Current Biology Vol 23 No 18
R834

Dispatches

Sleep Physiology: Setting the Right Tone

Humans prone to cataplexy experience sudden losses of postural muscle tone without a corresponding loss of conscious awareness. The brain mechanisms underlying this debilitating decoupling are now better understood, thanks to a new study using cataplectic mice.

Mark S. Blumberg

Some things just go well together. During the day, our waking activities — walking and running, eating and gesturing — entail large, coordinated muscle movements and supportive postural muscle tone, typically accompanied by a general awareness of what we are doing. Conversely, at night as we sleep, we are immobile, our muscle tone is low or absent, and we are largely unaware of our surroundings. And so go our lives, at least for most of us. But for some of us, our motor and cognitive experiences can become uncoupled in odd, often disturbing, and sometimes dangerous ways: we might walk in our sleep (*somnambulism*), wake up paralyzed in a semi-conscious state (*sleep paralysis*), seemingly 'act out our dreams' in ways that can endanger the dreamer and his/her bedroom companion (*REM sleep behavior disorder*), or suddenly lose all muscle tone and collapse to the ground, functionally paralyzed yet completely aware of what is happening in the immediate environment (*cataplexy*) [1]. Such disorders blur the boundaries between sleep and wake states, revealing inadequacies in the language we use to describe them. But this blurring also inspires and focuses our search for biological mechanisms. For those perplexed by the decoupling of muscle tone and arousal that characterizes cataplexy, a beautiful new study reported in this issue of *Current Biology* by Burgess and Peever [2] offers several clarifying insights.

Cataplexy is one of the prominent symptoms of narcolepsy, a neurodegenerative disorder that is also characterized by excessive daytime sleepiness [1]. In one of the great success stories of modern neuroscience [3], the co-discovery in the late-1990s [4,5] of a new neurotransmitter, called hypocretin (or orexin), led rapidly to the realization that this neurotransmitter is intimately connected with narcolepsy in dogs [6] and humans [7,8]. In short order, hypocretin knockout mice were developed and found to exhibit many of the features of human narcolepsy, including cataplexy [9,10].

The loss of muscle tone during cataplexy resembles the loss of tone that normally occurs during rapid eye movement (REM) sleep. This observation implicates those brain structures that control or mediate effects on sleep-wake activity and muscle tone. One potential candidate is the locus coeruleus, a dense collection of neurons in the midbrain that is a major source of noradrenaline throughout the brain (Figure 1). It has been known for many years that neurons in the locus coeruleus are most active during waking, less active during quiet sleep, and nearly silent during REM sleep [11], mirroring changes in muscle tone across those states. Locus coeruleus neurons also cease their activity during cataplectic episodes [12], just as they do during REM sleep. Moreover, anatomical connections between the noradrenergic neurons in the locus coeruleus and the hypocretin-producing neurons [13,14] — located exclusively within a small region of the hypothalamus — suggest that a dysfunctional relationship between the two systems underlies the uncoupling of muscle tone and arousal that characterizes cataplexy. Still, despite all these suggestive links, the hypothesis that the noradrenergic system (which comprises the locus coeruleus and several additional brainstem structures) is a key player in cataplexy had not been directly tested, until now.

Using adult wild-type and hypocretin knockout mice, Burgess and Peever [2] first demonstrated that the masseter muscle, a large jaw muscle, exhibits a sudden loss of muscle tone during cataplexy. (For the purposes of this study, the masseter muscle is a stand-in for all the postural muscles of the body.) With the loss of muscle tone established, they then focused their attention on a group of motoneurons in the brainstem — trigeminal motoneurons — that specifically control the masseter muscle. Similar to previous elegant studies from this lab using the trigeminal motor system [15,16], this was the model system used for the series of experiments in this new study.

In order to implicate a particular subset of adrenergic receptors (called α_1 receptors) in the modulation of cataplexy, Burgess and Peever [2] administered drugs systemically to activate or inhibit these receptors in the knockout mice. As predicted from similar experiments in dogs [17] and humans [18], activating α_1 receptors substantially reduced the incidence of cataplexy by 90% and inhibiting these receptors substantially increased the incidence of cataplexy by 92%. So far so good. The next step was to specifically tie these changes to the motoneurons controlling the masseter muscle. To do this, they had to infuse drugs in the vicinity of the trigeminal motoneurons as cataplectic episodes occurred. This was no easy feat as these episodes, even in hypocretin knockout mice, are exceedingly rare, normally occurring only once every several hours or so. Therefore, it was necessary to wait patiently for a cataplectic episode at night (when they occur more often), at which time the experimenter infused drugs to inhibit or activate α_1 receptors on trigeminal motoneurons.

