DOI: 10.5665/SLEEP.1112

Sleep Neurobiology from a Clinical Perspective

Rodrigo A. España, PhD[1]; Thomas E. Scammell, MD[2]

[1]Department of Physiology and Pharmacology, Wake Forest University Health Sciences, Winston Salem, NC; [2]Department of Neurology, Beth Israel Deaconess Medical Center, Boston, MA

Many neurochemical systems interact to generate wakefulness and sleep. Wakefulness is promoted by neurons in the pons, midbrain, and posterior hypothalamus that produce acetylcholine, norepinephrine, dopamine, serotonin, histamine, and orexin/hypocretin. Most of these ascending arousal systems diffusely activate the cortex and other forebrain targets. NREM sleep is mainly driven by neurons in the preoptic area that inhibit the ascending arousal systems, while REM sleep is regulated primarily by neurons in the pons, with additional influence arising in the hypothalamus. Mutual inhibition between these wake- and sleep-regulating regions likely helps generate full wakefulness and sleep with rapid transitions between states. This up-to-date review of these systems should allow clinicians and researchers to better understand the effects of drugs, lesions, and neurologic disease on sleep and wakefulness.

Keywords: Waking, arousal, locus coeruleus, tuberomammillary nucleus, dorsal raphe nucleus, thalamus, ventrolateral preoptic area

Citation: España RA; Scammell TE. Sleep neurobiology from a clinical perspective. *SLEEP* 2011;34(7):845-858.

INTRODUCTION

Sleep medicine physicians often encounter questions that require an understanding of the neurobiology of sleep: How do certain brain injuries produce coma or hypersomnolence? Why do antidepressants often reduce REM sleep? Why do people with narcolepsy have trouble staying awake? How do amphetamines improve alertness and wakefulness? To help with these and similar questions, this paper provides an overview of the basic circuits that control sleep and wakefulness. This paper has evolved from one we wrote several years ago[1] and has been updated to include many of the latest discoveries on the circuits and neurochemistry of sleep, more information on drugs that are used in clinical practice, and some thoughts on medications that are now in clinical trials. We hope this will provide the reader with useful perspectives on sleep disorders, how drugs influence sleep and wakefulness, and how injuries in different brain regions may affect sleep.

Almost 100 years ago, clinicians and pioneer neuroscientists began to identify the general brain regions that regulate sleep and wakefulness. After an epidemic of encephalitis lethargica around 1915-1920, Baron Constantin von Economo found that patients with encephalitis of the posterior hypothalamus and rostral midbrain often had crushing sleepiness, whereas those with injury to the preoptic area usually had severe insomnia.[2] He thus hypothesized that the preoptic area and adjacent anterior hypothalamus contained neurons that promoted sleep, whereas neurons in the posterior hypothalamus and rostral midbrain promoted wakefulness. In the 1940s, Moruzzi and Magoun found that stimulation of the rostral reticular formation caused the EEG of an anesthetized animal to switch from slow waves to the low-voltage desynchronized pattern typical of wakefulness, suggesting that this general region is capable of promoting arousal.[3] Soon after the discovery of REM sleep in the mid-1950s, Jouvet and others established that this state is driven by circuitry in the pons.[4-6] Over the last few decades, the latest generations of researchers and clinicians have built on these ideas and identified many distinct systems, each of which contributes to specific aspects of sleep-wake behavior.

Submitted for publication February, 2011
Submitted in final revised form March, 2011
Accepted for publication March, 2011

Address correspondence to: T. E. Scammell: Department of Neurology, Beth Israel Deaconess Medical Center, 330 Brookline Ave., Boston, MA 02215; Tel: (617)-735-3260; Fax: (617)-735-3252; E-mail: tscammel@bidmc.harvard.edu

The Reticular Formation

The reticular formation is a heterogeneous region that runs through the core of the brainstem from the medulla up to the midbrain and into the posterior hypothalamus. Soon after Moruzzi and Magoun showed that the rostral reticular formation can activate the cortex, experimental lesions in animals and clinical observations in patients with strokes or tumors confirmed that the rostral reticular formation is necessary for generating wakefulness, as these injuries often produce hypersomnolence or coma.[7,8] Thus, many researchers hypothesized that the reticular formation received inputs from a number of sensory systems and promoted wakefulness via excitatory projections to the thalamus, hypothalamus, and basal forebrain. More recently, researchers have reconsidered the idea of a monolithic reticular formation and instead attribute its functions to the activity of specific systems that promote arousal using acetylcholine, glutamate, or monoamine neurotransmitters (e.g., norepinephrine, histamine, serotonin, and dopamine). These neurotransmitters are generally considered to produce arousal through widespread, often excitatory effects on target neurons. In addition, they can act as neuromodulators to enhance other excitatory or inhibitory inputs to these cells. This modulation can thus amplify neuronal signals over much of the brain to recruit the many systems necessary for waking behaviors. Thus, the term reticular formation is helpful anatomically, but more insight can be gained from understanding the specific cells and pathways contained within this general region.

Wake-Promoting Neurochemical Systems

Acetylcholine (ACh)

The basal forebrain (BF) and brainstem contain large groups of cholinergic neurons that promote wakefulness and REM

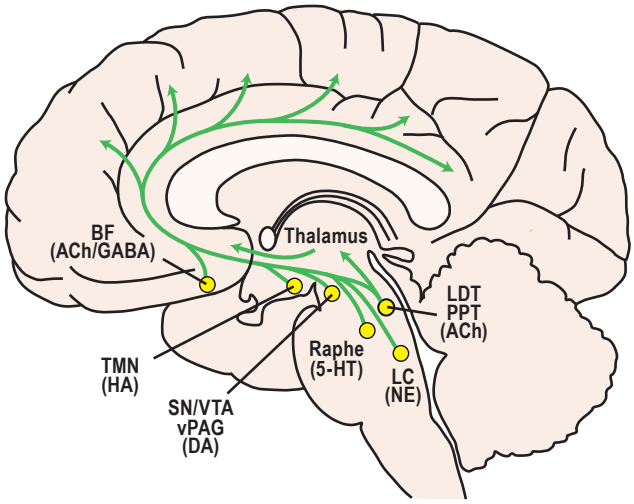

Figure 1—A variety of neurochemical systems promote arousal via projections to the forebrain. Cortical and subcortical regions are excited by monoaminergic neurotransmitters including norepinephrine (NE) from the locus coeruleus (LC), serotonin (5-HT) from the dorsal and median raphe nuclei, histamine (HA) from the tuberomammillary nucleus (TMN); and dopamine (DA) from the substantia nigra, ventral tegmental area, and ventral periaqueductal gray (SN/VTA/vPAG). Neurons of the basal forebrain (BF) promote cortical activation using acetylcholine (ACh) and γ-aminobutyric acid (GABA). Neurons in the laterodorsal and pedunculopontine tegmental nuclei (LDT/PPT) release ACh to excite neurons in the thalamus, hypothalamus, and brainstem.

Table 1—Activity profiles of neurotransmitter systems across sleep/wakefulness

Neurotransmitter	Wakefulness	NREM sleep	REM sleep
Acetylcholine	↑↑	—	↑↑
Monoamines	↑↑	↑	—
Orexin/Hypocretin	↑↑	—	—
MCH	—	—	↑↑
VLPO/MNPO	—	↑↑	↑↑

Neuronal activity: ↑↑, rapid firing rate; ↑, slower firing rate; —, little or no firing.

sleep and also participate in learning, memory, and cognition. The BF is a region surrounding the front of the hypothalamus that includes the medial septum, magnocellular preoptic nucleus, diagonal band of Broca, and substantia innominata (Figure 1). Most BF cholinergic neurons are active during wakefulness and REM sleep, and they directly promote fast EEG rhythms via projections to the cortex and hippocampus (Table 1).[9-12] The BF also contains a large population of neurons that produce the inhibitory neurotransmitter γ-aminobutyric acid (GABA), and these likely activate the cortex by reducing activity in inhibitory cortical interneurons.[13,14]

A second major group of cholinergic neurons is found in the pons within the laterodorsal and pedunculopontine tegmental nuclei (LDT/PPT). In contrast to the BF, LDT/PPT neurons primarily project to subcortical regions including the thalamus, lateral hypothalamus, and BF.[15,16] Like most BF neurons, cholinergic neurons in the LDT/PPT are mainly active during wakefulness and REM sleep, and promote cortical activation by releasing ACh into the thalamus.[17,18]

Pharmacological studies that manipulate ACh neurotransmission offer further evidence for its importance in the control of sleep and wakefulness. ACh, nicotine, and muscarinic receptor agonists such as pilocarpine produce desynchronized cortical activity and increases wakefulness.[19,20] Similar effects occur with physostigmine which blocks enzymatic degradation of ACh. In contrast, agents that reduce ACh signaling, including the muscarinic antagonists scopolamine and atropine, produce immobility and EEG slow waves.[21,22]

Norepinephrine (NE)

NE is produced by several brainstem nuclei and may help generate arousal during conditions that require high attention or activation of the sympathetic nervous system. The major source of NE to the forebrain is the locus coeruleus (LC), an elongated nucleus just beneath the floor of the fourth ventricle. LC neurons fire most rapidly during wakefulness, are much less active during NREM sleep, and are nearly silent during REM sleep.[23,24] Extracellular levels of NE are linearly related to LC neuronal activity, with the highest rates of release observed during wakefulness.[25] NE is also made by neurons in the ventral medulla that mediate autonomic responses, and though much less studied, these cells may also promote arousal.[26]

Pharmacological studies provide some of the strongest evidence that NE regulates wakefulness and sleep. For example, infusion of NE or the noradrenergic agonists isoproterenol and phenylephrine into the lateral ventricle or BF increases behavioral and EEG indices of wakefulness.[27] LC neurons are normally inhibited by NE via α_2 receptors, and blockade of this negative feedback with yohimbine increases LC activity and also increases wakefulness.[28,29] Conversely, bilateral inactivation of the LC with the α_2-agonist clonidine, or co-administration of both α_1 and β noradrenergic antagonists (prazosin and timolol) increases NREM sleep.[30,31]

The NE system may be especially important in promoting arousal under conditions that require responding to a behaviorally important stimulus, a cognitive challenge, or stress. In broad terms, an animal may be drowsy and inattentive if LC activity is too low, distractible and anxious if LC activity is too high, but optimally attentive and aroused with intermediate levels of activity. NE tone is clearly linked to cognition as LC neurons in monkeys fire phasically in response to a salient stimulus that signals a reward such as food, but these cells do not respond to a distracting stimulus.[32] Integrating these ideas, Aston-Jones and colleagues have proposed that activity in the LC may promote arousal in a way that optimizes attention and task performance.[33] In addition, the LC and NE neurons in the ventral medulla are active during stress.[23,34] The necessity of this system is clear in mice lacking NE because after exposure to a mild stressor, they fall asleep more rapidly than control mice.[35] Similarly, rats with lesions of the LC show considerably less behavioral arousal and cortical activation when confronted with novel stimuli.[36] On the other hand, excessive NE tone with anxiety might contribute to insomnia, and the α_1 antagonist prazosin can reduce the vivid nightmares and nighttime arousal of PTSD.[37]

Table 2—Effects of commonly used drugs on sleep and waking

Drug Type	Examples	Pharmacologic Effect	Neurobiologic Mechanism	Clinical Effects
Selective serotonin reuptake inhibitors (SSRIs)	Fluoxetine Fluvoxamine Citalopram	Increase extracellular levels of 5-HT	5-HT inhibits REM sleep-producing cells	Decreased REM sleep
Tricyclic antidepressants	Amitriptyline Nortriptyline Clomipramine Desipramine	Increase extracellular levels of 5-HT and NE	5-HT and NE inhibit REM sleep-producing cells	Decreased REM sleep
Traditional, amphetamine-like stimulants	Amphetamine Dextroamphetamine Methylphenidate	Increase extracellular levels of DA and NE	Increased DA and NE signaling	Increased wakefulness
Wake-promoting, non-traditional stimulants	Modafinil Armodafinil	Increase extracellular levels of DA	Increased DA signaling	Increased wakefulness
Benzodiazepines	Diazepam Clonazepam Lorazepam Triazolam	Enhance GABA signaling via $GABA_A$ receptors	GABA inhibits the arousal systems	Increased sleep
Non-benzodiazepine sedative hypnotics	Zolpidem Zaleplon Zopiclone	Enhance GABA signaling via $GABA_A$ receptors	GABA inhibits the arousal systems	Increased sleep
Classic antihistamines	Diphenhydramine Triprolidine	Block HA H_1 receptors	Reduced HA signaling	Increased sleep
Typical antipsychotics	Haloperidol Chlorpromazine	Block DA receptors	Reduced DA signaling	Increased sleep

