ALTERED SLEEP OSCILLATIONS AS EARLY BIOMARKERS OF PARKINSON’S DISEASE CHOLINOPATHY

Jasna Šaponjić

University of Belgrade, Department of Neurobiology, Institute for Biological Research - Sinisa Stankovic, 11 060 Belgrade, Serbia.
Definition of sleep

• The failure to define sleep as a single state lies in fact that it is not a homogenous state – sleep is continuum of number of mixed states

• No definition has succeeded in satisfying all aspects of sleep

• Sleep is global, complex behavioral state of all mammals that is homeostatically regulated

• Sleep is rapidly reversible state of immobility and greatly reduced sensory responsiveness

• The control mechanisms of sleep are manifested at every level of biological organization - from genes and intracellular mechanisms to networks of cell populations within the CNS
• Our knowledge of the neural substrates of sleep is based on animal studies, primarily involving cat and rat
What are the sleep functions?

We still do not have a meaningful explanation for the actual function of sleep.

Electrophysiological background

The discharge pattern activity of the LC noradrenergic neurons and DRN serotonergic neurons is opposite to the cholinergic PPT neurons.

(W > SWS > REM)

(McGinty and Harper, 1976; Lydic et al, 1987; Hobson et al., 1975; Foote et al., 1983)
FLIP-FLOP CONTROL OF REM SLEEP

GABA, galanin
VLPO

orixin
LH

REM - off

SLEEP and DEVELOPMENT

- REM sleep serves to direct the course of brain maturation

- REM sleep is not essential for survival (REM sleep suppression is not fatal)
  - (tricyclic antidepressants, clonidine, MAO inhibitors induce absence of dreams and reduction of REM sleep without major consequences)

- REM sleep provides endogenous activation at the time when the brain has little or no exogenous inputs
  - (activity-dependent concept of neuronal connectivity; high-frequency brainstem activation through PGO waves provides thalamocortical pathways maturation)
Figure 1—The developmental decrease in REM sleep and transmitter changes. A. Adapted from 78 to show the decrease in REM sleep from ~80% of sleep time to ~10% between 10 and 30 days in the rat, after which point it remains stable. B. Changes in the effects of transmitter systems on the membrane potential of PPN neurons. The y axis shows the mean depolarization above resting membrane potential (RMP, 0 mV) or hyperpolarization below RMP. Between 12 and 21 days, the same concentration of 1) NMDA was found to decrease from +12 mV to +4 mV (red line)118; 2) KA was found to increase from +4 mV to +13 mV (blue line)118; 3) a cholinergic agonist increased hyperpolarization from -8 mV to -10 mV127; 4) a α2 adrenergic agonist decreased hyperpolarization from -6 mV to -2 mV121; 5) a 5-HT1 agonist increased hyperpolarization from -2 mV to -7 mV101; 6) a 5-HT2 agonist did not change the degree of hyperpolarization (-2 mV)101; 7) a GABAa agonist first depolarized (+3 mV) then hyperpolarized (-1 mV)131; and 8) a GABAb agonist decreased hyperpolarization from -5 mV to -1 mV131.
REM sleep drive early in development is more related to high levels of electrical coupling then to changes in chemical transmission providing persistence of high frequency rhythms essential for waking and REM ???


- MODAFINIL (Provigil, Modiodal)
  diphenylmethyl sulfonyl-2 acetamide derivative
  approved for use in treating excessive sleepiness in narcolepsy, sleepiness in obstructive apnea and shift work sleep disorders, and in a number of neuropsychiatric conditions
  increase glutamatergic, adrenergic and histaminergic activity
  increase electrical coupling between cortical interneurons, thalamic reticular neurons and inferior olive neurons Urbano et al., PNAS, 2007, 104 (30), 12554-59.
  modafinil effects were blocked by the gap junction blocker mefloquine
Neurotransmitter systems in promoting wakefulness

Saper and Fuller, Current Opinion in Neurobiology 2017, 44, 186-192.
Neurotransmitter systems in promoting sleep

