

Cite as: Cold Spring Harb. Protoc.; 2007; doi:10.1101/pdb.ip35



## Information Panel

### Maintenance of a *Drosophila* Laboratory: General Procedures

**Michael Ashburner and John Roote**

Adapted from "Laboratory Culture of *Drosophila*," Chapter 35, in *Drosophila Protocols* (eds. Sullivan et al.). Cold Spring Harbor Laboratory Press, Cold Spring Harbor, NY, USA, 2000.

## INTRODUCTION

This article provides a general introduction to keeping *Drosophila* stocks, making and scoring crosses, mutagenesis, and controlling diseases in the laboratory.

## RELATED INFORMATION

General information on setting up a *Drosophila* laboratory can be found in [Culture of \*Drosophila\*: The Laboratory Setup](#).

## PROCEDURES

### Keeping Stocks

Most large fly laboratories maintain stocks that are not in everyday use at 18°C on a 4-5-week generation cycle. Stocks should be kept as two to four independent cultures, and it may be convenient to keep these on alternating generations, 2 weeks apart. Stocks are normally maintained in vials.

Most stocks can be kept by dump-transfer of flies to fresh vials. However, it is important to avoid too overcrowded cultures, and only 20 or so flies should be transferred. It is good practice to inspect the flies on transfer, to ensure that both sexes are present and that their phenotype is as expected. Fly laboratories may keep some stocks that require selection of each generation, and it is important that the stock keeper knows of any special requirements to keep any stock (these should be entered on the stock database, see below). The "sick tray" is an inevitable part of any stock room, a place where sickly stocks, or stocks going through a crisis, are kept under special attention. It is very good practice to keep the old cultures for 2 weeks (at 18°C) after transfer, so that they can be used as a backup should the new stocks fail for any reason.

### Collecting Virgins

Two general methods ensure that female *D. melanogaster* are virgin when used to set up a cross. These can be called the "biological" and "genetic" methods. Only the former is considered here; for genetic tricks useful for virgin collecting, see Chapter 12 of [Ashburner \(1989\)](#).

Although some variation between stocks exists, the general rule is that females will not accept a male mate until they are 10-12 hours old (i.e., after eclosion from the pupa). Thus, flies can be collected during this window (or, better, between 8 and 10 hr after eclosion), anesthetized, separated into males and females, and stored until needed in yeasted vials. The females will then usually be virgin when used. As a preliminary check, the vials that were used for storing the virgin females should be kept and inspected 3 or 4 days later for any signs of larvae. If

larvae are present, it is clear that at least one female in that vial was not virgin. Of course it does not matter too much if a single female is incorrectly stored with the males (as long as she is discarded); but a single male in the tube of females will play havoc. The rule for sexing for virgin collecting, especially when tired or rushed is: If in doubt, it is a male. The following is a convenient schedule for virgin collection.

*Day 0:* Clear all flies from emerging cultures in the late afternoon or early evening (e.g., 5:00 p.m. to 6:00 p.m.). Discard these flies. Store emerging cultures -- at 18°C in the dark.

*Day 1:* Put cultures at 25°C in the light, first thing in the morning. Clear all flies from the cultures ~1 hour later, anesthetize, separate into males and females, and store these in separate vials at 18°C until required. The young females, i.e., those that are relatively unpigmented and/or have unexpanded wings, will almost certainly be virgin. Check that the emerging cultures have no adult flies. Return the emerging cultures to 25°C in the light and possibly collect virgins last thing in the evening. Keep the "female" vials after using the virgins and inspect 3-4 days later for larvae. If present, presume that any females from that vial were nonvirgin at the time of use. (Note that the presence of eggs in the female-holding vials is not evidence of nonvirginity, even virgin females will lay eggs, albeit at a low rate in comparison with mated females.)

In practice, fly workers develop their own protocols for virgin collection that suit not only the flies, but also social and other activities. But please bear in mind, nonvirginity is *by far* the most common reason for an "unexpected" result from a cross. It is therefore very good practice to design crosses so that nonvirgin progeny will be evident by their phenotype, especially if unexpected nonvirgin progeny could confuse the analysis of an experiment.

