Why & How to do sleep manipulations in animals?

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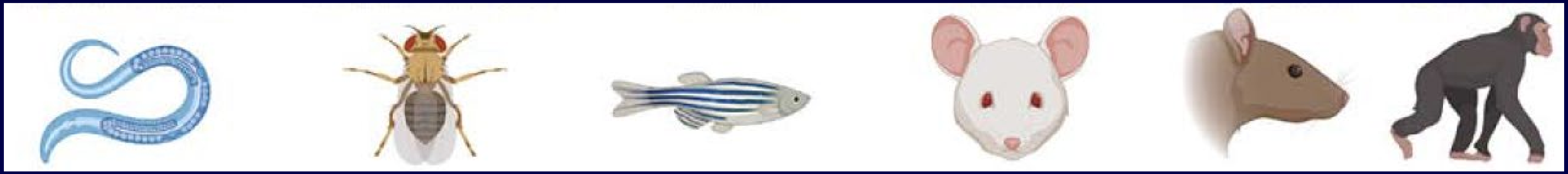
Why do sleep deprivation studies?

- Understand sleep physiology
- Mechanisms of sleep disorders
- Interaction between sleep and psychiatric diseases



(Modified from Moyer et al., Trends in Genetics, 2021)

Using rodents to study sleep

- Advantages
  - Genetics
  - Tools
  - Behavioral models
- Differences
  - Nocturnal
  - Polyphasic
- Similarities
  - NREM sleep, REM sleep
  - Homeostatic regulation
  - Conserved NREM and REM sleep regulatory brain regions

Sleep manipulation goals

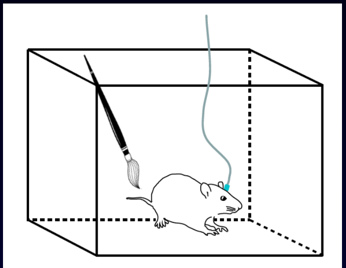
- Acute sleep deprivation
- Chronic sleep disturbance
- Sleep enhancement

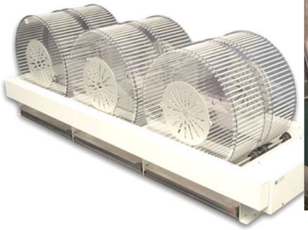
X

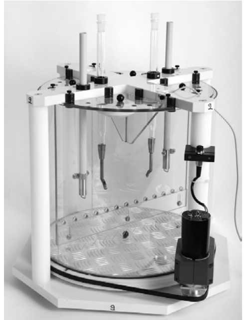
NREM/REM sleep selective manipulations

Sleep manipulation methods

- Gentle handling (acute; total sleep deprivation)
- Automated devices (chronic; NREM/REM selective)







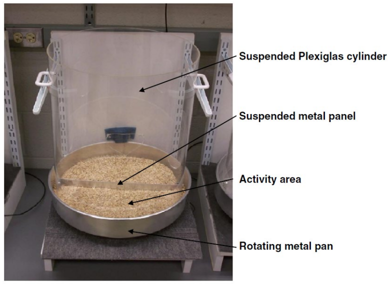
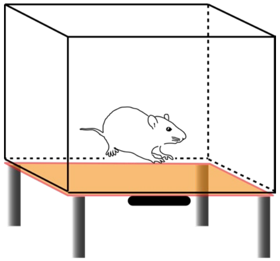
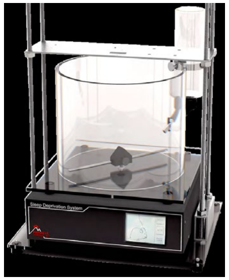
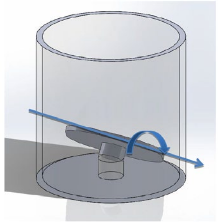


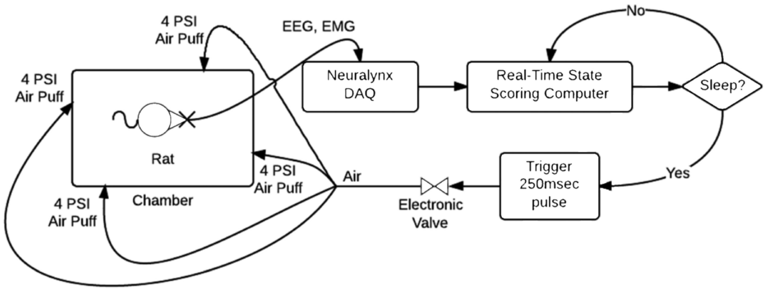
Fig. 1. Disc treadmill apparatus for chronic sleep restriction.