Histamine (HA)

HA plays an essential role in promoting wakefulness, yet little is known about which aspects of arousal it governs.[38] The tuberomammillary nucleus (TMN) is a small cluster of cells adjacent to the mammillary body at the base of the posterior hypothalamus. Though few in number, these cells innervate much of the forebrain and brainstem and are the sole source of HA in the brain. Similar to the pattern seen in the LC and other monoaminergic nuclei, TMN firing rates and HA release are highest during wakefulness, lower during NREM sleep and lowest during REM sleep.[39,40] Administration of HA or an H_1-receptor agonist increases cortical activation and wakefulness while reducing NREM and REM sleep.[41,42] In contrast, drugs that reduce HA signaling, including classical antihistamines, such as the H_1 receptor antagonists diphenhydramine, pyrilamine, and low dose doxepin increase NREM and REM sleep (Table 2).[41-46]

Over the last several years, a new class of wake-promoting drugs has been developed to target the autoinhibitory histamine H_3 receptors.[47,48] The clinical rationale for these agents appears strong, as many people with narcolepsy or idiopathic hypersomnia have reduced HA levels,[49] and blockade of H_3 receptors should increase HA signaling. For example, H_3 antagonists (or reverse agonists) such as ciproxifan or tiprolisant promote wakefulness and EEG desynchrony and improve the excessive daytime sleepiness observed with narcolepsy.[50-54] This wake-promoting effect is likely mediated by increased HA tone as the response to an H_3 antagonist is absent in mice lacking H_1 receptors.[55]

Which aspects of arousal are mediated by the HA system remains unclear. HA improves attention and psychomotor performance,[56] and it may promote motivated behaviors such as food seeking.[57] In addition, mice lacking HA have less wakefulness at the beginning of their active period,[58] suggesting that HA may be especially important for initiating arousal. As sleep inertia upon awakening is common in many patients with idiopathic hypersomnia, it is possible that low HA signaling is a contributing factor.

Serotonin (5-HT)

Understanding how 5-HT promotes arousal is challenging because: there are many sources of 5-HT; 5-HT binds to at least 15 different receptors with varied effects, and 5-HT has been shown to influence many other aspects of behavior including mood, anxiety, aggression, and appetite. 5-HT is produced by neurons in the dorsal raphe nucleus and other raphe nuclei scattered along the midline of the brainstem, and together these neurons innervate many brain regions that can influence sleep/wake behavior, including the preoptic area, basal forebrain, hypothalamus, and thalamus. Early studies suggested that 5-HT

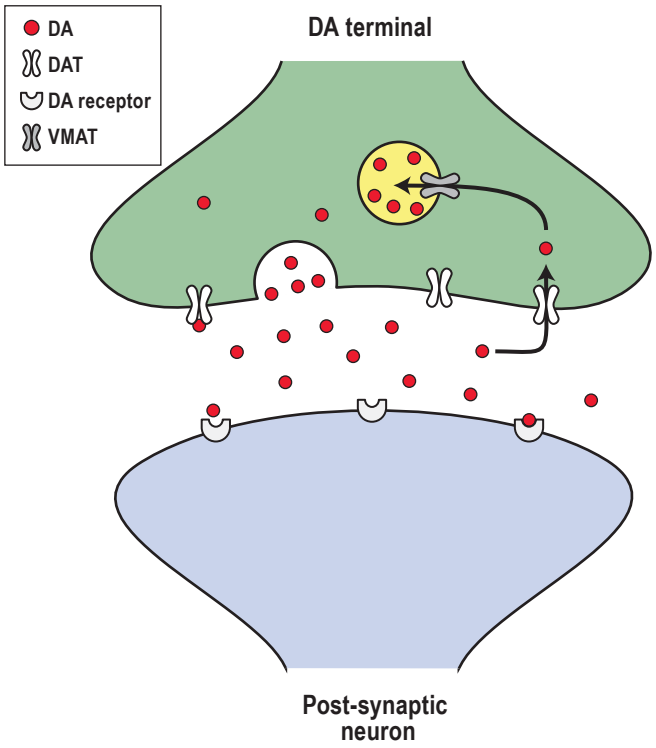

Figure 2—A prototypical dopamine synapse. Under normal conditions, action potentials in a DA nerve terminal cause DA-filled vesicles to fuse with the presynaptic membrane, and DA is released into the synaptic cleft where it can bind to postsynaptic DA receptors. DA is removed from the synaptic cleft primarily by the DA transporter (DAT). Once DA is back inside the presynaptic terminal, it is repackaged into synaptic vesicles for future release via the vesicular monoamine transporter (VMAT). Amphetamines increase the synaptic concentration of DA through two main mechanisms: Amphetamines interfere with the reuptake of DA through the DAT, and they disrupt vesicular packaging of DA which increases cytosolic levels of DA which can then leak out through the DAT via reverse transport.

might help produce NREM and possibly REM sleep, but more recent work indicates 5-HT generally promotes wakefulness and suppresses REM sleep. The firing rates of dorsal raphe neurons and extracellular 5-HT levels are highest during wakefulness, much lower during NREM sleep, and lowest during REM sleep—a pattern very similar to that of the NE and HA systems.[59,60] In support of this wake-promoting role, agonists of the 5-HT_{1A}, 5-HT_{1B}, 5-HT_2, or 5-HT_3 receptors increase wakefulness.[61-65] Of clinical relevance, similar effects occur with selective serotonin reuptake inhibitors (SSRIs) such as fluoxetine and citalopram that increase wakefulness and reduce REM sleep in both people and rodents.[66-70] In addition, drugs that block 5-HT_2 receptors such as ritanserin or agomelatine are thought to promote NREM sleep and thus are under development as treatments for insomnia.[62,64,71-73]

Dopamine (DA)

DA has been implicated in the regulation of a variety of behavioral and physiological processes including motor function, motivation, reward, and learning. Additionally, DA exerts potent wake-promoting effects that are of great clinical relevance. For example, sleepiness is common with DA antagonists such as haloperidol or chlorpromazine or in patients with Parkinson's disease who have a loss of DA-producing neurons.[74-76] Additionally, D_2 agonists like ropinirole can produce sleepiness via activation of autoinhibitory D_2 receptors that reduce DA signaling.[77,78]

However, it is unclear which DA neurons actually promote arousal. DA-producing neurons are most abundant in the substantia nigra and ventral tegmental area, yet cells in these regions fire in relation to movement or reward but, in general, have not been found to alter their rates of firing across sleep and wakefulness.[79-82] Nevertheless, extracellular levels of DA are high during periods of wakefulness and lower during NREM sleep, suggesting that some DA neurons must be wake-active.[81] One candidate population sits in the ventral periaqueductal gray of the pons, and lesions of these wake-active DA neurons produce moderate reductions in wakefulness.[83] The conditions under which these or other DA wake-promoting neurons fire are unknown, but in general, DA may naturally promote arousal when an individual is highly motivated or physically active.

Drugs that increase DA signaling are used frequently to improve excessive daytime sleepiness. Classical stimulants such as methylphenidate and amphetamine increase extracellular levels of DA by disrupting the function of the DA transporter (DAT), thereby increasing extracellular levels of DA (Figure 2).[84] These drugs are usually very effective, but because they enhance DA signaling in reward and motor pathways, they have high abuse potential and can elicit tics or other movement disorders. At higher doses, these stimulants can also block the reuptake of NE and 5-HT which can result in tachycardia, arrhythmias, mania, and psychosis.

Modafinil is frequently prescribed for treating the sleepiness of narcolepsy and some other disorders. Clinically, it promotes wakefulness effectively, usually with fewer side effects than encountered with classical stimulants. Like amphetamines, modafinil disrupts DAT function in humans and rodents,[85,86] and this is a necessary part of its wake-promoting mechanism, as mice lacking the DAT show no increase in wakefulness with modafinil,[87] and D_1 and D_2 receptor antagonists can block modafinil-induced wakefulness.[88] Still, if modafinil acts via the DAT, it seems surprising that it has less abuse potential than amphetamines. One possible explanation is that amphetamines produce a dramatic efflux of DA into the synapse via reverse transport through the DAT, and this may be very reinforcing. In contrast, modafinil may simply block reuptake of DA through the DAT, leading to more modest rises in DA that are not as reinforcing. A better understanding of these mechanisms could drive the discovery of even better wake-promoting medications.

Orexin/Hypocretin

The excitatory neuropeptides orexin-A and -B (also known as hypocretin-1 and -2) are synthesized by neurons in the lateral and posterior hypothalamus and play essential roles in the regulation of wakefulness and sleep.[89,90] The orexin neurons project widely and heavily innervate all the arousal regions described above, with particularly dense innervation of the LC and TMN (Figure 3).[89,91] Orexins excite target neurons through the OX1 and OX2 receptors. Like most other wake-promoting neurons, orexin neurons fire mainly during wakefulness, especially during active exploration, and are silent during NREM and REM sleep.[92,93] Orexin levels are highest during wakefulness,[94] and

when injected into the brain, orexins increase arousal and behavioral activity while suppressing NREM and REM sleep.[95-97] Consistent with this, selective optogenetic activation of orexin neurons can trigger brief awakenings from sleep.[98,99] Additionally, orexin receptor antagonists such as almorexant reduce sleep latency and increase the amounts of REM and NREM sleep.[100-102]

The most compelling evidence that orexins are necessary for the regulation of wakefulness and sleep was the discovery that narcolepsy with cataplexy is associated with a loss of orexin signaling.[103-107] Dogs with a mutation of the OX2 receptor gene display many of the classic symptoms of narcolepsy, including cataplexy when presented with palatable foods.[104] Further, mice lacking the orexin peptides or the orexin-producing neurons have severe sleepiness and cataplexy.[103,108-111] Most importantly, people with narcolepsy with cataplexy have a severe (85% to 95%) loss of the orexin neurons and very low CSF levels of orexin-A.[106,107,112] Less severe loss (20% to 60%) of the orexin neurons also occurs in other disorders that can cause sleepiness such as Parkinson disease, multiple system atrophy, and traumatic brain injury.[113-117]

In just the last 10 years, much has been learned about the ways in which orexins promote arousal. In general, it may be best to think of this as a system for *sustaining* wakefulness as people and mice with narcolepsy have approximately normal amounts of wakefulness, but have great difficulty maintaining long periods of wakefulness.[110] Orexins may also stabilize sleep as people with narcolepsy often have fragmented sleep, and orexins certainly regulate REM sleep as discussed below. In addition, orexins promote arousal responses to homeostatic challenges and drive motivated behaviors such as seeking food. Orexins directly excite neurons of the mesolimbic reward pathways, and orexin antagonists can reduce the motivation to seek drugs of abuse.[118-121] The orexin neurons are also activated by humoral indicators of hunger such as low glucose or high levels of ghrelin,[122,123] and while normal mice have a clear increase in arousal when deprived of food, mice lacking the orexin neurons show little response.[124] Thus, one can view the orexin system as helping sustain wakefulness across much of the day, and increasing arousal in motivating conditions.

Cortical and Thalamic Activity across Sleep and Wakefulness

All the arousal systems we have discussed thus far are located in the BF, hypothalamus, or brainstem and exert diffuse effects on the cortex and many other target regions. These subcortical systems are essential for the generation of sleep/wake states and for the regulation of the transitions between these states. However, patterns of EEG activity and consciousness itself arise from interactions between these subcortical systems, the thalamus, and the cortex.

