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CYTOKINES, HOST DEFENSE AND SLEEP

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Chapter Outline;

- a) Microbe-induced changes in sleep
- b) Molecular mechanisms of microbe-altered sleep
- c) Sleep and the immune response

Inflammatory states are associated with profound changes in sleep, sleepiness and fatigue. Cytokines mediate these effects; they form biochemical networks within the brain and immune system. Cytokines are group of proteins and glycoproteins that behave like hormones and neurotransmitters. The dynamic changes in these chemical entities that occur during normal sleep and over the course of inflammatory diseases have been characterized in animal models (Table 1).

Table 1: Abbreviations used in the Language of Cytokines

<p><i>A1AR, adenosine A1 receptor; CRH, corticotrophin releasing hormone; cry, cryptochrome; EGF, epidermal growth factor; GABA, gamma aminobutyric acid; GHRH, growth hormone releasing hormone; glu, glutamic acid; IL1, interleukin-1 beta; IL10, interleukin-10; IL1RA, IL1 receptor antagonist; IL4, interleukin-4; NFkB, nuclear factor kappa B; NGF, nerve growth factor; NO, nitric oxide; NOS, nitric oxide synthase; per, period; PG, prostaglandins; sIL1R, soluble IL1 receptor; sTNFR, soluble TNF receptor; TNF, tumor necrosis factor alpha</i></p>
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Table 2: Effects of Cytokines on Sleep

Cytokine	Brain Stimuli that Promote Production/Release	Sleep Effects: Promotes
Interlukin-1 beta	IL1, TNF, NFkB, sleep loss, microbes, neuronal activity, stress, feeding	NREMS/EEG SWA

Interleukin-6	IL1, TNF, NFkB, sleep loss, microbes, stress	NREMS
Tumor Necrosis Factor α	TNF, IL1, NFkB, sleep loss, microbes neuronal activity, stress, ambient temperature	NREMS/EEG SWA
Nerve Growth factor	IL1, TNF, NFkB, sleep loss, neuronal activity, microbes, stress	NREMS/REMS
Brain-Derived Neurotrophic Factor	Neuronal activity, stress, microbes, sleep loss	NREMS/REMS
Growth Hormone Releasing Hormone	IL1, sleep loss, microbes	NREMS/EEG SWA

Abbreviations: NREMS, non-rapid eye movement sleep; REMS, rapid-eye movement sleep; EEG SWA, electroencephalogram slow wave (1/2-4 Hz) activity.

In animals, microbes often enhance non-rapid eye movement sleep (NREMS) while rapid-eye movement sleep (REMS) is inhibited. These responses usually begin to occur within a few hours of exposure and last from several days to weeks. Their magnitude, duration and direction depend upon the specific microbe, host species, location of inflammatory site, and host physiological status, e.g., phase of circadian rhythm. Figure 1 shows the NREMS response of mice induced by intranasal challenge with influenza virus. Although the virus does not replicate within the brain, influenza viral positive and negative sense RNA and viral protein can be detected in the brain within 7 hours. These viral products induce cytokine production locally within brain tissue including the hypothalamus and these actions probably initiate the acute phase response. The virus also localizes in the lung where it replicates and induces large increases in cytokines.

