

Stress-induced sleep rebound: adaptive behavior and possible mechanisms

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Abstract

The role of stress as a triggering factor for insomnia is widely accepted. However, not everybody who is submitted to a stressful event develops insomnia, which indicates that it depends on individual vulnerability. In addition, the neurobiology of vulnerable individuals is far from known. Animal models are valuable instruments to disclose mechanisms involved in the installation of the disease, not to mention the development of potential therapeutic agents and preventive actions. This review seeks to present data on the effects of stress on sleep and the different approaches used to study this relationship and possible neurobiological underpinnings and mechanisms involved. The results of numerous studies in animals indicate that increased sleep following a stressful situation is an important and adaptive behavior for recovery. However, this endogenous strategy appears to be impaired in human beings that exhibit high levels of anxiety and in animal models of anxiety-type behavior.

Keywords: sleep, paradoxical sleep, stress, HPA axis, anxiety-related behavior, animal models.

Introduction

The impact of stress on sleep has been recognized for millennia since Hippocrates' stated that sleep loss is a signal of pain and suffering that may lead to mental disorders, whereas excessive diurnal somnolence indicates the existence of disease. He also emphasized the importance of sleep as a medicine for physical and psychological stress (1). However, there are more subtleties to this relation than previously thought. Therefore, the nature and the length of the stressor, and, most importantly, the individual variability to cope with stress determine the outcome. Moreover, not only stress alters the sleep pattern but also inadequate sleep influences the stress response, i.e., whether one is sleep-deprived or not, determines the ability to respond to stress. This bidirectional association can, thus, represent a

vicious circle with detrimental consequences to mental health.

Among the numerous sleep disorders, the closest related to stress seems to be insomnia. Several epidemiological studies indicate that stress is a triggering factor for insomnia and that individual vulnerability is a major influence. For instance, Cartwright and Wood (2) reported that people facing a divorce process exhibited a reduction of delta sleep, which was normalized when the process was over. However, for those who underwent a lawsuit, the sleep pattern resembles that of depressive patients, including increased percentage of REM sleep. Nonetheless, only rarely stress causes, by itself, permanent sleep problems (3).

Interestingly, an important feature of insomniacs is the fact that although they experience just as many minor life stressors as good sleepers, they interpret these events as more stressful. They may, therefore, have greater predisposition to ruminate about the

Received: Jul 25, 2009.

Accepted: Jul 31, 2009.

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stressors and, consequently, to suffer a greater impact of these events, which would lead to longer-lasting sleep disturbances (4). Moreover, it has been shown in a group of primary insomnia patients that intrusive thoughts of stressful nature impact on sleep quality, producing more beta activity in the EEG and less delta power (5). Insomniacs often exhibit symptoms of anxiety, which become exaggerated at bedtime, and they also display dysregulation of the hypothalamic-pituitary-adrenal (HPA) axis so as adrenocorticotrophic hormone (ACTH) and cortisol plasma levels are elevated above those of healthy volunteers at the nadir of the circadian rhythm (6). That is to say that, most likely, patients show signs of insomnia because they have a hyperactive HPA axis, since the elevation of stress hormones is seen before, and not after, the unrestful night of sleep.

Sleep disturbances are hallmarks of depression and post-traumatic stress disorder (PTSD). These psychiatric pathologies are also triggered by stressful events, but individual vulnerability is again a predominant feature. In depression, delta sleep is impaired and latency to REM sleep is shortened, whereas percentage of REM sleep is augmented (7,8). It has been reported that sleep disturbances in PTSD include decreased total sleep time due to shorter delta sleep, increased wake time after sleep onset with sleep disruptions due to nightmares and increased REM density (9,10).

Effects of stress hormones on sleep

Stress, as much as sleep, is a global phenomenon that involves the entire central nervous system (CNS). That means that numerous neurotransmitter systems participate in the regulation of the stress response and of sleep. On the one hand, noradrenaline (NA) and acetylcholine (ACh) stimulate, whereas serotonin (5-HT) and gamma aminobutyric acid (GABA) inhibit, the activity of the paraventricular nucleus of the hypothalamus (PVN), where neurons that synthesize corticotrophin releasing hormone (CRH) are located. On the other hand, these neurotransmitters operate in an orchestrated manner to regulate sleep, so much so that NA induces waking and GABA, sleep; administration of ACh into specific brain regions in the pons induces rapid eye movement (REM) sleep and 5-HT is involved in the regulation of delta sleep (11).

The stress response entails two main systems, the locus coeruleus/medulla adrenal and the HPA axis. However, other hormones do take part, as is the case of Prolactin (PRL), Oxytocin and Vasopressin (AVP). When facing either a physical or psychological threatening stimulus, the organism responds by activating two main systems, which major function is to promote behavioral and metabolic adaptations to the situation so as to guarantee survival. These systems are briefly described below (for review, see (12-14).

Locus coeruleus/adrenal medulla

The locus coeruleus-adrenal medulla is also known as the sympathetic-adrenomedullary system and is part of the sympathetic nervous system. The preganglionic neurons constitute the cell bodies and are located in the intermediolateral (IML) cell column, which exit the spinal cord via the ventral root to form cholinergic synapses on the chromaffin cells of the adrenal medulla. When the

chromaffin cells are stimulated, adrenaline (80%) and noradrenaline (NA - 20%) secretion ensues and these catecholamines affect organs and tissues by binding to specific alpha and beta adrenergic receptors. The net result of activation of these receptors involves vasodilatation of muscle and vasoconstriction of skin blood vessels, increased blood pressure and heart rate, increased oxygen and glucose supply to skeletal muscles and brain, so as to assure the best conditions for the fight or flight reaction.

Although neither Adrenaline nor NA cross the blood-brain barrier, activation of the locus coeruleus parallels the peripheral actions of these catecholamines so as vigilance, arousal and increased attention to the source of the stressful stimulus is ensured. In addition, this system also stimulates the neuroendocrine limb of the stress response, represented by the hypothalamic-pituitary-adrenal (HPA) axis.

Hypothalamic-pituitary-adrenal axis

The paraventricular nucleus of the hypothalamus (PVN) convey information from ascending pathways originating in the brainstem and midbrain, more specifically from the nucleus of the solitary tract, locus coeruleus, and raphe nucleus, and from descending pathways originating in the limbic system (hippocampus and amygdala) and prefrontal cortex, which signal, respectively, physical and psychological threats to homeostasis. Stimulation of the PVN triggers a cascade of neuroendocrine events, which involves the release of CRH and AVP, which stimulates the synthesis and release of ACTH from the anterior pituitary. ACTH reaches the adrenal cortex and induces the synthesis and secretion of glucocorticoids (GC) – cortisol in human beings and primates and corticosterone (CORT) in rodents. GC regulate the activity of the HPA axis by means of a feedback signal at the prefrontal cortex, hippocampus, hypothalamus and pituitary that results from the steroid binding to the low affinity glucocorticoid receptor (GR or type II), ubiquitously distributed throughout the body. These receptors are involved with the suppressive effects of GC, including immunosuppression, increased waking (suppression of sleep), and the negative feedback regulatory function (suppression of HPA axis activity). In addition to these receptors, high affinity mineralocorticoid receptors (MR or type I) are also involved in the basal, permissive effects of GC. These effects include the induction of digestive enzymes, stimulation of fibrinogen production (in case of tissue injury), regulation of memory consolidation and regulation of REM sleep. Binding of GC to their cytoplasmic receptors leads to changes in gene transcription, which explains the long time lag for initiation of their effects. Binding of GC to their cytoplasmic receptors leads to changes in gene transcription, which explains the long time lag for initiation of their effects.

