

## IS THERE ETHOLOGICAL ISOLATION AMONG *DROSOPHILA MELANOGASTER* STRAINS REARED FOR MORE THAN 35 GENERATIONS ON DIFFERENT FOOD?

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**Abstract** — Ethological isolation among three strains of *D. melanogaster* reared for more than 35 generations on different food (standard cornmeal-agar-yeast medium, banana, and tomato) was tested in a series of multiple choice tests. Long-term maintenance on different substrates did not contribute to sexual isolation among three "nutritional strains", as degrees of ethological isolation (Malogolowkin-Cohen's isolation indices) were not statistically significant. Males and females reared on different nutritional resources were equally successful in matings, i.e., significant differences in the number of copulations achieved by males and females from various lines were not observed.

**Key words:** *Drosophila melanogaster*, nutrition, ethological isolation, mating success, homogamic matings, heterogamic matings, multiple choice

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### INTRODUCTION

Courtship in *Drosophila melanogaster* is a very complex process consisting of different behavioral displays performed by both sexes, followed by an interchange of different sensory stimuli (visual, acoustic, olfactory, and tactile) (Spieth and Ringo, 1983; Hall, 1994; Yamamoto et al., 1997). This process precedes the sexual act and is very important in mate attraction and mate selection, as well as in ethological isolation, since stimuli used in attraction of the mating partners are also involved in behavioral identification and species recognition. Both sexual selection and sexual isolation have been studied in many *Drosophila* species (Spieth and Ringo, 1983; Partridge et al., 1987; Markow, 2002; Singh et al., 2002; Pavković-Lučić and Kekić, 2007).

Isolation mechanisms are usually divided into two groups (Mayr, 1963; McFarland, 2006): pre-mating (those that prevent interbreeding, including ecological, temporal, ethological, and mechanical isolation) and post-mating (ones that prevent success of interspecific crosses, including hybrid inviability and hybrid sterility). Reproductive isolation has been extensively studied in the genus

*Drosophila*: the results obtained in different species have shown that complete isolation exists between some species and incomplete between others (Singh and Chatterjee, 1992; Massie and Markow, 2005; Chang and Tai, 2007). Behavioral patterns by which *Drosophila* species attract mates are highly distinctive for each species and they are often very different in closely related species for whom the potential risk of hybrid mating is greatest. For example, *D. melanogaster* and *D. simulans*, which are sibling species, significantly differ in both courtship song parameters (Kawanishi and Watanabe, 1980) and courtship odor profiles (Coyne, 1996). Besides species-specific attraction, there may also exist within a species preferences for some mates rather than for others. *Drosophila* females are able to discriminate between different mutants, between the bearers of different chromosomal rearrangements, or between members of different geographic races (Ehrman, 1975 and references therein).

Ethological (sexual or behavioral) isolation constitutes one of the most important classes of reproductive isolating mechanisms in animals and is defined as the deviation from random mating in

mated individuals (Gilbert and Starmer, 1985). This type of isolation implies the tendency of individuals to avoid mating with those of another strain, race, or species, as a result of differences in courtship behavior (McFarland, 2006). Behavioral isolation is very effective, as it prevents the wastage of gametes and of food and space for developing inferior or sterile hybrids (Ehrman, 1975).

Sexual isolation in *Drosophila* species is controlled by a polygenic system (Ehrman, 1961), where many genes with a role in determination of important phenotypic traits (like color or morphology), when mutated, may be involved in changes of mating behavior (Koref-Santibañez and Waddington, 1958; Crossley, 1974; Stanić and Pavković-Lučić, 2005). By developing molecular techniques, many QTLs involved in *Drosophila* ethological isolation were mapped (Doi et al., 2001; Moehring et al., 2004). Besides genetic background, behavioral isolation in *Drosophila* can be modified by different factors. These are: temperature, density and overcrowding, previous mating experience, age, and female receptivity (Spieth and Ringo, 1983; Carracedo et al., 1987).

