VARIATION AMONG *DROSOPHILA MOJAVENSIS* POPULATIONS FOR LOCOMOTOR ACTIVITY DOES NOT CAUSE SEXUAL ISOLATION

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Received 20 June 1991, 23 August 1991

ABSTRACT Locomotor activity, mating success and mating speed were measured for males and females of six strains of *Drosophila mojavensis* from four geographically separated regions. Variation for ontogenic changes in locomotor activity, and correlations between activity and courtship components were measured to test the importance of locomotor activity to courtship success and the mating isolation which has been well documented for some *D. mojavensis* populations. Variation among strains was found for mean activity and for ontogenetic changes in males. Females of all strains declined in activity as they matured suggesting an age related behavioral switch from mate or host plant location to oviposition behaviors. Activity was not correlated with body size in either sex, but female activity was negatively correlated with courtship latency. Most importantly, there was no evidence that indicated locomotor activity differences among strains could mediate the asymmetrical mating isolation present between flies from mainland Sonora and the Baja peninsula.

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INTRODUCTION

Locomotor activity of adult *Drosophila* is assumed to affect dispersal, predation, and the search for food, oviposition sites, and mates (Burnet et al., 1988). Genetic variation among individuals and among populations has been established in *D. melanogaster* (reviewed in Costa et al., 1989), but locomotor activity has not been studied in other *Drosophila* species.

Populations of *D. mojavensis*, a cactophilic fly species in the mulleri complex of the Repleta group, utilize a number of cacti over a wide geographic range in south-western North America (Heed and Mangan, 1986). These environmental differences may contribute to variation in locomotor activity among races, and Brazner (1983) has suggested that it may also affect the asymmetrical behavioral isolation observed between flies from Sonora, Mexico and Arizona, and those from Baja California, Mexico (Koepfer, 1987a, b; Krebs and Markow, 1989). Cobb *et al.* (1990) have also suggested that locomotor activity differences may cause sexual isolation, but I have found no empirical evidence supporting or refuting this hypothesis.

In the present study, locomotor activity, size, and mating success of all geographic host races of *D. mojavensis* were compared as they matured. The populations used were two from the mainland Sonoran Desert that breed on organ pipe cactus (*Stenocereus thurberi*), two from the Baja Peninsula that breed on agria cactus (*S. gummosus*), one from Santa Catalina Island that breeds in prickly pear cactus (*Opuntia*), and one from the Mojave Desert that breeds in barrel cactus (*Ferrocactus*). These other races, while of potential significance because of their geographic isolation and shifts in host plant, have received surprisingly little attention in the overall research effort focused on *D. mojavensis*.

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Evolutionary Theory 10: 101-107 (December, 1991)
The editors thank Lee Ehrman and another referee for help in evaluating this paper.

MATERIALS AND METHODS

Strains and rearing conditions: Strains of *D. mojavensis* were obtained from the University of Arizona (with A stock numbers). For ease of reference, strains were denoted by abbreviations for geographic area: the Sonora desert mainland (S1 for A900, Santa Rosa Mountains, AZ and S2 for A891, Las Bocas, Sinaloa), the Baja Peninsula (B1, for A920, San Lucas and B2 for A859, Punta Prieta), Santa Catalina Island (SCI for A826), and southern California (CAL for A753, Vallecito Stage Station).

All flies were reared in half-pint milk bottles containing cornmeal-molasses-yeast medium with buffered propionic acid and seeded with live yeast. Strains were maintained in eight bottles with a minimum 50 pairs per bottle. Virgin males and females were separated under light ether anesthesia 1-6 hours after eclosion and stored in food vials, 2cm diameter and 5cm high, with live yeast, five flies/vial. The flies were kept in a 13:11 hour light:dark cycle with temperatures at 26-27°C day and 20-21°C night.

Locomotor activity (Ewing, 1963) was measured beginning one half hour after "lights on" which corresponds to the major daily mating peak in nature (Krebs and Bean, 1991). Single flies were introduced into a 30 cm long loop of clear tubing with inside diameter about 0.75 cm (Figure 1). The tubes allowed continous movement without edges and did not restrict movement other than flight. Flies were allowed to acclimate for 100s and were then measured for the number of lines crossed (48 lines spaced over 30 cm) during the next 100s (Burnet and Connolly, 1974) to reduce reactivity, or the response to being placed in the tube. No differentiation was made, however, between amount and speed of movement (Burnet *et al.*, 1988).