Thalamic neurons relay information to and from the cortex and have intrinsic electrical characteristics that help generate some of the cortical rhythms seen in NREM sleep.[125,126] The thalamus contains two major types of neurons, glutamatergic thalamocortical projection neurons that relay sensory, motor, and limbic information to the cortex, and GABAergic neurons in the reticular nucleus of the thalamus that are innervated by the projection neurons and cortex and in turn inhibit the projection neurons. These reciprocal connections are thought to drive some cortical rhythms, including sleep spindles.[127] Thalamic

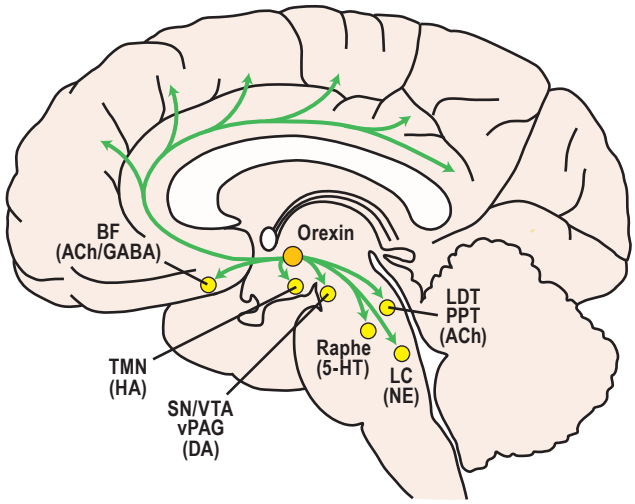

Figure 3—Orexin/hypocretin-producing neurons in the lateral hypothalamus innervate all of the ascending arousal systems, as well as the cerebral cortex.

neurons are hyperpolarized during NREM sleep, promoting a pattern of burst firing and reducing their responsiveness to incoming sensory stimuli.[128] During wakefulness and REM sleep, ACh depolarizes thalamic neurons to suppress spindles and slow waves and promote the transmission of single spikes that efficiently transmit information to the cortex and drive desynchronized cortical activity.[129] During wakefulness, monoamines bolster this effect.[119] Extensive damage to the thalamus severely impairs consciousness and the ability to interact with the environment, but the general patterns of wakefulness, NREM, and REM sleep persist, suggesting that the thalamus is not *required* for the basic generation of sleep states.[130-133]

The cortex contains a wide variety of neurons, and much less is known about their activity in relation to sleep/wake states. The EEG reflects broad patterns of excitatory and inhibitory post-synaptic potentials, mainly arising from the dendrites of pyramidal neurons. During wakefulness and REM sleep, these potentials are desynchronized, resulting in low-amplitude fast activity, but during NREM sleep these signals are synchronized, resulting in high-amplitude slow activity. Release of ACh and monoamines during wakefulness generally excites cortical neurons and increases their responsiveness to incoming sensory stimuli. Delta waves likely arise from interactions amongst cortical neurons and may also be influenced by the BF and other subcortical sites. Recent work has identified a population of widely projecting GABAergic neurons within the cortex that are uniquely active during NREM sleep, suggesting that these cells may broadly inhibit other cortical neurons, helping generate slow waves during NREM sleep.[134] In addition, the intensity of cortical slow waves may reflect prior local activity and changes in synaptic strength, as slow waves during NREM sleep are increased over supplementary motor cortex after learning a motor task but decreased with arm immobilization.[135-137]

The Arousal Network: Interactions among Wake-Promoting Neurotransmitter Systems

Each of the arousal systems presented above is independently capable of promoting wakefulness, yet these systems work

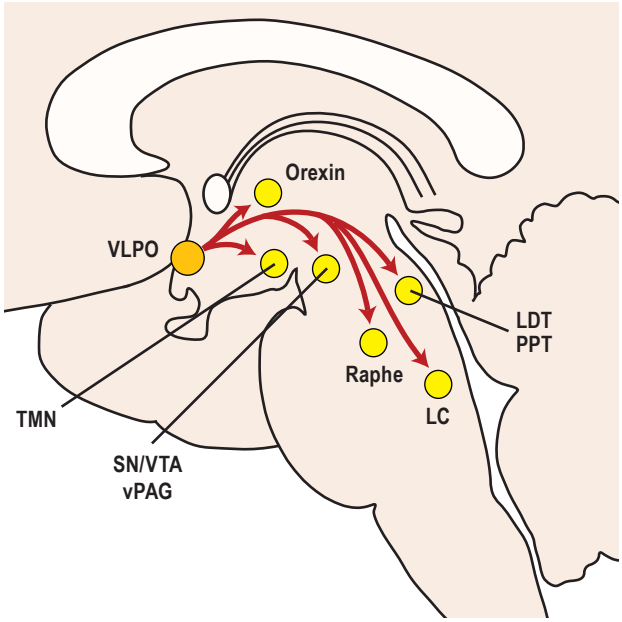

Figure 4—NREM sleep pathways. Ventrolateral preoptic area (VLPO) neurons are active during NREM sleep and reduce activity in the ascending arousal systems using GABA and galanin. A subset of VLPO neurons is also active during REM sleep.

together to generate behavioral arousal. Anatomically, there are many interconnections between the systems. For instance, ACh and 5-HT fibers innervate and excite LC neurons, and nearly all wake-promoting neurons respond to HA, NE, and orexin. In addition, these neurotransmitters often produce similar effects on their targets. For example, all the arousal systems excite thalamic and cortical neurons. These interconnections and parallel effects may explain why injury to any one of the arousal systems often produces little lasting effect on wakefulness. Functionally, this is adaptive, as it helps ensure that wakefulness will still occur after injury to any one of the arousal systems. In fact, there are only a few brain regions in which lesions produce lasting reductions in arousal. One is the rostral reticular formation in the midbrain and posterior hypothalamus in which lesions from strokes or tumors can produce severe hypersomnolence or even coma, probably from damage to many of the ascending monoaminergic and cholinergic pathways.

Wakefulness is a complex and dynamic state, arising from networks of neurons driven by homeostatic, affective, cognitive, and motivational processes. Thus, it is likely that each arousal system helps promote specific aspects of behavioral arousal so that individuals can detect sensory and internal stimuli and generate appropriate motor and affective responses.[138] For example, NE, HA, and ACh may be particularly important for enhancing attention and responding to novel, stressful, or salient stimuli. Similarly, through its limbic and striatal projections, DA may promote arousal especially when an individual is motivated or physically active. The orexin peptides help sustain wakefulness and also may help drive goal-oriented behaviors and locomotion. So, while lesions of some arousal systems appear to have little effect on the amounts of wakefulness, deficits in arousal may be best revealed by carefully examining the response to specific circumstances and challenges.

NREM Sleep-Promoting Systems

Preoptic area

In the early 20th century, most researchers thought that sleep was a passive consequence of inactivity in the arousal systems, but many experiments have now shown that specific neurons actively promote sleep. Baron von Economo first observed that insomnia was common in patients with encephalitis injuring the preoptic area (the rostral end of the hypothalamus, just above the optic chiasm) and the adjacent BF.[2] This observation suggested that this region might contain neurons that promote sleep, and subsequent research in animals identified sleep-active neurons in the ventrolateral preoptic area (VLPO) and median preoptic area (MNPO).[139,140] Many neurons in these nuclei fire most frequently during NREM sleep and to some degree during REM sleep but are virtually silent during wakefulness.[141-143] Interestingly, these sleep active VLPO neurons show particularly high rates of firing during deep NREM sleep, and MNPO neurons often begin firing just before NREM sleep. Lesions of the preoptic area and specifically of the VLPO markedly reduce sleep, and the sleep that does occur is light and fragmented.[144,145] Collectively, these observations suggest that MNPO neurons may help initiate sleep, whereas VLPO neurons may be necessary for the maintenance of sleep.

Anatomically, the VLPO and MNPO are well positioned to promote sleep. The neurons in these nuclei contain the inhibitory neurotransmitter GABA and the inhibitory neuropeptide galanin,[146,147] and they innervate all the arousal-promoting regions, including the LDT/PPT, LC, DR, TMN, and also the orexin neurons (Figure 4). Thus, the VLPO and MNPO are hypothesized to promote sleep by coordinating the inhibition of arousal regions during NREM and REM sleep.[146,148,149]

Other brain regions contain neurons active in NREM sleep, but these populations are less well understood. For example, parts of the BF and lateral hypothalamus contain scattered GABAergic neurons that are active during NREM sleep.[150-152] Some of these cells may directly innervate the cortex, and it is possible that they modulate cortical networks to promote slow wave activity.

Many of the medications now used to treat insomnia do so by promoting GABA signaling. Benzodiazepines, (e.g., diazepam), barbiturates (e.g., pentobarbital), and the newer non-benzodiazepine agents (e.g., zolpidem) all bind to GABA-A receptors to enhance the effects of GABA.[153,154] Gamma hydroxybutyrate (sodium oxybate) promotes very deep sleep, most likely by binding to GABA-B receptors.[155] These drugs may promote sleep by boosting signaling by the VLPO and other NREM sleep-active populations, but GABA can inhibit neurons throughout the brain, and the precise pathways through which these drugs work remain unclear.

REM Sleep-Promoting Systems

Soon after the discovery of REM sleep in the mid-1950s,[4,5] researchers learned that the pons plays an essential role in the generation of REM sleep.[6] After several decades of work, much has been learned, but the specific pathways that generate this state are still debated (Figure 5).

Acetylcholine

Many researchers have hypothesized that REM sleep is controlled by cholinergic neurons located in the LDT/PPT. These are

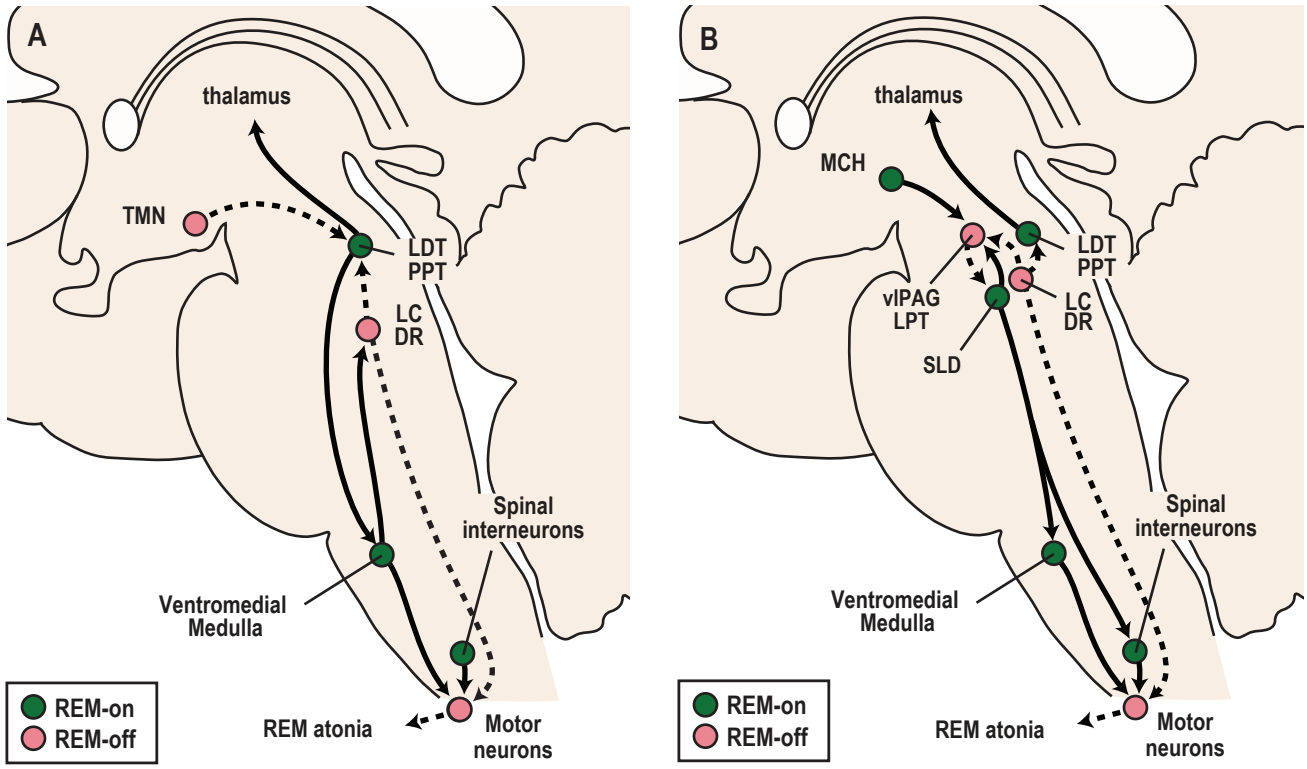

Figure 5—Pathways that control REM sleep. **(A)** A classic perspective on REM sleep control involves interactions between the cholinergic and aminergic systems. REM sleep-active cholinergic neurons in the LDT/PPT activate thalamo-cortical signaling and drive atonia by exciting neurons in the ventromedial medulla that inhibit motor neurons. During REM sleep, monoaminergic neurons including the LC, DR, and TMN become silent, which disinhibits the LDT/PPT and lessens the excitation of motor neurons by NE and 5-HT. **(B)** Recent observations have expanded on the classic view of REM sleep control. In this model, mutual inhibition between REM sleep-on neurons of the sublaterodorsal nucleus (SLD) and REM sleep-off neurons of the ventrolateral periaqueductal gray and lateral pontine tegmentum (vlPAG/LPT) is thought to regulate transitions into and out of REM sleep. During REM sleep, SLD neurons activate GABA/glycine neurons in the ventromedial medulla and spinal cord that inhibit motor neurons. At most times, the vlPAG/LPT inhibits the SLD, but during REM sleep, the vlPAG/LPT may be inhibited by neurons making melanin concentrating hormone (MCH) and other neurotransmitters. Solid lines depict pathways active during REM sleep, while dashed lines are pathways inactive during REM sleep.

the same nuclei that contain wake-promoting cells, but a subpopulation of these cholinergic neurons are active in both wakefulness and REM sleep or are selectively active in REM sleep.[9,156-158] When injected into the lateral pontine tegmentum (LPT; a heterogeneous region extending rostrally from the PPT that is lateral to the periaqueductal gray), drugs that enhance ACh signaling such as the cholinergic agonist carbachol or the acetylcholinesterase blocker, neostigmine, elicit intense and long-lasting REM sleep.[159-161] Conversely, cholinergic antagonists reduce the duration of REM sleep bouts.[162,163] Furthermore, large lesions that include the LDT/PPT produce significant reductions in REM sleep,[164,165] suggesting that the LDT/PPT is necessary for REM sleep.