As predicted, before and after a cataplectic episode when muscle tone is high, inhibition of α_1 receptors significantly decreased muscle tone; in contrast, during a cataplectic episode when muscle tone is already low, the same drug did not further reduce muscle tone. Given these results, it appears that noradrenergic drive to the

motoneurons is high before and after a cataplectic episode, but drops to low or minimal levels during an episode. However, because pharmacological inhibition of noradrenergic activity did not reduce masseter muscle tone to the same low paralysis-like levels as occur during cataplexy, other neurochemical systems must also be involved. Still, the prominent role of the noradrenergic system is clear.

Having shown that noradrenergic drive to trigeminal motoneurons is reduced during cataplexy, the next step was to show that increasing noradrenergic drive to these motoneurons during cataplexy is sufficient to restore muscle tone. To do this, the experimenter waited patiently as before for instances of cataplexy, at which time he selectively activated α_1 receptors on trigeminal motoneurons. Again as predicted, muscle tone in the masseter muscle, upon adrenergic activation, increased substantially from the low-tone levels of cataplexy to levels similar to those seen during waking. Separately, the authors also showed that α_1-receptor activation of trigeminal motoneurons similarly increased muscle tone during REM sleep, once again highlighting the similar mechanisms underlying low muscle tone in both REM sleep and cataplexy.

All together, these experiments help us to better understand how a mismatch can occur in the control of motor and arousal systems. With cataplexy, the muscle atonia of REM sleep and the cognitive awareness of waking are invoked simultaneously, because the brain structures that typically tie muscle activity and awareness together have become less stable (whether due to the neurodegeneration of hypocretin cells in narcoleptics or the absence of hypocretin neurons, beginning early in development [19], in knockout mice). As Burgess and Peever [2] suggest, their study "provides a new framework for understanding how the noradrenergic system controls normal physiology and behavior." Future work will need to address the interactions of the noradrenergic and hypocretinergic systems before, during, and after cataplectic episodes.

When I give lectures about sleep, most of the questions I receive concern phenomena that reside at the 'edges': Is sleepwalking more like walking in your sleep or sleeping while walking? If you open your eyes in the morning to discover you are frozen with paralysis, are you more awake or asleep? Is cataplexy a motor disorder of waking or a cognitive disorder of sleeping? The ambiguity presented by these conditions is fascinating and troubling. Our attempts to resolve these questions with words alone can feel more like a philosophical discussion — "Is a chair with only three legs still a chair?" — than a scientific one. Although under most circumstances our definitions of sleep and wake serve us well, their occasional failure reminds us that we should not be searching for essences of sleep and wake, but rather striving to understand these states as collections of components [20] — components that typically cohere but sometimes do not. Therefore, the best way to resolve the ambiguity posed by cataplexy and other similar phenomena is to leave the world of words and dive into the actual mechanisms involved.

The wonderfully precise and technically challenging experiments of Burgess and Peever [2] do exactly this by further dissecting the mechanisms underlying the ambiguity posed by cataplexy, namely, the point of interaction between systems governing muscle control and behavioral arousal. Therefore, with cataplexy, the rare but very real separability of two critical features of waking life has opened an avenue of research that is revolutionizing how we think about the most basic mammalian experiences of movement and awareness.

