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Som, Arundhati, and B.N. Singh. Department of Zoology, Banaras Hindu University, Varanasi, India. No effect of marking flies either by nail polish on scutellum or by wing clipping on mating success in *Drosophila ananassae*.

In different species of *Drosophila*, males and females of different strains are marked for identification in mating preference tests. For marking the flies, different methods have been used by various investigators. These methods are:

- a) wing-clipping -- margin of one wing is clipped in one of the strains (Ehrman, 1966, 1968)
- b) placing a small drop of quickly drying enamel paint on mesonotum just anterior to the scutellum (Arita and Kaneshiro, 1979)
- c) placing a small drop of nail polish on scutellum (Singh and Chatterjee, 1985)
- d) placing a small mark of ink on both wings (Zouros and D'Entremont, 1980)
- e) flies had been coloured with either pink or blue fluorescent dust (Markow, 1980) and
- f) flies were fed red and green coloured food (Wu *et al.*, 1995).

In these studies, no effect of marking was found on the performance of flies or the outcome of mating preference test, because similar results have been found when the strains marked and unmarked are alternated in successive replicates.

Rare-male mating advantage which is an example of frequency-dependent selection, has so far been reported in nine species of *Drosophila* (Singh and Sisodia, 1997). *Drosophila ananassae* is a cosmopolitan and domestic species. This species occupies unique status in the whole of the genus *Drosophila* due to certain peculiarities in its genetic behaviour (Singh, 1985). Extensive work on population and behaviour genetics of *D. ananassae* has been carried out by Singh and others (for references see Singh, 1996). Rare-male mating advantage has also been reported in *D. ananassae* (Singh, and Chatterjee, 1989). It has been suggested by Bryant *et al.* (1980) that rare-male mating advantage is induced by wing-clipping in housefly and thus it is nearly an artifact resulting from alternately marking the rare and the common strains. On the other hand, Knoppien (1984) questioned the arguments given by Bryant *et al.* (1980) and proposed that any artificial rare-male mating advantage caused by wing-clipping is less important than suggested by Bryant *et al.* (1980). Further, Markow (1980) has clearly demonstrated that rare-male effect is not induced by marking with fluorescent dust in *D. melanogaster*. In view of this, we have planned experiments to test the effect of marking on rare-male mating advantage in *D. ananassae*. Further, the phenomenon of rare-male mating advantage will be investigated in detail in *D. ananassae* by employing different wild type and mutant strains and inversion karyotypes as well as different experimental techniques. Before starting the detailed experiments, we have carried out preliminary experiments to test the effect of marking on mating success in *D. ananassae* and the results are reported in this note.

A wild type laboratory stock of *D. ananassae* (Bombay strain) established from a large number of flies collected from Bombay in 1985 was used. Virgin females and males were collected from this stock and aged for seven days.

Two marking procedures were used and for each procedure "male-choice" and "female-choice" techniques were employed:

A. Nail polish marking on scutellum

In "female-choice" experiments, males were marked by placing a small drop of quick drying nail polish on scutellum. Marking was done on lightly etherized flies 24 hr before the experiment. Twenty unmarked females with 10 marked and 10 unmarked males were introduced into an Elens-Wattiaux mating chamber and thus 20 pairs of flies were tested and the sex ratio was 1:1. Flies were observed for 60 minutes. When a pair commenced mating it was aspirated out and the type of male mated was recorded. In total five replicates were run.

In "male-choice" experiments, 20 unmarked males with 10 marked and 10 unmarked females were introduced into the mating chamber and were observed for 60 minutes. When a pair commenced mating it was aspirated out and the type of female mated was recorded. In total five replicates were run.

B. Marking by clipping the margin of wing of one side

Flies were lightly etherized and a small part of the distal tip of the right wing was clipped.

In "female-choice" experiments, 20 unmarked females with 10 marked and 10 unmarked males were introduced into the mating chamber. After commencement of mating, mated pair was aspirated out and the type of male mated was recorded. Observation continued for 60 minutes. In total five replicates were run.

In "male-choice" experiments, 20 unmarked males with 10 marked and 10 unmarked females were introduced into the mating chamber and were observed for 60 minutes. When a pair commenced mating it was aspirated out and the type of female mated was recorded. In total five replicates were run.

All the experiments were conducted in a room maintained at 24°C approximately under normal light conditions from 7 to 11 a.m.