It is well known that the process of adaptation to different environments can cause reproductive isolation (Etges, 1990). Genetic differentiation caused by adaptation to new environments may cause sexual isolation because those genes involved in adaptation may influence sexual isolation (Rice, 1987; Etges, 1990). In this study, we tested whether long-term maintenance of flies on different food can cause ethological isolation between different lines within a species. It was supposed that stock manipulations (long-term different diets) may alter the genetic background of the stock, i.e., that genes which adapt individuals to particular food resources may be indirectly involved in mate choice. Different nutritional regimes might be linked in this way with the chemical basis of courtship, and females would be capable of distinguishing among males from different laboratory stocks.

## MATERIALS AND METHODS

Fly strains: *Drosophila melanogaster* flies were main-

tained for more than 35 generations ( $F_{36}$ - $F_{39}$ ) on three substrates: standard laboratory food (cornmeal-agar-yeast medium, strain **C**), banana (strain **B**), and tomato (strain **T**). The banana and tomato substrates represent modified standard laboratory food (Kekić and Pavković-Lučić, 2003). Flies were maintained in 250-ml glass bottles without competition (about 100 individuals per bottle) under conditions of 25°C, relative humidity of 60%, and 12 h: 12 h light: dark cycles.

Flies used in the experiment were sexed without anesthesia every few hours after eclosion, as it may affect mating success (Joakim and Curtsinger, 1990). Virgin flies were kept separately according to sex and strain for 3-5 days in food vials until they were used.

Experimental procedure: Ethological isolation and mating success were measured in multiple choice experiments. The multiple choice method counts the instances of four types of matings when males and females of two strains are placed together, which is diagrammed in Fig. 1. All experiments were conducted in the morning (7.00 - 11.00 a.m.), at which time flies showed the highest mating activity, at room temperature (24 - 25°C). Three experimental groups were formed: MC 1 (multiple choice 1), MC 2 (multiple choice 2), and MC 3 (multiple choice 3). Flies were crossed as follows:

MC 1: 10 females (C) + 10 females (B) + 10 males (C) + 10 males (B)

MC 2: 10 females (C) + 10 females (T) + 10 males (C) + 10 males (T)

MC 3: 10 females (B) + 10 females (T) + 10 males (B) + 10 males (T)

Capital letters designate strains involved in the crosses, i.e., C = "cornmeal-agar-yeast" strain, B = "banana" strain, and T = "tomato" strain.

Ten trials were run for each experimental set, which means that 400 flies were tested per experimental group, i.e., a total of 1200 flies participated.

One trial (replica) was made in this way: virgin males and females were placed in adequate propor-

		females		Total
		1	2	
males	1	X <sub>11</sub>	X <sub>12</sub>	n <sub>1</sub>
	2	X <sub>21</sub>	X <sub>22</sub>	n <sub>2</sub>
Total		n <sub>3</sub>	n <sub>4</sub>	N

**Fig. 1.** Multiple choice experimental design. Abbreviations: 1 – strain 1; 2 – strain 2; X<sub>11</sub>, X<sub>22</sub> – number of homogamic matings; X<sub>12</sub>, X<sub>21</sub> – number of heterogamic matings; N – total number of achieved copulations.

tions in mating vials. At 24 h before testing, flies of alternative types (males and females from different "nutritional" strains) were marked by UV dust (red and green). Ten females of each of two strains were introduced to the mating vial with 10 males of each of two strains and observed for 60 min. Copulating pairs were gently aspirated, and the male and female types were recorded under binoculars using a UV lamp. Later, all flies (mating pairs, as well as non-mates) were put into separate eppendorphs filled with 70% ethanol for further morphological analyses.

Statistical analyses: Ethological isolation among strains would be observed if homogamic matings (within members of the same strain) significantly exceed heterogamic matings (between flies from different strains), which is statistically confirmed by various isolation indices (Spieth and Ringo, 1983; Gilbert and Starmer, 1985).

