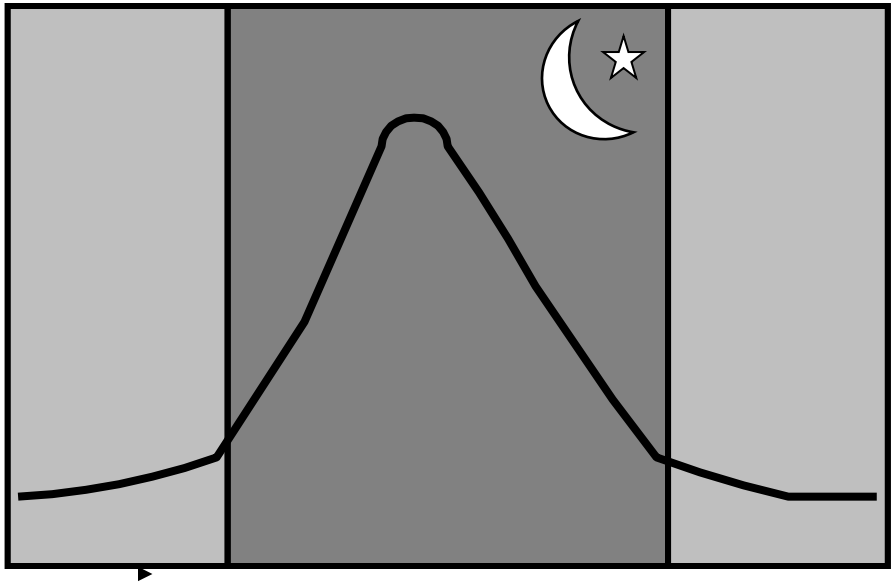
Biological rhythms

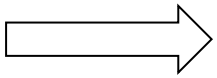


Research Importance

Medical importance: time of treatment, time of symptoms, synchronization of rhythms

Economical importance: accidents, efficiency.

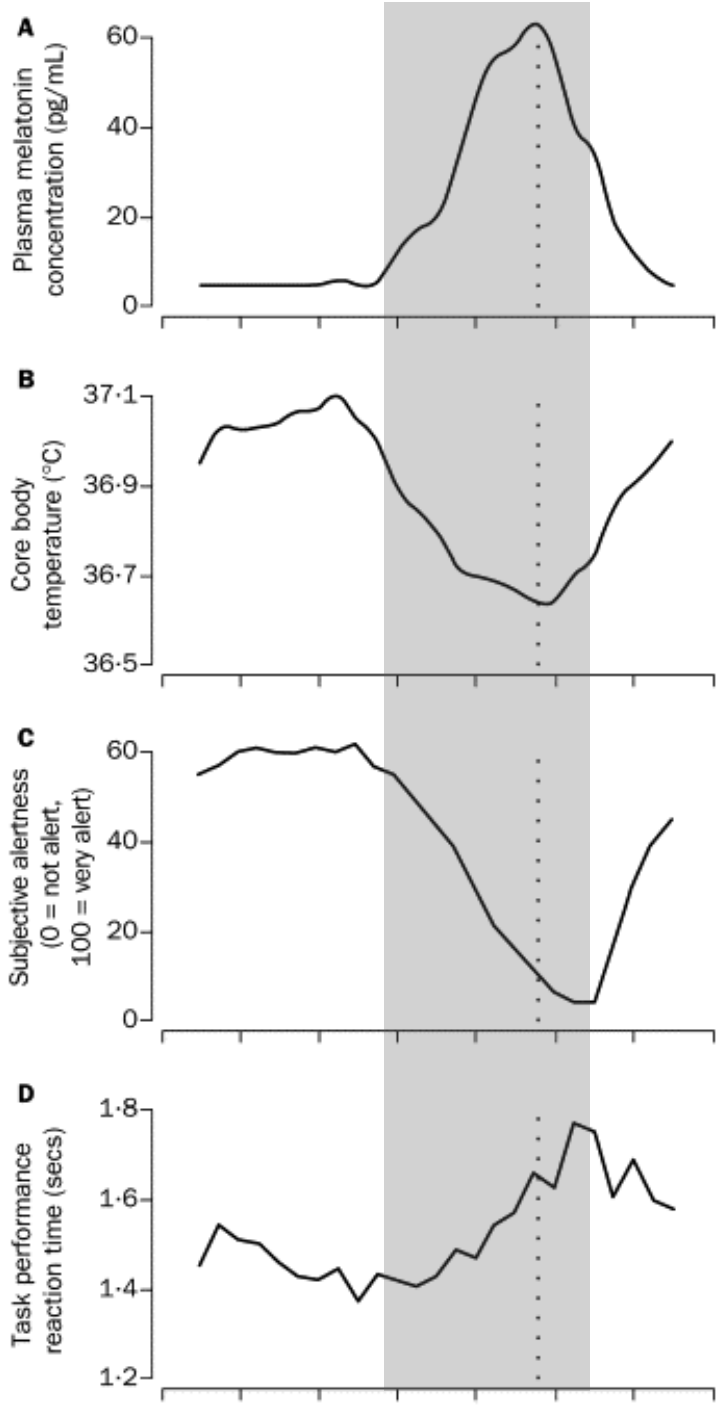
Sleep/wake cycles
Locomotor activity
Cognitive abilities
Reaction time
Body temperature
Metabolism
Hormone secretion
Enzymatic activities
Gene expression

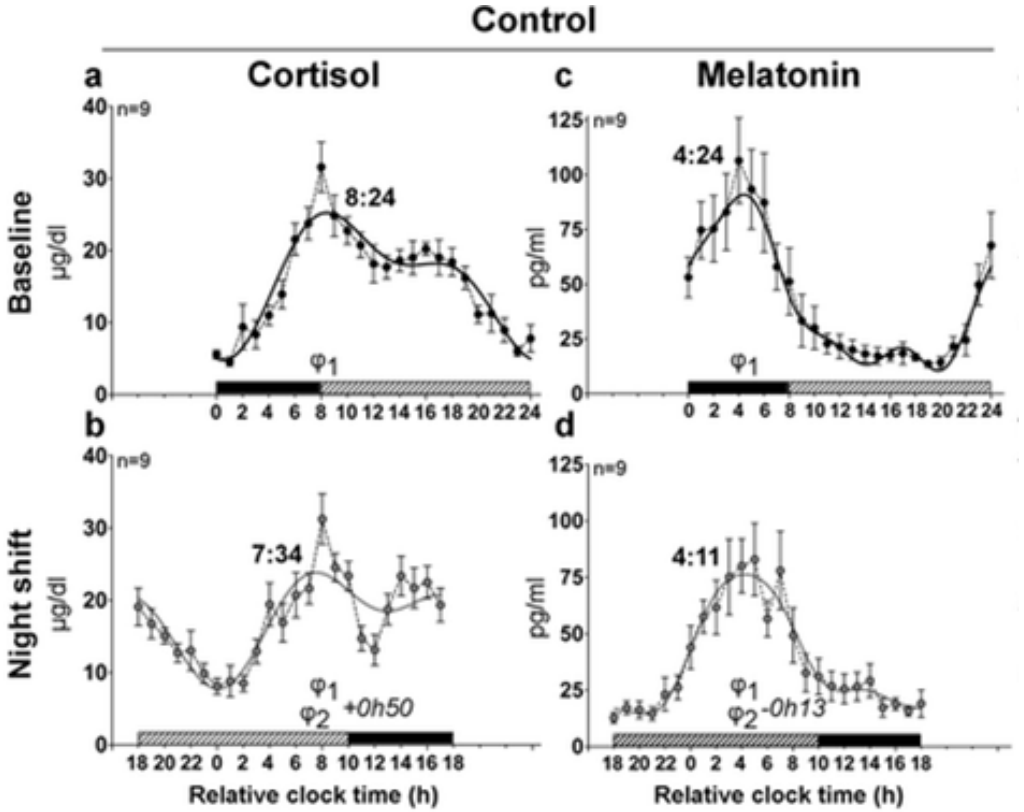


Biological clock

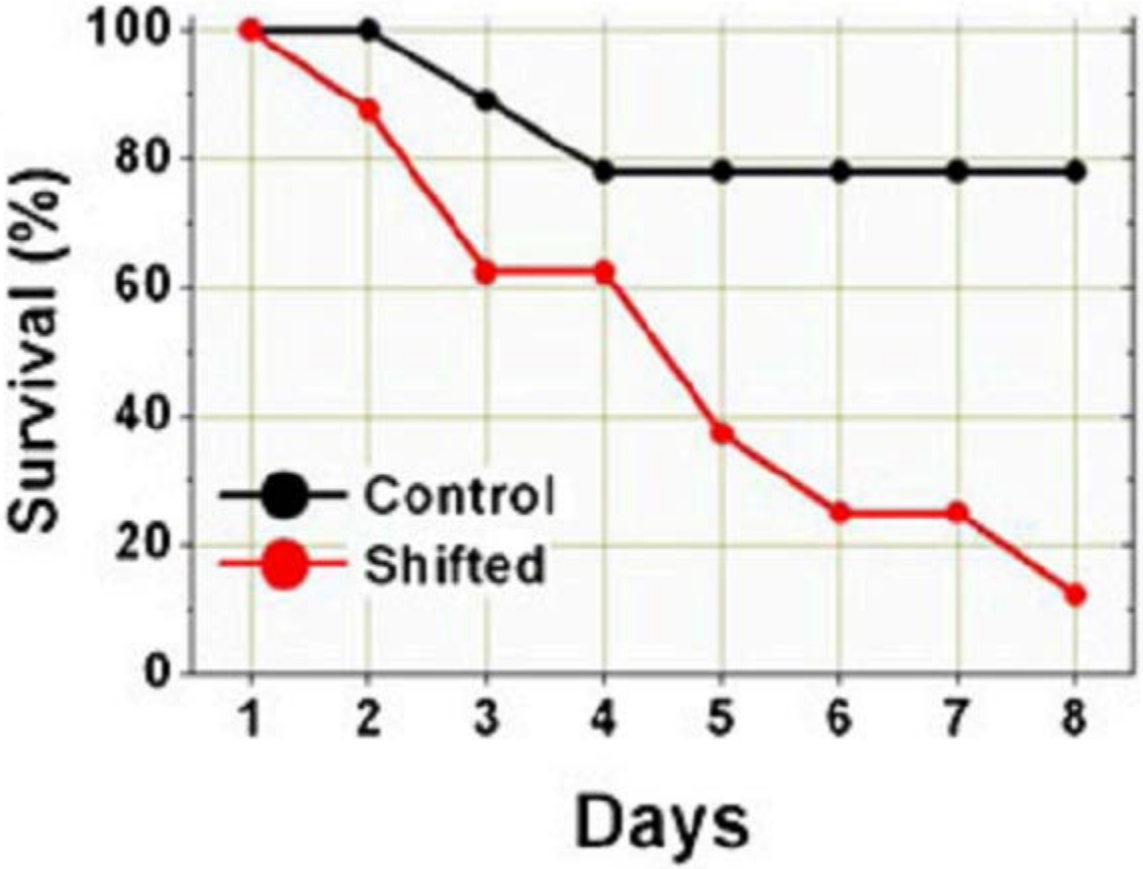
Why?

Is it important?

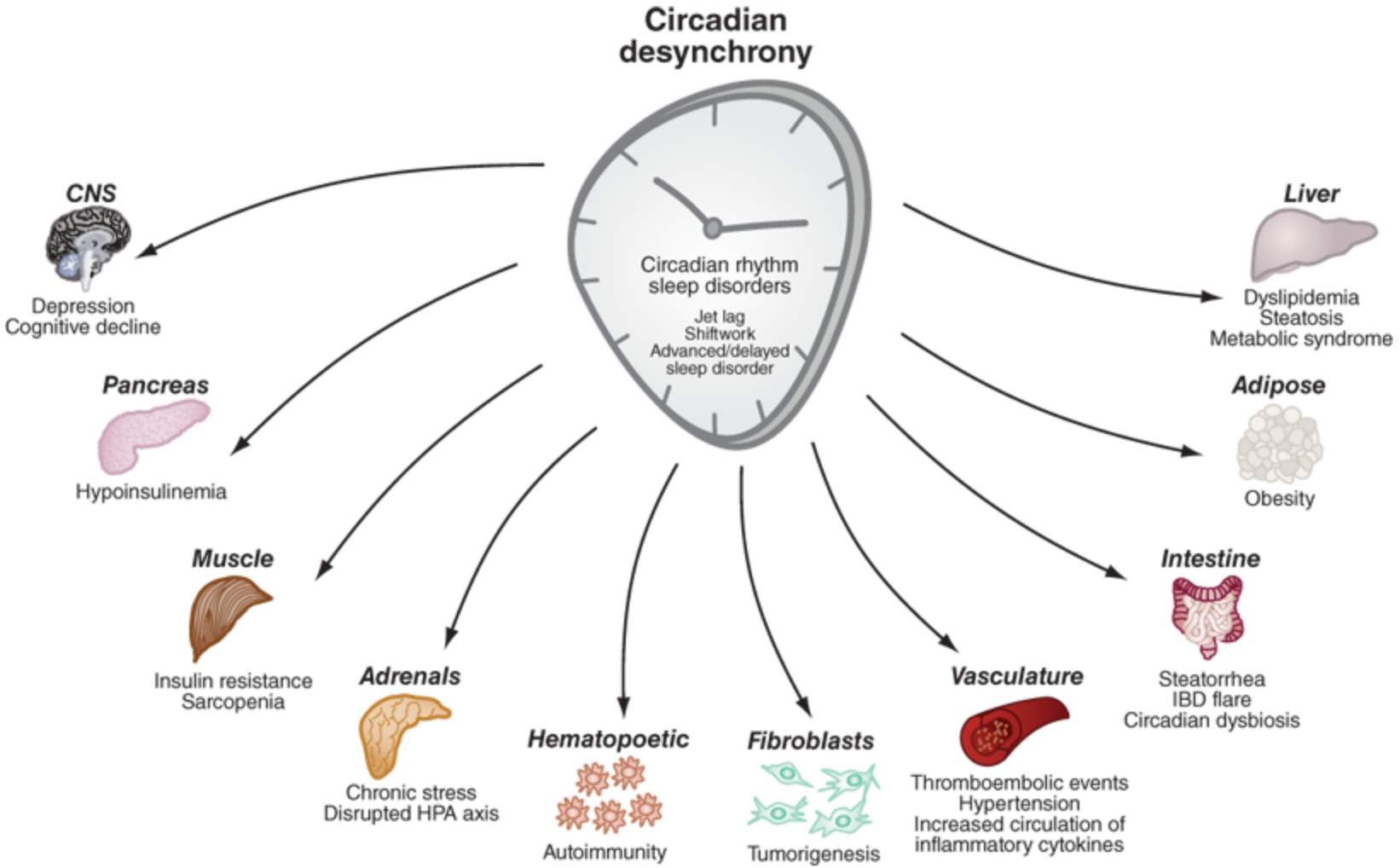

A
Plasma melatonin concentration (pg/mL)
60
40
20
0
B
Core body temperature (°C)
37·1
36·9
36·7
36·5
C
Subjective alertness (0 = not alert, 100 = very alert)
60
40
20
0
D
Task performance reaction time (secs)
1·8
1·6
1·4
1·2

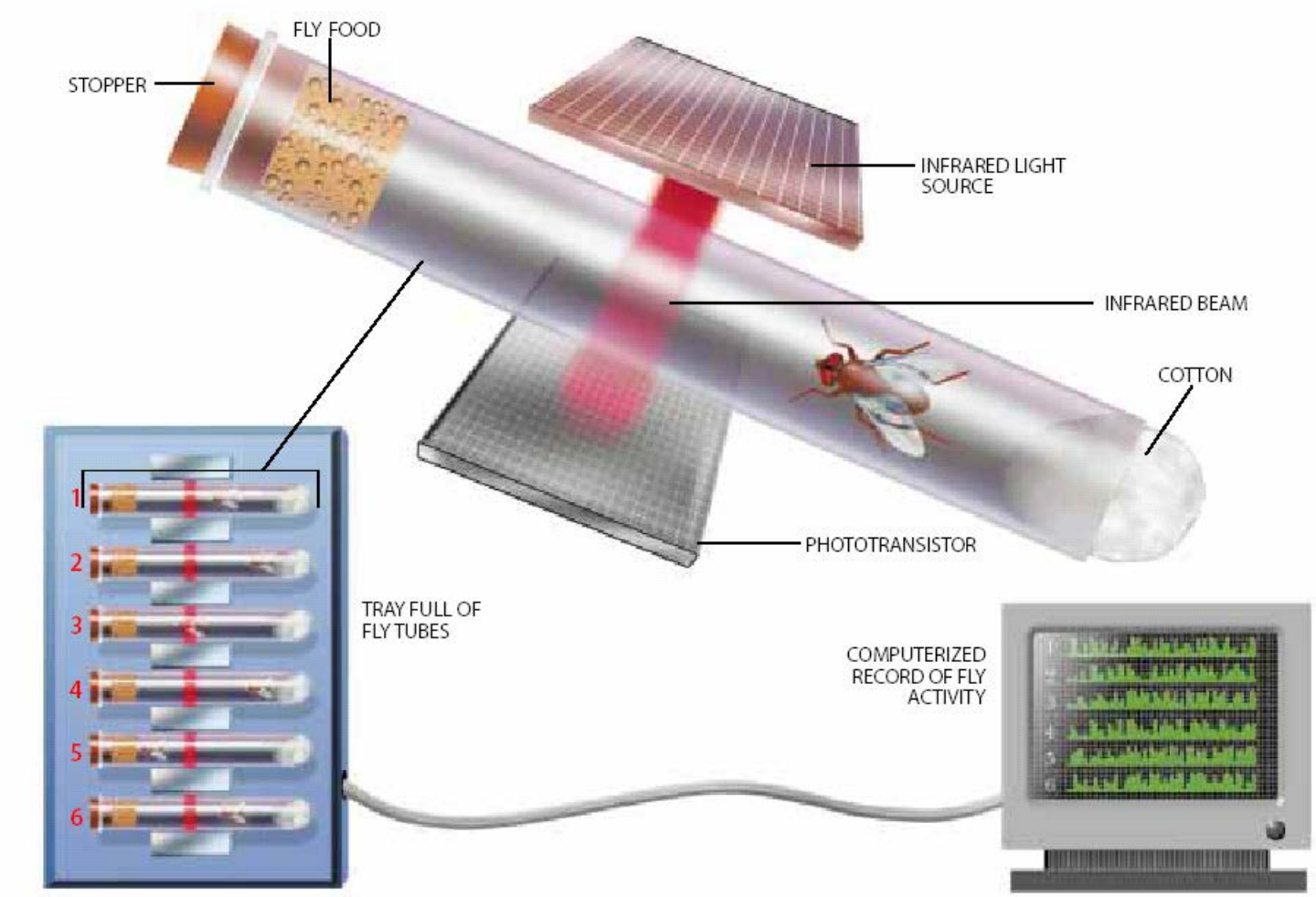

Control
a
Cortisol
c
Melatonin
Baseline
µg/dl
pg/ml
n=9
8:24
4:24
φ1
b
d
Night shift
7:34
4:11
φ2 +0h50
φ2 -0h13
0 2 4 6 8 10 12 14 16 18 20 22 24
18 20 22 0 2 4 6 8 10 12 14 16 18
Relative clock time (h)

