activated by short (blue to humans) wavelengths of light due to their expression of the photopigment melanopsin (Do, 2019). Light activates melanopsin to trigger a G protein cascade, causing membrane depolarization and the release of glutamate and the neuropeptide pituitary adenylate cyclase activating peptide (PACAP) (Hannibal et al., 2002). Although the number of ipRGCs in the mammalian retina is limited, they display remarkable heterogeneity with six different morphological subtypes, M1–M6 (reviewed in Do, 2019). The distinct subtypes of ipRGCs are thought to regulate specific light intensity or times of day responses. The sensitivity of ipRGCs specifically to blue light has led to an interest in regulating the circadian system by manipulating the wavelength of light environment. Studies in humans have shown that high intensity blue light can be disruptive to the circadian system, resulting in melatonin suppression and sleep loss (Chang et al., 2015; Hanifin et al., 2019; West et al., 2011). This has led to blue light filters in technology (e.g., laptops and smartphones) that eliminate these wavelengths of light at night. However, it is important to note that filtering out blue light is not a cure all: ipRGCs also receive input from rods and cones. Melanopsin knockout mice maintain some circadian responses to light, but mice lacking melanopsin coupled with disabled rod and cone phototransduction do not (Hattar et al., 2003).

Light input is transduced from an electrical signal – via propagation along the retinohypothalamic tract – to a chemical signal when the tract terminates in the SCN with the release of glutamate. Glutamate acts on NMDA receptors to increase calcium release in SCN neurons (Ding et al., 1997). This increased calcium activates the transcription factor CREB to increase Per transcription (Gau et al., 2002; Ginty et al., 1993; Schurov et al., 1999). For instance, a brief flash of light during the inactive

phase induces *de novo* expression of *Per* (Albrecht et al., 1997). Through these mechanisms, the SCN are optimized in mammals to link light timing in the environment with physiologic function.

1.2.3 Food, Exercise, and Other Factors Regulate Peripheral Circadian Rhythms

Although most strongly synchronized by light, the circadian system is also entrained by other factors. Early observations by Richter (1922) characterized anticipatory activity in response to timed feeding: rats fed one meal per day increase wheel running several hours prior to food availability. Indeed, both feeding (Boulos et al., 1980) and exercise (Edgar & Dement, 1991) can entrain circadian rhythms in locomotor activity. Moreover, when maintained in constant lighting conditions, rodents will also synchronize activity rhythms based on social cues (Paul et al., 2015). As discussed in Section 1.3, non-photic entrainment factors are typically more salient for extra-SCN clocks.

1.3 Circadian Rhythms Occur Throughout the Body: Extra-SCN Tissue-Specific Clocks

In addition to the central clock in the SCN, individual organs and cells outside the SCN rhythmically express core clock genes. These are termed peripheral or extra-SCN tissue-specific clocks. Individual cells contain autonomous clocks; importantly, all cells work in concert to time the occurrence of physiological events optimally. The SCN regulates peripheral clocks both through indirect and direct means. Direct regulation of peripheral processes by the SCN are evoked by neural or humoral signaling (e.g., Mohawk et al., 2012; Ramanathan et al., 2018). Indirectly, the SCN regulates the peripheral clocks via neural and humoral signaling factors, as well as by modulating the expression of circadian clock genes. For example, the SCN regulates secretion of hormonal cues that synchronize extra-SCN clocks, such as melatonin and glucocorticoids. Glucocorticoids are hormones released by the adrenal gland, and act via the glucocorticoid receptor in nearly all cells to regulate gene expression. Upon binding the cytosolic glucocorticoid receptor, the ligand-receptor complex enters the nucleus and binds to glucocorticoid response elements on DNA to activate or repress gene expression. Importantly, at baseline there are circadian rhythms in glucocorticoid levels and several circadian genes have glucocorticoid response elements in their regulatory regions (Reddy et al., 2007). Application of glucocorticoids to isolated cells can induce Per gene expression and thus serves as an important factor for regulating extra-SCN clocks (Balsalobre et al., 2000; Fonken et al., 2015).

In addition, physiologic cues coordinate or amplify circadian rhythms in extra-SCN cells; these cues include body temperature, feeding, and activity (Schibler et al.,

https://doi.org/10.1017/9781009057646.002 Published online by Cambridge University Press

2015). These neural, humoral, and physiologic factors are sensitive to entrainment by the SCN – but they are also regulated by other systemic factors (S. Zhang et al., 2020).

External events, such as food intake (Damiola et al., 2000) and physical exercise (Ripperger & Schibler, 2001), can indirectly reset peripheral clock rhythms in the liver and elsewhere (Chen et al., 2019; Landgraf et al., 2015). For example, restricting food intake to certain times of day (time-restricted feeding or TRF) can uncouple the SCN and extra-SCN clocks (Damiola et al., 2000). Indeed, timing of food intake strongly regulates the liver clock (Hatori et al., 2012) as well as cardiovascular function (see Chapter 11). Another mechanism of SCN-mediated peripheral clock regulation is by direct modulation of the autonomic nervous system, as described in Section 1.4.

Importantly, synchronizing clock gene expression in these extra-SCN tissues coordinates transcriptional programming of clock-controlled genes (Mavroudis et al., 2018). This means that myriad cellular functions are governed by the clock in peripheral tissues and also suggests that, during pathology, these intermediary circadian synchronizers may be susceptible to harmful perturbation that could desynchronize circadian oscillators. The remainder of this chapter will introduce circadian regulation of several major body systems; specific chapters in this book will then review clock disruption-elicited pathology in these systems.

1.4 Circadian Regulation of CNS Function

Central nervous system (CNS) function of animals displays distinct and overlapping circadian rhythms (Chapter 6). Indeed examples of circadian fluctuations in learning and memory, sensation and perception, attention, food intake, mating behaviors, maternal behaviors, aggression, drug and alcohol seeking behaviors, as well as regulation of locomotor activity have been reported (Nelson et al., 2021). These temporal variations are often overlooked and can significantly affect experimental outcomes. In this section we review some common examples of circadian regulation of CNS function. Notably, disrupted circadian rhythms negatively affect CNS function.

1.4.1 Locomotor Behavior

Early research on circadian rhythms focused on behavior as an output, especially locomotor behavior (Richter, 1922). Monitoring of activity cycles is adapted to the species under investigation. For example, small mammals are kept in a cage equipped with a running wheel connected to a counting device that automatically produces a continuous record of the individual's locomotor activity. The locomotor activity of small birds can be determined by monitoring perch-hopping activities around the

https://doi.org/10.1017/9781009057646.002 Published online by Cambridge University Press

clock. Humans can be equipped with electronic smart devices that transmit their locomotor activities to a central monitoring station.

Individuals of species are typically either diurnal or nocturnal in their locomotor activities. As noted, internal clocks display a period of about 24 hours and are set to precisely 24 hours by exposure to light. In the presence of constant lighting conditions (i.e., dim light, bright light, or darkness), locomotor rhythms display a temporal drift from 24 hours that mirrors the internal period (tau) of the circadian clock and is out of phase with the solar day.

Indeed, observing the locomotor activity of a colony of Syrian hamsters (*Mesocricetus auratus*) led to the discovery of an individual male with a very short tau (~22 hours) when housed in constant dark conditions (Ralph & Menaker, 1988). After a return to typical light–dark conditions, this individual displayed aberrant entrainment properties, beginning its locomotor activity about four hours prior to lights out, when hamsters typically begin their activities. This male was mated with three wild-type females with typical taus and, via standard cross-breeding studies, it was revealed that hamsters heterozygous for the mutation displayed periods of about 22 hours, whereas homozygous hamsters displayed locomotor rhythms with taus of about 20 hours. The tau mutant is encoded by casein kinase I epsilon (CKIε) and was the first gene identified that was associated with mammalian circadian rhythms (Lowrey et al., 2000). Animals display species-specific times of locomotor activity onset that are often linked to the timing of food intake, water consumption, and reproductive behavior, and have been a critical tool for understanding the genetics and other properties of circadian rhythms. Gene expression patterns are temporally similar in both nocturnal and diurnal animals (Challet, 2007).