The hormones of HPA axis are involved with waking. This seems to be obvious, for who will feel asleep under stress or under a threatening situation? The main purpose of secretion of CRH, ACTH, GC and adrenaline is to increase stimulus directed-attention and to promote metabolic changes required to provide energy for appropriate “fight or flight” response (Figure 1). Recent evidence shows that CRH is one of the main humoral mediators of waking. CRH intracerebroventricular (i.c.v.) administration leads to reduction of slow wave sleep in rats, and a typical waking EEG

pattern (15). In addition, blockade of CRH receptors induces slow wave sleep, and this effect seems to be mediated, at least partly, by interleukin-1 (IL-1), since administration of CRH antagonist inhibits the release of CORT and increases mRNA for IL-1 α and IL-1 β (16). Exogenous administration of IL-1 during waking increases slow wave sleep and decreases waking in rats (17).

As for the influence of CORT, it has been shown that circulating concentration of CORT is essential for sleep regulation. On one hand, adrenalectomy, i.e., removal of the adrenal gland, hence of CORT source, results in shorter sleep episodes, less slow wave sleep and more waking during the resting phase in the rat, and the opposite is seen during the active phase. Replacement of GC at the level of basal concentrations (approximately 5 μ g/dl), restores normal sleep pattern, whereas higher doses (resulting in twice as much plasma levels) inhibits slow wave sleep (18) and increases waking time (19). Therefore, there seems to be a need of a subtle balance between GC concentrations and sleep pattern, as well as with other medical conditions, which is likely to be mediated by the proportion of occupation of both GCs receptors, the MR or type I and the GR or type II. This balance results from the differential affinity of these receptors for CORT. MR or type I is the high affinity receptor and it is stimulated even by very low (basal) glucocorticoid concentrations throughout the day, whereas GR or type II receptor is the low affinity receptor and is only stimulated by high circulating levels of CORT (under stressful conditions or at the peak of the circadian release of the hormone). Interestingly, a negative relationship between CORT levels and post-stress SWS rebound has been shown, whereas the manifestation of post-stress REM sleep rebound follows an inverted U curve, with low or very high levels resulting in little rebound (these relationships are represented in Figure 2).

As mentioned before, other hormones that are also secreted under stressful conditions have a role in sleep regulation, with a well-established relationship in human beings, who exhibit a monophasic sleep pattern. Growth hormone (GH) large secretory pulses are observed during the first half of the night, when delta sleep predominates, and when the lowest activity of the stress systems (including the HPA axis and the LC/adrenal medulla) is seen, indicating a negative relationship between cortisol and

adrenaline levels, and delta sleep (20). On the other hand, PRL levels rise after sleep onset, reaching a peak during the first hours of the morning (21). REM sleep promoting effects of PRL have been shown in cats, rats and rabbits and seems to be dependent on the phase of the diurnal cycle, since REM is induced when PRL is administered during the day (at the resting phase for rats), whereas administration of this hormone during the night inhibits REM (22). Recently it was reported that i.c.v. administration of PRL-releasing peptide induces REM sleep (23). Moreover, ether stress is a strong elicitor of both PRL secretion and REM sleep in rats; however, because the effect of ether on REM sleep is abolished in hypophysectomized animals, it has been suggested that induction of this sleep phase is mediated by PRL secretion (24).

Animal models for the study of stress-induced sleep changes

The use of animal models to study human pathologies can be advantageous insofar as they can, not only, reveal mechanisms of disease but also help to test potentially therapeutic drugs. Despite these advantages, it is specially difficult to model psychopathologies, because of the involvement of higher cognitive functions that cannot be recreated in animals. Therefore, it is possible to model some, but not all the features of these pathologies. Nevertheless, these models are extremely useful to reveal the neurobiology and, in some cases, the triggering or preventive conditions for the development and pathologies.

Stress-induced sleep changes stem from the interaction between genetic, neuroendocrine and neurophysiologic mechanisms. Thus, the way the animal initially reacts to the stressor may determine its subsequent sleep pattern. Interestingly, the neuroendocrine and behavioral reactions to stress are determined by the interaction between the genetic background and the history of the individual. The main approaches for the study of stress – sleep relationship are: 1) Environmental influence, in which the effects of different kinds of stressors, whether ethologic or not, are assessed; 2) Genetic influence, in which the sleep pattern of animals with different genetic backgrounds is evaluated after exposure to a stressor.

Table 1 – Features of mineralocorticoid and glucocorticoid receptors (MR and GR, respectively) in regards to their affinity to natural and synthetic glucocorticoids (GC), distribution and the main functions.

Mineralocorticoid Receptors (MR)	Glucocorticoid receptors (GR)
High affinity for natural GC (cortisol or corticosterone) and for aldosterone	Low affinity for natural GC and high affinity for synthetic GC (Dexamethasone)
Full occupation throughout the day (90 - 100%)	Low occupation during the nadir of the circadian rhythm (10%) and high occupation during the peak of the rhythm or during stressful situations (~ 70%)
High receptor density in the hypothalamus, hippocampus, lateral septum, medial and central amygdala, olfactory nucleus (for corticosterone) and in the circumventricular organ (aldosterone)	Widely distributed in the central nervous system (CNS), pituitary and periphery. In the CNS, GR are located in the pre-frontal cortex, hippocampus (but CA3 area), septum, amygdala, paraventricular and supra-optic nuclei
Associated with permissive and basal functions (\uparrow digestive enzymatic activity, fibrinogen production, enzyme involved in the conversion of noradrenalin to adrenalin)	Associated with the effects of stressful situations (suppressive effects) and responsible for the GC negative feedback

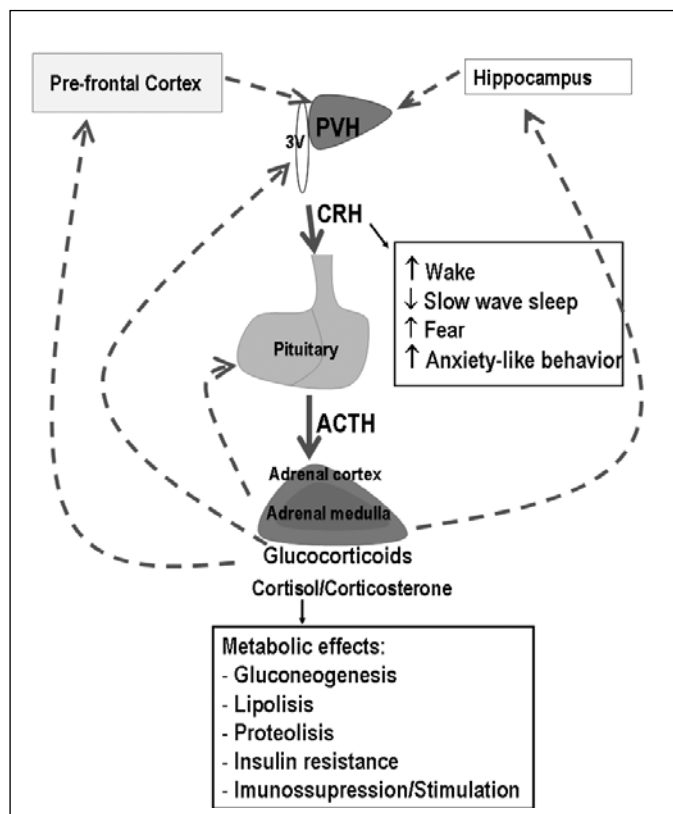


Figure 1 – Schematic representation of the hypothalamic-pituitary-adrenal (HPA) axis and main effects of corticotropin releasing hormone (CRH) and glucocorticoids (GC). Filled arrows represent stimulatory effects, whereas dotted arrows represent inhibition. The main sites of GC negative feedback are the pre-frontal cortex, hippocampus, hypothalamus and pituitary (adapted from (106)).

