

Baumann, J.L. and W.L. Bischoff. University of Toledo, Toledo, Ohio. A rapid reliable assay for glucose and fructose specific hexokinases in crude extracts of *D. melanogaster*.

The initiation of a series of experiments designed to cytologically localize the hexokinase loci in *D. melanogaster* necessitated the development of a satisfactory spectrophotometric assay for this system of enzymes. Published methods designed for use with various mammalian systems proved to be unreliable due to a lack

of linearity attributable primarily to inappropriate substrate concentrations and pH optima. These problems have been overcome in our laboratory through the use of the assay procedure described below.

A. Preparation of extracts: Three to ten adult males or females aged 5±1 day post-emergence are ground in 2 ml glass microhomogenizers containing 0.05 ml of 0.03 M tris-HCl buffer at pH 8.5/fly. Homogenates thus prepared are centrifuged at 12,000 xg in the cold for 20 minutes, the supernatant fraction serving as a source of enzyme. Assays were performed within three hours of homogenization. Storage of extracts even at -70°C results in a total loss of activity after 48 hours.

B. Assay: 0.56 ml of a reaction mixture composed of 0.015 M glucose or fructose, 0.02 M MgCl<sub>2</sub>·6H<sub>2</sub>O, 0.00013 M NADP, 0.00001 M EDTA (disodium salt) and 0.12 units of glucose-6-phosphate dehydrogenase in 0.02 M tris-HCl at pH 8.5 is mixed with 0.02 ml of the above enzyme extract in a 0.75 ml Helma quartz cuvette and gently agitated. After 1 minute 0.02 ml of 0.03 M ATP is added to initiate the reaction. To assay fructose phosphorylating activity 0.12 units of phosphoglucose isomerase are added to an otherwise identical reaction mixture. Reference cuvettes contain reaction mixture, enzyme, and 0.02 ml of tris-HCl buffer in place of the ATP. The reduction of NADP is monitored at 340 NM, and under the above condition is proportional to enzyme concentration. This assay is linear for at least 30 minutes and has been successfully utilized for the cytological localization of two hexokinase loci. The results of these studies will be reported elsewhere.

Boulétreau, M. and P. Fouillet. University of Lyon, Villeurbanne, France. An accurate and reliable olfactometer.

A new olfactometer was developed in order to measure the behavioral response of flies to various odoriferous substances. The greatest concern was to allow flies to move and fly freely in a sufficient space during the exper-

iment, to prevent the olfactory cue from interfering with other directional signals such as light, drafts or gravity, and to provide an accurate control of the composition and the concentration of the odor to be tested. The device has two main originalities: the use of a large, well aerated cage in which adults can fly and exhibit normal behavior towards an odoriferous source; the use of a gas mixing pump (Wüsthoff) which mixes gases in definite ratios and delivers mixtures of reliable composition.

The cages (Fig. 1, 6) are made up of clear plastic boxes (23 x 17 x 10 cm) with large gauze panels arranged on the top and the walls. The traps (Fig. 1) are fitted on the sides of the cages and can be renewed at given intervals without disturbing the flies. Traps are fed with gas mixtures to be tested. The whole arrangement is drawn in Fig. 1: atmospheric air is compressed with a diaphragm pump (1) and dried on silica gel columns (2). It is divided into two flows respectively saturated with vapors of substances A and B in convenient saturators (3) before reaching the pump (4). The pump mixes flows (a) and (b) in the required ratios and delivers two mixing ratios M1 and M2 each of which feeds two traps. The flow in each line is regulated using a flowmeter (5) and a needle valve so as to provide 25 ml/min to each trap.

The reliability of the device was tested by measuring the response of a wild strain of *D. melanogaster* to various concentrations of ethanol. Flow (a) is first saturated with ethanol vapor by bubbling twice in 100% ethanol and then conveniently diluted with flow (b), which is kept pure. The pump delivers two concentrations of M1 and M2 of ethanol vapor which are expressed as a percentage of concentration in flow (a), which is considered as saturated. Standardized flies, 6 days old and starved for 24 hours on water + agar are put in lots of 500 (250 of each sex) in each cage. Each cage is fitted with only one trap, so that four tests are run simultaneously. The cages are 1.5 m below two 40 W fluorescent lamps, and are kept at 25° and 30% R.H.

