Current Biology, Volume *28*

Supplemental Information

Fur Seals Suppress REM Sleep for Very Long Periods without Subsequent Rebound

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Seal	Sex /Age (years)	Month [1]	Baseline (days)	In water (days)[2]	Recovery (days)	Average water temp. °C
1	M, 3-4 yr	May-June	2	Days in water: 11 SWS: 10 (1-9,11) REM: 11 (1-11)	2	19.3±0.2
2	M, 2-3 yr	Apr	2	Days in water: 11 SWS: 6 (1,7-11) REM: 6 (1,7-11)	2	10.7±0.1
3	M, 2-3 yr	Aug	2	Days in water: 10 SWS: 10 (1-10) REM: 10 (1-10)	2	26.5±0.1
4	F, 4-5 yr	June	2	Days in water:14 SWS: 7 (1-2,7-8,12-14) REM: 11 (1-5,7-9,12-14)	2	23.4±0.1
Total number of days and number of days for which amounts of SWS and REM sleep were evaluated			8	Days in water: 46 SWS: 33 REM: 38	8	

[1] Studies of sleep in seawater (seals 1-4) were conducted between April and September. This includes the end of the migration period (October- May) and most of the breeding period (June-August). M and F – male and female. [2] The total number of days in seawater following the days for which SWS and REM sleep were scored and quantified.

Table S1. Characteristics of fur seals and experimental conditions. Related to STAR Methods.

Parameter	Units	B (1-2)	W1-5	W6-10	W11-14	W1-14	R1	R2	ANOVA	W vs B	R1 vs W R2 vs W	R1 vs B R2 vs B
REM sleep	% 24-h	5.46±0.54 (n=4)	0.06±0.04 (n=16)	0.29±0.06 (n=17)	0.21±0.06 (n=5)	0.18+0.04 (n=38)	7.43±0.99 (n=4)	6.64±1.24 (n=4)	p=3,21E-13, F13,33=27.506	W_{1-11}<B***	R_1 >W_{1-11}*** R_2>W_{1-11}***	ns, p=.381 ns, p=.959
	% B	100	1.2±0.8	5.2±1.2	3.8±1.1	3.3±0.7	138±18	122±23				
SWS	% 24-h	19.4±0.8 (n=4)	13.2±2.0 (n=13)	17.6±0.8 (n=15)	17.0±2.3 (n=5)	15.8+1.0 (n=33)	31.5±1.2 (n=4)	27.8±2.0 (n=4)	p=7.63E-10, F13,28=18.961	W_1<B***	R_1 >W_{1-11}** R_2> W_{1-11}*	p<.001 p<.01
	% B	100	68±10	91±4	87±12	81±5	162±6	143±5				
BSWS	% 24-h	7.5±1.2 (n=4)	0.5±0.2 (n=13)	1.3±0.4 (n=13)	1.0±0.5 (n=5)	0.9±0.2 (n=31)	17.0±1.8 (n=4)	15.0±2.1 (n=4)	p=1.18E-09 F12,25=22.868	W_1<B** $W_{2,5}$<B*	R_1 >W_{1-11}*** R_2> W_{1-11}***	p<.001 p<.01
	% B	100	7±3	17±5	13±6	12±3	227±25	200±28				
USWS	% SWS	62.0±5.3 (n=4)	96.0±1.8 (n=12)	91.8±2.5 (n=13)	94.0+2.4 (n=5)	93.8±1.4 (n=30)	42.9±3.4 (n=4)	44.9±7.2 (n=4)	p=2.03E-08, F12,25=17.540	W_{1-10}>B*	R_1 <W_{1-11}*** R_2<W_{1-11}**	ns, p=.288, ns, p=.451
	% B	100	155±3	148±4	152±4	151±2	69±6	72±12				
Duration of REM sleep episodes												
	sec	174±53 (n=4)	26±5 (n=3)	15±2 (n=13)	16±1 (n=5)	13±2 (n=21)	108±26 (n=4)	123±39 (n=4)	p=2.68-E05, F13,33=6.508	W_{1-3}<B*** W_{4-10}<B** W_{11}<B*	R_2>W_{1-3}*	ns, p=.714 ns, p=.942
	% B	100	15.2±2.6	8.5±1.3	9.2±0.7	7.2±1.2	62.3±14.9	70.8±19.7				
Number of REM sleep episodes												
	per day	35±8 (n=4)	1±1 (n=3)	5±2 (n=17)	7±2 (n=5)	4±1 (n=38)	69+15 (n=4)	52+11 (n=11)	P=9.99E-08, F13,33=10.754	$W_{1,3}$<B*	R_1 <W_{1-11}** R_2<W_{1-11}**	ns, p=.875 ns, p=.055
		100	4.0±2.2	15.6±4.5	20.2±5.3	11.4±2.5	198.6±44.8	150±31.0				