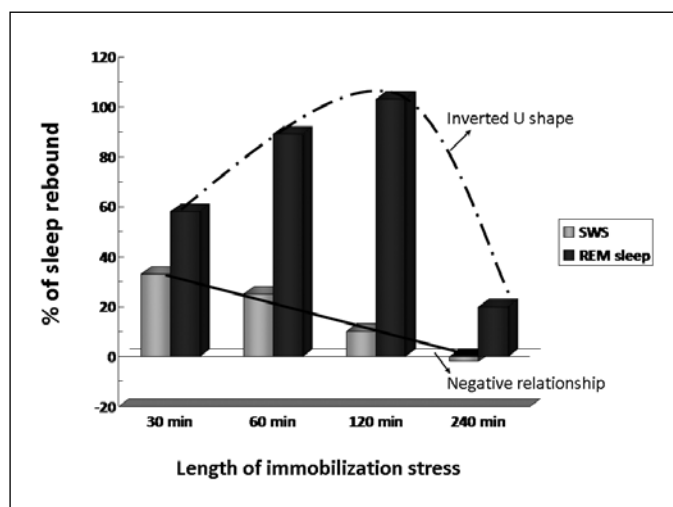


Figure 2 – Schematic representation the effects of stress on SWS and REM sleep rebound. The longer the stress period the higher the CORT secretion and consequently, there is a gradual inhibition of SWS rebound. REM sleep, however, is regulated by optimum levels of CORT, so either very low as well very high levels (induced by very short or very long periods of stress) result in meager rebound. Data is adapted from (25).

Environmental Influence

Several studies have shown that previous exposure to an acute stressor increases sleep time and that this sleep rebound is believed to represent a coping strategy that helps the organism recover. Nonetheless, the sleep rebound can be changed by many factors. For instance, there is a clear time course for this phenomenon to be manifested and what is generally reported is that during and a few hours after the stressor exposure, sleep is inhibited by the obvious mechanisms discussed above. This inhibition appears to be quite independent of the length of the stressor (25,26), suggesting a lack of habituation. After this initial inhibitory period, there is an increase in the time spent sleeping, with the highest levels being reached during the dark part of the light-dark cycle (26-31).

The length of stressor exposure is also an important variable that determines the magnitude of sleep rebound, inasmuch as 1 h of immobilization stress leads to a large increase in REM sleep (63% above baseline sleep) and a moderate increase of SWS (16% above baseline) (32), whereas 2 h of immobilization results in 32% increase in REM sleep and no change in slow wave sleep – SWS (33). By extending the period of stressor exposure to 4 h, the expression of sleep rebound is prevented, and the increase in REM or in SWS is no longer observed (25). One possible mediator of this wake-promoting effect of stress seems to be CORT, given the progressive increase of plasma levels of this hormone from 1 to 4 h of stressor exposure. These findings also indicate that inhibition of SWS by stress is a linear inverse function (the more intense the stressor, reflected by CORT levels, the less SWS rebound is obtained), whereas stress modulation of REM sleep follows an inverted U-shape curve, with very low or very high levels producing a smaller expression of REM rebound, and intermediate levels inducing a high expression (see Figure 2). Moreover, in adrenalectomized rats, i.e., removal of adrenal glands, and consequently, of the source of corticosterone production, REM sleep rebound is impaired. Administration of dexamethasone, a synthetic glucocorticoid that binds GR, also inhibits immobilization-induced REM sleep rebound (25), evidencing once again, the inverted U-shape curve effect of stress on sleep.

The exposure to stress in rats previously sleep-deprived is another form to challenge the system and to study the recovery effect of sleep. Prolonged sleep deprivation induced by instrumental methods is stressful per se (34-38). Interestingly, REM sleep deprivation produces sleep rebound, but this rebound can be differentially induced by related methods that lead to different stress levels, but similar changes in sleep electroencephalography (EEG) (35,36,39-43). Moreover, a further stressor can alter the expression of sleep rebound in sleep-deprived rats, such that REM sleep rebound is reduced, instead of increased, after immobilization of sleep-deprived rats (44). Therefore, association of different stressors can modify the expression of sleep rebound, possibly due to the interaction of different neurobiological systems that are recruited by these stressors. For instance, footshock increases serotonergic transmission (45,46), whereas immobilization increases dopaminergic transmission (47-49), two neurotransmitter involved with sleep regulation.

Even though sleep rebound takes place after many stressors, this is not a universal rule. For instance, several studies have shown that footshock, a fairly intense stressor, retards the onset of REM

sleep and prolongs the wake time following its application, sometimes even preventing the expression of sleep rebound during the night time (53). Rodents can learn to associate a neutral stimulus (sound, light) with an aversive stimulus, such as a footshock. After this association, the light or the sound is no longer neutral and the animal reacts to it. Single or multiple sound-shock pairings lead to shorter periods of REM and SWS although these reductions are more evident on the day following multiple pairings. Even 27 days after the training, presentation of tone results in reduction of REM, indicating that the strength of the pairing has a long-lasting effect on sleep (54). Shock can also be paired with a context, and when rats are left to sleep in a previously neutral, but currently aversive (due to pairing with a shock) cage, rats take longer to initiate sleep and REM sleep and they exhibit reduced percentage of REM sleep; however, when allowed to sleep in a neutral environment, these rats exhibit rebound of SWS and REM sleep indicating that they can distinguish between neutral and aversive contexts, and that distinction influences sleep differently (55).

In human beings, the most potent stressors are those of a social nature, including social hierarchy, family and/or work-related problems, inter-personal relationships. One example of social stressor in rodents is the social defeat paradigm, in which a male wild rat or mouse (resident) is housed with a female, establishing a territory that it will defend from intruders. The experimental animal (intruder) is introduced in this resident's cage and as soon as the resident attacks the intruder, a grid separates both animals to prevent physical injury, but, by then, the intruder is already defeated (56). The physiological changes seen in the defeated animal include an intense stress response, with increased catecholamine, corticosterone, prolactin and testosterone levels, increased heart rate, blood pressure and body temperature (57). The studies that employed social defeat report increased slow wave activity (SWA) during SWS, reflecting the augmented sleep intensity. Based on this result, it has been proposed that the function of SWA is to restore the inner balance following a traumatic event (50,58).

It is quite interesting that these responses can be also modulated by the genetic background and previous stress history of the animals. The gene-environment diathesis hypothesis has permeated numerous studies on the impact of stress on behavior, both in humans and in animal models. According to this hypothesis, the resulting resilience or vulnerability to stress-induced disorders are the consequence of the interplay between the genetic background and the environmental influence during specific time-windows, including infancy and adolescence. Adversity during these periods may result in augmented vulnerability, whereas social support throughout life may lead to resilience (59). A schematic representation of this proposition is presented in Figure 3.

With the idea that adversity during infancy might constitute a risk factor for psychopathologic-type behavior in rats, we performed a series of experiments employing the maternal separation (MS) and early handling paradigms. Numerous studies have shown that 3-6h of daily maternal separation – MS – results in increased novelty-induced fear (60) and propensity to consume alcohol (61), two well-known features of anxiety-like behavior. Moreover, these animals present hyper-responsiveness of the HPA axis to stress (62). Early handling, which consists of a brief separation from

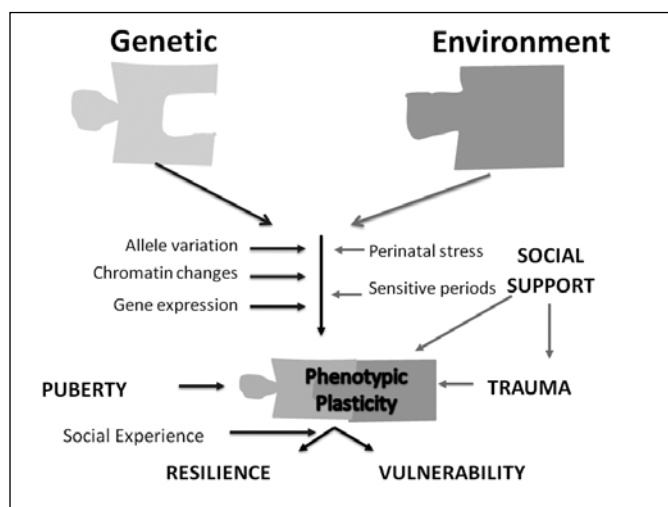


Figure 3 – The diathesis gene x environment hypothesis of resilience and vulnerability to stress-induced health problems. This schematic diagram is based on (59).

the mother for a period of 15 min/day, on the contrary, results in smaller response of the HPA axis to stressors and a more efficient negative feedback mechanism (62). Given the waking-promoting effect of CRH (63) and CORT (19), we hypothesized that MS would result in a phenotype of vulnerability to stress-induced insomnia. The rationale for the use of MS model was based on the fact that anxious individuals are at greater risk to develop chronic insomnia after a stressful situation (4,64,65).