“Shift workers” mice are less resistant to Lipopolysaccharides (LPS)-induced endotoxemic shock



Castanon-Cervantes et al., 2010

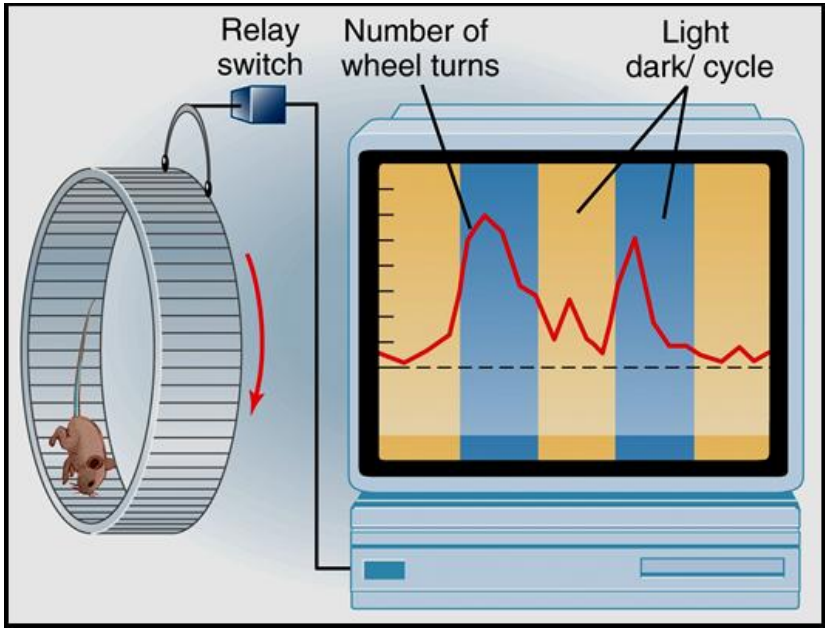


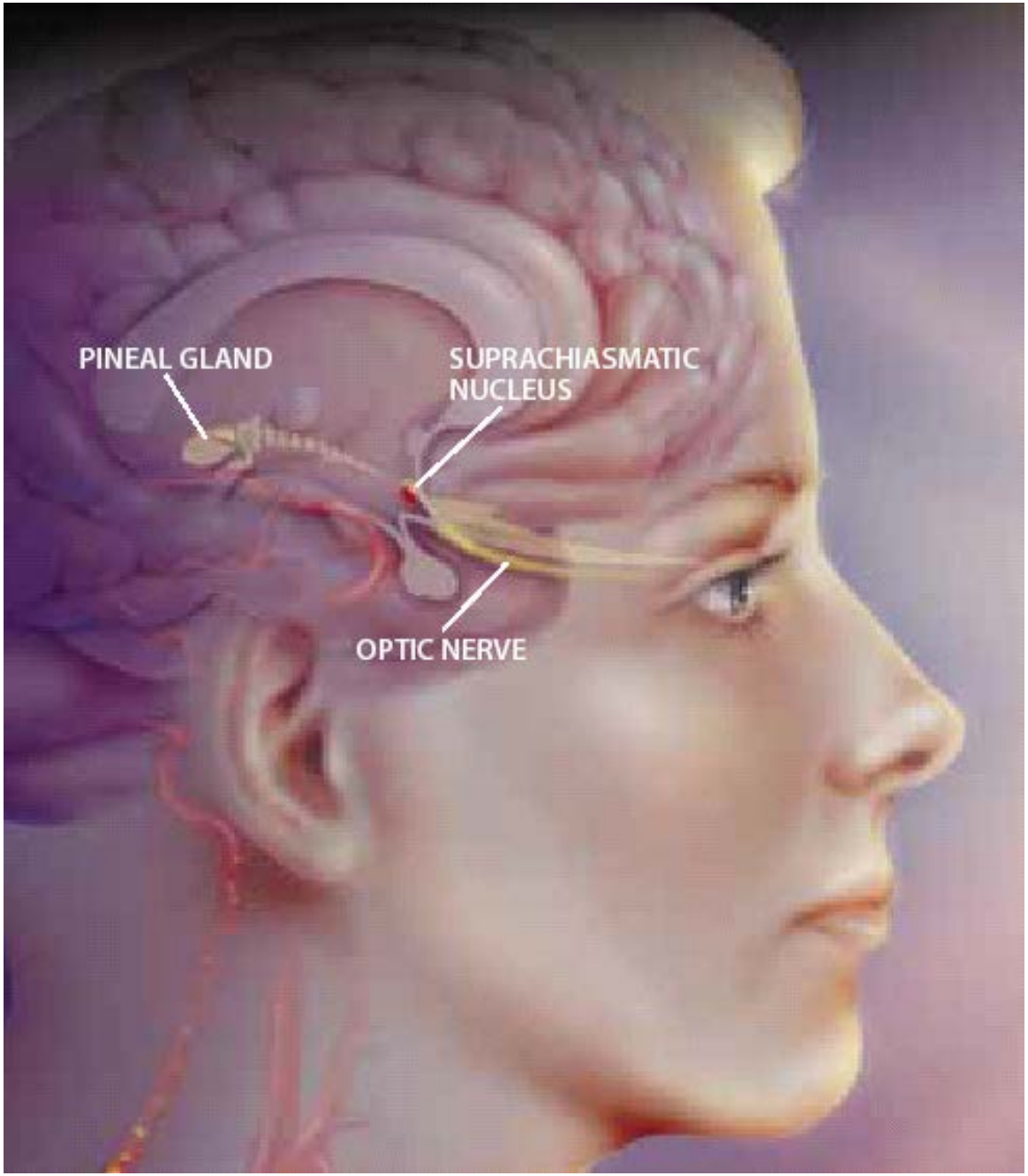

FLY FOOD
STOPPER
INFRARED LIGHT SOURCE
INFRARED BEAM
COTTON
PHOTOTRANSISTOR
1
2
3
4
5
6
TRAY FULL OF FLY TUBES
COMPUTERIZED RECORD OF FLY ACTIVITY

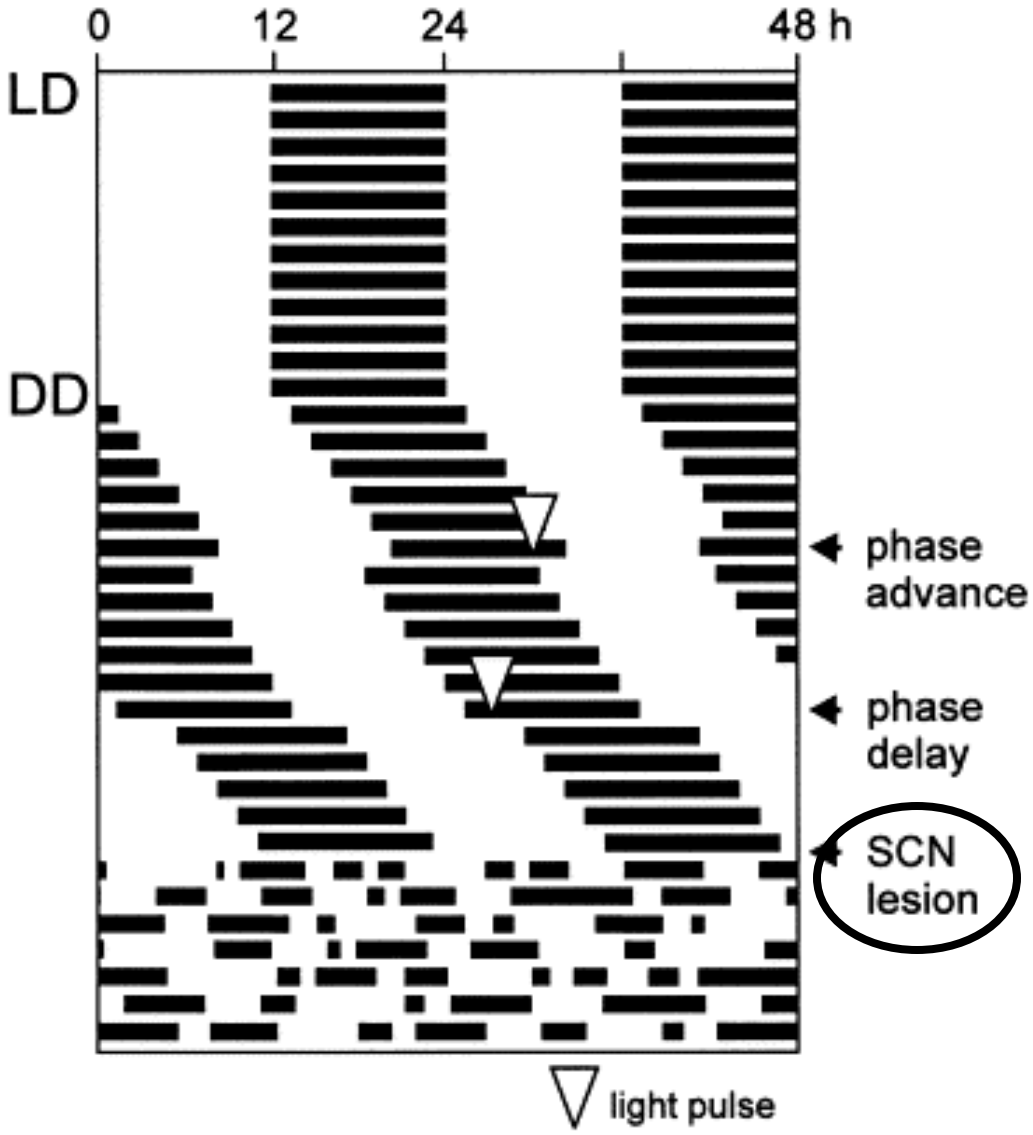
Monitoring circadian rhythms of activity in rodents.

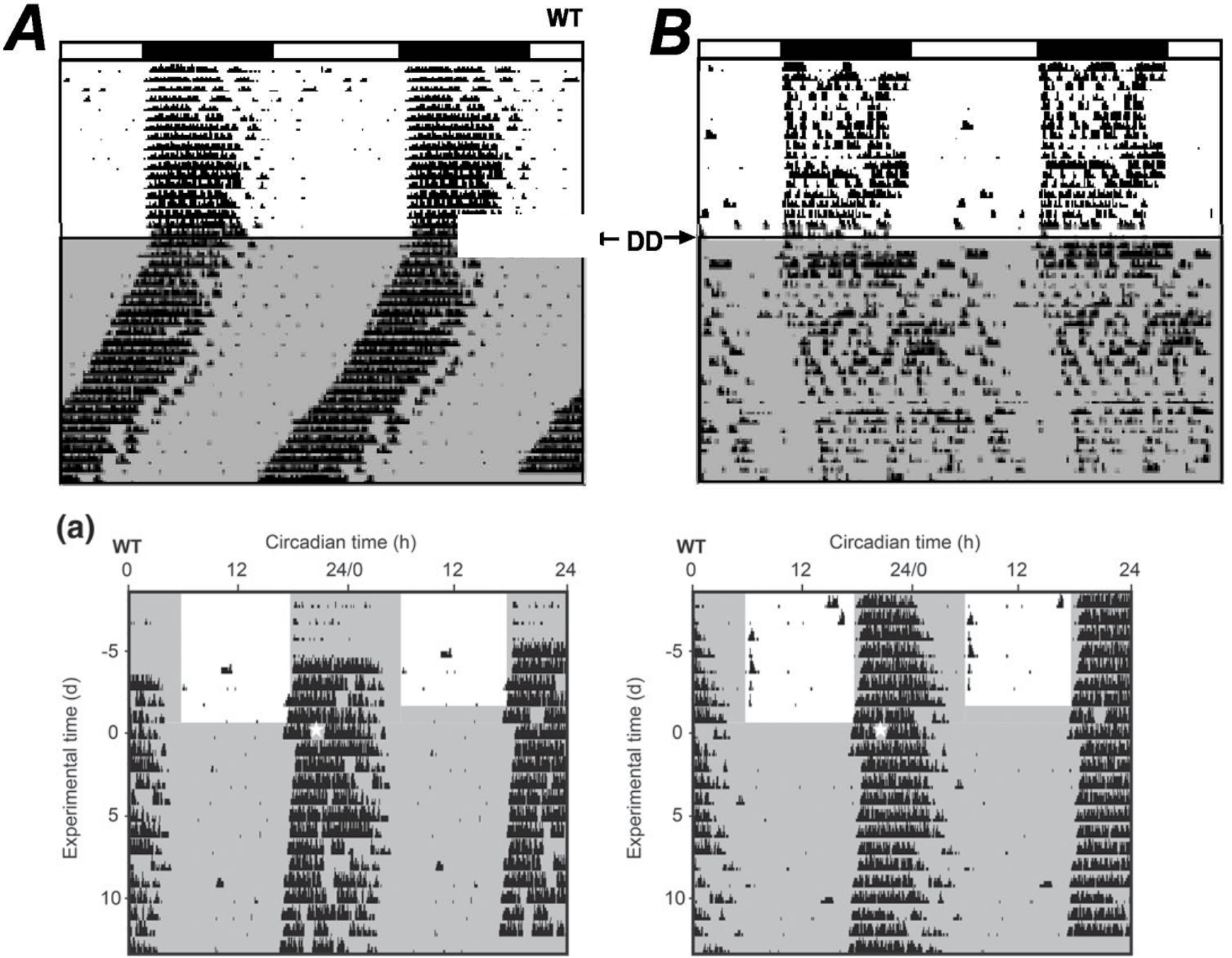
Actogram, the secret hand-shake of chronobiologists

