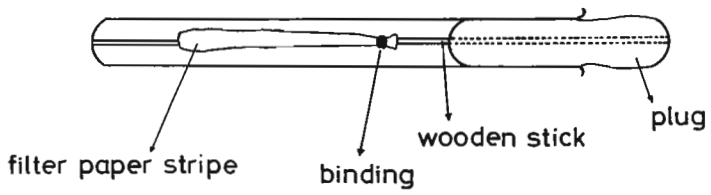
tube has also been flamed.) Holding the tube in a vertical position, the flies are shaken out of the pipette by knocking its tip against the wall.

Fig. 3



Collection of timed eggs: For this purpose, we use an empty test tube, the plug of which has an inserted wooden stick reaching down to the end of the tube. There is a strip of rough filter paper bound to and lying along the stick (Fig. 3). The tube and the insert are sterilized together, then the upper side of the filter paper strip is covered with a few drops of melted, sterile medium. When it has solidified, the plug with the insert is put into another empty, dry test tube into which the egg-laying flies have been transferred. The flies will lay their eggs onto the strip. The test tube is kept in the dark and the plug with the insert changed for new ones at convenient times, under sterile conditions. The paper strip with the eggs is cut off the stick and placed onto the surface of a sterile slant medium.

Sterility probes are not necessary to make for each subculture. However, it is advisable to avoid mixing the flies from different tubes and to separate, by marking, the tubes with eggs from different fly populations. In the case of any contamination, this helps in identifying and separating the possibly contaminated cultures. For checking yeast contamination, a small sample of the medium or a few flies are suspended in 5 ml of sterile broth (0.5 g yeast extract and 1 g glucose in 100 ml tap water, autoclaved) in a test tube and incubated at 29°C. Yeast growing in the settlings can be seen in 3 days after inoculation.

Parente, A. and R. Arking. Wayne State University, Detroit, Michigan. A convenient method of collecting moderately large numbers of eggs from D. melanogaster.

Previous techniques for collecting moderately large numbers of synchronous eggs have been tedious because they usually require large numbers of adults maintained in a population cage. We wish to present an alternative technique for collecting variable numbers (500-5000 eggs/collection) of highly synchronously developing embryos. Peak egg production in Drosophila occurs between 5-15 days after eclosion. To maintain this high level of efficiency it is essential to have an adequate supply of matured females available at all times.

In our laboratory this is accomplished by raising Drosophila in a rotational sequence under optimal conditions. Newly eclosed adults are placed on food and aged for 4 days at 22°C. On the 5th day, the adults are ready to be used for egg collection. This is done by placing 150 females and 75-100 males in a wide mouth glass bottle, the mouth of which is covered by a scored, yeasted, agar plate. The bottle is then inverted and placed in a totally dark incubator free from disturbances. The females are allowed to lay eggs, and at the end of the desired time periods the old plates are removed and new ones put in their places. The eggs may then be left on the agar plates to incubate for some predetermined time period or may be washed off the agar with a stream of water into a funnel containing an appropriately sized wire mesh. Adult females older than 10 days are discarded or used for stock and replaced by a new supply of mature females.

Table 1.

Collection period	No. females per bottle	No. eggs per plate Range	No. eggs per plate Mean	No. eggs per female per hr.	Percent synchrony	Percent abnormal*
30 min.	120	116-204	164	2.73	100	0
1 hr.	120	775-915	840	7.00	100	5-6
2 hr.	120	1000-2000	1500	6.25	81	10

*As measured 1-3 hours after collection.

Table 1 summarizes some of our experiences with this technique, showing the averaged results from three separate samples, each of which involved a 1 hr. pre-collection followed by an egg collection of the indicated time. It is evident that the production of eggs peaks at 1 hr. and appears to slowly decline thereafter. If the pre-collection has been sufficient, then the percentage of synchronously developing embryos stays very high and only decreases after 2 hours. The percentage of abnormally developing embryos also increases as collection time increases but never exceeds 10%. If synchronously developing embryos are not an important consideration, one can use longer egg laying periods (Table 2) and obtain much larger numbers of eggs per plate. It can seen, however, that egg production per female decreases linearly to very low levels by 6 hours. Therefore, females are not pushed for more than 4-6 hours of continuous collections. In our hands, then, the optimal collection time is one hour, which will produce a high level of synchrony, minimal numbers of abnormal embryos and a moderately large number of eggs per plate.

Table 2.

Hours	Collection	No. females per bottle	No. eggs per plate Range	No. eggs per plate Mean	No. eggs per female per hr.
0-2	Pre-collection	120	2000-4000	3000	13
2-4	Collection #1	120	1000-2000	1500	7
4-6	Collection #2	120	500-1000	750	4

In a 4 hour day it is, theoretically, possible to collect 20,000-40,000 eggs excluding the pre-collection. In practice, this technique has been able to produce a minimum of 28,000 embryos in an 8-day period. This figure is a minimum estimate since it does not include an approximately equal number of eggs which were discarded as surplus. These collections were done using anywhere from 8-15 collection bottles which held the adults obtained from 15-30 food bottles. This gave us an average yield of about 3,500 eggs per day, a figure which is in general agreement with the data contained in Table 1. As a Drosophilist may know, egg production is variable and there have been times when we have obtained little or no eggs; conversely, there have been times when we have obtained 40,000 eggs in one collection. In summary, we have devised a technique which is as productive as a population cage, and which can produce a moderately large number of eggs using a smaller number of adults, and which is more responsive to varying daily needs for embryos.

Pasic, T.R. and H. Nickla, Creighton University, Omaha, Nebraska. A BASIC program for construction of fate maps.

A computer program in BASIC has been developed to aid in the construction of blastula "fate maps". Early mitotic misdivisions in the embryo can be induced by an unstable ring X-chromosome. This results in two clones of nuclei randomly oriented with respect to the blastula cortex and differing in X-chromosome complement. When the nuclei migrate and become fixed to the cortex, a random plane of division divides the blastula. The probability of a plane of division falling between any two imaginal discs increases as the distance between two imaginal discs increases (Hotta and Benzer 1972).

Adult mosaics can be scored for any number of surface landmarks on the basis of tissue type (1 = male, 2 = female, 3 = mixed). Each scoring is entered into a data table at the location (A,B) where A is a unique integer for each fly (1,2,3, . . .) and B represents each landmark in coded form (1 = eye, 2 = wing, etc.). The computer effects a two-by-two pairing of landmarks and counts the number of times that the two scorings are different. This sum represents the number of times a plane of division fell between the imaginal discs. The sum is converted into a percentage and prints it as the distance (in Sturts) between each pair of imaginal discs. (Note: The total number of pairings is (N**2-N)/2 where N is the total number of landmarks.)

Other subroutines in the program include the distance between homologous landmarks; total percent male tissue, percent male tissue at each landmark and percent male tissue of each fly. The program was tested using data on 196 mosaics having 39 surface landmarks. The program is available upon request.

Reference: Hotta, Y. and S. Benzer 1972, Nature 240:527.