Insect Structure Function & Physiology

Circadian Rhythms

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Biological Rhythms

Topics

- Terms used in describing rhythms
- Circadian rhythms
- Neural basis of rhythms
- Peripheral and central rhythms
- Insect models have applications to biomedical research
- Genes molecules and cells associated with rhythmicity are being discovered

Biological Rhythms

Background

- Go to http://www.hhmi.org/biointeractive/museum/index.html and run through the slide series. Pay special attention to the insect examples.

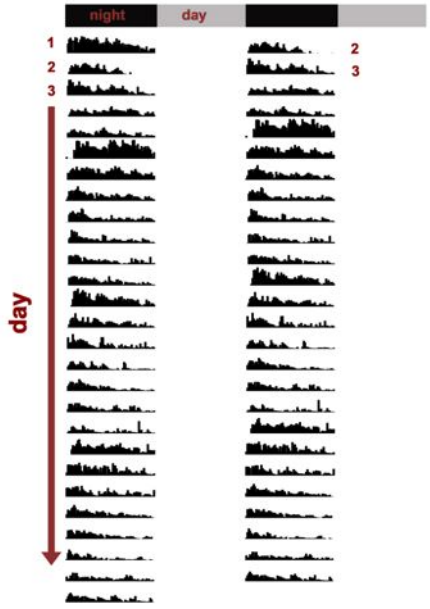
Mechanisms for Timing Behaviour

- Biological clocks or pacemakers are common regulators of behaviour
- Allows animal to change behavioural priorities over a day, month or year
- 24 hour clocks are most common: night & day have major impact on plants & animals
- Seasonality important for insects and diapause

- Many behaviours are restricted to time of day
- Closely related mosquito species restrict their prime feeding times to different very specific, narrow time frames after dusk
- Are they responding to environmental cues or are they responding to an *internal clock*?

Display methods

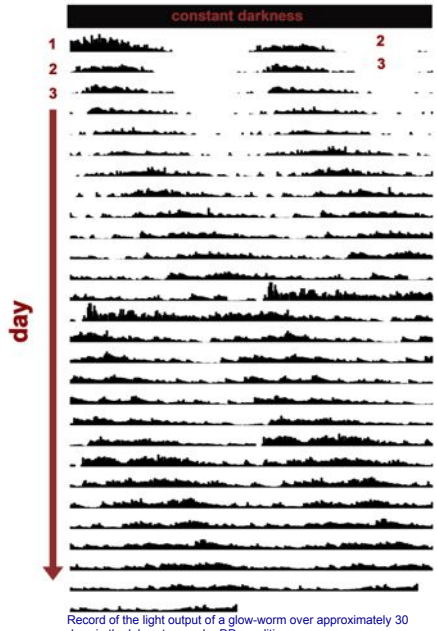
- Common to stack daily records so that trends are obvious
- Sometimes shown as stacked “double-plots” where each 24 h period is displayed twice
- Glow-worm bioluminescence is nocturnal
- Are they responding to environmental cues or are they responding to an *internal clock*?
- Best way to distinguish between possibilities is to place in constant dark (D:D), or constant light (L:L) and observe behaviour



Record of the light output of a glow-worm over approximately 30 days in the laboratory under LD conditions

DD

- Glow-worm bioluminescence “drifts” when placed in constant darkness (DD)
- Very characteristically not exactly 24 hours, and there is a day-to-day similar trend
- The fact that it is not exactly 24 hr cycle indicates it is obeying an internally-generated rhythm
- Approximately 24 hours = “circa” about, “dian” a day: **circadian**
- They show true circadian rhythmicity
- Free-running period > 24 h



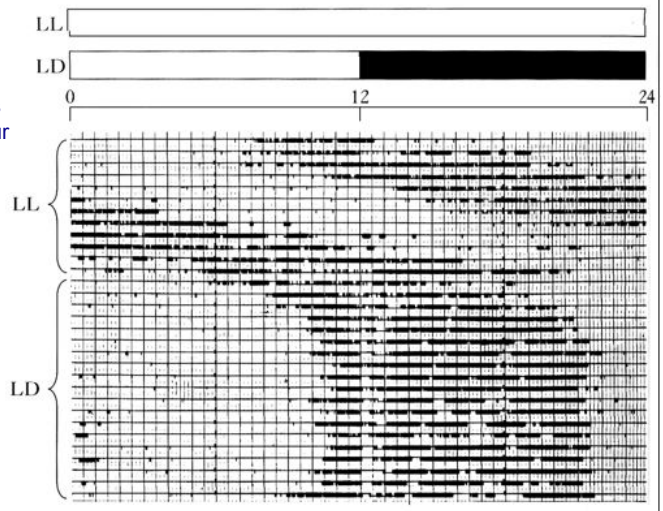
Record of the light output of a glow-worm over approximately 30 days in the laboratory under DD conditions

