CHAPTER 3

# Glial Modulation of Sleep and Electroencephalographic Rhythms

**Marcos G. Frank**
Department of Neuroscience, University of Pennsylvania School of Medicine

## INTRODUCTION

The three principal classes of glia in the mature brain are astrocytes, microglia, and oligodendrocytes. Astrocytes perform a number of "housekeeping" functions in the brain, including buffering ions, recycling neurotransmitter, and regulating metabolism. Microglia are the immune cells of the central nervous system and play critical roles in the response to neural injury and cellular stress. Oligodendrocytes produce myelin and enwrap axons and dysfunction in these cells leads to demyelination and disease. They are considered nonexcitable, as they do not produce action potentials and respond linearly to current injections. For these reasons, they have historically been viewed as supportive brain cells with no special or direct roles in brain activity or behavior. However, at least two classes of glial cells (astrocytes and microglia) are known to secrete substances that can alter the activity of surrounding neurons. This provides a means of influencing behavior and brain activity. In astrocytes this process is known as gliotransmission and is now recognized as an important means by which glial and neurons exchange chemical signals (Fiacco, Agulhon, & McCarthy, 2009; Halassa & Haydon, 2010; Hamilton & Atwell, 2010). Therefore, in conjunction with neuronal circuits and neuromodulatory nuclei, glia likely play important roles in brain activity patterns during sleep.

## EARLY CONCEPTS OF GLIAL ROLES IN SLEEP

In 1895, Ramon y Cajal hypothesized that astrocytes might control the expression of sleep and wakefulness (García-Marín, García-López, & Freire, 2007). He hypothesized that by either contracting or expanding their processes between synapses, astrocytes could modify the flow

*Sleep and Brain Activity*
DOI: http://dx.doi.org/10.1016/B978-0-12-384995-3.00003-4

of "nervous current" between neurons. The withdrawal of this physical barrier would promote wakefulness, while its expansion would promote sleep. Early empirical support for a role for glia in sleep came from a study of metabolic enzymes in the brainstem of sleeping rabbits (Hyden & Lange, 1965). Hyden and Lange found sleep-wake rhythms in succinooxidase (a key enzymatic step in the Krebs cycle) activity in neurons and glia (type not specified), with glia showing reduced activity and neurons heightened activity during sleep (Hyden & Lange, 1965). The authors suggested that "neuron and glia form a functional unit" and that sleep and wakefulness resulted in an exchange of signaling molecules between these cell types.

## GLIA, NEURAL METABOLISM, AND SLEEP

A more modern twist on the ideas of Hyden and Lange was proposed in 1995 by Joel Benington and Craig Heller. According to the Benington-Heller hypothesis, astrocytic glycogen—which acts as a reserve glucose store for neurons—is depleted during wakefulness and restored during NREM sleep. The depletion of glycogen is mediated by the heightened release of excitatory neurotransmitters during wake, which through enzymatic mechanisms convert astrocytic glycogen into glucose. The utilization of this glucose by neurons (and the subsequent hydrolysis of ATP to AMP) leads to an increase in neuronal adenosine production—which diffuses across the cell membrane and by acting on A1 receptors reduces neuronal excitability. The restoration of glycogen is then favored by states with reduced release of excitatory neurotransmitters, such as NREM sleep (Benington & Heller, 1995).

The Benington-Heller theory was very appealing because it connected the regulation of sleep to a function with clear adaptive value (cerebral metabolism) and elegantly incorporated previous work demonstrating a central role for adenosine in sleep regulation. The evidence for this theory, however, is equivocal. Sleep deprivation increases the activity of glycogen synthase (Petit, Tobler, Allaman, Borbely, & Magistretti, 2002), and one study showed decreases in brain glycogen content following sleep deprivation and increases following recovery sleep (Kong et al., 2002). These latter findings, however, have not been replicated by other labs (Franken, Gip, Hagiwara, Ruby, & Heller, 2003; Gip, Hagiwara, Ruby, & Heller, 2002; Gip et al., 2004; Zimmerman et al., 2004). Therefore, while it remains possible that astrocyte-neuronal metabolic interactions influence sleep (Magistretti, 2006; Scharf, Naidoo, Zimmerman, & Pack, 2008), the

specific role posited for astrocytes in the Benington-Heller hypothesis is not supported by current data.