### Setting Up and Scoring Crosses

It is impossible to give any universal rules for setting up or scoring crosses, since the precise protocol will vary from experiment to experiment. Crosses can be set up readily with a single pair of parents, although the failure rate can be quite high; normally one would use four pairs for crosses in vials and between five and ten pairs for crosses in bottles. Crosses can be transferred to new vials or bottles after 2 days. When scoring crosses, it is usually important to score at least once a day and to continue scoring for 9 days after the first progeny have emerged, since many genotypes have a delayed development (and/or a short life span). (Scoring for >9 days will cause confusion because F<sub>2</sub> flies may be emerging.)

### Mutagenesis

The three general techniques for mutagenesis in *Drosophila* are mutagenesis by irradiation, chemical mutagenesis, and genetic mutagenesis (i.e., by transposon insertion). Only the former two are discussed here. The choice between irradiation and chemical mutagenesis is determined by the objective of the experiment. In very general terms, only about one third of irradiation-induced mutations will be associated with chromosome aberrations, whereas most mutations induced with either of the two chemicals discussed here will be "point" mutations (usually due to single-base-pair changes; see Chapter 9 of Ashburner [1989] for a more systematic treatment, and Grigliatti [1998] for detailed protocols). In this section, we discuss only the general protocols for the mutagenesis itself, not the genetic schemes required to detect the desired mutations.

For all routine purposes, only 3-5-day-old males are mutagenized. After treatment, the males are mated immediately to harems of virgin females (usually as 20-pair bottles). These cultures should be transferred daily for 6 days. They can then be discarded or the males removed and the females further subcultured; this ensures that only postmeiotic stages are sampled and will avoid recovering clusters of identical mutations. Bottles should be labeled in such a way that identification of progeny from the same batch of parents is possible.

#### *Irradiation*

⚠ Flies may be irradiated with either  $\gamma$  or X rays. Many small laboratories use whatever source is conveniently available (e.g., a machine otherwise used for therapy); for purchase, a small industrial X-ray machine is strongly recommended. X-ray equipment, such as the Torrex TRX2800 and Torrex 120/150D, 24-inch cabinet, is available from Faxitron Corporation.  $\gamma$ -ray equipment is available from AEA Technology.

An X-ray machine has three advantages over a  $\gamma$ -ray source: (1) the relative biological effectiveness of X rays is higher than that of  $\gamma$  rays; (2) X-ray machines are safe when switched off, whereas  $\gamma$ -ray sources require expensive shielding that must be maintained; and (3) for  $\gamma$ -ray sources, the exposure time will need recalibration over time, since the source will be decaying (see Ashburner 1989 [p. 307]). Self-contained X-ray machines suitable for irradiation of flies, i.e., with an operating voltage of 100 kV or more and 5 mA, are available. These machines, designed for industrial use, need to be modified to remove damaging low-energy X rays for irradiating flies. This is done by inserting a 1-mm filter of aluminum or Perspex between the source and the flies. For irradiation, male flies are placed either in small plastic vials or gelatin capsules (use capsules with a 3-mm-thick wall for  $^{60}\text{Co}$   $\gamma$  rays). For the routine induction of mutations, and chromosome aberrations, use a dose of 4 kR with X rays and 5 kR with  $^{60}\text{Co}$   $\gamma$  rays. The dose rate of X-ray machines needs to be calibrated, which may be performed by the supplier, or a dosimeter can be used, for example, the Farmer Dosimeter 257D from NETechnology. A monitor should be used to test the machine for X-ray leakage, e.g., the Mini-monitor from Mini Instruments.

### Chemical Mutagenesis

⚠ *Ethylmethanesulfonate (EMS)*: The most convenient chemical for routine mutagenesis is EMS, administered by feeding to adult males. The standard dose for EMS (available from Sigma BioSciences) is 25 mM (0.24 ml of EMS in 100 ml of 1% aqueous sucrose, dispersed by repeated aspiration with a 10-ml syringe or P1000 micropipettor). Males are placed in an empty bottle and starved for 12-24 hours before being allowed to feed for 12-24 hours on the EMS solution. A piece of tissue paper (or a paper towel) is fitted tightly to the floor and sides of the bottle, so that the flies do not get trapped in nooks and crannies. Freshly made up EMS solutions can then be conveniently dispensed on the tissue paper with a P5000 micropipettor. (Starvation of males can lead to a high death rate, and consequent loss of yield; this is especially true if the males are already genetically weak. If so, keep them overnight in a food vial without any additional yeast.)