Signal Solutions LLC



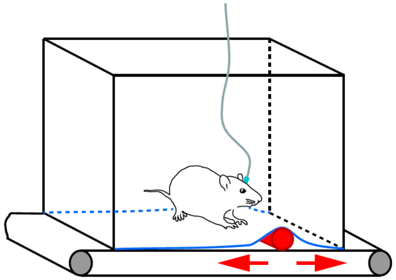




References: Gross et al., 2015; Bjorness et al., 2020; Stoebe et al., 2003; Leenaars et al., 2011; Puhl et al., 2013; Pinnacle brochure; Lafayette brochure

Sleep manipulation methods: chronic REM sleep deprivation/fragmentation

- Inverted flowerpot method
- Modified treadmill system



- Effectiveness for sleep disturbance
- Stress
- Software implementation
- Feasibility
- Cost
- IACUC approval

References: Jouvet et al., 1964; Cohen and Dement, 1965; Everson et al., 1989; Hughes, et al., 2013; Chen et al., 2015)

Sleep enhancement

- Sleep deprivation-rebound
- Temperature control
- Sleep network manipulations

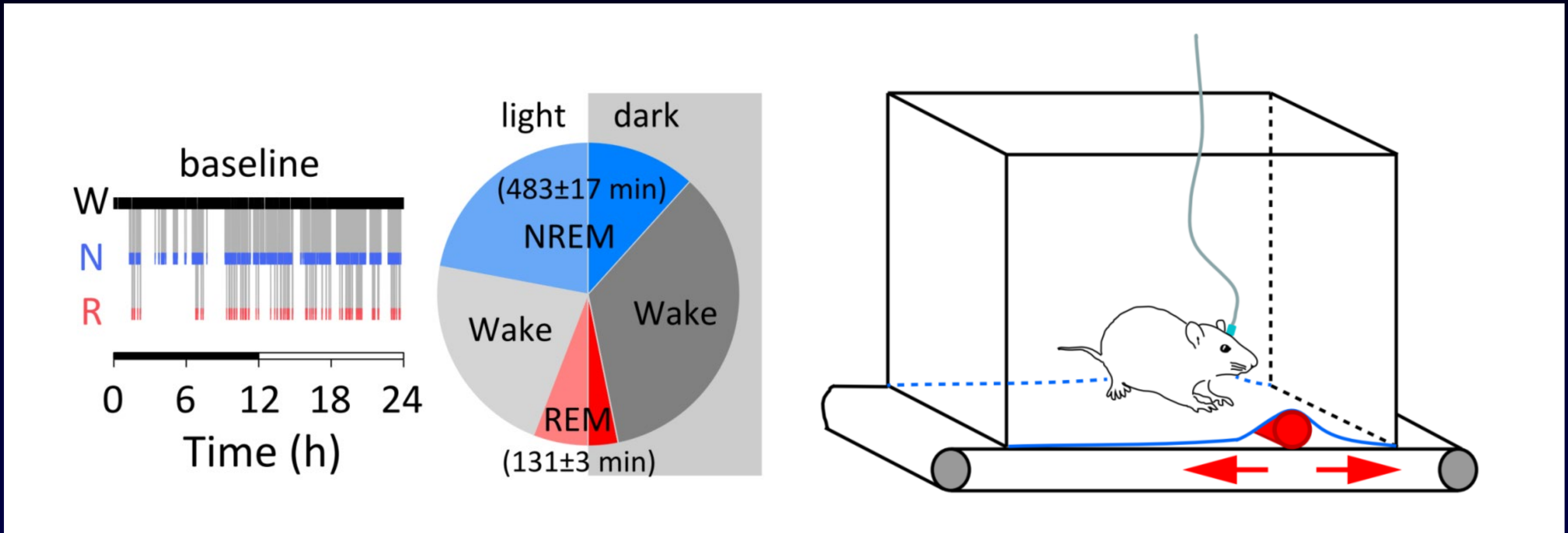
X

NREM/REM
selective manipulations

Sleep enhancement

- Sleep deprivation-rebound
- Temperature control
- Sleep network manipulations

Dark-phase REM sleep restriction/Light-phase rebound

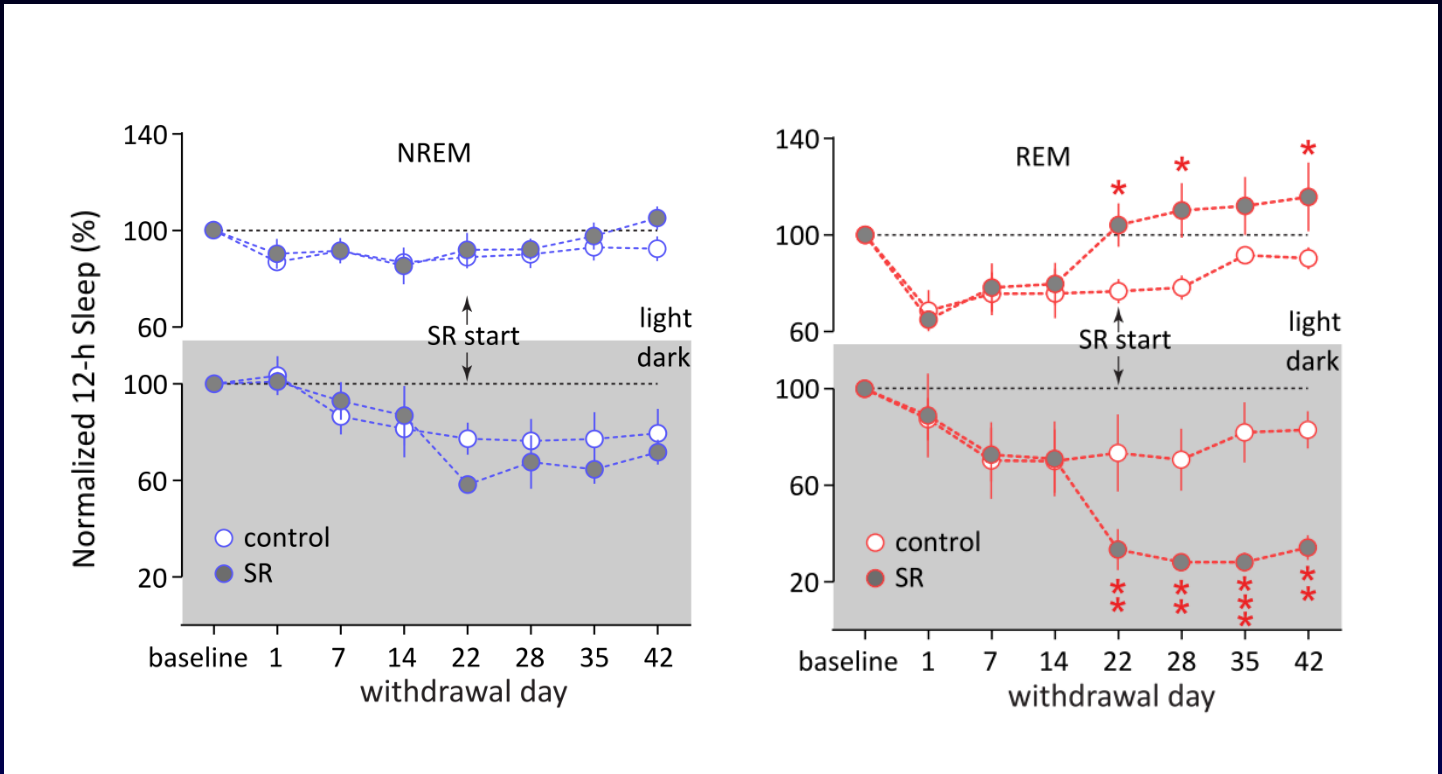


0 hr

12 hr

24 hr

Dark-phase sleep restriction: REM redistribution



(Chen et al., JNS, 2015)

Sleep enhancement

- Sleep deprivation-rebound
- Temperature control
- Sleep network manipulations

Ambient/body temperature and sleep

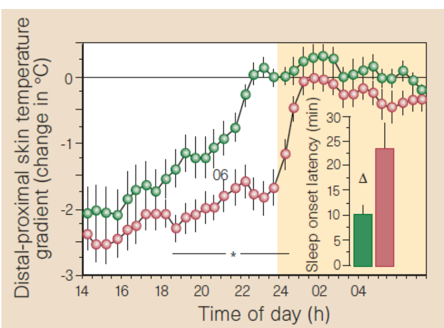
RESEARCH ARTICLE

Head cooling during sleep improves sleep quality in the luteal phase in female university students: A randomized crossover-controlled pilot study

