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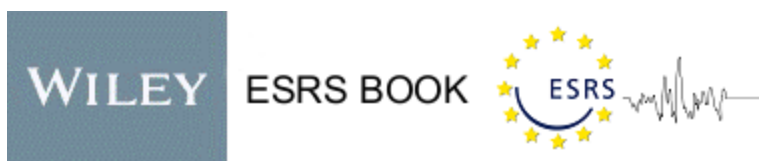
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The neurophysiology and neurobiology of paradoxical (REM) sleep

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Keywords: brainstem, melanin concentrating hormone, muscle atonia, glycine, acetylcholine, serotonin, noradrenaline, GABA, histamine, glutamate

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Abstract

Rapid eye movement (REM) sleep (or paradoxical sleep) is a state characterized by REMs, EEG activation and muscle atonia. In this review, we present the latest update on the neuronal network responsible for PS. We propose that muscle atonia during PS is due to activation of PS-on glutamatergic neurons localized in the caudal pontine sublaterodorsal tegmental nucleus (SLD) and the glycinergic/GABAergic pre-motoneurons localized in the ventro-medial medulla (vmM). The SLD neurons are inactivated during waking and slow wave sleep by PS-off GABAergic neurons localized in the ventrolateral periaqueductal gray and the adjacent deep mesencephalic reticular nucleus. PS onset and maintenance is due to the inhibition of these PS-off neurons by PS-on GABAergic neurons localized in the posterior hypothalamus, the ventrolateral periaqueductal gray and the lateral and dorsal paragigantocellular reticular nuclei. Finally, we show that only a few limbic cortical structures are activated during PS by the claustrum and the supramammillary nucleus and discuss the possible function of such activation.

Key points

- **REM sleep is produced by glutamatergic and GABA/glycinergic neurons located in the brainstem.**
- **GABA/MCH neurons located in the lateral hypothalamus control REM sleep by means of descending projections to the brainstem.**
- **A limited number of cortical and limbic areas are recruited during REM sleep.**

Brainstem network generating REM sleep**1. Glutamatergic neurons located in the SLD trigger PS**

REM sleep characterized by EEG activation and rapid eye movements (REMs) was discovered in 1953 by Aserinsky and Kleitman. It was shown to correlate in humans with dream activity (Aserinsky & Kleitman, 1953; Dement & Kleitman, 1957). In 1959, Jouvet and Michel discovered in cats that REM sleep is also characterized by a disappearance of muscle tone, paradoxically associated with a cortical activation and REMs and named this state paradoxical sleep (PS) (Jouvet & Michel, 1959; Jouvet, Michel, & Courjon, 1959). Soon after, It was shown that a state characterized by muscle atonia and REMs persists following decortication, cerebellar ablation or brain

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stem transections rostral to the pons and in the "pontine cat", a preparation in which structures rostral to the pons have been removed (Jouvet, 1962). These results suggested that brainstem structures are necessary and sufficient to trigger and maintain the state of PS. Using electrolytic and chemical lesions, it was then evidenced that the dorsal part of pontis oralis (PnO) and the pontis caudalis (PnC) nuclei contain the neurons responsible for PS onset (Jouvet, 1962). Furthermore, bilateral injections of a cholinergic agonist, carbachol, into these structures promote PS in cats. The dorsal part, where carbachol injection induces PS with the shortest latencies, was coined peri-locus coeruleus alpha nucleus (peri-LC α), pontine inhibitory area (PIA) or subcoeruleus nucleus (SubC) (Sakai, Sastre, Kanamori, & Jouvet, 1981). In contrast to data in cats, carbachol iontophoresis into the rat sublateralodorsal tegmental nucleus (SLD), the equivalent of the cat peri-LC α , induces waking with increased muscle activity (Boissard et al., 2002). Further, in rats, only a few of the numerous cFos labeled cells located in the laterodorsal tegmental nucleus (Ldt) and SLD after PS hypersomnia were cholinergic (Verret, Leger, Fort, & Luppi, 2005). Additionally, it has recently been shown that injection of scopolamine, a cholinergic muscarinic antagonist, in the SLD induces nearly no effect on PS (Grace, Vanstone, & Horner, 2014). In addition, it has been shown that cholinergic neurons of the Ldt and SLD are active both during waking and PS and therefore would play a role in cortical activation during both activated states rather than a specific role in PS genesis (Boucetta, Cisse, Mainville, Morales, & Jones, 2014). However, it was very recently shown that optogenetic activation of Ldt and pedunculopontine (PPTg) cholinergic neurons increases the probability of PS occurrence (Van Dort et al., 2014). Altogether, these results strongly suggest that the pontine cholinergic neurons play a modulatory role in PS genesis and a more central role in cortical activation both during waking and PS.

In addition, we recently showed that SLD PS-on neurons are not GABAergic. Indeed, the small number of cFos-positive neurons expressing a specific marker of GABAergic neurons (glutamate decarboxylase, GAD, enzyme of synthesis of GABA) in the SLD did not increase in rats displaying a PS rebound compared to control or PS deprived animals (Sapin et al., 2009). In contrast, it has been shown that most of the cFos-labeled neurons localized in the SLD after PS hypersomnia express a specific marker of glutamatergic neurons (vesicular glutamate transporter 2, vGlut2) (Clement, Sapin, Berod, Fort, & Luppi, 2011) and are therefore glutamatergic. Further, a large number of the PS-on neurons recorded in the SLD has been found to be glutamatergic (Boucetta et al., 2014). Confirming this hypothesis, we recently showed that inactivation of glutamatergic transmission in SLD neurons using genetic method induced a 30% decrease of PS quantities and removal of muscle atonia during the state and the occurrence of PS behavioral disorder (RBD) (Valencia Garcia et al., 2016). In addition,

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chemogenetic activation of SLD glutamatergic neurons induced muscle atonia whereas their inactivation induced RBD (Torontali, Fraigne, Sanghera, Horner, & Peever, 2019).

A number of results further indicate that SLD PS-on glutamatergic neurons generate muscle atonia via descending projections to medullary GABA/glycinergic neurons. Indeed, it has been shown that the SLD sends direct efferent projections to glycinergic neurons in the ventral (GiV) and alpha gigantocellular nuclei (Gia, corresponding to the cat magnocellular reticular nucleus, Mc) and the nucleus raphe magnus (RMg) (Valencia Garcia et al., 2016). In addition, glycinergic neurons of the Gia, GiV and RMg (forming the ventromedial medulla, vmM) express cFos after induction of PS by bicuculline (Bic, a GABA_A antagonist) injection in the SLD (Boissard et al., 2002) or PS hypersomnia (Valencia Garcia et al., 2018). Further, glycinergic neurons of vmM monosynaptically project to lumbar spinal motoneurons (Holstege & Bongers, 1991; Valencia Garcia et al., 2018). Finally, glycinergic interneurons in the spinal cord do not express cFos after PS hypersomnia (Valencia Garcia et al., 2018). These results suggest that vmM glycinergic neurons hyperpolarize motoneurons during PS. It is likely that these neurons are also GABAergic since a large majority of the cFos-labeled vmM neurons after 3h of PS recovery following 72h of PS deprivation express the mRNA of the synthesizing enzyme for GABA (GAD67mRNA) (Sapin et al., 2009). Further, it has recently been shown that combined microdialysis of bicuculline, strychnine and phaclophen (a GABA_B antagonist) in the trigeminal nucleus restored muscle tone during PS (P. Brooks & Peever, 2009; P. L. Brooks & Peever, 2012). Finally, we recently showed that inactivation of glycinergic and GABAergic transmission specifically in vmM neurons induces a state of PS without muscle atonia with abnormal and violent motor activity (Valencia Garcia et al., 2018). Altogether, these results indicate that the pre-motoneurons responsible for muscle atonia of PS are localized in the vmM and co-release GABA and glycine.