1.4.2 Cognition

There are strong daily rhythms in all aspects of cognition (Fisk et al., 2018; Schmidt et al., 2007; Smarr et al., 2014) (Chapter 6). Generally, memory formation peaks during individuals' active periods. Thus, rats and mice tend to display optimal memory for performance in the Morris water maze during the night, whereas diurnal grass rats display best memory performance during the day (Krishnan & Lyons, 2015; Martin-Fairey & Nunez, 2014).

In rodents, both sensory sensation and perception vary across the day. For example, visual sensation and perception and auditory sensation and perception change across the day in humans and nocturnal rodents (e.g., Basinou et al., 2017; Finlay & Sengelaub, 1981; Meltser et al., 2014). Tasks requiring attention display significant circadian fluctuations in both humans (van der Heijden et al., 2010) and rodents (Gritton et al., 2012). These fluctuations appear to reflect circadian changes in cholinergic activities (Hut & Van der Zee, 2011).

1.5 Circadian Regulation of Cardiac Function

Cardiac function is regulated by circadian rhythms (Liu et al., 2021; Melendez-Fernandez et al., 2021; Thosar et al., 2018) (Chapter 13). Cardiovascular regulation is associated with sleep–wake patterns (Bastianini et al., 2012; Smolensky et al., 2007) that are linked to underlying circadian rhythms. The circadian regulation of sympathetic and parasympathetic activation modulates heart rate, heart rate variability, blood pressure, vascular tone, and endothelial function (reviewed in Melendez-Fernandez et al., 2021). This temporal organization allows the vascular system to produce the necessary factors and mediators, such as prothrombotic and antithrombotic factors, and nitric oxide, at the appropriate time of the day to support activity during the active phase or support recovery and replenishment during the inactive phase. Dysregulation of these circadian fluctuations in cardiac function has been associated with cardiovascular pathology including myocardial infarction, ventricular tachycardia, and sudden cardiac death, which all peak during the early morning (Khan & Ahmad, 2003; Manfredini et al., 2013; Muller, 1999; Muller et al., 1987).

Cells comprising cardiovascular tissue display robust circadian oscillations including vascular smooth muscle, fibroblasts, cardiomyocytes, and cardiac progenitor-like cells, all of which regulate physiological functions including endothelial function, blood pressure, and heart rate (Paschos & FitzGerald, 2010). Disruption of these rhythms is associated with misalignment of cardiovascular dynamics, including endothelial (Etsuda et al., 1999), prothrombotic (Takeda et al., 2007), and clotting (Dalby et al., 2000) factors, which can provoke a pathological response (Rana et al., 2020).

Taken together, the available data indicate circadian regulation of the cardiovascular system. Indeed, peripheral clocks and clock genes are expressed in these tissues (Davidson et al., 2005; Storch et al., 2002). Rhythms in vascular function are also observed at the molecular level. RNA sequencing data indicate that 6 percent and 4 percent of protein-coding genes in the heart and aorta, respectively, are transcribed in a circadian fashion (Zhang et al., 2014). At the cellular level, the core clock genes, including *Bmal1*, *Clock*, *Per*, *Cry*, and *Rev-Erb*, serve an important role in maintaining physiological homeostasis of the cardiovascular system. For example, mice with *Per2* mutations display reduced nitric oxide production and decreased vasodilatory prostaglandins and elevated vasoconstrictors (Curtis et al., 2007). *Cry1/2* deletion leads to salt-sensitive hypertension and increased baroreflex sensitivity in mice (Stow et al., 2012). Given the importance of circadian organization for typical cardiovascular function, the potential of disrupted circadian rhythms for cardiovascular health is dramatic (see Chapter 13).

1.6 Circadian Regulation of Metabolism

Energy acquisition, storage, and utilization are critical for life. Metabolism regulates chemical changes in a cell or organism in order to generate energy or materials

https://doi.org/10.1017/9781009057646.002 Published online by Cambridge University Press

needed to grow, reproduce, and function appropriately. The circadian system helps optimize metabolic processes based on distinct metabolic requirements throughout the day to maintain homeostasis. Circadian rhythms in metabolism persist at multiple levels from the function of cellular mitochondria, to hormonal release, to behavioral rhythms in food intake.

Humans and other organisms face distinct metabolic demands based on time of day. A critical aspect of this is that daily behavior is partitioned into an active (wakefulness) and rest (sleep) phase. For understandable reasons, the majority of food intake occurs during an animals active phase, with circadian fluctuations in hunger and appetite contributing to this time of day difference in feeding (Scheer et al., 2013). There are also differences in cravings for specific foods based on time of day, with an increased preference for higher caloric foods as the onset of the sleep phase approaches (Scheer et al., 2013). This is thought to contribute to the increased risk for obesity and metabolic disorders that occurs in night shift workers – night shift workers are awake and active at a time where their bodies are primed for higher calorie food intake (Bouillon-Minois et al., 2022).

Importantly, metabolic regulation is not simply an output of the circadian system. Food intake feeds back on the clock to reinforce rhythms and to adapt physiology to tissue-specific needs. Along with changes in food intake, there are also time-of-day differences in whole body energy expenditure: metabolic rate is reduced during sleep compared to wakefulness (Fraser et al., 1989). Increases in energy expenditure occur during sleep restriction (although increases in energy expenditure are often countered by increased food intake) (Markwald et al., 2013; McHill et al., 2014). However, when humans are sleep restricted and maintained on bed rest, energy expenditure during the typical sleep phase is still lower than during the early active phase (Jung et al., 2011), suggesting the presence of an underlying endogenous rhythm.

Because of the differences in energy intake and expenditure that occur with predictable daily rhythm, there are also rhythms in the underlying hormonal and cellular processes associated with metabolism. Regulation of key metabolic hormones varies throughout the day both due to circadian regulation and as a direct consequence of timing of food intake. For example, because food intake occurs primarily during the active phase, there are increases in most intermediary metabolites including glucose, amino acids, and lipids in the blood during the active phase (reviewed in Reinke & Asher, 2019). The circadian clock, however, is critical for buffering against excessive fluctuations in metabolic factors. For example, blood glucose is regulated by the circadian system; glucose transporters oscillate in a circadian manner, presumably in anticipation of relative nutrient abundance during the active compared to the inactive phase (Reinke & Asher, 2019). Circadian function in key tissues and cells that mediate blood glucose are critical. Indeed, disrupting clock function in the liver and pancreas impacts glucose regulation (Lamia et al., 2008; Marcheva et al., 2010).

Given this tight regulation between the circadian system and metabolism, it follows that disruption of the circadian clock by genetic or environmental means results in metabolic disruption. Susceptibility to diet-induced obesity in a genetic circadian model was first shown in clock mutant mice (Turek et al., 2005). Subsequently, mutations in many clock linked genes have been associated with metabolic dysfunction (see table 2 in Fonken & Nelson, 2014). Environmental circadian disruption in rodent models, including exposure to light at night (Fonken, Aubrecht, et al., 2013; Fonken et al., 2010; Fonken & Nelson, 2013; Fonken, Weil, et al., 2013), constant light (Coomans et al., 2013; Fonken et al., 2010), simulated shift-work protocols (Barclay et al., 2012; Salgado-Delgado et al., 2013), and non-24 hour light cycles (Karatsoreos et al., 2011), are also associated with metabolic dysfunction. Furthermore, humans that are at risk for circadian disruption by engaging in activities such as shift work are at increased risk for developing metabolic syndrome (Pietroiusti et al., 2010). Perhaps not coincidental, the global obesity epidemic parallels rapid increases in disruptive nighttime light exposure in recent decades. This work is reviewed in Chapter 10. Overall, metabolism and the circadian system are integrally associated.