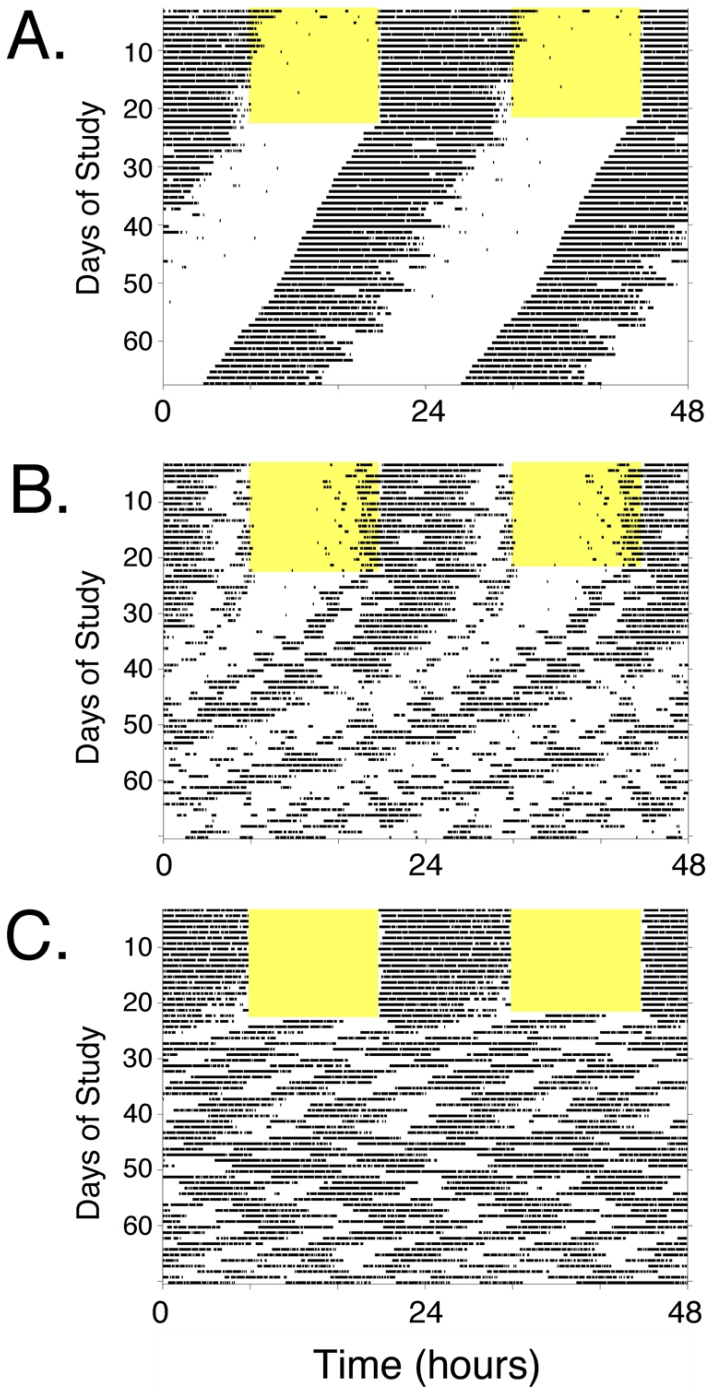
Free running



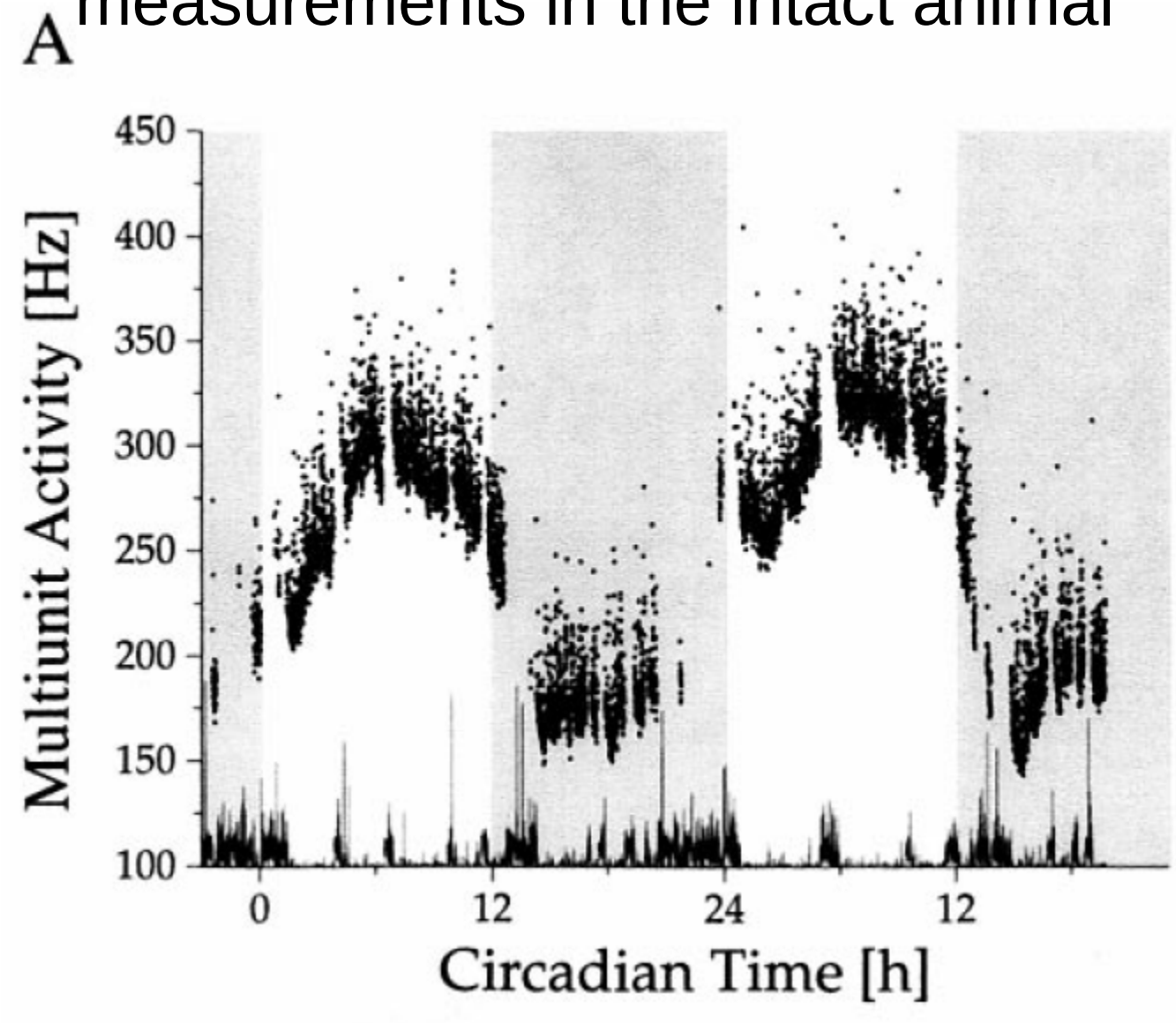

PINEAL GLAND
SUPRACHIASMATIC NUCLEUS
OPTIC NERVE


0
12
24
48 h
LD
DD
phase advance
phase delay
SCN lesion
light pulse


A
WT
B
DD
(a)
WT
Circadian time (h)
0
12
24/0
12
24
Experimental time (d)
-5
0
5
10
WT
Circadian time (h)
0
12
24/0
12
24
Experimental time (d)
-5
0
5
10

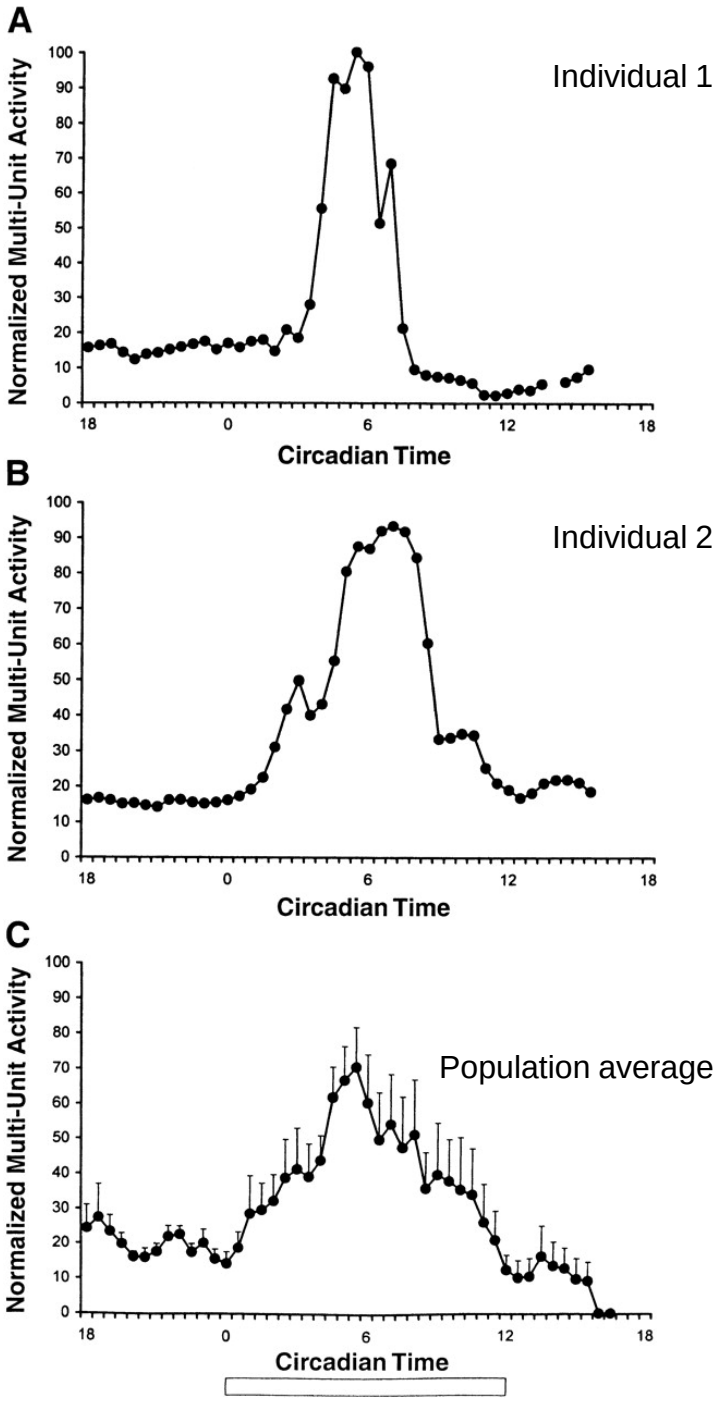

A.
B.
C.
Days of Study
10
20
30
40
50
60
0
24
48
Time (hours)

Rhythms of firing rate in the SCN, measurements in the intact animal



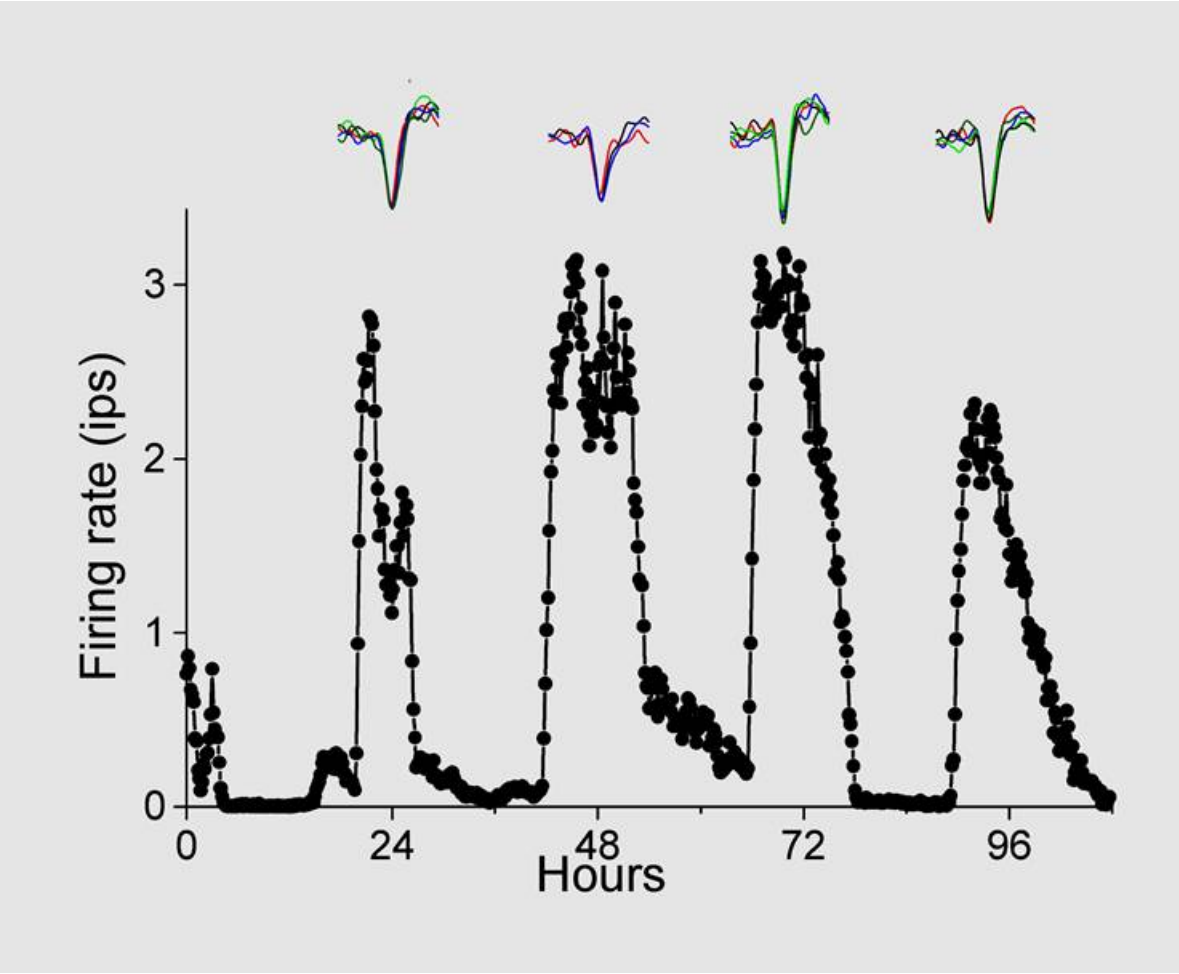
Meijer et al., 1998

Rhythms of firing rate in the SCN, measurements in tissue culture

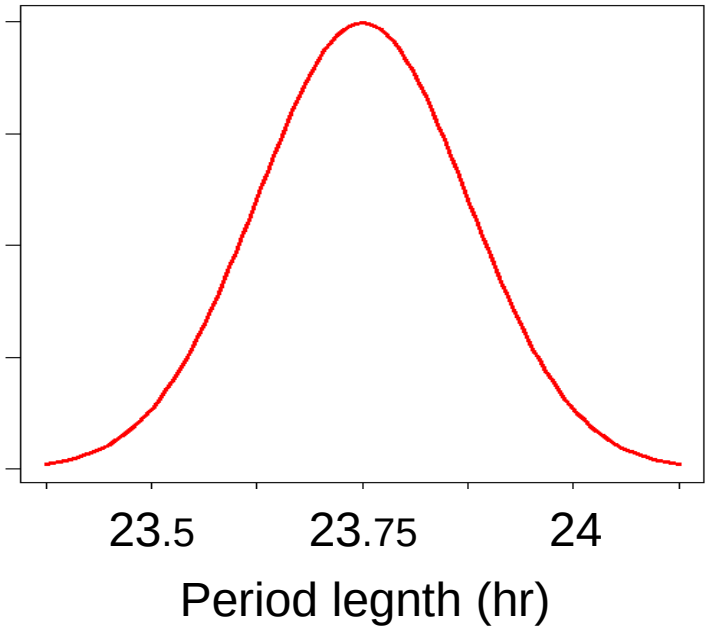


Mrugala et al., 2000

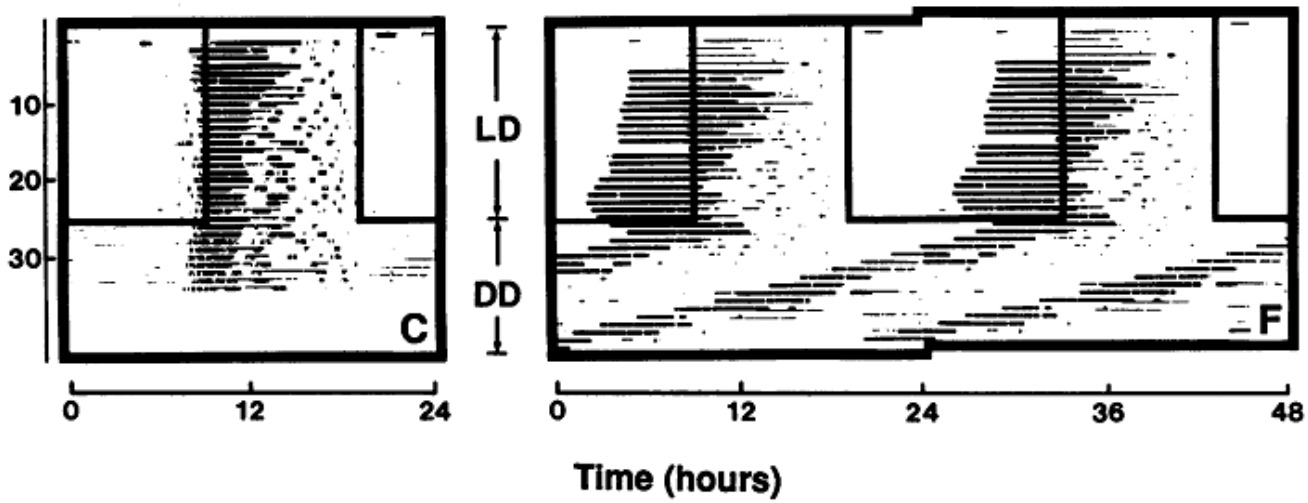
Rhythms of firing rate in the SCN, measurements in SCN cell culture