2. SLD glutamatergic neurons are inhibited by GABAergic neurons during W and SWS

A long-lasting PS-like episode can be pharmacologically induced with a short latency in head-restrained unanesthetized rats by iontophoretic applications of bicuculline or gabazine, two GABA_A receptor antagonists, specifically into the SLD. Further, PS-on neurons within the SLD are activated following bicuculline or gabazine iontophoresis (Boissard et al., 2002). Taken together, these data indicate that the onset of SLD PS-on neurons is mainly due to the removal during PS of a tonic GABAergic tone present during W and slow wave sleep (SWS). Combining retrograde tracing with cholera toxin b subunit (CTb) injected in the SLD and GAD immunostaining, we then identified neurons at the origin of the GABAergic innervation. These neurons are localized within the

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ventrolateral part of the periaqueductal gray (vlPAG) and the dorsal deep mesencephalic reticular nuclei (dDpMe) (Boissard, Fort, Gervasoni, Barbagli, & Luppi, 2003). Further, they are the only ponto-medullary structures containing a large number of cFos-positive neurons expressing GAD67mRNA after 72h of PS deprivation (Sapin et al., 2009). Injection of muscimol in the vlPAG and/or the dDpMe induces a strong increase in PS quantities (Sapin et al., 2009; Sastre, Buda, Kitahama, & Jouvet, 1996). In addition, neurochemical lesion of these two structures induces a profound increase in PS quantities (Lu, Sherman, Devor, & Saper, 2006). Further, optogenetic stimulation of PS-off GABAergic neurons within vlPAG/dDpMe almost completely suppresses PS by shortening the duration of PS episodes (Weber et al., 2015; Weber et al., 2018). Chemogenetic activation of vlPAG/dDpMe GABAergic neurons reduces PS and increases SWS. Conversely, their pharmacogenetic inhibition increases PS, mainly by increasing the number of PS episodes (Hayashi et al., 2015). Unit recording and calcium imaging revealed that most vlPAG GABAergic neurons start to be active at the end of PS episodes and decrease gradually their activity until the next PS episode. These results suggest that vlPAG GABAergic neurons powerfully gate PS and may contribute to the ultradian rhythm of PS and the SWS/PS alternation (Weber et al., 2018). Altogether, these congruent experimental data indicate that GABAergic neurons within the vlPAG/dDpMe are gating PS during W and SWS by tonically inhibiting PS-on glutamatergic neurons from the SLD.

3. Role of the monoaminergic neurons in the control of PS

A major achievement in the identification of the mechanisms controlling PS was the finding that serotonergic neurons from the raphe nuclei and noradrenergic neurons from the LC cease firing specifically during PS, i.e., show a PS-off firing activity, reciprocal to that of PS-on neurons (Hobson, McCarley, & Wyzinski, 1975). Later, it has been shown that histaminergic neurons from the tuberomammillary nucleus and hypocretinergic neurons from the perifornical hypothalamic area also depict a PS-off firing activity (M. G. Lee, Hassani, & Jones, 2005). These electrophysiological data were the basis for a well-accepted hypothesis suggesting that PS onset is gated by reciprocal inhibitory interactions between PS-on and PS-off monoaminergic neurons (Hobson et al., 1975). Supporting this neuronal model, drugs enhancing serotonin and noradrenergic transmission (monoamine oxidase inhibitors and serotonin and noradrenaline reuptake blockers) specifically suppress PS (Luppi et al., 2011). Further, applications of noradrenaline, adrenaline or benoxathian ($\alpha 2$ adrenergic agonist) into the peri-LC α inhibit PS but serotonin has no effect (Crochet & Sakai, 1999). Importantly, our recent data combining a marker of

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noradrenergic neurons (tyrosine hydroxylase, TH) and cFos staining after PS deprivation and recovery suggest that it is unlikely that the LC noradrenergic neurons are involved in the inhibition of PS, particularly during its deprivation. Indeed, the LC noradrenergic neurons do not display cFos after 72h of PS deprivation. Furthermore, no projection has been observed from the LC to the SLD (Boissard et al., 2003). Nevertheless, a substantial number of noradrenergic neurons from A1 and A2 cell groups display cFos after PS deprivation, indicating that noradrenergic neurons from these medullary cell groups might contribute to PS inhibition (Leger et al., 2008). In summary, it is clear that noradrenaline and serotonin inhibit PS but the targeted neurons remain to be identified. We propose that rather than inhibiting SLD PS-on neurons, noradrenaline and serotonin might inhibit PS by means of a tonic excitation of the dDPM/vIPAG PS-off GABAergic neurons.

4. GABAergic neurons are responsible for the inactivation of monoaminergic neurons during PS

The application of bicuculline on monoaminergic neurons during SWS or PS induces a tonic firing in serotonin and noradrenergic neurons (Gervasoni et al., 1998; Gervasoni et al., 2000). These results indicate the existence of a tonic GABA input to the LC and DRN that is active during sleep. These results strongly suggest that an increased GABA release is responsible for the PS-selective inactivation of monoaminergic neurons. By combining retrograde tracing with CTb and GAD immunohistochemistry in rats, we found that the LC and DRN receive GABAergic inputs from neurons located in a large number of distant regions from the forebrain to medulla (Gervasoni et al., 2000). Three brainstem areas contained numerous GABAergic neurons projecting both to the DRN and LC, and were thus candidates for mediating the PS-related inhibition of monoaminergic neurons: the vIPAG, the lateral and dorsal paragigantocellular nuclei (LPGi, DPGi) (Gervasoni et al., 2000). We then demonstrated by using cFos that these nuclei contain numerous LC-projecting neurons selectively activated during PS rebound following PS deprivation (Verret, Fort, Gervasoni, Leger, & Luppi, 2006). Since the LPGi/DPGi nuclei had not previously been considered in sleep mechanisms, we studied the firing activity of their neurons across the sleep-wake cycle in head-restrained rats (Goutagny et al., 2008; Sirieix, Gervasoni, Luppi, & Leger, 2012). In full agreement with our functional data using cFos, we found that the LPGi/DPGi contain numerous PS-on neurons that are silent during waking and SWS and fire tonically during PS. These PS-on neurons start discharging approximately 15 sec before PS onset and become silent around 10 sec before EEG signs of arousal. Further, inactivation of the DPGi with muscimol induced waking and cFos expression in LC and also A1 A2 and

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A5 noradrenergic neurons suggesting that GABAergic neurons of the DPGi inhibit the LC but also all the other brainstem noradrenergic neurons. Interestingly, a sub-population of Calbindin-D28K- glutamatergic neurons located in the DPGi are also active during PS, project to the three contralateral eye-muscle nuclei and augment their firing prior to the onset of REMs during PS. Optogenetic activation of these neurons triggers eye movements selectively during PS, while their genetic ablation or optogenetic silencing suppresses them (Gutierrez Herrera et al., 2019). None of these perturbations led to a change in the duration of PS episodes. These results indicate that the DPGi contains both inhibitory GABA and excitatory glutamatergic PS-active neurons.

In addition, it has been recently shown that optogenetic stimulation during SWS of GABAergic LPGi neurons or of their ascending fibers located in the vIPAG/dDPMe induced PS (Weber et al., 2015). It raises the possibility that LPGi GABAergic neurons also inhibit the GABAergic PS-off neurons in addition to the monoaminergic neurons. However, combining CTb injection in the vIPAG/dDPMe with cFos staining after PS hypersomnia, we did not find double-labeled cells in the LPGi (Clement et al., 2012). Taken together, these data strongly suggest that the LPGi and DPGi contain the neurons responsible for the inactivation of LC noradrenergic and the DRN serotonergic neurons during PS (Goutagny et al., 2008; Sirieix et al., 2012). A contribution from the vIPAG in this inhibitory mechanism is also likely. Indeed, an increase in cFos/GAD immunoreactive neurons has been reported in the vIPAG after a PS rebound induced by deprivation in rats (Sapin et al., 2009). In summary, a large body of data indicates that GABAergic PS-on neurons localized in the vIPAG and the LPGi/DPGi hyperpolarize the monoaminergic neurons during PS.