⚠ *Ethylnitrosourea (ENU)*: ENU is, like EMS, a very effective mutagen for *Drosophila*. Although EMS may induce chromosome aberrations, ENU is far less effective in this respect. ENU can be administered to flies in the same way as EMS. It can be purchased from Sigma as "Isopac" vials and should be freshly made up before use. To dissolve the ENU, make up a 0.01 M solution of sodium acetate buffer (pH 4.5), and inject it into the "Isopac" vial containing the ENU. Then dilute the ENU in a 1% sucrose solution until the final concentration of ENU is 5 mM (Grigliatti 1998).

### Controlling Plagues and Diseases

One reason for the success of *D. melanogaster* as a laboratory organism is that it is relatively resistant to plagues and diseases. The two most common problems are molds overgrowing the medium and mites. There have also been disturbing reports recently of viral infections in some laboratories. The culprit is apparently a picornavirus (*Drosophila C virus*, DCV) and the symptoms of infection are black, dying pupae. The extent to which fly laboratories suffer from molds and mites varies greatly, but there is some general advice that can be given. The most important advice is that prevention is better (*far* better) than cure; two main guidelines for preventing plagues and diseases are below.

*Cleanliness.* Keep a clean fly room, fly kitchen, and culture environment. For cleaning surfaces in fly rooms, use alcohol or a spray disinfectant, e.g., "Astell D."

*Isolation of new stocks.* Quarantine all incoming stocks, no matter from what source, even if the distributor swears that they are free of mold, mites, or viruses. A quarantine facility (e.g., a dedicated incubator) should be situated as distant from the normal fly and culture rooms as possible, and all materials, especially discarded vials, must be segregated from those in regular use. It is probably sufficient to quarantine for two generations, and only transfer the stocks to the regular facility when close inspection shows them to be free of infection or infestation. For flies brought to the laboratory straight from the wild, four generations of quarantine are recommended.

If an infection or infestation occurs, the first rule is to isolate all affected cultures to a quarantine facility. What is done next depends very much on the nature and extent of the problem.

### *Bacteria*

The most common bacterial problem is mucus-producing bacteria on the food, which often produce a reddish-brown pigment (e.g., *Acinebacter* sp.). The addition of antibiotics (streptomycin, tetracycline, or ampicillin) to the food at a concentration of 250 mg/liter is usually sufficient to cure the problem within one generation. If the problem is recurrent, then investigate the possible sources of the contamination (e.g., the yeast). The use of dextrose, rather than sucrose, in the fly medium should prevent most bacterial growth. The routine use of antibiotics in the food medium is not recommended, as this will inevitably lead to resistance.

### *Molds*

Molds, usually species of *Penicillium* or *Aspergillus*, are a common problem, as fly medium is an ideal substrate for their growth. With healthy cultures, the flies normally out-compete these fungi, but they can prove to be serious for weak stocks or for cultures at low density. It is now routine practice to include mold inhibitors in medium. Those most commonly used are Nipagin M or propionic acid. For both bacteria and molds, persistent infections that are refractory to treatment can best be overcome by washing eggs in 70% alcohol.

### *Mites*

Several species of mites can infect cultures of *Drosophila* (see Chapter 38 of **Ashburner 1989**). Broadly speaking, the mites may be interested either in the flies' food (food mites) or in the flies themselves. The fly mites are rarer than the food mites, but far more dangerous. Food mites often come in with the raw materials of fly medium (e.g., corn meal). For this reason, it is good practice to store bulk meal at -20°C and to be scrupulous about cleaning up any spills. One of the commonest causes of a serious mite infestation is allowing old fly cultures to fester in culture rooms or the fly room. These rooms must be inspected regularly by someone with the authority to autoclave old cultures without question. This is not an issue where the liberal social attitude so characteristic of fly labs can be allowed to constrain effective management.

If mites are found, then the affected cultures must be immediately quarantined (even better, autoclaved, but this is not always acceptable). If foam or muslin cotton-wool bungs are used, then replace these immediately with bungs of nonabsorbent, tightly balled cotton wool; these should prevent the mites spreading further.