## ION BUFFERING AND EEG ACTIVITY

Astrocytes through passive and active transport buffer ions in the extracellular space. This buffering may facilitate synchronized neuronal activity necessary for slow, cortical oscillations (slow-wave activity [SWA]) typical of NREM sleep (Crunelli et al., 2002). For example, astrocytes in thalamic slices (*in situ*) exhibit spontaneous intracellular calcium oscillations that fall within the slower EEG bands of slow-wave activity (<0.1 Hz). These oscillations can propagate within the slice and elicit NMDA currents in neighboring neurons. This particular coupling between neurons and astrocytes does not appear to be related to the EEG rhythms of sleep, as it predominates at ages when thalamocortical and intracortical EEG activity typical of adult sleep is absent (Crunelli & Hughes, 2010). More compelling evidence that astrocytes contribute to slow, EEG rhythms of sleep come from studies using dual intracellular recording in cortical astrocytes and neurons in the adult cat (Amzica, 2002; Amzica & Massimini, 2002; Amzica & Neckelmann, 1999). These investigators showed that astrocyte membrane polarization and capacitance oscillate in phase with slow EEG waves during natural NREM sleep, suggesting that the cation buffering by these cells is a critical component of neuronal "up" and "down" states.

## GLIAL SECRETION OF SOMNOGENIC SUBSTANCES: ASTROCYTES

Cultured astrocytes secrete and/or exocytose a variety of molecules that when injected either systemically or into the brain can increase sleep time or NREM SWA. For example, the cytokine IL-1 derived from cultured mouse astrocytes increases NREM sleep in rats when administered into the ventricles. (Tobler, Borbély, Schwyzer, & Fontana, 1984). Cultured astrocytes also secrete neurotrophins (e.g., brain-derived neurotrophin factor [BDNF]), prostaglandins (PGD2) and the cytokine TNFα that increase sleep time or intensity (e.g.,. NREM SWA) when injected intraventricularly and/or infused and applied to the neocortex (Faraguna, Vyazovskiy, Nelson, Tononi, & Cirelli, 2008; Hayaishi, 2002; Huang, Urade, & Hayaishi, 2007; Krueger, 2008; Kushikata, Fang, & Krueger, 1999). Astrocytes release some of these substances in response to neuronal signals, including ATP acting at

astrocytic PP2 receptors (Krueger, 2008). These findings support a mechanism by which neuronal activation of glia via purinergic receptors during wake leads to the release of substances that can increase sleep amounts and indices of sleep intensity (SWA) (Krueger, 2008).

Whether or not such mechanisms actually exist *in vivo* is unclear. Cultured cells can show very different properties than cells *in vivo*, or in brain slice preparations (Inagaki & Wada, 1994; Yamamoto, Miwa, Ueno, & Hayaishi, 1988). In addition, although mutant mice lacking the TNFα, IL-1, and PP2 receptors have sleep phenotypes consistent with this general hypothesis (i.e., reduced NREM sleep amounts and intensity), it is not known if this reflects neuronal or astrocytic influences, as neurons release and respond to many cytokines as well (Baracchi & Opp, 2008; Kapas et al., 2008; Krueger et al., 2010b). A recent study showed increased IL-β1 immunoreactivity in astrocytes following 2 hours of whisker stimulation in awake rodents (Hallett, Churchill, Taishi, De, & Krueger, 2010). While this latter finding is quite intriguing, it is not known if similar stimulation also produces IL-β1 secretion in astrocytes and subsequent sleep (Hallett et al., 2010). Therefore, determining the precise role of astrocyte cytokine signaling in sleep must await more selective manipulations of these signaling pathways *in vivo.*