5. Role of the hypothalamus in PS control

Surprisingly, a very large number of cFos-positive cells was observed in the posterior hypothalamus (PH), including zona incerta (ZI), perifornical area (PeF) and the lateral hypothalamic area (LHA) after PS hypersomnia (Verret et al., 2003). By using double-immunostaining, it has been further shown that around 75% of PH cells labeled for cFos after PS rebound express GAD67 mRNA and are likely GABAergic (Sapin et al., 2010). One third of these neurons were also immunoreactive for two neuropeptides, melanin concentrating hormone (MCH)(Verret et al., 2003) and nesfatin-1 (Jego et al., 2012). Further, it has been confirmed by unit recordings in head-restrained rats that MCH neurons fire exclusively during PS (Hassani, Lee, & Jones, 2009). Using calcium imaging, it has been recently confirmed that MCH neurons are strongly active during PS and not during SWS and quiet waking. Interestingly, the authors also reported activation of 70% of the MCH neurons during the exploration of new objects during active waking (Blanco-Centurion et al., 2019; Gonzalez, Iordanidou, Strom, Adamantidis,

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& Burdakov, 2016; Kosse & Burdakov, 2019). Rats receiving intracerebroventricular (icv) administration of MCH showed a strong dose-dependent increase in PS and, to a minor extent, SWS quantities (Verret et al., 2003). Such sleep increases were also obtained using chronic (24h) optogenetic activation of MCH neurons (Konadhode et al., 2013). Optogenetic activation of MCH neurons specifically at the onset of SWS epoch did not increase SWS duration but increase the probability of SWS-to-PS transitions. Similar results were reproduced by using a chemogenetic approach to modulate the firing activity of MCH neurons (Varin, Luppi, & Fort, 2018). More importantly, optogenetic stimulation of MCH neurons at PS onset significantly prolonged the duration of PS episodes (Jego et al., 2013). It was also recently shown that the increase in PS quantities induced by a rise in temperature was abolished when inactivating the MCH neurons by optogenetic (Komagata et al., 2019). Based on the finding that MCH neurons express GAD67, it was first thought that they are GABAergic and thus inhibitory in nature. However, three independent studies demonstrated that they do not express vGAT but rather vGluT2, the glutamate transporter (Mickelsen et al., 2019; Schneeberger et al., 2018) and that they release glutamate to excite neurons in lateral septum (Chee, Arrigoni, & Maratos-Flier, 2015).

Besides, it was shown that a large population of MCH-negative vGAT-positive neurons in PH discharge maximally during PS (Hassani, Henny, Lee, & Jones, 2010). Since these neurons anticipate PS onset, they could play a role in triggering the state. GABAergic neurons co-expressing Lhx6 localized in the ZI could constitute a subpopulation of these MCH-/vGAT+ PS-on neurons. Indeed, it has been shown that chemogenetic activation of Lhx6+ vGAT+ neurons induces an increase in PS whereas their inhibition decreases it (Liu et al., 2017). Further, it was shown that they are activated during PS hypersomnia (H. S. Lee et al., 2020).

To determine whether PH neurons indeed play a crucial role in PS, muscimol was bilaterally injected in this area. It induced a total inhibition of PS and an increase in SWS quantities. In the same experiments, the vIPAG/dDpMe region was shown to contain a large number of cFos/GAD67+ and of cFos/CTb+ neurons in animals with a CTb injection in the SLD. These results indicate that the activation of PH neurons is a necessary step for PS to occur. They further suggest that MCH+/vGluT2+ and MCH-/vGAT+ PS-on neurons control PS onset and maintenance by means of direct or indirect inhibitory projections to vIPAG/dDpMe PS-off GABAergic neurons. From these results, it can be proposed that these MCH+/ vGluT2+ and MCH-/vGAT+ neurons of the PH constitute a “master” generator of PS that controls a “slave” generator located in the brainstem. To reconcile Jouvet’s hypothesis (i.e. brainstem is necessary and sufficient to generate a state characterized by muscle atonia and REMs; (Jouvet, 1962) with these results, it can therefore be proposed that after removal of the forebrain, the brainstem generator is sufficient to induce a state with muscle atonia and REMs by means of a reorganization of the brainstem systems

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generating PS. However, the brainstem generator would be under control of the PH generator in intact animals.

In addition to the descending pathway to the PS-off GABAergic neurons, the MCH PS-on neurons might also promote PS by means of their projections to the histaminergic, monoaminergic and hypocretinergic PS-off neurons (Boissard et al., 2002; Jégo et al., 2013). Indeed, Jégo et al (2013) showed that optogenetic activation of MCH fibers in the TMN inhibit histaminergic cells through activation of GABA_A receptor. It was also shown that stimulation of MCH fibers in CA1 field of the hippocampus induced IPSP in local pyramidal cells (Izawa et al., 2019). These results in line with the initial hypothesis that MCH neurons are GABAergic are contradictory to recent results showing that they are glutamatergic and thus excitatory. One possibility is that they excite local inhibitory interneurons.

The mechanisms at the origin of the activation of the MCH/vGluT2+ neurons of the PH at the entrance into PS remain to be identified. A large number of studies indicate that MCH neurons also play a key role in metabolism control (Qu et al., 1996; Schneeberger et al., 2018). Therefore, the activation of these neurons at the onset and during PS could be influenced by the metabolic state. In addition, it is likely that yet undiscovered endogenous cellular or molecular clock-like mechanisms may play a role in their activation.

The cessation of activity of the MCH/vGluT2+ PS-on neurons and more largely of all the PS-on neurons at the end of PS episodes may be due to a different mechanism than the entrance into the state, possibly the reactivation of the arousal circuits, which are known to silence MCH neurons *in-vitro* (van den Pol, Acuna-Goycolea, Clark, & Ghosh, 2004). Indeed, animals enter PS slowly from SWS, while in contrast they exit from it abruptly by a micro-arousal. This indicates that the end of PS episodes is induced by the activation of the waking systems like the monoaminergic, hypocretin or histaminergic neurons. However, the precise mechanisms responsible for their activation remain to be identified.

6 . Cortical activation during PS

PET imaging in humans first pointed out that some cortical structures are more and other less activated during PS than during waking. It has indeed been shown that limbic and paralimbic areas and posterior cortices are more activated whereas the inferior and dorsolateral prefrontal cortices are less activated during PS compared to quiet waking (Braun et al., 1997; Maquet et al., 1996; Nofzinger, Mintun, Wiseman, Kupfer, & Moore, 1997). In the first few studies examining cortical state during PS in animals, it has been confirmed using cFos or C14 deoxyglucose that the cingulate cortex, hippocampus and amygdala are more active during PS than during waking

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(Lydic et al., 1991; Sastre, Buda, Lin, & Jouvet, 2000). Further, an increase in cyclic adenosine monophosphate (cAMP), phospho-mitogen-activated protein kinase (pMAPK), and phospho-cAMP response element-binding protein (pCREB) has been seen in CA1 and dentate gyrus during PS compared to waking and SWS (Luo, Phan, Yang, Garelick, & Storm, 2013). Theta oscillation occurs both during active waking and PS. Montgomery et al. (2008) found by coherence analysis that dentate/CA3 theta and gamma synchrony was significantly higher during PS compared with active waking and that, in contrast, gamma power in CA1 and CA3–CA1 gamma coherence showed significant decreases during PS. These results suggested that theta oscillation is not produced by the same mechanisms during active waking and PS. To determine the state of the cortex during PS at cellular level, we mapped the cortical expression of cFos, Bdnf and Arc in control rats, rats deprived of PS for 72h and rats allowed to recover and displaying PS hypersomnia during 6h. We also compared the distribution of cFos-labeled neurons in these conditions with a 3h period of waking (Renouard et al., 2015). We found out that, during PS hypersomnia compared to control and PS deprivation, the dentate gyrus, claustrum, parasubiculum, presubiculum, cortical amygdaloid nucleus, anterior cingulate, medial entorhinal and retrosplenial cortices were the only cortical structures containing neurons with an increased expression of Bdnf, cFos and/or Arc, known markers of activation and/or synaptic plasticity (Renouard et al., 2015). Further, the dentate gyrus was the only cortical structure containing more cFos-labeled neurons during PS hypersomnia than during waking (Renouard et al., 2015). In contrast, the number of cFos-labeled neurons was very low in the somatosensory cortices after PS hypersomnia compared to PS deprivation and furthermore waking indicating that these structures are rather deactivated during PS (Renouard et al., 2015). These results are in line with recent findings of the occurrence of slow waves in middle and superficial layers of primary sensory and motor cortices (Funk, Honjoh, Rodriguez, Cirelli, & Tononi, 2016). They fit also with data showing decreased pyramidal cell activity in the somatosensory cortices during PS compared to SWS and waking (Niethard et al., 2016). Such decrease was accompanied by activation of parvalbumin inhibitory interneurons.