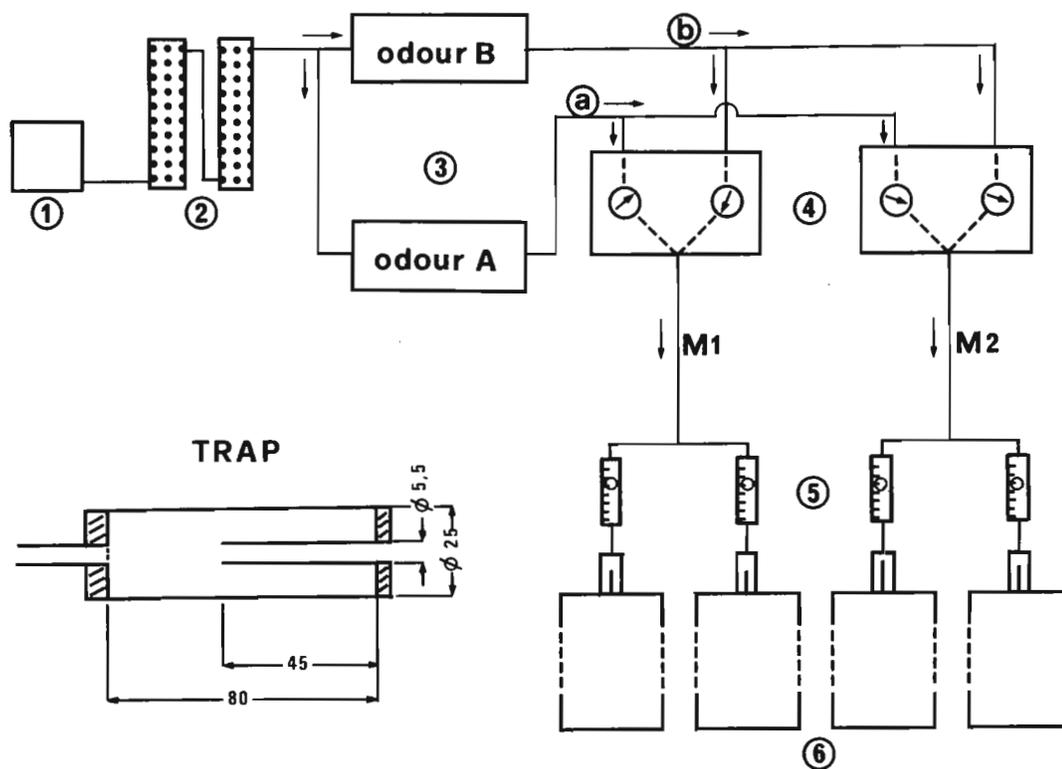


Fig. 1. Connection of the olfactometer with the air-delivery system (see text).

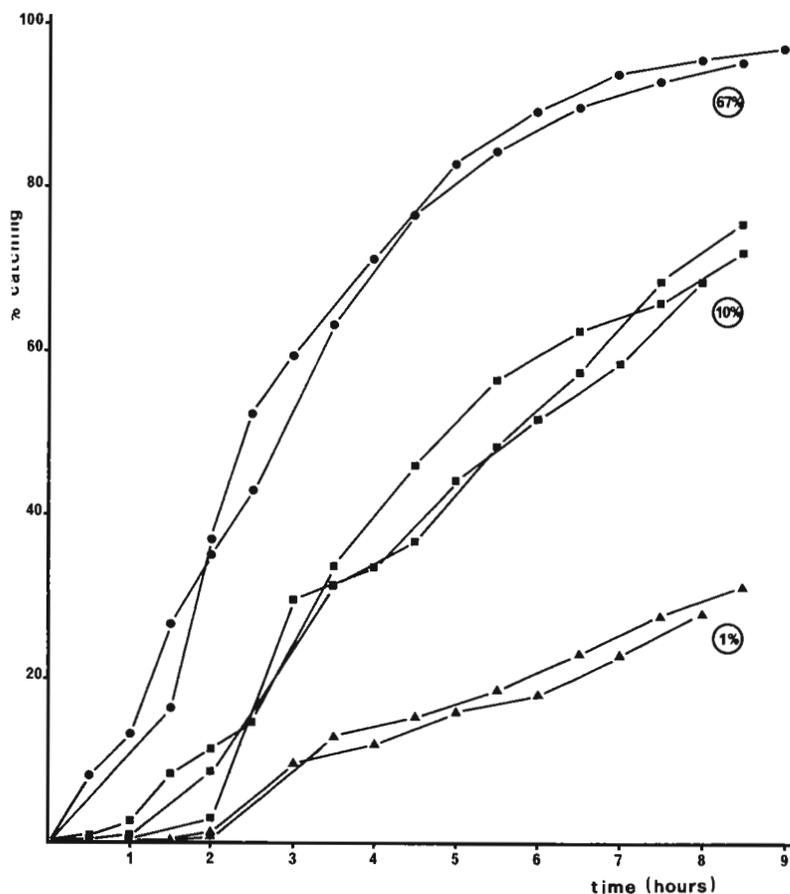


Fig. 2. Response of D.m. (wild strain) to various concentrations of ethanol vapor in dry air (1%, 10%, 67%).