Cricket calling under L:L and constant temperature

Onset of nightfall reschedules **calling rhythm** of cricket, re-entrainment

Free-running period in L:L calling starts 25-26 hours later than it did the day before

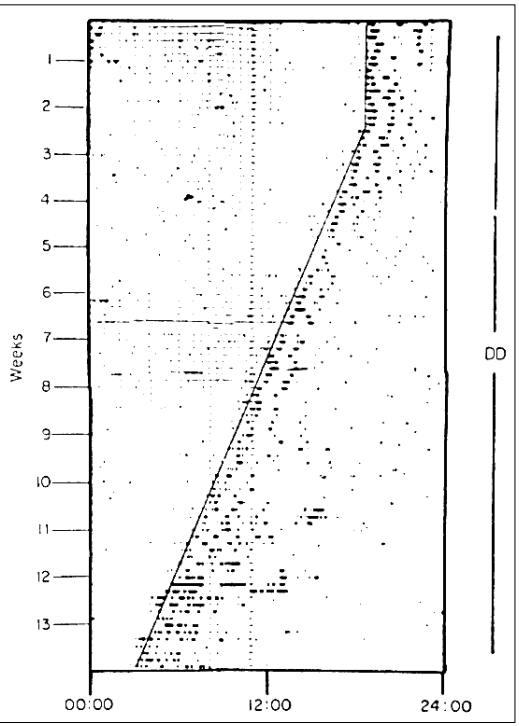
- Free-running period varies between individuals
- Under 12:12 L:D the free-running rhythm is **entrained** to a 24 hour cycle
- Accurately **predict** onset of dusk and dawn
- The circadian rhythm is **reset** every day
- Largely temperature independent (temperature-compensated)



Activity record of a cockroach

The activity record of an individual of the cockroach *Leucophaea maderae* in constant darkness (DD) for 13 weeks showing an abrupt change in its **free-running period** (T) in the third week from 24 hours to 23 hours 48 minutes.

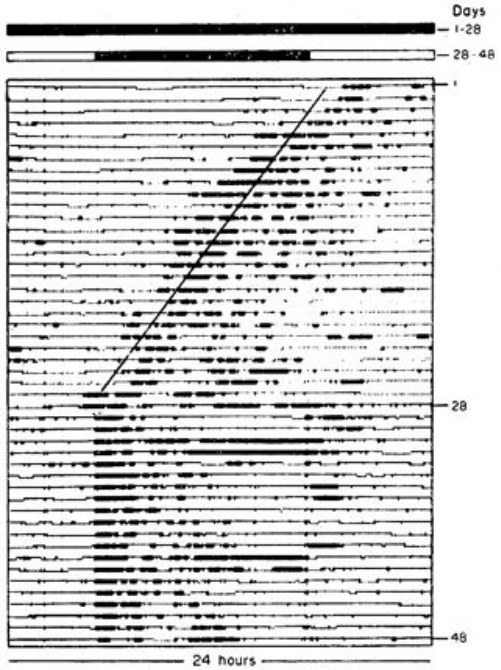
Exercise: calculate the free-running period (T)



Entrainment of the activity rhythm in an individual of *Leucophaea maderae* by a light/dark cycle (LD 12:12) following free-run in DD.

The natural period of the rhythm *(T)* during the first 28 days is about 23 hours 30 minutes; subsequently the rhythm is entrained to a 24 hour period to match the *Zeitgeber* (from Saunders, 1982.)

