GOODNIGHT BRAIN: THE SCIENCE OF HEALTHY SLEEP
Learning Objectives

• Describe the neurobiological and molecular bases of circadian rhythms and sleep-wake cycles

• Explain to patients how healthy sleep affects brain health and wellness

• Relate the neurobiology of sleep to the mechanisms of potential sleep treatment strategies
Arousal Spectrum

Circadian Rhythms Describe Events That Occur on a 24-hour Cycle
Zeitgebers

External cues to synchronize circadian rhythms:

• Light

• Melatonin

• Eating and drinking patterns

• Social interactions

Suprachiasmatic nucleus (SCN)

Retinohypothalamic tract
Processes Regulating Sleep

- **Awake**
- **Stage 1**
- **Stage 2**
- **Stage 3**
- **Stage 4**
- **REM**

**Time of Sleep**
- 0
- 1
- 2
- 3
- 4
- 5
- 6
- 7
- 8
Sleep Cycle

Hypocretin/Orexin

- Involved in sleep/wake, addiction, compulsivity, and overeating/obesity

Molecular Clock Genes

- An intricate group of transcription factors that regulate sleep/wake cycles by turning each other on/off
- Expression waxes/wanes approximately every 24 hours
- Turn on/off expression of other genes, including those involved in sleep, metabolism, cell division, and mood

- **CLOCK** (circadian locomotor output cycles kaput)
- **BMAL1** (brain and muscle ARNT-like-1)
- **PER** (period)
- **CRY** (cryptochrome)
- **REV-ERBα**
- **ROR** (retinoic acid-related orphan receptor)

Buhr ED, Takahashi JS. Handbook of Experimental Pharmacology 2013;217:3-27.
Transcription Factors
Involved in the Molecular Clock

- ROR
- CLOCK
- Period
- Cry
- REV-ERBα
- Bmal1
- Heterodimers

DNA
Promoter
Gene
## Clock Genes Associated With Psychiatric Disorders

<table>
<thead>
<tr>
<th>Clock Gene</th>
<th>Disorder</th>
<th>References</th>
</tr>
</thead>
<tbody>
<tr>
<td>Clock (or its homolog, NPAS)</td>
<td>Bipolar disorder</td>
<td>Benedetti et al, 2003; Soria et al, 2010.</td>
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</tbody>
</table>
Suprachiasmatic Nucleus Control of Sleep

VIP: vasoactive intestinal peptide
GRP: gastrin-releasing peptide
AVP: arginine vasopressin
PK2: prokineticin 2

Hypothalamus

Core
GRP
VIP
GABA

Shell
AVP
PK2
GABA

SCN

Other hypothalamic nuclei

Projections to secondary pacemakers throughout the brain and periphery

Misalignment Between Central and Peripheral Clocks

Cost and Consequences of Sleep/Wake Disorders

Sleep: How much is too much? Too little?

Sleep/Wake Disturbances Increase Risk of Work-Related Injury

- Sleep/wake disorders affect up to 70 million people in the US
- Workers with sleep/wake problems have a 1.62 times increased risk of being injured

Psychiatric Disorders

- Sleep/wake disorders may be a contributing cause or consequence of mood disorders
  - High rates of depression have been reported in shift workers
  - As many as 63% of patients with obstructive sleep apnea have a mood disorder

- Individuals with insomnia
  - 2X more likely to develop anxiety
  - 4X more likely to develop depression
  - 7X more likely to develop substance abuse disorder

- Many psychotropic agents can affect sleep/wake cycles

Synaptic Plasticity

• REM sleep may be essential for hippocampal-dependent cognitive function and synaptic plasticity

• Sleep deprivation (specifically REM sleep deprivation) affects the expression of genes involved in synaptic plasticity

• Consequences of 1 night of sleep deprivation
  • Similar effects to those seen with 1% blood alcohol level
  • 32% increase in number of errors by surgeons on a simulated surgery

