duced a great number of eggs. The first pupae were carefully transferred to small tubes (10 cm^3) - each tube received only a pupa. The virgin males and females obtained in such a way were then separated in couples, each couple being introduced into an individual bottle with the same culture medium. We obtained mating of *P. kerteszi* under controlled conditions in all the attempts carried out (15 couples). The courtship and also the behaviour of males and females, before and after the copula, were observed for the first time in the laboratory. The male stays in front of the female and moves the wings rapidly from sec to sec many times. Next, he curves the abdomen downwards and stays in this position while the female is in front of him. If the female goes away he repeats the operation from the beginning. When the female accepts the courtship she stands the abdomen up slowly from time to time; occasionally she may also move her wings. After the mating the male cleans the abdomen with his posterior legs and moves his wings again.

We registered the mating duration in only 2 cases; 32 and 90 sec, respectively. The 15 mated females produced from 25 to 93 descendants ($\bar{X} = 57 \pm 21.48$ descendants). In the 15 tubes we registered 432 females and 423 males (approximately 1:1; $\chi^2 = 0.90$, nonsignificant at 5% level). The relative viability for the descendants, from egg to imago, was determined in 8 out of the 15 tubes. The 8 mated females used in this experiment laid 531 eggs; the number of eggs laid by each female varied from 31 to 102 ($\bar{X} = 66.37 \pm 22.11$ eggs). The mean viability from egg to imago, considering the 8 experimental tubes, was around 59%. The minimum mean viability was observed in the tube number 4 (40.32%) and the maximum, in the tube number 3 (76.19%)[1].

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A note on differences in the mating behavior of *Drosophila heteroneura* and *D. silvestris*[1]

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Summary. Differences between *Drosophila heteroneura* and *D. silvestris* in the time taken to copulate and the preferred location for copulation are reported. The potential significance of these differences is discussed.

Watson[3] described differences in the mating display of *Drosophila heteroneura* (Grimshaw) and that of *D. silvestris* (Perkins) which might, in part, be responsible for the high level of premating isolation between these species[4,5]. This note reports 2 additional differences in mating behavior between these closely-related, sympatric drosophilids.

Methods and materials. All observations were made on courtship patterns of individuals from Kahuku Ranch, Southern Hawaii. Virgin females were reared in the laboratory either from laboratory stocks [University of Hawaii, Stock Numbers: U26B52, U26B57 *(heteroneura)* and U26B9 *(silvestris)*], or from larvae collected in the field by Kenneth Y. Kaneshiro, John Tonzetich and the author. All males were wild-caught.

Observations of mating behavior were made during experiments testing female discrimination. In each experiment, a mature virgin female was caged with a male of each species in a clear glass cylinder (26.5 cm diameter, 12.0 cm high) with a removable loose-weave muslin top and a polyethylene base covered with damp sand. A small dish of food and a piece of plant material (or artificial substitute) were placed in each cage. During some experiments absorbent paper was taped to part of the inner wall of each cage to facilitate locomotion of the flies; no significant effect on positions of successful copulations was noted.

All copulations were timed to the nearest 30-sec using a wall clock with sweep second hand, and the positions of successful courtships were noted. For this purpose the cages were considered to consist of 3 sections: the floor, the walls and plant material, and the roof. All statistical procedures follow the methods of Zar[6]. During the experiments 6 cages were observed for approximately 3 h per day (08.00-11.00 h). The results are based on 27 days of observation involving 77 trios (43 with *silvestris* females; 34 with *heteroneura* females); 39 copulations were observed.

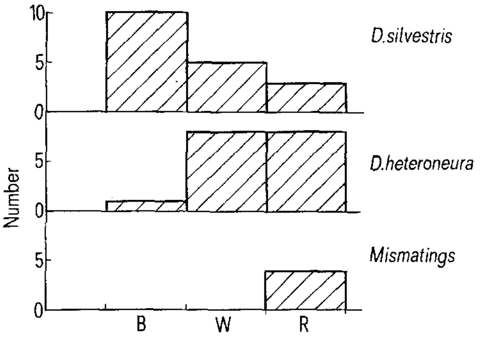

Fig. 1. Differences in positions of successful copulations in *Drosophila heteroneura* and *D. silvestris*. B, the base of the cage; W, the walls of the cage including plant material; R, the roof of the cage.

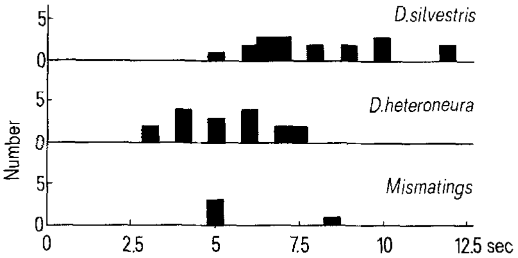

Fig. 2. Durations of copulation in *Drosophila heteroneura* and *D. silvestris*.