Number of matings in female- and male-choice experiments involving marked and unmarked flies are presented in Table 1. It is evident from the results that marked and unmarked males as well as marked and unmarked females are equally successful in mating as there is no significant difference in the number of matings between marked and unmarked flies in all the comparisons ($P > 0.05$). Further, marking by both the methods (placing a small

drop of nail polish on the scutellum and wing-clipping) produced identical results. Thus, it is concluded that marking the flies either by placing a drop of nail polish on scutellum or by wing-clipping has no effect on mating success in *D. ananassae*.

References: Arita, L.H., and K.Y. Kaneshiro 1979, Proc. Hawaiian Entomol. Soc. 13: 31-34; Bryant, E.H., A. Kence, and K.T. Kimball 1980, Genetics 96: 975-993; Ehrman, L., 1996, Anim. Behav. 14: 332-339; Ehrman, L., 1968, Genet. Res. 11: 135-140; Knoppien, P., 1984, Am. Nat. 123: 862-866; Markow, T.A., 1980, Behav. Genet. 10: 553-556; Markow, T.A., 1991, Evolution 45: 1525-1529; Singh, B.N., 1985, Nucleus 28: 169-176; Singh, B.N., 1996, Genetica 97: 321-329; Singh, B.N., and S. Chatterjee 1985, Can. J. Genet. Cytol. 27: 405-409; Singh, B.N., and S. Chatterjee 1989, Genet. Sel. Evol. 21: 447-455; Singh, B.N., and S. Sisodia 1997, Genetika 29: 41-48; Wu, C.-I., H. Hollocher, D.J. Begun, C.F. Aquadro, Y. Xu, and M.-L. Wu 1995, Proc. Natl. Acad. Sci. USA 92: 2519-2523; Zouros, E., and C.J. D'Entremont 1980, Evolution 34: 421-430.

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¹Depto. Genética – FMRP-USP. Av. Bandeirantes 3900 CEP 14049-900. ²Depto. Biologia – FFCLRP – USP. Av. Bandeirantes 3900 CEP 14040-901, Brasil. A chamber to record the courtship sound in *Drosophila*.

courtship sound an important character for behavioral, evolutionary and molecular studies. In order to obtain the sound pattern, it is necessary to record it with appropriate equipment, to observe courtship behavior, and to eliminate background noise such as female wing vibration and flies movement. We erected a mating chamber that can be easily handled and allows the observation of the fly couple. The chamber, presented in Figure 1, is composed by an acrylic block 3×3×1 cm. In the central part of its inferior base there is a concave chamber with a diameter of 1.5 cm and a height of 1.7 cm in its deepest part. This interior opening is sealed by a nylon nest allowing the sound produced during courtship to reach the microphone. In its superior face there is an inclined opening 0.6 cm wide which falls laterally in the enclosed space of the chamber. After the introduction of the flies through this opening in the chamber, we close it by plugging it with a cap. Because of its characteristics, this chamber is easily installed, fitting nicely on the exposed membrane of the microphone. In addition, it allows the flies movement in its interior and the observation of their behavior during courtship.

References: Manfrin *et al.*, 1997, Rev. Bras. Biol. 57:349-355; Miller, *et al.*, 1975, Evolution 29:531-544; Spieth, H.T., 1974, Ann. Rev. Entomol. 19:385-405.

Table 1. Number of matings in female- and male-choice experiments involving marked and unmarked flies

Replicates	Female-choice experiment			Male-choice experiment		
	Unmarked	Marked	Total	Unmarked	Marked	Total
A. Nail polish marking on scutellum						
1	11	6	17	10	9	19
2	9	7	16	10	10	20
3	9	10	19	7	9	16
4	9	8	17	10	7	17
5	9	5	14	8	7	15
Total	47	36	83	45	42	87
χ^2 for 1:1 ratio	1.44	P > 0.05		.10		P > 0.05
B. Wing Clipping						
1	8	6	14	8	9	17
2	8	6	14	9	8	17
3	9	8	17	8	8	16
4	7	6	13	8	9	17
5	8	9	17	9	9	18
Total	40	35	75	42	43	85
χ^2 for 1:1 ratio	0.33	P > 0.05		0.01		P > 0.05

In the genus *Drosophila* the most conspicuous element of the male's courtship is the wing vibration which emits an acoustic stimulus. This stimulus is species specific being a character that has been used to identify species in the genus (Spieth, 1974), and there is variation among populations and subspecies (Miller *et al.*, 1975; Manfrin *et al.*, 1997). This makes the