Neurons in the LDT/PPT may help generate the cortical activation and atonia of REM sleep. The LDT/PPT is the main source of ACh to the thalamus, and ACh depolarizes thalamic neurons to promote the transmission of information through the thalamus, driving the cortical activation that is probably required for the complex dreams of REM sleep. The LDT/PPT neurons may also activate atonia-promoting neurons in the ventromedial medulla.[158,166] These medullary cells release GABA and another inhibitory neurotransmitter glycine onto spinal and brainstem motor neurons during REM sleep, producing hyperpolarization and inhibition.[167] This descending inhibition is clearly important for atonia as drugs that block glycine signaling such as strychnine can markedly increase muscle tone in REM sleep and wakefulness.[168,169]

Monoamines

Monoamines such as NE and 5-HT increase muscle tone by directly exciting motor neurons.[170-173] In the genioglossus muscle, withdrawal of this excitatory tone contributes more to atonia than the inhibitory effects of GABA and glycine.[174,175] Whether this applies to most muscles is unknown, but it is clear that atonia during REM sleep is probably due to a combination of inhibition (GABA and glycine) and a loss of excitation (NE and 5-HT).

Monoamines also inhibit REM sleep itself. During wakefulness, and to some degree during NREM sleep, the REM-active cholinergic neurons are inhibited by 5-HT, NE, and HA.[176] This interaction between cholinergic and monoaminergic populations forms the foundation of the classic model explaining the alternation of NREM and REM sleep across the night (Figure 5A).[177]

These monoaminergic effects on motor tone and REM sleep may account for many phenomena commonly seen by sleep clinicians. NE and 5-HT reuptake inhibitors often increase muscle tone during sleep and can unmask REM sleep behavior disor-

der (RBD) and worsen periodic limb movements of sleep.[178] These drugs and other antidepressants also strongly suppress REM sleep, and thus can markedly reduce REM sleep during overnight polysomnograms or during the MSLT.[179]

GABA

Over the last few years, new observations have expanded on the classic model of REM sleep control (Figure 5B). One region that has received significant attention is the sublaterodorsal nucleus (SLD; also termed the subcoeruleus, or LCα), which is a small cluster of cells ventral to the LC that produce GABA or glutamate.[180] Many neurons in the SLD are active during REM sleep,[158,181-183] and they project to the ventromedial medulla and ventral horn of the spinal cord, providing pathways through which they may inhibit motor neurons. Activation of the SLD region elicits atonia and REM sleep-like EEG activity,[158] while inhibition of the SLD promotes wakefulness and reduces REM sleep. Most importantly, lesions of the SLD region disrupt REM sleep atonia and reduce REM sleep.[165,180,182,184,185] Neuronal loss near the SLD has been reported in some patients with RBD,[186] suggesting that injury to the SLD may contribute to the inadequate atonia of RBD.

Another new perspective on the classic view of REM sleep is that the SLD neurons may be strongly inhibited by REM sleep-suppressing neurons in the mid-pons.[149,182,187] These GABAergic cells are scattered from the ventral part of the periaqueductal gray out into the lateral pontine tegmentum (vlPAG/LPT) and lesions of this region substantially increase REM sleep.[188] The vlPAG/LPT inhibits the SLD, and the SLD may in turn inhibit the vlPAG/LPT, giving rise to a mutually inhibitory circuit that may regulate transitions between NREM and REM sleep.[149,182]

Melanin-concentrating hormone (MCH)

Mixed in with the orexin neurons of the lateral hypothalamus are a large number of REM sleep-active neurons that produce both MCH and GABA.[189-191] These cells innervate nearly all the same target regions as the orexin neurons including the DR and LC,[192,193] yet in contrast to the excitatory effects of orexins, both MCH and GABA are inhibitory. Electrophysiological recordings demonstrate that MCH neurons fire at a high rate during REM sleep, with much less firing during NREM sleep and complete inactivity during wakefulness.[152] The amount of REM sleep is increased by infusions of MCH into the lateral ventricles and decreased by a MCH antagonist.[191,194] Consistent with these observations, mice lacking MCH spend less time in NREM and REM sleep.[195] Thus, it seems likely that the MCH neurons promote REM sleep by inhibiting the arousal regions. This pattern is strikingly opposite to that of the orexin neurons and much remains to be learned about how the activity of these intertwined systems is organized.

Mechanisms that Regulate the Transitions between Sleep and Wakefulness

The systems that promote wakefulness, NREM, and REM sleep dynamically interact in a variety of ways to ensure rapid and complete transitions between sleep/wake states.[148,149] The VLPO and other sleep-promoting preoptic neurons inhibit monoaminergic and cholinergic wake-promoting neurons, and the preoptic neurons themselves are inhibited by NE, 5-HT, and ACh.[196,197] During wakefulness, high monoaminergic and cholinergic tone should thoroughly silence the VLPO, thus disinhibiting the arousal regions and helping ensure the production of complete wakefulness. Conversely, during sleep, preoptic neurons become active and inhibit the arousal regions, thus disinhibiting their own firing. This mutual inhibition should produce stable wakefulness and sleep while facilitating rapid transitions between sleep and wakefulness and minimizing time in drowsy, intermediate states. Similar mutually inhibitory circuits may regulate REM sleep as REM sleep-active neurons in the SLD inhibit and are inhibited by neurons in the vlPAG/LPT that are inactive in REM sleep.[182]

The orexin neuropeptides probably reinforce these mutually inhibitory systems. Orexins may stabilize wakefulness by enhancing activity in the arousal systems, ensuring full alertness and long periods of wakefulness despite rising homeostatic pressure across the day.[94] During wakefulness and perhaps to a lesser degree in NREM sleep, orexins may excite a variety of neurons that inhibit REM sleep, including monoaminergic neurons, the vlPAG/LPT, and GABAergic inputs to the SLD.[96,198-201] However, loss of the orexin neurons in narcolepsy with cataplexy results in persistent sleepiness, frequent transitions between states, and odd states such as cataplexy and hypnagogic hallucinations in which it seems that elements of REM sleep mix into wakefulness. Collectively, these symptoms may be best thought of as "behavioral state instability," a phenomena that is likely caused by loss of the stabilizing effects of orexins on the mutually inhibitory circuits that regulate wakefulness, NREM, and REM sleep.[110,202]

Somnogens

Most of the preceding text describes neural pathways that regulate sleep/wake states, but these states can also be influenced by diffusible or circulating factors that act upon many brain regions to promote sleep. In fact, more than 100 years ago, researchers found that the CSF of sleep deprived dogs contained somnogens, substances that promote sleep.[203,204] Much evidence now suggests that adenosine, cytokines, prostaglandins, and probably additional substances serve as natural sleep-generating signals.[205]

Adenosine

During wakefulness, brain metabolic activity is high, and adenosine may promote sleep in response to this metabolic challenge.[206-208] When cells have ample energy, nearly all adenosine is phosphorylated to ATP and adenosine levels are low. However, when cells are fatigued, ATP production is lower, adenosine levels rise, and then adenosine acts as an inhibitory neuromodulator. For example, adenosine reduces the activity of most wake-promoting neurons, but disinhibits VLPO neurons. With prolonged wakefulness, adenosine levels rise in the basal forebrain and other regions, and levels then fall during recovery sleep.[209] Most likely, the extracellular levels of adenosine are governed by the activity of astrocytes, the support cells of the brain, because manipulations of astrocytes reduce the usual increases in sleep and delta power after sleep deprivation.[207,209,210] Furthermore, adenosine receptor agonists increase sleep and NREM delta power, while caffeine and other drugs that block adenosine receptors promote wakefulness.[207,211-213] As caffeine promotes arousal after sleep depriva-

tion,[214] it seems likely that adenosine is an important mediator of everyday sleepiness.

Cytokines

Cytokines are intercellular signaling peptides released by immune cells, neurons, and astrocytes, and several cytokines, including interleukin-1β (IL-1β) and tumor necrosis factor-α (TNF-α), promote sleep.[215] Administration of IL-1β into the preoptic area of rats reduces firing rates of wake-active neurons and promotes NREM sleep.[216] Similarly, TNF-α infusions into the preoptic area also promote NREM sleep.[217] During infections, bacterial cell wall products such as lipopolysaccharide and muramyl dipeptide may trigger production of these cytokines that then increase NREM sleep and reduce REM sleep.[218] In addition, these cytokines may promote spontaneous, physiological sleep as IL-1β and TNF-α mRNA and protein levels are highest around sleep onset, and blockade of IL-1β and TNF-α signaling with antagonists, antibodies, or deletion of their receptors reduces spontaneous NREM sleep.[219-221]

Prostaglandin D2

Prostaglandin D2 (PGD2) is a lipid derived from fatty acids that potently promotes NREM sleep.[211] Unlike adenosine or cytokines which are made in the brain parenchyma, PGD2 is likely synthesized in the basal meninges just below the hypothalamus.[222] PGD2 levels in cerebrospinal fluid are highest during the sleep period,[223] and PDG2 levels increase with sleep deprivation.[224] Infusions of PGD2 just below or within the preoptic area activate neurons in the VLPO and increase NREM and REM sleep, perhaps by increasing local concentrations of adenosine.[225-229] Like cytokines, PGD2 may contribute to the sleepiness seen with inflammation as patients with African sleeping sickness display increased CSF levels of PGD2.[230]

Process C and Process S

The two-process model provides a useful macroscopic perspective on the dynamic control of sleep and wakefulness. It is likely that a homeostatic factor (process S) accumulates during wakefulness and declines during sleep, and this factor interacts with a circadian process (process C) that helps regulate the timing of wakefulness and REM sleep.[231-233] After a period of wakefulness, delta power in NREM sleep is thought to be a good indicator of Process S,[234,235] and somnogens such as adenosine may be the neurobiologic equivalent of Process S as disruption of adenosine signaling can blunt the usual increase in NREM sleep and the intense EEG delta power seen after sleep deprivation.[210]

Process C is driven by the suprachiasmatic nucleus (SCN), the master pacemaker that regulates the circadian rhythms of sleep, wakefulness, and most other physiologic rhythms.[236] The activity of individual SCN neurons is strikingly rhythmic, especially when coupled with other SCN neurons.[237] This rhythmicity arises from positive and negative feedback loops in the transcription and translation of several genes.[238] To synchronize its activity with the environmental light-dark cycle, the SCN uses luminance information from photosensitive retinal ganglion cells that contain the photopigment melanopsin.[239] SCN neurons then relay these timing signals to the adjacent subparaventricular zone, using neuropeptides such as prokineticin 2 or transforming growth factor-α.[240,241] This signal is then passed through the dorsomedial nucleus of the hypothalamus and on to brain regions that regulate sleep and wakefulness such as the LC, VLPO, and lateral hypothalamus.[242] The SCN also regulates the daily rhythm of body temperature, and through these cycles in temperature, the SCN can entrain circadian activity in cells throughout the body.[243] Circadian rhythms are closely linked to metabolism, and a breakdown in this coordination of central and peripheral rhythms may contribute to the obesity and glucose intolerance that is common in people with shift work sleep disorder or insufficient sleep.[244]

CONCLUSIONS

Since the days of von Economo and then Moruzzi and Magoun, much has been learned about the neurobiology of sleep and wakefulness. We now know that neurons producing ACh and monoamines such as NE, 5-HT, DA, and HA promote various aspects of wakefulness. In addition, orexins/hypocretins help sustain long periods of wakefulness while suppressing REM sleep. NREM sleep is mainly regulated by neural pathways originating in the VLPO and other preoptic regions, yet it is also influenced by diffusible somnogens such as adenosine. REM sleep is driven by neurons in the pons that make ACh and GABA. These discoveries provide a useful framework to better understand sleep disorders and the effects of medications on sleep.