In addition to neurotrophins, cytokines, and prostaglandins, there is evidence that astrocytes exocytose additional chemical transmitters that modulate neuronal excitability, including ATP, which is hydrolyzed to adenosine in the extracellular space (Halassa, Fellin, & Haydon, 2009; Pascual et al., 2005). Although the precise mechanisms of gliotransmission are unclear (Fiacco et al., 2009; Hamilton & Atwell, 2010), for some gliotransmitters, exocytosis may depend on the formation of a SNARE complex between vesicles and the target membrane (Scales, Bock, & Scheller, 2000). Conditional astrocyte-selective expression of the SNARE domain of the protein synaptobrevin II prevents both tonic and activity-dependent extracellular accumulation of adenosine that acts on A1 receptors *in situ* (Pascual et al., 2005). Fortunately, the role of this signaling pathway in sleep can be investigated *in vivo* using transgenic approaches in mice (Pascual et al., 2005).

We investigated this possibility (Halassa, Florian et al., 2009) using the tet-off system (Morozov, Kellendonk, Simpson, & Tronche, 2003) to allow conditional expression of a dnSNARE transgene selectively in astrocytes (Pascual et al., 2005). Astrocyte specificity of transgene expression is achieved by using the astrocyte-specific Glial Fibrillary Acidic Protein (GFAP) promoter to drive the expression of tetracycline transactivator (tTA) only in this subset of glia. GFAP.tTA mice were crossed

with tetO.dnSNARE mice. The tet-operator (tet.O) drives the expression of dnSNARE and the EGFP reporter. Thus, in bigenic offspring of this mating transgenes are only expressed in GFAP-positive astrocytes (Pascual et al., 2005). Conditional suppression of transgene expression is achieved by including doxycycline (Dox) in the diet. Dox binds to tTA preventing it from activating the tet.O promoter, and by maintaining all bigenic mice on DOX throughout gestation and early development, the transgene was only expressed, when desired, in adult mice (Halassa, Florian, et al., 2009).

Suppressing gliotransmission *in vivo* had surprisingly little effect on baseline sleep-wake architecture, except for a reduction in the normal accumulation of NREM EEG slow-wave activity—a classic index of sleep pressure in mammals (Halassa, Florian, et al., 2009). This suggested that in the absence of gliotransmission, the accumulation of sleep need was reduced. This was confirmed by examining compensatory responses to sleep deprivation in dnSNARE mutant mice and wild-type controls. While wild-type mice showed normal compensatory increases in NREM sleep time, bout duration, and SWA after 6 hours of sleep deprivation, these changes did not occur (or were greatly attenuated) in the mutant mice. Interestingly, the cognitive effects of sleep deprivation were also absent in the mutant mice. Subsequent investigations showed that this phenotype could be copied in wild-type mice by antagonists to the A1 (but not A2) receptor, demonstrating that the gliotransmitter of interest was ATP (Halassa, Florian, et al., 2009). Manipulations of adenosine receptors in wild-type mice duplicated most of the effects of the dnSNARE mutation; however, subsequent investigations showed that gliotransmission of D-serine is also necessary for SWA (Fellin et al., 2009). These findings represent the first direct demonstration that astrocytes *in vivo* influence mammalian sleep.

Further studies *in situ* and *in vivo* in the dnSNARE mouse demonstrated that gliotransmission also modulates sleep EEG rhythms (Fellin et al., 2009). Mutant mice normally show reduced levels of NREM SWA under baseline conditions, and after sleep deprivation—which suggests impairment intracortically in the ability to generate slow, neuronal oscillations (Fellin et al., 2009; Halassa, Florian, et al., 2009). This was confirmed using a combination of extracellular local field potential (LFP) and patch-clamp recordings *in vivo* from the somatosensory cortex of urethane anesthetized dnSNARE animals and wild-type littermates. Attenuation of gliotransmission in transgenic animals significantly decreased the power of slow oscillations (<1 Hz), pyramidal neurons from dnSNARE animals have a significantly lower probability of being at the depolarized