In summary, recent results strongly suggest that only a few limbic cortical structures are strongly activated during PS whereas most of the other cortices in particular the somatosensory ones are rather deactivated.

7 . Subcortical structures activating the cortex during PS

It was first demonstrated that theta oscillations and neocortical desynchrony characteristics of PS are absent in atropinized rats and cats suggesting that cholinergic neurons play a key role in cortical activation during PS

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(Robinson, Kramis, & Vanderwolf, 1977). It was then shown that cholinergic neurons in the basal forebrain directly project to the neocortex and hippocampus while those located in Ldt and PPTg pontine nuclei project to the thalamus, hypothalamus and basal forebrain (Jones, 1993). Then, it has been recently shown that PPT and Ldt cholinergic neurons are maximally active during waking and PS in positive correlation with fast (gamma) cortical activity (Boucetta et al., 2014). However, optogenetic excitation of PPT and Ldt cholinergic neurons promotes PS but did not modify spectrally the EEG (Van Dort et al., 2014). In addition, specific chemogenetic activation of PPT cholinergic neurons did not modify PS or the EEG spectral composition (Kroeger et al., 2017). However, continuous light pulse stimulation during SWS evoked tonic spiking in cholinergic PPT and Ldt neurons that elicited a shift from irregular slow wave activity to rhythmic theta and enhanced gamma activity on the cortex without behavioral arousal (Cisse et al., 2018). It has been also demonstrated that basal forebrain cholinergic neurons depict bursty discharge in phase with theta during active waking and PS while they virtually cease firing during SWS (M. G. Lee, Hassani, Alonso, & Jones, 2005; Xu et al., 2015). Importantly, GABAergic and glutamatergic neurons located in the basal forebrain intermingled with the cholinergic neurons were also shown to be more active during PS and waking than SWS and to be phased locked with theta (Manns, Alonso, & Jones, 2000; Xu et al., 2015). Surprisingly, medial septum neurons projecting to the dentate gyrus do not express cFos after PS hypersomnia (Renouard et al., 2015) in contrast to those of the supramammillary nucleus (see below). This might be due to the fact that they are not selectively activated during PS or that their mode of discharge does not trigger cFos expression. Nevertheless, it was recently shown that silencing by optogenetic the GABAergic neurons of the medial septum during PS dramatically reduces theta power (Boyce, Glasgow, Williams, & Adamantidis, 2016). In contrast, silencing of cholinergic or GABAergic basal forebrain neurons did not induce a decrease in theta power or cortical desynchrony during PS (Anacleit et al., 2015; Shi, Han, Su, Yang, & Yu, 2015). In summary, it seems clear that GABAergic neurons of the medial septum play a key role in theta generation during PS while glutamatergic and cholinergic neurons (from the basal forebrain and the pons) do not seem to play a crucial role in cortical activation during PS.

In addition, we previously showed that SLD neurons directly project to the intralaminar thalamic nuclei which also contain cFos-labeled neurons after 90 minutes of SLD pharmacological activation inducing PS-like state (Boissard et al., 2002). Based on these results, we proposed that glutamatergic neurons of the SLD are specifically active during PS and contribute to cortical activation during this state by means of their projection to the thalamus. However, we recently showed that SLD neurons ascending to the thalamus and intralaminar thalamic neurons themselves are not cFos-positive after PS hypersomnia (Renouard et al., 2015; Valencia Garcia et al., 2016).

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Further, it has been shown that glutamate SLD neurons do not project to the intralaminar thalamic nucleus or the basal forebrain (Krenzer et al., 2011). These results cast a strong doubt on our hypothesis that glutamate SLD neurons activate the cortex through their projection to the intralaminar thalamic nuclei during PS. In contrast, we found out that the claustrum was the only subcortical structure containing a large number of cFos-positive neurons projecting to the anterior cingulate and retrosplenial cortices after PS hypersomnia (Renouard et al., 2015). In addition, we showed that the lateral supramammillary nucleus (SumL) was the only subcortical structure containing cFos-positive neurons projecting to the dorsal part of the dentate gyrus (Renouard et al., 2015) while the medial entorhinal cortex was the only structure containing a large number of cFos+ neurons projecting to the ventral part of the DG (Billwiller, Renouard, Clement, Fort, & Luppi, 2016). After lesion of the entire Sum, cFos+ neurons were no longer visible in the dorsal but not in the ventral DG after PS hypersomnia and theta power was decreased during PS (Billwiller et al., 2016). Our results and those of others also recently demonstrated that SumL neurons activated during PS hypersomnia and projecting to the DG display a dual glutamatergic and GABAergic phenotype (Billwiller et al., 2016; Soussi, Zhang, Tahtakran, Houser, & Esclapez, 2010). Based on these results, it has been proposed that SumL neurons could simultaneously hyperpolarize the soma and depolarize the dendrites of dentate granule neurons (Soussi et al., 2010). The release of glutamate on the dendrites of granule neurons during PS could activate NMDA receptors and initiate the intracellular cascade inducing the expression of genes involved in synaptic plasticity like cFos, Bdnf, Arc and Cox2. The fact that only a subset of granule cells expresses these proteins during PS hypersomnia suggest that these neurons might receive additional excitatory input during PS from other afferent structures such as the medial entorhinal cortex. It has also recently been shown that glutamatergic neurons located in the claustrum strongly project to the medial entorhinal cortex (Kitanishi & Matsuo, 2017). It is therefore likely that the claustrum is also responsible for the direct and indirect activation of the medial entorhinal cortex and the ventral dentate gyrus although it remains to be demonstrated.

Altogether, these results indicate that GABAergic neurons of the medial septum induce theta oscillation in hippocampal neurons during PS while those of the SumL activate specifically dorsal DG granule cells and increase theta pacing. They also strongly suggest that glutamatergic neurons of the claustrum activate anterior cingulate and retrosplenial cortices and also likely the medial entorhinal cortex during PS.

8 . Function (s) of the limbic cortical activation during PS

The cortical structures activated during PS belong to the limbic system and have been implicated in spatial and emotional memory in rodents. The retrosplenial, medial entorhinal and anterior cingulate cortices and the dentate

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gyrus have all been shown to play crucial roles in spatial learning in rodents (Cho & Sharp, 2001; Czajkowski et al., 2014; Frankland, Bontempi, Talton, Kaczmarek, & Silva, 2004; Katche, Dorman, Slipczuk, Cammarota, & Medina, 2013; Kheirbek et al., 2013; Takehara-Nishiuchi, 2014). Further, these structures are densely interconnected (van Strien, Cappaert, & Witter, 2009; Wyss & Van Groen, 1992). The firing properties of neurons of these cortical structures in behaving animals are also consistent with such hypothesis. Indeed, they contain place cells, head direction cells, grid cells and/or boundary cells, each of which has a characteristic firing pattern that encodes spatial parameters relating to the animal's current position and orientation (Hartley, Lever, Burgess, & O'Keefe, 2014). In view of these and our results, the activation of neurons from these structures during PS is likely involved in contextual memory consolidation. Supporting such an hypothesis, optogenetic inhibition during PS of GABAergic neurons in the medial septum known to project to the hippocampus, medial entorhinal and retrosplenial cortices (Unal, Joshi, Viney, Kis, & Somogyi, 2015), strongly decreased theta power and induced contextual memory impairment (Boyce et al., 2016). Surprisingly, it was also recently shown that activation or inhibition of MCH neurons impaired or improved hippocampus-dependent memory, respectively suggesting that the activation of MCH neurons during PS induces forgetting of hippocampus-dependent memories (Izawa et al., 2019). It is now necessary to determine whether neurons of the limbic structures are activated both during learning tasks and subsequent PS episodes and also to establish their specific role. Dreaming also could be due to the coordinated activation of neurons of these structures and imagery in humans could provide evidence supporting such hypothesis.