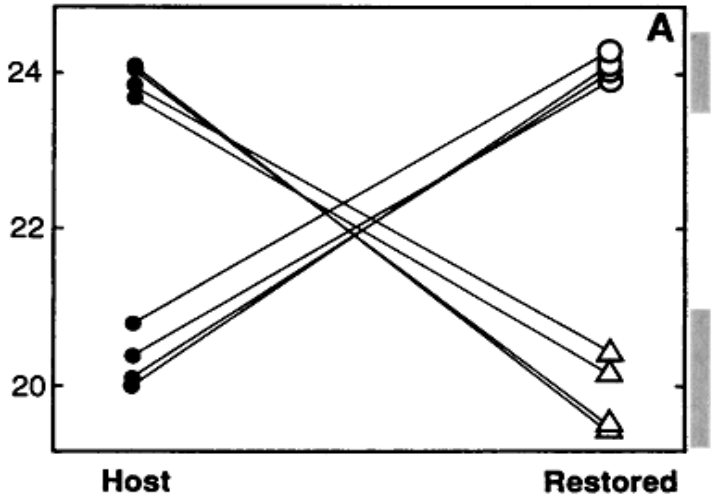


Mike Menaker

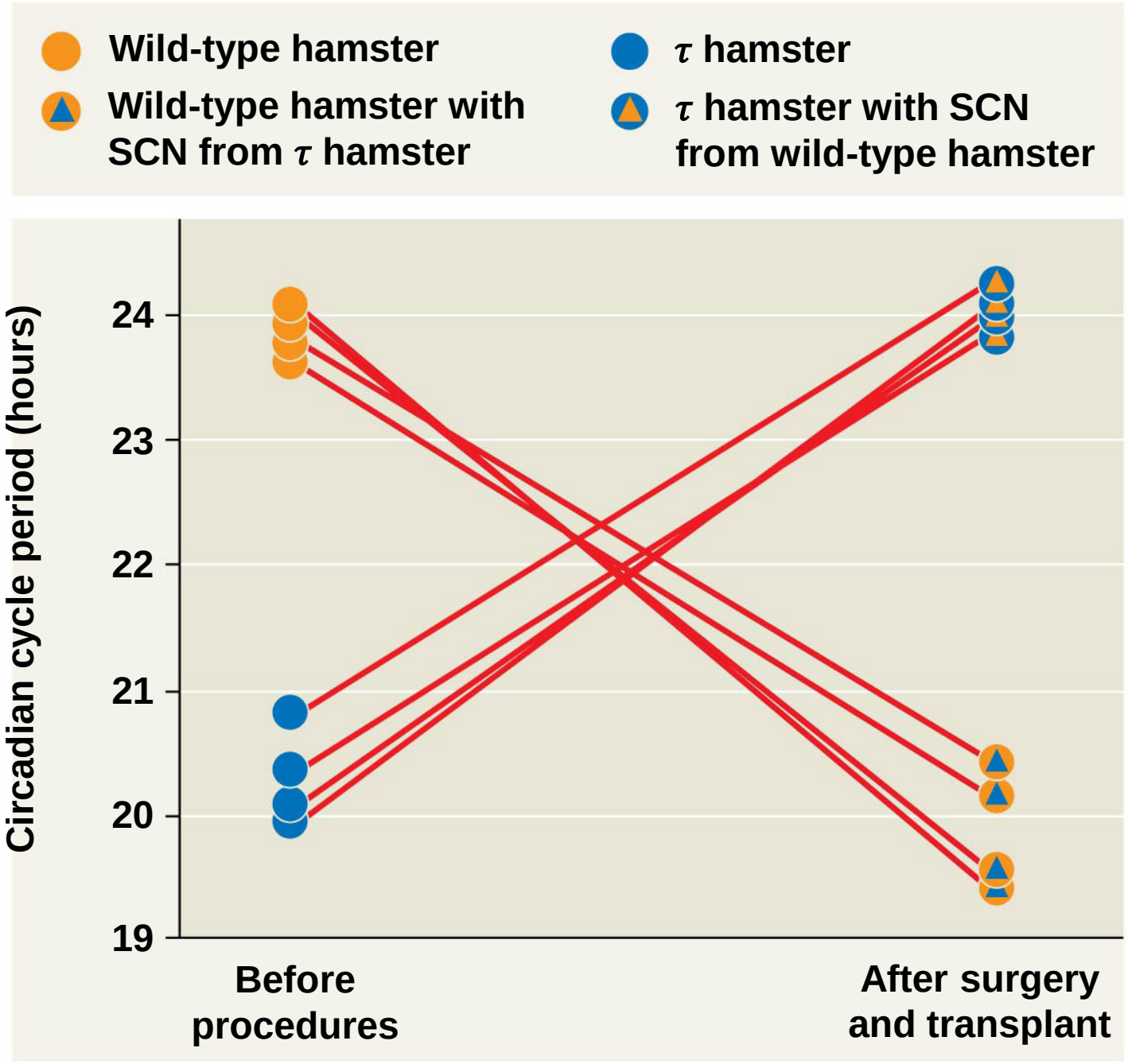


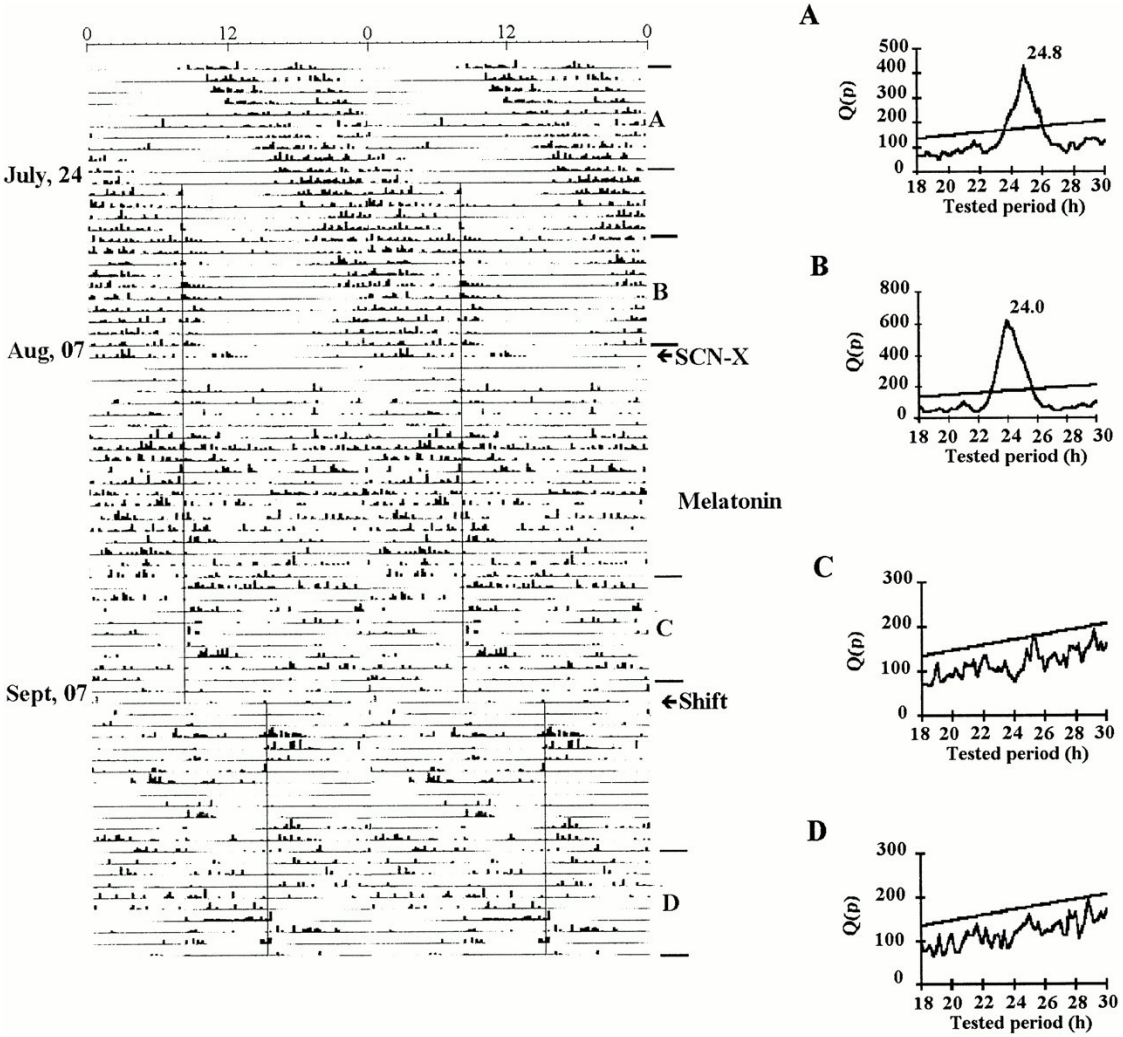
Ralph et al., 1990





Ralph et al., 1990


Wild-type hamster
Wild-type hamster with SCN from τ hamster
τ hamster
τ hamster with SCN from wild-type hamster
Circadian cycle period (hours)
24
23
22
21
20
19
Before procedures
After surgery and transplant

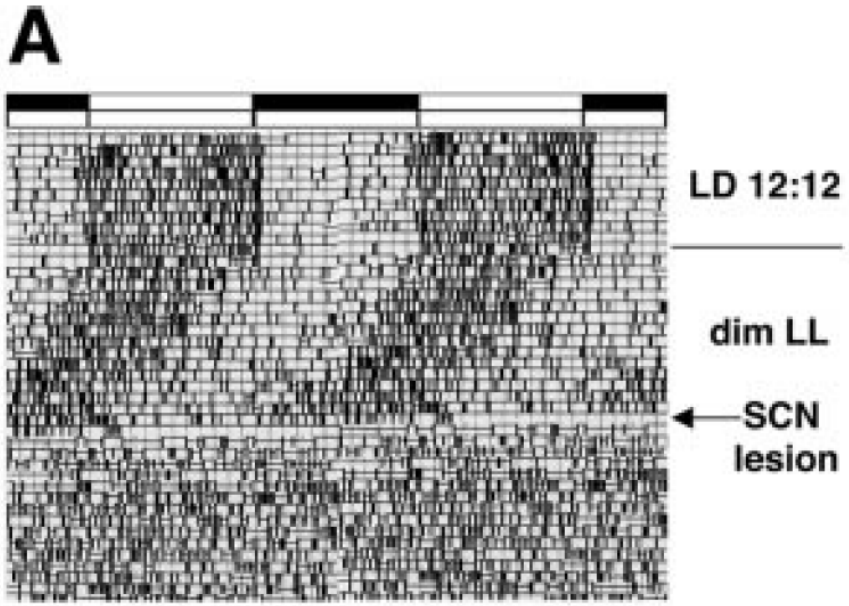


Free running

Entrainment

After SCN ablation

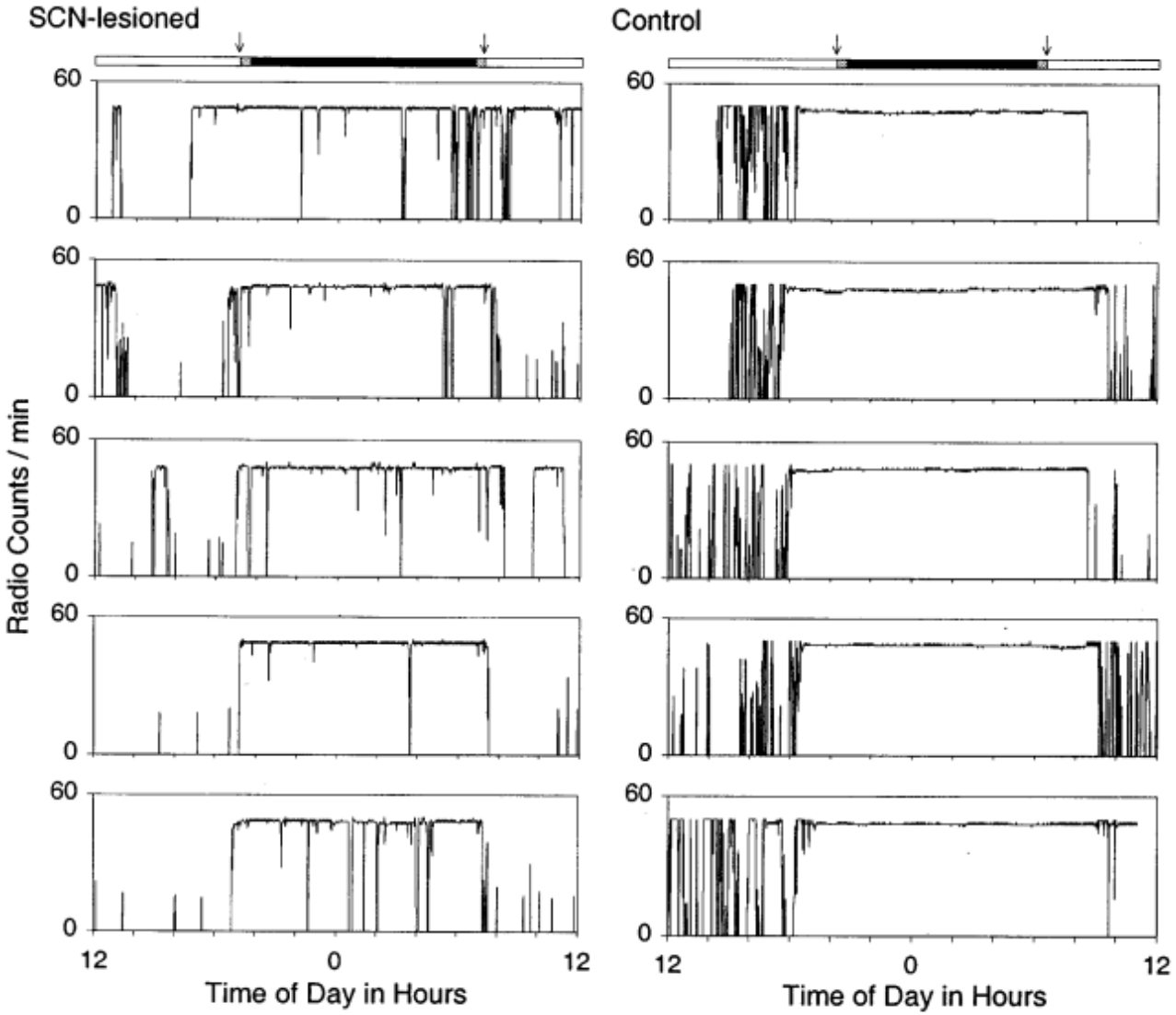
After SCN ablation

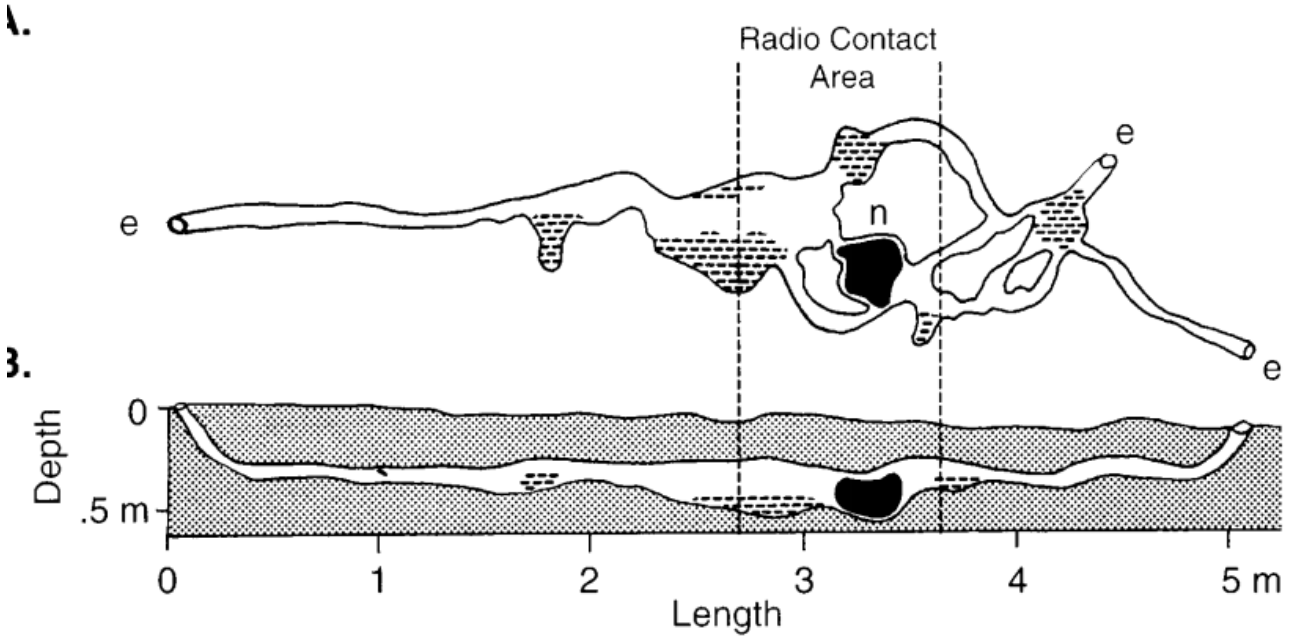

A
LD 12:12
dim LL
SCN
lesion

What are the evidence that the SCN is the central pacemaker in mammals?