The data were analyzed in two ways. Chi-square tests were used to detect deviations from random mating. The degrees of ethological isolation among strains were calculated as the joint isolation index (*I*). The joint isolation index (*I*) and standard error of *I* were calculated according to formulas proposed by Malogolowkin-Cohen (Malogolowkin-Cohen et al., 1965). The joint isolation index was calculated as:  $I = [(X_{11} + X_{22}) - (X_{21} + X_{12})]/N$ . This index calculates the number of homogamic (i.e., of the same strain) minus heterogamic (i.e., between strains) matings and divides it by the total number of matings (N), giving positive values in the case of an

excess of homogamic matings and negative values in the opposite case. If *I* = 0, then random matings occur.

Male (*Z<sub>m</sub>*) and female (*Z<sub>f</sub>*) selection indices were calculated according to formulas given by Levine (1949):

$$Z_m = \sqrt{\frac{(X_{11} \cdot X_{12})}{(X_{21} \cdot X_{22})}} \quad \text{and} \quad Z_f = \sqrt{\frac{(X_{11} \cdot X_{21})}{(X_{12} \cdot X_{22})}}$$

The selection indices vary from infinity to zero; values of the index higher than one show an advantage for strain 1, i.e., if *Z<sub>m</sub>* (*Z<sub>f</sub>*) > 1, then individuals that belong to strain 1 are more successful in matings than individuals from strain 2, and when *Z<sub>m</sub>* (*Z<sub>f</sub>*) < 1, the more successful in matings are individuals that belong to strain 2.

## RESULTS

Results of the multiple choice tests are summarized in Table 1. Table 1 (a-c) records the actual number of homogamic vs. heterogamic copulations observed during a 60-min interval. In the MC 1 experimental group, out of a possible 200 matings, 144 copulations (or 72%) were achieved (Table 1a); 156 copulations (78%) were observed in the MC 2 group (Table 1b) and 152 (76%) in the MC 3 group (Table 1c). The results presented in Table 1 (a-c) were used in estimating the degrees of ethological isolation among nutritional strains (Table 2).

All multiple choice tests (MC 1, MC 2, and MC 3) yielded non-significant results, i.e., no evidence of discrimination and choice was found, which means that matings occurred randomly. The Chi-square value was calculated in all experimental groups in order to determine the difference between homogamic and heterogamic matings under the assumption of random mating. These values indicated non-significant deviations from random mating in all experimental groups (Table 2). A slight excess of homogamic over heterogamic matings was detected, i.e., the number of homogamic matings was larger than heterogamic in all experimental groups (Table 1), although these differences were not significant (Table 2).

**Table 1.** Number of copulations achieved in multiple choice tests (a-c). Abbreviations: C – strain C, B – strain B; T – strain T.

a) “cornmeal” x “banana” (MC 1).

		females		Total
		C	B	
males	C	48	32	80
	B	30	34	64
Total		78	66	144

b) “cornmeal” x “tomato” (MC 2).

		females		Total
		C	T	
males	C	46	38	84
	T	30	42	72
Total		76	80	156

c) “banana” x “tomato” (MC 3).

		females		Total
		B	T	
males	B	41	32	73
	T	33	46	79
Total		74	78	152

Isolation estimates were calculated for each experimental group separately. The degree of ethological isolation among three nutritional strains (the joint isolation index) was non-significant: in all experimental groups, random mating (no preferences, no sexual isolation) resulted in a coefficient of zero (Table 2).

The relative mating success of two types of males and females was also compared. Neither of the two competing types was significantly more successful in

achieving copulations than the other type, i.e., neither different “nutritional strain” was significantly more successful in achieving copulations than the strain with which it was compared (Table 3).