Nevertheless, despite these advances, many questions of clinical importance remain unanswered. What goes wrong in these circuits to cause parasomnias such as sleepwalking and periodic limb movements of sleep? Under what conditions are specific wake- and sleep-promoting systems especially necessary? How is sleep restorative? What are the functions of NREM and REM sleep? Undoubtedly, future sleep research will provide helpful insights into the underlying causes of sleep disorders and lead to new and more powerful therapeutics to treat them.

ACKNOWLEDGMENTS

The authors thank Dr. P. Valko, D. Kroeger, and C. Burgess for their thoughtful comments on this manuscript. Writing of this article was partially supported by research grants from the NIH (NS055367, HL095491).

DISCLOSURE STATEMENT

This was not an industry supported study. Dr. Scammell has consulted for Merck and Cephalon. Dr. España has indicated no financial conflicts of interest.

REFERENCES

1. España RA, Scammell TE. Sleep neurobiology for the clinician. Sleep 2004;27:811-20.
2. von Economo C. Sleep as a problem of localization. J Nerv Ment Dis 1930;71:249-59.
3. Moruzzi G, Magoun HW. Brain stem reticular formation and activation of the EEG. Electroencephalogr Clin Neurophysiol 1949;1:455-73.
4. Aserinsky E, Kleitman N. A motility cycle in sleeping infants as manifested by ocular and gross bodily activity. J Appl Physiol 1955;8:11-8.
5. Dement W, Kleitman N. Cyclic variations in EEG during sleep and their relation to eye movements, body motility, and dreaming. Electroencephalogr Clin Neurophysiol 1957;9:673-90.
6. Jouvet M, Michel F. New research on the structures responsible for the "paradoxical phase" of sleep. J Physiol (Paris) 1960;52:130-1.

7. Lindsley DB, Bowden JW, Magoun HW. Effect upon the EEG of acute injury to the brain stem activating system. Electroencephalogr Clin Neurophysiol 1949;1:475-86.
8. Posner J, Saper CB, Schiff N, Plum F. Plum and Posner's diagnosis of stupor and coma. 4th ed. Oxford University Press, 2007.
9. el Mansari M, Sakai K, Jouvet M. Unitary characteristics of presumptive cholinergic tegmental neurons during the sleep-waking cycle in freely moving cats. Exp Brain Res 1989;76:519-29.
10. Steriade M, Datta S, Pare D, Oakson G, Curro Dossi RC. Neuronal activities in brain-stem cholinergic nuclei related to tonic activation processes in thalamocortical systems. J Neurosci 1990;10:2541-59.
11. Lee MG, Manns ID, Alonso A, Jones BE. Sleep-wake related discharge properties of basal forebrain neurons recorded with micropipettes in head-fixed rats. J Neurophysiol 2004;92:1182-98.
12. Boucetta S, Jones BE. Activity profiles of cholinergic and intermingled GABAergic and putative glutamatergic neurons in the pontomesencephalic tegmentum of urethane-anesthetized rats. J Neurosci 2009;29:4664-74.
13. Gritti I, Mainville L, Mancia M, Jones BE. GABAergic and other noncholinergic basal forebrain neurons, together with cholinergic neurons, project to the mesocortex and isocortex in the rat. J Comp Neurol 1997;383:163-77.
14. Henny P, Jones BE. Projections from basal forebrain to prefrontal cortex comprise cholinergic, GABAergic and glutamatergic inputs to pyramidal cells or interneurons. Eur J Neurosci 2008;27:654-70.
15. Satoh K, Fibiger HC. Cholinergic neurons of the laterodorsal tegmental nucleus: efferent and afferent connections. J Comp Neurol 1986;253:277-302.
16. Hallanger AE, Levey AI, Lee HJ, Rye DB, Wainer BH. The origins of cholinergic and other subcortical afferents to the thalamus in the rat. J Comp Neurol 1987;262:105-24.
17. Williams JA, Comisarow J, Day J, Fibiger HC, Reiner PB. State-dependent release of acetylcholine in rat thalamus measured by in vivo microdialysis. J Neurosci 1994;14:5236-42.
18. Marrosu F, Portas C, Mascia MS, et al. Microdialysis measurement of cortical and hippocampal acetylcholine release during sleep-wake cycle in freely moving cats. Brain Res 1995;671:329-32.
19. Yamamoto KI, Domino EF. Cholinergic agonist-antagonist interactions on neocortical and limbic EEG activation. Int J Neuropharmacol 1967;6:357-73.
20. Davila DG, Hurt RD, Offord KP, Harris CD, Shepard JW Jr. Acute effects of transdermal nicotine on sleep architecture, snoring, and sleep-disordered breathing in nonsmokers. Am J Respir Crit Care Med 1994;150:469-74.
21. Spehlmann R, Norcross K. Cholinergic mechanisms in the production of focal cortical slow waves. Experientia 1982;38:109-11.
22. Vanderwolf CH. Neocortical and hippocampal activation relation to behavior: effects of atropine, eserine, phenothiazines, and amphetamine. J Comp Physiol Psychol 1975;88:300-23.
23. Foote SL, Aston-Jones G, Bloom FE. Impulse activity of locus coeruleus neurons in awake rats and monkeys is a function of sensory stimulation and arousal. Proc Natl Acad Sci U S A 1980;77:3033-7.
24. Aston-Jones G, Bloom FE. Activity of norepinephrine-containing locus coeruleus neurons in behaving rats anticipates fluctuations in the sleep-waking cycle. J Neurosci 1981;1:876-86.
25. Berridge CW, Abercrombie ED. Relationship between locus coeruleus discharge rates and rates of norepinephrine release within neocortex as assessed by in vivo microdialysis. Neuroscience 1999;93:1263-70.
26. España RA, Vlasaty J, McCormack SL, Llewellyn-Smith IJ, Scammell TE. Aminergic inputs to the hypocretin/orexin neurons. Society for Neuroscience Meeting, Washington, DC, 2005.
27. Berridge CW, Isaac SO, España RA. Additive wake-promoting actions of medial basal forebrain noradrenergic alpha1- and beta-receptor stimulation. Behav Neurosci 2003;117:350-9.
28. De Sarro GB, Ascioti C, Froio F, Libri V, Nistico G. Evidence that locus coeruleus is the site where clonidine and drugs acting at alpha 1- and alpha 2-adrenoceptors affect sleep and arousal mechanisms. Br J Pharmacol 1987;90:675-85.
29. Berridge CW, Foote SL. Effects of locus coeruleus activation on electroencephalographic activity in neocortex and hippocampus. J Neurosci 1991;11:3135-45.
30. Berridge CW, Page ME, Valentino RJ, Foote SL. Effects of locus coeruleus inactivation on electroencephalographic activity in neocortex and hippocampus. Neuroscience 1993;55:381-93.
31. Berridge CW, España RA. Synergistic sedative effects of noradrenergic alpha(1)- and beta- receptor blockade on forebrain electroencephalographic and behavioral indices. Neuroscience 2000;99:495-505.
32. Aston-Jones G, Rajkowski J, Kubiak P, Alexinsky T. Locus coeruleus neurons in monkey are selectively activated by attended cues in a vigilance task. J Neurosci 1994;14:4467-80.
33. Aston-Jones G, Cohen JD. An integrative theory of locus coeruleus-norepinephrine function: adaptive gain and optimal performance. Annu Rev Neurosci 2005;28:403-50.
34. Dayas CV, Buller KM, Crane JW, Xu Y, Day TA. Stressor categorization: acute physical and psychological stressors elicit distinctive recruitment patterns in the amygdala and in medullary noradrenergic cell groups. Eur J Neurosci 2001;14:1143-52.
35. Hunsley MS, Palmiter RD. Norepinephrine-deficient mice exhibit normal sleep-wake states but have shorter sleep latency after mild stress and low doses of amphetamine. Sleep 2003;26:521-6.
36. Gompf HS, Mathai C, Fuller PM, et al. Locus ceruleus and anterior cingulate cortex sustain wakefulness in a novel environment. J Neurosci 2010;30:14543-51.
37. Byers MG, Allison KM, Wendel CS, Lee JK. Prazosin versus quetiapine for nighttime posttraumatic stress disorder symptoms in veterans: an assessment of long-term comparative effectiveness and safety. J Clin Psychopharmacol 2010;30:225-9.
38. Haas HL, Sergeeva OA, Selbach O. Histamine in the nervous system. Physiol Rev 2008;88:1183-241.
39. Sakai K, el Mansari M, Lin JS, Zhang ZG, Vanni-Mercier G. The posterior hypothalamus in the regulation of wakefulness and paradoxical sleep. In: Mancia M, Marini M, eds. The diencephalon and sleep. New York: Raven Press, 1990:171-98.
40. Mochizuki T, Yamatodani A, Okakura K, Horii A, Inagaki N, Wada H. Circadian rhythm of histamine release from the hypothalamus of freely moving rats. Physiol Behav 1992;51:391-4.
41. Lin JS, Sakai K, Jouvet M. Evidence for histaminergic arousal mechanisms in the hypothalamus of cat. Neuropharmacology 1988;27:111-22.
42. Monti JM, Pellejero T, Jantos H. Effects of H1- and H2-histamine receptor agonists and antagonists on sleep and wakefulness in the rat. J Neural Transm 1986;66:1-11.
43. Roehrs TA, Tietz EI, Zorick FJ, Roth T. Daytime sleepiness and antihistamines. Sleep 1984;7:137-41.
44. Tasaka K, Chung YH, Sawada K, Mio M. Excitatory effect of histamine on the arousal system and its inhibition by H1 blockers. Brain Res Bull 1989;22:271-5.
45. Hajak G, Rodenbeck A, Voderholzer U, et al. Doxepin in the treatment of primary insomnia: a placebo-controlled, double-blind, polysomnographic study. J Clin Psychiatry 2001;62:453-63.
46. Krystal AD, Durrence HH, Scharf M, et al. Efficacy and safety of doxepin 1 mg and 3 mg in a 12-week sleep laboratory and outpatient trial of elderly subjects with chronic primary insomnia. Sleep 2010;33:1553-61.
47. Celanire S, Wijtmans M, Talaga P, Leurs R, de Esch IJ. Keynote review: histamine H3 receptor antagonists reach out for the clinic. Drug Discov Today 2005;10:1613-27.
48. Lin JS, Sergeeva OA, Haas HL. Histamine H3 receptors and sleep-wake regulation. J Pharmacol Exp Ther 2011;336:17-23.
49. Kanbayashi T, Kodama T, Kondo H, et al. CSF histamine contents in narcolepsy, idiopathic hypersomnia and obstructive sleep apnea syndrome. Sleep 2009;32:181-7.
50. Lin JS, Sakai K, Vanni-Mercier G, et al. Involvement of histaminergic neurons in arousal mechanisms demonstrated with H3-receptor ligands in the cat. Brain Res 1990;523:325-30.
51. Monti JM, Jantos H, Ponzoni A, Monti D. Sleep and waking during acute histamine H3 agonist BP 2.94 or H3 antagonist carboperamide (MR 16155) administration in rats. Neuropsychopharmacology 1996;15:31-5.
52. Bonaventure P, Letavic M, Dugovic C, et al. Histamine H3 receptor antagonists: from target identification to drug leads. Biochem Pharmacol 2007;73:1084-96.
53. Ligneau X, Perrin D, Landais L, et al. BF2.649 [1-{3-[3-(4-Chlorophenyl)propoxy] propyl}piperidine, hydrochloride], a nonimidazole inverse agonist/antagonist at the human histamine H3 receptor: Preclinical pharmacology. J Pharmacol Exp Ther 2007;320:365-75.
54. Lin JS, Dauvilliers Y, Arnulf I, et al. An inverse agonist of the histamine H(3) receptor improves wakefulness in narcolepsy: studies in orexin-/- mice and patients. Neurobiol Dis 2008;30:74-83.

