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## Behavioral Facilitation of Reproduction in Sexual and Parthenogenetic *Drosophila*

Abstract. *In a normal bisexual laboratory strain of* Drosophila mercatorum, *females housed with either fertile or sterile males lay more eggs than do females housed in pairs or as isolates. Females of a derived parthenogenetic strain have suffered genetic loss of this behavioral facilitation of egg production, a loss comparable to the loss of sexual receptivity. Despite these losses there has been a large increase in fecundity in the parthenogenetic strain. These findings are compared with those in a parthenogenetic lizard.*

Morphological, physiological, and behavioral components of reproduction coevolve under different selection pressures. In vertebrates, the presence and behavior of the male facilitates ovarian growth in the conspecific female. In the green anole lizard, *Anolis carolinensis*, courtship behavior of the male is critical for normal pituitary gonadotropin secretion in the female (*1*). Behavioral facilitation of reproduction is also seen in unisexual lizards. The whiptail lizard, *Cnemidophorus uniparens*, reproduces parthenogenetically and consists only of female individuals. Individuals perform both complementary male-like and femalelike displays similar to the displays seen during courtship and copulation in closely related sexual species (*2*). The presence and behavior of conspecifics have been experimentally shown to facilitate reproduction in this unisexual lizard (*3*).

We studied the effect of different "social" conditions on reproduction in both sexual and parthenogenetic strains of *Drosophila mercatorum*. We present evidence for a measurable behavioral facilitation of egg-laying in sexual *D. mercatorum* and show that females of this strain of parthenogenetic flies have lost the behavioral facilitation of oviposition characteristic of their bisexual ancestor. There is, however, an increased oviposition rate that does not depend on social factors.

Individual virgin females of a sexual strain of *D. mercatorum*, denoted S (H62.60 LaPalma, El Salvador), and a parthenogenetic strain derived from it by artificial selection in 1961 (S-1) were separately exposed to one of the following social conditions (*4*): housing in isolation, housing with another female, housing with one sterile (XO) male, housing with two sterile males, housing with one fertile (XY) male, and housing with two fertile males. Eggs produced by each female were counted on a daily basis for four consecutive days (*5*).

In the sexual strain, virgin females housed with males laid the most eggs ($\chi^2(9) = 28.01$, $P < 0.001$); females housed with males produced 13 times more eggs than isolated females and 6 times more than females housed in pairs (Table 1). Although isolated virgin females had fully developed ovaries, they deposited eggs only sporadically (*6*). Females housed together laid more eggs per female than isolated females ($\chi^2(7) = 47.71$, $P < 0.01$) (Fig. 1). Courtship behaviors, not copulatory stimuli, from the male may be responsible for this facilitation of reproduction, but this is not yet known. Analysis with 2 by 2 contingency tables shows that egg production in females with sperm in the seminal receptacle was not statistically different from that of unfertilized females with males ($t(18) = 0.64$, $P > 0.05$) (*7*). Further, there was no statistically significant difference among females housed with sterile males compared with fertile males ($t(38) = 0.25$, $P > 0.05$) (*8*). The average daily egg production of females housed with two males, fertile or sterile, was less than that of females housed with one male (44.65 ± 2.19 and 53.55 ± 2.92, respectively; $t(38) = 2.44$, $P < 0.02$). Aggression between males may inhibit ovarian activity in the female, as has been shown with the green anole lizard (*9*).

There was no effect of social condition on egg production in the parthenogenetic strain ($F(5, 231) = 0.19$, $P > 0.05$) (Table 1 and Fig. 1). Parthenogenetic female isolates laid approximately 15 times more eggs than sexual female isolates (mean, 55.9 eggs per day compared with 3.8 eggs). Mean egg production by females of the parthenogenetic strain S-1 when first reported in 1962, 6 months after the origin of the strain, was 6.1 eggs per day; about 1 year later it was 10.4 eggs per day (*10*). Thus, the current rate is more than five times greater than the rate observed in 1963. This is a significant change in egg production, presumably resulting from natural selection for fecundity in the stock over the 23 years of its existence. This change is especially noteworthy in view of the fact that the parthenogenetic rate of the S-1 stock (number of impaternate females per un-

about 2 percent since the strain was established (*11*). Accordingly, the S-1 stock has increased its egg production but suffered a loss in two behavioral traits, facilitation of egg production (our data) and sexual receptivity (*12*).