9. A network model for PS onset and maintenance (Fig. 1)

As described above, most of the populations of neurons responsible for PS were identified by means of cFos labeling induced by PS deprivation and PS hypersomnia. In the future, it will be important to employ additional experimental approaches to fully determine their respective role, including tract-tracing, single unit recordings, inactivation and activation by genetic or pharmacological tools. Furthermore, several regions that contain a large number of cFos-labeled neurons require additional studies, including the lateral paraventricular nucleus, the lateral parabrachial nucleus, the nucleus raphe obscurus and the dorsal PAG (Verret et al., 2005).

The observation that PS episodes in the rat start from SWS after a relatively long intermediate state during which the EEG displays a mix of spindles and theta activity, and then terminate abruptly by a short microarousal deserves further attention. These findings suggest that different mechanisms are responsible for the entrance into and the exit from PS. Altogether, these characteristics, as well as our current knowledge of the neuronal network, lead us

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to propose an updated model of the mechanisms controlling PS onset and maintenance.

The onset of PS would be due to the activation by intrinsic "clock like" mechanisms of PS-on MCH+/vGluT2+ and MCH-/vGAT+ hypothalamic neurons and PS-on GABAergic neurons localized in the LPGi/DPGi and vIPAG. These neurons would inactivate the vIPAG/dDpMe PS-off GABAergic neurons and the aminergic and Hcrt waking neurons. Descending PS-on SLD neurons would induce muscle atonia via their excitatory projections to glycinergic/GABA neurons localized in the vmM. Cortical activation limited to the retrosplenial, anterior cingulate and medial entorhinal cortices and the dentate gyrus would be induced by projections from the claustrum and the lateral supramammillary nucleus. The exit from PS would be due to the activation of waking systems. The waking systems would inhibit the MCH+/vGAT+ neurons and the brainstem GABAergic PS-on neurons.

For Review Only

Legends of the Figures

Figure 1 : State of the network responsible for PS

During W, PS-off GABAergic neurons located in the vIPAG and dDPMe tonically inhibits SLD glutamatergic neurons. These PS-off GABAergic neurons are excited by monoaminergic and hypocretin inputs. W-on neurons activate the cortex either directly or by means of a relay in the intralaminar thalamic nuclei. The onset and maintenance of PS is due to the intrinsic "clock-like" activation of GABAergic and glutamatergic neurons localized in the posterior hypothalamus (part of them containing also MCH), the vIPAG/dDPMe and the DPGi. These neurons inhibit all W neurons and the PS-off GABAergic neurons of the vIPAG and dDPMe. SLD glutamatergic neurons are disinhibited and start to generate muscle atonia by means of their descending projections. They excite glycinergic/GABAergic neurons of the GiV which in turns hyperpolarize spinal and cranial motoneurons. Direct pathways from the noradrenergic and serotonergic neurons to the cortex contributing to W are not represented for sake of clarity of the figure.

Abbreviations: 5HT, serotonin; Ach, acetylcholine; BF, Basal Forebrain, DPGi, dorsal paragigantocellular reticular nucleus; dDPMe, dorsal deep mesencephalic reticular nucleus; DRN, dorsal raphe nucleus; GiV, ventral gigantocellular reticular nucleus; Glu, glutamate; Gly, glycine; Hcrt, hypocretin- (orexin) containing neurons; His, histamine; LC, locus coeruleus; Ldt, laterodorsal tegmental nucleus, LPGi, lateral paragigantocellular reticular nucleus; MCH, melanin concentrating hormone; PH, posterior hypothalamus; PPT, pedunculo pontine nucleus; vIPAG, ventrolateral periaqueductal gray; VLPO, ventrolateral preoptic nucleus; SCN, suprachiasmatic nucleus; SLD, sublaterodorsal nucleus. Add definitions for the following mENT, RSC, dDG & vDG, ACA, CLA, and, SumL

References

- Anaclet, C., Pedersen, N. P., Ferrari, L. L., Venner, A., Bass, C. E., Arrigoni, E., & Fuller, P. M. (2015). Basal forebrain control of wakefulness and cortical rhythms. *Nat Commun*, 6, 8744. doi:10.1038/ncomms9744
- Aserinsky, E., & Kleitman, N. (1953). Regularly occurring periods of eye motility and concomitant phenomena during sleep. *Science*, 118, 273-274.

- Billwiller, F., Renouard, L., Clement, O., Fort, P., & Luppi, P. H. (2016). Differential origin of the activation of dorsal and ventral dentate gyrus granule cells during paradoxical (REM) sleep in the rat. *Brain Struct Funct.* doi:10.1007/s00429-016-1289-7
- Blanco-Centurion, C., Luo, S., Spergel, D. J., Vidal-Ortiz, A., Oprisan, S. A., Van den Pol, A. N., . . . Shiromani, P. J. (2019). Dynamic Network Activation of Hypothalamic MCH Neurons in REM Sleep and Exploratory Behavior. *J Neurosci*, *39*(25), 4986-4998. doi:10.1523/JNEUROSCI.0305-19.2019
- Boissard, R., Fort, P., Gervasoni, D., Barbagli, B., & Luppi, P. H. (2003). Localization of the GABAergic and non-GABAergic neurons projecting to the sublaterodorsal nucleus and potentially gating paradoxical sleep onset. *Eur J Neurosci*, *18*(6), 1627-1639.
- Boissard, R., Gervasoni, D., Schmidt, M. H., Barbagli, B., Fort, P., & Luppi, P. H. (2002). The rat ponto-medullary network responsible for paradoxical sleep onset and maintenance: a combined microinjection and functional neuroanatomical study. *Eur J Neurosci*, *16*(10), 1959-1973.
- Boucetta, S., Cisse, Y., Mainville, L., Morales, M., & Jones, B. E. (2014). Discharge profiles across the sleep-waking cycle of identified cholinergic, GABAergic, and glutamatergic neurons in the pontomesencephalic tegmentum of the rat. *J Neurosci*, *34*(13), 4708-4727. doi:10.1523/JNEUROSCI.2617-13.2014
- Boyce, R., Glasgow, S. D., Williams, S., & Adamantidis, A. (2016). Causal evidence for the role of REM sleep theta rhythm in contextual memory consolidation. *Science*, *352*(6287), 812-816. doi:10.1126/science.aad5252
- Braun, A. R., Balkin, T. J., Wesenten, N. J., Carson, R. E., Varga, M., Baldwin, P., . . . Herscovitch, P. (1997). Regional cerebral blood flow throughout the sleep-wake cycle. An H2(15)O PET study. *Brain*, *120* (Pt 7), 1173-1197.
- Brooks, P., & Peever, J. (2009). Role for GABAB-mediated inhibition in the control of somatic motoneurons during REM sleep. *SfN (Abstract)*.
- Brooks, P. L., & Peever, J. H. (2012). Identification of the transmitter and receptor mechanisms responsible for REM sleep paralysis. *J Neurosci*, *32*(29), 9785-9795. doi:10.1523/JNEUROSCI.0482-12.2012
- Chee, M. J., Arrigoni, E., & Maratos-Flier, E. (2015). Melanin-concentrating hormone neurons release glutamate for feedforward inhibition of the lateral septum. *J Neurosci*, *35*(8), 3644-3651. doi:10.1523/JNEUROSCI.4187-14.2015
- Cho, J., & Sharp, P. E. (2001). Head direction, place, and movement correlates for cells in the rat retrosplenial cortex. *Behav Neurosci*, *115*(1), 3-25.
- Cisse, Y., Toossi, H., Ishibashi, M., Mainville, L., Leonard, C. S., Adamantidis, A., & Jones, B. E. (2018). Discharge and Role of Acetylcholine Pontomesencephalic Neurons in Cortical Activity and Sleep-Wake States Examined by Optogenetics and Juxtacellular Recording in Mice. *eNeuro*, *5*(4). doi:10.1523/ENEURO.0270-18.2018
- Clement, O., Sapin, E., Berod, A., Fort, P., & Luppi, P. H. (2011). Evidence that Neurons of the Sublaterodorsal Tegmental Nucleus Triggering Paradoxical (REM) Sleep Are Glutamatergic. *Sleep*, *34*(4), 419-423.
- Clement, O., Sapin, E., Libourel, P. A., Arthaud, S., Brischoux, F., Fort, P., & Luppi, P. H. (2012). The Lateral Hypothalamic Area Controls Paradoxical (REM) Sleep by Means of Descending Projections to Brainstem GABAergic Neurons. *J Neurosci*, *32*(47), 16763-16774. doi:10.1523/JNEUROSCI.1885-12.2012
- Crochet, S., & Sakai, K. (1999). Alpha-2 adrenoceptor mediated paradoxical (REM) sleep inhibition in the cat. *Neuroreport*, *10*(10), 2199-2204.
- Czajkowski, R., Jayaprakash, B., Wiltgen, B., Rogerson, T., Guzman-Karlsson, M. C., Barth, A. L., . . . Silva, A. J. (2014). Encoding and storage of spatial information in the