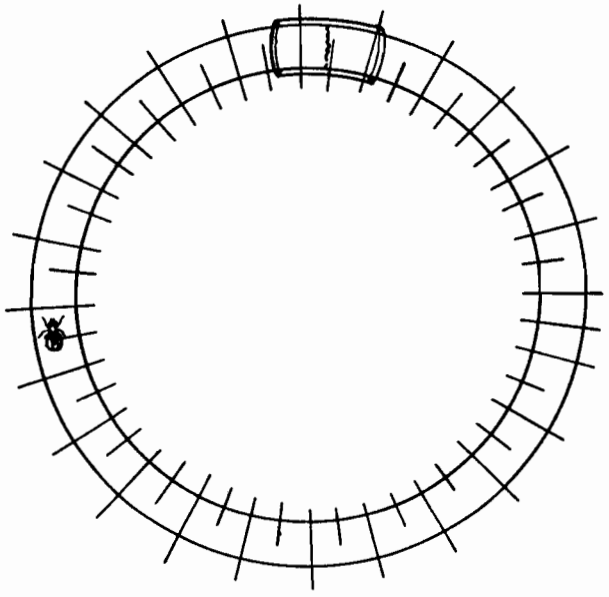

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Figure 1. A representation of the device for measuring locomotor activity in *Drosophila mojavensis* (after S. Crossley, pers. comm.). The clear tube is 30 cm long and is divided by 48 lines.

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Table 1. Activity in mean lines crossed of seven day old males in activity tubes conditioned by previous presence of flies (x+se).

Male Strain	Conditioning Treatment Males	S1 Females	B1 Females
S1 (n=17)	19.4 ± 4.8	20.6 ± 6.0	19.9 ± 5.9
B1 (n=20)	28.1 ± 3.8	25.5 ± 3.3	27.0 ± 4.2

Drosophila mojavensis is not sexually mature until seven days after eclosion for males and three days after eclosion for females (Markow, 1982). Activity was therefore measured for males at 1, 3, 5, and 7 days after eclosion and for females at 1, 3, and 5 days. Flies were transferred individually to food vials after each test. On the eighth day for males and sixth for females, each fly was paired with one mature virgin fly of the opposite sex of the same strain in a fresh food vial for 20 minutes. The number of seconds until the onset of courtship, female acceptance [indicated by wing spreading (Markow, 1981)], and the time until successful copulation (not all acceptances lead to copulation) were recorded. Following the mating experiments, thorax lengths of all flies were measured with an ocular micrometer.

Because locomotor activity was measured for isolated males but courtship latency was measured for a male paired with a female, a test was conducted to verify that locomotor activity, as measured, was unaffected by odor or volatile pheromones a female *D. mojavensis* may produce. In the experimental group, four six-day-old virgin females were placed in the activity tubes for five minutes. These females were gently tapped out to minimize air removal prior to introducing the males. Control tubes had males similarly placed within them. Activity of seven-day-old males (mean lines crossed in the activity chamber) was unaffected by prior female presence (Table 1).

RESULTS

Locomotor activity differences among strains and ontogenetic changes are shown in Figure 2. Strain and age, the main effects of the two-way ANOVA for activity (GLM procedure with main effects fixed, SAS, 1985), were both significant for females, but males differed only among strains. Activity declined significantly with female age ($F = 39.93_{[2,392]}$, $P < 0.0001$) with strain differences persisting over time (strain x age, ns). For males, the changes in activity with age differed among strains (interaction $F = 1.97_{[15,499]}$, $P = 0.016$). Strains varied significantly in activity when males were sexually immature, but activity of males from initially "slow" strains increased with age while activity in "fast" strains declined. Therefore, at sexual maturity (seven days of age), no differences remained among males from different strains ($F = 2.11_{[5,163]}$, ns).

Within any strain, no significant correlations were found between activity and thorax length (Table 2a). Combining flies across localities also failed to reveal any significant activity/size relationship. Activity of females was correlated negatively with courtship latency in one strain and in all strains combined, but greater activity in males was not related to how quickly they initiated courtship (Table 2b). No correlations were found between activity and

Table 2. Correlations of activity with size (Pearson) and courtship latency (Spearman rank) in *Drosophila mojavensis* for seven day old males and five day old females.