Rapid (i.e., daily) transfer of stocks or cultures can rid them of mites, but this can be dangerous for weak stocks. An alternative is to collect eggs and wash them free of any mite eggs before transfer to clean vials, or to collect pupae on paper inserts and wash them free of mites and mite eggs in 70% ethanol, again before transfer to clean vials.

Tedion has been found to be effective against some common species of food mite. Tedion is available from the Sigma Aldrich Library of Rare Chemicals and also from local suppliers of agrochemicals. Dilute the commercial product (usually 8% active compound) to 5000 ppm in acetone and soak 7-cm filter papers in this solution. Allow filters to dry completely and introduce one into each culture.

Serious endemic mite infestations should never be allowed to build up. If they do, then seek professional advice to combat them, since they will require complete fumigation of all fly-handling rooms and equipment (see [Ashburner 1989](#) [p. 1218]).

### Viruses

In contradiction to the statements in [Ashburner \(1989, p. 1192\)](#), infection with the double-stranded RNA virus DCV has been found to be a serious problem in a few fly laboratories in recent years. Its symptoms are the presence of dying black pupae, particularly in old cultures. In addition, DCV infection seems to block the induction of transgenes under the Hsp70 heat shock promoter (T. Tully, pers. comm.). T. Tully (pers. comm.) has developed protocols for eradicating viral infection.

## REFERENCES

- Ashburner, M. 1989. *Drosophila. A laboratory handbook*. Cold Spring Harbor Laboratory Press, Cold Spring Harbor, NY.
- Grigliatti, T. 1998. Mutagenesis. In *Drosophila. A practical approach*, 2nd ed, (ed. D. Roberts), pp. 55–83. IRL Press at Oxford University Press, UK.
- 



### Caution

#### Ethyl methane sulfonate (EMS)

Ethyl methane sulfonate (EMS) is a volatile organic solvent that is a mutagen and carcinogen. It is harmful if inhaled, ingested, or absorbed through the skin. Discard supernatants and washes containing EMS in a beaker containing 50% sodium thiosulfate. Decontaminate all material that has come in contact with EMS by treatment in a large volume of 10% (w/v) sodium thiosulfate. Use extreme caution when handling. When using undiluted EMS, wear protective appropriate gloves and use in a chemical fume hood. Store EMS in the cold. DO NOT mouth-pipette EMS. Pipettes used with undiluted EMS should not be too warm; chill them in the refrigerator before use to minimize the volatility of EMS. All glassware coming in contact with EMS should be immersed in a large beaker of 1 n NaOH or laboratory bleach before recycling or disposal.

---



### Caution

#### EthylNitrosourea (ENU)

EthylNitrosourea (ENU) see *N*-Nitroso-*N*-ethylurea



### Caution

#### X-rays

X-rays, see Radioactive substances



## Information Panel

### Culture of *Drosophila*: The Laboratory Setup

Michael Ashburner and John Roote

Adapted from "Laboratory Culture of *Drosophila*," Chapter 35, in *Drosophila Protocols* (eds. Sullivan et al.). Cold Spring Harbor Laboratory Press, Cold Spring Harbor, NY, USA, 2000.

## INTRODUCTION

Since its introduction to experimental biology more than 90 years ago, *Drosophila melanogaster* has proved to be an easily cultured and robust laboratory animal. Although culture techniques and the ways in which flies are handled have changed over the years, if he were to enter a fly room today, T.H. Morgan would clearly recognize what is being done, and why. This article provides the basic methods for the laboratory culture of *D. melanogaster*. The intelligent culture of *Drosophila* requires a basic understanding of the life cycle of this fly. For this reason, we begin by describing the life cycle of *Drosophila* and then present information for setting up a fly laboratory.

## RELATED INFORMATION

For more detailed information on laboratory setup and maintenance, see [Ashburner and Thompson \(1978\)](#) and [Ashburner \(1989\)](#). [Greenspan \(1997\)](#) and [Roberts \(1998\)](#) are also very useful sources of information.

General procedures for maintaining *Drosophila* stocks, making and scoring crosses, performing mutagenesis, and controlling diseases in the laboratory can be found in [Maintenance of a \*Drosophila\* Laboratory: General Procedures](#).