Neurogenesis

## Circadian Function and Alzheimer’s Disease

### Links Between AD and Circadian Rhythms

<table>
<thead>
<tr>
<th>Description</th>
<th>Details</th>
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</thead>
<tbody>
<tr>
<td>Melatonin production is decreased with aging and even further in AD</td>
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<tr>
<td>Beta-amyloid levels increase during periods of wakefulness</td>
<td></td>
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<tr>
<td>SCN degeneration and loss of molecular clock gene expression rhythmicity is seen in AD</td>
<td></td>
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<tr>
<td>Beta-amyloid reduces expression of the molecular clock genes that regulate circadian rhythms</td>
<td></td>
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<tr>
<td>Reduced ability of the retina to transmit light (e.g., from cataracts) is seen in aging individuals and patients with AD</td>
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<tr>
<td>Individuals with sleep/wake and circadian disorders have a 1.49-fold increased risk of developing AD</td>
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<tr>
<td>Sleep-deprived mice show increased levels of insoluble tau</td>
<td></td>
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<tr>
<td>Levels of orexin (involved in maintaining wakefulness) are increased in patients with AD and correlate with increased levels of tau protein. Orexin may also inhibit clearance of beta-amyloid</td>
<td></td>
</tr>
<tr>
<td>Circadian rhythm disorders may affect clearance of proteins (such as beta-amyloid)</td>
<td></td>
</tr>
<tr>
<td>Sleep deprivation increases neuroinflammation</td>
<td></td>
</tr>
</tbody>
</table>

Melatonin and Alzheimer’s Disease

Hossain MF et al. Mol Neurobiol 2019; Epub ahead of print.
Cardiometabolic Consequences

- Sleep deprivation is associated with:
  - Obesity and diabetes
  - Decreased levels of leptin (anorectic hormone)
  - Impaired ability to lose weight
- Shift work is associated with cardiovascular disease, obesity, and type 2 diabetes
- The prevalence of cardiovascular disease is higher in patients with restless leg syndrome
- 83% of patients with drug-resistant hypertension have obstructive sleep apnea (OSA)
- 28% of patients with type 2 diabetes have OSA
- 77% of obese patients have OSA
- Obesity is a risk factor for insomnia to become chronic

Cardiometabolic Consequences (cont’d)

• Many hormones involved in metabolism (e.g., ghrelin, leptin) exhibit circadian oscillation
  – The expression of these hormones is regulated by molecular clock genes/transcription factors
  – Many of these hormones also regulate the expression of molecular clock genes/transcription factors

• CLOCK polymorphisms are associated with an increased risk of obesity and metabolic syndrome

• BMAL1 polymorphisms are associated with susceptibility to hypertension and type 2 diabetes

• Chronic misalignment of feeding cycles and sleep cycles results in metabolic disorders and DNA damage

Cancer

• Shift workers have a higher incidence of cancer

• Several cell cycle genes (e.g., MYC, WEE1) are regulated by molecular clock genes/transcription factors

• PER interacts with proteins involved in the DNA damage response

• PER expression is deregulated in breast cancer cells

• DNA damage can also act as a zeitgeber (reset the molecular clock)

• Circadian rhythm/cell cycle synchronization may prevent DNA replication during times of high exposure to damaging UV rays or byproducts of intense metabolism

Walsh et al. Sleep Med 2009;10:859-64;
WEE1 ensures that cells are large enough to divide

1. Cell Growth Checkpoint
2. DNA Synthesis Checkpoint
3. Mitosis Checkpoint

S: synthesis
G: growth
M: mitosis

Cyclin D drives cell proliferation
MYC induces cell proliferation
P53 senses DNA damage

Cancer and Circadian Rhythms

Sleep and Immunity

Sleep and Obesity

- Impaired sleep/wake cycle
- Decreased leptin
- Increased ghrelin
- Gut microbiota dysbiosis
- Increased risk of obesity, type 2 diabetes, and cardiovascular disease

References:
Nutrition and the Molecular Clock

CLOCK: circadian locomotor output cycles kaput
BMAL1: brain and muscle ARNT-like protein
Per: period
Cry: cryptochrome
REV-ERBα: nuclear hormone receptor-related protein
Dbp: albumin D element-binding protein
Nampt: nicotinamide phosphoribosyltransferase
Upp2: uridine phosphorylase 2

