The Biological Link Between Time & Space

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- Circadian Rhythms are integral in the daily fluctuations of physiological activity that allow for normal functioning in most organisms.
- Endogenous circadian rhythms depend on the expression of *Clock* genes, such as *Period*, *Timer*, *Tau*, and *Clock*.
- Alterations in the genes that code for heterodimer proteins *Period* and *Timer* result in altered circadian rhythms in *Drosophila melanogaster*.
- Even with endogenous rhythms in place, environmental "zeitgebers" can entrain new rhythms, just as humans with 24 hour circadian clocks can adjust to different time zones.

The Affects of Altering Circadian Rhythms in Fruit Flies

Objective:
Determine how entrainment into non-endogenous rhythms influences spacial memory in *D. melanogaster*.

Experimental Methods:

Experimental Groups:

1. Period Short Flies entrained to 24 hour incubator
3. Period Short Flies entrained to 21 hour incubator
5. Wild Type Flies entrained to 24 hour incubator
4. Wild Type Flies entrained to 21 hour incubator

Hypothesis: Flies who are entrained to circadian rhythms that do not match their endogenous rhythms will experience deficits in spatial learning not experienced by their control counterparts.

Experimentation took place in four 5-minute intervals.

1. **Preference Test:** Flies placed in enclosure for 5 minutes to determine side preference.
2. **Training Period One:** Boiling water is poured into a container positioned beneath the preferred side of the enclosure.
3. **Training Period Two:** 24 hours later. Same procedure as training period one.
4. **Memory Test:** Flies were again placed in enclosure without aversive stimuli

A Time budget was recorded during each phase of the experiment using Jwatcher.

Experimental and Statistical Results:

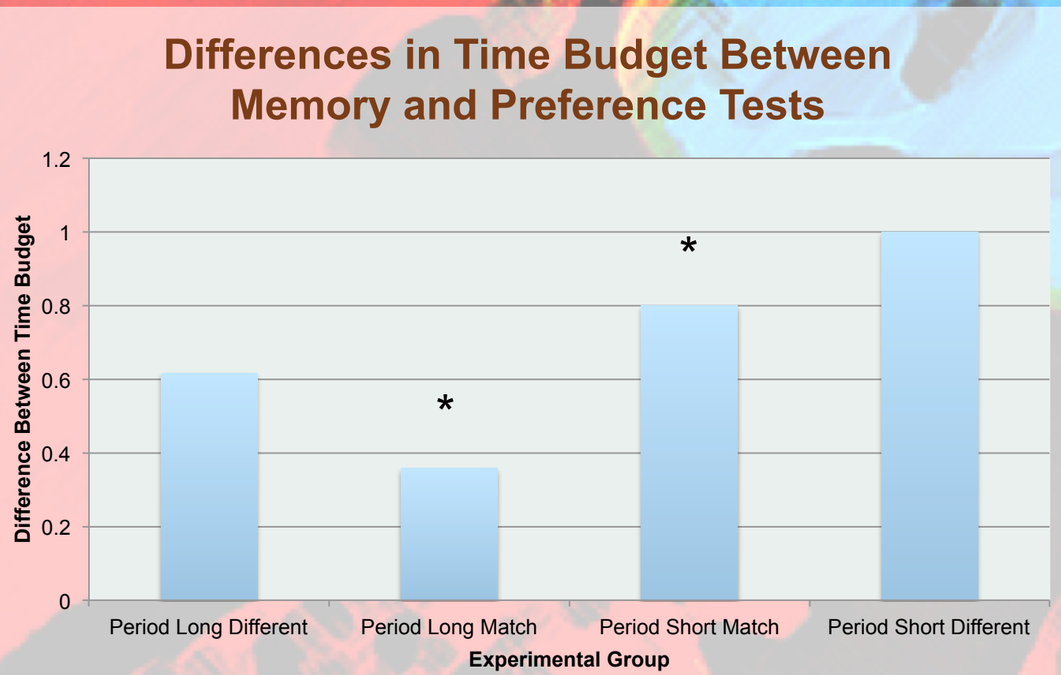

Figure 1 shows the difference in time spent on non-preferred side normalized by the time spent on the preferred side between the memory and preference test trials. Values for each trial were reached by subtracting the amount of time spent on the preferred side (seconds) from the amount of time spent on the non-preferred side (seconds) and dividing the difference by total trial time (300 seconds). The value obtained for the preference test was then subtracted from the value obtained from the memory test to determine bar length.

Statistical Results

- WT Match: t = 3.2915, df = 8, p-value < 0.02
- WT Different: t = -1.9272, df = 4, p-value = 0.1262
- Per Short Match: t = -2.4495, df = 8, p-value < 0.04
- Per Short Different: t = -1, df = 2, p-value = 0.4226

- We performed a paired t-test for each group between their time spent in the non-preferred side normalized by the time spent on the preferred side for each of the four groups.
- This difference was found to be significant for the two groups whose entrained rhythms matched their endogenous rhythms.
- This difference was not significant for the two groups whose entrained rhythms did not match their endogenous rhythms

Conclusions & New Directions:

- Our results supported our hypothesis that alterations in the synchrony between entrained and endogenous rhythms affect D. melanogaster's capacity for spatial memory processing.
- This is interesting in that it implies imperfection in the ability of external "zeitgebers" to fully and effectively entrain new circadian rhythms.
- This also suggests a possible mechanism within the neurocircuitry of *D. melanogaster* that relies on the regulated expression of the Period gene.
- However, due to our small sample sizes, our results must be viewed with caution. Ideally, we would be able to replicate our experiment with much larger sample sizes and attempt the replicate our results.

References:

- Baylies, M. K., et al. "New Short-Period Mutations of the Drosophila Clock Gene Per." Neuron 9.3 (1992): 575-81. Print.
- "Memories in Drosophila Heat-box Learning." Learning and Memory. Web. 09 Dec. 2011. <http://www.ncbi.nlm.nih.gov/pmc/articles/PMC187128/.>