

Lee, William R. The University of Texas. Feeding radioactive isotopes to specific larval stages of Drosophila melanogaster.

In previous work on feeding radioactive isotopes to Drosophila, flies during their entire life cycle have been kept in contact with or close to media containing the isotope. As a result the proportion of mutations induced by beta rays

from the media is high in relation to those induced by the incorporation of the isotope into the genetic material. Pulse labeling can be accomplished by injection; however, results reported by Kaplan et al. (Genetics 49: 701-714) (also observed in this laboratory) show poor labeling of sperm from males injected with tritiated thymidine. Kaplan obtained better labeling by feeding larvae. Cytological considerations indicate the desirable time to feed would be just before synthesis of DNA prior to meiosis. Therefore, it is desirable to pulse label larvae during the first 24 hours of the third instar.

To accomplish feeding a radioactive isotope during a specific larval stage, 63 mm diameter and 29 mm deep Stender dishes are filled half full of corn meal Drosophila media. A sheet of black satin through which larvae cannot pass is pressed into contact with the surface of the media and is allowed to extend up on the sides of the Stender dish to prevent larvae from crawling over its edge into the media below. 0.05 ml of a solution containing 50 microcuries of phosphorus-32 as inorganic phosphate is pipetted onto the surface of the satin which initially absorbs most of the P-32. The surface of the satin is then sprayed with yeast and the Stender dish allowed to incubate at 34°C for three days. At the end of the three days the satin is covered with a growth of P-32 labeled yeast.

Larvae from a three day old Drosophila culture are floated out with a concentrated sugar solution and third instar larvae picked out with a brush. For convenience these larvae are stored in petri dishes on moistened filter paper during the collecting process. The larvae are then washed into a funnel screened with a disk of nylon chiffon. This concentrates the larvae into a cluster which is then brushed onto a 4 cm diameter disk of nylon chiffon. This disk is then transferred by 10 inch tongs to a Stender dish containing the labeled yeast.

Larvae are allowed to feed on the labeled yeast for 24 hours; then, the satin containing the larvae on its upper surface is removed with the tongs and placed in a funnel over a jar to collect radioactive materials. The funnel is lined with nylon chiffon which retains the larvae but allows the radioactive yeast and bits of media to be washed through. A fine stream of water from a polyethylene squeeze bottle is directed at the larvae and they are washed until further washing does not reduce the radioactivity. The nylon chiffon containing the washed larvae is then placed into a half-pint bottle with Drosophila media for pupation.

Radioautographs of sperm from females inseminated by males treated in the above manner with either P-32 or tritiated thymidine showed heavy uniform labeling during the first three days of mating yet the male germ cells had been subjected to radiation from radioactive media for only one day and during the relatively insensitive spermatogonial stage.

The 4 mm thick glass of the Stender dishes provides good shielding against the Beta rays of P-32. A wrist film badge worn during these operations has not recorded any significant radiation.

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Strickberger, M. W. and L. Harth. St. Louis University. Ready-Made heated dispensers for Drosophila medium.

A simple dispenser for Drosophila medium is an ordinary automatic coffee percolator of the 32 cup variety or larger that is sold quite cheaply in hardware stores as "party" coffeemakers (our's cost \$10.00). They are fitted with a

spigot that easily controls the amount of medium dispensed and they have a warming element which keeps the medium hot and fluid. To use, remove the inside coffee "basket", add the cooked hot medium, and connect the apparatus to electricity. The hot medium will prevent the high voltage "percolator" element from operating and will restrict heating to the low voltage "warming" element. If desired, the high voltage heating element can be permanently disconnected by reconnecting the wiring so that its circuit is by-passed. In those instances when all of the medium is quickly dispensed through this device, use of the electrical warming element is not necessary.