**Table 3.1** Glial Substances Linked to Sleep

| Substance | Effects on Sleep | Evidence That Glial Secretion *in vivo* Regulates Sleep? |
|---|---|---|
| ATP>> adenosine | Mediates NREM sleep homeostasis (EEG slow-wave activity (SWA) and state architecture) (Halassa, Florian et al., 2009) | Yes |
| D-serine | Necessary for normal SWA (Fellin et al., 2009) | Yes |
| TNFα | Intracranial infusions increase sleep time and SWA (Krueger, 2008) | No |
| IL-1 | Intracranial infusions increase sleep time and SWA (Tobler et al., 1984) | No |
| PDG2 | Intracranial infusions increase sleep time and SWA (Huang et al., 2007) | No |
| BDNF | Intracranial infusions increases sleep time and SWA (Faraguna et al., 2008; Kushikata et al., 1999) | No |

"Intracranial" infusions include intraventricular injections, topical application to the cortex, and intracortical infusions.

potential (up-state probability) compared to controls, and up-state transitions occurred at lower frequency in the mutants (Fellin et al., 2009). Subsequent studies *in situ* and *in vivo* demonstrated that the attenuation of slow, EEG rhythms in sleep was due to the absence of two gliotransmitters: D-serine and ATP (hydrolyzed to adenosine) (Fellin et al., 2009).

## GLIAL SECRETION OF SOMNOGENIC SUBSTANCES: MICROGLIA AND OLIGODENDROCYTES

Cerebral microglia and oligodendrocytes cells secrete a number of substances *in vitro* known to influence sleep or brain activity in sleep (e.g., cytokines, prostaglandins, and nitric oxide, see Table 3.1) (Matsui et al., 2010). Because sleep deprivation is associated with an increase in markers of cellular stress, it has been proposed that substances secreted by microglia may play a central role in sleep regulation (Wisor, Clegern, & Schmidt, 2011; Wisor, Schmidt, & Clegern, 2011). For example, attenuation of microglia reactivity with minocycline reduces the normal compensatory increases in NREM SWA in mice following sleep deprivation (Wisor & Clegern, 2011). Interestingly, microglia contain membrane-bound purinergic receptors, providing a means of interaction with astrocyte-derived ATP and adenosine

(Gyoneva, Orr, & Traynelis, 2009; Haynes et al., 2006). On the other hand, a putative transducer of microglial-mediated effects (the toll-like receptor 4 (TLR4)) does not appear to play a central role in sleep brain activity. Constitutive deletion of TLR4 minimally impacts NREM EEG SWA under baseline conditions or after sleep deprivation) (Wisor, Clegern, et al., 2011). Oligodendrocytes are a source of prostaglandin D2 in the mature brain, which has been shown to be a potent sleep-inducing substance when applied exogenously (Urade & Hayaishi, 2011). In addition to increasing behavioral indices of sleep, stimulation of D2 receptors increases NREM SWA in a physiological manner(Urade & Hayaishi, 2011). However, in contrast to astrocytes, much less is known about exocytosis and secretion in microglia and oligodendrocytes. It is also unknown what signals normally trigger the release of these substances across the sleep-wake cycle.

