A remarkable microscale differentiation has been established in a *D. melanogaster* natural population at "Evolution Canyon", Lower Nahal Oren, Mount Carmel (Israel) (Nevo, 1997a; Nevo et al., 1997; Derzhavets et al., 1996a, b, 1997). The studies were conducted on isofemale lines set up from flies of the opposite slopes of the Canyon and maintained in normal laboratory conditions for 1-2 years. The measured traits included viability and longevity changes caused by short-term and lifetime temperature treatments, changes in fly weight due to desiccation/starvation treatments at different temperatures, fluctuating asymmetry test of wing scores, and rates of mutation and recombination. The obtained evidence leads to the conclusion that strong microclimatic natural selection can override migration and generate a complex slope-specific adaptive syndrome contributing to fitness at a microsite.

The foregoing tests also showed (both for *D. melanogaster* and *D. simulans*) that adaptation to contrasting ecological conditions of the opposite slopes has resulted in strong genetic divergence for habitat choice (Nevo et al., 1997). The direction of the revealed interslope difference in the oviposition preferred temperatures is exactly the one expected in accordance with habitat selection and habitat choice models (Hedrick, 1990). Theoretical models predict that selection in systems with spatially heterogeneous selection can bring about a genetically determined reduction in the migration rate (Wiener and Feldman, 1993). In view of that, our results established a promising natural model to bridge theory and evidence in an in-depth analysis of behavioral adaptation under heterogeneous stressful conditions (Parsons, 1993) including the interactions between selection, migration, and habitat choice. This model is one of several model organisms currently studied at the "Evolution Canyon" research program.

Inter-population geographical isolation (allopatry) may promote the evolution of sexual isolation as a first step of speciation (Dobzhansky and Pavlovsky, 1957; Mayr, 1963). Incipient sexual isolation among geographic races of the same species has been reported in a number of studies (e.g., Wu et al., 1995; Noor, 1995; Wade et al., 1995). Habitat choice caused by strong microsite ecological-genetic differences can also develop a tendency toward within-population reproductive isolation (sympathy). However, the model of sympatric speciation is by far more controversial than the allopatric model and suffers from scarce evidence in natural populations. Here we report the results of some pilot tests on sexual behavior conducted on *D. melanogaster* genotypes derived from subpopulations of "Evolution Canyon" that display sharp adaptive divergence over a short distance, of 200 m, presumably resulting from the contrasting interslope ecological differences.

The experiments were conducted on two specially designed testing groups (one per slope) that have been constructed by crossing 25 isofemale lines from each slope. The lines were derived from inseminated females caught at mid-slope stations of the opposite slopes of the Canyon and maintained in normal laboratory conditions for about 2 years (see Nevo et al., 1997). To establish the testing groups, 10 males and 10 virgin females of each line were crossed and then maintained under random mating for 12 generations. We conducted three tests, aimed to examine (i) mate choice, (ii) dynamics of mating pair formation, and (iii) mating speed and copulation duration.

**Mate-choice:** The first test aimed to measure biases in choices of males 'exposed' to females of their own and opposite slopes. The experiments were conducted in a special apparatus made of 2 mm plexiglass. The apparatus included 3 compartments. The main chamber (11x11x11 cm) and offshoots (3x3x10 cm) were separated by thin nylon nets, permitting flies to see and smell each other, but not to cross the nets. Virgin flies used for the test were aged for 7 days after eclosion. Fifty tested males were introduced without etherization into the main chamber of the apparatus; simultaneously, 25 females of the testing group of the more stressful warm and xeric south-facing slope (SFS) were introduced into one offshoot and 25 females of the testing group of the cool and mesic north-facing slope (NFS) into the other one. Direct observations of males choice were made over a period of 60 minutes. The number of males interested in females from one or another offshoots was recorded (Table 1).

Males of both testing groups discriminate against females of the groups of the opposite slopes. Out of 50 males from the NFS-group 70.4% were oriented to females from their own slope and only 46.4% to SFS-females. Similarly, 49.6% of males from SFS preferred females of their own origin and only 26.8% - females from NFS (note, that the sums of these frequency indices in some combinations are more than 100%, because males were often interested not only in females of one sample, but also in a second-batch female). The differences between the number of homo- and heteroslope choices (*D* = homo-hetero) were highly significant and rather similar for males of both slopes: for SFS males *D* = 11.4, Student's test *t* = 6.69, *df* = 8, *P* < 0.001; for NFS males *D* = 12.0, *t* = 5.59, *df* = 8, *P* < 0.001). Thus, with respect to sexual activity, males of both slopes demonstrated a bias toward females of their own origin..
According to the data in Table 1, males of the SFS-group evolved a lower level of sexual activity than NFS-males. This difference is manifested when comparisons $ d = NFS-SFS $ are made for both homo- and heteroslope choices. The interslope difference ($ d $) of the mean scores was: for homoslope choice $ d = 10.4, t = 4.61, df = 8, P < 0.01 $; for heteroslope choices $ d = 9.8, t = 6.31, df = 8, P < 0.001 $. These results fit the generalization of a reduced metabolic rate and energy-saving behavior in populations adapted to stressful environments (Parsons, 1993).

