Neurotransmitter and synaptic function mutants of Drosophila melanogaster display unique distinct recovery behaviors from ether anesthesia.

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Introduction

- Drosophila melanogaster has electrical and chemical synapses in neurons similar to those of humans.
- Mutations in chemical synapses involve disruption of neurotransmitter function:
  - Cha-ts1, TBh[nm18], DDC[DEI]
- Mutations in electrical synapses involve disruption of gap junction function:
  - ShakB[pos]
- Locomotion of many mutant fruit flies has been examined and explained.
- The effects of ether use as an anaesthetic is not well understood; Some research exists on a the ShakB[pos] mutant’s vigorous leg movement under ether.
- Drosophila melanogaster’s recovery from ether is understudied.

Objectives

- Develop consistent procedure to study recovery of Drosophila melanogaster from ether.
- Use procedure to find unique recovery patterns.

Methods

- Automated analysis of Locomotion in Mutant Drosophila:
  - The IowaFLI tracking system is used to outline the fly’s movement. The program can measure interactions, inactivity, shaking and other behaviors. (A) A webcam connected to computer records flies in a petri dish. (B) Image is recorded and flies are marked and distinguished from background to track. (C) Fly tracking plot for a 30 second interval. Each color correlates to a single fly.

- Ether dosing of Drosophila mutants:
  - (A) A bottle contains a sponge that absorbs ether. Enough ether is poured and absorbed by the sponge such that a pool forms at bottom. (B) Flies are then placed in an opening on top that enters into a wide circular tube with holes allowing vapor to reach flies. (C) Once down, flies are then transferred to prepared chamber.

Results

- Figure 3: Wild type fly under ether; Laying on back and immobile.
- Figure 4: Wild type fly partially recovered; Standing on legs.
- Figure 5 – Sample tracks of WT (CsJL), Cha-ts1, and ShakB[pos] with and without ether:
  - Set of four 30 second frames. Four flies of each genotype recorded and tracked. Each color corresponds to each fly. Flies in tracks (A), (C) & (E) were not dosed with ether. Flies in tracks (B), (D), and (F) were dosed with ether.

- CsJL: (A) Control video- Wt flies generally move in a smooth circular motion around the chamber. (B) Ether video- Wt flies eventually reach full recovery approximately in frame 210.
- Cha-ts1: (C) Control video- mutant flies move significantly less than Wt flies. (D) Ether video- flies move while recovering and then settle and remain stationary the majority of the time.
- ShakB[pos]: (E) Control video- mutant flies show greater activity than Wt flies. (F) Ether video- mutant flies reach full recovery by frame 870 with lower level of activity than control group.

- Figure 6 – Average total distance travelled in millimeters per second for each genotype:
  - (A) Graph of the three genotypes without any ether dosage. ShakB clearly is the most active while Cha is the least. (B) Graph of three genotypes after ether dosage. Little distance covered because flies are recovering. ShakB mutants did not return to full activity as they covered less distance than Cs during recovery. Cha remains the least active. (C) Graph of control and ether for each individual genotype.

Conclusion

- Cha-ts1 mutants has locomotion deficit.
- ShakB[pos] mutants show increased activity.
- CsJL flies recover to same level of activity of pre ether dosage while ShakB[pos] mutants don’t.
- Variances of ether recovery in mutants of different genotypes suggests that ether affects signalling between neurons.

Implications

- Developed protocol can be used to study para mutants, which spin and vibrate their wings under ether.
- Mosaic flies, with mutant and normal parts, would be interesting to study as well.
- Protocol can be modified to study other fly anaesthetics: CO2, ice, flynap.
- Results could shed light on how anaesthesia works on humans as well.

References