We initiated this series of studies by challenging Wistar rats submitted to early handling, also named brief maternal separation (BMS - 15 min of separation from the mother during the first two weeks of life) with a 1 h restraint stress and assessed their sleep pattern before and after the challenge. We reasoned that BMS rats would display more sleep rebound than control rats, but to our surprise, they showed the same amount as control non-manipulated rats, i.e., increased time spent in SWS and in REM (28). We then proceeded to test MS rats, expecting that they would exhibit changes compatible with anxiety- and depressive-like behaviors and, consequently, less sleep rebound. But again, to our surprise we found that male rats displayed more paradoxical sleep in the light period of the baseline sleep than control and BMS rats, and a sleep rebound in the dark period of the light-dark cycle that was similar to the other groups (26).

Because women are in greater risk of developing depression and increased percentage of REM sleep is one of the hallmarks of depression, we proceeded by testing female rats, exposed to these early life manipulations, as to their sleep pattern after the same stressor used in male rats. We observed that MS females exhibited the largest REM rebound in the dark period after 1 h of cold stress, although all groups exhibited sleep rebound. Moreover, both manipulated groups (BMS and MS) exhibited a smaller secretion of CORT, although this did not reach statistical significance (29). Therefore, although we found gender differences in stress-induced sleep rebound we were unable to establish a model of vulnerability to insomnia induced by stress.

Another paradigm that has produced stress hyper-responsive is pre-natal stress. The most effective period to expose pregnant rats to the stressor so as the offspring becomes hyper-responsive is the third – and last – week of pregnancy. The offspring of mothers exposed to gestational stress present hormonal and behavioral changes that are compatible with depression-type behavior (66), including altered sleep regulation, with shorter latency to REM, more REM time and more awakenings (67).

Genetic Influence

The genetic background is one of the most important variables for determination of the behavioral expression. Therefore, the same stimulus may result in different outcomes, depending on how the animal reacts to it. Animal models that try to explore individual differences do so by two approaches: 1) Selection of animals within the same strain that respond in opposite ways to a given stimulus; 2) comparison of behaviors between different genetic and phenotypic strains.

Regarding the first approach, the selection of rats from the Wistar strain on the elevated plus maze (EPM), a test of anxiety-like behavior, show that some animals explore the open arms of this maze (and are, thus, classified as low anxiety-type behavior – LAB rats), whereas others avoid the open arms (and are, thus classified as high anxiety-type behavior – HAB rats). These substrains of the Wistar rats provide neuroendocrine and behavioral phenotypes that represent two different strategies of coping and fear behaviors in new situations (68,69). In response to a stressful situation, HAB rats exhibit lower percentage of sleep, specifically of SWS, than LAB rats. When treated with a CRH receptor 1 antagonist, the lack of stress-induced SWS rebound is prevented, confirming the hypothesis that sleep impairment in these rats is associated to a hyperactive CRH system (70).

Another behavior that can be selected, other than anxiety, is the level of locomotor activity that rats exhibit when placed in a new environment (high - HL or low locomotion - LL). HL rats also display higher CORT plasma levels, due to a prolonged secretion of the hormone, indicating an impairment of the negative feedback system. Interestingly, HL rats present more baseline waking and less SWS than LL rats; however, in response to a stressor, HL exhibit the characteristic sleep rebound, whereas LL rats do not show any change in sleep pattern (71).

The second approach consists of comparing different strains which basically differ in their behavioral and neuroendocrine responsiveness to stress. One such study compared Lewis (LEW) rats, which are deficient in CRH production with Fisher 344 (F344) rats, which are hyper-responsive to stress, and to Sprague-Dawley (Sp-D) rats. LEW and F344 strains are derived from Sp-D, therefore, the latter is used as a control strain. At basal situation, LEW rats exhibit less waking time and more SWS during the active phase, whereas F344 and Sp-D strains do not differ in their sleep pattern (72). In another study, comparison of F344 rats to Sp-D or to the outbred Wistar strains, demonstrated an adrenocortical and prolactin hyper-responsiveness to footshock stress and more anxiety-related behavior in the open field and elevated plus maze. In their home cages, F344 and Wistar rats are more active during the dark period and baseline sleep indicate that these two

strains also sleep less than LEW and Sp-D rats. The smaller time of sleep is due to reduction of both SWS and REM, but interestingly, the percentage of REM is similar for all strains (73). In response to a conditioning fear paradigm, performed in two days, LEW rats exhibit the largest sleep loss during the daytime period and the least recovery during the night-time period, compared to F344 and Wistar rats. Curiously, F344 and LEW exhibit similar levels of freezing behavior, which are higher than those exhibited by Wistar rats, indicating that the reaction to an aversive stimulus may not predict the sleep alteration (74).

In mice strains that differ in reactivity to noxious stimuli such as footshock and contextual fear conditioning, a clear and persistent loss of SWS and REM is observed in the more “anxious” BALB/cJ mice, whereas the less “anxious” C57BL/6J mice only show a loss of REM within the hour that follows shock presentation (75,76). A similar pattern of sleep reactivity is also seen in response to non-noxious manipulations, such as exposure to an open-field (77) or restraint stress (78): C57BL/6J mice display the characteristic rebound, whereas BALB/cJ strain displays sleep impairment.

Finally, the use of Wistar-Kyoto rat, a genetic model of depression has shed some light on the regulation of sleep in this disorder. These rats present the typical changes in sleep architecture found in depressive patients, i.e., shorter REM sleep latency, increased REM sleep percentage and increased sleep fragmentation (79). Treatment with antidepressants, however, was not shown to reduce REM in Wistar-Kyoto as much as in Sprague-Dawley rats, suggesting that they may be less sensitive to this class of drugs (80). Nonetheless, we recently performed a study with Wistar-Kyoto rats treated with citalopram and showed that not only REM sleep was reduced after the first administration of the drug but also the prolonged treatment resulted in a normalization of REM sleep in response to a learning paradigm that involves footshock (unpublished data). A recent study also demonstrated that REM sleep deprivation, an effective, although transient, treatment for depression, increases the expression of orexin in both Wistar-Kyoto and Wistar rats, indicating that orexin does not seem to be the mediator of REM sleep deprivation-induced antidepressant effect (81).

Mechanisms and mediators of stress-induced sleep changes

The first hypothesis to explain the mechanisms by which acute stressors induce sleep rebound involved the serotonergic system. It is based on a classical study which shows that blockade of serotonin synthesis or lesions of the Raphe nuclei induce insomnia (82). Classical studies show that serotonin is differentially released, depending on the phase of the light-dark cycle so that during waking, serotonin is released from the axon, whereas during sleep, it is released from the dendrites (83). The axonal release prepares the brain to sleep, due to induction of hypnogenic substances synthesis in target structures, such as the arcuate nucleus and pre-optic area (84), and likely initiates a cascade of genomic events (85,86). In the arcuate nucleus, serotonin induces the synthesis of corticotropin-like intermediate lobe peptide (CLIP), which might be directly involved in sleep rebound (83). Numerous evidence points CLIP

as a major candidate mediator of stress-induced sleep rebound. For instance, increased serotonin transmission induces pro-opiomelanocortin (POMC) mRNA in the arcuate nucleus, which, in turn, increases POMC derivatives, such as ACTH, α -melanocyte stimulating hormone (α -MSH) and CLIP (or ACTH18-39).