55. Huang ZL, Mochizuki T, Qu WM, et al. Altered sleep-wake characteristics and lack of arousal response to H3 receptor antagonist in histamine H1 receptor knockout mice. Proc Natl Acad Sci U S A 2006;103:4687-92.
56. van RP, Vermeeren A, Riedel WJ. Cognitive domains affected by histamine H(1)-antagonism in humans: a literature review. Brain Res Rev 2010;64:263-82.
57. Passani MB, Blandina P, Torrealba F. The histamine H3 receptor and eating behavior. J Pharmacol Exp Ther 2011;336:24-9.
58. Parmentier R, Ohtsu H, Djebbara-Hannas Z, Valatx JL, Watanabe T, Lin JS. Anatomical, physiological, and pharmacological characteristics of histidine decarboxylase knock-out mice: evidence for the role of brain histamine in behavioral and sleep-wake control. J Neurosci 2002;22:7695-711.
59. Trulson ME, Jacobs BL. Raphe unit activity in freely moving cats: correlation with level of behavioral arousal. Brain Res 1979;163:135-50.
60. Portas CM, Bjorvatn B, Fagerland S, et al. On-line detection of extracellular levels of serotonin in dorsal raphe nucleus and frontal cortex over the sleep/wake cycle in the freely moving rat. Neuroscience 1998;83:807-14.
61. Dzoljic MR, Ukponmwan OE, Saxena PR. 5-HT1-like receptor agonists enhance wakefulness. Neuropharmacology 1992;31:623-33.
62. Dugovic C, Wauquier A, Leysen JE, Marrannes R, Janssen PA. Functional role of 5-HT2 receptors in the regulation of sleep and wakefulness in the rat. Psychopharmacology (Berl) 1989;97:436-42.
63. Bjorvatn B, Ursin R. Effects of the selective 5-HT1B agonist, CGS 12066B, on sleep/waking stages and EEG power spectrum in rats. J Sleep Res 1994;3:97-105.
64. Boutrel B, Franc B, Hen R, Hamon M, Adrien J. Key role of 5-HT1B receptors in the regulation of paradoxical sleep as evidenced in 5-HT1B knock-out mice. J Neurosci 1999;19:3204-12.
65. Ponzoni A, Monti JM, Jantos H. The effects of selective activation of the 5-HT3 receptor with m-chlorophenylbiguanide on sleep and wakefulness in the rat. Eur J Pharmacol 1993;249:259-64.
66. Bakalian MJ, Fernstrom JD. Effects of L-tryptophan and other amino acids on electroencephalographic sleep in the rat. Brain Res 1990;528:300-7.
67. Maudhuit C, Jolas T, Lainey E, Hamon M, Adrien J. Effects of acute and chronic treatment with amoxapine and cericlamine on the sleep-wakefulness cycle in the rat. Neuropharmacology 1994;33:1017-25.
68. Vasar V, Appelberg B, Rimon R, Selvaratnam J. The effect of fluoxetine on sleep: a longitudinal, double-blind polysomnographic study of healthy volunteers. Int Clin Psychopharmacol 1994;9:203-6.
69. Monaca C, Boutrel B, Hen R, Hamon M, Adrien J. 5-HT 1A/1B receptor-mediated effects of the selective serotonin reuptake inhibitor, citalopram, on sleep: studies in 5-HT 1A and 5-HT 1B knockout mice. Neuropsychopharmacology 2003;28:850-6.
70. Vazquez-Palacios G, Hernandez-Gonzalez M, Guevara Perez MA, Bonilla-Jaime H. Nicotine and fluoxetine induce arousing effects on sleep-wake cycle in antidepressive doses: a possible mechanism of antidepressant-like effects of nicotine. Pharmacol Biochem Behav 2010;94:503-9.
71. Monti JM. Serotonin 5-HT(2A) receptor antagonists in the treatment of insomnia: present status and future prospects. Drugs Today (Barc) 2010;46:183-93.
72. Teegarden BR, Al SH, Xiong Y. 5-HT(2A) inverse-agonists for the treatment of insomnia. Curr Top Med Chem 2008;8:969-76.
73. Lemoine P, Guilleminault C, Alvarez E. Improvement in subjective sleep in major depressive disorder with a novel antidepressant, agomelatine: randomized, double-blind comparison with venlafaxine. J Clin Psychiatry 2007;68:1723-32.
74. Neylan TC, van Kammen DP, Kelley ME, Peters JL. Sleep in schizophrenic patients on and off haloperidol therapy. Clinically stable vs relapsed patients. Arch Gen Psychiatry 1992;49:643-9.
75. Ongini E, Bonizzoni E, Ferri N, Milani S, Trampus M. Differential effects of dopamine D-1 and D-2 receptor antagonist antipsychotics on sleep-wake patterns in the rat. J Pharmacol Exp Ther 1993;266:726-31.
76. Arnulf I, Leu S, Oudiette D. Abnormal sleep and sleepiness in Parkinson's disease. Curr Opin Neurol 2008;21:472-7.
77. Paus S, Brecht HM, Koster J, Seeger G, Klockgether T, Wullner U. Sleep attacks, daytime sleepiness, and dopamine agonists in Parkinson's disease. Mov Disord 2003;18:659-67.
78. Arnulf I. Excessive daytime sleepiness in parkinsonism. Sleep Med Rev 2005;9:185-200.
79. Schultz W. Predictive reward signal of dopamine neurons. J Neurophysiol 1998;80:1-27.
80. Schultz W. Multiple dopamine functions at different time courses. Annu Rev Neurosci 2007;30:259-88.
81. Trulson ME. Simultaneous recording of substantia nigra neurons and voltammetric release of dopamine in the caudate of behaving cats. Brain Res Bull 1985;15:221-3.
82. Trulson ME, Preussler DW. Dopamine-containing ventral tegmental area neurons in freely moving cats: activity during the sleep-waking cycle and effects of stress. Exp Neurol 1984;83:367-77.
83. Lu J, Jhou TC, Saper CB. Identification of wake-active dopaminergic neurons in the ventral periaqueductal gray matter. J Neurosci 2006;26:193-202.
84. Schmitt KC, Reith ME. Regulation of the dopamine transporter: aspects relevant to psychostimulant drugs of abuse. Ann N Y Acad Sci 2010;1187:316-40.
85. Volkow ND, Fowler JS, Logan J, et al. Effects of modafinil on dopamine and dopamine transporters in the male human brain: clinical implications. JAMA 2009;301:1148-54.
86. Mignot E, Nishino S, Guilleminault C, Dement WC. Modafinil binds to the dopamine uptake carrier site with low affinity. Sleep 1994;17:436-7.
87. Wisor JP, Nishino S, Sora I, Uhl GH, Mignot E, Edgar DM. Dopaminergic role in stimulant-induced wakefulness. J Neurosci 2001;21:1787-94.
88. Qu WM, Huang ZL, Xu XH, Matsumoto N, Urade Y. Dopaminergic D1 and D2 receptors are essential for the arousal effect of modafinil. J Neurosci 2008;28:8462-9.
89. Sakurai T, Amemiya A, Ishii M, et al. Orexins and orexin receptors: a family of hypothalamic neuropeptides and G protein-coupled receptors that regulate feeding behavior. Cell 1998;92:1.
90. de Lecea L, Kilduff TS, Peyron C, et al. The hypocretins: hypothalamus-specific peptides with neuroexcitatory activity. Proc Natl Acad Sci U S A 1998;95:322-7.
91. Peyron C, Tighe DK, van Den Pol AN, et al. Neurons containing hypocretin (orexin) project to multiple neuronal systems. J Neurosci 1998;18:9996-10015.
92. Mileykovskiy BY, Kiyashchenko LI, Siegel JM. Behavioral correlates of activity in identified hypocretin/orexin neurons. Neuron 2005;46:787-98.
93. Lee MG, Hassani OK, Jones BE. Discharge of identified orexin/hypocretin neurons across the sleep-waking cycle. J Neurosci 2005;25:6716-20.
94. Zeitzer JM, Buckmaster CL, Parker KJ, Hauck CM, Lyons DM, Mignot E. Circadian and homeostatic regulation of hypocretin in a primate model: implications for the consolidation of wakefulness. J Neurosci 2003;23:3555-60.
95. Hagan JJ, Leslie RA, Patel S, et al. Orexin A activates locus coeruleus cell firing and increases arousal in the rat. Proc Natl Acad Sci U S A 1999;96:10911-6.
96. Bourgin P, Huitron-Resendiz S, Spier AD, et al. Hypocretin-1 modulates rapid eye movement sleep through activation of locus coeruleus neurons. J Neurosci 2000;20:7760-5.
97. España RA, Baldo BA, Kelley AE, Berridge CW. Wake-promoting and sleep-suppressing actions of hypocretin (orexin): basal forebrain sites of action. Neuroscience 2001;106:699-715.
98. Adamantidis AR, Zhang F, Aravanis AM, Deisseroth K, de Lecea L. Neural substrates of awakening probed with optogenetic control of hypocretin neurons. Nature 2007;450:420-4.
99. Carter ME, Adamantidis A, Ohtsu H, Deisseroth K, de Lecea L. Sleep homeostasis modulates hypocretin-mediated sleep-to-wake transitions. J Neurosci 2009;29:10939-49.
100. Brisbare-Roch C, Dingemanse J, Koberstein R, et al. Promotion of sleep by targeting the orexin system in rats, dogs and humans. Nat Med 2007;13:150-5.
101. Dugovic C, Shelton JE, Aluisio LE, et al. Blockade of orexin-1 receptors attenuates orexin-2 receptor antagonism-induced sleep promotion in the rat. J Pharmacol Exp Ther 2009;330:142-51.
102. Scammell TE, Winrow CJ. Orexin receptors: pharmacology and therapeutic opportunities. Annu Rev Pharmacol Toxicol 2011;51:243-66.
103. Chemelli RM, Willie JT, Sinton CM, et al. Narcolepsy in orexin knockout mice: molecular genetics of sleep regulation. Cell 1999;98:437-51.
104. Lin L, Faraco J, Li R, et al. The sleep disorder canine narcolepsy is caused by a mutation in the hypocretin (orexin) receptor 2 gene. Cell 1999;98:365-76.
105. Nishino S, Ripley B, Overeem S, Lammers GJ, Mignot E. Hypocretin (orexin) deficiency in human narcolepsy. Lancet 2000;355:39-40.
106. Peyron C, Faraco J, Rogers W, et al. A mutation in a case of early onset narcolepsy and a generalized absence of hypocretin peptides in human narcoleptic brains. Nat Med 2000;6:991-7.

