Effects of Sleep Fragmentation on Alcohol Response in *Drosophila melanogaster*

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Introduction

- Previous research has found an association between increased incidents of alcohol abuse and individuals who experience regular circadian rhythm or sleep disturbance, such as shift workers (Colten & Altevogt, 2006).
- Gaining a deeper understanding of the effect of circadian rhythm disruption on drug tolerance and recovery is a crucial first step in tackling the issue of increased alcohol toxicity and abuse in sleep fragmented human populations (Nobrega & Lyons, 2016).
- *Drosophila melanogaster* are ideal models for studying human disease as the majority of genes responsible for human diseases also exist in the fruit fly genome (Chien *et al*., 2002; Lessing & Bonini, 2009).

Methods

Maintenance: Wild type Canton-S (CS) *Drosophila* were maintained on 12:12h light-dark (LD) cycles between 0-3 days of age. Zeitgeiber Time (ZT) 0 corresponds with lights on and ZT12 with lights off. Flies were maintained in cornmeal-molasses vials and were transferred to new vials every 3 days.

Sleep fragmentation occurred between 3-7 days of age on either a less disrupted paradigm or on a more highly disrupted paradigm. Drosophila were placed on automatic shakers to stimulate a disruption in sleep pattern. Drosophila on the more disrupted paradigm experienced 3 min sleep deprivation:7 min rest for 4 consecutive days. Drosophila on the mild paradigm experienced 18 min sleep deprivation:42 min rest for 4 consecutive days.

Alcohol exposure: Flies were exposed to alcohol using an alcohol vapor delivery system called the FlyBar (De Nobrega and Lyons, 2016) (**Fig 1**). Exposure occurred at ZT 9.0 at 25°C on day 11 using 50% alcohol vapor.

Sedation was measured in 5 minute intervals during hour-long alcohol exposures. Flies were deemed to be fully sedated once they were no longer capable of coordinated movement (with the exception of spontaneous twitching of the hind legs).

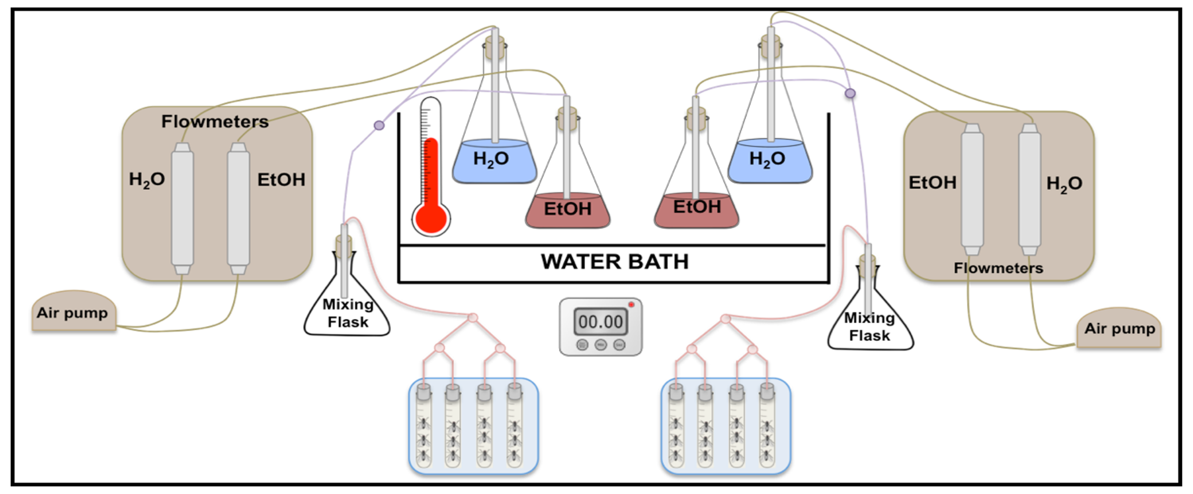
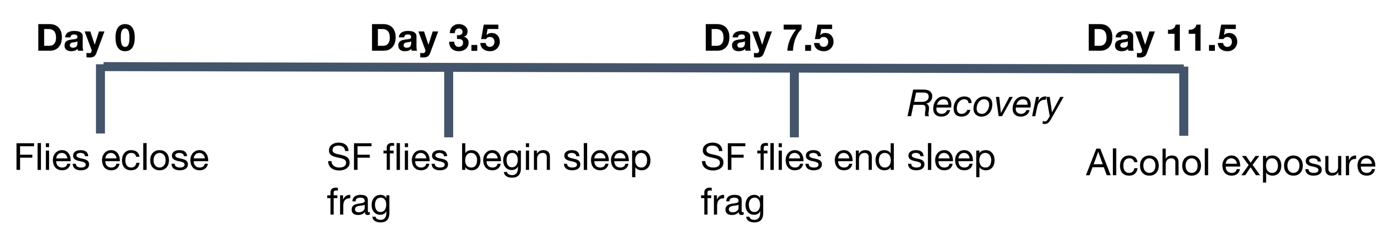