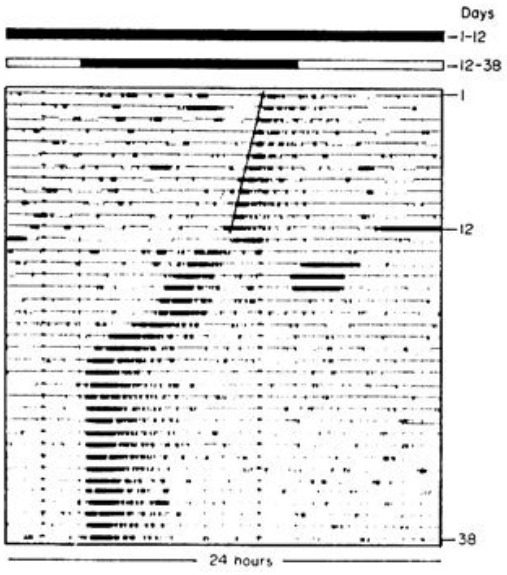
The **zeitgeber** is the entraining stimulus



Resetting the cockroach activity rhythm

Resetting with a light cycle (LD*12:12)* initially out of phase with the free-running rhythm and with "dusk" falling during the insect's subjective day. The onset of activity is gradually phase-advanced until entrainment is achieved after about eleven transient cycles.

phase-advance

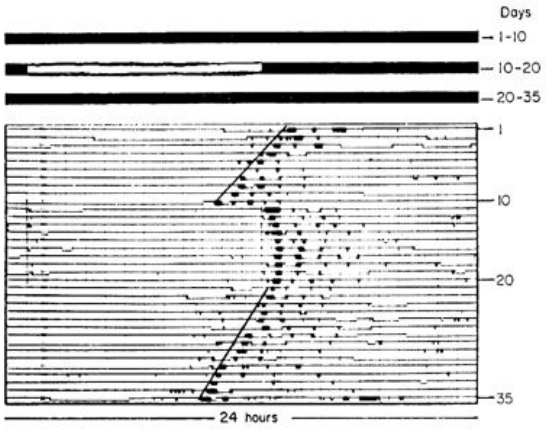


Activity record of a cockroach

Resetting with a light cycle (*LD12:I2*) initially out of phase and with "dusk" falling during the insect's subjective night. The onset of running is rapidly phase-delayed and the rhythm becomes entrained. Subsequent transfer to DD (on day *20)* shows that the phase of the endogenous rhythm has been shifted by the light treatment. (From Saunders, 1982.)

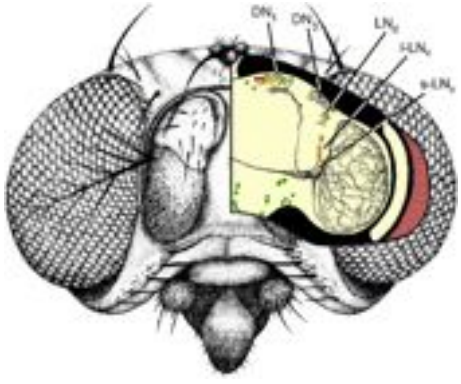
phase-delay

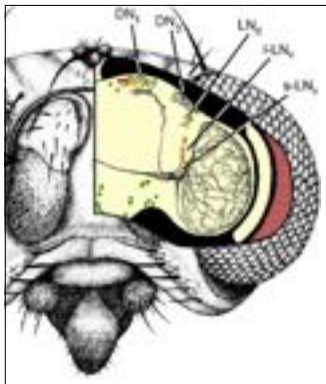
Phase changes are the cause of jet-lag in humans

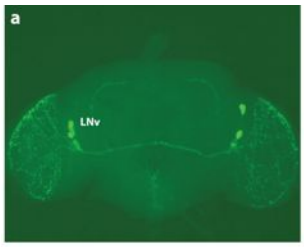


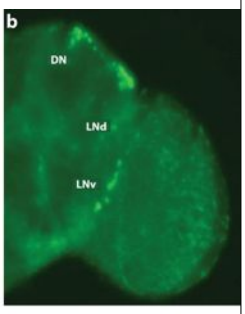
- Experimental manipulations help track down the source of the rhythm
- In cricket, cut nerves from eye to optic lobes. Rhythm (a) persists and (b) becomes free running. The eyes are not source of rhythm but are necessary for entrainment.

