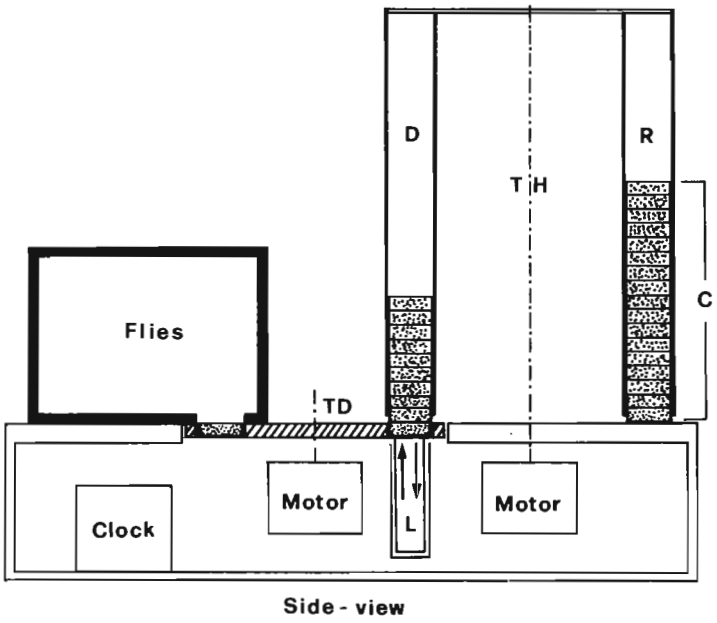
Allemand, R., J. Biston and P.M. Mallet. University of Lyon I, Villeurbanne Cedex, France. An apparatus for recording free-running oviposition rhythm in Drosophila.

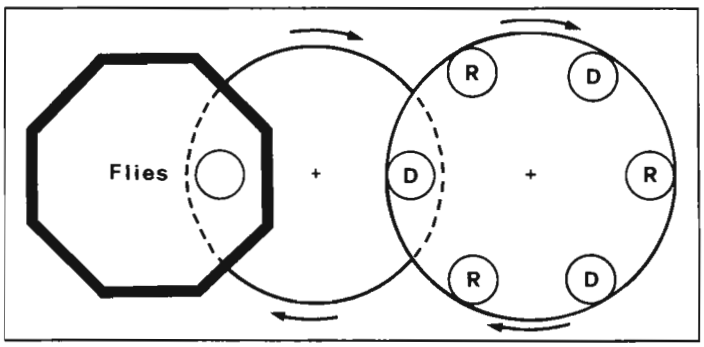
To measure the oviposition rhythm in Drosophila, several devices have already been constructed. In these apparatuses the medium on which the flies lay their eggs is moved at a constant speed so that placement of the eggs corresponds to time of deposition. This was achieved either by sliding the medium under the flies (David and Fouillet 1973) or by shifting the cage with the flies over the medium (Jungen and Locher 1970). In both kinds of apparatuses, the food-bearing plates must be changed every day and this change acts as an external synchronizer. Free-running experiments, in which conditions must be kept absolutely constant, are thus impossible with these devices.

The apparatus which is described here allows free-running experiments since no handling of the flies is needed. The principle idea is to collect the eggs on small plates which are periodically changed automatically. Every hour a cup containing fresh medium is taken from a distributor, given to the flies for one hour and then put into storage in which the order corresponds to oviposition time.

Description: (Fig. 1) The cage containing the flies (200 cm^3) is made of plastic (methylmethacrylate) which a circular hole through the floor (diameter 2 cm) adapted to the size of the cups containing the medium. The cups (C) are plastic, cylindrical plates (diameter 2 cm, height 0.5 cm, depth 0.4 cm). The surface of medium is 2 cm^2. The transferring-disc (TD) is of duraluminium and is pierced by two holes (diameter 2 cm) in order to transfer the cups. Its thickness is the same as the height of the cups (0.5 cm). The tube-holder (TH) bears six store-tubes consisting of three distributors (D) and three alternated recuperators (R). Each tube can contain 35 stacked cups, this disposition preventing the dessication of the medium.



Side-view



Top-view

Functioning: Every hour an electric pulse given by the clock causes the change of the laying plate. This change is obtained by successive linked movements which occur in the following order:

1. A distributor-tube (D) being above the transferring-disc (TD), a downward movement of the lift (L) causes the lower cup to enter a hole of the transferring disc.
2. The transferring disc turns (180°) until the fresh plate reaches the hole at the bottom of the cage. Simultaneously the plate used by the flies during the previous hour is transferred by the same movement under the tube holder (TH).
3. The tube-holder turns 60° so that a recuperator-tube (R) comes above the lift.
4. The lift which was in the lower position is moved upward and pulls up the plate into the recuperator tube. The lift then stays in the upper position.

