Grossfield, J. and J. Smith. Purdue University, Lafayette, Indiana.
Video taping Drosophila behavior.

In the course of working out some details of the behavior of species that require light in order to mate, it was necessary to ascertain whether or not the beasts made contact with one another in darkness (They do). The system we used has general applicability for the analysis of Drosophila behavior. A TV camera with its lens removed (A in Fig. 1) is mounted vertically on a trinocular dissecting scope. This allows the microscope adjustment to focus the camera. A 10X eyepiece is located in the phototube supporting the camera. A lower power would give a wider field of view. The problems of glare from wings and thorax and heavy shadows can be compensated for by diffusing incident light, placing a set of polarizers in the light path, balancing light with aluminum foil reflectors, and using a deep pile underlay (velvet) on the bottom of the lucite observation chamber. A 1/4 wave plate

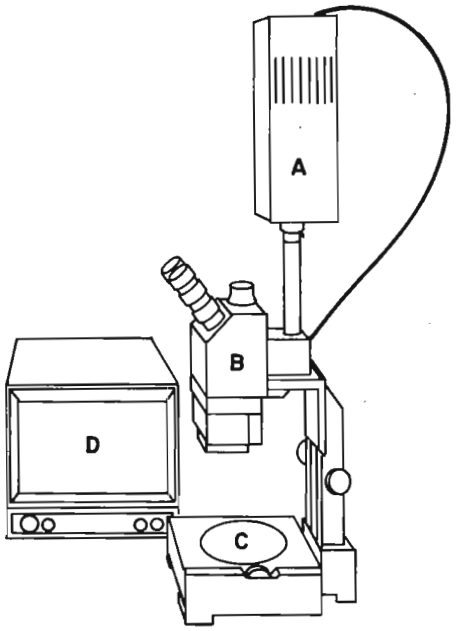


Figure 1. A. TV Camera; B. Dissecting scope with trinocular head; C. Observation Chamber, flat surface; D. TV monitor.

can also be used to cut glare. Any remaining glare can be compensated for by turning down the automatic gain control on both the camera and the monitor. A light coat of vaseline on the inside vertical surfaces of the observation chamber is reasonably effective in keeping the flies from assuming poorly photogenic positions on the corners or walls of the chamber.

For work in the dark a flashlight with a red filter (650 nm cut off, no UV transmittance) is a sufficient light source since the vidicon tube in the TV camera is sensitive to infrared light. IR Image Converter equipment can be used to work with wavelengths further towards that region of the spectrum (RCA laboratories, David Sarnoff Research Center, Princeton, N.J.).

A videotape recorder can be interposed in the system. This yields the capability to stop action at any point in a behavioral sequence and measure distances (angles of body parts, etc.) on the face of the TV monitor during playback. The videotape records can, of course, be stored to form a library of behavioral activities. In the long run this method is less expensive than using and processing 16mm film.

If you have sufficient funds to think about color TV, we'd be glad to hear where you got them.

Zalokar, M. Centre de Génétique Moléculaire, CNRS 91, Gif sur Yvette (France).
Fixation of Drosophila eggs without pricking.

Because of the impermeability of the vitelline membrane, the usual cytological fixatives can not penetrate the Drosophila egg and the egg has to be pricked to facilitate their entry. Only Carnoy fixative can be used directly and then only if its content of chloroform is higher than in recommended formulas, but this fixation shrinks eggs very badly.

Lipid solvents can penetrate the vitelline membrane, and if they contain a fixative in solution, they can carry it across the membrane. Any fixative which is soluble both in the solvent and in water will diffuse into the ooplasm and partition itself between its aqueous phase and the solvent according to the phase rule. If we want to fix an egg with 50% acetic acid, we should shake the solvent with the acid of this concentration. The solvent will take up the acid at the proper concentration so that the acid entering an egg submerged in the solution will reach 50%.

If we use a solvent which does not disrupt the egg lipids too drastically, we can achieve

fixation which is equivalent to fixation by the corresponding aqueous fixative. It was found that heptane or octane did not injure the cytoplasm unduly, while penetrating well through the vitelline membrane. An egg remains alive if submerged in these solvents for 10 minutes or more. The eggs become fixed in heptane loaded with acetic acid, picric acid, acrolein or glutaraldehyde, in less than one minute and can remain in the fixative for several minutes before beginning to shrink.

In order to facilitate the penetration of post-fixatives, colorants or dehydrating liquids, the vitelline membrane should be removed after initial fixation. To do this, the egg is transferred into the aqueous phase of the fixative and the membrane torn away with sharp needles. Surface tension helps to remove the membrane and the egg falls into the liquid. This operation can be performed best in 30% acetic acid, but after some practice, one can do it also in other fixatives.

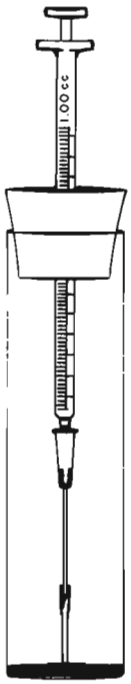
Fixation in heptane containing acrolein or glutaraldehyde is quite adequate for electron microscopy. Cell inclusions and organelles are well fixed, the ergastoplasm has its normal appearance and mitochondria have well perserved cristae. The following procedure is used:

1. Dechorionate eggs.
2. Fix in heptane which has been shaken with a 10% solution of acrolein or 25% solution of glutaraldehyde, for 1 to 2 minutes.
3. Remove the vitelline membrane in a buffered glutaraldehyde solution (conventional electron microscopy fixative).
4. Fix in the same solution for 1 hour.
5. Wash with buffered physiological solution.
6. Post fix with osmic acid 2 to 24 hours.
7. Further processing for embedding like any other tissue.

This fixation may be useful also in other cases where lipophilic membranes prevent the penetration of the usual fixatives, e.g. to fix Drosophila larvae and adults.

Félix, R. National Commission of Nuclear Energy, Mexico City, Mexico. A system for feeding liquids to adult flies.

The following method may be used as an alternative to injection of solutions to Drosophila flies, especially when several treatments with liquids should be tested in adults at separate time intervals. This system is particularly effective as the solution is administered during a period of time that may be lengthened to several days. It proved effectual for feeding cyclamates and cyclohexylamine to Drosophila melanogaster.



The liquid is gradually injected by means of a thin hypodermic syringe that goes through a hole of a rubber plug occluding the 2.8 x 9.0 cm vial, into a double layer of filter paper. The piece of polietilene tubing (Intramedic, Clay Adams, Inc.) adapted to the needle of the syringe, touches the filter paper, assuring a continuous delivery of the solution, when the embolus is pushed in.

The quality of filter paper cut to fit the bottom of the vial is important because it must be sufficiently absorbent to remain moist, without retaining an excessive amount of solution, which would drown the flies. Whatman 3 filter paper was used for such a purpose.

The syringe may be removed without the removal of the rubber plug, thus avoiding the escape, as well as the squashing of the flies, that occurs if the plug is removed and replaced. The amount of solution contained in the syringe (1.00 cc B.D. Yale turbeculin, Becton Dickinson) is enough to feed flies during several days. An additional pasteur pipette made at the laboratory with thin glass tube may be adapted through another hole, assuring the proper aeration of the vial, if the system is to be used during a period of several days without the removal of the flies.