Although ACTH is well-known for its waking properties, α -MSH and CLIP induce sleep (87). CLIP content and its phosphorylated form increase in the dorsal Raphe nucleus immediately after immobilization stress, whereas such an increase takes place in the arcuate nucleus 4 h after the end of the stress, when REM sleep rebound reaches its maximum (88). Exogenous administration of CLIP or its N-terminal fragments ACTH20-24 and ACTH18-24 induces a significant increase of REM sleep, by increasing episode length (87,89).

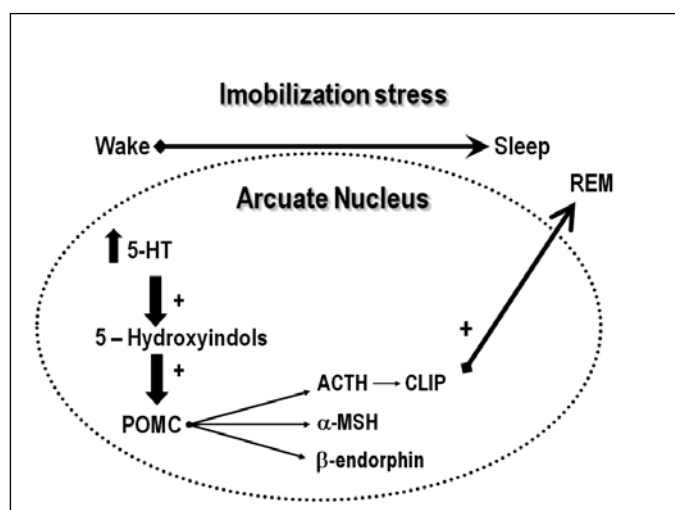


Figure 4 – The serotonergic hypothesis of acute stress-induced sleep rebound. According to this hypothesis, stress induces the release of serotonin, which is metabolized to 5-hydroxyindol substances that stimulates the cleavage of proopi melanocortin, originating adrenocorticotrophic hormone (ACTH), alpha-melanocyte stimulating hormone (α -MSH) and β -endorphin. ACTH is metabolized originating corticotropin-like intermediate lobe peptide (CLIP), which is a major inducer of REM sleep.

Interestingly, serotonin is a major stimulator of PRL secretion (90) and PRL increases the activity of cholinergic neurons in the mesopontine tegmental neurons (91), which are involved in the induction of REM sleep. These effects could, at least, partly explain the role of serotonin in stress-induced sleep rebound. This hormone seems to be a protective factor against harmful effects of stress, as indicated by a recent study showing that PRL protects hippocampal neurons from chronic stress-induced neuronal death, facilitating neurogenesis in the dentate gyrus (92).

Several neurotransmitter systems are altered by stress, one of these is the opioid system, since β -endorphin is one of the products of POMC cleavage, being released together with ACTH. The effects of β -endorphin on sleep are somehow controversial, since morphine i.c.v. administration inhibits both SWS and REM sleep, whereas morphine administration intra-LC or nucleus of the solitary tract induces sleep. Naltrexone, a selective blocker of μ -opioid recep-

tors prevents stress-induced sleep rebound, without interfering with CORT levels, indicating, counter-intuitively, the dissociation between the endocrine and sleep responses to stress. Interestingly, in stress-free conditions, naltrexone does not alter sleep pattern, indicating that this effect takes place only during stressful situations, and can be explained by the fact that stimulation of μ -opioid receptors inhibits the activity of LC neurons, inducing sleep (99). Therefore, blockade of opioidergic system with naltrexone maintains LC activity, inhibiting sleep rebound (100).

As mentioned earlier, PRL is closely related to REM sleep. Additional evidence of this relationship comes from studies in which stressors such as restraint and exposure to ether vapors also augment prolactin levels and the time spent in REM sleep (24,78). In mice strain that differ in anxiety-type behavior – C57BL/6J and BALB/cJ, restraint stress induces sleep rebound only in the less anxious strain, C57BL/6J, which also secretes significant amounts of PRL, whereas the more anxious strain, BALB/cJ, exhibit neither REM sleep rebound nor increased PRL secretion. Interestingly, both strains present a similar CORT response to the stressor (78). Recently, we showed that REM sleep deprived rats submitted to an intense, repeated stressor display the longest REM sleep rebound, with very long episodes and very high PRL levels, indicating that this peptide may be partially involved in the REM sleep rebound seen after exposure to stress (30).

Final Remarks

In human beings, insomnia may be triggered by stressful situations, especially by social stressors. Nonetheless, this is not a general rule and stress-induced insomnia depends on individual characteristics, particularly on anxiety-related personality features. This seems also to be the case for rodents, insofar as acute stressors normally elicit a sleep rebound that is considered a highly adaptive response, since sleep is believed to be a major recovery behavior. However, rats and mice which exhibit anxiety-type behavior show impairment of sleep.

The central nucleus of the amygdala (CeA) is closely involved with anxiety and may be a neural substrate for stress-induced insomnia. A large part of the evidence about the hyper-arousal produced by aversive stimuli comes from studies that employ the classical conditioning paradigm, which is highly dependent on the amygdala, a brain structure involved in the appraisal of emotional valence of several stimuli. The CeA show reciprocal connections with brain stem regions involved in the regulation of sleep-wake cycle, including the dorsal Raphe nucleus, the LC and the tegmental pedunculopontine and laterodorsal nuclei (101).

Despite the well-established role of the CeA on anxiety, there is some controversy on its role on sleep. On one hand, it has been reported that the electrical stimulation of CeA inhibits the discharge of LC neurons, resulting in increased REM sleep, and that global reduction of CeA activity by muscimol, a GABAergic agonist, reduces REM sleep, whereas intra-CeA administration of bicuculline, a GABAergic antagonist, increases REM sleep (102). On the other hand, a recent study showed that placement of rats in a cage previously dwelled by a dominant male, which represents a stressful

situation, inhibits REM sleep and activates neurons of arousal and waking-promoting areas, such as the LC, infralimbic cortex and the CeA. Interestingly, when rats are subjected to lesions in the CeA do not exhibit the delay in sleep onset or inhibition of REM sleep under stress, suggesting that the CeA is a major brain structure mediating hyper-vigilance induced by stress (103). Reasons for the discrepancy between these studies may lay in the distinct experimental conditions under which rats were tested. In the Sanford's study (102), rats were tested under non-stress conditions and pharmacologically challenged, whereas in the Cano's study (103), rats were tested during the stressful situation. Although these studies are hardly comparable or complementary, they highlight the importance of animal models to reveal the neurobiological underpinnings of the relationship between stress and sleep.

Stress-induced sleep rebound is an adaptive strategy that represents a major evolutionary acquisition, and inability to engage in it may have disastrous consequences. A good example of the importance of sleep rebound after a stressful situation is attested by the sleep assessment of individuals who experienced a traumatic event; those who exhibited long episodes of REM sleep did not develop post-traumatic stress disorder (PTSD), whereas those who had several, but very short, episodes of REM sleep developed the disorder. Moreover, the individuals who did not develop PTSD displayed more relative beta frequency during REM sleep, indicating high cognitive activation during this sleep phase. This frequency was negatively correlated with the occurrence of nightmares and with symptoms of PTSD (104). These findings suggest that long episodes of and consolidated REM sleep are essential for elaboration and integration of traumatic memories into the auto-biography of the patient (105), so they can resume their quotidian.