## LIFE CYCLE

*Drosophila* is a typical holometabolous insect, i.e., its life can be divided into four stages--embryo, larva, pupa, and adult--there being a complete metamorphosis of body form from larva to pupa. In the laboratory, *D. melanogaster* is usually cultured at 25°C or 18°C, and we provide timings of developmental stages appropriate to these temperatures only.

Eggs are deposited by females on, or inserted in, the surface of the culture medium. A female may lay ~100 eggs per day at her peak. Embryonic development takes ~24 hours at 25°C (~40 hr at 18°C); the first-instar larva begins to feed immediately. The larva passes through two molts: from first to second instar and from second to third instar. The first-instar larvae feed on the surface of the medium; second- and third-instar larvae burrow into the medium. The mature third-instar larva leaves the food medium at ~96 hours (25°C) and wanders, searching for a site to pupariate. Pupariation occurs at ~120 hours (25°C), and it is within the immobile confines of the pupal case (in fact, the tanned third-instar larval cuticle) that metamorphosis occurs. Eclosion of adult flies occurs from the pupal case about 9 days (25°C) (19 days at 18°C) after egg laying. Adult females will not mate until they are ~12 hours old; segregation of males and females during the first 8-10 hours of adult life is a convenient way of ensuring that the females are virgin (see [Maintenance of a \*Drosophila\* Laboratory: General Procedures](#)).

## LABORATORY SETUP

For the simple culture of *D. melanogaster* and routine genetic experiments, the following equipment and services are either necessary or to be greatly desired. **Table 1** lists the specifications and sources of items discussed below. Much of the basic equipment is adapted, rather than specifically made, for fly culture. For this reason, only examples of sources are provided in **Table 1**, and the best advice for those newly setting up a fly laboratory is to talk to groups in their locality. There are specialist companies that deal in equipment for fly laboratories; examples are LabScientific, Inc. and Applied Scientific.

### Fly Food Kitchen

The facility for the production of fly food and for washing used vials and bottles can range from a very simple kitchen to one that is complex and expensive, depending on need. To maintain a handful of stocks, or set up a few crosses a week, all that is required is a domestic saucepan, a gas or electric ring, and a wooden spoon. We describe here a kitchen suitable for a reasonably large laboratory, producing 100 liters of food (enough for 10,000 vials) a week. Although this could be done entirely by hand, some investment in automation is well worthwhile. The most important equipment includes a steam kettle for preparing medium and a peristaltic pump for dispensing medium into bottles and vials.

*Bottles and vials:* *D. melanogaster* are usually cultured in 250-ml bottles, 40-ml vials (100 x 25-mm diameter), or 30-ml vials (75 x 25-mm diameter), which may be either plastic, and hence disposable, or glass, and hence reusable after washing. To keep the flies within the bottles or vials, a variety of stoppers can be used, which, again, may be disposable or washable and reusable (at least until they fall apart). The most commonly used plugs are made of polyurethane foam (3 x 3-cm or 5 x 5-cm diameter) or nonabsorbent cotton wool. Plugs can be washed and reused, but are not mite-proof, whereas cotton wool has the disadvantage that some people are irritated by cotton fibers in the air. Applied Scientific now supplies a disposable mite-detering plug, called the Buzz Plug, made from cigarette filters (cellulose acetate). This has the advantage of not shedding but the disadvantage of cost. Stock or cross labels can be written on the bottle, or special tags (e.g., those available from LabScientific) can be used.

*Medium (fly food):* A number of different recipes for fly food are in use, and examples of the most popular, with general sources of their ingredients, are given here:

-  General-purpose medium for *Drosophila*,
-  Yeast-glucose medium for *Drosophila* (suitable for growing larvae for polytene chromosome preparations).

**Table 2** lists the sources of ingredients used in the preparation of these media, as well as commercial suppliers of instant media.