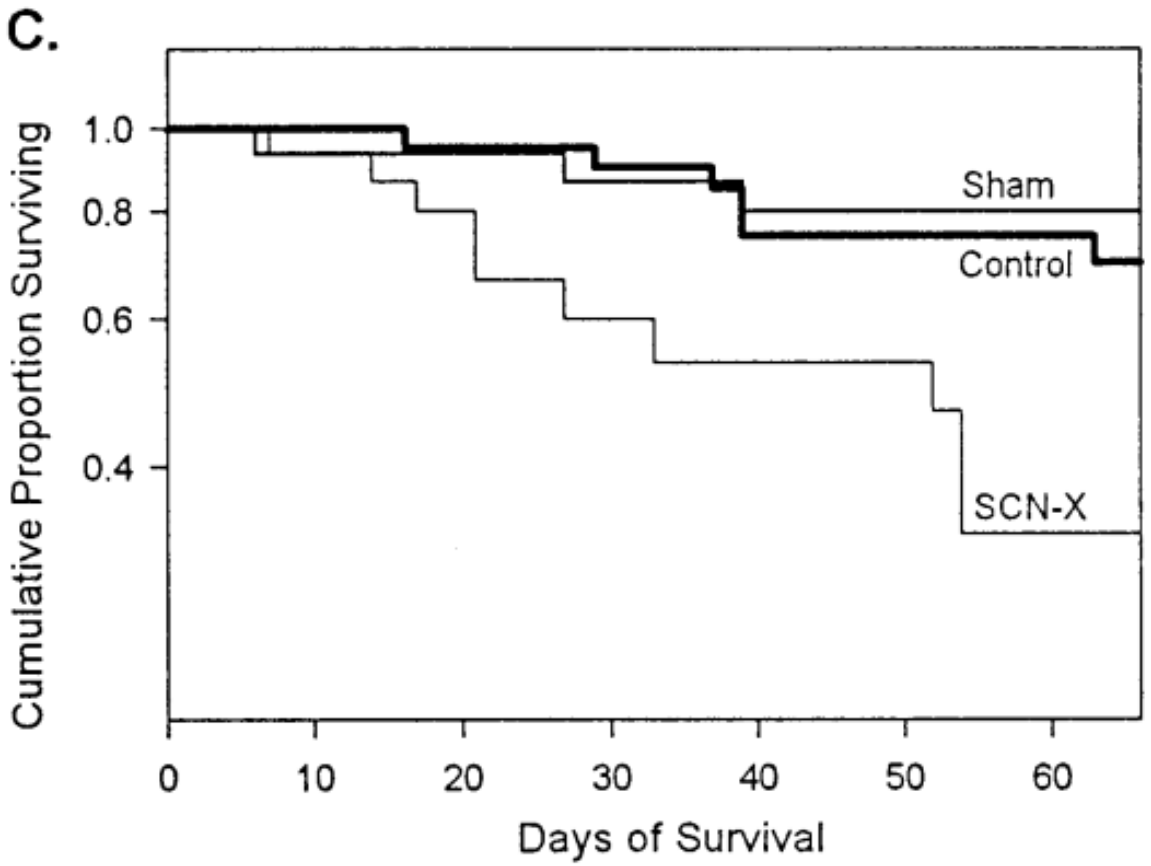
- SCN lesion led to loss of rhythms
- SCN neurons have an intrinsic clock
- Stimulation of SCN neurons synchronized rhythms
- An implanted SCN dictates the rhythm

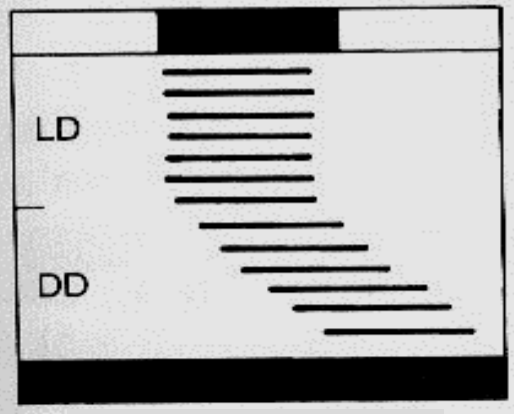
How important is the SCN? A wild experiment



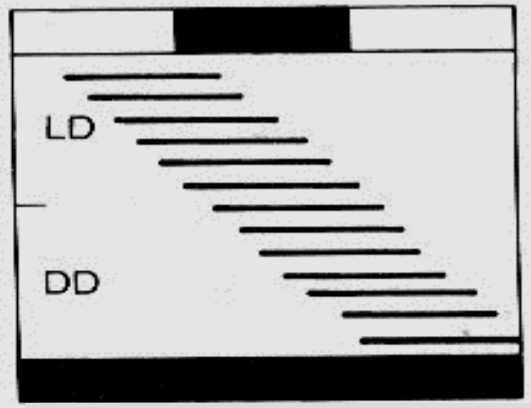

A.
Radio Contact
Area
e
n
e
e
B.
Depth
0
.5 m
0
1
2
3
4
5 m
Length

SCN-lesioned animals did not survive well

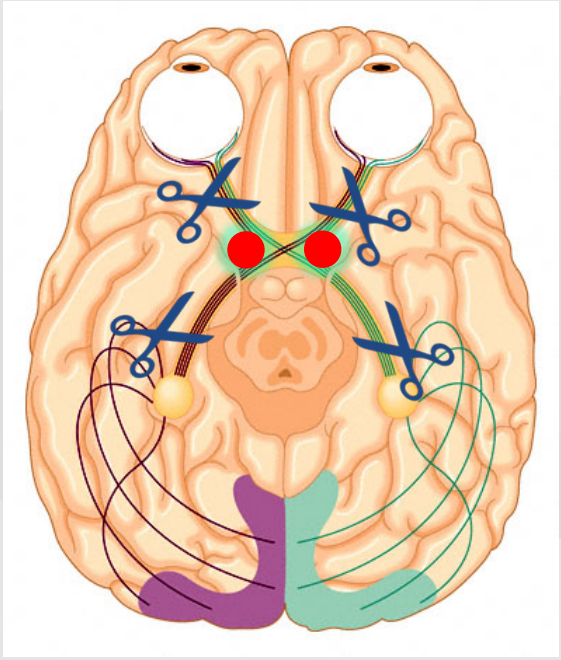




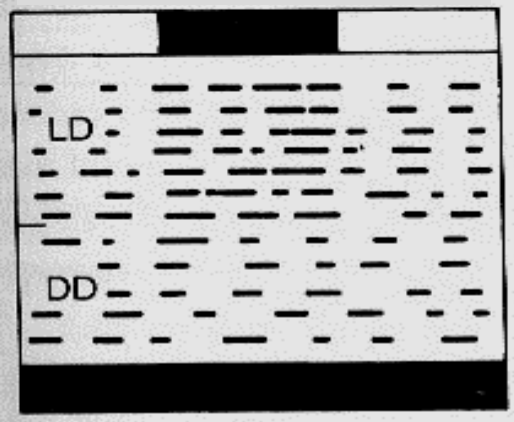
No manipulation



Pre-chiasmatic dissection of the optic nerve

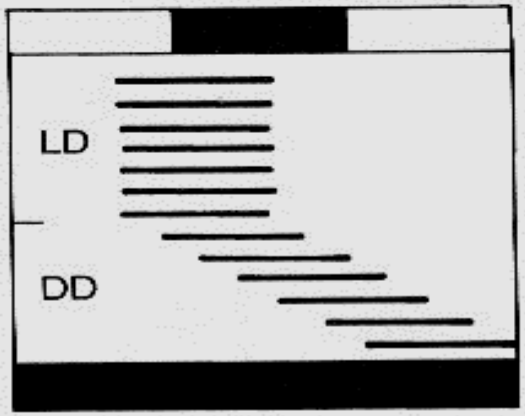


SCN lesion

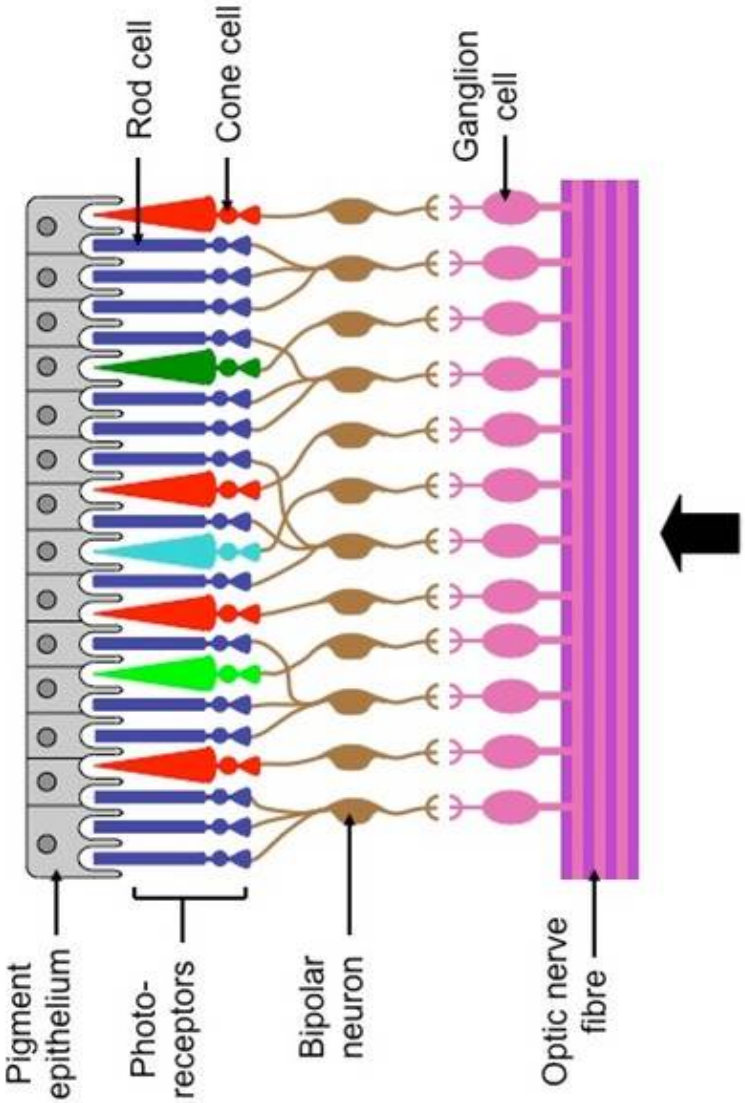


(c)

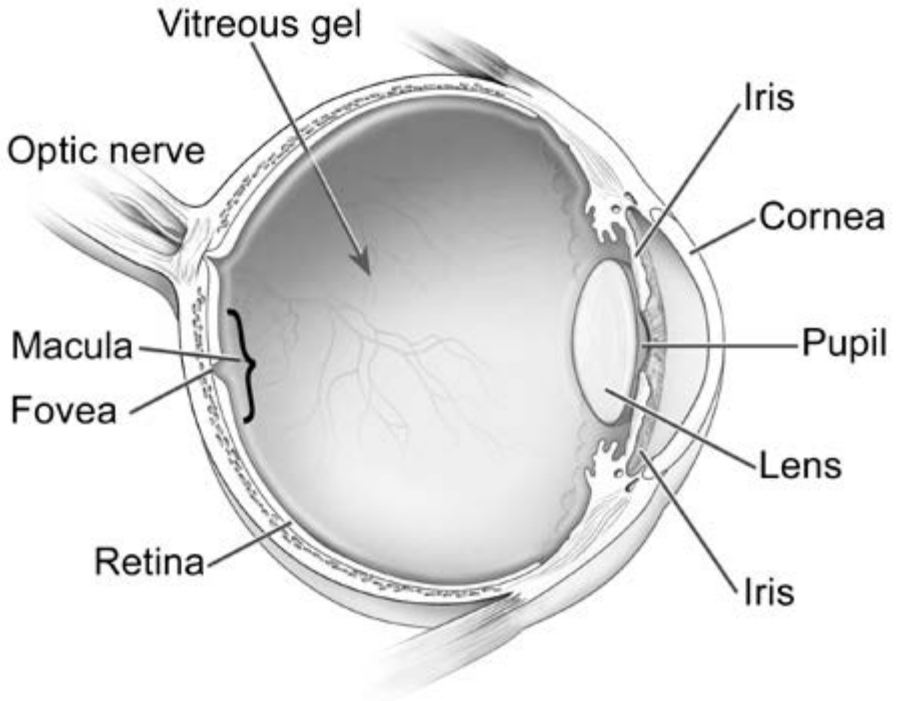
Post-chiasmatic dissection of the optic nerve

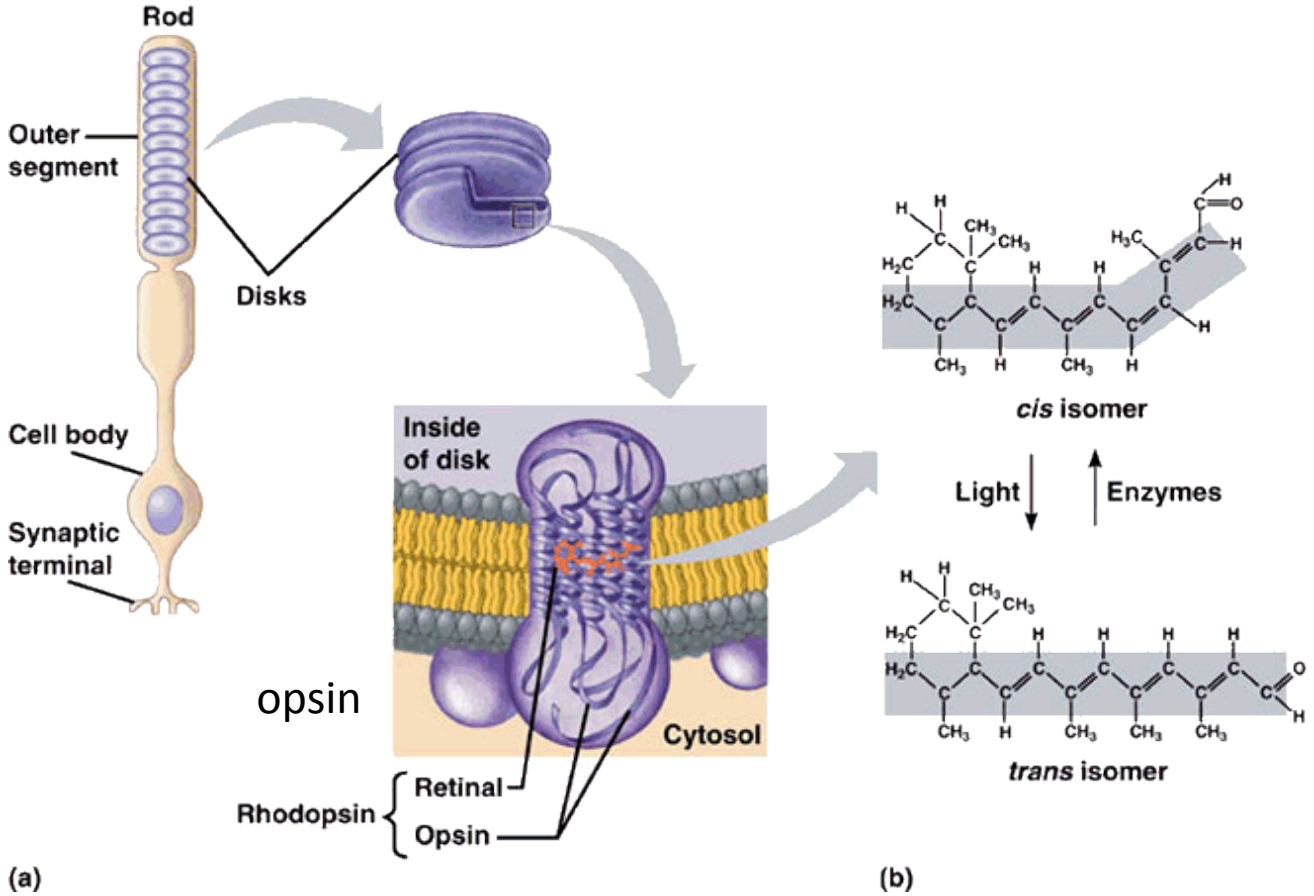


(d)

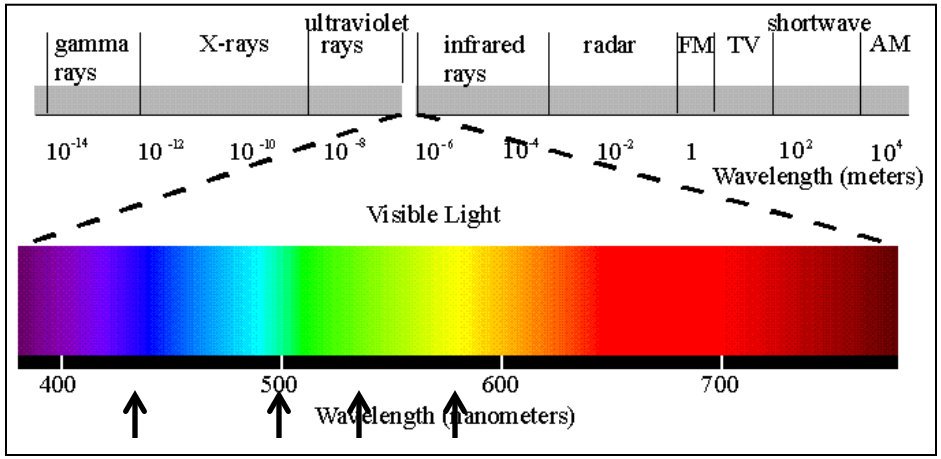

Rod cell
Cone cell
Ganglion cell
Pigment epithelium
Photo-receptors
Bipolar neuron
Optic nerve fibre
DIRECTION OF LIGHT