**The dynamics of mating pairs formation:** In order to evaluate the dynamics of mating pairs formation, females and males of each testing group were mated in all possible combinations (referred to as homo- and hetero-crosses) in a mating chamber. For each cross, the mating experiment was conducted in three independent blocks, each with 25 virgin females and 25 males. Number of couples was recorded at 5-min intervals for an hour. The greatest number of mating couples was recorded in the homo-cross (SFS$xSFS$) after 15-20 minutes from the beginning of the experiment; here matings were virtually finished during the first 30 min. The second homo-cross (NFS$xNFS$) manifested a slower dynamics of pair formation. The greatest number of mating pairs was recorded after 25-30 min and the maximum number of couples per registration was lower. Nevertheless, these differences did not reach the border of significance, hence further tests are needed.

**Mating speed and duration of copulation:** In this test, 25 individual pairs were observed for each of the four possible crossing combinations (SFS$xSFS$, NFS$xNFS$, SFS$xNFS$ and NFS$xSFS$). The foregoing two parameters were registered during the first 120 min from the beginning of the test. The pairs that did not mate during this period were recorded as 'non-mated' and were not taken into account. In order to get 25 mated couples, the test was continued with additional pairs. The obtained estimates indicated a significantly higher mating speed in intraslope matings than in interslope ones, i.e. a positive assortative mating, especially strongly manifested by NSF females (Table 2).

In both types of crosses (homo- and hetero-), males attempted to court immediately, showing no significant differences in their courtship. Mating success and, therefore, mating speed are influenced, to a large extent, by the level of females' receptivity. The results suggest that with respect to homogeneous mating, females of the SFS-subpopulation have evolved a decreased receptivity to their own males as compared to NFS-females (11.8 versus 8.6 mean scores, $ t = 2.54, df = 48, P < 0.05 $). This fits the generalization of a reduced metabolic rate in natural populations adapted to stressful environments (Parsons, 1993). The foregoing data on heterogeneous crosses imply that interslope population differentiation brought about a highly significant mating selectivity of NFS-females in favor of males of their own origin (compare the mean scores 20.0 and 8.6 for hetero- and homo-crosses, $ t = 9.45, P < 0.001 $). The same, but less pronounced, trend is exhibited by SFS-females ($ t = 2.12, P < 0.05 $). In accordance with the results on mating speed, NFS flies manifest also significantly longer copulation in homogeneous crosses than do SFS flies ($ t = 5.23, P < 0.001 $). Likewise, a highly significant reduction in copulation duration was observed in the heterogeneous cross NFS (females) $ \times $ SFS (males) as compared to the homogeneous one, NFS (females) $ \times $ NFS (males) ($ t = 9.44, P < 0.001 $). However, an opposite although less significant trend was observed in the heterogeneous cross SFS (females) $ \times $ NFS (males) ($ t = 2.40, P < 0.05 $). With the exception of the last difference, all results of the third experiment correspond to the assumption of non-random mating behavior of the tested populations with a tendency to positive assortative mating.

The foregoing results allow us to suggest that population differentiation at "Evolution Canyon" caused by strong interslope ecological contrasts has resulted in an incipient premating sexual isolation system, in spite of the extremely small distance between the slopes. Further tests are needed for an in-depth study of this evolving sympatric isolation system.

Gaining Access to the BIOSCI/bionet.DROSOPHILA Newsgroup

This information was provided by Dave Kristofferson, BIOSCI/bionet Manager.

The BIOSCI/bionet DROSOPHILA newsgroup can be accessed on USENET by reading bionet.drosophila using your local USENET newsreader or on the World Wide Web at

http://www.bio.net/hypermail/DROS/.

The Web address includes access to the complete newsgroup archive going back to the group's beginning in July 1993. If your browser is configured to send e-mail, you can also post to the newsgroup via this Web site.

For readers who prefer to use e-mail exclusively, there are two sign-up procedures based on your location.

For users in the Americas and Pacific Rim countries, address a message to biosci-server@net.bio.net and include the line

subscribe dros

(NOT "subscribe drosophila" as printed in DIS 77)

in the *body* of the message. Anything put on the Subject: line will be ignored. To cancel your subscription repeat the same procedure using the

unsubscribe dros

command. Your address on the From: line of your mail message must match the address on the list for this to work. Please address any problems to biosci-help@net.bio.net for technical assistance.

For users in Europe, Africa and Central Asia, address a message to mxt@dl.ac.uk and include the line

sub bionet-news.bionet.drosophila

in the body (not the Subject:) of the message. To unsubscribe send in the command

unsub bionet-news.bionet.drosophila