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We recently reported that a gastropod mollusc, Lymnaea stagnalis, exhibits a sleep-like quiescent state. However quiescence differed from mammalian sleep in that both circadian and homeostatic regulation seemed to be absent in the snail. In the present paper I explore the possibility that the clustered pattern of quiescence observed in the snail may provide insight into underlying regulatory mechanisms. Specifically, I hypothesize that clustering of quiescence arises from feedback modulation of a stochastic wake-quiescence oscillator. The feedback loop is postulated to limit cumulative wakefulness and to exert probabilistic modulation of wake duration. Computer simulations using this model succeeded in simulating snail wake-quiescence behavior, confirming the plausibility of the hypothesis. Implications of this analysis for our understanding of mechanisms and evolution of sleep homeostasis are discussed.

In a recent paper¹ we reported that adult specimens of an aquatic gastropod mollusc, Lymnaea stagnalis, become spontaneously quiescent at apparently irregular intervals. Quiescence was found to be reversible, both spontaneously and in response to moderate stimuli, and we found that their behavioral sensitivity to appetitive and aversive stimuli was reduced during quiescence relative to that during periods of activity (hereafter referred to as “wake”). This combination of behavioral characteristics distinguishes sleep from vigilant rest and other inert states such as hibernation² and we therefore concluded that quiescence in the freshwater snail is sleep-like. However we also noted some interesting departures from sleep, insofar as it is understood in many other invertebrate and vertebrate species. In contrast to most other species studied to date, quiescence in Lymnaea was minimally influenced by time of day and instead followed a clustered pattern spanning an infradian timescale of 2–3 days. The durations of individual bouts of wake and quiescence were found to be random, with quiescence bouts lasting at least 5 min, but thereafter having a half-life of approximately 11 min. Furthermore, we observed neither a rebound in quiescence following various kinds of sensory stimuli, nor serial correlations between durations of quiescence bouts and contiguous wake bouts. We therefore suggested that the behavioral state of Lymnaea may be regulated by a stochastic oscillator with little or no modulation by circadian or homeostatic processes. However the fact that the quiescence bouts were grouped into clusters indicates that state transitions cannot be entirely random over the longer term (i.e., over a multiple-bout timescale). Thus, the temporal organization of behavioral state may harbor clues about the properties of underlying control mechanisms. Here I explore these issues further using a theoretical model of behavioral state regulation. I also briefly discuss some potential implications of this analysis for the sleep homeostasis concept in general.

Random events can exhibit clustering if event probability is modulated over time in a periodic or quasi-periodic pattern. Such fluctuations in event probability could arise by several mechanisms, of which the two main types are feedforward
The key parameters in the model were determined as the lag time (τω) and the rate of decline of Wω and ω during quiescence exceeds the rate of increase during intervening wake bouts. Hence, after several quiescence bouts ω falls below Tω and the cluster ends. A new cycle then begins.

The parameters in the model were found to be the lag time for Wω and Wω min, which together strongly influenced the durations of clusters and inter-cluster intervals, respectively. By definition, the values of Ptw either side of Tω determined the mean duration of wake bouts during clusters and during inter-cluster intervals.
Thus the feedback loop in the present model can be considered to be "homeostatic" if it is viewed as serving to keep cumulative wakefulness within tolerable, or perhaps optimal, limits (or as constraining the intervals between quiescence bouts). Alternatively, the feedback loop may be considered to be "non-homeostatic" in that it acts to generate variability (clustering), not to suppress it. Negative feedback loops are often considered to be the quintessential example of a homeostatic control mechanism because they tend to promote stability by opposing perturbations of a regulated variable. However, they only accomplish this when the control loop has the appropriate set of dynamic characteristics. Under certain conditions (e.g., high loop gain or delay in the system), the control loop will become unstable and give rise to an oscillatory output, as is shown in this study. Hence a feedback loop can be "designed" to introduce fluctuations in the controlled variable, which can be interpreted as non-homeostatic.

As can be seen in Figure 2, the inclusion of $W_{\text{min}}$ and $\omega$-delay in the model was essential for the development of clustering of quiescence bouts. An example of a simulated "hypnogram" is presented in Figure 2 and compared with data from a representative snail. Simulations have found that the appropriate adjustment of model parameters can accommodate inter-animal differences.