Seiji Hamanishi[1,2]*, **Eri Eguchi**[1], **Tatsuo Ito**[1‡], **Kenjiro Nagaoka**[1‡], **Keiki Ogino**[1]

1 Graduate School of Medicine Density and Pharmaceutical Sciences, Okayama University, Okayama, Japan, **2** Department of Nursing, Kansai University of Social Welfare, Ako, Japan

(Hamanishi et al., PLOS ONE, 2019)



Physiology

Warm feet promote the rapid onset of sleep

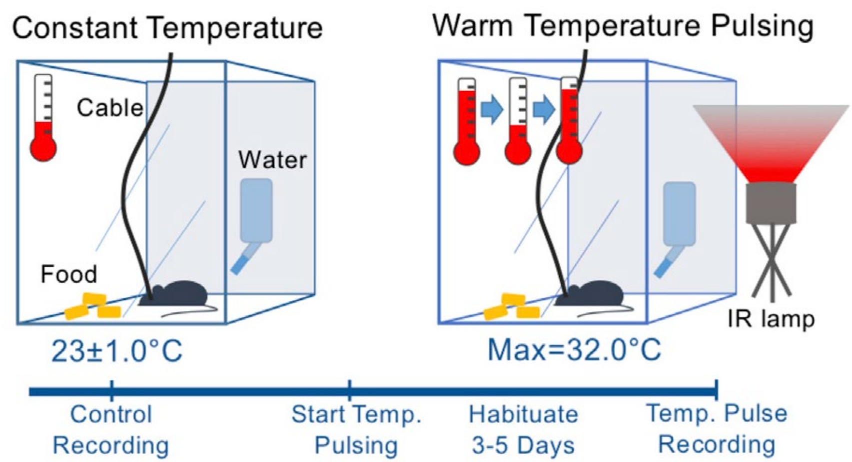
Even healthy people occasionally have difficulty falling asleep. Psychological relaxation techniques, hot baths, soothing infusions of plant extracts, melatonin and conventional hypnotics are all invoked in the search for a good night's sleep. Here we show that the degree of dilation of blood vessels in the skin of the hands and feet, which increases heat loss at these extremities, is the best physiological predictor for the rapid onset of sleep. Our findings provide further insight into the thermoregulatory cascade of events that precede the initiation of sleep[1].

(Kräuchi et al., Nature, 1999)

Article

Current Biology

Dynamic REM Sleep Modulation by Ambient Temperature and the Critical Role of the Melanin-Concentrating Hormone System