## DISCUSSION

Different *Drosophila* species that occupy similar feeding and breeding habitats do not interbreed mostly because of differences expressed on the behavioral level. Ethological isolation may result both when animals are capable of discrimination both between members of other species and between those of their own species. It may arise as a result of behavioral differences expressed in the acoustic, visual, olfactory, or other sensory domains. In behavioral recognition, flies may use one (i.e., the most important) or more (combined) stimuli, which are both sexually attractive and species-specific. Pheromones play an important role in both mate stimulation and discrimination, and variation in the profiles of adult cuticular hydrocarbons can influence mate choice in many *Drosophila* species (Etges, 2002) and play a role in behavioral isolation (Savarit et al., 1999). Furthermore, it was previously observed that part of the pattern of cuticular hydrocarbons may be adaptive: in several *Drosophila* species they have been shown to vary with environmental conditions such as temperature or humidity (Savarit and Ferveur, 2002 and references therein). In *D. melanogaster*, hydrocarbons on the cuticle of mature flies play a crucial role in mate recognition (Savarit and Ferveur, 2002). On the other hand, in *D. montana* inbred strains, flies mated as actively with individuals of their own strain as with those of alien strains, even though the hydrocarbon profiles of the strains differed considerably (Suvanto et al., 2000).

It was previously shown that in the cactophilic species *D. mojavensis*, pre-mating isolation was determined by larval rearing substrates (Brazner and Etges, 1993). In *D. grimshawi*, males fed high-protein diets were in better physical condition, courted more vigorously, and mated more often and sooner than males fed low-protein diets, whereas females preferred males fed high-protein food (Droney, 1996). *Drosophila melanogaster* is a fruit specialist (Basden, 1954), although it may use decaying plants

**Table 2.** Degree of ethological isolation estimated by the joint isolation index (I) and Chi-square ( $\chi^2$ ) in three experimental groups. Abbreviations: C – strain C; B – strain B; T – strain T; n.s. – non-significant.

Experimental group	Type of mating	Joint isolation index (I) <i>I</i> ± S. E.	$\chi^2$	df	P
MC 1	C x B	0.14 ± 0.07	2.78	1	n.s.
MC 2	C x T	0.13 ± 0.07	2.56	1	n.s.
MC 3	B x T	0.14 ± 0.07	3.18	1	n.s.

**Table 3.** Selection indices of females and males reared on different food and Chi-square values ( $\chi^2$ ). Abbreviations: C – strain C; B – strain B; T – strain T; Zf – female selection index; Zm – male selection index; n.s. – non-significant.

Type of mating	Female selection index (Zf)	$\chi^2$	df	P	Male selection index (Zm)	$\chi^2$	df	P
C x B	1.15	1	1	n.s.	1.23	1.78	1	n.s.
C x T	0.93	0.10	1	n.s.	1.18	0.92	1	n.s.
B x T	0.96	0.10	1	n.s.	0.93	0.24	1	n.s.

like tomato and onion (Atkinson and Shorrocks, 1977). In this experiment, by choosing standard (cornmeal-agar-yeast), banana, or tomato nutritive substrates, suboptimal diets that may disturb the metabolic pattern of the developing larvae were avoided. On the contrary, those substrates permitted normal growth of *D. melanogaster* strains (Kekić and Pavković-Lučić, 2003). However, the nutritional resources used in the experiment differed chemically. For instance, pure banana substrates contain a higher percentage of carbohydrates and a lower percentage of proteins and lipids than in the standard cornmeal medium (Carsten et al., 2005).

As different populations of the same species may develop ethological isolation as a result of genetic divergence, it was hypothesized that ethological isolation among these strains might develop as a byproduct of genetic divergence resulting from the action of different selection pressures operating in different nutritional conditions over 35 generations. However, results of multiple choice experiments involving three "nutritional" strains of *D. melanogaster* showed that the flies mated actively in both inter- and intrastrain combinations. Females did not strongly discriminate during mate choice against males of the opposite nutritional strain. It is well known that in many *Drosophila* species, including *D. melanogaster*, mating occurs when

a female attains the excitation level necessary for male acceptance (Bastock, 1956; Singh and Singh, 1999). This means that courtship of males belonging to different strains was "good enough" to cause the necessary level of female receptivity. In all crosses, the isolation estimate was close to 0, and deviation from randomness was not significant, which indicates random mating and no isolation. Variation in nutritional rearing regimes among strains (the other environmental parameters were constant and optimal for the species) did not increase divergence in the mate recognition system. Similarly, under unusual environmental conditions (total darkness), despite a very long period (for about 27 years), the mate recognition system of individuals from "dark" *D. melanogaster* strains did not change significantly from their controls (Lambert and Harper, 1985). There was stability in the mate recognition system, and darkness did not block any stimulus-response component of mating behavior.