Saper and Fuller, Current Opinion in Neurobiology 2017, 44, 186-192.
Levels of sleep organization

Brain state dependent differences of cortical neuronal (pyramidal neurons) firing patterns

Watson et al., Neuron 90, 2016, 839-852.
Impact of brain state on the population distribution of neuronal firing rates

Watson et al., Neuron 90, 2016, 839-852.
97% RBD cases are α-synucleinopathy – α-synuclein positive intracellular inclusions
~33-60% PD; 50-80% in DLB; 80-95% of MSA patients
RBD tend to precede by years or decades (5-40 years) the onset of motor and cognitive features of neurodegenerative disorders such as PD, MSA, DLB

Table 3  Updated clinicopathological experience at Mayo Clinic from January 1990 to December 2006 of REM sleep behaviour disorder associated with dementia and/or parkinsonism

<table>
<thead>
<tr>
<th>Pathological diagnoses</th>
<th>N</th>
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<tbody>
<tr>
<td>Lewy body disease(^a)</td>
<td>31</td>
</tr>
<tr>
<td>Multiple system atrophy</td>
<td>4</td>
</tr>
<tr>
<td>Progressive supranuclear palsy(^b)</td>
<td>1</td>
</tr>
<tr>
<td>Total</td>
<td>36</td>
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</tbody>
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\(^a\)Twelve of the LBD cases also met criteria for intermediate or high likelihood of Alzheimer’s disease.
\(^b\)Coexisting vascular and Alzheimer pathology was present.

Boeve et al., Brain 2007, 130: 2770-2788.
Parkinson’s disease

- Progressive neurodegenerative disorder affecting about 1 person out of every 1000, up to 50 years old; 19 persons out of every 1000, up to ≥ 80 years old

- Main histopathological changes - progressive loss of the nigrostriatal dopaminergic pathway and dopaminergic neurons in the SNpc

- Neurodegeneration affects also NA, 5-HT, Ach
Parkinson’s disease at cellular level

• **Synucleinopathy**
  accumulation of misfolded protein aggregates

• **Synaptopathy**
  abnormal synaptic connectivity within the nigrostriatal pathways and intrastriatal interneuronal connections (redistribution of synaptic proteins – SNARE, SNAP-25, syntaxin-1, synaptobrevin-2)

• **Cholinopathy**
  (PPT degeneration – RBD, gait impairment, frequent falls).
PPT as the higher relay nucleus in control of the integrated brain cholinergic function
Schematic representation of the REM sleep generation process.

Hobson et al. (2000)
Material and Methods
Excitotoxic lesion (IBO lesion)

Stereotaxically guided microinfusion of the 100 nl 0.1M IBO single 60 s pulse duration

NB (A/P: - 1.4; R/L: 3.0; D/V: 7.0);
PPT (A/P: - 7.8; R/L: 1.9; D/V: 7.0)
Material and Methods
- Operative procedure -

Chronic implantation of the EEG and EMG electrodes for sleep recording

MCx: A/P: +1.0; R/L: 2.0

SMCx: A/P: -3.0; R/L: 2.0

CA1: A/P: -3.60; R/L: 2.5; D/V: 2.5
Material and Methods

Sleep recording 6 h during the normal inactive circadian phase for rats from 9 a.m. - 3 p.m.
NADPH-diaphorase histochemical verification of the PPT lesion
There was no progression of the NB and PPT cholinergic neuronal loss within their overall rostro-caudal dimensions and across the overall aging follow-up period.
NREM EEG microstructure in PPT lesion vs. control

**NREM**

**(RA)_{b} = \frac{\sum_{b} \text{Amp}_{b}}{\sum_{\text{tot}} \text{Amp}_{\text{tot}}}**
REM EEG microstructure in PPT lesion vs. control

Petrovic et al., Experimental Neurology 247 (2013), 562-571.
Topography of the Wake/NREM/REM differentiation
REM without atonia (RBD) in bi-PPT lesion?