## UNANSWERED QUESTIONS AND FUTURE DIRECTIONS

An important future area of investigation is to determine where glial cells exert their effects on sleep and/or brain activity. Glia are dispersed widely in subcortical and cortical brain areas (Zhang & Haydon, 2005) including regions known to trigger sleep and wakefulness (Halassa, Florian, et al., 2009). Therefore, they may regulate sleep and brain activity by acting within specific neocortical circuits or via modulation of basal forebrain and hypothalamic sleep and arousal centers (Benington & Heller, 1995; Krueger et al., 2008; Strecker et al., 2000; Szymusiak, Gvilia, & McGinty, 2007). It is presently unknown which of these two models is valid. Addressing this issue is complicated by several factors. First, it is not yet clear if glia in different brain regions secrete the same or different sets of somnogenic substances. As discussed above, cultured cells appear to secrete a variety of neuroactive molecules, but evidence for similar secretion *in vivo* is very sparse, and regional patterns of secretion within the intact brain are poorly understood. Second, cultured glia express numerous neurotransmitter receptors (including glutamate NMDA and metabotropic, neurotrophin (TRKb), and purinergic receptors), raising the possibility that substances secreted by astrocytes also feedback onto their sites of release. These potential feedback loops add yet another layer of complexity to any proposed model of neuronal-glial interactions (Fiacco et al., 2009; Halassa & Haydon, 2010; Hamilton & Atwell, 2010).

A related important question to address is the relative role of different glial-secreted substances in the sleeping brain. Astrocytic adenosine (acting

at A1 receptors) is likely to be a key mediator of sleep behavior and brain activity. This is because (a) gliotransmission of ATP provides a large amount of extracellular adenosine in the brain (Halassa, Florian, et al., 2009; Pascual et al., 2005), (b) activation of gliotransmission in cultured astrocytes increases adenosine release (Figueiredo et al., 2010), (c) similar activation *in vivo* leads to a suppression of surrounding neuronal activity consistent with activation of A1 receptors (Gradinaru, Mogri, Thompson, Henderson, & Deisseroth, 2009), and (d) adenosine is widely recognized as an endogenous sleep-inducing substance (Bjorness & Greene, 2009; Strecker et al., 2000). However, other signaling pathways and secreted substances may play complementary roles (Frank, 2011). For example, glial-derived adenosine might activate A2 receptors in sleep-promoting hypothalamic neurons (Szymusiak & McGinty, 2008) and glial-derived glutamate might excite sleep-promoting neurons in the forebrain and brainstem (Kaushik, Kumar, & Mallick, 2010; Luppi et al., 2007). This may act in concert with A1-mediated inhibition of wake-promoting neurons to increase sleep drive. There may also be important roles for purinergic receptors activated directly by ATP (Krueger et al., 2010a) and other substances secreted by glia (e.g., cytokines (Krueger, 2008; Opp, 2005)).

## REFERENCES

Amzica, F. (2002). In vivo electrophysiological evidences for cortical neuron-glia interactions during slow (<1 Hz) and paroxysmal sleep oscillations. *Journal of Physiology-Paris*, *96*(3–4), 209–219.

Amzica, F., & Massimini, M. (2002). Glial and neuronal interactions during slow wave and paroxysmal activities in the neocortex. *Cerebral Cortex*, *12*(10), 1101–1113.

Amzica, F., & Neckelmann, D. (1999). Membrane capacitance of cortical neurons and glia during sleep oscillations and spike-wave seizures. *Journal of Neurophysiology*, *82*(5), 2731–2746.

Baracchi, F., & Opp, M. R. (2008). Sleep-wake behavior and responses to sleep deprivation of mice lacking both interleukin-1[beta] receptor 1 and tumor necrosis factor-[alpha] receptor 1. *Brain, Behavior, and Immunity*, *22*(6), 982–993. doi:10.1016/j.bbi.2008.02.001.

Benington, J., & Heller, H. C. (1995). Restoration of brain energy metabolism as the function of sleep. *Progress in Neurobiology*, *45*(4), 347–360.

Bjorness, T. E., & Greene, R. W. (2009). Adenosine and sleep. *Current Neuropharmacology*, *7*(3), 238–245.

Crunelli, V., Blethyn, K. L., Cope, D. W., Hughes, S. W., Parri, H. R., Turner, J. P., et al. (2002). Novel neuronal and astrocytic mechanisms in thalamocortical loop dynamics. *Philosophical Transactions of the Royal Society of London Series B, Biological Sciences*, *357*(1428), 1675–1693.