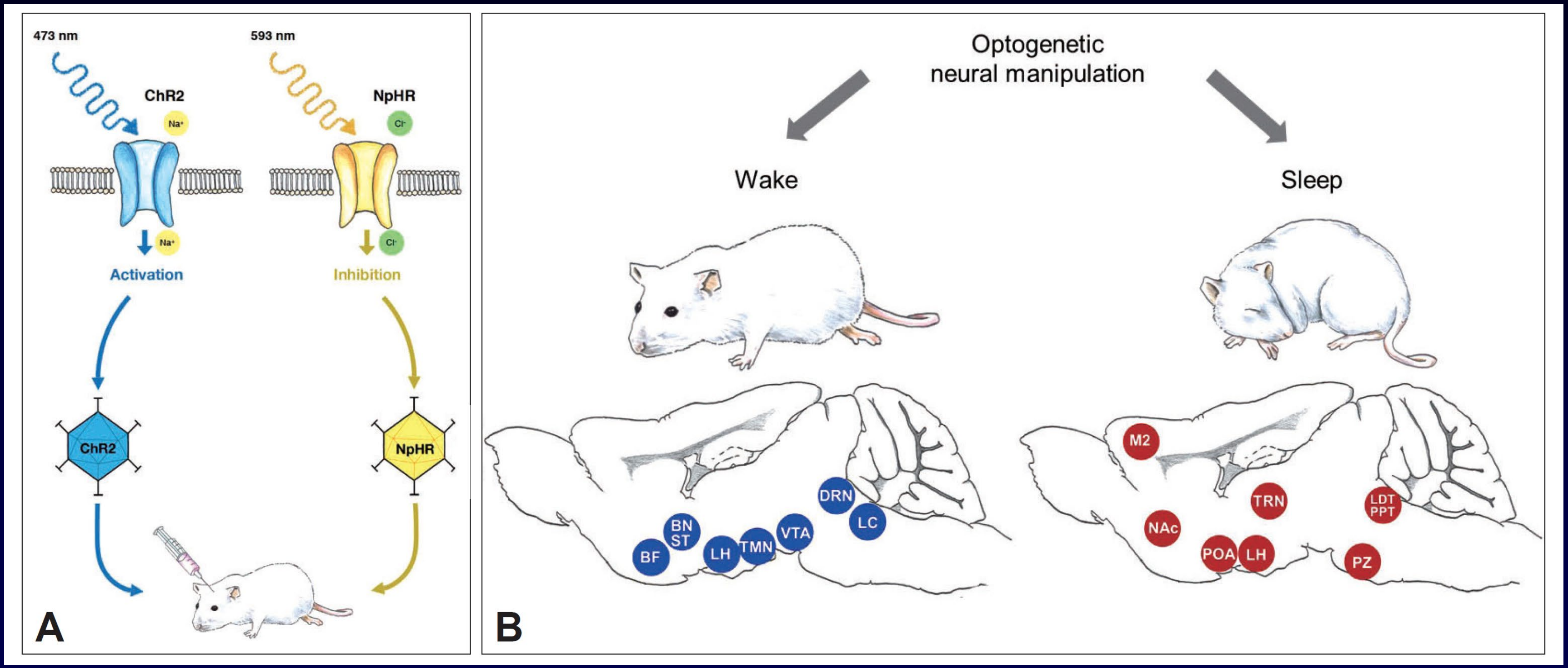


(Komagata et al., Curr Biol, 2019)

Sleep enhancement

- Sleep deprivation-rebound
- Temperature control
- Sleep network manipulations

Sleep network manipulations: Optogenetics



(Drew et al., Sleep Med Res, 2018)

Sleep network manipulations: Optogenetics

Table 1. Summarization of recent publications on the sleep-wake control using optogenetics

Cell type	Opsin	Marker protein	Animal subject	Vector	Results	Reference
Wake-related						
BF^{ACh}	ChR2	ChAT	ChAT-Cre mice	AAV5-DIO-ChR2-eYFP	BF^{ACh} neurons are wake-promoting	Ozen Irmak et al. [26]
BF^{ACh}, BF^{Glu}, BF^{GABA}	ChR2	ChAT, VGLUT, PV, SOM	ChAT-Cre, VGLUT2-Cre, PV-Cre, SOM-Cre mice	AAV2-FLEX-ChR2-eYFP, AAV2-FLEXeYFP, AAV-DJ-FLEX-ChR2-mCherry, AAV-DJ-FLEX-eNpHR3.0-eYFP	BF^{ACh}, BF$^{PV/GABA}$, BF^{Glu} are wake-promoting, BF$^{SOM/GABA}$ sleep-promoting	Xu et al. [25]
BF^{ACh}	ChR2	ChAT	ChAT-ChR2-eYFP mice	No vector	BF^{ACh} neurons promote wakefulness via activation of BFPV neurons	Zant et al. [24]
BF^{ACh} → Cortex$^{nNOS/NKR}$	ChR2	ChAT	ChAT-IRES-Cre;Ai32 mice	AAV-ChR2(H134R)-eYFP	BF^{ACh} neurons excite but reduce glutamatergic inputs to Cortexn$^{NOS/NK1}$ neurons	Williams et al. [37]
BF$^{PV/GABA}$	ChR2	PV	PV-Cre mice	AAV5-DIO-ChR2-EYFP, AAV5-FLEX-ArchT-GFP	BF$^{PV/GABA}$ neurons regulate cortical GBO and are wake-active	Kim et al. [38]
BNST^{GABA}	ChR2	GAD67	GAD67-Cre mice	AAV-DIO-ChR2-EYFP	BNST^{GABA} neurons promote wakefulness	Kodani et al. [39]
DRN^{5-HT}	ChR2	Tph2	Tph2-ChR2 mice	No vector	DRN^{5-HT} neurons are wake-promoting	Ito et al. [21]
DRN^{DA}	ChR2	Tyrosine hydroxylase	TH-IRES-Cre mice	AAV-DIO-ChR2-eYFP	DRN^{DA} neurons are wake-promoting	Cho et al. [22]
LC^{NE}	ChR2	Tyrosine hydroxylase	TH-Cre mice	AAV-DIO-ChR2-eYFP	LC^{NE} neurons are wake-promoting	Carter et al. [36]
LH^{Hcrt}	ChR2	Hcrt	Hcrt-EGFP mice	Lenti-Hcrt-ChR2-mCherry	LH^{Hcrt} neurons promote wakefulness	Adamantidis et al. [30]
LH^{Hcrt}	ChR2	Hcrt	C57BL/6 mice	Hcrt::ChR2–mCherry lentivirus	Sleep pressure moderates Hcrt-mediated transitions to wakefulness	Carter et al. [31]
LH^{Hcrt}	NpHR	Hcrt	Hcrt-NpHR mice	No vector	Inhibition of LH^{Hcrt} neurons induces sleep	Tsunematsu et al. [33]
LH^{Hcrt}, LC^{NE}	ChR2	Tyrosine hydroxylase	TH-IRES-Cre mice	Lent-Hcrt::ChR2-mCherry, AAV-DIO-eNpHR3.0-eYFP or eYFP	LH^{Hcrt} neurons need LC^{NE} neurons to induce wakefulness	Carter et al. [32]
TMN^{HA}	Arch3.0	Hdc	HDC-Cre mice	AAV5-FLEX-Arch3.0-EYFP	Silencing of TMN^{HA} neurons promotes NREM, but not REM, sleep during periods of low sleep pressure	Fujita et al. [40]
TMN^{HA}, TMN^{GABA}	ChR2	HDC	HDC-ChR mice	AAV-flex-ChR2(H134R)-EYFP	TMN$^{HA/GABA}$ neurons are wake-promoting	Yu et al. [23]
VTA^{DA}	ChR2	Tyrosine hydroxylase	TH-Cre mice	AAV5-DIO-ChR2-eYFP	VTA^{DA} neurons promote wakefulness	Eban-Rothschild et al. [27]

