**Male fly response to spider and mantid ques, 2016**

May 19, 2016

**Prediction**: Male flies in vials with predator cues will be less active during the day than male flies in control vials. (see spider activity data: no activity in the dark, then a nice inverted U with peak in the afternoon. We still don’t have mantid activity data).

**Protocol for spiders**.

Use males from our wild fly population. Use 32 DAM vials and caps with holes.

Do egg laying for 3 days 12 to 10 days prior to test.

Prepare humidified chambers with lights on at 10 AM and off at 10 PM for holding the predators. Also prepare the 2 tupperwares for the DAMs and their lights. Food vials: each vial will contain ~4 ml food medium (12 mm high). Vials are 22 mm wide and 48 mm long with snap cap containing a ventilation hole in the centre. The horizontal distance between the food and the IF sensors is about 7 mm. An LED light bulb will provide illumination with lights on at 10 AM and off at 10 PM.

Get spiders and place them in B123 on 10-10 light cycle (Ian keeps them on 8-8).

Day 1. Thu. Feed spiders by adding 3 flies into each spider vial. Prepare 64 DAM vials with regular food (for both spiders and mantids).

Day 2. Fri. 1 PM. Move spiders individually into 16 DAM vials with regular medium + filter paper. Place in B123 chamber on 10 to 10 . Also place in Tupperware chamber in B132 16 empty food vials with filter paper.

Day 5. Mon 7:30 AM. Clear flies.

Day 5. Mon 11:30 AM. Remove spiders and silk, then place newly eclosed live-sexed males in each vial.

Monitors will be in their default vertical position with the vials being in horizontal position as they were with the flies.

Monitor 1: Top row: control. Bottom row: spider cues

Monitor 2: Top row: spider cues. Bottom row: control.

Start DAM.

Day 6. Tue. Stop DAM after a bit longer than 24 h.

Spider and Fly need: 16 spiders, 32 males.

**Protocol for mantids**.

Get mantids and place them on 10-10 light cycle in B123.

Day 1. Fri. Place many flies and a food bottle inside the mantid cage. Mantids will be with no food from Mon until Thu.

Day 4. Mon. Move mantids individually into 16 DAM vials with regular medium + filter paper. Place in B123 chamber on 10 to 10 . Also place in Tupperware chamber in B133D 16 empty food vials with filter paper.

Day 7. Thu 7:30 AM. Clear flies.

Day 7. Thu 11 AM. Remove mantids + feed, then place newly eclosed live-sexed males in each vial.

Monitors will be in their default vertical position with the vials being in horizontal position as they were with the flies.

Monitor 1: Top row: mantid cues. Bottom row: control.

Monitor 2: Top row: control. Bottom row: mantid cues

Start DAM.

Day 8. Fri. Stop DAM after a bit longer than 24 h.

Mantid and Fly need: 16 mantids, 32 males.