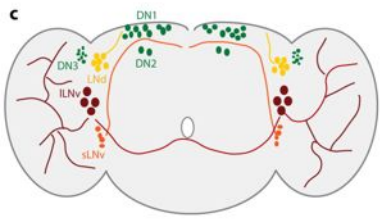
  - Removal of optic lobes removes rhythmicity.
  - The pacemaker lies in the optic lobes.
  - More recent use of antibody staining shows small clusters of neurons in brain are the pacemaker cells









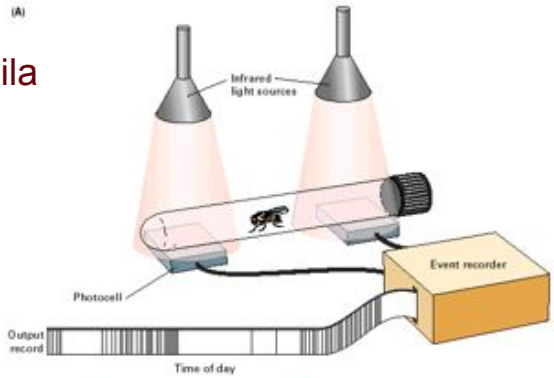


pacemaker cells

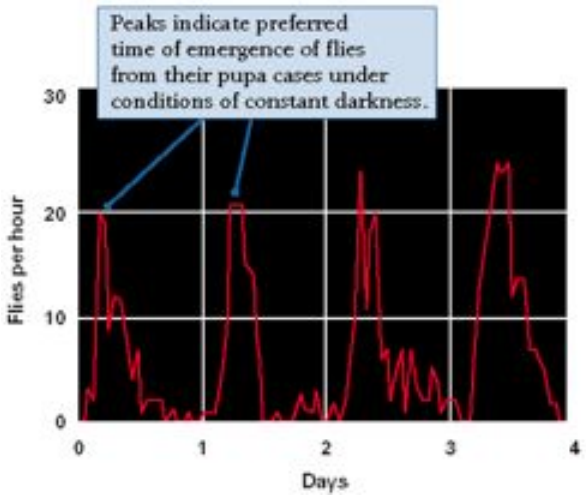
Allada R, Chung BY. 2010.
Annu. Rev. Physiol. 72:605–24

per gene in Drosophila

- The molecular and genetic basis of rhythmic behaviour is being teased apart in Drosophila
- Drosophila show circadian activity rhythms
- Genetic screens start with no assumptions about what genes are involved

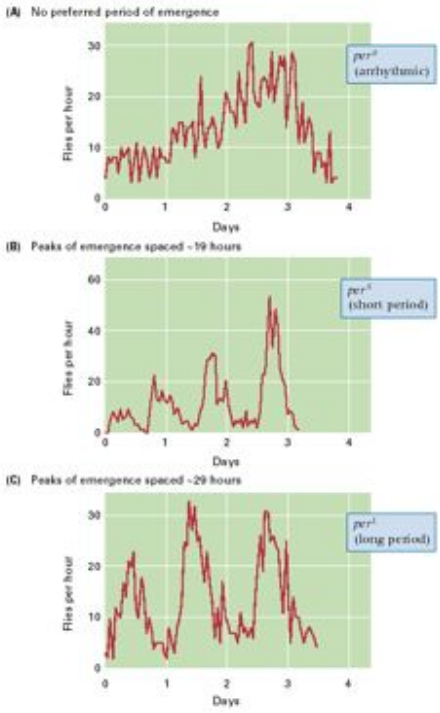


- show pupal emergence rhythms



Drosophila

- Mutagenesis screen revealed mutations that affect the *period* of the rhythm,
- *Per* zero, *per* short and *per* long
- Gene has been sequenced
- Further genes involved in the cascade have been identified: *timeless*, *cycle*, and *Clock*
- blue light-absorbing photoreceptor cryptochrome (CRY)



