Trends in Neurosciences

CellPress

Opinion

Call it Worm Sleep

Nicholas F. Trojanowski[1] and David M. Raizen[1,*]

The nematode *Caenorhabditis elegans* stops feeding and moving during a larval transition stage called lethargus and following exposure to cellular stressors. These behaviors have been termed 'sleep-like states'. We argue that these behaviors should instead be called sleep. Sleep during lethargus is similar to sleep regulated by circadian timers in insects and mammals, and sleep in response to cellular stress is similar to sleep induced by sickness in other animals. Sleep in mammals and *Drosophila* shows molecular and functional conservation with *C. elegans* sleep. The simple neuroanatomy and powerful genetic tools of *C. elegans* have yielded insights into sleep regulation and hold great promise for future research into sleep regulation and function.

Behavioral and Electrophysiological Properties of Sleep

The core function of sleep is a long-standing mystery. Theories for sleep function include roles in brain energetics [1], brain recovery [2], somatic functions and thermoregulation [3], biosynthesis [4], neural plasticity [5,6], and allocation of energetic resources [7]. A key challenge for sleep researchers is to determine whether identified sleep functions are particular to the organism of study, or if they represent core functions that led to the evolutionary maintenance of sleep states. Comparative physiology across phylogeny is a powerful approach to distill conserved sleep functions.

Until the mid-1930s, sleep in all animals was identified by three behavioral characteristics: decreased responsiveness to environmental stimuli, rapid reversibility to strong stimuli, and an increased threshold to arousal [8]. The development and use of the electroencephalogram (EEG) revealed that brain activity patterns during sleep are variable [9] and that sleep can be divided into physiologically distinct states based on electrical activity patterns [10]. The subsequent discovery of rapid eye movement (REM) and non-REM sleep [11] demonstrated that these different sleep states are generated by distinct physiological processes [12]. Experiments using the EEG have also revealed that slow wave activity can serve as a biomarker for sleep need in certain settings [13,14].

In the 1960s, the ontogenetic hypothesis of sleep, which suggested sleep played an important role in the development of the nervous system specifically in young animals, came to prominence [15]. More recently, it was found that sleep can be triggered by both circadian [16] and noncircadian factors [17], such as bacterial infection [18], through at least partly distinct neural pathways [19,20]. Together, this indicates that there is additional heterogeneity in sleep. While the physiological differences between mechanisms generating REM and non-REM sleep are widely appreciated, the function and regulation of these states are incompletely understood [21]. Likewise, little is known about the physiological and functional differences between infection-induced and circadian sleep, or between sleep at different developmental stages [22].

The past 15 years have witnessed the introduction of nonmammalian model organisms, such as *Danio rerio*, a zebrafish [23], and *Drosophila melanogaster*, a fruit fly [24,25], to sleep research, providing insights into the function and regulation of sleep. Because EEGs are not performed in non-mammals, sleep in such species must be defined by behavioral criteria so as to differentiate

Trends

C. elegans has two states that meet the behavioral criteria for sleep, developmentally timed sleep (DTS) and stress-induced sleep (SIS).

DTS and SIS are regulated by the same signaling pathways that regulate mammalian sleep, including pigment dispersing factor (PDF), cAMP, epidermal growth factor receptor (EGFR), dopamine, and protein kinase (PK)G signaling.

C. elegans sleep has proposed functions similar to those of mammalian sleep, including synaptic plasticity, anabolic metabolism, and stress response, and is similarly prominent during development.

Animals in DTS and SIS display behaviorally identical cessation of feeding and locomotion, but this quiescence is produced by different mechanisms in each state.

Homeostatic responses to disruption of DTS by gentle and harsh stimuli occur through molecularly distinct pathways.

[1]Department of Neurology, Perelman School of Medicine, University of Pennsylvania, Philadelphia, PA, USA

*Correspondence: raizen@mail.med.upenn.edu (D.M. Raizen).

Table 1. *C. elegans* Fulfills Behavioral Criteria of Sleep

	Mammals	*Drosophila*	*C. elegans*	Refs for *C. elegans*
Reversible quiescent behavior	Yes	Yes	Yes	[29,31,89]
Increased arousal threshold	Yes	Yes	Yes	[29,35]
Stereotypical posture	Yes	Yes	Yes	[32–34]
Homeostatic response to deprivation	Yes	Yes	Yes	[29,37,93]
Sleep deprivation can be lethal	Yes	Yes	Yes	[37]

this state from quiet wakefulness or pathological quiescence. In addition to locomotion and feeding quiescence, rapid reversibility, and reduced sensory responsiveness, Campbell and Tobler [26] and others [27,28] have proposed that a stereotypical body posture and a homeostatic response to sleep deprivation should be included in these criteria.

States that fulfill the behavioral criteria for sleep have been also identified in *C. elegans*, a nematode (Table 1). However, unlike in zebrafish and *Drosophila*, in *C. elegans* these behavioral states have been referred to as 'sleep-like states' rather than sleep states [29,30]. We argue here that these nematode behavioral states should be referred to as sleep.

C. elegans Sleeps

C. elegans has two states that fulfill all the behavioral criteria for sleep: Developmentally timed sleep (DTS) or lethargus, and stress-induced sleep (SIS).

DTS occurs for two to three hours following each of the four larval stages [29,31], and is characterized by behavioral quiescence, stereotypical posture, decreased response to sensory stimuli, and homeostatic response to sleep deprivation [29,32]. Feeding and locomotion, which persist throughout the development of the worm (Video S1 in the supplemental information online), cease during DTS (Video S2), and worms tend to assume a hockey stick-shaped posture [32–34]. The worms are more difficult to arouse during this time [29], partially due to decreases in evoked Ca^{2+} response in sensory neurons [35,36]. Deprivation of DTS, which can be lethal [37], is followed by a homeostatic rebound [29]. These bouts of immobility are interspersed with activity bouts, during which animals move but do not feed [32].

SIS occurs following exposure to environmental stimuli that cause cellular stress, and its duration depends on the severity of the stressor [30,38]. Like DTS, SIS is characterized by a similar cessation of feeding and locomotion and increased arousal threshold [30] (Video S3). Although a homeostatic response to deprivation of SIS has not been documented, impaired SIS results in increased mortality [30], demonstrating that this sleep state has an adaptive physiological function. SIS can occur during any developmental state, but the possibility that prolonged waking is stressful, and therefore causes SIS, has not been tested.

Satiety behavior is induced by feeding on high-quality food or by refeeding after starvation and shares behavioral quiescence with SIS and DIS, but its other sleep properties have not yet been assessed [39,40].

The Importance of Nomenclature

Why is the correct nomenclature important? One could argue that the term 'sleep-like' has sufficed to advance the field. However, 'sleep-like' carries the connotation that *C. elegans* sleep is like sleep but somehow not quite the same, as if there is a universally accepted definition of sleep that is met by other organisms but not *C. elegans*. What is this universally accepted definition of sleep that *C. elegans* 'sleep-like' states fail to meet? Should a qualifying 'like' be