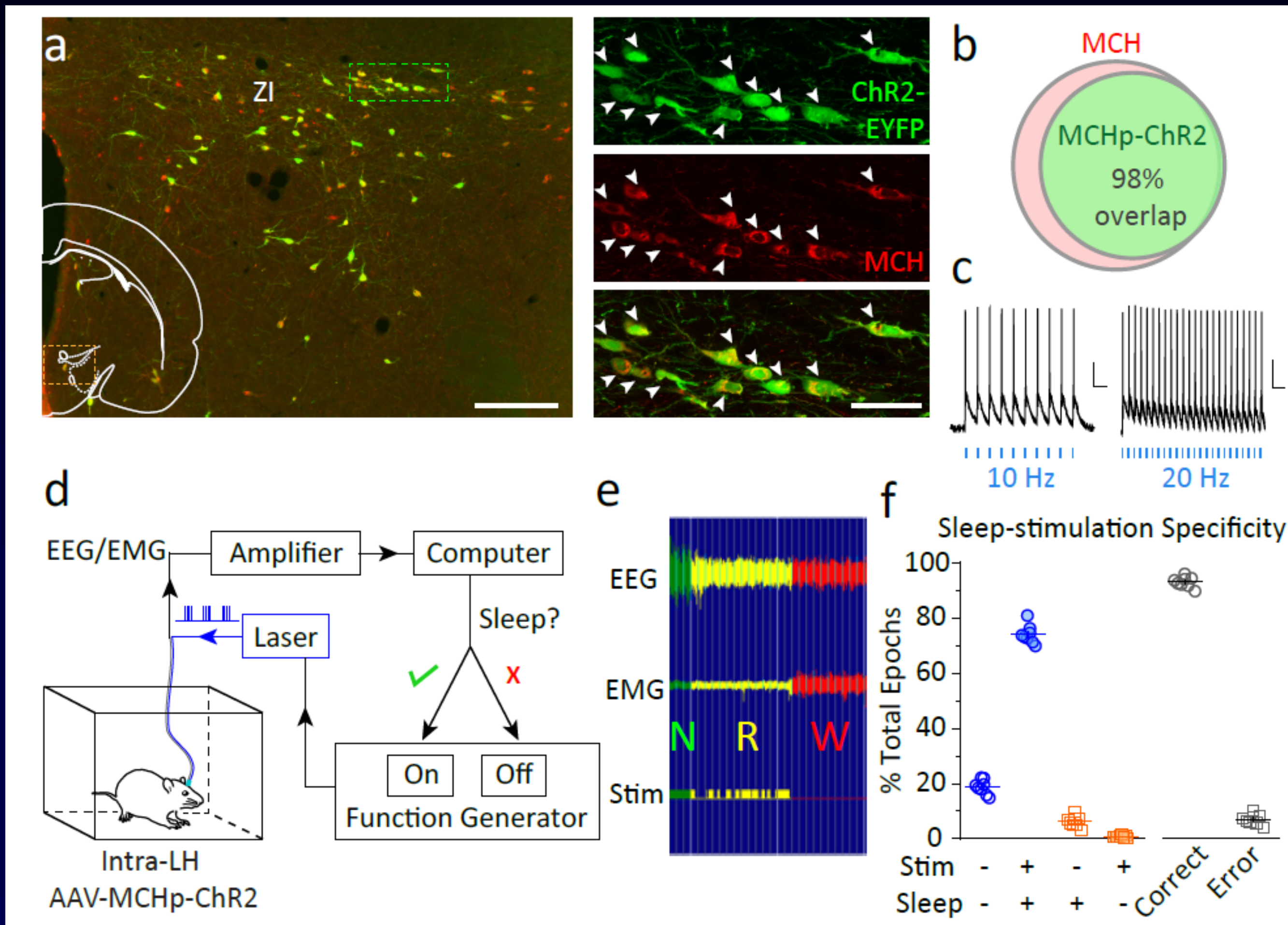
Table 1. Summarization of recent publications on the sleep-wake control using optogenetics (continued)