When considering the multiple choice test, it is important to note that lengthening of the observation period increased the number of copulations achieved. During multiple choices, the relative numbers of homogamic and heterogamic matings may change; thus, the measured isolation values depend on how long observations are continued (Gilbert and Starmer, 1985). In fact, it has been demonstrat-

ed that mate choice can change in multiple choice experiments as the trial is lengthened to obtain the maximum number of mates. When the sex ratio is F: M = 1: 1, any single individual can find its mating partner if behavioral differences are not too inappropriate. In the presence of this condition (equal sex ratio), it is better for a female to mate with any available male than to stay unmated. Furthermore, in multiple choice experiments, there is the possibility of comparing and selecting from individuals of both strains included in the test. Regardless of whether females are mating partners that accept or reject courting male, it is possible that females of different strains vary in their attractiveness and that males also respond differently to females from various lines. It was previously recorded that sex pheromones enable *Drosophila* males to discriminate between conspecific females from different laboratory stocks (Tompkins and Hall, 1984). However, in this experiment, it would appear that males and females from various lines were equally attractive. Furthermore, as only virgin flies were used in the experiment, they were highly sexually motivated and possible minor differences among strains did not contribute to the increasing of behavioral isolation. It is also possible that mate choice may differ during a second mating if a fertilized female prefers signals different from those it preferred when it was a virgin. Finally, it is possible that there is an adaptive basis of polymorphism in "smell attractiveness", if it is partly linked with the different types of food which are seasonally or from time to time available to *Drosophila* flies in nature.

When considering mating success of flies in the multiple choice test, sexual selection is not expected to be as intense as in, for example, female choice experiments. Actually, in female choice tests, when the sex ratio was F: M = 1: 2, "banana" males were the most successful in matings (Pavković-Lučić, 2006). It seems that male mating success differs according to the sex ratio used, and that "environmental (i.e., food) strain origin" was more important in competitive conditions, which is a commonly seen situation in this species in nature (Markow, 2002), since one female is usually courted by more than one male at the same time (Markow, 1988).

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## REFERENCES

- Atkinson, W., and B. Shorrocks (1977). Breeding site specificity in the domestic species of *Drosophila*. *Oecologia* **29**, 223-233.
- Basden, E. B. (1954). The distribution and biology of Drosophilidae (Diptera) in Scotland, including a new species of *Drosophila*. *Trans. Roy. Soc. Edinb.* **62**, 602-654.
- Bastock, M. (1956). A gene mutation that changes a behavior pattern. *Evolution* **10**, 421-439.
- Brazner, J. C., and W. J. Etges (1993). Pre-mating isolation is determined by larval rearing substrates in cactophilic *Drosophila mojavensis*. II. Effects of larval substrates on time to copulation, mate choice, and mating propensity. *Evol. Ecol.* **7**, 605-624.
- Carracedo, M. C., Casares, P., and E. San Miguel (1987). Sexual isolation between *Drosophila melanogaster* females and *D. simulans* males. II. Influence of female receptivity on hybridization. *Genome* **29**, 334-339.
- Carsten, L. D., Watts, T., and T. A. Markow (2005). Gene expression patterns accompanying a dietary shift in *Drosophila melanogaster*. *Mol. Ecol.* **14**, 3202-3208.
- Chang, H.-Y., and Y.-T. Tai (2007). Asymmetrical reproductive isolation between *Drosophila albomicans* and *D. nasuta*. *Zool. Stud.* **46**, 638-646.
- Coyne, J. A. (1996). Genetics of differences in pheromonal hydrocarbons between *Drosophila melanogaster* and *D. simulans*. *Genetics* **143**, 353-364.
- Crossley, S. A. (1974). Changes in mating behavior produced by selection for ethological isolation between ebony and vestigial mutants of *Drosophila melanogaster*. *Evolution* **28**, 631-647.
- Doi, M., Matsuda, M., Tomaru, M., Matsubayashi, H., and Y. Oguma (2001). A locus for female discrimination behavior causing sexual isolation in *Drosophila*. *Proc. Natl. Acad. Sci. USA* **98**, 6714-6719.
- Droney, D. (1996). Environmental influences on male courtship and implications for female choice in a lekking Hawaiian *Drosophila*. *Anim. Behav.* **51**, 821-830.
- Ehrman, L. (1961). The genetics of sexual isolation in *Drosophila paulistorum*. *Genetics* **46**, 1025-1038.
- Ehrman, L. (1975). Reproductive isolation in *Drosophila*. In: *Animal Behavior in Laboratory and Field*, 71-74. W. H. Freeman and Company.
- Etges, W. J. (1990). Direction of life history evolution in *Drosophila mojavensis*. In: *Ecological and Evolutionary Genetics of Drosophila* (Eds. J. S. F. Barker et al.), 37-56.