Carson *et al.* (*12*) have observed that individuals of the S-1 parthenogenetic strain of *D. mercatorum* are less likely to mate with males than are females of the sexual strain from which it arose. This loss of sexual receptivity was interpreted as evidence for the disappearance of a genetic trait (sexual receptivity) due to the absence of a selective force (the need for males for reproduction). The difference between parthenogenetic whiptail lizards, which continue to exhibit both sexual behavior and behavioral facilitation of reproduction, and parthenogenetic *D. mercatorum* may lie in the nature of the chromosomal origin of the respective species. *Cnemidophorus uniparens* is a triploid and arose from the hybridization of two closely related sexual species (*13*). The parthenogenetic *D. mercatorum* stock, on the other hand, arose from isolated virgin females of a single bisexual strain (*10*). It may be significant that in the parthenogenetic whiptail lizard, the offspring of sexual interaction, the neuroendocrine mechanisms subserving the behavioral aspects of reproduction have been retained, whereas in *D. mercatorum*, the offspring of virgin females, the comparable neuroendocrine mechanisms have been lost. Alternatively, the mode of parthenogenesis may be critical for the heritability of neuroendocrine controlling mechanisms. In *C. uniparens*, triploidy is restored by endoduplication prior to meiosis, resulting in fixed heterozygosity (*14*), whereas in *D. mercatorum*, diploidy is restored largely by duplication of single pronuclei, resulting in completely homozygous clones (*15*).

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Table 1. Behavioral facilitation of reproduction in sexual and parthenogenetic strains of *Drosophila mercatorum*. Each experimental group consisted of ten vials (*4*). Eggs were removed and counted on four consecutive days (*5*).

| Housing | Females ovipositing | Days on which eggs found (%) | Females fertilized | Eggs produced in 4 days |
|---|---|---|---|---|
| | | *Sexual strain* | | |
| Isolation | 6 | 22 | 0 | 150 |
| 2 females | 8 | 45 | 0 | 750 (375)* |
| 1 XO male | 10 | 88 | 0 | 2199 |
| 2 XO males | 10 | 83 | 0 | 1764 |
| 1 XY male | 10 | 92 | 1 | 2113 |
| 2 XY males | 10 | 88 | 3 | 1803 |
| | | *Parthenogenic strain* | | |
| Isolation | 10 | 92 | 0 | 2236 |
| 2 females | 10 | 100 | 0 | 4186 (2093)* |
| 1 XO male | 10 | 100 | 0 | 2029 |
| 2 XO males | 10 | 100 | 0 | 2045 |
| 1 XY male | 10 | 90 | 2 | 2053 |
| 2 XY males | 10 | 90 | 3 | 2187 |

*The numbers in parentheses represent egg production per female with both females contributing equally.