Acknowledgements

This work is supported by Associação Fundo de Incentivo à Psicofarmacologia (AFIP) and Fundação de Amparo à Pesquisa do Estado de São Paulo (FAPESP – CEPID, grant # 98/14303-3). Paula A. Tiba is the recipient of a post-doc fellowship from FAPESP and Deborah Sucbecki is the recipient of a research fellowship from the National Research Council (CNPq).

References

1. Vgontzas AN, Bixler EO, Kales A. Sleep, sleep disorders, and stress. San Diego: Academic Press; 2000. 449-457 p.
2. Cartwright RD, Wood E. Adjustment disorders of sleep: the sleep effects of a major stressful event and its resolution. *Psychiatry Res.* 1991;39(3):199-209.
3. Hall M, Thayer JF, Germain A, Moul D, Vasko R, Puhl M, Miewald J, Buysse DJ. Psychological stress is associated with heightened physiological arousal during NREM sleep in primary insomnia. *Behav Sleep Med.* 2007;5(3):178-93.
4. Morin CM, Rodrigue S, Ivers H. Role of stress, arousal, and coping skills in primary insomnia. *Psychosom Med.* 2003;65(2):259-67.
5. Hall M, Buysse DJ, Nowell PD, Nofzinger EA, Houck P, Reynolds CF, 3rd, Kupfer DJ. Symptoms of stress and depression as correlates of sleep in primary insomnia. *Psychosom Med.* 2000;62(2):227-30.
6. Vgontzas AN, Bixler EO, Lin HM, Prolo P, Mastorakos G, Vela-Bueno A, Kales A, Chrousos GP. Chronic insomnia is associated with nyctohemeral activation of the hypothalamic-pituitary-adrenal axis: clinical implications. *J Clin Endocrinol Metab.* 2001;86(8):3787-94.
7. Holsboer-Trachsler E, Seifritz E. Sleep in depression and sleep deprivation: a brief conceptual review. *World J Biol Psychiatry.* 2000;1(4):180-6.
8. Riemann D, Berger M, Voderholzer U. Sleep and depression--results from psychobiological studies: an overview. *Biol Psychol.* 2001;57(1-3):67-103.
9. Kobayashi I, Boarts JM, Delahanty DL. Polysomnographically measured sleep abnormalities in PTSD: a meta-analytic review. *Psychophysiology.* 2007;44(4):660-9.
10. Raboni MR, Tufik S, Sucbecki D. Treatment of PTSD by eye movement desensitization reprocessing (EMDR) improves sleep quality, quality of life, and perception of stress. *Ann N Y Acad Sci.* 2006;1071:508-13.
11. Pace-Schott EF, Hobson JA. The neurobiology of sleep: genetics, cellular physiology and subcortical networks. *Nat Rev Neurosci.* 2002;3(8):591-605.
12. Sapolsky RM, Romero LM, Munck AU. How do glucocorticoids influence stress responses? Integrating permissive, suppressive, stimulatory, and preparative actions. *Endocr Rev.* 2000;21(1):55-89.
13. Gunnar M, Quevedo K. The neurobiology of stress and development. *Annu Rev Psychol.* 2007;58:145-73.
14. Morilak DA, Barrera G, Echevarria DJ, Garcia AS, Hernandez A, Ma S, Petre CO. Role of brain norepinephrine in the behavioral response to stress. *Prog Neuropsychopharmacol Biol Psychiatry.* 2005;29(8):1214-24.
15. Ehlers CL, Reed TK, Henriksen SJ. Effects of corticotropin-releasing factor and growth hormone-releasing factor on sleep and activity in rats. *Neuroendocrinology.* 1986;42(6):467-74.
16. Chang FC, Opp MR. Blockade of corticotropin-releasing hormone receptors reduces spontaneous waking in the rat. *Am J Physiol.* 1998;275(3 Pt 2):R793-802.
17. Chang FC, Opp MR. IL-1 is a mediator of increases in slow-wave sleep induced by CRH receptor blockade. *Am J Physiol Regul Integr Comp Physiol.* 2000;279(3):R793-802.
18. Bradbury MJ, Dement WC, Edgar DM. Effects of adrenalectomy and subsequent corticosterone replacement on rat sleep state and EEG power spectra. *Am J Physiol.* 1998;275(2 Pt 2):R555-65.
19. Vazquez-Palacios G, Retana-Marquez S, Bonilla-Jaime H, Velazquez-Moctezuma J. Further definition of the effect of corticosterone on the sleep-wake pattern in the male rat. *Pharmacol Biochem Behav.* 2001;70(2-3):305-10.
20. Born J, Fehm HL. The neuroendocrine recovery function of sleep. *Noise Health.* 2000;2(7):25-38.
21. Roky R, Obal F, Jr., Valatx JL, Bredow S, Fang J, Pagano LP, Krueger JM. Prolactin and rapid eye movement sleep regulation. *Sleep.* 1995;18(7):536-42.
22. Roky R, Valatx JL, Jouvet M. Effect of prolactin on the sleep-wake cycle in the rat. *Neurosci Lett.* 1993;156(1-2):117-20.
23. Zhang SQ, Inoue S, Kimura M. Sleep-promoting activity of prolactin-releasing peptide (PrRP) in the rat. *Neuroreport.* 2001;12(15):3173-6.
24. Bodosi B, Obal F, Jr., Gardi J, Komlodi J, Fang J, Krueger JM. An ether stressor increases REM sleep in rats: possible role of prolactin. *Am J Physiol Regul Integr Comp Physiol.* 2000;279(5):R1590-8.
25. Marinesco S, Bonnet C, Cespuoglio R. Influence of stress duration on the sleep rebound induced by immobilization in the rat: a possible role for corticosterone. *Neuroscience.* 1999;92(3):921-33.
26. Tiba PA, Tufik S, Sucbecki D. Effects of maternal separation on baseline sleep and cold stress-induced sleep rebound in adult Wistar rats. *Sleep.*