- Plenum Press, New York.
- Etges, W. (2002). Divergence in mate choice systems: does evolution play by rules? *Genetica* **116**, 151-166.
- Gilbert, D. G., and W. T. Starmer (1985). Statistics of sexual isolation. *Evolution* **39**, 1380-1383.
- Hall, J. C. (1994). The mating of flies. *Science* **264**, 1702-1714.
- Joakim, D., and J. W. Curtsinger (1990). Genotype and anesthetic determine mate choice in *Drosophila melanogaster*. *Behav. Genet.* **29**, 73-81.
- Kawanishi, M., and T. K. Watanabe (1980). Genetic variations of courtship song of *Drosophila melanogaster* and *D. simulans*. *Japan. J. Genet.* **55**, 235-240.
- Kekić, V., and S. Pavković-Lučić (2003). Fruit and vegetable food media suitable for maintaining *Drosophila melanogaster* flies. *Drosophila Inf. Serv.* **86**, 147.
- Koref-Santibañez, S., and C. H. Waddington (1958). The origin of isolation between different lines within a species. *Evolution* **12**, 485-493.
- Lambert, D. M., and A. A. Harper (1985). Mating behavior stability in strains of *Drosophila melanogaster* which have been kept under constant darkness for about 27 years. *Japan. J. Genet.* **4**, 281-291.
- Levine, H. (1949). A new measure of sexual isolation. *Evolution* **3**, 315-321.
- Malogolowkin-Cohen, C., Simmons, S., and H. Levene (1965). A study of sexual isolation between certain strains of *Drosophila paulistorum*. *Evolution* **19**, 95-103.
- Markow, T. A. (1988). Reproductive behavior of *Drosophila* in the laboratory and in the field. *J. Comp. Physiol.* **102**, 169-174.
- Markow, T. A. (2002). Perspective: female remating, operational sex-ratio, and the arena of sexual selection in *Drosophila* species. *Evolution* **56**, 1725-1734.
- Massie, K. R., and T. A. Markow (2005). Sympatry, allopatry, and sexual isolation between *Drosophila mojavensis* and *D. arizonae*. *Hereditas* **142**, 51-55.
- Mayr, E. (1963). *Animal Species and Evolution*. Harvard University Press, Cambridge, Mass.
- McFarland, D. (2006). *A Dictionary of Animal Behavior*. Oxford University Press.
- Moehring, A. J., Li, J., Schug, M. D., Smith, S. G., de Angelis, M., Mackay, T. F. C., and J. A. Coyne (2004). Quantitative trait loci for sexual isolation between *Drosophila simulans* and *D. mauritiana*. *Genetics* **167**, 1265-1274.
- Partridge, L., Hoffmann, A., and J. S. Jones (1987). Male size and mating success in *Drosophila melanogaster* and *Drosophila pseudoobscura* under field conditions. *Anim. Behav.* **35**, 468-476.
- Pavković-Lučić, S. (2006). Seksualna selekcija kod *Drosophila melanogaster*. 340 pp. Doctoral Dissertation, Faculty of Biology, University of Belgrade.
- Pavković-Lučić, S., and V. Kekić (2007). Is body size a sexually-selected trait in *Drosophila hydei* males? *Arch. Biol. Sci. (Belgrade)* **59**, 21P-22P.