opsins


Vitreous gel
Optic nerve
Iris
Cornea
Macula
Fovea
Pupil
Lens
Retina
Iris

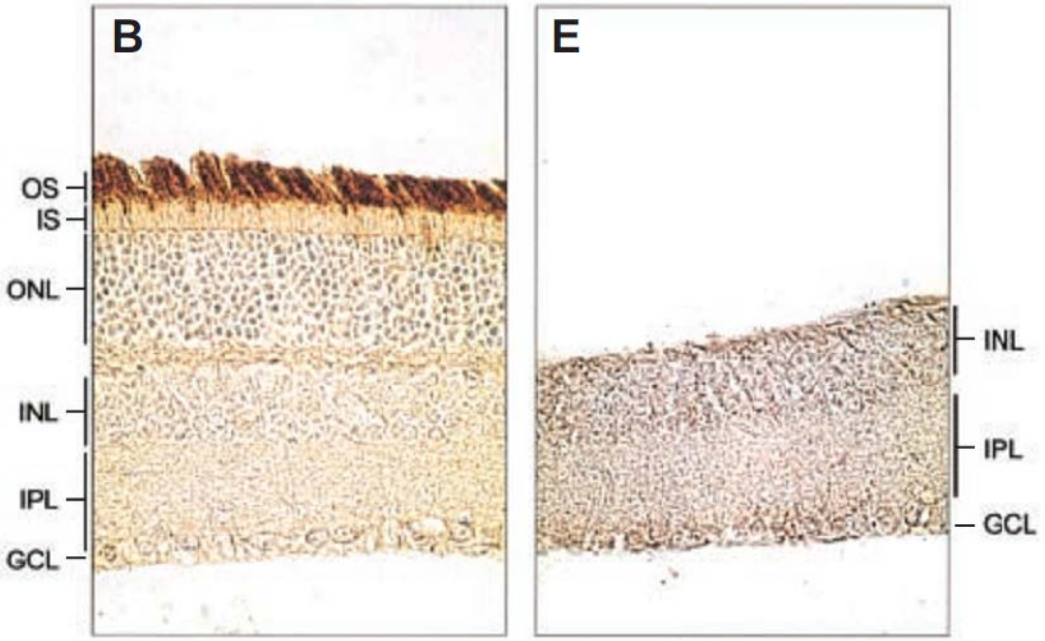


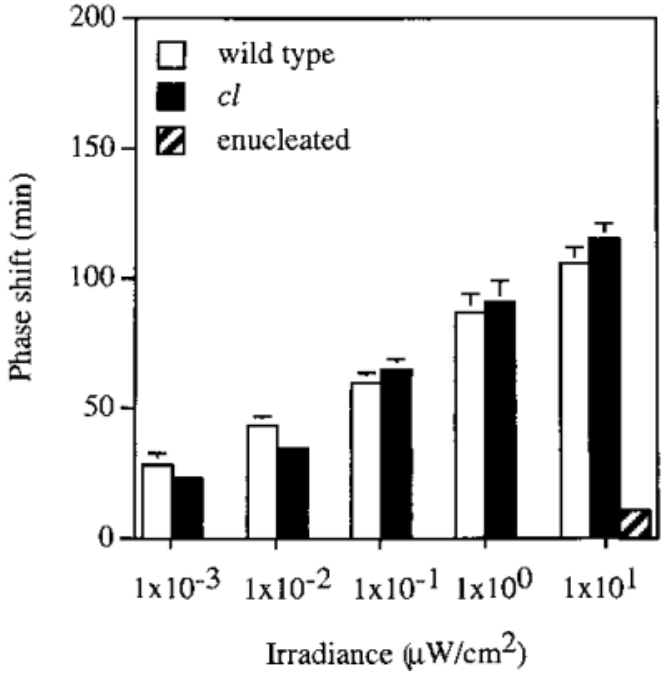
Four opsins in the human eyes are sensitive to different wave lengths

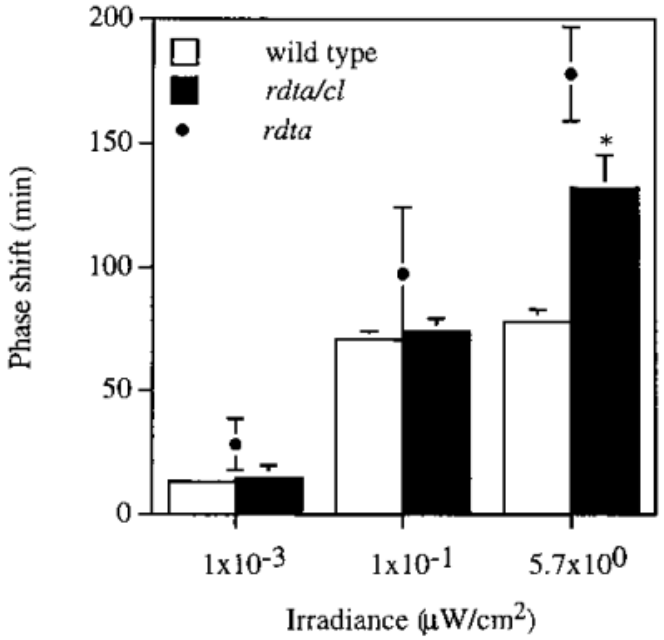


Genetic ablation of both rods and cones using diphtheria toxin did not affect light-induced phase shifts

"The eye contains additional photoreceptors that regulate the circadian clock."
Freedman et al, 1999

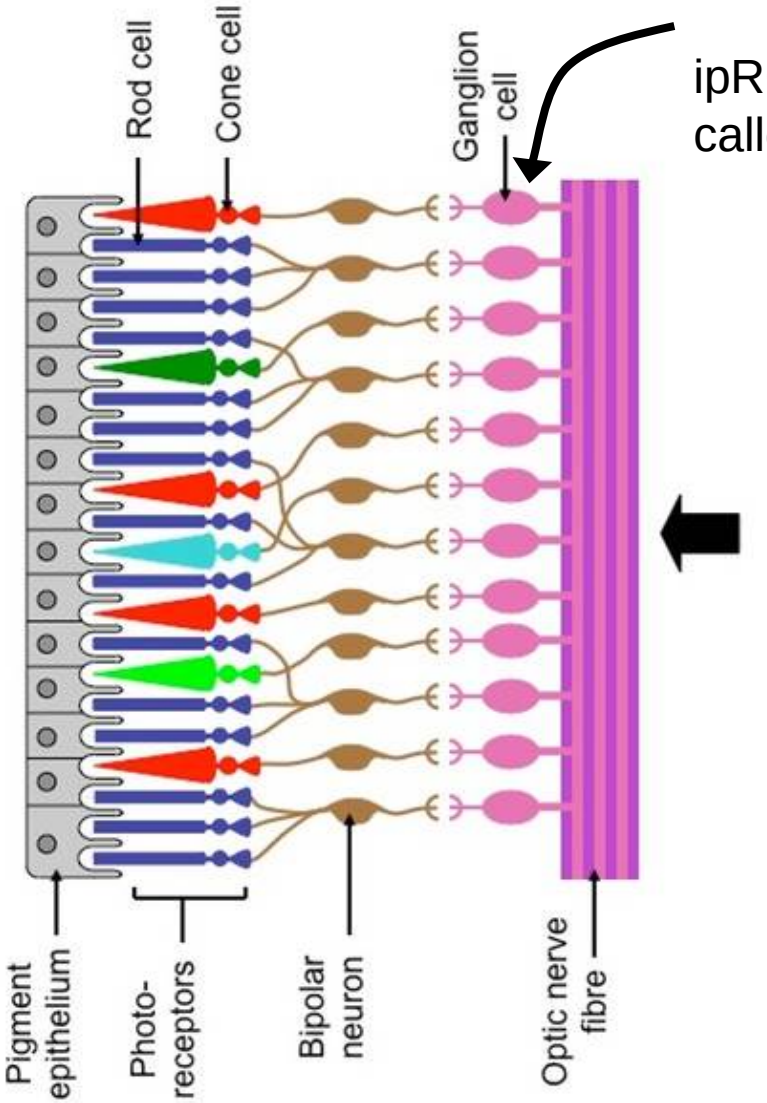




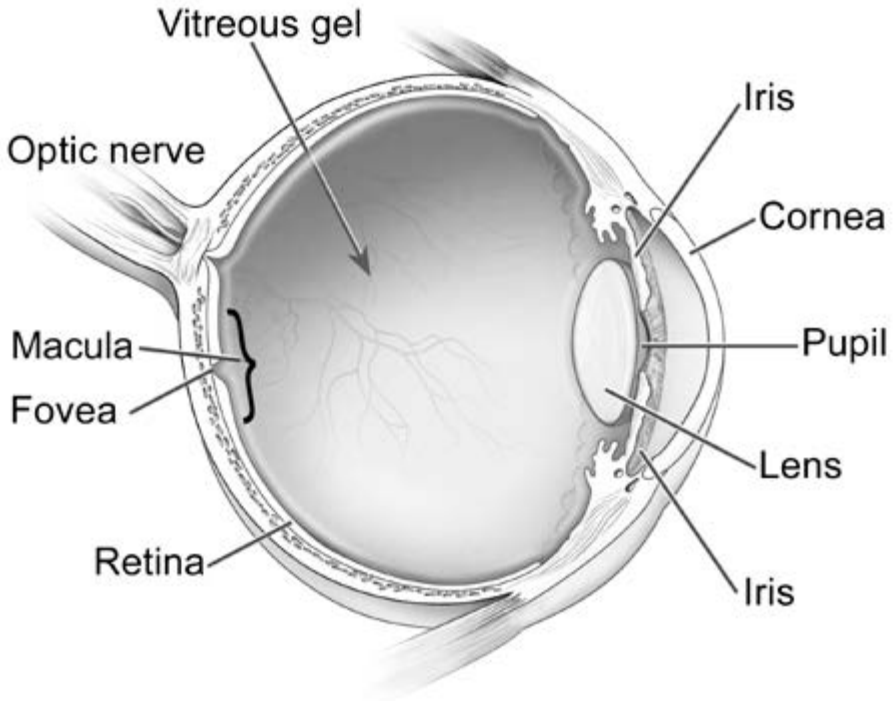


ipRGC - intrinsically photosensitive retinal ganglion cells

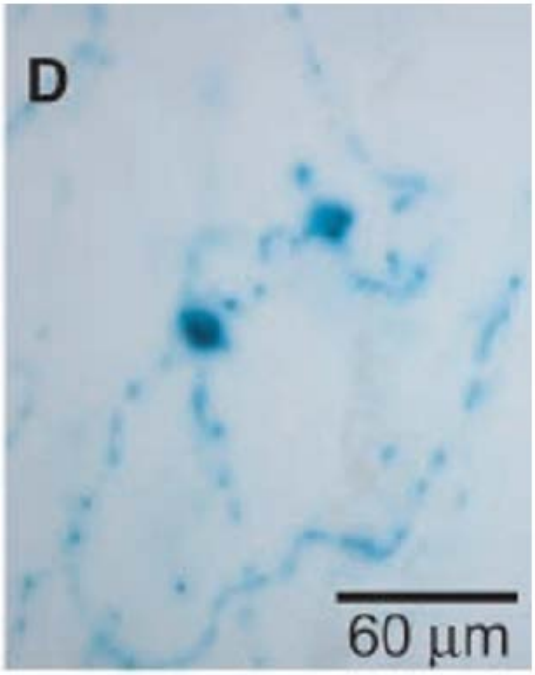
ipRGC express another form of opsin called melanopsin