Strain	S1	S2	B1	B2	SCI	CAL	Total
A. SIZE							
Males	0.05	-0.25	0.02	0.31	-0.08	-0.26	-0.10
	p=.82	p=.33	p=.89	p=.07	p=.72	p=.37	p=.20
	(27)	(17)	(35)	(25)	(23)	(14)	(141)
Females	0.04	0.36	0.11	-0.09	0.06	-0.15	0.01
	p=.83	p=.06	p=.56	p=.63	p=.74	p=.40	p=.85
	(31)	(29)	(33)	(30)	(33)	(32)	(188)
B. COURTSHIP LATENCY							
Males	0.12	-0.07	-0.07	-0.27	0.12	0.25	0.01
	p=.54	p=.78	p=.48	p=.18	p=.60	p=.51	p=.90
	(27)	(17)	(34)	(24)	(22)	(13)	(137)
Females	-.036	-0.28	-0.20	-0.04	0.04	-0.22	-0.22
	p=.05	p=.06	p=.34	p=.86	p=.84	p=.32	p=.01
	(30)	(37)	(23)	(22)	(30)	(20)	(162)

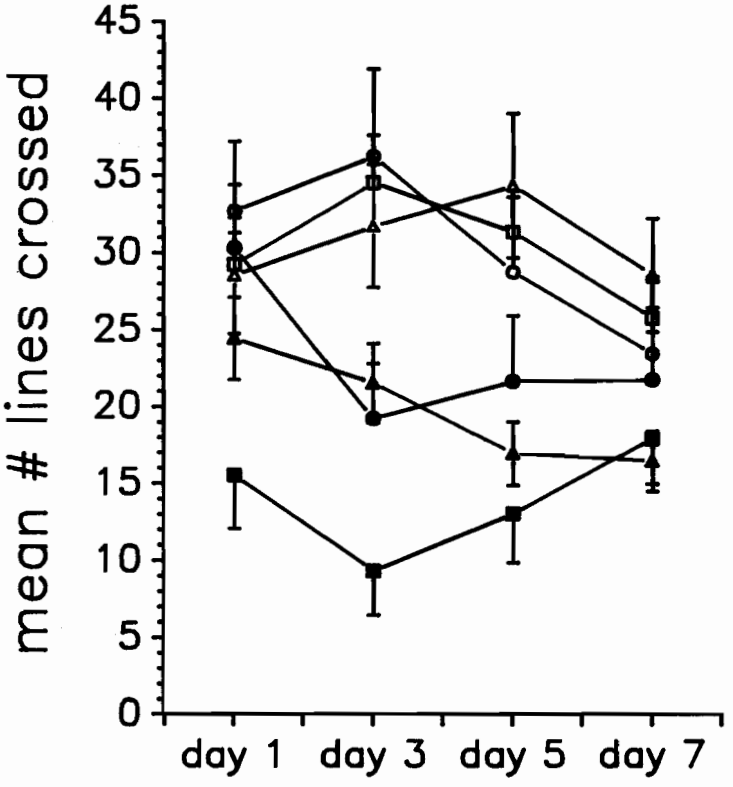
other courtship factors (receptivity, copulation) in either males or females of any strain or of all strains combined (not shown). Interestingly, neither male nor female size was correlated with any mating behavior measured in any strain.