Crunelli, V., & Hughes, S. W. (2010). The slow (<1 Hz) rhythm of non-REM sleep: A dialogue between three cardinal oscillators. *Nature Neuroscience*, *13*(1), 9–17.

Faraguna, U., Vyazovskiy, V. V., Nelson, A. B., Tononi, G., & Cirelli, C. (2008). A causal role for brain-derived neurotrophic factor in the homeostatic regulation of sleep. *The Journal of Neuroscience*, *28*(15), 4088–4095.

Fellin, T., Halassa, M. M., Terunuma, M., Succol, F., Takano, H., Frank, M., et al. (2009). Endogenous nonneuronal modulators of synaptic transmission control cortical slow

oscillations in vivo. *Proceedings of the National Academy of Sciences of the United States of America*, *106*(35), 15037–15042.
Fiacco, T. A., Agulhon, C., & McCarthy, K. D. (2009). Sorting out astrocyte physiology from pharmacology. *Annual Review of Pharmacology and Toxicology*, *49*, 151–174.
Figueiredo, M., Lane, S., Tang, F., Liu, B. H., Hewinson, J., Marina, N., et al. (2010). Optogenetic experimentation on astrocytes. *Experimental Physiology*, *96*(1), 40–50.
Frank, M. G. (2011). Beyond the neuron: Astroglial regulation of mammalian sleep. *Current Topics in Medicinal Chemistry*, *11*(19), 2452–2456.
Franken, P., Gip, P., Hagiwara, G., Ruby, N. F., & Heller, H. C. (2003). Changes in brain glycogen after sleep deprivation vary with genotype. *American Journal of Physiology Regulatory, Integrative and Comparative Physiology*, *285*(2), R413–419.
García-Marín, V., García-López, P., & Freire, M. (2007). Cajal's contributions to glia research. *Trends in Neurosciences*, *30*(9), 479–487.
Gip, P., Hagiwara, G., Ruby, N. F., & Heller, H. C. (2002). Sleep deprivation decreases glycogen in the cerebellum but not in the cortex of young rats. *American Journal of Physiology Regulatory, Integrative and Comparative Physiology*, *283*(1), R54–59.
Gip, P., Hagiwara, G., Sapolsky, R. M., Cao, V. H., Heller, H. C., & Ruby, N. F. (2004). Glucocorticoids influence brain glycogen levels during sleep deprivation. *American Journal of Physiology Regulatory, Integrative and Comparative Physiology*, *286*(6), R1057–1062.
Gradinaru, V., Mogri, M., Thompson, K. R., Henderson, J. M., & Deisseroth, K. (2009). Optical deconstruction of parkinsonian neural circuitry. *Science*, *324*(5925), 354–359.
Gyoneva, S., Orr, A. G., & Traynelis, S. F. (2009). Differential regulation of microglial motility by ATP/ADP and adenosine. *Parkinsonism & Related Disorders*, *15*(*Supplement 3*), S195–S199.
Halassa, M. M., Fellin, T., & Haydon, P. G. (2009). Tripartite synapses: Roles for astrocytic purines in the control of synaptic physiology and behavior. *Neuropharmacology*, *57*(4), 343–346.
Halassa, M. M., Florian, C., Fellin, T., Munoz, J. R., Lee, S. Y., Abel, T., et al. (2009). Astrocytic modulation of sleep homeostasis and cognitive consequences of sleep loss. *Neuron*, *61*(2), 213–219.
Halassa, M. M., & Haydon, P. G. (2010). Integrated brain circuits: Astrocytic networks modulate neuronal activity and behavior. *Annual Review of Physiology*, *72*, 335–355.
Hallett, H., Churchill, L., Taishi, P., De, A., & Krueger, J. M. (2010). Whisker stimulation increases expression of nerve growth factor- and interleukin-1[beta]-immunoreactivity in the rat somatosensory cortex. *Brain Research* (*In Press, Accepted Manuscript*).
Hamilton, N. B., & Atwell, D. (2010). Do astrocytes really exocytose neurotransmitters? *Nature Reviews Neuroscience*, *11*, 227–238.
Hayaishi, O. (2002). Functional genomics of sleep and circadian rhythm: Invited review: Molecular genetic studies on sleep-wake regulation, with special emphasis on the prostaglandin D2 system. *Journal of Applied Physiology*, *92*(2), 863–868.
Haynes, S. E., Hollopeter, G., Yang, G., Kurpius, D., Dailey, M. E., Gan, W. B., et al. (2006). The P2Y12 receptor regulates microglial activation by extracellular nucleotides. *Nature Neuroscience*, *9*(12), 1512–1519.
Huang, Z.-L., Urade, Y., & Hayaishi, O. (2007). Prostaglandins and adenosine in the regulation of sleep and wakefulness. *Current Opinion in Pharmacology*, 7(1), 33–38.
Hyden, H., & Lange, P. W. (1965). Rhythmic enzyme changes in neurons and glia during sleep. *Science*, *149*(3684), 654–656.
Inagaki, N., & Wada, H. (1994). Histamine and prostanoid receptors on glial cells. *Glia*, *11*, 102–109.
Kapas, L., Bohnet, S. G., Traynor, T. R., Majde, J. A., Szentirmai, E., Magrath, P., et al. (2008). Spontaneous and influenza virus-induced sleep are altered in TNF-α double-receptor deficient mice. *Journal of Applied Physiology*, *105*(4), 1187–1198.
Kaushik, M. K., Kumar, V. M., & Mallick, H. N. (2010). Glutamate microinjection at the medial preoptic area enhances slow wave sleep in rats. *Behavioural Brain Research*, *217*(1), 240–243.