- retrosplenial cortex. *Proc Natl Acad Sci U S A*, 111(23), 8661-8666. doi:10.1073/pnas.1313222111
- Dement, W., & Kleitman, N. (1957). The relation of eye movements during sleep to dream activity: an objective method for the study of dreaming. *Journal of Experimental Psychology. Learning, Memory, and Cognition*, 53, 339-346.
- Frankland, P. W., Bontempi, B., Talton, L. E., Kaczmarek, L., & Silva, A. J. (2004). The involvement of the anterior cingulate cortex in remote contextual fear memory. *Science*, 304(5672), 881-883. doi:10.1126/science.1094804
- 304/5672/881 [pii]
- Funk, C. M., Honjoh, S., Rodriguez, A. V., Cirelli, C., & Tononi, G. (2016). Local Slow Waves in Superficial Layers of Primary Cortical Areas during REM Sleep. *Curr Biol*, 26(3), 396-403. doi:10.1016/j.cub.2015.11.062
- Gervasoni, D., Darracq, L., Fort, P., Souliere, F., Chouvet, G., & Luppi, P. H. (1998). Electrophysiological evidence that noradrenergic neurons of the rat locus coeruleus are tonically inhibited by GABA during sleep. *Eur J Neurosci*, 10(3), 964-970.
- Gervasoni, D., Peyron, C., Rampon, C., Barbagli, B., Chouvet, G., Urbain, N., . . . Luppi, P. H. (2000). Role and origin of the GABAergic innervation of dorsal raphe serotonergic neurons. *J Neurosci*, 20(11), 4217-4225.
- Gonzalez, J. A., Iordanidou, P., Strom, M., Adamantidis, A., & Burdakov, D. (2016). Awake dynamics and brain-wide direct inputs of hypothalamic MCH and orexin networks. *Nat Commun*, 7, 11395. doi:10.1038/ncomms11395
- Goutagny, R., Luppi, P. H., Salvert, D., Lapray, D., Gervasoni, D., & Fort, P. (2008). Role of the dorsal paragigantocellular reticular nucleus in paradoxical (rapid eye movement) sleep generation: a combined electrophysiological and anatomical study in the rat. *Neuroscience*, 152(3), 849-857. doi:S0306-4522(07)01555-2 [pii]
- 10.1016/j.neuroscience.2007.12.014
- Grace, K. P., Vanstone, L. E., & Horner, R. L. (2014). Endogenous Cholinergic Input to the Pontine REM Sleep Generator Is Not Required for REM Sleep to Occur. *J Neurosci*, 34(43), 14198-14209. doi:10.1523/JNEUROSCI.0274-14.2014
- Gutierrez Herrera, C., Girard, F., Bilella, A., Gent, T. C., Rocco-Waldmeyer, D. M., Adamantidis, A., & Celio, M. R. (2019). Neurons in the Nucleus papilio contribute to the control of eye movements during REM sleep. *Nat Commun*, 10(1), 5225. doi:10.1038/s41467-019-13217-y
- Hartley, T., Lever, C., Burgess, N., & O'Keefe, J. (2014). Space in the brain: how the hippocampal formation supports spatial cognition. *Philos Trans R Soc Lond B Biol Sci*, 369(1635), 20120510. doi:10.1098/rstb.2012.0510
- Hassani, O. K., Henny, P., Lee, M. G., & Jones, B. E. (2010). GABAergic neurons intermingled with orexin and MCH neurons in the lateral hypothalamus discharge maximally during sleep. *Eur J Neurosci*. doi:EJN7295 [pii]
- 10.1111/j.1460-9568.2010.07295.x
- Hassani, O. K., Lee, M. G., & Jones, B. E. (2009). Melanin-concentrating hormone neurons discharge in a reciprocal manner to orexin neurons across the sleep-wake cycle. *Proc Natl Acad Sci U S A*, 106(7), 2418-2422. doi:0811400106 [pii]
- 10.1073/pnas.0811400106
- Hayashi, Y., Kashiwagi, M., Yasuda, K., Ando, R., Kanuka, M., Sakai, K., & Itohara, S. (2015). Cells of a common developmental origin regulate REM/non-REM sleep and wakefulness in mice. *Science*, 350(6263), 957-961. doi:10.1126/science.aad1023
- Hobson, J. A., McCarley, R. W., & Wyzinski, P. W. (1975). Sleep cycle oscillation: reciprocal discharge by two brainstem neuronal groups. *Science*, 189(4196), 55-58.