In conclusion, this theoretical analysis has succeeded in simulating the clustered infradian patterns of quiescence exhibited by Lymnaea. However, it is worth emphasizing that successful model simulations do not constitute positive evidence that such a mechanism is present in real animals. Simulations do, however, establish the plausibility of the hypothesis that clustering of quiescence can arise by modulation of a stochastic sleep-wake mechanism via an unstable feedback loop. The real acid test of the model will depend upon the outcome of experimental tests of quantitative predictions derived from the model.

These data and theoretical analysis raise several interesting points pertaining to the general concept of sleep homeostasis. In Lymnaea, it is the duration of wake episodes, not quiescence, which is subject to modulation. From the point of view of adaptive regulation of behavior there is no difference between whether this is interpreted as signifying modulation of wake maintenance or modulation of sleep onset; in principle both can achieve the same pattern of behavior. However, the implications of these two interpretations differ, particularly in relation to the neural and molecular mechanisms of state control, as well as for our understanding of the possible functional aspects of sleep-wake behavior.

In the proposed model, the feedback loop serves to limit cumulative excess wakefulness. Is this a homeostatic control system? Both "pro-homeostasis" and "anti-homeostasis" arguments can be made. Homeostasis is often incorrectly defined as the maintenance of internal constancy, whereas it is actually the maintenance of variables within appropriate limits. Thus the feedback loop in the present model can be considered to be "homeostatic" if it is viewed as serving to keep cumulative wakefulness within tolerable, or perhaps optimal, limits (or as constraining the intervals between quiescence bouts).

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**Figure 2.** (A) "Hypnogram" showing 7 days of data from a representative snail. $W$, wakefulness (activity); $Q$, quiescence. Note the infradian clustering of behavioral state. (B) Model simulation using the values of $P_n$ measured in the snail shown in (A). Computer simulation was run using the following model parameters: $W_{\text{min}}$, 60 min; $\omega$-delay, 180 min. Note that the stochastic nature of the model precludes an exact match between data and simulation—every simulation is different, as is every week of data from each snail. However this simple model is clearly able to account for the key dynamic properties of snail quiescence behavior. (C) Simulation run without $W_{\text{min}}$ and $\omega$-delay (parameter values set at 0), and $\omega$ at suprathreshold levels throughout. (D) Simulation run without $W_{\text{min}}$ and $\omega$-delay, and $\omega$ at sub-threshold levels throughout. Note that clustering does not occur (i.e., durations of simulated wake and quiescence bouts are random over time) in the absence of delays in the feedback loop.
To take this speculation one step further it is conceivable that a feedback loop may have evolved, not for homeostatic regulation of wakefulness or quiescence, but as a means to elicit clustered patterns of behavior. After all, it would not be the first time that such a mechanism had evolved—the circadian timing system is the classic example of this principle. Thus, in Lymnaea (and other animals too?) a feedback control system may be more concerned with regulating the timing, rather than the quantity of wakefulness and/or quiescence. Sleep rebounds following experimentally-induced extension of wakefulness may merely reflect the dynamic response of an unstable feedback loop, instead of a homeostatic compensation for sleep loss.

Our recent study failed to demonstrate a rebound in quiescence following deprivation, probably mainly due to methodological difficulties. However, even if the methods had been problem-free this negative result would have been inconclusive because absence of a sleep rebound does not necessarily imply absence of sleep homeostasis. A feedback system is neither necessary nor sufficient to establish the existence of homeostatic control. Many other classes of control system can achieve homeostatic regulation with or without concurrent feedback, including simple paced oscillators and feedforward commands. If snails don’t “need” much sleep (in our study they spent an average of about 10% of the total recording time in a quiescent state), or if extension of wakefulness has little effect on them (high tolerance limits), then a random oscillator, tuned by natural selection to deliver the appropriate overall quantities of wakefulness and sleep, can be considered to be a rudimentary yet sufficient homeostatic system. A simple wake-limiting feedback loop of the type proposed in this model might represent a first step in the evolution of a more precise control system and could possibly be the evolutionary precursor of sleep-wake regulation mechanisms seen in animals such as ourselves. As such, studies of phylogenetically distant animals such as Lymnaea may yield insights into the evolutionary origin and adaptive significance of sleep-wake regulation.

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