1.7 Circadian Regulation of Immune Function

The diverse activities in which humans and other animals engage throughout the day come with different risks for encountering pathogens, toxins, and injuries. This suggests that coordinating responses to such threats would also be adaptive, with the immune system representing a major responder. Under healthy conditions, the immune systems may promote a state of anticipation and enhanced vigilance prior to the onset of the active phase, and repair and rejuvenation at the end of the active phase (Curtis et al., 2014).

The immune system differentially responds to challenges based on time of day (Haspel et al., 2020). For example, exposure to the same *E. coli* endotoxin challenge during the active versus inactive phase produces striking differences in mortality: rats that receive *E. coli* endotoxin during their inactive phase exhibit approximately 10 percent mortality versus approximately 80 percent mortality to the exact same dose during the active phase (Halberg et al., 1960). Similarly, humans with rheumatoid arthritis show increased pain and inflammatory markers during the nighttime (rest phase) and early morning (Gibbs & Ray, 2013; Ingpen, 1968; Perry et al., 2009). These changes in immune function are associated with direct circadian regulation of immune cells as well as due to circadian regulation of hormones that gate immune responses (e.g., glucocorticoids).

Recent work has illuminated how the circadian system drives healthy daily rhythms in immune responsivity and migration. Every immune cell examined expresses the circadian clockwork necessary for approximately 24 hour rhythms entrained by intermediary oscillators, and these clock genes refine expression of

immune-related genes. In mouse macrophages, the clock gene *Rev-erba* (e.g., *Nr1d1*) peaks around Zeitgeber time (ZT) 12 (late inactive phase, where ZT represents 24 hours of the day with ZT0 = lights on) (Alexander et al., 2020; Gibbs et al., 2012); this transcriptional repressor inhibits expression of the core clock gene *Bmal1*, while also repressing expression of inflammatory genes. At the nadir of *Rev-erb* expression – during the active phase – genes with REV-ERB regulatory binding sites are derepressed and are present at higher levels. Thus, REV-ERB provides an example of how a clock-related gene can drive daily oscillations in the functional outputs of immune cells.

Similar daily patterns in expression and reactivity are observed for other clock genes and cell types, respectively. Interestingly, there are also rhythms in immune cell release into blood and extravasation into inflamed tissue: most subsets show highest release and migration in mice during the inactive phase, and *Bmal1* deletion in either endothelial cells or leukocyte subsets ablate these migratory rhythms (He et al., 2018). The existence of this intrinsic daily rhythm in immunity suggests it may have an adaptive benefit for optimally balancing preparation of immunity for experiencing infection or injury during active phases versus undergoing rejuvenation or repair during rest (Westwood et al., 2019).

1.8 How Do We Disrupt the Clock?

As noted, circadian rhythms can be entrained by several external cues, primarily light exposure and food intake; thus, disrupting these entraining cues can perturb circadian regulation of physiology and behavior (Vetter, 2020).

1.8.1 Environmental Lighting

As mentioned, life evolved on Earth with an internal timing system aligned with bright days and dark nights. The invention of electric lighting approximately 150 years ago initiated social and economic revolutions, but also effectively ended completely dark nights (Figure 1.1). Artificial light currently floods the skies with a night glow known as "light pollution." Light pollution is defined as the alteration of natural night light caused by anthropogenic sources of light (Falchi et al., 2016). According to Falchi et al., 80 percent of the world and over 99 percent of Europe and the United States live under polluted night skies. Sources of outdoor artificial light include vehicles, buildings, street and traffic lights, and signs. Of course, light has also been brought indoors; sources of indoor artificial light includes light bulbs, TVs, computer screens, e-books, tablets, phones, and other electronic devices. Incandescent light bulbs initially emitted light of a full spectral composition. Technological advances and ecological concerns have driven the development of more cost- and energy-efficient sources of light, *viz*., light-emitting diodes (LEDs). The spectral composition of LED lights negatively affect the

https://doi.org/10.1017/9781009057646.002 Published online by Cambridge University Press

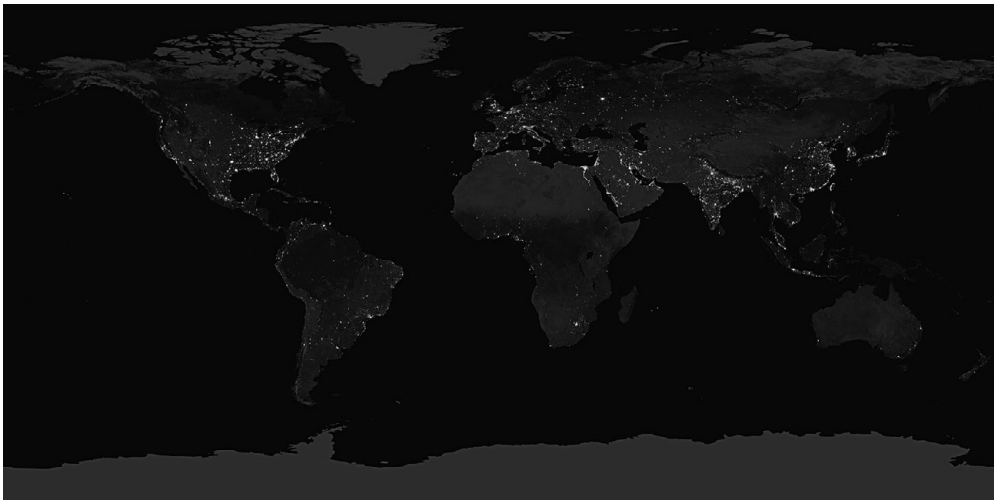

Figure 1.1 Satellite image illustrating nighttime skyglow across the globe.
Credit: NASA Earth Observatory images by Joshua Stevens, using Suomi NPP VIRS data from Miguel Roman, NASA's Goddard Space Flight Center.

environment (Gaston et al., 2015), as well as circadian rhythms. LEDs emit light spectra with a short-wavelength peak, that, as previously described, coincides with the maximal sensitivity of melanopsin, the primary photopigment that conveys environmental light information to the SCN to entrain circadian rhythms. Activation of this pathway during the evening or night may disrupt the internal cycle of clock gene expression/interactions, reset the circadian clock(s), and lead to disrupted circadian rhythms in humans (Brown et al., 2022) as well as other animals exposed to anthropogenic light pollution (see Chapter 15).

1.8.2 Night Shift Work, Jet Lag, and Social Jetlag

Night shift work, jet lag, and social jet lag all combine altered exposure to light as well as altered timing of food intake; as such, these factors can potently and significantly disrupt circadian rhythms. Night shift work has become common across the globe coincident with increased use of electricity, light at night, and industrial development. Among Americans and Europeans, approximately 15 and 30 percent of the population work night shifts, at least part time (Boivin & Boudreau, 2014). Although many economic and other benefits arise from night shift work, many longitudinal studies have reported that night shift work disrupts circadian rhythms and is associated with negative consequences on health and wellness (e.g., Dutheil et al., 2020; Hansen, 2017; Q. Zhang et al., 2020). Use of animal models of night shift work has revealed a causative effect on several diseases (e.g., Arble et al., 2010; Evans & Davidson, 2013; IARC Monographs Vol 124 group, 2019).

https://doi.org/10.1017/9781009057646.002 Published online by Cambridge University Press

Another relatively recent technological development that can dysregulate circadian rhythms is travel by jet. Jet travel across four or more time zones induces a syndrome termed jet lag. Jet lag occurs in response to simultaneous shifts in zeitgebers that desynchronize internal circadian rhythms. Symptoms include sleep disruption, disruption of digestive processes, impaired psychological processes, including attention, perception, and motivation, as well as a general feeling of malaise. Most people report that jet lag is worse on eastward compared to westward flights (Herxheimer, 2014). However, extensive jet travel and jet lag is relatively uncommon among the general population.

However, a phenomenon termed social jetlag is relatively common among us. Social jet lag is defined as the difference in the time of sleep onset on work days compared to the time of sleep onset on so-called free days (e.g., weekends (Roenneberg et al., 2003; Sudy et al., 2019)). Thus, it is common for people to shift their wake–sleep and other circadian rhythms by 3–6 hours in both directions every weekend voluntarily. Both chronic night shift work and social jet lag uncouple central and peripheral clocks and impair physiological and behavioral functioning.

