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BRIEF REPORT

Mating Success of Photoreceptor Mutants of *Drosophila melanogaster*[1]

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The compound eye of *Drosophila* contains 700–800 individual ommatidia, each made up of six outer retinula cells (R_{1-6}), and two central retinula cells (R_7 and R_8). Each retinula cell contains a rhabdomere which is the location of the visual pigment. Mutants have been recovered which specifically eliminate function in R_{1-6} or in R_7. Other mutants, such as $norp^{AP24}$, result in a loss of the receptor potential for the entire eye. In a series of "female choice" experiments, the relative importance of the outer and inner retinula cells for male courtship success was investigated. The results suggest that in *D. melanogaster* an intact visual system, expecially R_{1-6}, is important for male mating behavior. In the absence of functional R_{1-6}, a small but significant role for R_7 was detectable.

The main photoreceptors of *Drosophila melanogaster* are the two large compound eyes, each of which is composed of about 700–800 ommatidia. A detailed description of the structure and function of the *Drosophila* compound eye is found in the comprehensive review by Pak and Grabowski (1978). Light is prevented from passing between adjacent ommatidia by the screening pigments which give *Drosophila* eyes their characteristic reddish-brown appearance. Each ommatidium contains eight retinula cells arranged in a highly organized fashion. Six retinula cells (R_1–R_6) form a peripheral ring around the two central cells R_7 and R_8, R_8 being located directly beneath R_7. Within each retinula cell is a rhabdomere which contains the visual pigment.

We were interested in the relative importance of the peripheral and central retinula cells for male courtship success. Recently the application of genetic techniques to the study of *Drosophila* vision has generated a series of mutants with known, highly specific neuroanatomical and physiological characteristics. One mutant, *ora* (III—65.3 ± 0.4, "outer rhabdomeres absent," Koenig & Merriam, 1977), results in a loss of function in the six peripheral rhabdomeres. Another, *sev* (I—33.2 ± 0.2, "seven-

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less," Harris et al., 1976) eliminates the function of R_7. The receptor potential of the entire compound eye may be eliminated by mutants at the *norp*^{A} locus ("no receptor potential," I—6.5 ± 0.1, Pak et al., 1976). We used "female choice experiments" to compare the relative mating success of wild-type males and males of the above mutant phenotypes in order to assess the importance of the inner and outer rhabdomeres for courtship outcomes.

Strains of flies. Mutant strains were obtained from the laboratories of Dr. John Merriam at UCLA and Dr. William Pak at Purdue University. The Canton-S (CS) control strain was also obtained from UCLA. Mutant strains are characterized by abnormal countercurrent behavior and/or by specific alterations of the electroretinogram (ERG). The strains used were *norp*^{AP24}, *ora*^{JK84}, *sev*, and the double mutant *sev; ora*^{JK84}. All strains had the wild-type reddish-brown eye color phenotype.

Fly culturing. Flies were reared on standard cornmeal molasses agar medium at 24 ± 1°C. Virgin males and females were separated under light ether anesthesia and stored separately until use in experiments at 4 days of age.

Female choice experiments. Four-day-old CS females were aspirated into 8-dram shell vials each containing one mutant and one wild type male. Males were distinguished by small wing clips made during initial separation. The time until mating and genotype of the successful male was recorded. Vials showing no mating were discarded after 1 hr. Several replications of about 10–20 matings each were carried out for each type of experiment. The genotype of the male clipped was alternated between replications even though no effect of small clips has been found (Markow et al., 1978).

Male mating success is shown in Table 1. In most experiments where females have a choice between CS and mutant males, CS males are at a significant advantage. Other female choice tests show that when one male is *sev*, all other competing mutant males are at a significant disadvantage. In the case where one male is *ora* and the other male is *sev; ora*, the *ora* male was more successful. Both males had an intact R_8 and both were missing outer rhabdomeres, but only one had R_7. Those with R_7 tended to be more successful. Further evidence for the importance of R_7 comes from comparing the last two experiments in Table 1. In the *ora–norp*^{A} choice, males having an intact R_7 are significantly more successful than males lacking any receptor potential. But when R_7 is taken away, this advantage disappears.