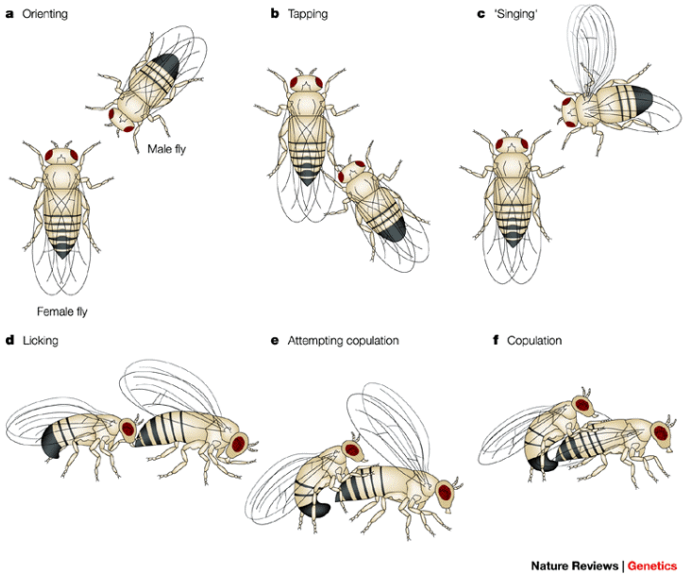
Where is the clock?

Central clock: brain neurons
Peripheral clocks:
Malpighian tubules
epidermis (rhythms of cuticle deposition),
eyes (light receptors)
antennae (olf receptors?)

Behaviours

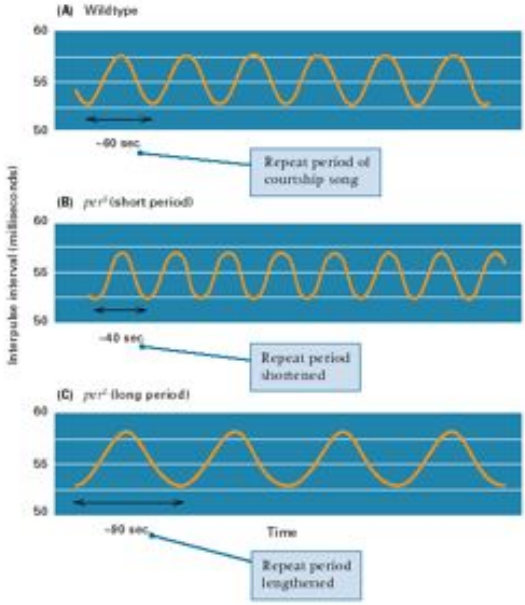
Sleep
Locomotion
Learning & memory
Eclosion
Sensory sensitivity
Courtship & mating
immunity

The Courtship Song

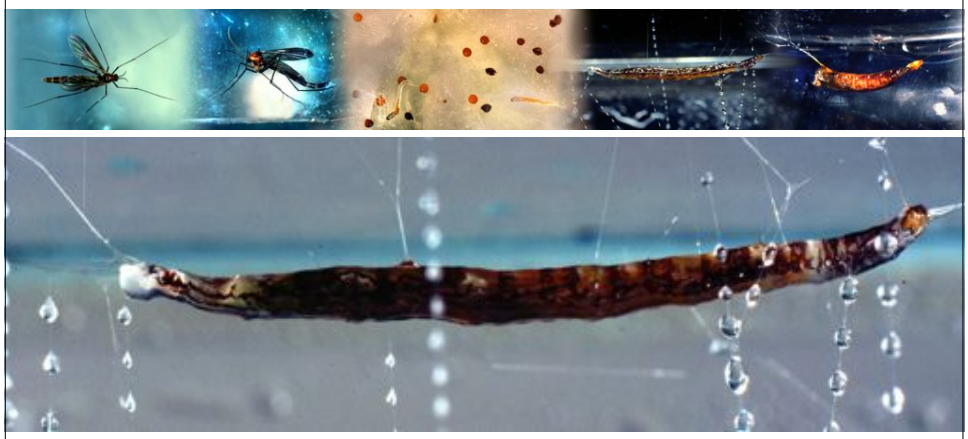


Short term cycles

- An unexpected finding was that mutations in the *per* gene affected very short term rhythms as well.
- The 60 sec cycle of the male mating song is affected
- Per gene from a related species placed in a per null *Drosophila melanogaster* restores the rhythmicity of the song — to that of the donor species.



