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juvant (CFA) and carrageenan (CAR). To identify the nociceptive fibers responsible for changes in sleep patterns, we also tested the effects on sleep of capsaicin (CAP) and mustard oil (MO), two compounds that respectively activate TRPV1 and TRPA1 channels expressed by specific subsets of nociceptors.

Methods: Adult C57Bl/6J male mice were instrumented with EEG/EMG electrodes for sleep recordings and telemetric transmitters (DSI) for body temperature and locomotor activity monitoring. After baseline recordings, we performed intraplantar injections of CFA, CAR, CAP, or MO (20 μl) at 12:30 pm and examined sleep-wake behavior and body temperature.

Results: Compared to saline, intraplantar administration of all compounds caused spontaneous pain behaviors (flinches of the injected paw) and hypersensitivity to mechanical and thermal stimuli (i.e. evoked pain). Unexpectedly, mice injected with CFA exhibited more sleep during the 3 subsequent dark periods compared to baseline. This hypersomnia dissipated on day 4. CAR, CAP and MO administration also increased sleep during the first dark period following the injection. Importantly, none of these compounds increased body temperature, indicating that the hypersomnia was not related to a febrile state.

Conclusion: Our results provide evidence of a putative protective sleep response after a painful challenge. CFA and CAR intraplantar injections are both standard models for the study of chronic inflammation. However, our data suggest that they might not be ideal for studying sleep disturbances associated with chronic inflammation and question the limited translational aspects of these models.

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0178

RIGID RED BLOOD CELL MEMBRANES AND LOW IRON RESULT FROM CHRONIC SLEEP DEFICIENCY IN RATS

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Introduction: The mammalian bone marrow and blood system is adapted to carrying out diverse, life-sustaining functions. Experimental sleep deficiency in otherwise healthy humans results in signs of an affected blood system, such as granulocytosis, iron deficiency, and a proinflammatory state. Insomnia is associated with anemia, and restless legs syndrome is associated with iron deficiency. The purpose of the present study was to investigate the extent to which red blood cells (RBCs) become more fragile due to sleep loss and the association of fragility with indices of iron.

Methods: Adult male Sprague-Dawley rats were chronically sleep restricted (SR, N = 5–7) for 72 days by disrupting their sleep (33% reduction) for six 10-day periods, separated by 2-day periods of sleep ad libitum according to a validated schedule. Ambulation controls (AC, N = 4–8) were produced by consolidating the ambulation requirements of sleep-restricted rats to allow for longer periods of uninterrupted sleep. Peripheral blood was tested for RBC mechanical and osmotic fragility. Decalcified bone marrow sections were stained with Prussian Blue for iron stores. Haptoglobin, which binds hemoglobin released by RBC lysis, was measured in heparinized plasma, as were plasma total iron binding capacity (TIBC), transferrin, and plasma osmolality.

Results: RBC hemolysis in response to increased concentrations of hypotonic solution was normal in AC rats, while that for SR rats was shifted to the right, indicating rigid membranes. Plasma osmolality was normal. Marrow iron stores were greatly reduced in SR rats compared with AC rats (0.07 vs. 1.0% area). Haptoglobin exceeded to upper limit of assay detection (> 1200 μg/ml) in SR rats, compared with AC rats (733 ± 367 [SD] μg/ml; P < 0.01). Both TIBC and transferrin were increased in SR rats (TIBC, SR vs. AC: 139 vs. 61 μg/dl, P < 0.01; transferrin: 1931 vs. 1367 μg/ml, P < 0.001, respectively).

Conclusion: These results indicate that chronic sleep restriction decreases the deformability of RBCs, which is a sign of iron deficiency and decreased RBC lifespan. Iron deficiency is indicated by low iron stores and increased TIBC and transferrin. Dramatically increased haptoglobin rules out excessive hemolysis and usually reflects low numbers of RBCs and inflammatory processes. Changes to blood cell production and function are expected to contribute to signs and disease risk associated with sleep deficiency because RBCs circulate throughout the body, potentially affecting cell-cell signaling and functions in many organs and systems.

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0179

HEART RATE VARIABILITY MEASURES OF WATCHSTANDING IN SIMULATED NAVAL WATCH SCHEDULES

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Introduction: Watch bills in Naval surface operations assign personnel to watches in rotating or fixed schedules to operate the ship around the clock. We simulated shipboard watch schedules in a laboratory and assigned continuous cognitive tasks during watch periods. Heart rate (HR) and HR variability (HRV) were measured as indices of psychologic state during simulated watchstanding.

Methods: N = 15 healthy, male subjects (ages 18–29) spent 5 consecutive days and nights in a laboratory. Subjects were assigned to one of four Naval watch schedules, each with 6 h of simulated watchstanding and 6.5 h opportunity to sleep daily on average. During simulated watches, subjects continuously performed cognitively challenging tasks. EKG was recorded throughout the study with Holter monitors. HR and low frequency (LF) and high frequency (HF) components of HRV were extracted in 5 min intervals. We compared these measures during watchstanding versus 30 min intervals immediately pre-watch and post-watch, controlling for time awake and time of day. For reference, we also compared these measures between scheduled wake and sleep periods. Statistical analyses employed mixed-effects ANOVA.

Results: HR was lower during simulated watches than immediately pre- and post-watch (F = 346, p < 0.001), and lower during sleep than during wakefulness (F = 7323, p < 0.001). HF and LF/HF ratio indicated greater vagal tone during watches than immediately pre- and post-watch (F > 34.4, p < 0.001), and even greater vagal tone during sleep (F > 1465, p < 0.001). LF, a speculative measure of sympathetic tone, was marginally increased during watches compared to 30 min pre-watch, and remained high 30 min post-watch (F = 6.2, p = 0.002). LF was substantially lower during other scheduled wakefulness and further decreased during sleep (F = 523, p < 0.001).

Conclusion: In this pilot study, HR and HRV indices differentiated sympathovagal balance during simulated watches from other wakefulness and from sleep. In particular, HR, HF and LF/HF ratio provided congruent evidence for increased vagal tone during watches compared to other waking periods. Using differences in HR and HRV measures between wakefulness and sleep as reference, this finding suggests that simulated watchstanding with continuous performance of cognitively challenging tasks was associated with physiologic symptoms that may indicate increased sleepiness—due to task load, reduced physical activity and/or sedentary posture.

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