B, baseline. B (1-2), average for two baseline days. R1 and R2, the 1st and 2nd recovery days. W_{1-14}, days after the seal has been transferred to seawater. REM sleep, rapid eye movement sleep. SWS, slow wave sleep (the total amount of asymmetrical and bilateral SWS). ASWS – asymmetrical SWS. The data are means ± SEM (in parentheses). The average values are counted for individual days (B1-2, R1 and R2) and periods (W_{1-5}, first 5 days in seawater; W_{6-10}, the 6-10th days in seawater, W_{10-14}, the 10-14th days in seawater). ANOVA, one way ANOVA followed by Tukey post hoc test. *p<0.05, **p<0.01, ***p<0.001.

Table S2. Sleep parameters in fur seals during the baseline (B), in seawater (W) and recovery (R) conditions. Related to Figure 3.

seal	Days In water	Baseline %24-h B1	Baseline %24-h B2	Baseline %24-h B1-2	Baseline Min B1-2	Recovery %24-h R1	Recovery %24-h R2	Accumulated loss on the 10th day in water Min	Accumulated loss on the 10th day in water %B1-2	Accumulated loss on the last day in water Min	Accumulated loss on the last day in water %B1-2	% of REM lost recovered R1	% of REM lost recovered R2
1	11	6.7%	5.7%	6.2%	89	10.2%	9.9%	853	954%	936	1047%	6.1%	6.0%
2	11	4.5%	4.9%	4.7%	68	7.5%	4.5%	658	972%	724	1070%	5.6%	-0.3%
3	10	4.4%	4.3%	4.4%	63	5.8%	4.9%	627	993%	627	993%	3.2%	1.1%
4	14	5.7%	7.4%	6.6%	94	6.3%	7.3%	921	976%	1292	1369%	-0.3%	0.8%
Mean (seals 1-3, n=3)		5.2%	5.0%	5.1%	73	7.8%	6.4%	713	973%	762	1037%	5.0%	2.3%
SEM		0.7%	0.4%	0.6%	8	1.3%	1.7%	71	11%	91	23%	0.9%	1.9%
Mean (seals 1-4, n=4)		5.3%	5.6%	5.5%	79	7.4%	6.6%	765	974%	895	1120%	3.6%	1.9%
SEM		0.5%	0.7%	0.5%	8	1.0%	1.2%	72	8%	147	85%	1.5%	1.4%

B, baseline. B1-2, average for two baseline days. R1 and R2, the 1st and 2nd recovery days. Min, minutes. There was no significant correlation between the amounts of REM sleep lost by the last day in seawater and REM sleep rebound on R1. There were a non-significant negative regression statistics on R1 (Pearson Product Moment Correlation, r=-0.489, P=0.511, n=4) and R2 (r=0.229, P=0.77, n=4).

Table S3. Amounts of REM sleep in fur seals during baseline and recovery days after 10-14 days in seawater. Related to Figure 4.