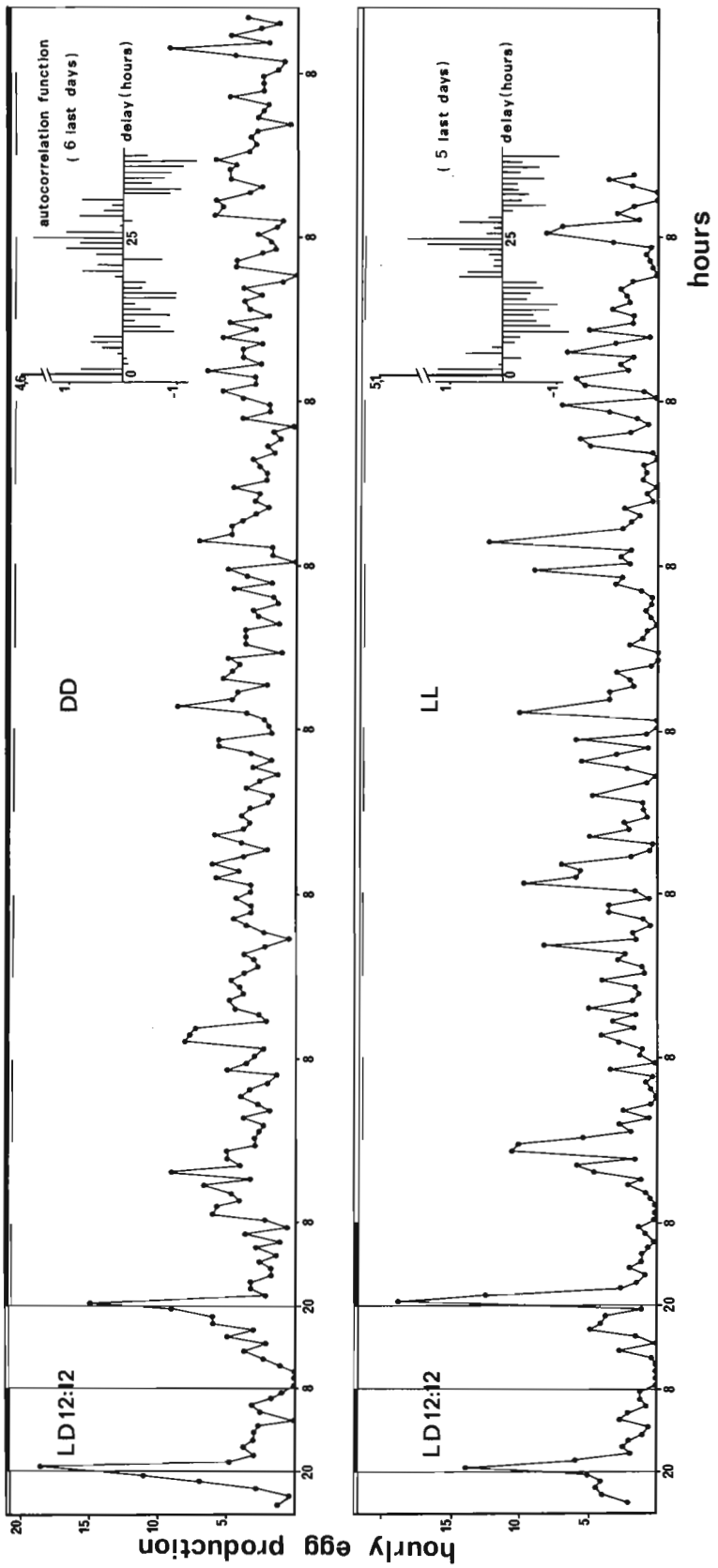


Fig. 2.

5. The tube-holder turns and stops when the next distributor tube is just above the lift (60°). The apparatus has then returned to starting position.

The onset of each movement is electrically dependent on the completion of the former one, making the apparatus more reliable. When the cup is transferred from the cage to the recuperator tube, glycerol (50%) is poured over the medium in order to avoid its dessication and to prevent embryo development. The autonomy of the apparatus is 4 days (about 100 cups) but longer experiments can be carried out since the tube-holder can be changed without disturbing the flies, even in darkness.

Results: Fig. 2 shows two examples of oviposition rhythms in D. melanogaster studied during 7 days in free-running conditions. In both cases the flies lived under a LD 12:12 photoperiod and then were transferred under constant darkness or constant light. Under LD 12:12 a peak of egg deposition occurs after the light-off (Allemand 1976a). Upon suppressing the light cycle, the oviposition pattern is modified: there are no more large peaks but only small peaks. A statistical analysis showed that in both cases a free-running rhythm remained with a period of about 25 hours (see autocorrelation functions, Fig. 2), a weak amplitude and a maximum during the virtual photophases. This oviposition rhythm in free-running conditions seems to correspond to endogenous ovarian rhythm of vitellogenesis (Allemand 1976b).

References: Allemand, R. 1976a, J. Insect Physiol. 22: 1031-1035; ____ 1976b, J. Insect Physiol. 22:1075-1080; David, J. and P. Fouillet 1973, Rev. Comp. Anim. 7:197-202; Jungen, H. and R. Locher 1970, DIS 45:201.