Figure 1. The Fly-bar

Timeline of experiments:



Results

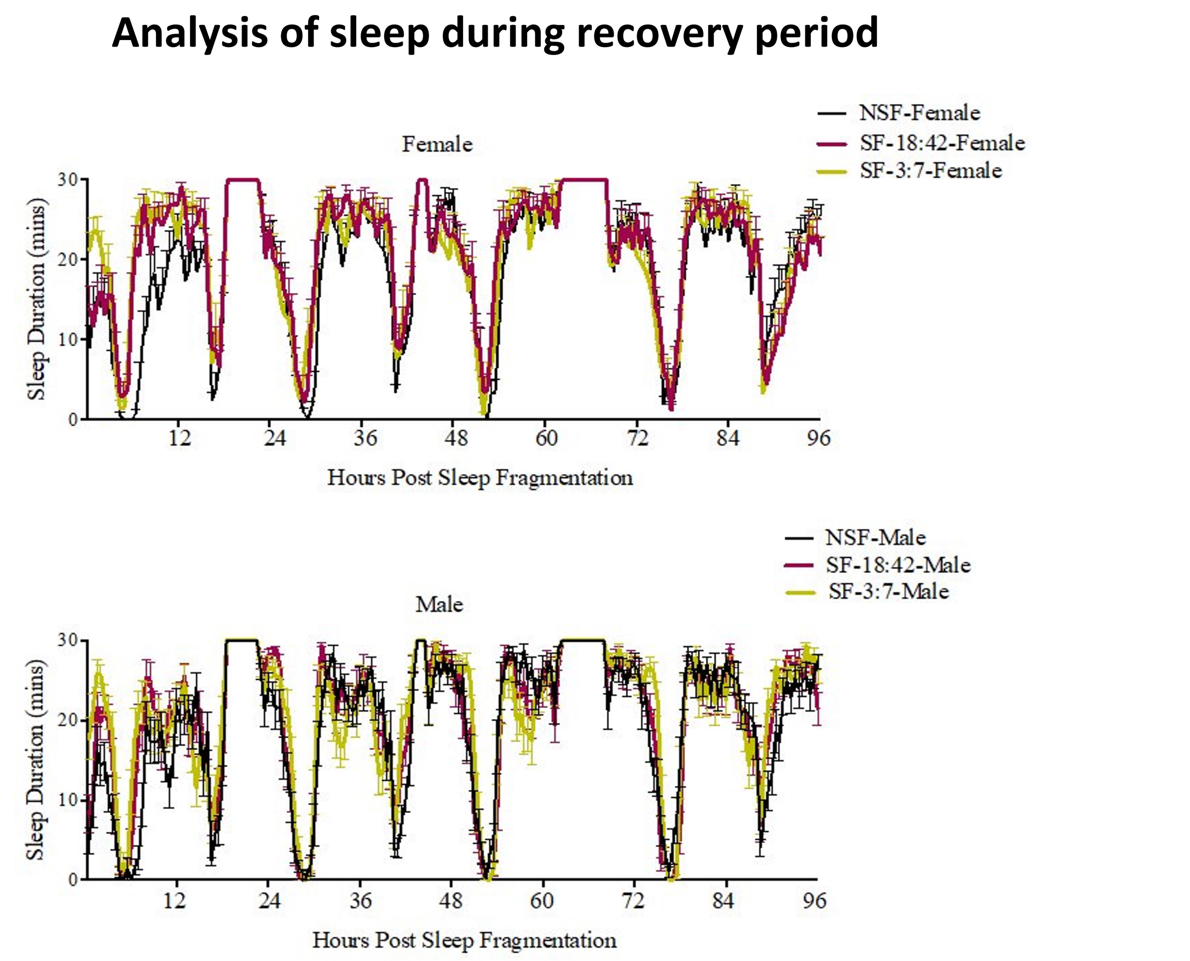


Figure 2. Analysis of sleep in male and female wild-type CS flies using locomotor activity rhythms in the DAMs monitoring system.