- Holstege, J. C., & Bongers, C. M. (1991). A glycinergic projection from the ventromedial lower brainstem to spinal motoneurons. An ultrastructural double labeling study in rat. *Brain Res*, 566(1-2), 308-315.
- Izawa, S., Chowdhury, S., Miyazaki, T., Mukai, Y., Ono, D., Inoue, R., . . . Yamanaka, A. (2019). REM sleep-active MCH neurons are involved in forgetting hippocampus-dependent memories. *Science*, 365(6459), 1308-1313. doi:10.1126/science.aax9238
- Jego, S., Glasgow, S. D., Herrera, C. G., Ekstrand, M., Reed, S. J., Boyce, R., . . . Adamantidis, A. R. (2013). Optogenetic identification of a rapid eye movement sleep modulatory circuit in the hypothalamus. *Nat Neurosci*. doi:10.1038/nn.3522
- Jego, S., Salvert, D., Renouard, L., Mori, M., Goutagny, R., Luppi, P. H., & Fort, P. (2012). Tuberal Hypothalamic Neurons Secreting the Satiety Molecule Nesfatin-1 Are Critically Involved in Paradoxical (REM) Sleep Homeostasis. *PLoS ONE*, 7(12), e52525. doi:10.1371/journal.pone.0052525
- Jones, B. E. (1993). The organization of central cholinergic systems and their functional importance in sleep-waking states. *Prog Brain Res*, 98, 61-71.
- Jouvet, M. (1962). Recherches sur les structures nerveuses et les mécanismes responsables des différentes phases du sommeil physiologique. *Arch Ital Biol*, 100, 125-206.
- Jouvet, M., & Michel, F. (1959). Corrélations électromyographiques du sommeil chez le chat décortiqué et mésencéphalique chronique. *CR Soc. Biol.*, 153, 422-425.
- Jouvet, M., Michel, F., & Courjon, J. (1959). Sur un stade d'activité électrique cérébrale rapide au cours du sommeil physiologique. *CR Seances Soc Biol*, 153, 1024-1028.
- Katche, C., Dorman, G., Slipczuk, L., Cammarota, M., & Medina, J. H. (2013). Functional integrity of the retrosplenial cortex is essential for rapid consolidation and recall of fear memory. *Learn Mem*, 20(4), 170-173. doi:10.1101/lm.030080.112
- Kheirbek, M. A., Drew, L. J., Burghardt, N. S., Costantini, D. O., Tannenholz, L., Ahmari, S. E., . . . Hen, R. (2013). Differential control of learning and anxiety along the dorsoventral axis of the dentate gyrus. *Neuron*, 77(5), 955-968. doi:10.1016/j.neuron.2012.12.038
- Kitanishi, T., & Matsuo, N. (2017). Organization of the Claustrum-to-Entorhinal Cortical Connection in Mice. *J Neurosci*, 37(2), 269-280. doi:10.1523/JNEUROSCI.1360-16.2017
- Komagata, N., Latifi, B., Rusterholz, T., Bassetti, C. L. A., Adamantidis, A., & Schmidt, M. H. (2019). Dynamic REM Sleep Modulation by Ambient Temperature and the Critical Role of the Melanin-Concentrating Hormone System. *Curr Biol*, 29(12), 1976-1987 e1974. doi:10.1016/j.cub.2019.05.009
- Konadhode, R. R., Pelluru, D., Blanco-Centurion, C., Zayachivsky, A., Liu, M., Uhde, T., . . . Shiromani, P. J. (2013). Optogenetic stimulation of MCH neurons increases sleep. *J Neurosci*, 33(25), 10257-10263. doi:10.1523/JNEUROSCI.1225-13.2013
- Kosse, C., & Burdakov, D. (2019). Natural hypothalamic circuit dynamics underlying object memorization. *Nat Commun*, 10(1), 2505. doi:10.1038/s41467-019-10484-7
- Krenzer, M., Anacleit, C., Vetrivelan, R., Wang, N., Vong, L., Lowell, B. B., . . . Lu, J. (2011). Brainstem and spinal cord circuitry regulating REM sleep and muscle atonia. *PLoS ONE*, 6(10), e24998. doi:10.1371/journal.pone.0024998
- Kroeger, D., Ferrari, L. L., Petit, G., Mahoney, C. E., Fuller, P. M., Arrigoni, E., & Scammell, T. E. (2017). Cholinergic, Glutamatergic, and GABAergic Neurons of the Pedunculopontine Tegmental Nucleus Have Distinct Effects on Sleep/Wake Behavior in Mice. *J Neurosci*, 37(5), 1352-1366. doi:10.1523/JNEUROSCI.1405-16.2016
- Lee, H. S., Yamazaki, R., Wang, D., Arthaud, S., Fort, P., DeNardo, L. A., & Luppi, P. H. (2020). Targeted recombination in active populations as a new mouse genetic model to study sleep-active neuronal populations: Demonstration that Lhx6+ neurons in the

- ventral zona incerta are activated during paradoxical sleep hypersomnia. *J Sleep Res*, e12976. doi:10.1111/jsr.12976
- Lee, M. G., Hassani, O. K., Alonso, A., & Jones, B. E. (2005). Cholinergic basal forebrain neurons burst with theta during waking and paradoxical sleep. *J Neurosci*, 25(17), 4365-4369. doi:10.1523/JNEUROSCI.0178-05.2005
- Lee, M. G., Hassani, O. K., & Jones, B. E. (2005). Discharge of identified orexin/hypocretin neurons across the sleep-waking cycle. *Journal of Neuroscience*, 25(28), 6716-6720.
- Leger, L., Goutagny, R., Sapin, E., Salvert, D., Fort, P., & Luppi, P. H. (2008). Noradrenergic neurons expressing Fos during waking and paradoxical sleep deprivation in the rat. *J Chem Neuroanat*. doi:S0891-0618(08)00164-6 [pii] 10.1016/j.jchemneu.2008.12.008
- Liu, K., Kim, J., Kim, D. W., Zhang, Y. S., Bao, H., Denaxa, M., . . . Blackshaw, S. (2017). Lhx6-positive GABA-releasing neurons of the zona incerta promote sleep. *Nature*, 548(7669), 582-587. doi:10.1038/nature23663
- Lu, J., Sherman, D., Devor, M., & Saper, C. B. (2006). A putative flip-flop switch for control of REM sleep. *Nature*, 441(7093), 589-594.
- Luo, J., Phan, T. X., Yang, Y., Garelick, M. G., & Storm, D. R. (2013). Increases in cAMP, MAPK activity, and CREB phosphorylation during REM sleep: implications for REM sleep and memory consolidation. *J Neurosci*, 33(15), 6460-6468. doi:10.1523/JNEUROSCI.5018-12.2013
- Luppi, P. H., Boissard, R., Gervasoni, D., Verret, L., Goutagny, R., Peyron, C., . . . Fort, P. (2004). The network responsible for paradoxical sleep onset and maintenance: a new theory based on the head-restrained rat model. In P. H. Luppi (Ed.), *Sleep: circuits and function* (pp. 272): CRC Press.
- Luppi, P. H., Clement, O., Sapin, E., Gervasoni, D., Peyron, C., Leger, L., . . . Fort, P. (2011). The neuronal network responsible for paradoxical sleep and its dysfunctions causing narcolepsy and rapid eye movement (REM) behavior disorder. *Sleep Med Rev*, 15(3), 153-163. doi:10.1016/j.smrv.2010.08.002
- Lydic, R., Baghdoyan, H. A., Hibbard, L., Bonyak, E. V., DeJoseph, M. R., & Hawkins, R. A. (1991). Regional brain glucose metabolism is altered during rapid eye movement sleep in the cat: a preliminary study. *J Comp Neurol*, 304(4), 517-529.
- Manns, I. D., Alonso, A., & Jones, B. E. (2000). Discharge profiles of juxtacellularly labeled and immunohistochemically identified GABAergic basal forebrain neurons recorded in association with the electroencephalogram in anesthetized rats. *J Neurosci*, 20(24), 9252-9263.
- Maquet, P., Peters, J., Aerts, J., Delfiore, G., Degueldre, C., Luxen, A., & Franck, G. (1996). Functional neuroanatomy of human rapid-eye-movement sleep and dreaming. *Nature*, 383(6596), 163-166.
- Mickelsen, L. E., Bolisetty, M., Chimileski, B. R., Fujita, A., Beltrami, E. J., Costanzo, J. T., . . . Jackson, A. C. (2019). Single-cell transcriptomic analysis of the lateral hypothalamic area reveals molecularly distinct populations of inhibitory and excitatory neurons. *Nat Neurosci*, 22(4), 642-656. doi:10.1038/s41593-019-0349-8
- Montgomery, S. M., Sirota, A., & Buzsaki, G. (2008). Theta and gamma coordination of hippocampal networks during waking and rapid eye movement sleep. *J Neurosci*, 28(26), 6731-6741. doi:28/26/6731 [pii] 10.1523/JNEUROSCI.1227-08.2008
- Niethard, N., Hasegawa, M., Itokazu, T., Oyanedel, C. N., Born, J., & Sato, T. R. (2016). Sleep-Stage-Specific Regulation of Cortical Excitation and Inhibition. *Curr Biol*, 26(20), 2739-2749. doi:10.1016/j.cub.2016.08.035