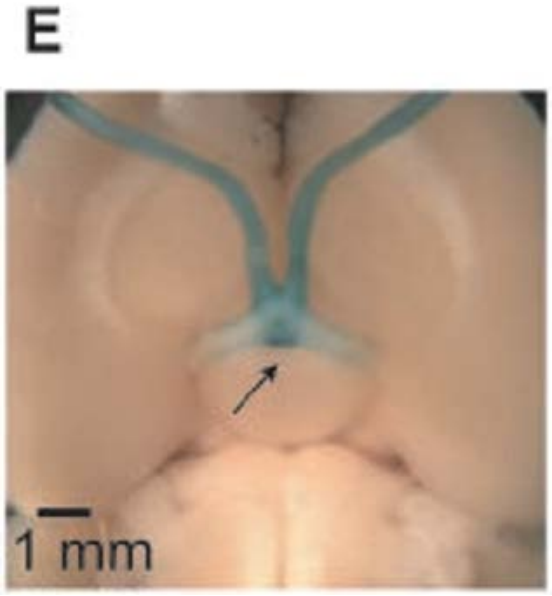


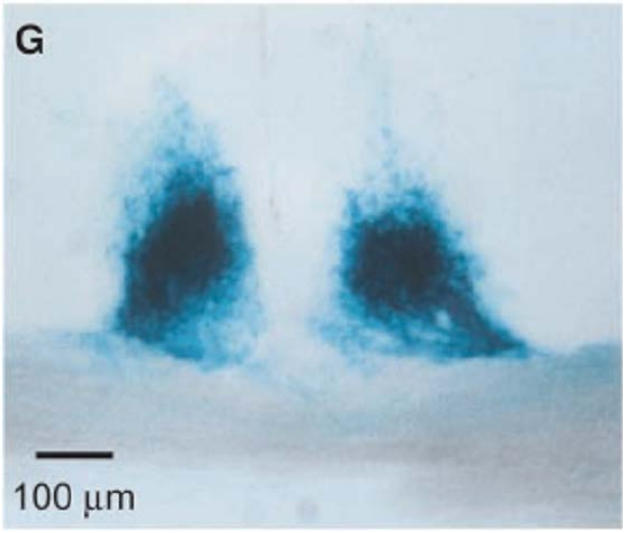
DIRECTION OF LIGHT



Transgenic mice expressing x-gal under the control of the melanopsin gene

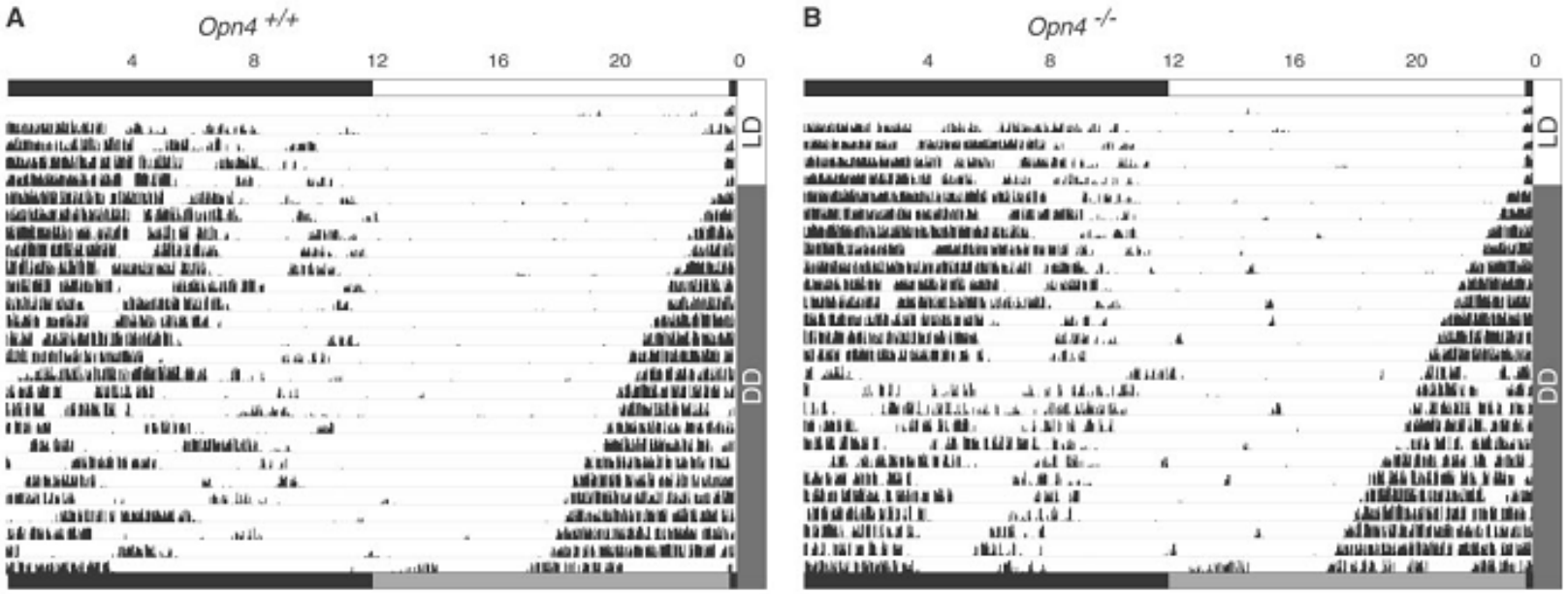






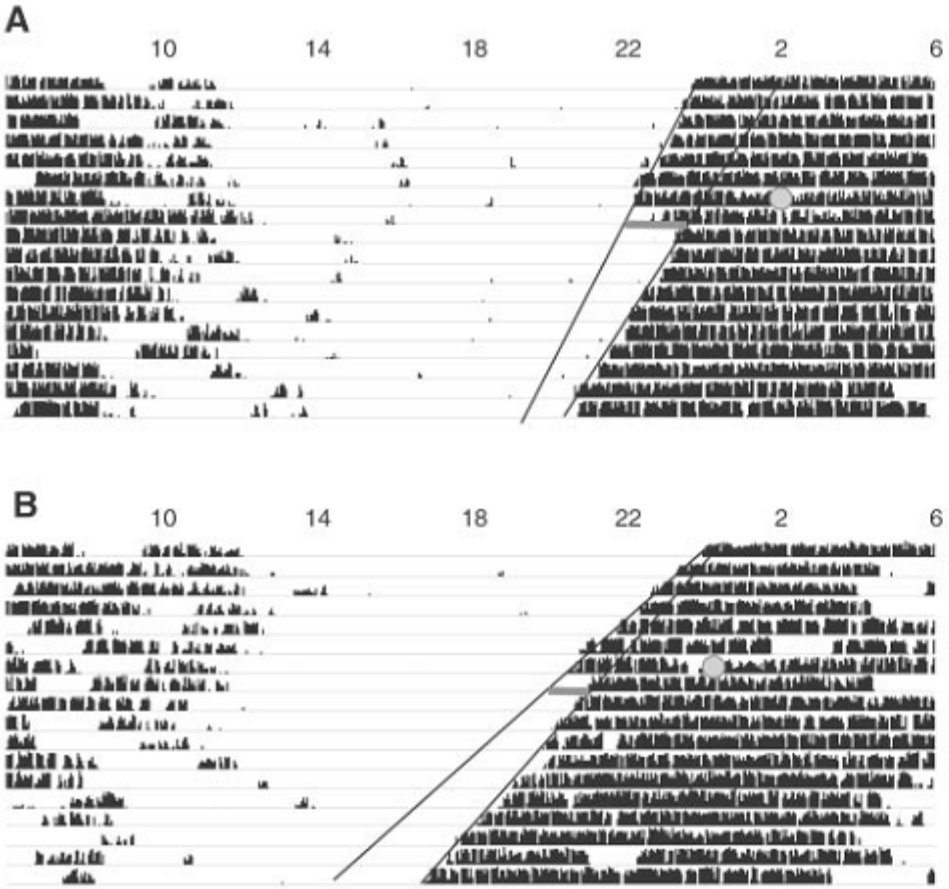
Hattar et al, 2002

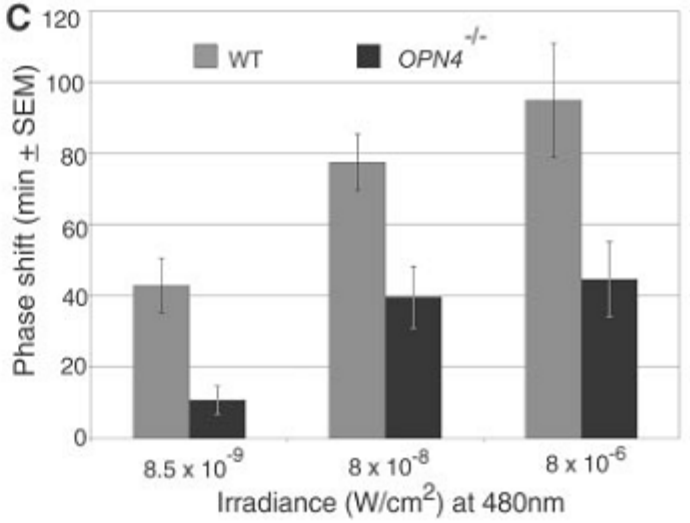
Knock out of the melanopsin gene



Panda et al, 2002

Knock out of the melanopsin gene

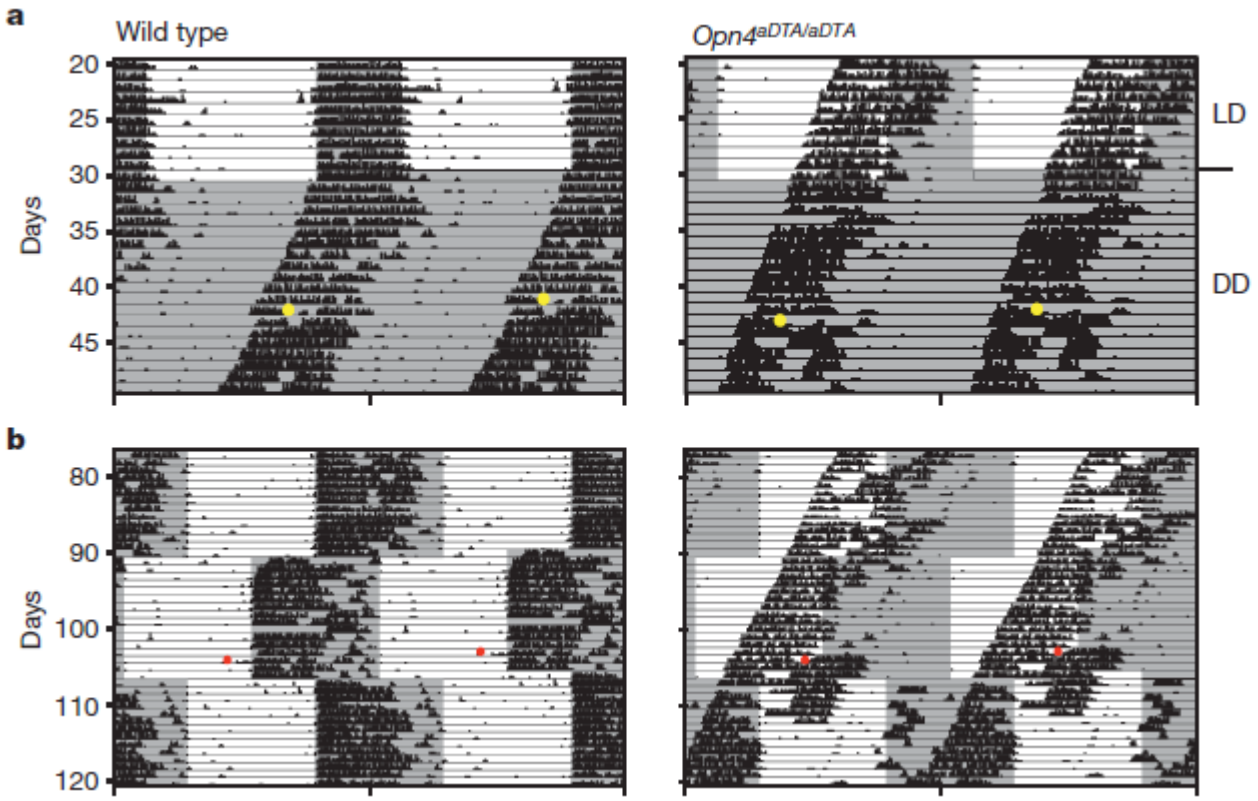




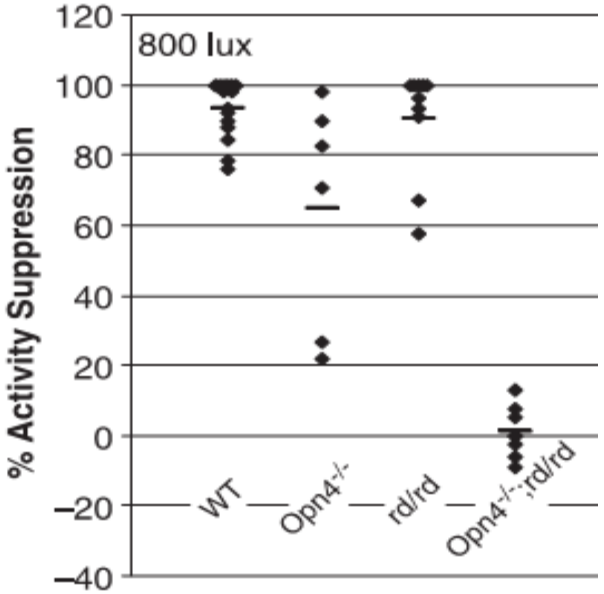
←480 nm 15 min.