- 2004;27(6):1146-53.
27. Koehl M, Bouyer JJ, Darnaudery M, Le Moal M, Mayo W. The effect of restraint stress on paradoxical sleep is influenced by the circadian cycle. *Brain Res.* 2002;937(1-2):45-50.
 28. Tiba PA, Palma BD, Tufik S, Suchecki D. Effects of early handling on basal and stress-induced sleep parameters in rats. *Brain Res.* 2003;975(1-2):158-66.
 29. Tiba PA, Tufik S, Suchecki D. Long lasting alteration in REM sleep of female rats submitted to long maternal separation. *Physiol Behav.* 2008;93(3):444-52.
 30. Machado RB, Tufik S, Suchecki D. Chronic stress during paradoxical sleep deprivation increases paradoxical sleep rebound: Association with prolactin plasma levels and brain serotonin content. *Psychoneuroendocrinology.* 2008;33(9):1211-24.
 31. Dewasmes G, Loos N, Delanaud S, Dewasmes D, Ramadan W. Pattern of rapid-eye movement sleep episode occurrence after an immobilization stress in the rat. *Neurosci Lett.* 2004;355(1-2):17-20.
 32. Cespuglio R, Marinesco S, Baubet V, Bonnet C, el Kafi B. Evidence for a sleep-promoting influence of stress. *Adv Neuroimmunol.* 1995;5(2):145-54.
 33. Rampin C, Cespuglio R, Chastrette N, Jouvet M. Immobilisation stress induces a paradoxical sleep rebound in rat. *Neurosci Lett.* 1991;126(2):113-8.
 34. Rechtschaffen A, Gilliland MA, Bergmann BM, Winter JB. Physiological Correlates of Prolonged Sleep-Deprivation in Rats. *Science.* 1983;221(4606):182-184.
 35. Suchecki D, Tufik S. Social stability attenuates the stress in the modified multiple platform method for paradoxical sleep deprivation in the rat. *Physiol Behav.* 2000;68(3):309-16.
 36. Suchecki D, Lobo LL, Hipolide DC, Tufik S. Increased ACTH and corticosterone secretion induced by different methods of paradoxical sleep deprivation. *J Sleep Res.* 1998;7(4):276-81.
 37. Coenen AM, van Luijtelaar EL. Stress induced by three procedures of deprivation of paradoxical sleep. *Physiol Behav.* 1985;35(4):501-4.
 38. Tobler I, Murison R, Ursin R, Ursin H, Borbely AA. The effect of sleep deprivation and recovery sleep on plasma corticosterone in the rat. *Neurosci Lett.* 1983;35(3):297-300.
 39. Machado RB, Hipolide DC, Benedito-Silva AA, Tufik S. Sleep deprivation induced by the modified multiple platform technique: quantification of sleep loss and recovery. *Brain Res.* 2004;1004(1-2):45-51.
 40. Suchecki D, Duarte Palma B, Tufik S. Sleep rebound in animals deprived of paradoxical sleep by the modified multiple platform method. *Brain Res.* 2000;875(1-2):14-22.
 41. Suchecki D, Tiba PA, Tufik S. Hormonal and behavioural responses of paradoxical sleep-deprived rats to the elevated plus maze. *J Neuroendocrinol.* 2002;14(7):549-54.
 42. Suchecki D, Tiba PA, Tufik S. Paradoxical sleep deprivation facilitates subsequent corticosterone response to a mild stressor in rats. *Neurosci Lett.* 2002;320(1-2):45-8.
 43. Machado RB, Suchecki D, Tufik S. Comparison of the sleep pattern throughout a protocol of chronic sleep restriction induced by two methods of paradoxical sleep deprivation. *Brain Res Bull.* 2006;70(3):213-20.
 44. Altman JL, Whitehead WE, Rechtschaffen A. Effects of 5 Hours of Restraint Stress on Subsequent Sleep in Rat. *Psychonomic Science.* 1972;26(3):152-154.
 45. Lenard NR, Dunn AJ. Mechanisms and significance of the increased brain uptake of tryptophan. *Neurochem Res.* 2005;30(12):1543-8.
 46. Jorgensen HS. Studies on the neuroendocrine role of serotonin. *Dan Med Bull.* 2007;54(4):266-88.
 47. Copeland BJ, Neff NH, Hadjiconstantinou M. Enhanced dopamine uptake in the striatum following repeated restraint stress. *Synapse.* 2005;57(3):167-74.
 48. Rasheed N, Ahmad A, Pandey CP, Chaturvedi RK, Lohani M, Palit G. Differential Response of Central Dopaminergic System in Acute and Chronic Unpredictable Stress Models in Rats. *Neurochem Res.* 2009.
 49. Saavedra JM. Changes in dopamine, noradrenaline and adrenaline in specific septal and preoptic nuclei after acute immobilization stress. *Neuroendocrinology.* 1982;35(5):396-401.
 50. Meerlo P, de Bruin EA, Strijkstra AM, Daan S. A social conflict increases EEG slow-wave activity during subsequent sleep. *Physiol Behav.* 2001;73(3):331-5.
 51. Palma BD, Suchecki D, Tufik S. Differential effects of acute cold and foot-shock on the sleep of rats. *Brain Res.* 2000;861(1):97-104.
 52. Adrien J, Dugovic C, Martin P. Sleep-wakefulness patterns in the helpless rat. *Physiol Behav.* 1991;49(2):257-62.
 53. Vazquez-Palacios G, Velazquez-Moctezuma J. Effect of electric foot shocks, immobilization, and corticosterone administration on the sleep-wake pattern in the rat. *Physiol Behav.* 2000;71(1-2):23-8.
 54. Sanford LD, Fang J, Tang X. Sleep after differing amounts of conditioned fear training in BALB/cJ mice. *Behav Brain Res.* 2003;147(1-2):193-202.
 55. Pawlyk AC, Jha SK, Brennan FX, Morrison AR, Ross RJ. A rodent model of sleep disturbances in posttraumatic stress disorder: the role of context after fear conditioning. *Biol Psychiatry.* 2005;57(3):268-77.
 56. Koolhaas JM, De Boer SF, De Rutter AJ, Meerlo P, Sgoifo A. Social stress in rats and mice. *Acta Physiol Scand Suppl.* 1997;640:69-72.
 57. Bohus B, Benus RF, Fokkema DS, Koolhaas JM, Nyakas C, van Oortmerssen GA, Prins AJ, de Ruiter AJ, Scheurink AJ, Steffens AB. Neuroendocrine states and behavioral and physiological stress responses. *Prog Brain Res.* 1987;72:57-70.
 58. Meerlo P, Pragt BJ, Daan S. Social stress induces high intensity sleep in rats. *Neurosci Lett.* 1997;225(1):41-4.
 59. Plotsky PM, Owens MJ, Nemeroff CB. Psychoneuroendocrinology of depression. Hypothalamic-pituitary-adrenal axis. *Psychiatr Clin North Am.* 1998;21(2):293-307.
 60. Caldji C, Francis D, Sharma S, Plotsky PM, Meaney MJ. The effects of early rearing environment on the development of GABAA and central benzodiazepine receptor levels and novelty-induced fearfulness in the rat. *Neuropsychopharmacology.* 2000;22(3):219-29.
 61. Huot RL, Thiruvikraman KV, Meaney MJ, Plotsky PM. Development of adult ethanol preference and anxiety as a consequence of neonatal maternal separation in Long Evans rats and reversal with antidepressant treatment. *Psychopharmacology (Berl).* 2001;158(4):366-73.
 62. Plotsky PM, Meaney MJ. Early, postnatal experience alters hypothalamic corticotropin-releasing factor (CRF) mRNA, median eminence CRF content and stress-induced release in adult rats. *Brain Res Mol Brain Res.* 1993;18(3):195-200.
 63. Chang FC, Opp MR. Corticotropin-releasing hormone (CRH) as a regulator of waking. *Neurosci Biobehav Rev.* 2001;25(5):445-53.
 64. Waters WF, Adams SG, Jr., Binks P, Varnado P. Attention, stress and negative emotion in persistent sleep-onset and sleep-maintenance insomnia. *Sleep.* 1993;16(2):128-36.
 65. Larsson MR, Backstrom M, Johanson A. The interaction between baseline trait anxiety and trauma exposure as predictor of post-trauma symptoms of anxiety and insomnia. *Scand J Psychol.* 2008;49(5):447-50.
 66. Weinstock M. The long-term behavioural consequences of prenatal stress. *Neurosci Biobehav Rev.* 2008;32(6):1073-86.
 67. Dugovic C, Maccari S, Weibel L, Turek FW, Van Reeth O. High corticosterone levels in prenatally stressed rats predict persistent paradoxical sleep