- Nofzinger, E. A., Mintun, M. A., Wiseman, M., Kupfer, D. J., & Moore, R. Y. (1997). Forebrain activation in REM sleep: an FDG PET study. *Brain Res*, 770(1-2), 192-201.
- Qu, D., Ludwig, D. S., Gammeltoft, S., Piper, M., Pellemounter, M. A., Cullen, M. J., . . . Maratos-Flier, E. (1996). A role for melanin-concentrating hormone in the central regulation of feeding behaviour. *Nature*, 380(6571), 243-247.
- Renouard, L., Billwiller, F., Ogawa, K., Clément, O., Camargo, N., Abdelkarim, M., . . . Luppi, P.-H. (2015). The supramammillary nucleus and the claustrum activate the cortex during REM sleep. *Science Advances*, 1(3). doi:10.1126/sciadv.1400177
- Robinson, T. E., Kramis, R. C., & Vanderwolf, C. H. (1977). Two types of cerebral activation during active sleep: relations to behavior. *Brain Res*, 124(3), 544-549.
- Sakai, K., Sastre, J. P., Kanamori, N., & Jouvet, M. (1981). State-specific neurones in the ponto-medullary reticular formation with special reference to the postural atonia during paradoxical sleep in the cat. In O. Pompeiano & C. Aimone Marsan (Eds.), *Brain Mechanisms of Perceptual Awareness and Purposeful Behavior* (pp. 405-429). New York: Raven Press.
- Sapin, E., Berod, A., Leger, L., Herman, P. A., Luppi, P. H., & Peyron, C. (2010). A Very Large Number of GABAergic Neurons Are Activated in the Tuberal Hypothalamus during Paradoxical (REM) Sleep Hypersomnia. *PLoS ONE*, 5(7), e11766. doi:10.1371/journal.pone.0011766
- Sapin, E., Lapray, D., Berod, A., Goutagny, R., Leger, L., Ravassard, P., . . . Luppi, P. H. (2009). Localization of the brainstem GABAergic neurons controlling paradoxical (REM) sleep. *PLoS ONE*, 4(1), e4272. doi:10.1371/journal.pone.0004272
- Sastre, J. P., Buda, C., Kitahama, K., & Jouvet, M. (1996). Importance of the ventrolateral region of the periaqueductal gray and adjacent tegmentum in the control of paradoxical sleep as studied by muscimol microinjections in the cat. *Neuroscience*, 74(2), 415-426.
- Sastre, J. P., Buda, C., Lin, J. S., & Jouvet, M. (2000). Differential c-fos expression in the rhinencephalon and striatum after enhanced sleep-wake states in the cat. *Eur J Neurosci*, 12(4), 1397-1410.
- Schneeberger, M., Tan, K., Nectow, A. R., Parolari, L., Caglar, C., Azevedo, E., . . . Friedman, J. M. (2018). Functional analysis reveals differential effects of glutamate and MCH neuropeptide in MCH neurons. *Mol Metab*, 13, 83-89. doi:10.1016/j.molmet.2018.05.001
- Shi, Y. F., Han, Y., Su, Y. T., Yang, J. H., & Yu, Y. Q. (2015). Silencing of Cholinergic Basal Forebrain Neurons Using Archaelhodopsin Prolongs Slow-Wave Sleep in Mice. *PLoS ONE*, 10(7), e0130130. doi:10.1371/journal.pone.0130130
- Sirieux, C., Gervasoni, D., Luppi, P. H., & Leger, L. (2012). Role of the lateral paragigantocellular nucleus in the network of paradoxical (REM) sleep: an electrophysiological and anatomical study in the rat. *PLoS ONE*, 7(1), e28724. doi:10.1371/journal.pone.0028724
- Soussi, R., Zhang, N., Tahtakran, S., Houser, C. R., & Esclapez, M. (2010). Heterogeneity of the supramammillary-hippocampal pathways: evidence for a unique GABAergic neurotransmitter phenotype and regional differences. *Eur J Neurosci*, 32(5), 771-785. doi:EJN7329 [pii]
- 10.1111/j.1460-9568.2010.07329.x
- Takehara-Nishiuchi, K. (2014). Entorhinal cortex and consolidated memory. *Neurosci Res*, 84, 27-33. doi:10.1016/j.neures.2014.02.012
- Torontali, Z. A., Fraise, J. J., Sanghera, P., Horner, R., & Peever, J. (2019). The Sublaterodorsal Tegmental Nucleus Functions to Couple Brain State and Motor Activity during REM Sleep and Wakefulness. *Curr Biol*, 29(22), 3803-3813 e3805. doi:10.1016/j.cub.2019.09.026

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- Unal, G., Joshi, A., Viney, T. J., Kis, V., & Somogyi, P. (2015). Synaptic Targets of Medial Septal Projections in the Hippocampus and Extrahippocampal Cortices of the Mouse. *J Neurosci*, 35(48), 15812-15826. doi:10.1523/JNEUROSCI.2639-15.2015
- Valencia Garcia, S., Brischoux, F., Clement, O., Libourel, P. A., Arthaud, S., Lazarus, M., . . . Fort, P. (2018). Ventromedial medulla inhibitory neuron inactivation induces REM sleep without atonia and REM sleep behavior disorder. *Nat Commun*, 9(1), 504. doi:10.1038/s41467-017-02761-0
- Valencia Garcia, S., Libourel, P. A., Lazarus, M., Grassi, D., Luppi, P. H., & Fort, P. (2016). Genetic inactivation of glutamate neurons in the rat sublaterodorsal tegmental nucleus recapitulates REM sleep behaviour disorder. *Brain*. doi:10.1093/brain/aww310
- van den Pol, A. N., Acuna-Goycolea, C., Clark, K. R., & Ghosh, P. K. (2004). Physiological properties of hypothalamic MCH neurons identified with selective expression of reporter gene after recombinant virus infection. *Neuron*, 42(4), 635-652.
- Van Dort, C. J., Zachs, D. P., Kenny, J. D., Zheng, S., Goldblum, R. R., Gelwan, N. A., . . . Brown, E. N. (2014). Optogenetic activation of cholinergic neurons in the PPT or LDT induces REM sleep. *Proc Natl Acad Sci U S A*. doi:10.1073/pnas.1423136112
- van Strien, N. M., Cappaert, N. L., & Witter, M. P. (2009). The anatomy of memory: an interactive overview of the parahippocampal-hippocampal network. *Nat Rev Neurosci*, 10(4), 272-282. doi:10.1038/nrn2614
- Varin, C., Luppi, P. H., & Fort, P. (2018). Melanin-concentrating hormone-expressing neurons adjust slow-wave sleep dynamics to catalyze paradoxical (REM) sleep. *Sleep*, 41(6). doi:10.1093/sleep/zsy068
- Verret, L., Fort, P., Gervasoni, D., Leger, L., & Luppi, P. H. (2006). Localization of the neurons active during paradoxical (REM) sleep and projecting to the locus coeruleus noradrenergic neurons in the rat. *J Comp Neurol*, 495(5), 573-586. doi:10.1002/cne.20891
- Verret, L., Goutagny, R., Fort, P., Cagnon, L., Salvart, D., Leger, L., . . . Luppi, P. H. (2003). A role of melanin-concentrating hormone producing neurons in the central regulation of paradoxical sleep. *BMC Neurosci*, 4(1), 19.
- Verret, L., Leger, L., Fort, P., & Luppi, P. H. (2005). Cholinergic and noncholinergic brainstem neurons expressing Fos after paradoxical (REM) sleep deprivation and recovery. *Eur J Neurosci*, 21(9), 2488-2504. doi:EJN4060 [pii] 10.1111/j.1460-9568.2005.04060.x
- Weber, F., Chung, S., Beier, K. T., Xu, M., Luo, L., & Dan, Y. (2015). Control of REM sleep by ventral medulla GABAergic neurons. *Nature*, 526(7573), 435-438. doi:10.1038/nature14979
- Weber, F., Hoang Do, J. P., Chung, S., Beier, K. T., Bikov, M., Saffari Doost, M., & Dan, Y. (2018). Regulation of REM and Non-REM Sleep by Periaqueductal GABAergic Neurons. *Nat Commun*, 9(1), 354. doi:10.1038/s41467-017-02765-w
- Wyss, J. M., & Van Groen, T. (1992). Connections between the retrosplenial cortex and the hippocampal formation in the rat: a review. *Hippocampus*, 2(1), 1-11. doi:10.1002/hipo.450020102
- Xu, M., Chung, S., Zhang, S., Zhong, P., Ma, C., Chang, W. C., . . . Dan, Y. (2015). Basal forebrain circuit for sleep-wake control. *Nat Neurosci*, 18(11), 1641-1647. doi:10.1038/nn.4143

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