Cell type	Opsin	Marker protein	Animal subject	Vector	Results	Reference
Sleep-related						
LH$^{MCH/GABA}$	ChETA, SSFO	proMCH	Pmch-Cre mice	AAVdj-EF1-DIO-ChETA-EYFP	LH$^{MCH/GABA}$ neurons increase REM sleep	Jego et al. [52]
LH$^{MCH/GABA}$	ChR2	MCH	C57BL/6J mice	rAAV-MCH-hChR2(H134R)-EYFP	LH$^{MCH/GABA}$ neurons increase sleep	Konadhode et al. [50]
M2^{SOM}	C1V1, ChR2	SOM	SOM-Cre mice	AAV5-DIO-C1V1(E122T/E162T)-TS-EYFP	M2$^{GABA/SOM}$ neurons promote SWS	Funk et al. [47]
NAc Core$^{GABA/A2AR}$	ChR2	A2AR	A2AR-Cre mice	AAV-DIO-ChR2-mCherry	NAc core$^{GABA/A2AR}$ neurons promote SWS	Oishi et al. [45]
POA^{GABA} → TMN^{HA}	ChR2, iC++	GAD2	GAD2-Cre mice	AAV-DIO-ChR2-eYFP, Lenti-rEIAV-DIO-TLoop-iC++-eYFP	POA^{GABA} → TMN neurons are sleep-promoting	Chung et al. [43]
PPT^{ACh}	ChR2	ChAT	ChAT-ChR2 mice	No vector	PPT^{ACh} and LDT^{ACh} neurons promote REM sleep	Van Dort et al. [48]
PZGABA	ChR2	Vgat, Vglut2	Vgat-IRES-Cre, Vglut2-IRES-Cre mice	AAV-DIO-ChR2-mCherry	PZGABA neurons promote SWS	Anaclet et al. [46]
TRN^{GABA}	ChR2	Thy1	Thy1-ChR2 mice	No vector	Activating TRN with "spindle-like" protocol increases NREM sleep	Kim et al. [44]
Other functions						
BLA^{Glu}	ChR2, eNpHR3.0	CaMKIIα	C57BL/6J mice	AAV-CaMKIIα-eNpHR3.0-eYFP, CaMKIIα-ChR2-eYFP	BLA^{Glu} mediates stress-induced REM sleep reduction caused by fear memory	Machida et al. [54]
C1^{NE}	ChR2	TH	TH-Cre rats	AAV-DIO-ChR2-EYFP	RVLM^{NE} neurons (C1 neurons) increase wakefulness	Burke et al. [66]
V1-L6^{CT}	ChR2	Ntsr1	Ntsr1-ChR2 mice	AAV-FLEX-Arch-GFP	NREM sleep is necessary for transferring information from the visual thalamus to V1	Durkin et al. [64]
DRN^{5-HT}	ChR2	Sert	Sert-Cre;orexin-ataxin 3 mice	AAV-DIO-SSFO-EYFP	Cataplexy is inhibited by DRN^{5-HT} neurons	Hasegawa et al. [75]
LH^{Hcrt}	ChR2	Hcrt	C57BL/6 mice	Lenti-Hrct-ChR2-mCherry	Sleep fragmentation impairs memory consolidation	Rolls et al. [57]
M2^{Glu}	ArchT, ChR2	CaMKIIa	CaMKIIa-Cre, Thy1-ChR2 mice	AAV-FLEX-ArchT	M2^{Glu} neurons are necessary for memory consolidation during NREM sleep	Miyamoto et al. [62]