- alterations. *J Neurosci*. 1999;19(19):8656-64.
68. Liebsch G, Montkowski A, Holsboer F, Landgraf R. Behavioural profiles of two Wistar rat lines selectively bred for high or low anxiety-related behaviour. *Behav Brain Res*. 1998;94(2):301-10.
 69. Neumann ID, Wigger A, Liebsch G, Holsboer F, Landgraf R. Increased basal activity of the hypothalamo-pituitary-adrenal axis during pregnancy in rats bred for high anxiety-related behaviour. *Psychoneuroendocrinology*. 1998;23(5):449-63.
 70. Lancel M, Muller-Preuss P, Wigger A, Landgraf R, Holsboer F. The CRH1 receptor antagonist R121919 attenuates stress-elicited sleep disturbances in rats, particularly in those with high innate anxiety. *J Psychiatr Res*. 2002;36(4):197-208.
 71. Bouyer JJ, Vallee M, Deminiere JM, Le Moal M, Mayo W. Reaction of sleep-wakefulness cycle to stress is related to differences in hypothalamo-pituitary-adrenal axis reactivity in rat. *Brain Res*. 1998;804(1):114-24.
 72. Opp MR. Rat strain differences suggest a role for corticotropin-releasing hormone in modulating sleep. *Physiol Behav*. 1997;63(1):67-74.
 73. Tang X, Liu X, Yang L, Sanford LD. Rat strain differences in sleep after acute mild stressors and short-term sleep loss. *Behav Brain Res*. 2005;160(1):60-71.
 74. Tang X, Yang L, Sanford LD. Rat strain differences in freezing and sleep alterations associated with contextual fear. *Sleep*. 2005;28(10):1235-44.
 75. Sanford LD, Tang X, Ross RJ, Morrison AR. Influence of shock training and explicit fear-conditioned cues on sleep architecture in mice: strain comparison. *Behav Genet*. 2003;33(1):43-58.
 76. Sanford LD, Yang L, Tang X. Influence of contextual fear on sleep in mice: a strain comparison. *Sleep*. 2003;26(5):527-40.
 77. Tang X, Xiao J, Liu X, Sanford LD. Strain differences in the influence of open field exposure on sleep in mice. *Behav Brain Res*. 2004;154(1):137-47.
 78. Meerlo P, Easton A, Bergmann BM, Turek FW. Restraint increases prolactin and REM sleep in C57BL/6J mice but not in BALB/cJ mice. *Am J Physiol Regul Integr Comp Physiol*. 2001;281(3):R846-54.
 79. Dugovic C, Solberg LC, Redei E, Van Reeth O, Turek FW. Sleep in the Wistar-Kyoto rat, a putative genetic animal model for depression. *Neuroreport*. 2000;11(3):627-31.
 80. Ivarsson M, Paterson LM, Hutson PH. Antidepressants and REM sleep in Wistar-Kyoto and Sprague-Dawley rats. *Eur J Pharmacol*. 2005;522(1-3):63-71.
 81. Allard JS, Tizabi Y, Shaffery JP, Manaye K. Effects of rapid eye movement sleep deprivation on hypocretin neurons in the hypothalamus of a rat model of depression. *Neuropeptides*. 2007;41(5):329-37.
 82. Jouvret M, Renault J. Persistence of insomnia after lesions of the nuclei of the raphe in the cat. *C R Seances Soc Biol Fil*. 1966;160(7):1461-5.
 83. Cespuoglio R, Houdouin F, Oulrich M, El Mansari M, Jouvret M. Axonal and somato-dendritic modalities of serotonin release: their involvement in sleep preparation, triggering and maintenance. *J Sleep Res*. 1992;1(3):150-156.
 84. Jouvret M. Sleep and serotonin: an unfinished story. *Neuropsychopharmacology*. 1999;21(2 Suppl):24S-27S.
 85. Garcia de Yebenes E, Li S, Pelletier G. Role of serotonin in the regulation of prolactin gene expression in the male rat as evaluated by in situ hybridization. *Brain Res Bull*. 1994;34(5):437-40.
 86. Churrua I, Portillo MP, Casis L, Gutierrez A, Macarulla MT, Echevarria E. Effects of fluoxetine administration on hypothalamic melanocortin system in obese Zucker rats. *Neuropeptides*. 2008;42(3):293-9.
 87. Wetzell W, Balschun D, Janke S, Vogel D, Wagner T. Effects of CLIP (corticotropin-like intermediate lobe peptide) and CLIP fragments on paradoxical sleep in rats. *Peptides*. 1994;15(2):237-41.
 88. Bonnet C, Leger L, Baubert V, Debilly G, Cespuoglio R. Influence of a 1 h immobilization stress on sleep states and corticotropin-like intermediate lobe peptide (CLIP or ACTH18-39, Ph-ACTH18-39) brain contents in the rat. *Brain Res*. 1997;751(1):54-63.
 89. Wetzell W, Wagner T, Vogel D, Demuth HU, Balschun D. Effects of the CLIP fragment ACTH 20-24 on the duration of REM sleep episodes. *Neuropeptides*. 1997;31(1):41-5.
 90. Balsa JA, Sanchez-Franco E, Pazos F, Lara JI, Lorenzo MJ, Maldonado G, Cacicedo L. Direct action of serotonin on prolactin, growth hormone, corticotropin and luteinizing hormone release in cocultures of anterior and posterior pituitary lobes: autocrine and/or paracrine action of vasoactive intestinal peptide. *Neuroendocrinology*. 1998;68(5):326-33.
 91. Takahashi K, Koyama Y, Kayama Y, Yamamoto M. The effects of prolactin on the mesopontine tegmental neurons. *Psychiatry Clin Neurosci*. 2000;54(3):257-8.
 92. Torner L, Karg S, Blume A, Kandasamy M, Kuhn HG, Winkler J, Aigner L, Neumann ID. Prolactin prevents chronic stress-induced decrease of adult hippocampal neurogenesis and promotes neuronal fate. *J Neurosci*. 2009;29(6):1826-33.
 93. Wilson SJ, Bailey JE, Rich AS, Adrover M, Potokar J, Nutt DJ. Using sleep to evaluate comparative serotonergic effects of paroxetine and citalopram. *Eur Neuropsychopharmacol*. 2004;14(5):367-72.
 94. Sanchez C, Brennum LT, Sturustovu S, Kreilgard M, Mork A. Depression and poor sleep: the effect of monoaminergic antidepressants in a pre-clinical model in rats. *Pharmacol Biochem Behav*. 2007;86(3):468-76.
 95. Simon GE, Heiligenstein JH, Grothaus L, Katon W, Revicki D. Should anxiety and insomnia influence antidepressant selection: a randomized comparison of fluoxetine and imipramine. *J Clin Psychiatry*. 1998;59(2):49-55.
 96. Vermetten E, Vythilingam M, Schmahl C, C DEK, Southwick SM, Charney DS, Bremner JD. Alterations in stress reactivity after long-term treatment with paroxetine in women with posttraumatic stress disorder. *Ann N Y Acad Sci*. 2006;1071:184-202.
 97. Boldrini M, Underwood MD, Hen R, Rosoklija GB, Dwork AJ, John Mann J, Arango V. Antidepressants increase neural progenitor cells in the human hippocampus. *Neuropsychopharmacology*. 2009.
 98. Jankord R, Herman JP. Limbic regulation of hypothalamo-pituitary-adrenocortical function during acute and chronic stress. *Ann N Y Acad Sci*. 2008;1148:64-73.
 99. Yang YR, Lee EH, Chiu TH. Electrophysiological and behavioral effects of Tyr-D-Arg-Phe-Sar on locus coeruleus neurons of the rat. *Eur J Pharmacol*. 1998;351(1):23-30.
 100. Vazquez-Palacios G, Retana-Marquez S, Bonilla-Jaime H, Velazquez-Moctezuma J. Stress-induced REM sleep increase is antagonized by naltrexone in rats. *Psychopharmacology (Berl)*. 2004;171(2):186-90.
 101. Morrison AR, Sanford LD, Ross RJ. The amygdala: a critical modulator of sensory influence on sleep. *Biol Signals Recept*. 2000;9(6):283-96.
 102. Sanford LD, Parris B, Tang X. GABAergic regulation of the central nucleus of the amygdala: implications for sleep control. *Brain Res*. 2002;956(2):276-84.
 103. Cano G, Mochizuki T, Saper CB. Neural circuitry of stress-induced insomnia in rats. *J Neurosci*. 2008;28(40):10167-84.
 104. Mellman TA, Pigeon WR, Nowell PD, Nolan B. Relationships between REM sleep findings and PTSD symptoms during the early aftermath of trauma. *J Trauma Stress*. 2007;20(5):893-901.
 105. Stickgold R. Of sleep, memories and trauma. *Nat Neurosci*. 2007;10(5):540-2.
 106. Arborelius L, Owens MJ, Plotsky PM, Nemeroff CB. The role of corticotropin-releasing factor in depression and anxiety disorders. *J Endocrinol*. 1999;160(1):1-12.