Genetic Dissection of the Contribution of Central and Peripheral Circadian Clocks to Drosophila Feeding Rhythms

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Introduction

The circadian system produces ~24-hr rhythms and consists of three major components: a central molecular clock in the brain that keeps time, input pathways that allow organisms to stay synchronized with changes in their environment, and output pathways that couple the clock to various behavioral and physiological processes such as locomotion [1]. Recent studies have demonstrated circadian control of feeding independent of locomotor activity [2], but the neuronal circuitry governing feeding rhythms is not understood. In addition to the central brain clock, circadian clocks are present in many peripheral tissues, such as the Drosophila melanogaster fat body, which is homologous to the mammalian liver and regulates metabolism [3]. Here, we investigated the feeding behavior of transgenic flies in which we eliminated or changed the speed of the brain or fat body clock to identify the contributions of central and peripheral circadian clocks to feeding rhythms. We additionally conducted immunohistochemical analysis to confirm molecular clock speed alterations in these flies.

Methods

Feeding experiments
- Circadian feeding behavior was assayed using the Fly Liquid-food Interaction Counter (FLIC) [4]. Male were entrained to light-dark cycles for ≥ 3 d before being transferred to constant darkness for feeding monitoring.

Data Analysis
- Feeding data were binned into 30 min intervals and analyzed with X2 periodogram to measure period and strength (power) of feeding rhythms (Clock Lab software, Actimetrics).
- Group data were compared with Tukey posthoc test following one-way ANOVA.

Immunohistochemical Staining
- Male brains or abdomens were dissected in PBST and fixed with 4% paraformaldehyde. Dissected tissues were blocked in normal donkey serum and incubated overnight in primary antibodies, then incubated overnight in secondary antibodies before confocal imaging.

References


Acknowledgements

Work was funded by Mulchay Fellowship awarded by The College of Arts and Sciences Dean’s office of Loyola University Chicago

Conclusions

- Changing the speed of the central, but not fat body clock, alters feeding behavior by altering feeding rhythm period and strength.
- Eliminating the central clock, but not the fat body clock, reduces strength of feeding rhythms.
- The brain clock functions as the chief regulator of feeding rhythms.