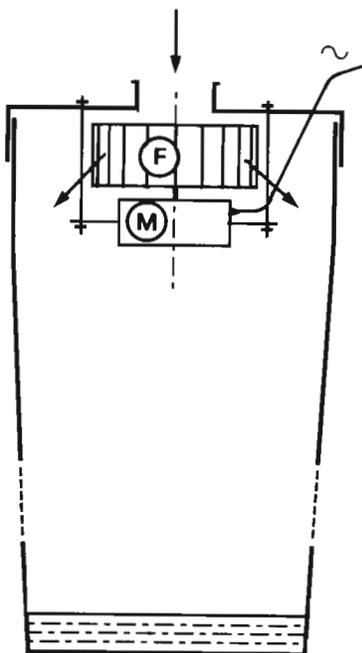
Fig. 2 gives the kinetics for three concentrations of ethanol. Each curve corresponds to the mean of two parallel boxes and is based on 1000 flies. The different curves given for each concentration were obtained in different experiments. Their similarity demonstrates the reliability of the device. The curves fit well with an exponential model, thus indicating the constancy of individual response throughout the experiment. Each concentration can thus be simply characterized by the proportion of flies which enter the trap per unit of time. No decrease in ethanol attraction occurs at higher concentrations. This observation is opposite to previous results (Fuyama 1976; Carton 1977). This difference is likely to result from the quite different methods: our device allows the humidity to be kept constant (here: 0%) whatever the concentration of ethanol. The only effect to be measured is that of ethanol. Other methods

lead necessarily to a simultaneous variation of ethanol and water contents in the tested air flow. Since in these experiments flies had to choose between this air flow and a control saturated with water, the authors actually measured some interaction between the response of flies towards ethanol and their response towards humidity. High concentrations of ethanol probably act as air dessicators, which could explain the observed repulsion.

Choice experiments can also be carried out with this olfactometer: by adjusting pump inputs and fitting two traps to each box, flies can be given a choice either between two different concentrations, or between two odor mixtures.

References: Carton, Y. 1977, Coll. Int. CNRS Tours (Fr.) 285-303; Fuyama, Y. 1976, Behavioral Genetics 6:407-420.

Boulétreau, M. and O. Terrier. University of Lyon, Villeurbanne, France. A device for getting rid of excess adult flies.



Routine rearings or experimental plans often require the daily destruction of large numbers of flies. A simple device was developed to prevent flies from escaping in the lab and to avoid disadvantages of traditional devices.

A weak electric motor (M), fitted with a plastic fan (F), hangs on the cover of a cylindrical plastic spice jar (1.5 liter). A 30 mm hole is pierced through the wall of the jar, 10 cm above the bottom. 100 ml water, added with a few drops of household detergent, are poured into the device.

By gentling drumming inverted vials or tubes above the upper hole, flies are allowed to be sucked down by the air swirl. They immediately sink to the bottom. None escape or float on the surface, thus allowing the quick drowning of next victims and making the capacity unlimited.

Once the daily holocaust is completed, the cover is removed, the jar is water rinsed and provided again with water + detergent. Years of daily use proved the device to be very efficient and suitable.

Crespí, S. and O. Cabré. Autonomous University of Barcelona, Bellaterra, Barcelona, Spain. A simple method for electron-microscope visualization of *D. melanogaster* embryo polysomes.

of the traduction complex. Here, we propose a very simple analytic method, with mild conditions, and material proceeding from only one egg. It allows the study by electron microscopy of processes related to translation, with minimum interference between the experimental treatment and its visualization.

The method consists of dechorionizing one egg in the embryonic stage that is to be studied. The egg is disrupted in 50  $\mu$ l of Na borate buffer  $\mu$ M pH 8.5, and left 10 min. at room temperature. 20  $\mu$ l of the sample is placed on a carbon-coated grid (300 mesh), and

The common techniques of polysome and ribosome preparation are based on relatively complex methods in which tissue homogenates, gradient centrifugations, etc., are used. These preparative methodologies are characterized in subjecting the samples to drastic treatments which can alter the native stage