1.9 Conclusions

Billions of years of daily light–dark cycles led to the evolution and refinement of the circadian system. In mammals, the primary pacemakers are the SCN, which help entrain peripheral clocks via secreted cues and direct nervous system input. These cues, in combination with extra-SCN signals, control the timing of molecular clocks in nearly every cell of the body. Molecular clocks also regulate cell-specific processes, leading to circadian regulation of nearly every bodily function – here, we introduced how the circadian system regulates homeostasis throughout the body by regulating the function of major body systems including the CNS, metabolism, the cardiovascular system, and immunity. Circadian regulation of physiology is adaptive, as it optimizes body functions for predictable daily activities during active–inactive cycles. Unfortunately, circadian function in humans and other animals are disrupted by technologies developed over the past 150 years, such as artificial light at night. Overall, the circadian system is an extraordinary and evolutionarily conserved feature of animals on Earth that helps optimize physiology and biological output for the time-of-day. Future research will further explore how the circadian system is interwoven with nearly every bodily system and how this ubiquitous system can be manipulated to improve health and survival of life on Earth.

References

Abrahamson, E. E., & Moore, R. Y. (2001). The posterior hypothalamic area: Chemoarchitecture and afferent connections. *Brain Res*, *889*(1–2), 1–22.

Albers, H. E., Ferris, C. F., Leeman, S. E., & Goldman, B. D. (1984). Avian pancreatic polypeptide phase shifts hamster circadian rhythms when microinjected into the suprachiasmatic region. *Science*, *223*(4638), 833–835.

https://doi.org/10.1017/9781009057646.002 Published online by Cambridge University Press

Albrecht, U., Sun, Z. S., Eichele, G., & Lee, C. C. (1997). A differential response of two putative mammalian circadian regulators, mper1 and mper2, to light. *Cell*, *91*(7), 1055–1064.

Alexander, R. K., Liou, Y. H., Knudsen, N. H., Starost, K. A., Xu, C., Hyde, A. L., Liu, S., Jacobi, D., Liao, N. S., & Lee, C. H. (2020). Bmal1 integrates mitochondrial metabolism and macrophage activation. *Elife*, *9*, https://doi.org/10.7554/eLife.54090.

Arble, D. M., Ramsey, K. M., Bass, J., & Turek, F. W. (2010). Circadian disruption and metabolic disease: Findings from animal models. *Best Pract Res Clin Endocrinol Metab*, *24*(5), 785–800.

Aton, S. J., Colwell, C. S., Harmar, A. J., Waschek, J., & Herzog, E. D. (2005). Vasoactive intestinal polypeptide mediates circadian rhythmicity and synchrony in mammalian clock neurons. *Nat Neurosci*, *8*(4), 476–483.

Aton, S. J., & Herzog, E. D. (2005). Come together, right . . . now: Synchronization of rhythms in a mammalian circadian clock. *Neuron*, *48*(4), 531–534.

Balsalobre, A., Brown, S. A., Marcacci, L., Tronche, F., Kellendonk, C., Reichardt, H. M., Schutz, G., & Schibler, U. (2000). Resetting of circadian time in peripheral tissues by glucocorticoid signaling. *Science*, *289*(5488), 2344–2347.

Barclay, J. L., Husse, J., Bode, B., Naujokat, N., Meyer-Kovac, J., Schmid, S. M., Lehnert, H., & Oster, H. (2012). Circadian desynchrony promotes metabolic disruption in a mouse model of shiftwork. *PLoS One*, *7*(5), e37150.

Basinou, V., Park, J. S., Cederroth, C. R., & Canlon, B. (2017). Circadian regulation of auditory function. *Hear Res*, *347*, 47–55.

Bastianini, S., Silvani, A., Berteotti, C., Martire, V. L., & Zoccoli, G. (2012). Mice show circadian rhythms of blood pressure during each wake–sleep state. *Chronobiol Int*, *29*(1), 82–86.

Beier, C., Zhang, Z., Yurgel, M., & Hattar, S. (2021). Projections of ipRGCs and conventional RGCs to retinorecipient brain nuclei. *J Comp Neurol*, *529*(8), 1863–1875.

Berson, D. M., Dunn, F. A., & Takao, M. (2002). Phototransduction by retinal ganglion cells that set the circadian clock. *Science*, *295*(5557), 1070–1073.

Boivin, D. B., & Boudreau, P. (2014). Impacts of shift work on sleep and circadian rhythms. *Pathol Biol (Paris)*, *62*(5), 292–301.

Bouillon-Minois, J. B., Thivel, D., Croizier, C., Ajebo, E., Cambier, S., Boudet, G., Adeyemi, O. J., Ugbolue, U. C., Bagheri, R., Vallet, G. T., Schmidt, J., Trousselard, M., & Dutheil, F. (2022). The negative impact of night shifts on diet in emergency healthcare workers. *Nutrients*, *14*(4), 829.

Boulos, Z., Rosenwasser, A. M., & Terman, M. (1980). Feeding schedules and the circadian organization of behavior in the rat. *Behav Brain Res*, *1*(1), 39–65.

Brainard, G. C., Hanifin, J. P., Greeson, J. M., Byrne, B., Glickman, G., Gerner, E., & Rollag, M. D. (2001). Action spectrum for melatonin regulation in humans: Evidence for a novel circadian photoreceptor. *J Neurosci*, *21*(16), 6405–6412.

Brown, T. M., Brainard, G. C., Cajochen, C., Czeisler, C. A., Hanifin, J. P., Lockley, S. W., Lucas, R. J., Munch, M., O'Hagan, J. B., Peirson, S. N., Price, L. L. A., Roenneberg, T., Schlangen, L. J. M., Skene, D. J., Spitschan, M., Vetter, C., Zee, P. C., & Wright, K. P., Jr. (2022). Recommendations for daytime, evening, and nighttime indoor light exposure to best support physiology, sleep, and wakefulness in healthy adults. *PLoS Biol*, *20*(3), e3001571.

Bunger, M. K., Wilsbacher, L. D., Moran, S. M., Clendenin, C., Radcliffe, L. A., Hogenesch, J. B., Simon, M. C., Takahashi, J. S., & Bradfield, C. A. (2000). Mop3 is an essential component of the master circadian pacemaker in mammals. *Cell*, *103*(7), 1009–1017.

Challet, E. (2007). Minireview: Entrainment of the suprachiasmatic clockwork in diurnal and nocturnal mammals. *Endocrinology*, *148*(12), 5648–5655.

Chang, A. M., Aeschbach, D., Duffy, J. F., & Czeisler, C. A. (2015). Evening use of light-emitting eReaders negatively affects sleep, circadian timing, and next-morning alertness. *Proc Natl Acad Sci USA*, *112*, 1232–1237.

Chen, S., Feng, M., Zhang, S., Dong, Z., Wang, Y., Zhang, W., & Liu, C. (2019). Angptl8 mediates food-driven resetting of hepatic circadian clock in mice. *Nat Commun*, *10*(1), 3518.

Colwell, C. S., Michel, S., Itri, J., Rodriguez, W., Tam, J., Lelievre, V., Hu, Z., Liu, X., & Waschek, J. A. (2003). Disrupted circadian rhythms in VIP- and PHI-deficient mice. *Am J Physiol Regul Integr Comp Physiol*, *285*(5), R939–R949.

Coomans, C. P., van den Berg, S. A., Houben, T., van Klinken, J. B., van den Berg, R., Pronk, A. C., Havekes, L. M., Romijn, J. A., van Dijk, K. W., Biermasz, N. R., & Meijer, J. H. (2013). Detrimental effects of constant light exposure and high-fat diet on circadian energy metabolism and insulin sensitivity. *Faseb J*, *27*(4), 1721–1732.