Kong, J., Shepel, P. N., Holden, C. P., Mackiewicz, M., Pack, A. I., & Geiger, J. D. (2002). Brain glycogen decreases with increased periods of wakefulness: Implications for homeostatic drive to sleep. *The Journal of Neuroscience*, *22*(13), 5581–5587.

Krueger, J. M. (2008). The role of cytokines in sleep regulation. *Current Pharmaceutical Design*, *14*(32), 3408–3416.

Krueger, J. M., Rector, D. M., Roy, S., Van Dongen, H. P., Belenky, G., & Panksepp, J. (2008). Sleep as a fundamental property of neuronal assemblies. *Nature Reviews Neuroscience*, *9*(12), 910–919.

Krueger, J. M., Taishi, P., De, A., Davis, C. J., Winters, B. D., Clinton, J., et al. (2010a). ATP and the purine type 2 X 7 receptor affect sleep. *Journal of Applied Physiology*, *109*(5), 1318–1327.

Krueger, J. M., Taishi, P., De, A., Davis, C. J., Winters, B. D., Clinton, J., et al. (2010b). ATP and the purine type 2 X 7 receptor affect sleep. *Journal of Applied Physiology* (japplphysiol.00586.02010).

Kushikata, T., Fang, J., & Krueger, J. M. (1999). Brain-derived neurotrophic factor enhances spontaneous sleep in rats and rabbits. *AJP – Regulatory, Integrative and Comparative Physiology*, *276*(5), R1334–1338.

Luppi, P. H., Gervasoni, D., Verret, L., Goutagny, R., Peyron, C., Salvert, D., et al. (2007). Paradoxical (REM) sleep genesis: The switch from an aminergic-cholinergic to a GABAergic-glutamatergic hypothesis. *Journal of Physiology, Paris*.

Magistretti, P. J. (2006). Neuron-glia metabolic coupling and plasticity. *The Journal of Experimental Biology*, *209*(12), 2304–2311.

Matsui, T., Svensson, C. I., Hirata, Y., Mizobata, K., Hua, X.-Y., & Yaksh, T. L. (2010). Release of prostaglandin E2 and nitric oxide from spinal microglia is dependent on activation of p38 mitogen-activated protein kinase. *Anesthesia & Analgesia*, *111*(2), 554–560.