It was apparent during the female choice experiments that wild-type males mated before mutant males even began courting. A series of experiments was therefore designed to test courtship latency, that time from the initial introduction of a single male into a chamber containing a CS female to when courtship was first observed. The results appear in Table 2. A

TABLE 1
Relative Mating Success of Visually Mutant Males in Female Choice Experiments[a]

Male genotype			Successful male				
A	B	Mating	A		B		χ^2
		(%)	Obs	Exp.	Obs.	Exp.	
CS	*sev*	87.3	29	27	25	27	.296
CS	ora^{JK84}	92.7	50	25	0	25	50.000*
CS	*sev*;ora^{JK84}	85.5	67	34.5	2	34.5	61.232*
CS	$norp^{AP24}$	98.5	56	29	2	29	50.276*
sev	ora^{JK84}	96.2	50	26	2	26	44.30 *
sev	*sev*;ora^{JK84}	93.0	44	22.5	1	22.5	41.09 *
ora^{JK84}	*sev*;ora^{JK84}	50.5	61	40.5	30	40.5	5.44 *
sev	$norp^{AP24}$	100	48	25	2	25	42.32 *
ora^{JK84}	$norp^{AP24}$	62	53	41	29	41	7.024*
$norp^{AP24}$	*sev*;ora^{JK84}	42.3	36	30	24	30	2.40

[a] All females were from the Canton-S wild-type strain.
* $p < .01$.

Duncan multiple range test at α .05 places the males in two separate categories with respect to courtship latency. Wild-type CS males and *sev* males appear in subset A which is significantly different ($p < .05$) from subset B. Not only did it take males from subset B substantially longer to locate females in order to begin courting them, but these males frequently showed courtship elements that were inappropriately directed. Males were seen to circle and vibrate when no female was in the immediate vicinity, while this was not observed among wild-type or *sev* males. These males frequently lost contact with the females once they had begun courting.

The occurrence of mutants which specifically eliminate the function of R_{1-6} or R_7 is suggestive of a distinct developmental origin of the peripheral cells from R_7 and R_8. The two central cells probably have a different developmental relationship from each other. Such an idea is supported by

TABLE 2
Courtship Latency of Males from Each Strain, Tested Individually with CS Females

Male genotype	Courtship latency (in sec)
CS	51.74 ± 7.37
ly^3	64.30 ± 8.12 Subset A*
ora^{JK84}	290.14 ± 31.62
ly^3;ora^{JK84}	341.86 ± 39.44 Subset B*
$norp^{AP24}$	326.73 ± 45.11

[a] $\bar{X}$ ± SE.
Note. A Duncan multiple range test gives two significantly different subsets at $p < .05$.

the fact that each of the different types of cells show differences in spectral sensitivity peaks, R_{1-6} having a λ max of $\simeq$ 465 nm, R_7 with a λ max of $\simeq$ 370 nm (uv), and R_8 showing a λ max of $\simeq$ 485 nm. Each is felt to have spectrally distinct visual pigments (Pak & Grabowski, 1978). It has been suggested that R_{1-6} are somewhat important for motion detection and acuity (Pak & Grabowski, 1978). While the visual pigments in *norp*^{A} are apparently normal, the lesion evidently affects a part of the phototransduction process occurring after the rhodopsin–metarhodopsin transition, resulting in complete blindness for flies homozygous or hemizygous for extreme *norp*^{A} alleles (Pak et al., 1976). R_7 is supposed to function somewhat specifically as a uv detector and with white light, *sev* shows a near normal ERG and only slightly aberrant countercurrent behavior.

It is obvious from the present study that males which have serious visual defects also show markedly reduced competitive courtship success. The fact that males lacking function in R_{1-6} show a reduced success similar to that seen among blind *norp*^{A} males would imply that the outer rhabdomeres are very important at some stage of the courtship process. Since in general, flies with an intact R_7 show greater mating success relative to *sev* males, R_7 would appear to have some role for mate location and/or proper execution of courtship behaviors. Males from the *sev; ora* strain and *norp*^{A} strain do not differ significantly in mating success, a finding which suggests that R_8 might be of little importance in the courtship process. Unfortunately, this idea cannot be more critically examined since no mutants have yet been found which only alter R_8.

While the mating behavior of *D. melanogaster* has been classified as independent of light (Bastock, 1956; Grossfield, 1971) several lines of evidence contradict this idea. First of all, among wild-type flies, matings occur more quickly in the light than in darkness (Markow, 1975). Mutants which alter screening pigments affect the quantitative and qualitative aspects of male courtship (Sturtevant, 1915; Connolly, Burnet, & Sewell, 1969). Furthermore, mutants which affect screening pigments are rapidly lost from laboratory populations maintained in the light but not from populations kept in darkness (Burnet & Connolly, 1973). These observations are not surprising since screening pigments have been shown to affect visual acuity (Kalmus, 1943).

Burnet and Connolly (1973) suggest several levels at which photoreceptors may function in mating behavior. Among them are ability to locate the female and properly direct courtship toward her. A loss of photoreception or of visual acuity such as occurs with altered screening pigments or of rhabdomere function may impede mate localization and courtship behavior. Our findings support this idea and provide evidence that R_{1-6} and R_7 are important for these events to occur normally. While there is absolutely no evidence that the photoreceptor mutants used in the present study affect processes or structures beyond those described in the

visual system, it is remotely possible that their effect upon mating success could result from some unknown pleiotropy.

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