107. Thannickal TC, Moore RY, Nienhuis R, et al. Reduced number of hypocretin neurons in human narcolepsy. Neuron 2000;27:469-74.
108. España RA, McCormack SL, Mochizuki T, Scammell TE. Running promotes wakefulness and increases cataplexy in orexin knockout mice. Sleep 2007;30:1417-25.
109. Hara J, Beuckmann CT, Nambu T, et al. Genetic ablation of orexin neurons in mice results in narcolepsy, hypophagia, and obesity. Neuron 2001;30:345-54.
110. Mochizuki T, Crocker A, McCormack S, Yanagisawa M, Sakurai T, Scammell TE. Behavioral state instability in orexin knock-out mice. J Neurosci 2004;24:6291-300.
111. Scammell TE, Willie JT, Guilleminault C, Siegel JM. A consensus definition of cataplexy in mouse models of narcolepsy. Sleep 2009;32:111-6.
112. Crocker A, Espana RA, Papadopoulou M, et al. Concomitant loss of dynorphin, NARP, and orexin in narcolepsy. Neurology 2005;65:1184-8.
113. Thannickal TC, Lai YY, Siegel JM. Hypocretin (orexin) cell loss in Parkinson's disease. Brain 2007;130:1586-95.
114. Fronczek R, Overeem S, Lee SY, et al. Hypocretin (orexin) loss in Parkinson's disease. Brain 2007;130:1577-85.
115. Benarroch EE, Schmeichel AM, Sandroni P, Low PA, Parisi JE. Involvement of hypocretin neurons in multiple system atrophy. Acta Neuropathol 2007;113:75-80.
116. Baumann CR, Bassetti CL, Valko PO, et al. Loss of hypocretin (orexin) neurons with traumatic brain injury. Ann Neurol 2009;66:555-9.
117. Fronczek R, Baumann CR, Lammers GJ, Bassetti CL, Overeem S. Hypocretin/orexin disturbances in neurological disorders. Sleep Med Rev 2009;13:9-22.
118. Borgland SL, Chang SJ, Bowers MS, et al. Orexin A/hypocretin-1 selectively promotes motivation for positive reinforcers. J Neurosci 2009;29:11215-25.
119. Aston-Jones G, Smith RJ, Moorman DE, Richardson KA. Role of lateral hypothalamic orexin neurons in reward processing and addiction. Neuropharmacology 2009;56 Suppl 1:112-21.
120. España RA, Oleson EB, Locke JL, Brookshire BR, Roberts DCS, Jones SR. The hypocretin-orexin system regulates cocaine self-administration via actions on the mesolimbic dopamine system. Eur J Neurosci 2010;31:336-48.
121. España RA, Melchior JR, Roberts DCS, Jones SR. Hypocretin 1/orexin A in the ventral tegmental area enhances dopamine responses to cocaine and promotes cocaine self-administration. Psychopharmacology 2010;1-12.
122. Moriguchi T, Sakurai T, Nambu T, Yanagisawa M, Goto K. Neurons containing orexin in the lateral hypothalamic area of the adult rat brain are activated by insulin-induced acute hypoglycemia. Neurosci Lett 1999;264:101-4.
123. Burdakov D, Gerasimenko O, Verkhratsky A. Physiological changes in glucose differentially modulate the excitability of hypothalamic melanin-concentrating hormone and orexin neurons in situ. J Neurosci 2005;25:2429-33.
124. Yamanaka A, Beuckmann CT, Willie JT, et al. Hypothalamic orexin neurons regulate arousal according to energy balance in mice. Neuron 2003;38:701-13.
125. McCormick DA, Bal T. Sleep and arousal: thalamocortical mechanisms. Annu Rev Neurosci 1997;20:185-215.
126. Huguenard JR, McCormick DA. Thalamic synchrony and dynamic regulation of global forebrain oscillations. Trends Neurosci 2007;30:350-6.
127. Steriade M, Domich L, Oakson G, Deschenes M. The deafferented reticular thalamic nucleus generates spindle rhythmicity. J Neurophysiol 1987;57:260-73.
128. Livingstone MS, Hubel DH. Effects of sleep and arousal on the processing of visual information in the cat. Nature 1981;291:554-61.
129. Hu B, Steriade M, Deschenes M. The cellular mechanism of thalamic ponto-geniculo-occipital waves. Neuroscience 1989;31:25-35.
130. Vanderwolf CH, Robinson TE. Reticulo-cortical activity and behavior: A critique of the arousal theory and a new synthesis. Behavioral Brain Science 1981;4:459-514.
131. Buzsaki G, Bickford RG, Ponomareff G, Thal LJ, Mandel R, Gage FH. Nucleus basalis and thalamic control of neocortical activity in the freely moving rat. J Neurosci 1988;8:4007-26.
132. Kinney HC, Korein J, Panigrahy A, Dikkes P, Goode R. Neuropathological findings in the brain of Karen Ann Quinlan. The role of the thalamus in the persistent vegetative state. N Engl J Med 1994;330:1469-75.
133. Fuller P, Sherman D, Pedersen NP, Saper CB, Lu J. Reassessment of the structural basis of the ascending arousal system. J Comp Neurol 2011;519:933-56.
134. Gerashchenko D, Wisor JP, Burns D, et al. Identification of a population of sleep-active cerebral cortex neurons. Proc Natl Acad Sci U S A 2008;105:10227-32.
135. Vyazovskiy VV, Cirelli C, Pfister-Genskow M, Faraguna U, Tononi G. Molecular and electrophysiological evidence for net synaptic potentiation in wake and depression in sleep. Nat Neurosci 2008;11:200-8.
136. Huber R, Ghilardi MF, Massimini M, Tononi G. Local sleep and learning. Nature 2004;430:78-81.
137. Huber R, Ghilardi MF, Massimini M, et al. Arm immobilization causes cortical plastic changes and locally decreases sleep slow wave activity. Nat Neurosci 2006;9:1169-76.
138. Pfaff D, Banavar JR. A theoretical framework for CNS arousal. Bioessays 2007;29:803-10.
139. Sherin JE, Shiromani PJ, McCarley RW, Saper CB. Activation of ventrolateral preoptic neurons during sleep. Science 1996;271:216-9.
140. Gong H, McGinty D, Guzman-Marin R, Chew KT, Stewart D, Szymusiak R. Activation of c-fos in GABAergic neurones in the preoptic area during sleep and in response to sleep deprivation. J Physiol 2004;556:935-46.
141. Szymusiak R, Alam N, Steininger TL, McGinty D. Sleep-waking discharge patterns of ventrolateral preoptic/anterior hypothalamic neurons in rats. Brain Res 1998;803:178-88.
142. Takahashi K, Lin JS, Sakai K. Characterization and mapping of sleep-waking specific neurons in the basal forebrain and preoptic hypothalamus in mice. Neuroscience 2009;161:269-92.
143. Suntsova N, Szymusiak R, Alam MN, Guzman-Marin R, McGinty D. Sleep-waking discharge patterns of median preoptic nucleus neurons in rats. J Physiol 2002;543:665-77.
144. McGinty DJ, Sterman MB. Sleep suppression after basal forebrain lesions in the cat. Science 1968;160:1253-5.
145. Lu J, Greco MA, Shiromani P, Saper CB. Effect of lesions of the ventrolateral preoptic nucleus on NREM and REM sleep. J Neurosci 2000;20:3830-42.
146. Sherin JE, Elmquist JK, Torrealba F, Saper CB. Innervation of histaminergic tuberomammillary neurons by GABAergic and galaninergic neurons in the ventrolateral preoptic nucleus of the rat. J Neurosci 1998;18:4705-21.
147. Gaus SE, Strecker RE, Tate BA, Parker RA, Saper CB. Ventrolateral preoptic nucleus contains sleep-active, galaninergic neurons in multiple mammalian species. Neuroscience 2002;115:285-94.
148. Saper CB, Cano G, Scammell TE. Homeostatic, circadian, and emotional regulation of sleep. J Comp Neurol 2005;493:92-8.
149. Saper CB, Fuller PM, Pedersen NP, Lu J, Scammell TE. Sleep state switching. Neuron 2010;68:1023-42.
150. Manns ID, Alonso A, Jones BE. Discharge profiles of juxtacellularly labeled and immunohistochemically identified GABAergic basal forebrain neurons recorded in association with the electroencephalogram in anesthetized rats. J Neurosci 2000;20:9252-63.
151. Hassani OK, Lee MG, Henny P, Jones BE. Discharge profiles of identified GABAergic in comparison to cholinergic and putative glutamatergic basal forebrain neurons across the sleep-wake cycle. J Neurosci 2009;29:11828-40.
152. Hassani OK, Henny P, Lee MG, Jones BE. GABAergic neurons intermingled with orexin and MCH neurons in the lateral hypothalamus discharge maximally during sleep. Eur J Neurosci 2010;32:448-57.
153. Gottesmann C. GABA mechanisms and sleep. Neuroscience 2002;111:231-9.
154. Sanger DJ. The pharmacology and mechanisms of action of new generation, non-benzodiazepine hypnotic agents. CNS Drugs 2004;18 Suppl 1:9-15.
155. Vienne J, Bettler B, Franken P, Tafti M. Differential effects of GABAB receptor subtypes, {gamma}-hydroxybutyric Acid, and Baclofen on EEG activity and sleep regulation. J Neurosci 2010;30:14194-204.
156. Steriade M, Datta S, Pare D, Oakson G, Curro Dossi RC. Neuronal activities in brain-stem cholinergic nuclei related to tonic activation processes in thalamocortical systems. J Neurosci 1990;10:2541-59.
157. Kayama Y, Ohta M, Jodo E. Firing of 'possibly' cholinergic neurons in the rat laterodorsal tegmental nucleus during sleep and wakefulness. Brain Res 1992;569:210-20.

158. Boissard R, Gervasoni D, Schmidt MH, Barbagli B, Fort P, Luppi PH. The rat ponto-medullary network responsible for paradoxical sleep onset and maintenance: a combined microinjection and functional neuroanatomical study. Eur J Neurosci 2002;16:1959-73.
159. Velazquez-Moctezuma J, Gillin JC, Shiromani PJ. Effect of specific M1, M2 muscarinic receptor agonists on REM sleep generation. Brain Res 1989;503:128-31.
160. Marks GA, Birabil CG. Comparison of three muscarinic agonits injected into the medial pontine reticular formation of rats to enhance REM sleep. Sleep Res Online 2001;4:17-24.
161. Baghdoyan HA. Location and quantification of muscarinic receptor subtypes in rat pons: implications for REM sleep generation. Am J Physiol 1997;273:R896-R904.
162. Imeri L, Bianchi S, Angeli P, Mancia M. Selective blockade of different brain stem muscarinic receptor subtypes: effects on the sleep-wake cycle. Brain Res 1994;636:68-72.
163. Benington JH, Heller HC. Monoaminergic and cholinergic modulation of REM-sleep timing in rats. Brain Res 1995;681:141-6.
164. Webster HH, Jones BE. Neurotoxic lesions of the dorsolateral pontomesencephalic tegmentum-cholinergic cell area in the cat. II. Effects upon sleep-waking states. Brain Res 1988;458:285-302.
165. Shouse MN, Siegel JM. Pontine regulation of REM sleep components in cats: integrity of the pedunculopontine tegmentum (PPT) is important for phasic events but unnecessary for atonia during REM sleep. Brain Res 1992;571:50-63.
166. Morales FR, Engelhardt JK, Soja PJ, Pereda AE, Chase MH. Motoneuron properties during motor inhibition produced by microinjection of carbachol into the pontine reticular formation of the decerebrate cat. J Neurophysiol 1987;57:1118-29.
167. Curtis DR, Hosli L, Johnston GA, Johnston IH. The hyperpolarization of spinal motoneurones by glycine and related amino acids. Exp Brain Res 1968;5:235-58.
168. Chase MH, Soja PJ, Morales FR. Evidence that glycine mediates the postsynaptic potentials that inhibit lumbar motoneurons during the atonia of active sleep. J Neurosci 1989;9:743-51.
169. Yamuy J, Fung SJ, Xi M, Morales FR, Chase MH. Hypoglossal motoneurons are postsynaptically inhibited during carbachol-induced rapid eye movement sleep. Neuroscience 1999;94:11-5.
170. Fedirchuk B, Dai Y. Monoamines increase the excitability of spinal neurones in the neonatal rat by hyperpolarizing the threshold for action potential production. J Physiol 2004;557:355-61.
171. Jelev A, Sood S, Liu H, Nolan P, Horner RL. Microdialysis perfusion of 5-HT into hypoglossal motor nucleus differentially modulates genioglossus activity across natural sleep-wake states in rats. J Physiol 2001;532:467-81.
172. Perrier JF, Delgado-Lezama R. Synaptic release of serotonin induced by stimulation of the raphe nucleus promotes plateau potentials in spinal motoneurons of the adult turtle. J Neurosci 2005;25:7993-9.
173. Lai YY, Kodama T, Siegel JM. Changes in monoamine release in the ventral horn and hypoglossal nucleus linked to pontine inhibition of muscle tone: an in vivo microdialysis study. J Neurosci 2001;21:7384-91.
174. Morrison JL, Sood S, Liu H, et al. Role of inhibitory amino acids in control of hypoglossal motor outflow to genioglossus muscle in naturally sleeping rats. J Physiol 2003;552:975-91.
175. Fenik V, Davies RO, Kubin L. Combined antagonism of aminergic excitatory and amino acid inhibitory receptors in the XII nucleus abolishes REM sleep-like depression of hypoglossal motoneuronal activity. Arch Ital Biol 2004;142:237-49.
176. Leonard CS, Llinas R. Serotonergic and cholinergic inhibition of mesopontine cholinergic neurons controlling REM sleep: an in vitro electrophysiological study. Neuroscience 1994;59:309-30.
177. McCarley RW, Hobson JA. Neuronal excitability modulation over the sleep cycle: a structural and mathematical model. Science 1975;189:58-60.
178. Hoque R, Chesson AL Jr. Pharmacologically induced/exacerbated restless legs syndrome, periodic limb movements of sleep, and REM behavior disorder/REM sleep without atonia: literature review, qualitative scoring, and comparative analysis. J Clin Sleep Med 2010;6:79-83.
179. Wilson S, Argyropoulos S. Antidepressants and sleep: a qualitative review of the literature. Drugs 2005;65:927-47.
180. Sakai K, Sastre JP, Salvert D, Touret M, Tohyama M, Jouvet M. Tegmentoreticular projections with special reference to the muscular atonia during paradoxical sleep in the cat: an HRP study. Brain Res 1979;176:233-54.
181. Verret L, Leger L, Fort P, Luppi PH. Cholinergic and noncholinergic brainstem neurons expressing Fos after paradoxical (REM) sleep deprivation and recovery. Eur J Neurosci 2005;21:2488-504.
182. Lu J, Sherman D, Devor M, Saper CB. A putative flip-flop switch for control of REM sleep. Nature 2006;441:589-94.
183. Xi MC, Morales FR, Chase MH. Interactions between GABAergic and cholinergic processes in the nucleus pontis oralis: neuronal mechanisms controlling active (rapid eye movement) sleep and wakefulness. J Neurosci 2004;24:10670-8.
184. Sastre JP, Jouvet M. Oneiric behavior in cats. Physiol Behav 1979;22:979-89.
185. Hendricks JC, Morrison AR, Mann GL. Different behaviors during paradoxical sleep without atonia depend on pontine lesion site. Brain Res 1982;239:81-105.
186. Boeve BF, Silber MH, Saper CB, et al. Pathophysiology of REM sleep behaviour disorder and relevance to neurodegenerative disease. Brain 2007;130:2770-88.
187. Boissard R, Fort P, Gervasoni D, Barbagli B, Luppi PH. Localization of the GABAergic and non-GABAergic neurons projecting to the sublaterodorsal nucleus and potentially gating paradoxical sleep onset. Eur J Neurosci 2003;18:1627-39.
188. Sastre JP, Buda C, Kitahama K, Jouvet M. Importance of the ventrolateral region of the periaqueductal gray and adjacent tegmentum in the control of paradoxical sleep as studied by muscimol microinjections in the cat. Neuroscience 1996;74:415-26.
189. Alam MN, Gong H, Alam T, Jaganath R, McGinty D, Szymusiak R. Sleep-waking discharge patterns of neurons recorded in the rat perifornical lateral hypothalamic area. J Physiol 2002;538:619-31.
190. Koyama Y, Takahashi K, Kodama T, Kayama Y. State-dependent activity of neurons in the perifornical hypothalamic area during sleep and waking. Neuroscience 2003;119:1209-19.
191. Verret L, Goutagny R, Fort P, et al. A role of melanin-concentrating hormone producing neurons in the central regulation of paradoxical sleep. BMC Neurosci 2003;4:19.
192. Bittencourt JC, Presse F, Arias C, et al. The melanin-concentrating hormone system of the rat brain: an immuno- and hybridization histochemical characterization. J Comp Neurol 1992;319:218-45.
193. Kilduff TS, de Lecea L. Mapping of the mRNAs for the hypocretin/orexin and melanin- concentrating hormone receptors: Networks of overlapping peptide systems. J Comp Neurol 2001;435:1-5.
194. Ahnaou A, Drinkenburg WH, Bouwknecht JA, Alcazar J, Steckler T, Dautzenberg FM. Blocking melanin-concentrating hormone MCH1 receptor affects rat sleep-wake architecture. Eur J Pharmacol 2008;579:177-88.
195. Willie JT, Sinton CM, Maratos-Flier E, Yanagisawa M. Abnormal response of melanin-concentrating hormone deficient mice to fasting: hyperactivity and rapid eye movement sleep suppression. Neuroscience 2008;156:819-29.
196. Chou TC, Bjorkum AA, Gaus SE, Lu J, Scammell TE, Saper CB. Afferents to the ventrolateral preoptic nucleus. J Neurosci 2002;22:977-90.
197. Gallopin T, Fort P, Eggermann E, et al. Identification of sleep-promoting neurons in vitro. Nature 2000;404:992-5.
198. Brown RE, Sergeeva O, Eriksson KS, Haas HL. Orexin A excites serotonergic neurons in the dorsal raphe nucleus of the rat. Neuropharmacology 2001;40:457-9.
199. Eggermann E, Serafin M, Bayer L, et al. Orexins/hypocretins excite basal forebrain cholinergic neurones. Neuroscience 2001;108:177-81.
200. Marcus JN, Aschkenasi CJ, Lee CE, et al. Differential expression of orexin receptors 1 and 2 in the rat brain. J Comp Neurol 2001;435:6-25.
201. Arrigoni E, Saper CB, Lu J. Modulation of the spinal cord projecting neurons of the sublaterodorsal nucleus. Society for Neuroscience Meeting, Chicago, IL, 2009.
202. Broughton R, Valley V, Aguirre M, Roberts J, Suwalski W, Dunham W. Excessive daytime sleepiness and the pathophysiology of narcolepsy-cataplexy: a laboratory perspective. Sleep 1986;9:205-15.
203. Ishimori K. True cause of sleep: A hypnogenic substance as evidenced in the brain of sleep-deprived animals. Tokyo Igakkai Zasshi 1909;23:429-57.
204. Legendre R, Pieron H. Recherches sur le besoin de sommeil consecutif à une veille prolongée. Z Allgem Physiol 1913;14:235-62.
205. Obal F, Jr., Krueger JM. Biochemical regulation of non-rapid-eye-movement sleep. Front Biosci 2003;8:d520-d550.
206. Radulovacki M, Virus RM, Djuricic-Nedelson M, Green RD. Adenosine analogs and sleep in rats. J Pharmacol Exp Ther 1984;228:268-74.