Results. Figure 1 shows the positions of successful courtships. A χ^2 test on these data (excluding the interspecific matings) shows that the differences are highly significant ($0.01 > p > 0.005$).

Details of the durations of copulations in each species are shown in figure 2. A Mann-Whitney U-test on these data (excluding the interspecific matings) indicates that these interspecific differences are highly significant ($p \ll 0.01$).

Discussion. The biological importance of the differences in mating behavior reported here is not clear. The copulation times of 3 of the 4 interspecific matings are those of the species to which the male belongs. If the male terminated copulation then the mismatings observed in this study may be less successful than intraspecific matings, because the female may have insufficient time to complete a mating sequence. For example, a difference in times of copulation could affect the efficiency of sperm transfer in interspecific matings.

Differences in preferred sites for courtship and mating may play a more direct role in interspecific ethological isolation. The results indicate that *D. heteroneura* and *D. silvestris* prefer different areas for courtship; thus there may be microgeographic isolation between them. All the mismatings occurred in the preferred area of the male involved, suggesting that lack of site discrimination by the female may contribute to interspecific hybridization. Such hybridization has been reported from Kahuku Ranch and morphological studies of hybrid individuals indicate that mismating is occurring in only 1 direction, viz. *silvestris* ♀ × *heteroneura* ♂[7].

Few complete mating sequences have been observed in the field despite intensive observations[8]. However, it is clear that copulation normally does not take place on the lek site where males advertise. On the basis of the observations made in this study it seems possible that mating in *D. heteroneura* commonly occurs on vertical surfaces of the undersides of horizontal structures in elevated positions, whereas mating in *D. silvestris* takes place on or near the ground.

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Evolution of behavioral reproductive isolation in a laboratory stock of *Drosophila silvestris*

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Summary. A laboratory stock of *D. silvestris* has become partially sexually isolated since its origin from 1 natural population of this species. Evolution of reproductive isolation is probably incidental to reorganization of the genome, particularly the genetics of sexual behavior, following random drift in a small population.

Essential to speciation is the origin of reproductive isolating mechanisms. Speciation, as conceptualized by Mayr[2], can occur when a geographical barrier fragments a population of interbreeding organisms. Subsequent to cessation of gene exchange, genetic divergence proceeds in the isolate either in response to selection leading to adaptation to new environmental conditions, or merely due to drift. But without establishment of reproductive barriers the newly derived species could be assimilated into the ancestral population were the geographical barrier to be removed. Indeed, reproductive isolation is an integral part of the biological species concept.

In the Hawaiian Islands speciation of Drosophila has occurred by a series of founder events. Carson[3] has theorized that very few individuals (perhaps but a single gravid female) reach a previously uninhabited island where a new species evolves. It has been established that courtship behavior is of primary importance as an isolating mechanism in Hawaiian picture-winged Drosophila[4-6]. Spieth[7] has observed that courtship of picture-winged Drosophila involves a complex pattern of stimulus-response interactions between male and female. It seems valid to assume the elements of courtship behavior are genetically determined and under strong selection. Furthermore, the genes determining elements of courtship behavior should be subject to genetic drift during the founder event as is the rest of the genome.

Kaneshiro[5] has proposed that 'a few of the elements of the courtship pattern of the ancestral population are changed ('lost') in the genetic revolution which accompanies the founder event in the derived population'. According to his hypothesis, females of the derived species are stimulated to accept copulation by males of the ancestral species but in the converse situation derived males do not perform all the courtship patterns necessary to stimulate ancestral females to copulate. Several serendipitous occurrences in the lab provided an experimental situation in which Kaneshiro's hypothesis could be tested.

Materials and methods. 2 stocks of *Drosophila silvestris*, R59G4 and U28T2, were established from single native females collected at Kilauea Forest Reserve at 1510 m near Hawaii Volcanoes National Park. Female R59G4 was collected in November 1972, and the isoline suffered at least 2 major crashes when the effective population size may have been as low as two, once in 1974 and once in 1975. Female U28T2 was brought to the laboratory in January 1977, and the isoline was maintained at an effective population size of about 100 until mating preference

Mating preference tests between two laboratory stocks of *Drosophila silvestris*

Male[a]	Mating					
	Homogamic	Heterogamic	n	I[b]	c[c]	p
U	10	22	32	−0.38	−2.17	<0.05
R	24	3	27	0.78	4.06	<0.01

[a] U = U28T2, R = R59G4; [b] Stalker Isolation Index; [c] c = t with infinite degrees of freedom; calculated by the method of proportions.