### Microscopes

Although simple fly culture does not require microscopes, and males and females can readily be sexed with the naked eye after a little practice (especially by the young), any serious fly work requires many hours of careful observation of living flies under a dissecting microscope. Important considerations for the purchase of a dissecting microscope are (1) overall optical quality (which should be as good as one can afford); (2) magnification range (6X-40X is adequate for most purposes); and (3) a light source (a "cold" light, such as from a fiber-optic source or a halogen lamp with a heat filter, is recommended). Many investigators prefer a microscope with a zoom, rather than fixed magnification settings. For the detection of GFP (green fluorescent protein) in living flies, a microscope with a suitable UV light source is required.

## Autoclave

Old cultures should be autoclaved. For cultures in glass bottles or vials, this is essential before they are washed. Autoclaving is also recommended for cultures in plastic vials because it will prevent release of flies, in particular the release of transgenic flies. The size of autoclave used will depend on the volume of use.

## Anesthetization Equipment

Traditionally, fly pushers anesthetized their flies with diethyl ether, but because it is both safer and generally more pleasant, most (except the elderly) now use carbon dioxide. Normal commercial-grade CO<sub>2</sub> as well as gas regulators for CO<sub>2</sub> tanks are readily available from many suppliers. To accommodate CO<sub>2</sub> tanks, a fly room should have racks that can be conveniently made or purchased, and automatic equipment for switching between tanks can save much grief. For any large fly laboratory, permanent piping of CO<sub>2</sub> at the bench is very worthwhile. CO<sub>2</sub> should be humidified before use, which can easily be done by passing the gas through a large flask of water. Our only recommendation here is to use a heavy-duty flask and bungs to avoid blowouts due to pressure. If an air line is available in the laboratory, it can usually be easily adapted by a professional plumber to provide CO<sub>2</sub>.

Each user should have a CO<sub>2</sub> pipeline from the humidified CO<sub>2</sub> source, which passes through a local valve that allows the supply pressure to be regulated and turned on and off. The pipeline then divides into two arms (using a Y-junction), a meter or so from the end. One arm goes to the CO<sub>2</sub> plate and the other to a tube that can be inserted past the bung into a vial or bottle. The second arm should have a spring-clip regulator.

The CO<sub>2</sub> functions to deliver the gas to flies and to keep them anesthetized while being examined or handled. The plate is normally a shallow vessel that sits on the microscope stage. Gas is passed into this vessel and escapes through the surface of the plate, which is constructed of a white porous material, typically sintered glass or porous polyethylene. (If the surface of the CO<sub>2</sub> pad becomes clogged with the debris of dead flies, it can be cleaned with sandpaper.) Plates can be purchased from fly equipment suppliers or constructed from Perspex by a competent workshop technician. Post a large notice--"HAVE YOU TURNED THE GAS OFF?"--that is visible to all as they leave the fly room.

## Controlled Temperature and Humidity Environment

Although *D. melanogaster* can be cultured happily at room temperature, any serious use requires a controlled temperature and humidity environment. These can either be stand-alone biological incubators, from any good manufacturer, or specially constructed rooms. Some incubators suffer from serious corrosion problems, especially if propionic acid is being used as a mold inhibitor. The cooling coils should be protected, for example, by enamel paint, or protected coils should be obtained at the time of purchase from the manufacturer. The temperature range should be between 18°C and 29°C (these being the extremes for most normal purposes), and the specification should demand a maximum deviation of ±0.5°C. Humidity is also important, but the requirement for humidity control will vary with geographical location and season. In general, 70% relative humidity (RH) is necessary.

Humidifiers should be of the type that releases a fine mist (rather than steam) into the environment. It is preferable that constant temperature rooms also be air-conditioned, with the circulating air being prehumidified. It is difficult to maintain a high humidity in most laboratory incubators. Those marketed by LMS have, with *Drosophila* culture in mind, modified the cooling coils to reduce the loss of humidity normally caused by condensation on the coils.

Whatever controlled environment is used, some factors are critical. Most important of these is that the temperature control is fail-safe. The environment should be designed so that overheating can never occur. The cooling system must be such that it cannot, under any circumstances, freeze the flies. There must be continuous monitoring of

temperature and humidity, noted in a permanent record. In any large facility, a hand-held, digital temperature probe should be regularly used to test for hot or cold spots in the controlled chamber. Finally, any serious fly laboratory must have its temperature-controlled environments wired to a commercial service that alerts users by phone at any time of day or night (usually, in our experience, at ~4.30 a.m.) if the temperature deviates from predetermined tolerances (typically more than 1°C above or 3°C below 25°C, with a decreased higher tolerance at 29°C, e.g., 0.5°C, as well as a decreased lower tolerance at 18°C, e.g., 1°C).