Immature stage of keroplatid fly



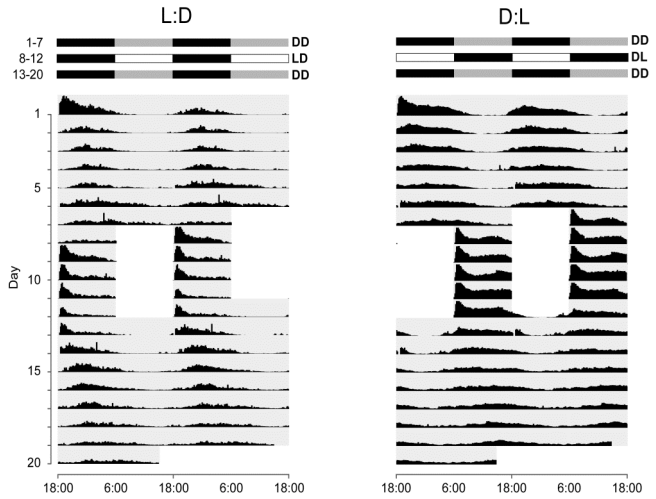
Larvae live in rainforest or caves

If you lived in a cave without any outside contact would you lose track of time?

circadian (= approximately 24h)

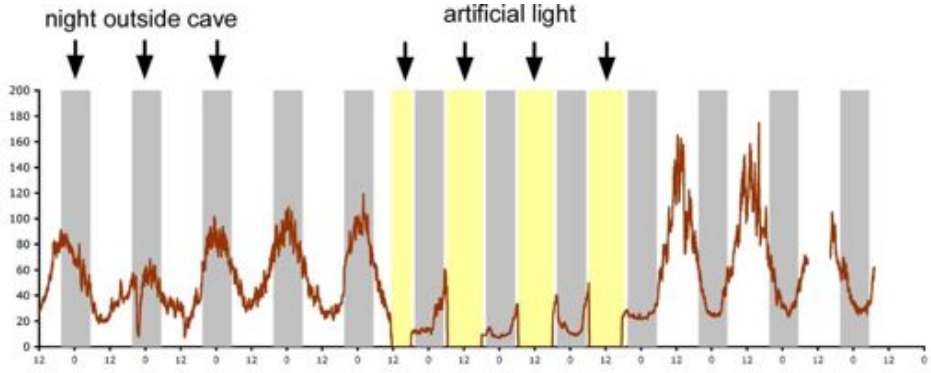
Yes: and in a consistent way. Individuals show cycles of approx 25.1 hours. Humans possess rhythms in sleep, activity, body temperature and other physiological processes. So do insects

Rainforest
Arachnocampa flava
tested in lab
1: exposed to DD to free-run
2. Exposed to LD to show L represses biolum
3. On return to DD the light has re-entrained the rhythms to match the dark phase



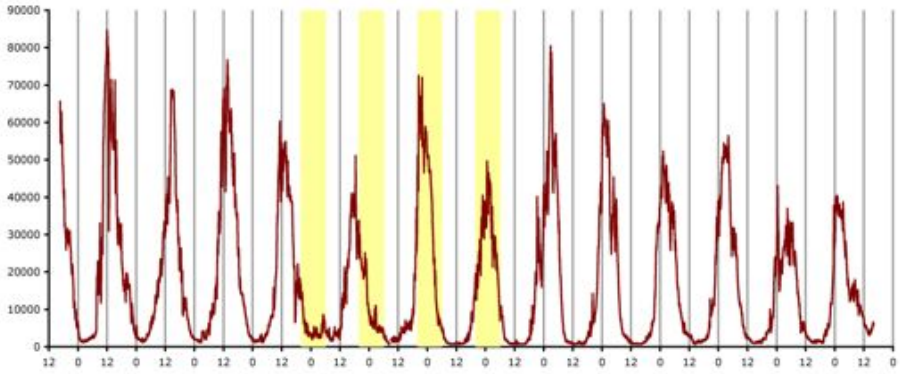
All species:
light is a *masking* agent, i.e. it blocks bioluminescent output
Light is an entraining agent

CAVES



In caves (A. tasmaniensis) cycle but don't free-run.
Exposure to artificial light masks and re-entrains ***to the light cycle***

Light: circadian rhythms



Lab experiments with weak point-sources show same synchronisation but light not strong enough to mask

Conclusion

Synchronise to each others' glows

References

- Chapter 6 Alcock
- Chapman 20.5.6
- Circadian rhythms and genes. Genetics, by Hartl and Jones (4th edition)
- Insect Clocks. D.S. Saunders
- Of interest: a good read: "Sync: the emerging science of spontaneous order" Steven Strogatz