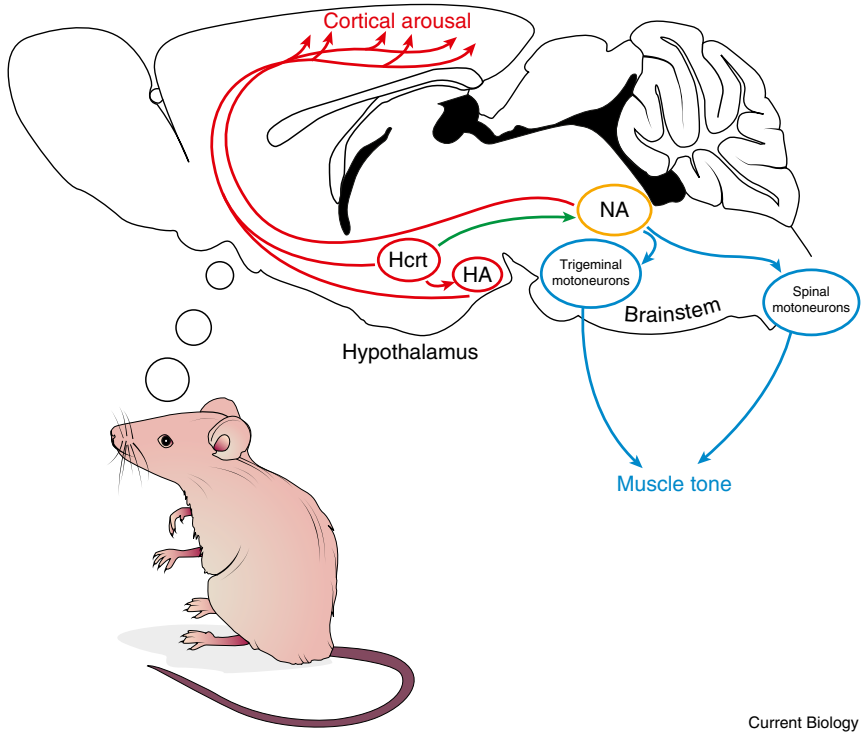

Figure 1. Neural systems linking arousal and motor control in mice.

Behavioral arousal in an awake mouse, as in other mammals, entails the combined activation of postural muscles and heightened arousal and alertness. Within the brain, the activated 'motor state' is supported by brainstem structures that include noradrenergic (NA) cells in the brainstem (including the locus coeruleus) projecting directly or indirectly to motoneurons within the brainstem and spinal cord. The heightened 'arousal state' is supported by structures that include the NA neurons in the brainstem, but also hypothalamic neurons that produce hypocretin (Hcrt) and histamine (HA); these neurons project widely within the cerebral cortex and are most active during wakefulness. During REM sleep, all of these systems become less active together. However, during cataplexy, an uncoupling occurs whereby a heightened arousal state is maintained at the same time as the animal exhibits a REM-like depressed motor state. Cataplectic episodes are very common in mice lacking hypocretinergic neurons, due in part to the loss of a functional linkage between them and noradrenergic neurons in the locus coeruleus (green arrow).

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http://dx.doi.org/10.1016/j.cub.2013.07.040

Memory Processing: The Critical Role of Neuronal Replay during Sleep

Patterns of neuronal activity present during learning in the hippocampus are replayed during sleep. A new study highlights the functional importance of this neurophysiological phenomenon by showing that neuronal replay is critical for memory processing over a night of sleep.

Jocelyn Breton and Edwin M. Robertson

As we sleep, facts, events and skills learnt during the day continue to be processed. Our memories become enhanced, stabilized and integrated with older memories, a process known as consolidation [1–3]. The biological mechanisms underlying memory consolidation remain poorly understood. Over forty years ago, David Marr [4] suggested that while we sleep, memories formed during the day may be replayed within the hippocampus, the part of our brain involved in memory formation. A new study [5] reported in this issue of *Current Biology* has sought to determine whether there is a critical connection between hippocampal replay and memory consolidation.

In a recent series of innovative studies, sensory cues were used to reactivate memories during a night of sleep [6–8]. In one such study, participants learnt to associate object-locations with the odor of rose-petals. During sleep, specifically during slow wave sleep (SWS), participants were exposed to either an odorless vehicle or to the rose-petal odor they were exposed to while awake. In the morning, participants were better able to recall object-locations if the rose-petal odor had been applied during the night compared to the odorless, control group. Furthermore, participants underwent functional brain imaging while in SWS and indeed, hippocampal activity increased during odor presentation. These findings demonstrate that the odor was able to reactivate, and thereby strengthen hippocampal-dependent memories [6].

Subsequent studies have gone on to demonstrate that memory reactivation can occur through the use of auditory cues. Various object-locations, each associated with characteristic sounds, were more accurately recalled if their associated sound was played during SWS [7]. Thus, auditory cues were able to selectively strengthen individual memories. Now, Fuentemilla and colleagues [5] have used auditory cues to reactivate memories during sleep in groups of patients, to test for a critical contribution of the hippocampus to memory reactivation and consolidation.

In this study [5], patients with chronic temporal lobe epilepsy, with damage restricted to the hippocampus, were tested in a memory reactivation task. Three groups of individuals were studied: those with unilateral hippocampal sclerosis; those with bilateral hippocampal sclerosis; and a healthy age-matched control group. On the first night, participants learned a list of 28 words, each of which was cued by an unrelated, unique sound. A sound was played and then a word would appear in one of four locations on a