Any enclosed space within which flies are cultured should be designed for ease of cleaning. This means an open floor space (e.g., shelving should end at least 12 inches above the floor), with rounded, rather than right-angular, junctions of walls and floor. There should be no nooks or crannies in which nasties can hide and the shelves should be of the types that are easily cleaned.

### Ancillary Equipment

Fly workers have their own favorite collection of equipment for handling flies, including a selection of fine (camel hair) brushes (e.g., grade 00), forceps (e.g., watchmaker's forceps, size 4 or 5), and perhaps carefully manicured feathers (flight feathers of geese are said to be the best) for pushing flies around a plate. In addition, small hand-held digital counters are useful for those with a poor short-term memory, and a morgue for dead and discarded flies is essential. Morgues are simply vessels filled either with a very light oil or methylated spirits. A thick rubber mat is useful for deadening the sound of bottles being banged against the laboratory bench.

Flies escape into the fly room or culture room even in the best-run laboratories, and it is essential to trap as many of these as possible. Three methods can (and should) be used. Old-fashioned, sticky fly papers are remarkably effective. Liquid traps, of which there are a variety of beautiful Victorian designs, filled with a solution of live yeast and glucose are also effective. Finally, some laboratories use commercial fly electrocution devices, such as are typically sold to food shops, but these are not effective because the space between the wires is generally too large for *Drosophila*.

## REFERENCES

- Ashburner, M. 1989. *Drosophila. A laboratory handbook*. Cold Spring Harbor Laboratory Press, Cold Spring Harbor, NY.
- Ashburner, M. and Thompson, J. 1978. The laboratory culture of *Drosophila*. In *The genetics and biology of Drosophila* (eds. M. Ashburner and T.R.F. Wright). Vol. 2a, pp. 100–109. Academic Press, London.
- Greenspan, R. 1997. *Fly pushing*. Cold Spring Harbor Laboratory Press, Cold Spring Harbor, NY.
- Roberts, D. 1998. *Drosophila. A practical approach*, 2nd ed., (ed. D. Roberts). IRL Press at Oxford University Press, UK.

Copyright © 2007 by Cold Spring Harbor Laboratory Press. Online ISSN: 1559-6095 [Terms of Service](#) All rights reserved. Anyone using the procedures outlined in these protocols does so at their own risk. Cold Spring Harbor Laboratory makes no representations or warranties with respect to the material set forth in these protocols and has no liability in connection with their use. All materials used in these protocols, but not limited to those highlighted with the Warning icon, may be considered hazardous and should be used with caution. For a full listing of cautions, [click here](#).

All rights reserved. No part of these pages, either text or images, may be used for any reason other than personal use. Reproduction, modification, storage in a retrieval system or retransmission, in any form or by any means-electronic, mechanical, or otherwise-for reasons other than personal use is strictly prohibited without prior written permission.

## This article has been cited by other articles:



Cold Spring Harbor Protocols

▶ HOME

R. M. Parton, A. M. Valles, I. M. Dobbie, and I. Davis

### **Isolation of Drosophila Egg Chambers for Imaging**

Cold Spring Harb Protoc, April 1, 2010; 2010(4): 10.1101/pdb.prot5402.

[\[Abstract\]](#) [\[Full Text\]](#)

---



Cold Spring Harbor Protocols

▶ HOME

R. M. Parton, A. M. Valles, I. M. Dobbie, and I. Davis

### **Collection and Mounting of Drosophila Embryos for Imaging**

Cold Spring Harb Protoc, April 1, 2010; 2010(4): 10.1101/pdb.prot5403.

[\[Abstract\]](#) [\[Full Text\]](#)

---



Cold Spring Harbor Protocols

▶ HOME

R. M. Parton, A. M. Valles, I. M. Dobbie, and I. Davis

### **Drosophila Macrophage Preparation and Screening**

Cold Spring Harb Protoc, April 1, 2010; 2010(4): 10.1101/pdb.prot5404.

[\[Abstract\]](#) [\[Full Text\]](#)

---