(Drew et al., Sleep Med Res, 2018)

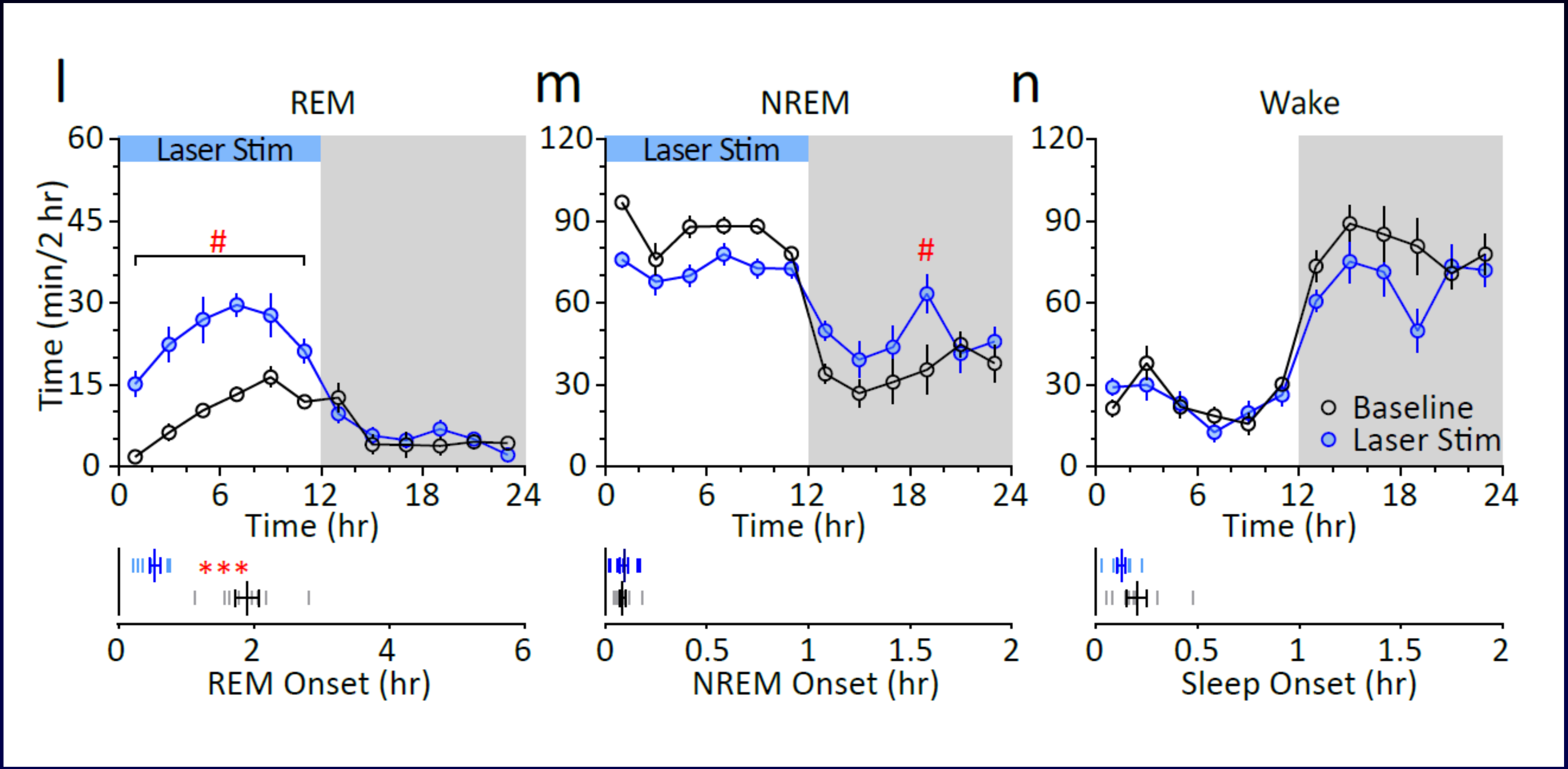
Closed-loop, sleep state-dependent stimulation

Jidong Fang
(Penn State)



Rong Guo

Sleep-stimulation of LH MCH neurons increase REM sleep



(Guo et al., Biological Psychiatry, 2022)

Sleep manipulation goals

- Acute sleep deprivation
- Chronic sleep disturbance
- Sleep enhancement

X

NREM/REM
selective manipulations

Acknowledgement

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