Cox, K. H., & Takahashi, J. S. (2019). Circadian clock genes and the transcriptional architecture of the clock mechanism. *J Mol Endocrinol*, *63*(4), R93–R102.

Curtis, A. M., Bellet, M. M., Sassone-Corsi, P., & O'Neill, L. A. (2014). Circadian clock proteins and immunity. *Immunity*, *40*(2), 178–186.

Curtis, A. M., Cheng, Y., Kapoor, S., Reilly, D., Price, T. S., & Fitzgerald, G. A. (2007). Circadian variation of blood pressure and the vascular response to asynchronous stress. *Proc Natl Acad Sci USA*, *104*(9), 3450–3455.

Czeisler, C. A., Duffy, J. F., Shanahan, T. L., Brown, E. N., Mitchell, J. F., Rimmer, D. W., Ronda, J. M., Silva, E. J., Allan, J. S., Emens, J. S., Dijk, D. J., & Kronauer, R. E. (1999). Stability, precision, and near-24-hour period of the human circadian pacemaker. *Science*, *284*(5423), 2177–2181.

Czeisler, C. A., Shanahan, T. L., Klerman, E. B., Martens, H., Brotman, D. J., Emens, J. S., Klein, T., & Rizzo, J. F. (1995). Suppression of melatonin secretion in some blind patients by exposure to bright light [see comments]. *N Engl J Med*, *332*, 6–11.

Dalby, M. C., Davidson, S. J., Burman, J. F., & Davies, S. W. (2000). Diurnal variation in platelet aggregation with the PFA-100 platelet function analyser. *Platelets*, *11*(6), 320–324.

Damiola, F., Le Minh, N., Preitner, N., Kornmann, B., Fleury-Olela, F., & Schibler, U. (2000). Restricted feeding uncouples circadian oscillators in peripheral tissues from the central pacemaker in the suprachiasmatic nucleus. *Genes Dev*, *14*(23), 2950–2961.

Davidson, A. J., London, B., Block, G. D., & Menaker, M. (2005). Cardiovascular tissues contain independent circadian clocks. *Clin Exp Hypertens*, *27*(2–3), 307–311.

Ding, J. M., Faiman, L. E., Hurst, W. J., Kuriashkina, L. R., & Gillette, M. U. (1997). Resetting the biological clock: Mediation of nocturnal CREB phosphorylation via light, glutamate, and nitric oxide. *J Neurosci*, *17*(2), 667–675.

Do, M. T. H. (2019). Melanopsin and the intrinsically photosensitive retinal ganglion cells: Biophysics to behavior. *Neuron*, *104*(2), 205–226.

Dutheil, F., Baker, J. S., Mermillod, M., De Cesare, M., Vidal, A., Moustafa, F., Pereira, B., & Navel, V. (2020). Shift work, and particularly permanent night shifts, promote dyslipidaemia: A systematic review and meta-analysis. *Atherosclerosis*, *313*, 156–169.

Edgar, D. M., & Dement, W. C. (1991). Regularly scheduled voluntary exercise synchronizes the mouse circadian clock. *Am J Physiol*, *261*(4 Pt 2), R928–R933.

Etsuda, H., Takase, B., Uehata, A., Kusano, H., Hamabe, A., Kuhara, R., Akima, T., Matsushima, Y., Arakawa, K., Satomura, K., Kurita, A., & Ohsuzu, F. (1999). Morning attenuation of endothelium-dependent, flow-mediated dilation in healthy young men: Possible connection to morning peak of cardiac events? *Clin Cardiol*, *22*(6), 417–421.

Evans, J. A., & Davidson, A. J. (2013). Health consequences of circadian disruption in humans and animal models. *Prog Mol Biol Transl Sci*, *119*, 283–323.

Falchi, F., Cinzano, P., Duriscoe, D., Kyba, C. C., Elvidge, C. D., Baugh, K., Portnov, B. A., Rybnikova, N. A., & Furgoni, R. (2016). The new world atlas of artificial night sky brightness. *Sci Adv*, *2*(6), e1600377.

Fernandez, D. C., Chang, Y. T., Hattar, S., & Chen, S. K. (2016). Architecture of retinal projections to the central circadian pacemaker. *Proc Natl Acad Sci USA*, *113*(21), 6047–6052.

Finlay, B. L., & Sengelaub, D. R. (1981). Toward a neuroethology of mammalian vision: Ecology and anatomy of rodent visuomotor behavior. *Behav Brain Res*, *3*(2), 133–149.

Fisk, A. S., Tam, S. K. E., Brown, L. A., Vyazovskiy, V. V., Bannerman, D. M., & Peirson, S. N. (2018). Light and cognition: Roles for circadian rhythms, sleep, and arousal. *Front Neurol*, *9*, 56.

Fonken, L. K., Aubrecht, T. G., Melendez-Fernandez, O. H., Weil, Z. M., & Nelson, R. J. (2013). Dim light at night disrupts molecular circadian rhythms and increases body weight. *J Biol Rhythms*, *28*(4), 262–271.

Fonken, L. K., Frank, M. G., Kitt, M. M., Barrientos, R. M., Watkins, L. R., & Maier, S. F. (2015). Microglia inflammatory responses are controlled by an intrinsic circadian clock. *Brain Behav Immun*, *45*, 171–179.

Fonken, L. K., & Nelson, R. J. (2013). Dim light at night increases depressive-like responses in male C3H/HeNHsd mice. *Behav Brain Res*, *243*, 74–78.

Fonken, L. K., & Nelson, R. J. (2014). The effects of light at night on circadian clocks and metabolism. *Endocr Rev*, *35*(4), 648–670.

Fonken, L. K., Weil, Z. M., & Nelson, R. J. (2013). Dark nights reverse metabolic disruption caused by dim light at night. *Obesity (Silver Spring)*, *21*(6), 1159–1164.

Fonken, L. K., Workman, J. L., Walton, J. C., Weil, Z. M., Morris, J. S., Haim, A., & Nelson, R. J. (2010). Light at night increases body mass by shifting the time of food intake. *Proc Natl Acad Sci USA*, *107*(43), 18664–18669.

Fraser, G., Trinder, J., Colrain, I. M., & Montgomery, I. (1989). Effect of sleep and circadian cycle on sleep period energy expenditure. *J Appl Physiol (1985)*, *66*(2), 830–836.

Gaston, K. J., Visser, M. E., & Holker, F. (2015). The biological impacts of artificial light at night: The research challenge. *Philosophical Transactions of the Royal Society B*, *370*, 20140133.

Gau, D., Lemberger, T., von Gall, C., Kretz, O., Le Minh, N., Gass, P., Schmid, W., Schibler, U., Korf, H. W., & Schutz, G. (2002). Phosphorylation of CREB Ser142 regulates light-induced phase shifts of the circadian clock. *Neuron*, *34*(2), 245–253.

Gekakis, N., Staknis, D., Nguyen, H. B., Davis, F. C., Wilsbacher, L. D., King, D. P., Takahashi, J. S., & Weitz, C. J. (1998). Role of the CLOCK protein in the mammalian circadian mechanism. *Science*, *280*(5369), 1564–1569.

Gibbs, J. E., Blaikley, J., Beesley, S., Matthews, L., Simpson, K. D., Boyce, S. H., Farrow, S. N., Else, K. J., Singh, D., Ray, D. W., & Loudon, A. S. (2012). The nuclear receptor REV-ERBalpha mediates circadian regulation of innate immunity through selective regulation of inflammatory cytokines. *Proc Natl Acad Sci USA*, *109*(2), 582–587.

Gibbs, J. E., & Ray, D. W. (2013). The role of the circadian clock in rheumatoid arthritis. *Arthritis Res Ther*, *15*(1), 205.

Ginty, D. D., Kornhauser, J. M., Thompson, M. A., Bading, H., Mayo, K. E., Takahashi, J. S., & Greenberg, M. E. (1993). Regulation of CREB phosphorylation in the suprachiasmatic nucleus by light and a circadian clock. *Science*, *260*(5105), 238–241.