- Rice, W. R. (1987). Speciation via habitat specialization: the evolution of reproductive isolation as a correlated character. *Evol. Ecol.* **1**, 301-314.
- Savarit, F., and J.-F. Ferveur (2002). Temperature affects the ontogeny of sexually dimorphic cuticular hydrocarbons in *Drosophila melanogaster*. *J. Exp. Biol.* **205**, 3241-3249.
- Savarit, F., Sureau, G., Cobb, M., and J.-F. Ferveur (1999). Genetic elimination of pheromones reveals the fundamental chemical bases of mating and isolation in *Drosophila*. *Proc. Natl. Acad. Sci. USA* **96**, 9015-9020.
- Singh, B. N., and S. Chatterjee (1992). Intraspecific sexual isolation in *Drosophila*. *Indian J. Exp. Biol.* **30**, 260-263.
- Singh, B. N., and S. R. Singh (1999). Female remating in *Drosophila ananassae*: shorter duration of copulation during second mating as compared to first mating. *J. Biosci.* **24**, 427-431.
- Singh, S. R., Singh B. N., and H. F. Hoenigsberg (2002). Female remating, sperm competition, and sexual selection in *Drosophila*. *Genet. Mol. Res.* **1**, 178-215.
- Spieth, H. T., and J. M. Ringo (1983). Mating behavior and sexual isolation in *Drosophila*, In: *The Genetics and Biology of Drosophila* (Eds. M. Ashburner, H. L. Carson and J. N. Thompson), 223-284. Academic Press.
- Stanić, S., and S. Pavković-Lučić (2005). Mating success of wild type and sepia mutants *Drosophila melanogaster* in different choice. *Riv. Biol.* **98**, 513-524.
- Suvanto, L., Liimatainen J. O., Tregenza, T., and A. Hoikkala (2000). Courtship signals and mate choice of the flies of inbred *Drosophila montana* strains. *J. Evol. Biol.* **13**, 583-592.
- Tompkins, L., and J. C. Hall (1984). Sex pheromones enable *Drosophila* males to discriminate between conspecific females from different laboratory stocks. *Anim. Behav.* **32**, 349-352.
- Yamamoto, D., Jallon, J.-M., and A. Komatsu (1997). Genetic dissection of sexual behavior in *Drosophila melanogaster*. *Annu. Rev. Entomol.* **42**, 551-585.

**ДА ЛИ ПОСТОЈИ ЕТОЛОШКА ИЗОЛАЦИЈА ИЗМЕЂУ ЛИНИЈА  
*DROSOPHILA MELANOGASTER* КОЈЕ СУ ВИШЕ ОД 35 ГЕНЕРАЦИЈА  
ГАЈЕНЕ НА РАЗЛИЧИТИМ СУПСТРАТИМА?**

СОФИЈА ПАВКОВИЋ-ЛУЧИЋ

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Постојање етолошке изолације између три линије *Drosophila melanogaster* које су гајене преко 35 генерација на различитим супстратима (стандардни кукурузни медијум, подлоге од банане и парадајза) је тестирано у серијама тзв. теста ва вишеструког избора. Дуготрајно одржавање линија на различитим супстратима није довело до сексуалне изолације између ових линија, јер

изолациони индекси (Malogolowkin-Cohen-ови изолациони индекси) нису били статистички значајни. Такође, мужјаци и женке који су били одгајани на различитим хранљивим подлогама су били подједнако успешни у парењу, с обзиром да статистички значајне разлике у броју копулација које су остварили мужјаци и женке из различитих линија нису уочене.