FIGHTING FLIES KRAVITZLAB 2004

MAKING A CHAMBER

Required components:

1 package plain microscope slides (Fisherbrand)

diamond glass cutter

steel or plastic ruler

Dow corning DAP 100% silicon household adhesive for ceramics and china foil lined closure for scintillation vial (15x10 mm – Kimble)

vial of standard fly food

Pasteur pipette with broken and fire polished tip to transfer food

agarose powder (Shelton molecular biology reagent)

dry yeast

small mortar and pestle

wooden toothpicks

black filter paper circles (8.5 cm) with 2 cm hole cut in center polystyrene disposable sterile Petri dishes (100x15mm) (VWR)

MilliQ water

Straight dissecting needle with birch handle

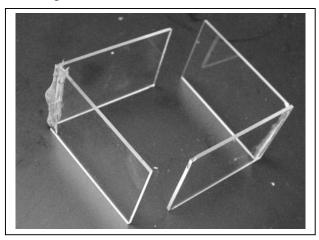
Colored lab label tape for covering hole in lid

Microwave oven and Bunsen burner



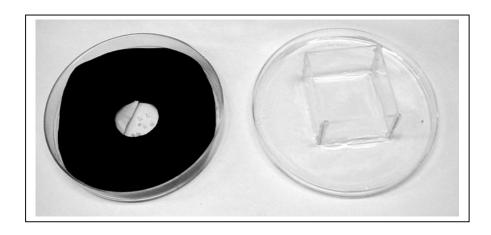
1. Assembling the chamber walls:

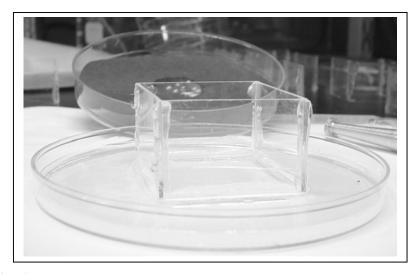
Take two plain glass slides, mark the middle at the two edges and using a straight edged ruler as a guide, score the glass with the diamond cutter. Slide the diamond cutter back and forth a few times until you hear a scraping noise. Wash the glass and holding it with two paper towels over a broken glass receptacle, apply gentle pressure close to the scored line with the scored side facing away from you to break the slide into two halves. Generously apply the glass adhesive along an edge of one piece of glass and push a second piece of glass against the adhesive at a right angle to the first piece and place down on a flat surface (aluminum foil, waxed paper or regular paper) and allow to set for at least an hour. After an hour repeat the application of the adhesive step to form a square chamber and allow to harden overnight.



2. Placing the walls in a Petri dish:

Take the top of a Petri dish and fill to a depth of at least 5 mm with a heated solution of 2% agarose (microwave the agarose in deionized water until it fully dissolves). Wait a few minutes, but while still dissolved, place the walls of the chamber in the center of the dish. Take the bottom of the Petri dish, place it bottom side up, and using a flame heated straight dissecting needle, make a set of small holes (>1mm) in the center of the dish for ventilation and a larger hole (ca. 4 mm diameter) slightly to the side for introducing flies to the chamber. Place a piece of removable label tape over the larger hole.





3. Making the food cup:

Heat a vial of fly food to the point where the food is melted and use a Pasteur pipette to transfer food to a scintillation vial closures filling it to form a flat surface at the top. Be careful to avoid air bubbles and allow to cool. Make fresh yeast paste by grinding a small amount of dry yeast in a mortar and pestle and adding a few drops of water. Add more yeast paste or water until the consistency is thick enough to allow a small visible drop to be picked up by a toothpick and applied to the center of the food surface. Place the vial of food in the center of the chamber and place the bottom of the Petri dish, inverted so that the sides are up and not obscuring the view, on top of the chamber. Introduce two 3-5 day old flies by aspiration into the chamber through the larger hole in the top, cover that hole with tape and place a piece of black filter paper with a 2 cm hole cut in it into the top dish. Place a desk lamp above the chamber, far enough away that it will not heat the chamber, and positioned so that it will illuminate the food surface. The entire assembly should ideally be placed in an environment with high humidity and a temperature of 22-25°C.



4. Begin videotaping through front wall of the chamber when both flies are on the food surface.