Griffin, E. A., Jr., Staknis, D., & Weitz, C. J. (1999). Light-independent role of CRY1 and CRY2 in the mammalian circadian clock. *Science*, *286*(5440), 768–771.

Gritton, H. J., Kantorowski, A., Sarter, M., & Lee, T. M. (2012). Bidirectional interactions between circadian entrainment and cognitive performance. *Learn Mem*, *19*(3), 126–141.

Halberg, F., Johnson, E. A., Brown, B. W., & Bittner, J. J. (1960). Susceptibility rhythm to E. coli endotoxin and bioassay. *Proc Soc Exp Biol Med*, *103*, 142–144.

Hanifin, J. P., Lockley, S. W., Cecil, K., West, K., Jablonski, M., Warfield, B., James, M., Ayers, M., Byrne, B., Gerner, E., Pineda, C., Rollag, M., & Brainard, G. C. (2019). Randomized trial of polychromatic blue-enriched light for circadian phase shifting, melatonin suppression, and alerting responses. *Physiol Behav*, *198*, 57–66.

Hannibal, J., Hindersson, P., Knudsen, S. M., Georg, B., & Fahrenkrug, J. (2002). The photopigment melanopsin is exclusively present in pituitary adenylate cyclase-activating polypeptide-containing retinal ganglion cells of the retinohypothalamic tract. *J Neurosci*, *22*(1), RC191.

Hansen, J. (2017). Night shift work and risk of breast cancer. *Curr Environ Health Rep*, *4*(3), 325–339.

Harmar, A. J., Marston, H. M., Shen, S., Spratt, C., West, K. M., Sheward, W. J., Morrison, C. F., Dorin, J. R., Piggins, H. D., Reubi, J. C., Kelly, J. S., Maywood, E. S., & Hastings, M. H. (2002). The VPAC(2) receptor is essential for circadian function in the mouse suprachiasmatic nuclei. *Cell*, *109*(4), 497–508.

Haspel, J. A., Anafi, R., Brown, M. K., Cermakian, N., Depner, C., Desplats, P., Gelman, A. E., Haack, M., Jelic, S., Kim, B. S., Laposky, A. D., Lee, Y. C., Mongodin, E., Prather, A. A., Prendergast, B. J., Reardon, C., Shaw, A. C., Sengupta, S., Szentirmai, E., … Solt, L. A. (2020). Perfect timing: Circadian rhythms, sleep, and immunity: An NIH workshop summary. *JCI Insight*, *5*(1).

Hatori, M., Vollmers, C., Zarrinpar, A., DiTacchio, L., Bushong, E. A., Gill, S., Leblanc, M., Chaix, A., Joens, M., Fitzpatrick, J. A., Ellisman, M. H., & Panda, S. (2012). Time-restricted feeding without reducing caloric intake prevents metabolic diseases in mice fed a high-fat diet. *Cell Metab*, *15*(6), 848–860.

Hattar, S., Kumar, M., Park, A., Tong, P., Tung, J., Yau, K. W., & Berson, D. M. (2006). Central projections of melanopsin-expressing retinal ganglion cells in the mouse [Comparative Study Research Support, N.I.H., Extramural]. *J Comp Neurol*, *497*(3), 326–349.

Hattar, S., Lucas, R. J., Mrosovsky, N., Thompson, S., Douglas, R. H., Hankins, M. W., Lem, J., Biel, M., Hofmann, F., Foster, R. G., & Yau, K. W. (2003). Melanopsin and rod-cone photoreceptive systems account for all major accessory visual functions in mice. *Nature*, *424*(6944), 76–81.

He, W., Holtkamp, S., Hergenhan, S. M., Kraus, K., de Juan, A., Weber, J., Bradfield, P., Grenier, J. M. P., Pelletier, J., Druzd, D., Chen, C. S., Ince, L. M., Bierschenk, S., Pick, R., Sperandio, M., Aurrand-Lions, M., & Scheiermann, C. (2018). Circadian expression of migratory factors establishes lineage-specific signatures that guide the homing of leukocyte subsets to tissues. *Immunity*, *49*(6), 1175–1190.

van der Heijden, K. B., de Sonneville, L. M., & Althaus, M. (2010). Time-of-day effects on cognition in preadolescents: A trails study. *Chronobiol Int*, *27*(9–10), 1870–1894.

Herxheimer, A. (2014). Jet lag. *BMJ Clin Evid*, *2014*, 2303.

Hogenesch, J. B., Gu, Y. Z., Jain, S., & Bradfield, C. A. (1998). The basic-helix-loop-helix-PAS orphan MOP3 forms transcriptionally active complexes with circadian and hypoxia factors. *Proc Natl Acad Sci USA*, *95*(10), 5474–5479.

Hut, R. A., & Van der Zee, E. A. (2011). The cholinergic system, circadian rhythmicity, and time memory. *Behav Brain Res*, *221*(2), 466–480.

IARC Monographs Vol 124 group. (2019). Carcinogenicity of night shift work. *Lancet Oncol*, *20*(8), 1058–1059.

Ingpen, M. L. (1968). The quantitative measurement of joint changes in rheumatoid arthritis. *Ann Phys Med*, *9*(8), 322–327.

Jin, X., Shearman, L. P., Weaver, D. R., Zylka, M. J., de Vries, G. J., & Reppert, S. M. (1999). A molecular mechanism regulating rhythmic output from the suprachiasmatic circadian clock. *Cell*, *96*(1), 57–68.

Jones, S. E., Lane, J. M., Wood, A. R., van Hees, V. T., Tyrrell, J., Beaumont, R. N., Jeffries, A. R., Dashti, H. S., Hillsdon, M., Ruth, K. S., Tuke, M. A., Yaghootkar, H., Sharp, S. A., Jie, Y., Thompson, W. D., Harrison, J. W., Dawes, A., Byrne, E. M., Tiemeier, H., … Weedon, M. N. (2019). Genome-wide association analyses of chronotype in 697,828 individuals provides insights into circadian rhythms. *Nat Commun*, *10*(1), 343.

Jung, C. M., Melanson, E. L., Frydendall, E. J., Perreault, L., Eckel, R. H., & Wright, K. P. (2011). Energy expenditure during sleep, sleep deprivation and sleep following sleep deprivation in adult humans. *J Physiol*, *589*(Pt 1), 235–244.

Karatsoreos, I. N., Bhagat, S., Bloss, E. B., Morrison, J. H., & McEwen, B. S. (2011). Disruption of circadian clocks has ramifications for metabolism, brain, and behavior. *Proc Natl Acad Sci USA*, *108*(4), 1657–1662.

Khan, M. S., & Ahmad, S. I. (2003). Circadian variation: Increased morning incidence of acute myocardial infarction in patients with coronary artery disease. *J Pak Med Assoc*, *53* (2), 84–87.

Krishnan, H. C., & Lyons, L. C. (2015). Synchrony and desynchrony in circadian clocks: Impacts on learning and memory. *Learn Mem*, *22*(9), 426–437.

Kume, K., Zylka, M. J., Sriram, S., Shearman, L. P., Weaver, D. R., Jin, X., Maywood, E. S., Hastings, M. H., & Reppert, S. M. (1999). mCRY1 and mCRY2 are essential components of the negative limb of the circadian clock feedback loop. *Cell*, *98*(2), 193–205.

Lamia, K. A., Storch, K. F., & Weitz, C. J. (2008). Physiological significance of a peripheral tissue circadian clock. *Proc Natl Acad Sci USA*, *105*(39), 15172–15177.

Landgraf, D., Tsang, A. H., Leliavski, A., Koch, C. E., Barclay, J. L., Drucker, D. J., & Oster, H. (2015). Oxyntomodulin regulates resetting of the liver circadian clock by food. *Elife*, *4*, e06253.

Leak, R. K., & Moore, R. Y. (2001). Topographic organization of suprachiasmatic nucleus projection neurons. *J Comp Neurol*, *433*(3), 312–334.