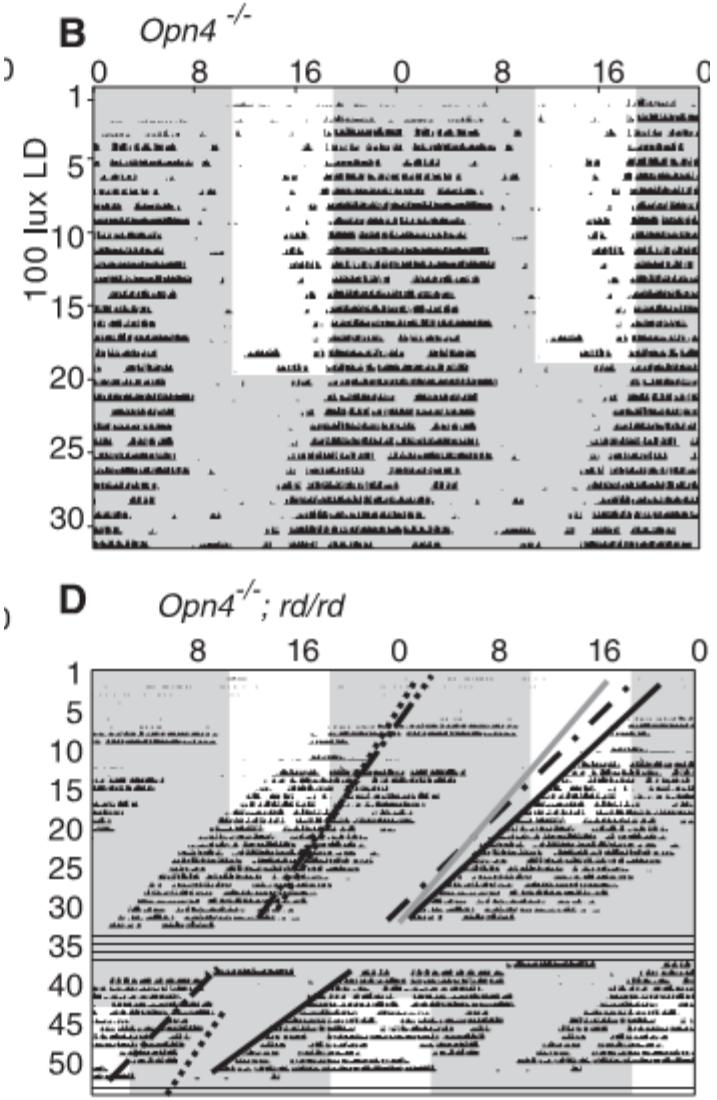
Panda et al, 2002

Ablation of the ipRGCs



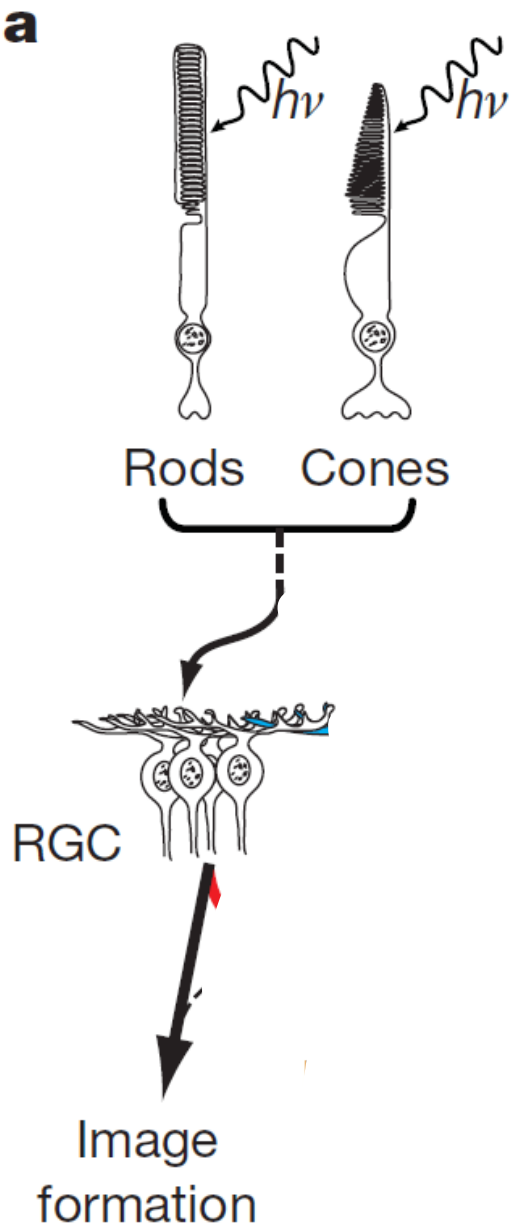
Guler et al, 2008





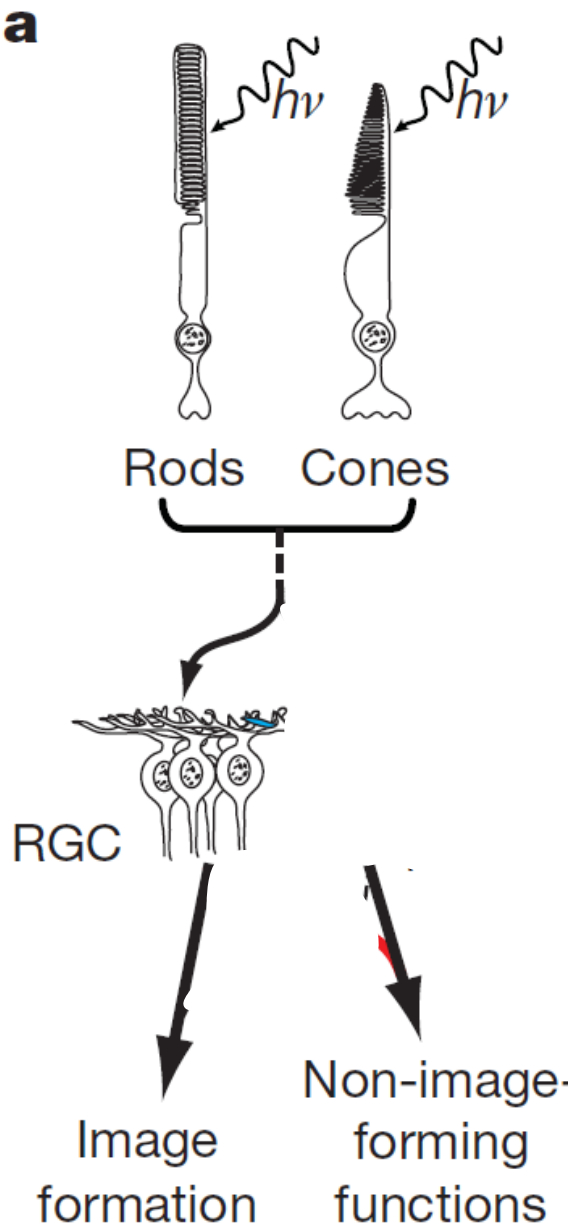
Panda et al, 2003

RGC - retinal ganglion cells



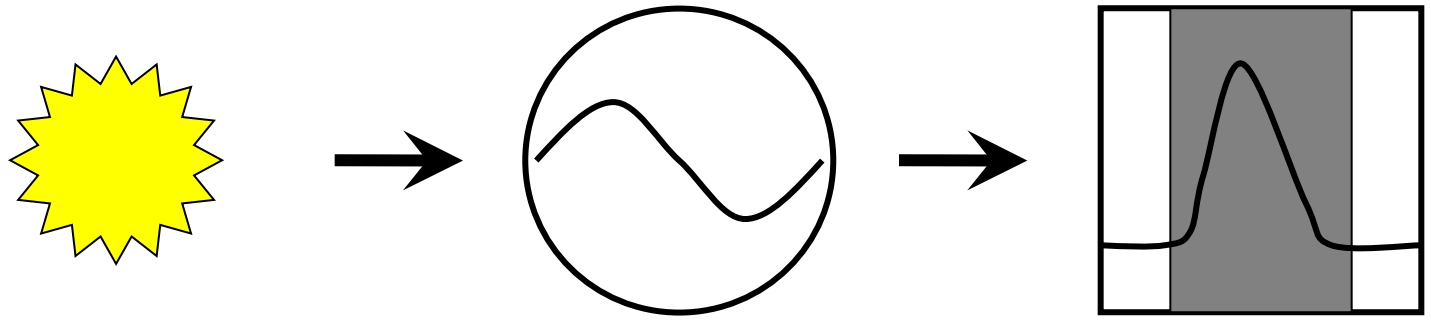
RGC - retinal ganglion cells

ipRGC - intrinsically photosensitive retinal ganglion cells



Organization of the mammalian clock system

Light input | **Oscillator** | **Output rhythm**



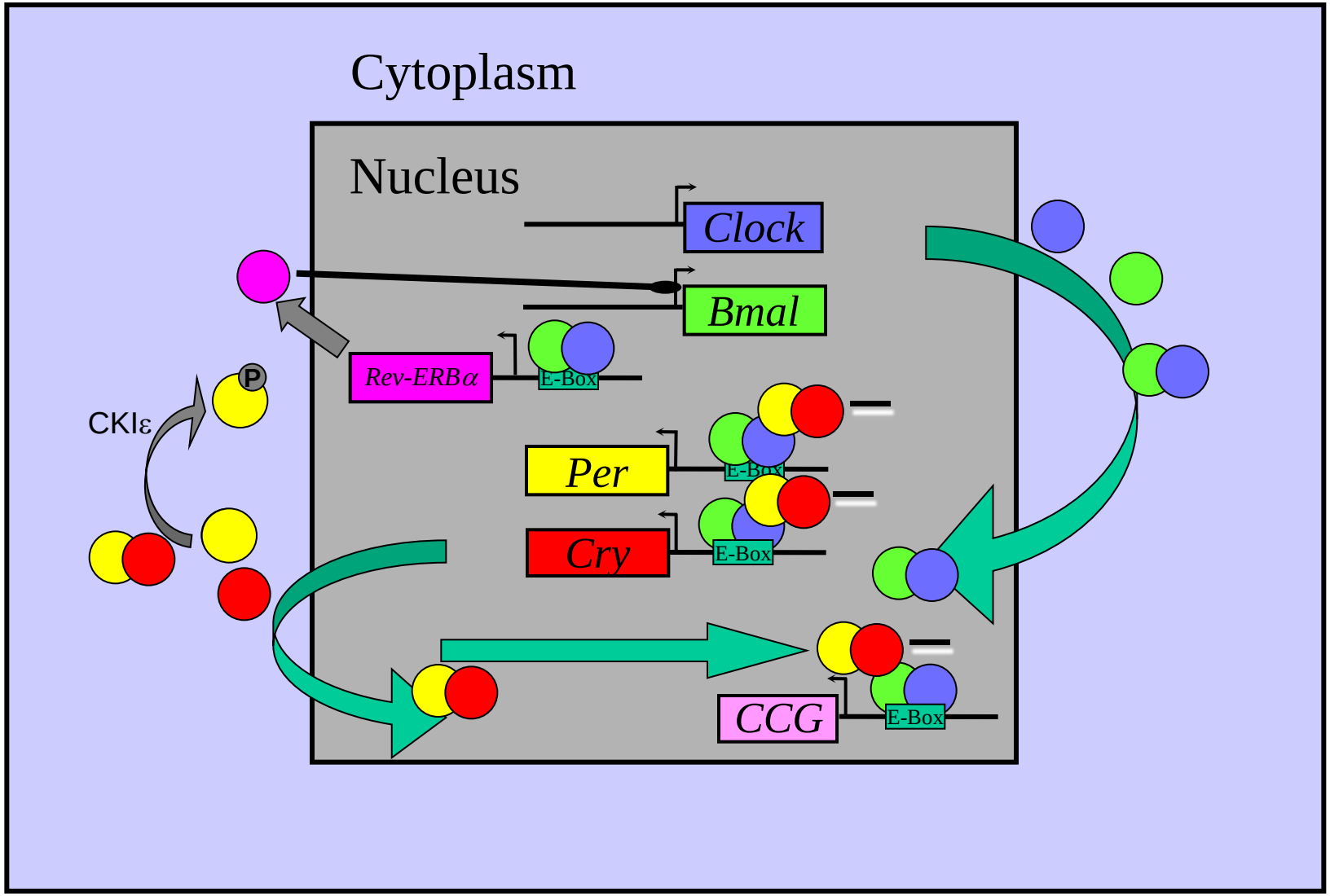
ipRGC | **SCN** | **Circadian rhythms**

Feeding
Activity
Social interaction

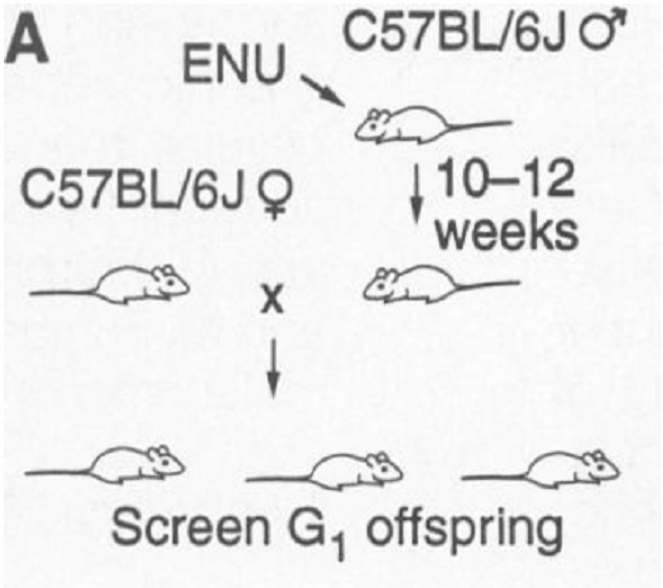
Independent
Light-entrained

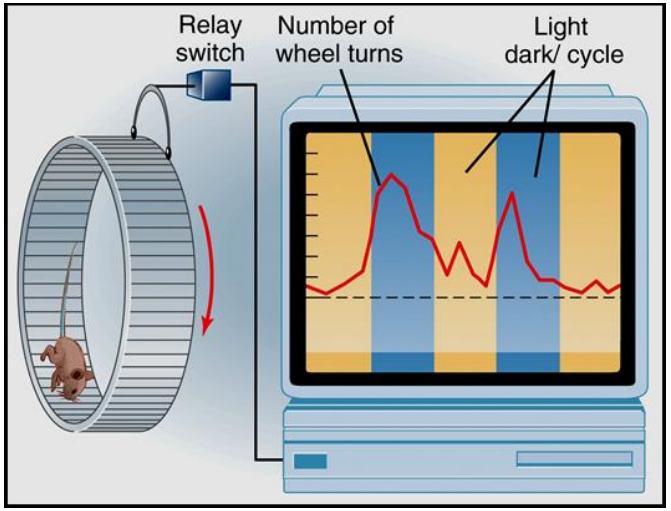
Sleep/wake cycles
Locomotor activity
Cognitive abilities
Reaction time
Body temperature
Metabolism
Hormone secretion
Enzymatic activities
Gene expression

Molecular oscillator based on a transcription feedback loops

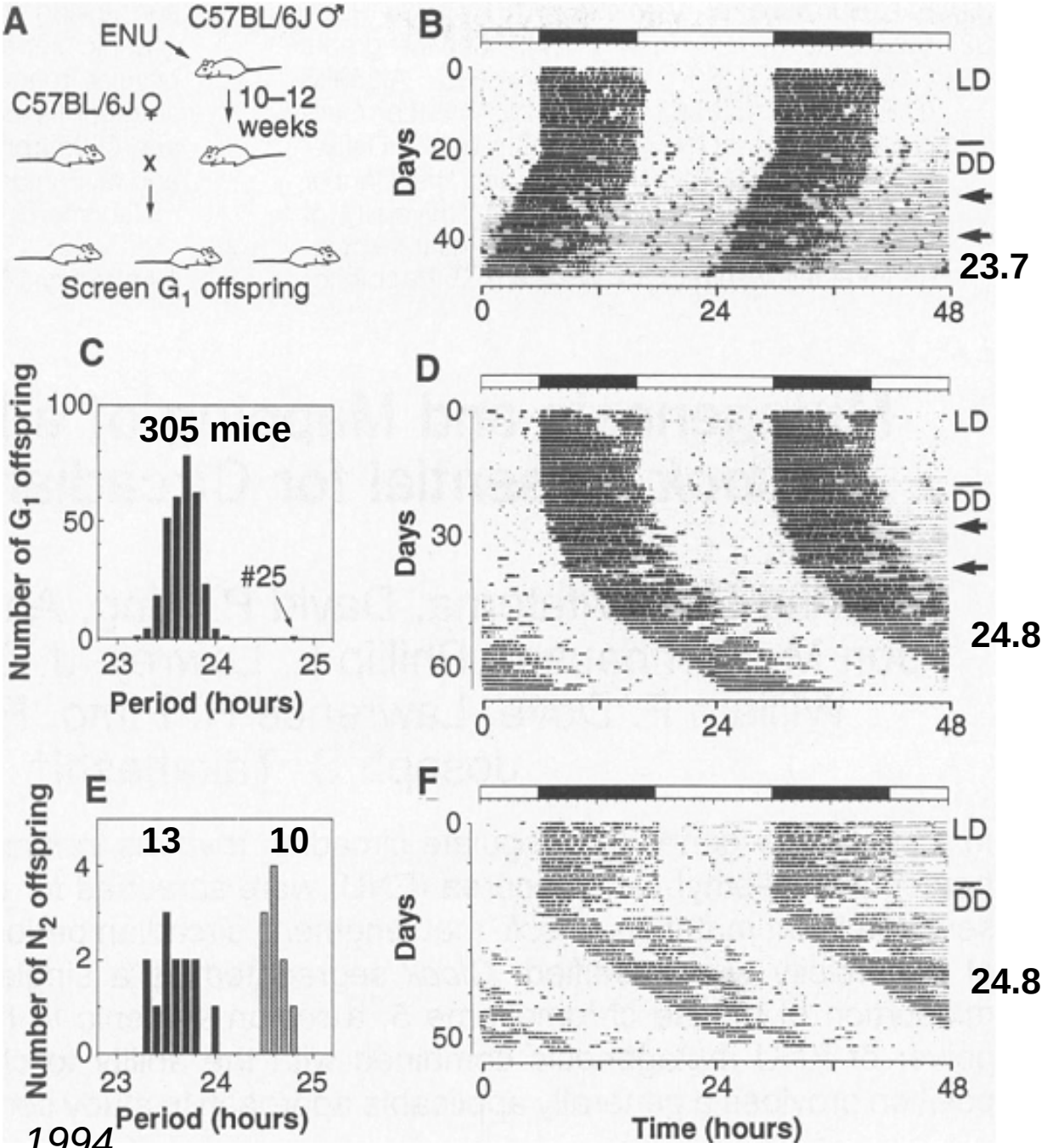


Discovering ***clock***

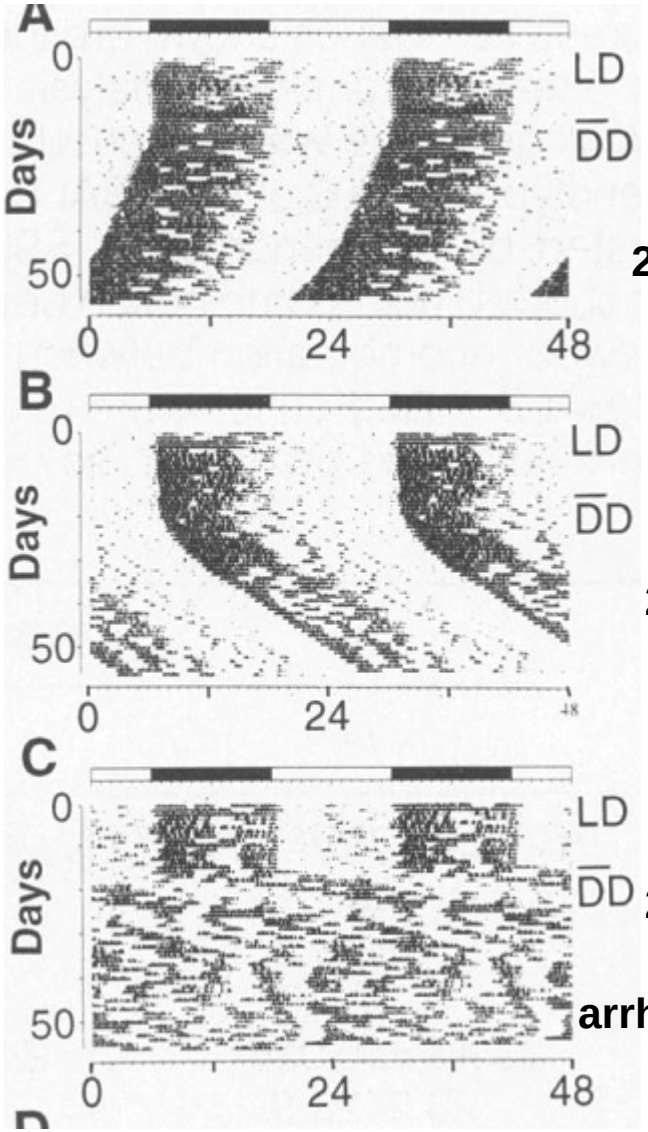




Vitaterna et al., 1994



Vitaterna et al., 1994



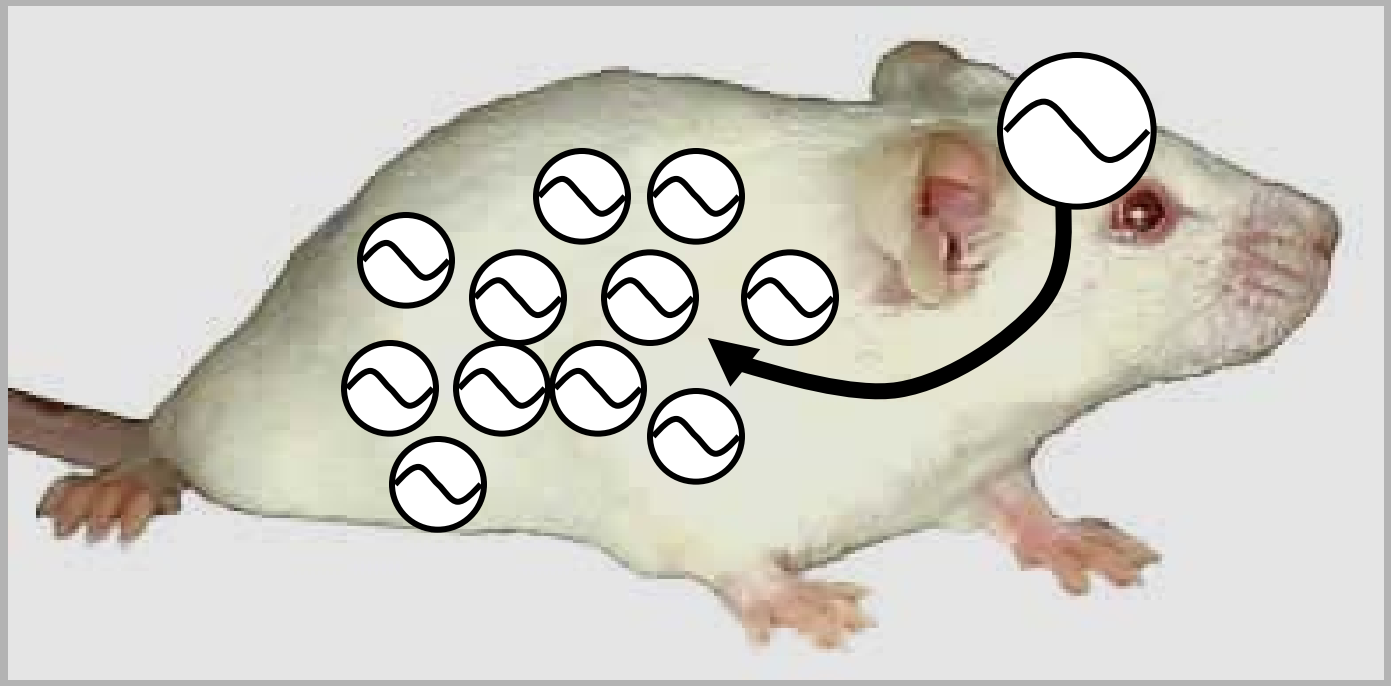
+/+ **23.6**

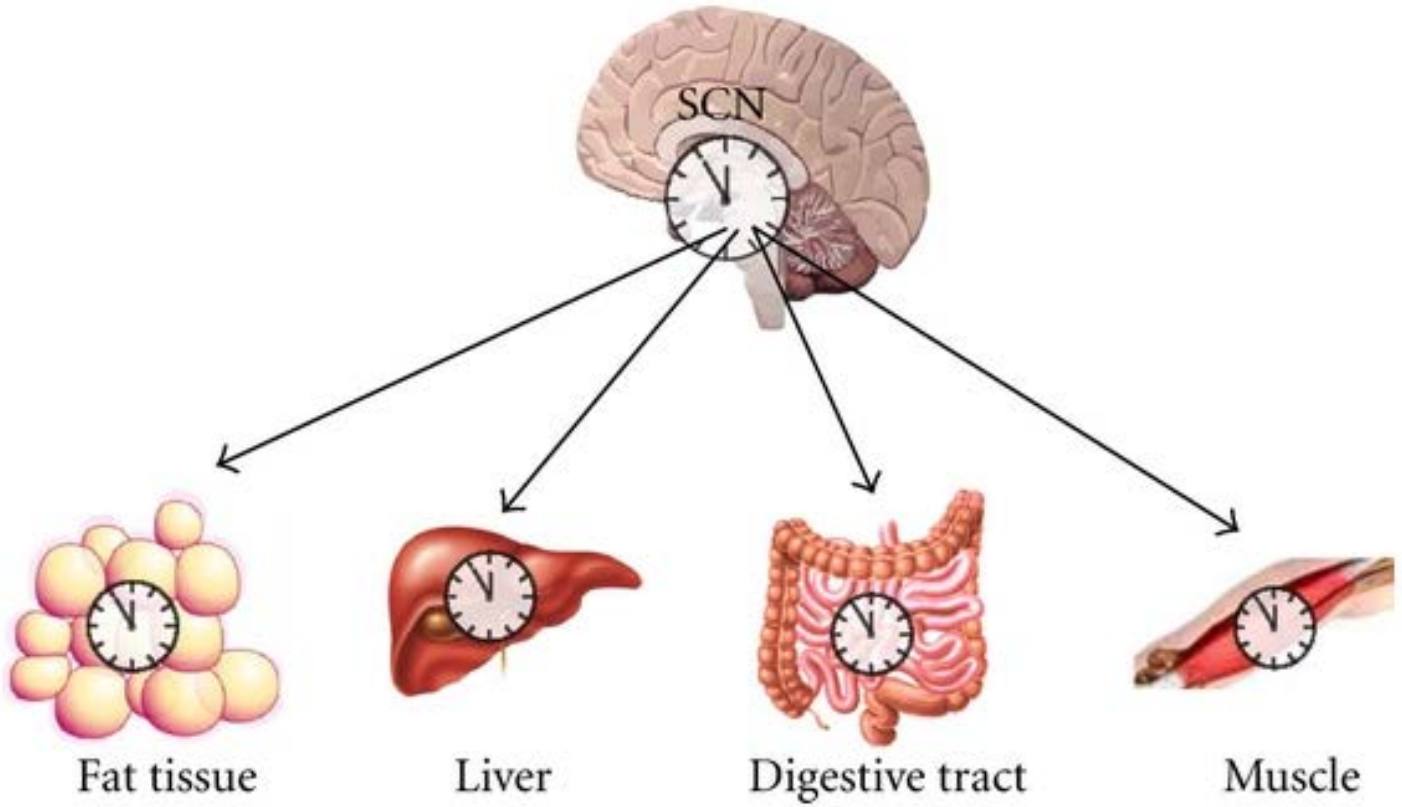
+/- **24.8**

-/- **27.1**

arrhythmic

Vitaterna et al., 1994




SCN
Fat tissue
Liver
Digestive tract
Muscle