Species	Total Sleep duration	REM duration	nonREM duration	Waking duration
Carnivores				
Eastern American mole *Scalopus aquaticus*	8.4	2.1	6.3	16
Phalanger *Trichosurus vulpecula*	13.7	1.8	11.9	10
European hedgehog *Erinaceus eoropaeus*	10.1	3.5	6.6	14
Tenrec *Tenrec ecaudatus*	15.6	2.3	13.3	8
Greater short-tailed shrew *Blarina brevicauda*	14.9	2.3	12.6	9
Lesser short-tailed shew *Cryptotis parva*	9.1	1.4	7.7	15
Star-nosed mole *Condylura cristata*	10.3	2.2	8.1	14
Little brown bat *Myotis lucfugus*	19.9	2	17.9	4
Big brown bat *Eptesicus fuscus*	19.7	3.9	15.8	4
Giant armadillo *Priodontes maximus*	18.1	6.1	12	6
Long-nosed armadillo *Dasypus novemcinctus*	17.4	3.1	14.3	7
European polecat *Mustela putorius*	14.5	6	8.5	10
Dog *Canis familiaris*	10.1	2.9	7.2	14
Cat *Felis catus*	12.5	3.2	9.3	12
Red fox *Vulpes vulpes*	9.8	2.4	7.4	14
Omnivores				
Tree shrew *Blarina brevicauda*	8.9	2.6	6.3	15
Squirrel monkey *Saimiri sciureus*	9.6	1.4	8.2	14
North American opossum *Didelphis marsupialis*	18	4.9	13.1	6
Thick-tailed opossum *Lutreolina crassicaudata*	19.4	6.6	12.8	5
Galago *Galago senegalensis*	9.8	1.1	8.7	14
Owl monkey *Aotus trivirgatus*	17	1.8	15.2	7
Grivet *Ceropithecus aethiops*	10	0.7	9.3	14
Patas monkey *Erythrocebus patas*	10.9	1.1	9.8	13
Macaque *Macaca mulatta*	10.1	1.2	8.9	14
Baboon *Papio papio*	9.4	1	8.4	15
Chimpanzee *Pan troglodytes*	9.7	1.4	8.3	14
Man *Homo sapiens*	*8*	1.9	6.1	16
Pig *Sus scrofa*	9.1	2.4	6.7	15
Thirteen-lined ground squirrel Spermophilus tridecemlineatus	13.8	3.4	10.4	10
Cotton rat *Sigmodon hispdus*	11.3	1.1	10.2	13
Mole rat *Spalax leucodon*	10.6	2.4	8.2	13
Norway rat *Rattus norvegicus*	13	2.4	10.6	11

House mouse *Mus musculus*	12.5	1.4	11.1	12
Genet *Genet genetta*	6.3	1.3	5	18
Herbivores				
Horse *Equus caballus*	2.9	0.6	2.3	21
Donkey *Equus asinus*	3.1	0.4	2.7	21
Domestic cattle *Bos taurus*	4	0.7	3.3	20
Goat *Capri hircus*	5.3	0.6	4.7	19
Sheep *Ovis aries*	3.8	0.6	3.2	20
Three-toed sloth *Bradypus tridactylus*	14.4	2.2	12.2	10
Rock hyrax *Procavia habessinica*	5.4	0.5	4.9	19
Gray hyrax *Heterohyrax bruccei*	6.3	0.6	5.7	18
Tree hyrax *Dendrohyrax validus*	5.3	0.5	4.8	19
Tapir *Tapirus terrestris*	4.4	1	3.4	20
Rabbit *Oryctolagus cuniculus*	8.4	0.9	7.5	16
Mountain beaver *Aplodontia rufa*	14.4	2.4	12	10
Golden hamster *Mesocricetus auratus*	14.3	3.1	11.2	10
Mongolian gerbil *Meriones unguiculatus*	14.2	1.9	12.3	10
Guinea pig *Cavia porcellus*	9.4	0.8	8.6	15
Chinchilla *Chinchilla laniger*	12.5	1.5	11	12
Degu Octodon *degu*	7.7	0.9	6.8	16

Across species (N=51), daily REM sleep duration increases with SWS (nonREM) duration, decreases with wakefulness (W) nonREM duration is strongly and negatively correlated with waking in land mammals. We excluded species where REM was not determined with EEG. We excluded the platypus & echidna because REM & nonREM states are mixed. See: Siegel et al. PMID: 8627382, PMCID: 10336087 and PMCID: 16251951)

CORRELATIONS		R	p
REM to nonREM		+0.488	2.79E-04
REM to W		-0.716	3.42E-09
nonREM to W		-0.959	2.12E-28
		N=51	

Table S4. Amounts of REM nonREM sleep and waking in mammals (hours). Related to Figure 3, 4 and Discussion.

Seal	Month	Average Water Temp C	REM in water (% of 24)
1, m	May 22-June 5	19.3+0.2	0.30%
2, m	April 11-23	10.7+0.1	0.24%
3, m	August 1-12	26.5+0.1	0.03%
4, f	June 19-July 7	23.4+0.1	0.17%
Pearson moment correlation between the amount of REM sleep and water temperature [1]			R^2=0.469, r=- 0.684, (p=0.949)

REM sleep was suppressed at all water temperatures. m and f, male and female. Breeding season is June-August. There was no significant correlation between the amounts of REM sleep in water in fur seals and the average water temperature during their stay in water.

Table S5. Amounts of REM sleep in fur seals, and seawater temperature and month when experiments were conducted. Related to Star Methods.