207. Benington JH, Kodali SK, Heller HC. Stimulation of A1 adenosine receptors mimics the electroencephalographic effects of sleep deprivation. Brain Res 1995;692:79-85.
208. Basheer R, Strecker RE, Thakkar MM, McCarley RW. Adenosine and sleep-wake regulation. Prog Neurobiol 2004;73:379-96.
209. Porkka-Heiskanen T, Strecker RE, Thakkar M, Bjorkum AA, Greene RW, McCarley RW. Adenosine: a mediator of the sleep-inducing effects of prolonged wakefulness. Science 1997;276:1265-8.
210. Halassa MM, Florian C, Fellin T, et al. Astrocytic modulation of sleep homeostasis and cognitive consequences of sleep loss. Neuron 2009;61:213-9.
211. Huang ZL, Urade Y, Hayaishi O. Prostaglandins and adenosine in the regulation of sleep and wakefulness. Curr Opin Pharmacol 2007;7:33-8.
212. Oishi Y, Huang ZL, Fredholm BB, Urade Y, Hayaishi O. Adenosine in the tuberomammillary nucleus inhibits the histaminergic system via A1 receptors and promotes non-rapid eye movement sleep. Proc Natl Acad Sci U S A 2008;105:19992-7.
213. Scammell TE, Gerashchenko DY, Mochizuki T, et al. An adenosine A2a agonist increases sleep and induces Fos in ventrolateral preoptic neurons. Neuroscience 2001;107:653-63.
214. Penetar D, McCann U, Thorne D, et al. Caffeine reversal of sleep deprivation effects on alertness and mood. Psychopharmacology (Berl) 1993;112:359-65.
215. Imeri L, Opp MR. How (and why) the immune system makes us sleep. Nat Rev Neurosci 2009;10:199-210.
216. Alam MN, McGinty D, Bashir T, et al. Interleukin-1beta modulates state-dependent discharge activity of preoptic area and basal forebrain neurons: role in sleep regulation. Eur J Neurosci 2004;20:207-16.
217. Kubota T, Li N, Guan Z, Brown RA, Krueger JM. Intrapreoptic microinjection of TNF-alpha enhances non-REM sleep in rats. Brain Res 2002;932:37-44.
218. Krueger JM, Majde JA. Microbial products and cytokines in sleep and fever regulation. Crit Rev Immunol 1994;14:355-79.
219. Bredow S, Guha-Thakurta N, Taishi P, Obal F, Jr., Krueger JM. Diurnal variations of tumor necrosis factor alpha mRNA and alpha-tubulin mRNA in rat brain. Neuroimmunomodulation 1997;4:84-90.
220. Floyd RA, Krueger JM. Diurnal variation of TNF alpha in the rat brain. Neuroreport 1997;8:915-8.
221. Taishi P, Bredow S, Guha-Thakurta N, Obal F, Jr., Krueger JM. Diurnal variations of interleukin-1 beta mRNA and beta-actin mRNA in rat brain. J Neuroimmunol 1997;75:69-74.
222. Mizoguchi A, Eguchi N, Kimura K, et al. Dominant localization of prostaglandin D receptors on arachnoid trabecular cells in mouse basal forebrain and their involvement in the regulation of non-rapid eye movement sleep. Proc Natl Acad Sci U S A 2001;98:11674-9.
223. Pandey HP, Ram A, Matsumura H, Satoh S, Hayaishi O. Circadian variations of prostaglandins D2, E2, and F2 alpha in the cerebrospinal fluid of anesthetized rats. Biochem Biophys Res Commun 1995;213:625-9.
224. Ram A, Pandey HP, Matsumura H, et al. CSF levels of prostaglandins, especially the level of prostaglandin D2, are correlated with increasing propensity towards sleep in rats. Brain Res 1997;751:81-9.
225. Scammell T, Gerashchenko D, Urade Y, Onoe H, Saper C, Hayaishi O. Activation of ventrolateral preoptic neurons by the somnogen prostaglandin D2. Proc Natl Acad Sci U S A 1998;95:7754-9.
226. Ueno R, Honda K, Inoue S, Hayaishi O. Prostaglandin D2, a cerebral sleep-inducing substance in rats. Proc Natl Acad Sci U S A 1983;80:1735-7.
227. Inoue S, Honda K, Komoda Y, Uchizono K, Ueno R, Hayaishi O. Differential sleep-promoting effects of five sleep substances nocturnally infused in unrestrained rats. Proc Natl Acad Sci U S A 1984;81:6240-4.
228. Onoe H, Ueno R, Fujita I, Nishino H, Oomura Y, Hayaishi O. Prostaglandin D2, a cerebral sleep-inducing substance in monkeys. Proc Natl Acad Sci U S A 1988;85:4082-6.
229. Kantha SS. Histamine-interleukin-prostaglandin pathway: a hypothesis for a biochemical cycle regulating sleep and wakefulness. Med Hypotheses 1994;42:335-9.
230. Pentreath VW, Rees K, Owolabi OA, Philip KA, Doua F. The somnogenic T lymphocyte suppressor prostaglandin D2 is selectively elevated in cerebrospinal fluid of advanced sleeping sickness patients. Trans R Soc Trop Med Hyg 1990;84:795-9.
231. Borbely AA, Achermann P. Sleep homeostasis and models of sleep regulation. J Biol Rhythms 1999;14:557-68.
232. Achermann P. The two-process model of sleep regulation revisited. Aviat Space Environ Med 2004;75:A37-A43.
233. Dijk DJ, Czeisler CA. Contribution of the circadian pacemaker and the sleep homeostat to sleep propensity, sleep structure, electroencephalographic slow waves, and sleep spindle activity in humans. J Neurosci 1995;15:3526-38.
234. Dijk DJ, Brunner DP, Borbely AA. Time course of EEG power density during long sleep in humans. Am J Physiol 1990;258:R650-R661.
235. Franken P, Dijk DJ, Tobler I, Borbely AA. Sleep deprivation in rats: effects on EEG power spectra, vigilance states, and cortical temperature. Am J Physiol 1991;261:R198-R208.
236. Dibner C, Schibler U, Albrecht U. The mammalian circadian timing system: organization and coordination of central and peripheral clocks. Annu Rev Physiol 2010;72:517-49.
237. Welsh DK, Takahashi JS, Kay SA. Suprachiasmatic nucleus: cell autonomy and network properties. Annu Rev Physiol 2010;72:551-77.
238. Ukai H, Ueda HR. Systems biology of mammalian circadian clocks. Annu Rev Physiol 2010;72:579-603.
239. Hattar S, Liao HW, Takao M, Berson DM, Yau KW. Melanopsin-containing retinal ganglion cells: architecture, projections, and intrinsic photosensitivity. Science 2002;295:1065-70.
240. Cheng MY, Bullock CM, Li C, et al. Prokineticin 2 transmits the behavioural circadian rhythm of the suprachiasmatic nucleus. Nature 2002;417:405-10.
241. Kramer A, Yang FC, Snodgrass P, et al. Regulation of daily locomotor activity and sleep by hypothalamic EGF receptor signaling. Science 2001;294:2511-5.
242. Chou TC, Scammell TE, Gooley JJ, Gaus SE, Saper CB, Lu J. Critical role of dorsomedial hypothalamic nucleus in a wide range of behavioral circadian rhythms. J Neurosci 2003;23:10691-702.
243. Buhr ED, Yoo SH, Takahashi JS. Temperature as a universal resetting cue for mammalian circadian oscillators. Science 2010;330:379-85.
244. Bass J, Takahashi JS. Circadian integration of metabolism and energetics. Science 2010;330:1349-54.