Morozov, A., Kellendonk, C., Simpson, E., & Tronche, F. (2003). Using conditional mutagenesis to study the brain. *Biological Psychiatry*, *54*(11), 1125–1133.

Opp, M. R. (2005). Cytokines and sleep. *Sleep Medicine Reviews*, *9*(5), 355–364.

Pascual, O., Casper, K. B., Kubera, C., Zhang, J., Revilla-Sanchez, R., Sul, J. Y., et al. (2005). Astrocytic purinergic signaling coordinates synaptic networks. *Science*, *310*(5745), 113–116.

Petit, J. M., Tobler, I., Allaman, I., Borbely, A. A., & Magistretti, P. J. (2002). Sleep deprivation modulates brain mRNAs encoding genes of glycogen metabolism. *The European Journal of Neuroscience*, *16*(6), 1163–1167.

Scales, S. J., Bock, J. B., & Scheller, R. H. (2000). The specifics of membrane fusion. *Nature*, *407*(6801), 144–146.

Scharf, M. T., Naidoo, N., Zimmerman, J. E., & Pack, A. I. (2008). The energy hypothesis of sleep revisited. *Progress in Neurobiology*, *86*(3), 264–280.

Strecker, R. E., Morairty, S., Thakkar, M. M., Porkka-Heiskanen, T., Basheer, R., Dauphin, L. J., et al. (2000). Adenosinergic modulation of basal forebrain and preoptic/anterior hypothalamic neuronal activity in the control of behavioral state. *Behavioural Brain Research*, *115*(2), 183–204.

Szymusiak, R., Gvilia, I., & McGinty, D. (2007). Hypothalamic control of sleep. *Sleep Medicine Advances in Sleep Medicine*, *8*(4), 291–301.

Szymusiak, R., & McGinty, D. (2008). Hypothalamic regulation of sleep and arousal. *Annals of the New York Academy of Sciences*, *1129*(1), 275–286.

Tobler, I., Borbély, A. A., Schwyzer, M., & Fontana, A. (1984). Interleukin-1 derived from astrocytes enhances slow wave activity in sleep EEG of the rat. *European Journal of Pharmacology*, *104*(1–2), 191–192.

Urade, Y., & Hayaishi, O. (2011). Prostaglandin D2 and sleep/wake regulation. *Sleep Medicine Reviews*, *15*(6), 411–418.

Wisor, J. P., & Clegern, W. C. (2011). Quantification of short-term slow wave sleep homeostasis and its disruption by minocycline in the laboratory mouse. *Neuroscience Letters*, *490*(3), 165–169.

Wisor, J. P., Clegern, W. C., & Schmidt, M. A. (2011). Toll-like receptor 4 is a regulator of monocyte and electroencephalographic responses to sleep loss. *Sleep*, *34*(10), 1335–1345.

Wisor, J. P., Schmidt, M. A., & Clegern, W. C. (2011). Evidence for neuroinflammatory and microglial changes in the cerebral response to sleep loss. *Sleep*, *34*(3), 261–272.

Yamamoto, K., Miwa, T., Ueno, R., & Hayaishi, O. (1988). Muramyl dipeptide-elicited production of PGD2 from astrocytes in culture. *Biochemical and Biophysical Research Communications*, *156*(2), 882–888.

Zhang, Q., & Haydon, P. G. (2005). Roles for gliotransmission in the nervous system. *Journal of Neural Transmission*, *112*(1), 121–125. doi:10.1007/s00702-004-0119-x.

Zimmerman, J. E., Mackiewicz, M., Galante, R. J., Zhang, L., Cater, J., Zoh, C., et al. (2004). Glycogen in the brain of *Drosophila melanogaster*: Diurnal rhythm and the effect of rest deprivation. *Journal of Neurochemistry*, *88*(1), 32–40.