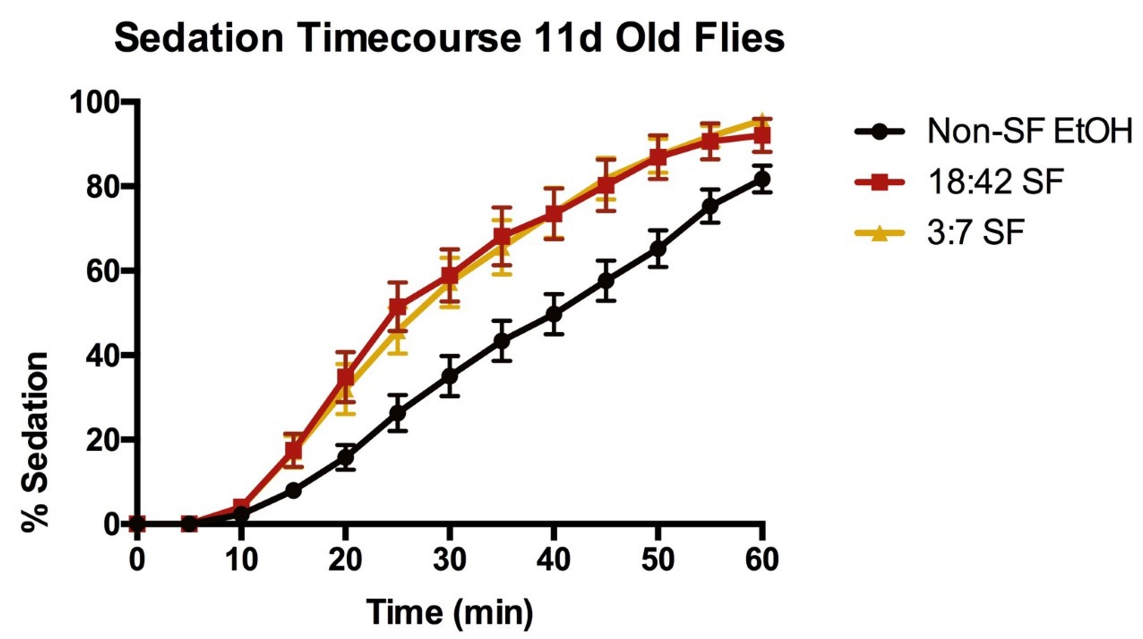


Figure 3. Timecourse of alcohol sedation in 11-day old sleep fragmented and non-sleep fragmented flies using 50% alcohol vapor.

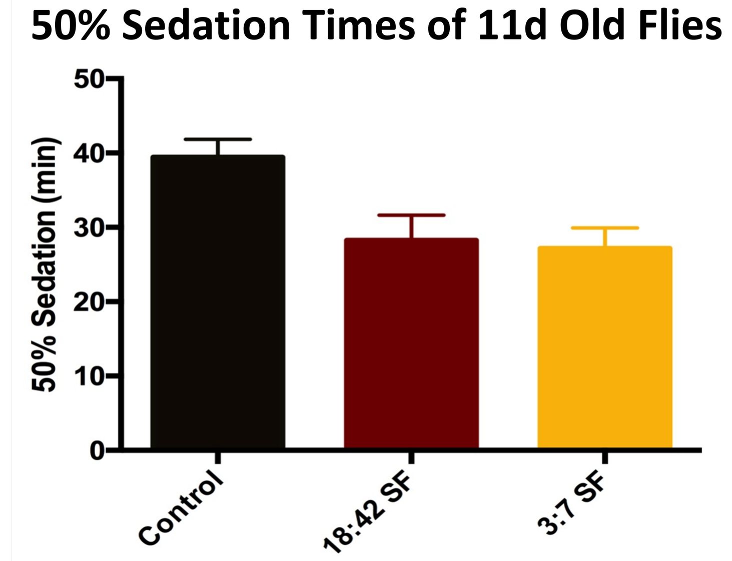


Figure 4. The 50% sedation rate shows that sleep fragmentation significantly increases alcohol sensitivity.

Conclusion

- Four days of sleep fragmentation causes persistent changes in alcohol sensitivity.
- Even with four days of recovery sleep, sleep fragmentation significantly increases alcohol sensitivity as compared to non sleep fragmented controls.
- Surprisingly, sleep fragmented flies on a less disrupted paradigm exhibited the same long term sensitivity to alcohol as flies on a more disrupted paradigm.
- Locomotor activity analysis of sleep patterns during the four day recovery period revealed continued sleep fragmentation and aberrant sleep patterns, specifically for the first 18 hours after sleep fragmentation.

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