Topography of the NREM EEG microstructure (NB vs. PPT lesion)

NREM

A

B

C

D

Topography of the REM EEG microstructure (NB vs. PPT lesion)
Topography of the REM sleep alterations following the PPT lesion

A

SMCx

MCx

Saponjic et al., Challenges in Parkinson’s Disease, 2016, Ch7, pp. 135-153.
EEG microstructures during REM/REM1/REM2 states

Saponjic et al., Challenges in Parkinson’s Disease, 2016, Ch7, pp. 135-153.
REM/REM1/REM2 coherence spectra

C

SMCx

MCx

Coherence

R

R1

R2

Frequency (Hz)

Saponjic et al., Challenges in Parkinson's Disease, 2016, Ch7, pp. 135-153.
Cortico-muscular coherence (CMC)

- Calculated using the "cohere" routine of the MATLAB 6.5 Signal Processing Toolbox

- The magnitude squared coherence between x (EEG) and y (EMG) as:

\[ C_{xy}(f) = \frac{|P_{xy}(f)|^2}{P_{xx}(f)P_{yy}(f)} \]

where \( P_{xy}(f) \) stands for the cross spectrum of \( x \) and \( y \), while \( P_{xx}(f) \) and \( P_{yy}(f) \) denote the power spectra of the two signals.

- All \( P_{xy} \), \( P_{xx} \) and \( P_{yy} \) values were determined for every 10 s of each 6 h recording, and for each frequency within the overall 0.3 - 50 Hz range, with 0.1 Hz resolution.

- The CMCs values were averaged within each conventional EEG frequency band for each spectrum, and their means were calculated for each state and each group.
Cortico-muscular coherence (CMC)

- measure of propagated oscillation from the cortex to dorsal nuchal muscles;
- a method for detecting an early markers of healthy aging;
- a marker of functional coupling between primary motor cortex and peripheral muscles;
- a method for quantifying the functional coupling between the motor cortex and contralateral peripheral muscles in frequency domain;
- a measure of the pyramidal system integrity;
- CMC plays a crucial role for sensorimotor integration, representing a key mechanism for appropriate motor control.
Sleep architecture within the motor cortex (MCx) during aging

Ciric et al., Translational Brain Rhythmicity 2017, 2 (1): 2-11.
Topography of ageing altered REM EEG microstructure

Ciric et al., Behav Brain Res 2016, 301:273-286.
Individual examples of the **MCx REM spectrograms** with their typical 10 s analog EEG signals throughout the overall ageing follow-up period in the control vs. PPT lesioned rat.

Ciric et al., Behav Brain Res 2016, 301:273-286.
REM “enriched with” sigma oscillations in MCx

Ciric et al., Translational Brain Rhythmicity 2017, 2 (1): 2-11.
Sleep spindles (SS) during REM “enriched” with sigma oscillations

Ciric et al., Translational Brain Rhythmicity 2017, 2 (1): 2-11.
Sleep spindle dynamics (density, duration, intrinsic frequency)

Ciric et al., Translational Brain Rhythmicity 2017, 2 (1): 2-11.
Distinct MCx and Hipp sleep during PD cholinopathy

Quantification of the PPT cholinergic neuronal loss
Quantification of the remote cholinergic neuronal loss in the caudate putamen

Altered EEG microstructure of hippocampal NREM sleep as the earliest and long-lasting

NREM sleep EEG microstructure 91 days after the PPT lesion

High voltage sleep spindles during REM

Control - 414 HVS

PPT lesion – 1338 HVS

High voltage sleep spindles during REM

c-Fos immunostaining in hippocampus

Control

PPT lesion

Altered neuronal activity in the DG during PD cholinopathy – no activation of interneurons
Delayed hypoactivity

D-AMPH induced hyperactivity with underlying c-Fos immunoactivity suppression in the caudate putamen

Conclusion

Disorders (alterations) of:

**EEG microstructure during sleep,**

cortical drives during sleep,

and **sleep spindles**

are the hallmarks of PD cholinopathy,

and the early biomarkers of brain re-organizational processes
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