



Steps in the establishment of parabiosis between houseflies. A) Fly with incised mesothoracic tergum. B) 'Primary' immobilized by staples. C) 'Primary' and 'Secondary' with their thoracic openings in close apposition, held firm and immobile by a grid of staples. D) Top view of a parabiotic pair. E) Side view of a parabiotic pair.

Duration of survival of parabiotic pairs of different strains of houseflies

Experiment No.	Flies (parabionts)		No. pairs	Pairs surviving/time after union (h)				
	1	2		1	4	8	12	24
1	R ^a	R	22	22	22	22	22	18
2	S ^b	S	10	10	10	10	10	7
3	R	S	15	15	15	15	15	11

^a Resistant (U2) strain. ^b Susceptible (WHO/SRS/*Musca domestica*/1) strain.

the 'secondary' was similarly incised and placed on the immobilized primary in such a manner that the incisions of the 2 animals were opposed to each other. Once the flies had been thus placed back to back, more staples were gently pressed on the venter of the secondary, encompassing the 2 animals in a grid of staples (Figure C).

Upon the removal of the staples after about 30 min, the 2 individuals remained attached together (Figure D and E) and were not separated in spite of hectic movements of the flies. An examination of these individuals under the binocular microscope showed that they were firmly held together and no part of the incision in either was visible. No exudation of tissue or fluid could be detected in any of the several pairs examined. Manual separation of the pairs under the binocular microscope revealed that the dorsal muscles of the 2 partners had become firmly wedged to each other. This contiguity between the 2 individuals was so firm in some cases that separation caused the dislocation of a block of muscle strands of one or both individuals. Exposed dorsal muscles of a recently separated pair were freely bathed in haemolymph showing that actual parabiosis had been established – mere mechanical adhesion being ruled out as incised flies permitted free exposure to air developed a dark brown, fairly brittle scar tissue.

Establishment of parabiosis was further confirmed by demonstrating the actual exchange of haemolymph between the fly parabionts. For this, flies were paired as described and a union of 1 h allowed. One of the partners of each pair was injected with 1 µl of 0.1% Bromophenol blue solution in Insect Ringer through its pleuron. The flies were permitted to resume activity while still paired and examinations of both the injected and the corresponding uninjected partner were made at different time intervals after injection. Presence of the dye in the muscles and haemolymph could be made out as early as 5 min after the injection. By the 15th min after injection, the dye was visible in the alimentary canal of both the

injected and the uninjected parabionts. After 20 min, the distribution of the injected dye in the 2 animals was identical, leaving no doubt about the parabiotic nature of the link.

Once parabiosis had been established by these methods, the success of such operations was determined by keeping the parabiotic pairs on sucrose and water under constant observation and noting their mortality or survival at different time intervals after the operation. All those individuals which were injured during operation or which got apart as a result of their own movements within 1 h of the operation, were discarded. Pairs which did not fall apart within 1 h after the parabiotic operation would not separate from each other later. These were regarded as the successful parabionts.

Observations of such operated individuals belonging to 2 different strains of flies are given in the Table. It was noted that all successful parabionts were viable for at least 12 h after the operation, whereafter some of them died. Even so, nearly 70% of the parabionts could readily survive up to 24 h after the parabiotic operation.

These observations indicated that the technique of parabiosis described above could be most profitably employed for suitably designed short term in vivo experiments. Thus, the technique was found quite suitable for examining the production or otherwise of neurotoxins other than the insecticide in dieldrin-treated flies by using two strains showing a gross difference in sensitivity to the toxicant in question. The results of this study will be published in detail elsewhere later.

Zusammenfassung. Es wird eine neue, sehr einfache Methode zur Erzeugung parabiotischer, 12–24 h lang lebensfähiger Fliegenpaare (*Musca domestica* L.) beschrieben.

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Corrigendum

J. HANNERZ: *Discharge Properties of Motor Units in Man*, *Experientia* 29, 45 (1973). At the top of the Figure 1 the time bar is too long, so that the frequencies of the motor units seems to be twice as high as these given in the text; the time bar (100 msec) should therefore be reduced at 4 mm.