NREMS

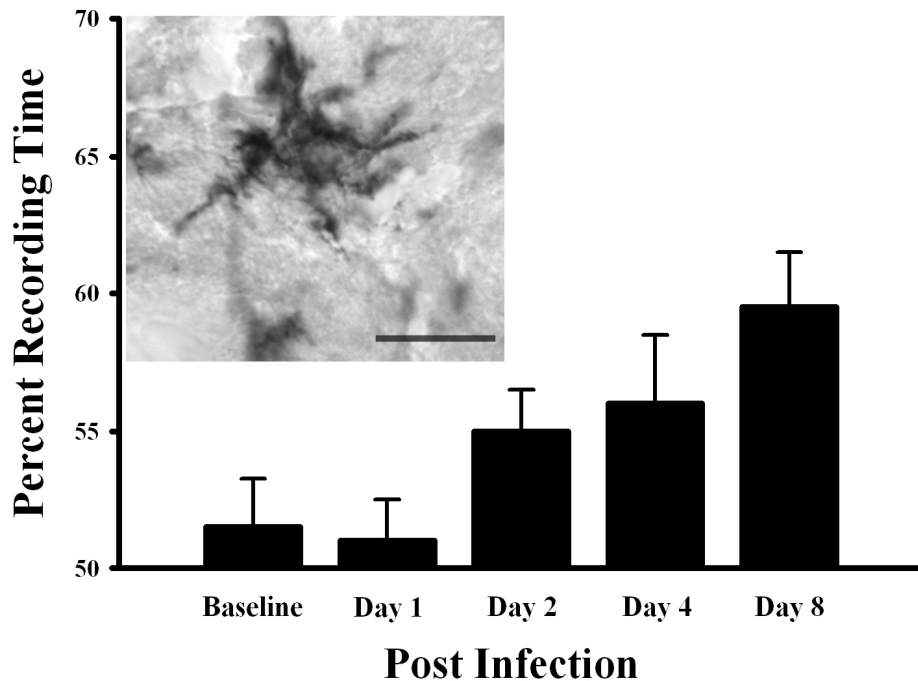


Figure 1: Influenza virus infection greatly increases non-rapid eye movement sleep (NREMS). By day 2 after intranasal inoculation of mice with influenza virus NREMS increases. These increases persist for a week or more. The mice recover from the infection after about 2 weeks and sleep returns towards normal values at that time. The insert shows the detection of viral antigen within the olfactory bulb after inoculation. The cell shown has characteristics of microglia and appears to be activated. Microglia are responsible, in part, for the cellular host defenses in brain. Such cells likely produce cytokines and they in turn are involved in sleep regulation.

Systemically-produced cytokines also influence brain functions such as sleep (Figure 2) and are thought to be responsible for the longer-term maintenance of the acute phase sleep responses.

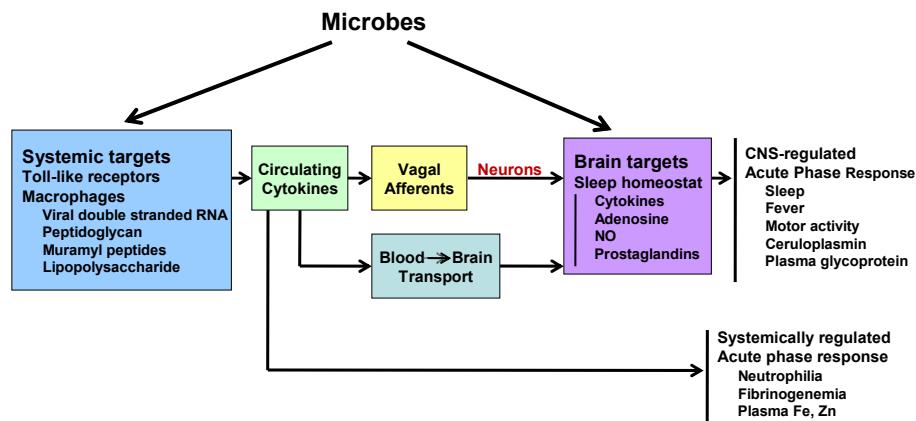


Figure 2: Microbes affect sleep via steps involving systemic immunocytes such as macrophages or if they infiltrate the brain directly via glia and neurons. Components of bacterial cell walls such as lipopolysaccharide from Gram-negative bacteria, muramyl peptides from either Gram-negative or Gram-positive bacterial peptidoglycan, and double-stranded RNA from viruses interact with Toll-like receptors. This interaction leads to enhanced cytokine production. Cytokines released systemically reach the brain via specific blood-to-brain transporters or can signal the brain via vagal nerve afferents. Cytokines released in the brain directly interact with brain targets to enhance sleep. There are several brain-regulated acute phase responses including sleep, fever, social

withdrawal, reduced locomotor activity and some serum proteins. Systemically released cytokines also act on other organs such as the liver and spleen to affect a variety of additional acute phase reactants such as plasma iron and zinc levels.

The molecular steps by which microbes and associated inflammation alter sleep involve the amplification of those mechanisms responsible for physiological sleep (Figure 3).