Sleep/Wake Issues in the Military

• Military members are often subject to a number of situations/issues that impact sleep duration and quality
  • Reduced sleep
    • 60% get 6 hours of sleep or less
  • Early mornings
    • e.g., 4am
  • Shift work
    • 24-hour operations
  • Less-than-optimal sleep conditions
    • Loud noises
    • Excessive lighting
• May have unique impacts in the military population
  • e.g., for every hour of sleep lost, combat effectiveness is lowered by 15-20%

Good CH et al. Neuropsychopharmacol 2019; Epub ahead of print.
### Sleep/Wake Issues
- Sleep deprivation
- Restricted sleep (<6 hrs)
- Insomnia
- Interrupted sleep
- Circadian rhythm disorders (e.g., 4 am wake time)
- Shift work

### Consequential Impairments
- Cognition
- Memory
- Vigilance
- Excessive daytime sleepiness
- Risk of PTSD and other psychiatric disorders
- Continued sleep/wake issues after deployment/retirement
- Metabolic/endocrine issues

### Sample Military Duties Affected
- Personal safety
- Unit performance
- National security
- Marksmanship
- Pilot errors
Sleep Hygiene

Sleep Time
- No stimulants before bed
- Dark room
- Cool environment
- No disturbances

Wake Time
- Activity
Theoretical Pharmacological Targets

• To promote wakefulness
  • Inhibit
    • GABA
    • Galanin
  • Enhance
    • DA
    • NE
    • 5HT
    • Hcrt
    • ACh
    • HA

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  • Inhibit
    • DA
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  • Enhance
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Melatonin

• Endogenous melatonin is secreted by the pineal gland during periods of darkness

• Acts on the suprachiasmatic nucleus to regulate circadian rhythms

• Melatonin may help to adjust circadian rhythms if taken 3 hours before dim-light melatonin onset

Caffeine

• Both adenosine 2A receptors and dopamine D2 receptors are localized on GABAergic neurons in the striatum

• When adenosine stimulates adenosine 2A receptors it reduces the affinity of nearby D2 receptors for dopamine

• By blocking adenosine from binding to adenosine 2A receptors, caffeine prevents the lowered affinity of D2 receptors for dopamine, resulting in enhanced dopaminergic neurotransmission
Summary

• The neurobiology and molecular underpinnings of sleep are complex
• Disordered sleep/wake cycles affect many physiological and psychological functions
• As our understanding of the neurobiology of sleep increases, we are beginning to recognize how sleep/wake issues may increase risk of cancer, cardiovascular disease, psychiatric disorders, etc.
• In addition to attaining proper sleep hygiene, there are many neurobiological targets that can enhance sleep or wakefulness
Posttest Question 1

Paul is a 21-year-old college student who is interested in using over-the-counter melatonin to help with his sleep-wake cycle while studying for final exams. Which of the following statements is true regarding endogenous melatonin?

1. Melatonin is released from the pineal gland during periods of darkness
2. Melatonin is released from the pineal gland during periods of light
3. Melatonin is released from the suprachiasmatic nucleus during periods of darkness
4. Melatonin is released from the suprachiasmatic nucleus during periods of light
Mitchell is a 42-year-old patient with shift work sleep disorder (SWSD) who reports that he is having difficulty in his job as a police officer due to excessive sleepiness during his shift. Which of the following is a potential therapeutic mechanism to promote wakefulness?

1. Inhibit hypocretin activity
2. Promote GABA activity
3. Promote histamine activity
4. All of the above
5. None of the above
Diane is a 54-year-old Air Force veteran who suffers with insomnia. Her clinician has suggested initiating the hypocretin/orexin antagonist, suvorexant. Endogenous hypocretin/orexin has which of the following effects?

1. Decreases serotonin release from the raphe nucleus
2. Decreases GABA release from the striatum
3. Increases release of dopamine from the ventral tegmental area (VTA)
4. Decreases release of glutamate from the prefrontal cortex (PFC)