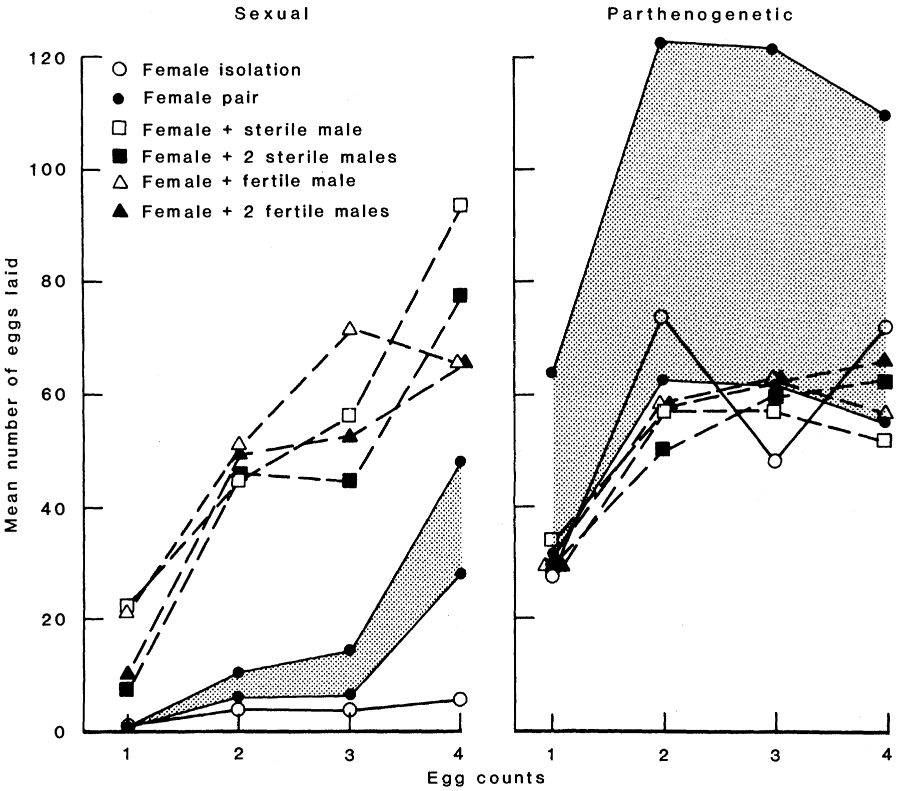

Fig. 1. Egg production over four consecutive days by sexual and parthenogenetic female *Drosophila mercatorum* in various social conditions. It was not possible to determine the extent to which each female contributed to the total egg count in the female pair category. Therefore both the total egg count (the maximum each female could lay) and one-half that amount (if both females were contributing equally) are shown (shaded area).

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4. The stocks were maintained on standard cornmeal medium. Flies were taken from stock bottles on the day of eclosion. Each female was housed in a separate vial until the start of the experiment; males were caged in groups. All females were 4 to 5 days old and all males were 14 days or older. Females for egg counts were housed in plastic vials (102 by 33 m) stoppered with cotton. Each vial contained a plastic cap (volume 5.5 ml) filled with food. This food cap served as an oviposition site. All vials were kept in a room with abnormal diurnal photoperiod and maintained at 22° ± 1°C.
5. The food cap containing the eggs was removed from each vial on each of four consecutive days beginning the day after the establishment of the groups. Food caps were replaced and the eggs were counted.
6. Because of the large number of days during which isolated and paired females of the sexual strain failed to lay eggs, it was necessary to analyze these groups by nonparametric statistical methods. Egg laying among sexual females in the other social groups and in all of the parthenogenetic females was normally distributed and was analyzed by two-way analysis of variance and *t*-tests.
7. At the end of the experiment all females were dissected and the seminal receptacle examined for sperm.
8. Sterile males lacking a Y chromosome (XO) occasionally appear in the parthenogenetic stocks of *D. mercatorum* (*11*). Although S-1 itself does not produce any, 60 XO males were found among 15,558 impaternate progeny from virgins isolated from two bisexual bridge stocks [H. L. Carson and S. H. Snyder, *Egypt. J. Gen. Cytol.* **1**, 256 (1972)] related to S-1 (S-1-Bridge 67 and S-1-Bridge 88).
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11. The eggs of five parthenogenetic females from each of the social conditions (except those housed with fertile males) were allowed to develop through pupation to eclosion. The rate of parthenogenesis is then expressed as a percentage by dividing the number of impaternate females produced ($n = 98$) by the number of unfertilized eggs counted ($n = 4842$). This gives about 2 percent, not significantly different from the rate measured in 1963 (*10*).
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