The Sleep Homeostat

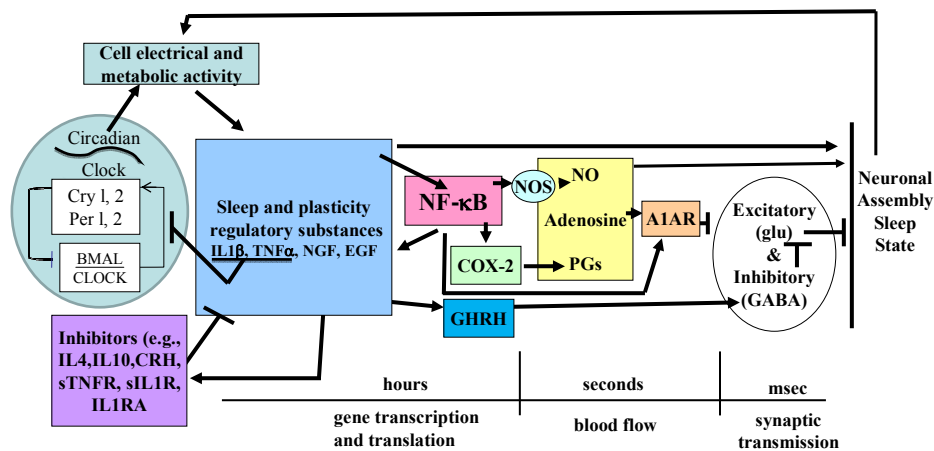


Figure 3: Microbes act to amplify the production of many of the components of the sleep homeostat. The cytokines such as IL1, TNF, IL4, IL10, NGF, EGF and associated soluble and membrane-bound receptors all form part of the sleep biochemical regulatory network. Microbial products affect immunocyte and brain production of these substances. Within brain and immunocytes, ATP, co-released for example during neurotransmission, induces the release of IL1 and TNF from glia. These substances induce their own production and multiple other substances via nuclear factor kappa B

activation. These actions are associated with gene transcription and translation and take several hours. Down-stream events include well-known immune response modifiers and regulators of the microcirculation such as NO, adenosine and prostaglandins. They in turn affect neurotransmission on an even faster time scale leading to state oscillations within local networks (Krueger et al 2007). See Table 1 for abbreviation.

Tumor necrosis factor alpha (TNF) is but one of many such substances that collectively constitute the sleep homeostat, an ultra-complex neuronal and glial biochemical network. Hypothalamic and cerebral cortical levels of TNF mRNA or TNF protein have diurnal variations (2 and 10 fold respectively) with higher levels associated with greater sleep propensity. Sleep loss is associated with enhanced brain TNF. Central or systemic TNF injection enhances sleep. Inhibition of TNF using the soluble TNF receptor, or anti-TNF antibodies, or a TNF small inhibitory RNA reduces spontaneous sleep. Mice lacking TNF receptors have less spontaneous sleep. Injection of TNF into sleep regulatory circuits, e.g., the hypothalamus, promotes sleep. In normal humans, plasma levels of TNF co-vary with EEG slow wave activity and in multiple disease states plasma TNF increases in parallel with sleep propensity. Downstream mechanisms of TNF-enhanced sleep include nitric oxide, adenosine, prostaglandins and activation of nuclear factor kappa B. That many of the molecules implicated in physiological sleep regulation (Figure 3) are also known regulatory components in immunocytes suggests an evolutionary link between sleep and host defenses.

Sleep *per se* may feedback to affect the efficacy of the host defenses. There are numerous studies in humans demonstrating that sleep loss alters many immune parameters including, antibody responses to vaccines, bacterial translocation from the intestine, lymphocyte mitogenesis, phagocytosis, antigen uptake, circulating immune complexes, circulating immunoglobulin, and natural killer cell and T lymphocyte populations. Sleep loss and several sleep pathologies, such as sleep apnea and insomnia affect circulating levels of certain cytokines such as TNF and interleukin-6; both of these are pro-inflammatory cytokines critical to the development of the acute phase response. Although such findings strongly suggest that sleep plays a role in host defenses the important question of whether sleep and/or sleep loss alters microbial-associated morbidity or mortality remains unanswered. This is a difficult issue to address because it is not possible to isolate sleep as the independent variable; all physiological functions vary with state.

Suggested Readings:

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