Lee, C., Etchegaray, J. P., Cagampang, F. R., Loudon, A. S., & Reppert, S. M. (2001). Posttranslational mechanisms regulate the mammalian circadian clock. *Cell*, *107*(7), 855–867.

Liu, J. A., Walton, J. C., DeVries, A. C., & Nelson, R. J. (2021). Disruptions of circadian rhythms and thrombolytic therapy during ischemic stroke intervention. *Front Neurosci*, *15*, 675732.

Lowrey, P. L., Shimomura, K., Antoch, M. P., Yamazaki, S., Zemenides, P. D., Ralph, M. R., Menaker, M., & Takahashi, J. S. (2000). Positional syntenic cloning and functional characterization of the mammalian circadian mutation tau. *Science*, *288*(5465), 483–492.

Manfredini, R., Boari, B., Salmi, R., Fabbian, F., Pala, M., Tiseo, R., & Portaluppi, F. (2013). Twenty-four-hour patterns in occurrence and pathophysiology of acute cardiovascular events and ischemic heart disease. *Chronobiol Int*, *30*(1–2), 6–16.

Marcheva, B., Ramsey, K. M., Buhr, E. D., Kobayashi, Y., Su, H., Ko, C. H., Ivanova, G., Omura, C., Mo, S., Vitaterna, M. H., Lopez, J. P., Philipson, L. H., Bradfield, C. A., Crosby, S. D., JeBailey, L., Wang, X. Z., Takahashi, J. S., & Bass, J. (2010). Disruption of the clock components CLOCK and BMAL1 leads to hypoinsulinaemia and diabetes. *Nature*, *466*(7306), 627–631.

Markwald, R. R., Melanson, E. L., Smith, M. R., Higgins, J., Perreault, L., Eckel, R. H., & Wright, K. P., Jr. (2013). Impact of insufficient sleep on total daily energy expenditure, food intake, and weight gain. *Proc Natl Acad Sci USA*, *110*(14), 5695–5700.

Martin-Fairey, C. A., & Nunez, A. A. (2014). Circadian modulation of memory and plasticity gene products in a diurnal species. *Brain Res*, *1581*, 30–39.
Mavroudis, P. D., DuBois, D. C., Almon, R. R., & Jusko, W. J. (2018). Modeling circadian variability of core-clock and clock-controlled genes in four tissues of the rat. *PLoS One*, *13*(6), e0197534.
Maywood, E. S., Chesham, J. E., O'Brien, J. A., & Hastings, M. H. (2011). A diversity of paracrine signals sustains molecular circadian cycling in suprachiasmatic nucleus circuits. *Proc Natl Acad Sci USA*, *108*(34), 14306–14311.
McHill, A. W., Melanson, E. L., Higgins, J., Connick, E., Moehlman, T. M., Stothard, E. R., & Wright, K. P., Jr. (2014). Impact of circadian misalignment on energy metabolism during simulated nightshift work. *Proc Natl Acad Sci USA*, *111*(48), 17302–17307.
Melendez-Fernandez, O. H., Walton, J. C., DeVries, A. C., & Nelson, R. J. (2021). Clocks, rhythms, sex, and hearts: How disrupted circadian rhythms, time-of-day, and sex influence cardiovascular health. *Biomolecules*, *11*(6), 883.
Meltser, I., Cederroth, C. R., Basinou, V., Savelyev, S., Lundkvist, G. S., & Canlon, B. (2014). TrkB-mediated protection against circadian sensitivity to noise trauma in the murine cochlea. *Curr Biol*, *24*(6), 658–663.
Mohawk, J. A., Green, C. B., & Takahashi, J. S. (2012). Central and peripheral circadian clocks in mammals. *Annu Rev Neurosci*, *35*, 445–462.
Moore, R. Y., & Eichler, V. B. (1972). Loss of a circadian adrenal corticosterone rhythm following suprachiasmatic lesions in the rat. *Brain Res*, *42*(1), 201–206.
Moore, R. Y., & Qavi, H. B. (1971). Circadian rhythm in adrenal adenyl cyclase and corticosterone abolished by medial forebrain bundle transection in the rat. *Experientia*, *27*(3), 249–250.
Moore, R. Y., Speh, J. C., & Leak, R. K. (2002). Suprachiasmatic nucleus organization. *Cell Tissue Res*, *309*(1), 89–98.
Mrosovsky, N., Lucas, R. J., & Foster, R. G. (2001). Persistence of masking responses to light in mice lacking rods and cones. *J Biol Rhythms*, *16*(6), 585–588.
Muller, J. E. (1999). Circadian variation and triggering of acute coronary events. *Am Heart J*, *137*(4 Pt 2), S1–S8.
Muller, J. E., Ludmer, P. L., Willich, S. N., Tofler, G. H., Aylmer, G., Klangos, I., & Stone, P. H. (1987). Circadian variation in the frequency of sudden cardiac death. *Circulation*, *75*(1), 131–138.
Nelson, R. J., Bumgarner, J. R., Walker, W. H., 2nd, & DeVries, A. C. (2021). Time-of-day as a critical biological variable. *Neurosci Biobehav Rev*, *127*, 740–746.
Paschos, G. K., & FitzGerald, G. A. (2010). Circadian clocks and vascular function. *Circ Res*, *106*(5), 833–841.
Patton, A. P., & Hastings, M. H. (2018). The suprachiasmatic nucleus. *Curr Biol*, *28*(15), R816–R822.
Paul, M. J., Indic, P., & Schwartz, W. J. (2015). Social synchronization of circadian rhythmicity in female mice depends on the number of cohabiting animals. *Biol Lett*, *11* (6), 20150204.
Perry, M. G., Kirwan, J. R., Jessop, D. S., & Hunt, L. P. (2009). Overnight variations in cortisol, interleukin 6, tumour necrosis factor alpha and other cytokines in people with rheumatoid arthritis. *Ann Rheum Dis*, *68*(1), 63–68.
Pietroiusti, A., Neri, A., Somma, G., Coppeta, L., Iavicoli, I., Bergamaschi, A., & Magrini, A. (2010). Incidence of metabolic syndrome among night-shift healthcare workers. *Occup Environ Med*, *67*(1), 54–57.
Preitner, N., Damiola, F., Lopez-Molina, L., Zakany, J., Duboule, D., Albrecht, U., & Schibler, U. (2002). The orphan nuclear receptor REV-ERBalpha controls circadian