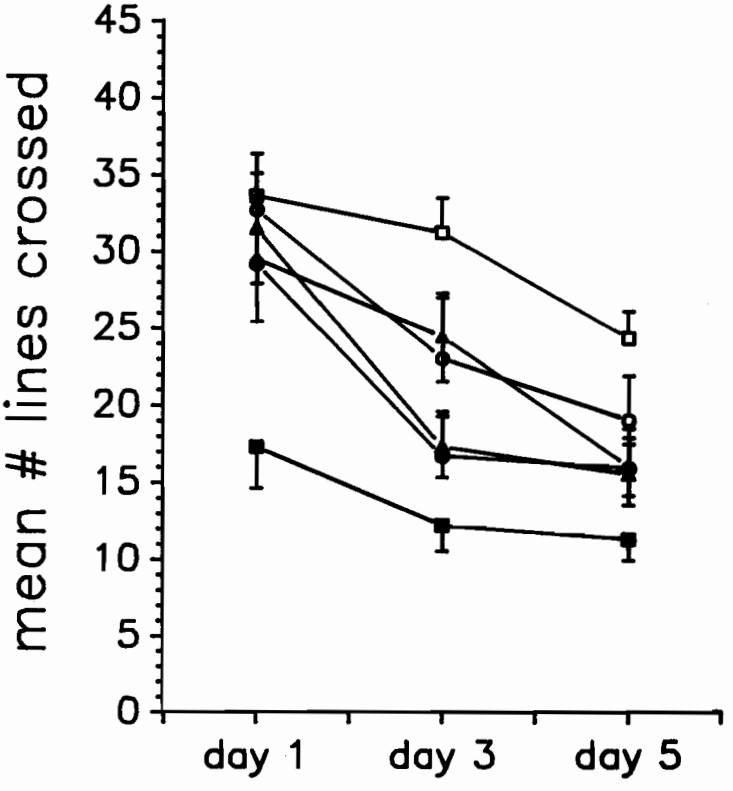
DISCUSSION

Activity differences among strains, as well as differences in ontogenic changes occuring with sexual maturity are present among *D. mojavensis* populations. The hypothesis that locomotor activity can cause sexual isolation was not supported because the strains showing sexual isolation, Sonora and Baja (Krebs and Markow, 1989), were not different in locomotor activity. Fast (S2 and B1) and slow (S1 and B2) strains were present for both populations. Additionally, flies from the least and most active strains overall, from Southern California and Santa Catalina Island respectively, show no sexual isolation from either Sonora or Baja flies (T. A. Markow, pers. comm.).

Locomotor activity differences during adult maturation may reflect race and gender differences for dispersal times in nature. Age and sex differences in dispersal are as yet unknown, yet males take at least twice as long as females to become sexually mature (Markow, 1982). That females of all strains become less active at sexual maturity may be due to a switch from searching behaviors, either

Figure 2. Locomotor activity of *Drosophila mojavensis* males (measured on four days as they matured) and females (measured on day 1, day 3 [maturity], and day 5: Sonora (circles), S1 (closed) and S2 (opened); Baja (triangles), B1 (opened) and B2 (closed); CAL (closed squares) and SCI (opened squares). Bars are ± 1SE.


mean # lines crossed
45
40
35
30
25
20
15
10
5
0
day 1
day 3
day 5
day 7


mean # lines crossed
45
40
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10
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0
day 1
day 3
day 5

for host plants or mates, to oviposition behaviors. Males of some strains increase their activity during maturation, while activity of males from other strains decreases. These activity differences may reflect on such factors as differences in population structure or host plant availability.

The cactophilic *D. mojavensis* differs from the cosmopolitan *D. melanogaster* with respect to associations among activity, size, and male mating success (Cobb et al., 1987), differences that reflect the natural mating ecology of these species. In *D. melanogaster*, males court females at the site where matings take place (Markow, 1988). Males continually move around to out-maneuver each other for positions behind females. In *D. mojavensis*, males position themselves at non-resourse based territories and wait, usually motionless, for females to arrive (Markow and Toolson, 1990). Male activity is therefore not as large a component of the mating system as in *D. melanogaster*. Female movement, on the other hand, is correlated with shorter courtship latencies in both *D. melanogaster* and *D. mojavensis*, and in nature, female movement triggers courtship in both species (Cobb *et al.*, 1987; Krebs and Bean, 1991).

Activity at sexual maturity is therefore a function of the mating system of a species which probably varies under differing enviromental conditions. Activity is important to courtship initiation, but not mating success either within or between populations. The pattern of variation in locomotor activity is also inconsistent with its having an important role in the sexual isolation observed among some of the races of *D. mojavensis*.

ACKNOWLEDGMENTS

This work was supported by NSF grants BSR 8600105 and BSR 8708531 to T. A. Markow, who also provided many suggestions necessary to the completion of this report. I thank William Heed and William Etges for strains of flies, and Lee Ehrman for comments on the manuscript.

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