transcription within the positive limb of the mammalian circadian oscillator. *Cell*, *110*(2), 251–260.
Provencio, I., Rollag, M. D., & Castrucci, A. M. (2002). Photoreceptive net in the mammalian retina. This mesh of cells may explain how some blind mice can still tell day from night. *Nature*, *415*(6871), 493.
Ralph, M. R., & Menaker, M. (1988). A mutation of the circadian system in golden hamsters. *Science*, *241*, 1225–1227.
Ramanathan, C., Kathale, N. D., Liu, D., Lee, C., Freeman, D. A., Hogenesch, J. B., Cao, R., & Liu, A. C. (2018). mTOR signaling regulates central and peripheral circadian clock function. *PLoS Genet*, *14*(5), e1007369.
Rana, S., Prabhu, S. D., & Young, M. E. (2020). Chronobiological influence over cardiovascular function: The good, the bad, and the ugly. *Circ Res*, *126*(2), 258–279.
Reddy, A. B., Maywood, E. S., Karp, N. A., King, V. M., Inoue, Y., Gonzalez, F. J., Lilley, K. S., Kyriacou, C. P., & Hastings, M. H. (2007). Glucocorticoid signaling synchronizes the liver circadian transcriptome. *Hepatology*, *45*(6), 1478–1488.
Reinke, H., & Asher, G. (2019). Crosstalk between metabolism and circadian clocks. *Nat Rev Mol Cell Biol*, *20*(4), 227–241.
Richter, C. P. (1922). A behavioristic study of the activity of the rat. *Compar Psychol Monogr*, *1*, 1–55.
Ripperger, J. A., & Schibler, U. (2001). Circadian regulation of gene expression in animals. *Curr Opin Cell Biol*, *13*(3), 357–362.
Roenneberg, T., Wirz-Justice, A., & Merrow, M. (2003). Life between clocks: Daily temporal patterns of human chronotypes. *J Biol Rhythms*, *18*(1), 80–90.
Rusak, B., & Groos, G. (1982). Suprachiasmatic stimulation phase shifts rodent circadian rhythms. *Science*, *215*(4538), 1407–1409.
Salgado-Delgado, R. C., Saderi, N., Basualdo Mdel, C., Guerrero-Vargas, N. N., Escobar, C., & Buijs, R. M. (2013). Shift work or food intake during the rest phase promotes metabolic disruption and desynchrony of liver genes in male rats. *PLoS One*, *8*(4), e60052.
Scheer, F. A., Morris, C. J., & Shea, S. A. (2013). The internal circadian clock increases hunger and appetite in the evening independent of food intake and other behaviors. *Obesity (Silver Spring)*, *21*(3), 421–423.
Schibler, U., Gotic, I., Saini, C., Gos, P., Curie, T., Emmenegger, Y., Sinturel, F., Gosselin, P., Gerber, A., Fleury-Olela, F., Rando, G., Demarque, M., & Franken, P. (2015). Clock-talk: Interactions between central and peripheral circadian oscillators in mammals. *Cold Spring Harb Symp Quant Biol*, *80*, 223–232.
Schmidt, C., Collette, F., Cajochen, C., & Peigneux, P. (2007, Oct). A time to think: Circadian rhythms in human cognition. *Cogn Neuropsychol*, *24*(7), 755–789.
Schurov, I. L., McNulty, S., Best, J. D., Sloper, P. J., & Hastings, M. H. (1999). Glutamatergic induction of CREB phosphorylation and Fos expression in primary cultures of the suprachiasmatic hypothalamus in vitro is mediated by co-ordinate activity of NMDA and non-NMDA receptors. *J Neuroendocrinol*, *11*(1), 43–51.
Silver, R., LeSauter, J., Tresco, P. A., & Lehman, M. N. (1996). A diffusible coupling signal from the transplanted suprachiasmatic nucleus controlling circadian locomotor rhythms. *Nature*, *382*(6594), 810–813.
Smarr, B. L., Jennings, K. J., Driscoll, J. R., & Kriegsfeld, L. J. (2014). A time to remember: The role of circadian clocks in learning and memory. *Behav Neurosci*, *128*(3), 283–303.
Smolensky, M. H., Hermida, R. C., Castriotta, R. J., & Portaluppi, F. (2007). Role of sleep–wake cycle on blood pressure circadian rhythms and hypertension. *Sleep Med*, *8*(6), 668–680.

Stephan, F. K., & Zucker, I. (1972). Circadian rhythms in drinking behavior and locomotor activity of rats are eliminated by hypothalamic lesions. *Proc Natl Acad Sci USA*, *69*(6), 1583–1586.

Storch, K. F., Lipan, O., Leykin, I., Viswanathan, N., Davis, F. C., Wong, W. H., & Weitz, C. J. (2002). Extensive and divergent circadian gene expression in liver and heart. *Nature*, *417*(6884), 78–83.

Stow, L. R., Richards, J., Cheng, K. Y., Lynch, I. J., Jeffers, L. A., Greenlee, M. M., Cain, B. D., Wingo, C. S., & Gumz, M. L. (2012). The circadian protein period 1 contributes to blood pressure control and coordinately regulates renal sodium transport genes. *Hypertension*, *59*(6), 1151–1156.

Sudy, A. R., Ella, K., Bodizs, R., & Kaldi, K. (2019). Association of social jetlag with sleep quality and autonomic cardiac control during sleep in young healthy men. *Front Neurosci*, *13*, 950.

Takahashi, J. S., DeCoursey, P. J., Bauman, L., & Menaker, M. (1984). Spectral sensitivity of a novel photoreceptive system mediating entrainment of mammalian circadian rhythms. *Nature*, *308*(5955), 186–188.

Takeda, N., Maemura, K., Horie, S., Oishi, K., Imai, Y., Harada, T., Saito, T., Shiga, T., Amiya, E., Manabe, I., Ishida, N., & Nagai, R. (2007). Thrombomodulin is a clock-controlled gene in vascular endothelial cells. *J Biol Chem*, *282*(45), 32561–32567.

Thosar, S. S., Butler, M. P., & Shea, S. A. (2018). Role of the circadian system in cardiovascular disease. *J Clin Invest*, *128*(6), 2157–2167.

Turek, F. W., Joshu, C., Kohsaka, A., Lin, E., Ivanova, G., McDearmon, E., Laposky, A., Losee-Olson, S., Easton, A., Jensen, D. R., Eckel, R. H., Takahashi, J. S., & Bass, J. (2005). Obesity and metabolic syndrome in circadian clock mutant mice. *Science*, *308* (5724), 1043–1045.

Vetter, C. (2020). Circadian disruption: What do we actually mean? *Eur J Neurosci*, *51*(1), 531–550.

Vitaterna, M. H., King, D. P., Chang, A. M., Kornhauser, J. M., Lowrey, P. L., McDonald, J. D., Dove, W. F., Pinto, L. H., Turek, F. W., & Takahashi, J. S. (1994). Mutagenesis and mapping of a mouse gene, clock, essential for circadian behavior. *Science*, *264* (5159), 719–725.

Weaver, D. R. (1998). The suprachiasmatic nucleus: A 25-year retrospective. *J Biol Rhythms*, *13*(2), 100–112.

Welsh, D. K., Logothetis, D. E., Meister, M., & Reppert, S. M. (1995). Individual neurons dissociated from rat suprachiasmatic nucleus express independently phased circadian firing rhythms. *Neuron*, *14*(4), 697–706.

West, K. E., Jablonski, M. R., Warfield, B., Cecil, K. S., James, M., Ayers, M. A., Maida, J., Bowen, C., Sliney, D. H., Rollag, M. D., Hanifin, J. P., & Brainard, G. C. (2011). Blue light from light-emitting diodes elicits a dose-dependent suppression of melatonin in humans. *J Appl Physiol (1985)*, *110*(3), 619–626.

Westwood, M. L., O'Donnell, A. J., de Bekker, C., Lively, C. M., Zuk, M., & Reece, S. E. (2019). The evolutionary ecology of circadian rhythms in infection. *Nat Ecol Evol*, *3*(4), 552–560.

Yamazaki, S., Numano, R., Abe, M., Hida, A., Takahashi, R., Ueda, M., Block, G. D., Sakaki, Y., Menaker, M., & Tei, H. (2000). Resetting central and peripheral circadian oscillators in transgenic rats. *Science*, *288*(5466), 682–685.

Yoo, S. H., Yamazaki, S., Lowrey, P. L., Shimomura, K., Ko, C. H., Buhr, E. D., Siepka, S. M., Hong, H. K., Oh, W. J., Yoo, O. J., Menaker, M., & Takahashi, J. S. (2004). PERIOD2:: LUCIFERASE real-time reporting of circadian dynamics reveals persistent circadian oscillations in mouse peripheral tissues. *Proc Natl Acad Sci USA*, *101*(15), 5339–5346.

Zhang, Q., Chair, S. Y., Lo, S. H. S., Chau, J. P., Schwade, M., & Zhao, X. (2020). Association between shift work and obesity among nurses: A systematic review and meta-analysis. *Int J Nurs Stud*, *112*, 103757.

Zhang, R., Lahens, N. F., Ballance, H. I., Hughes, M. E., & Hogenesch, J. B. (2014). A circadian gene expression atlas in mammals: Implications for biology and medicine. *Proc Natl Acad Sci USA*, *111*(45), 16219–16224.

Zhang, S., Dai, M., Wang, X., Jiang, S. H., Hu, L. P., Zhang, X. L., & Zhang, Z. G. (2020). Signalling entrains the peripheral circadian clock. *Cell Signal*, *69*, 109433.