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Frequency dependence of mating success in *Drosophila pseudoobscura*

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1. INTRODUCTION

Direct observations have been made, using Elens–Wattiaux observation chambers (1964), of mating frequencies within groups of *Drosophila pseudoobscura* containing different proportions of individuals homokaryotypic for the third chromosome gene arrangements, ARrowhead and CHiricahua. The proportions of both sexes varied from 18:2 of CH/CH:AR/AR to 2:18 of CH/CH:AR/AR (Ehrman, 1968, and references therein). Minority males mated relatively more frequently than the majority ones. When the two kinds of males were equally frequent, the matings they participated in were usually also equally frequent. Attempts to identify the sensory cues by means of which the females perceive the relative frequency of the courting males in the chamber have thus far not produced conclusive results (Ehrman, 1968).

It would seem then, that under the conditions of these experiments the Darwinian fitness of a genotype may depend on its frequency in a population, with increased fitness tending to be a consequence of rarity. Frequency-dependent changes in Darwinian fitness were also found by Tobari & Kojima (1967) and Kojima & Yarbrough (1967) with heretofore unreported frequency-dependent selection involving inversion karyotypes in *Drosophila ananassee* and also the esterase 6 locus in *D. melanogaster*. Frequency-dependent selection could theoretically maintain balanced polymorphisms even without the heterokaryotypes having selective advantages. A further study, to be reported here, has been undertaken to ascertain the role of frequency-dependent selection when some of the competing individuals are heterokaryotypic.

2. MATERIALS AND METHODS

The ARrowhead and CHiricahua populations utilized here were the same as used by Ehrman, Spassky, Pavlovsky & Dobzhansky (1965) and Ehrman (1966). The 'positive' and 'negative' designations are attached to them in order to distinguish them from other sets of strains with which these experiments were repeated. Positive and negative are the directions of the now long-relaxed selection for geotaxis formerly practised upon these AR and CH strains (Dobzhansky & Spassky, 1962). In this paper they are referred to as 'P strains' and 'N strains', respectively.

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The technique of direct observation of the matings, and the simple apparatus used, have been described and figured by Ehrman (1965). The method permits recording of four types of mating, i.e. A♀ × A♂, A♀ × B♂, B♀ × A♂ and B♀ × B♂. It is possible to record also the time when each mating occurs and its position in the sequence of matings. Females and males, aged separately for at least 3 days, were introduced into a chamber, and were observed at 6-min intervals or more often for 3 h. Most of the matings took place within the first hour, when observations were made more often than 6 min apart. To make the flies from different strains distinguishable in the observation chamber, with the aid of a four-power hand lens, the distal margin of one wing was clipped in one of the strains; the strains so marked and unmarked were alternated in successive runs. The wing clipping was done to very lightly etherized flies. Introduction into the chambers is made without new etherization.

3. RESULTS AND DISCUSSION

Tables 1 and 2 summarize the results of experiments in which flies homozygous for either the AR or the CH gene arrangements were placed in observation chambers with flies heterozygous for the same arrangements, in the ratios 1:1, 1:9 and 9:1. CH/AR refers to individuals with CH/CH mothers and AR/AR fathers; reciprocally, AR/CH = the offspring from an AR/AR♀ × CH/CH♂ cross. Each observation chamber always had forty individuals—twenty females plus twenty males. Several runs were made with each proportion of the two karyotypes, as shown in the Tables. The data are reported in two ways, taking into account 'all matings' or only the first half of the matings observed ('early matings'). This is necessary because while a female can mate only once during the period of observation, a male can mate repeatedly. In many of the runs all or almost all of the females have mated, making calculations of chi-squares for the females meaningless in the 'all matings' series. The Tables report the numbers of the females and males of each kind that were observed in copula; repeated matings of the same male were counted as separate matings. The chi-squares indicate the significance of the deviations observed from random mating, i.e. from the numbers expected if the probability of an individual mating was independent of the frequencies of the two genotypes in the chamber. These chi-squares have 1 degree of freedom. (A chi-square of 3·84 is significant at the 5% level, while 6·63 is significant at the 1% level.)

The data for the set of N strains may be considered first (Table 1). When AR/AR and AR/CH, AR/AR and CH/AR, or CH/CH and CH/AR are present in equal numbers (the 10:10 ratio), both kinds of male and female mate in proportion to their numbers. The highest, but still not significant, chi-square in these instances is only 2·33. When CH/CH males compete with equally numerous AR/CH males the heterozygote is more successful (giving a significant chi-square of 6·69). When CH/CH is made rare (the 2:18 ratio) this superiority is lost, but is expressed again when AR/CH is rare. The other three combinations clearly give positive frequency-

Table 1. *Matings observed with different proportions of homokaryotypic AR or CH and heterokaryotypic AR/CH (from an AR♀ × CH♂ cross) or CH/AR (from a CH♀ × AR♂ cross) flies per observation chamber, N strains*

(Ho = homokaryotype; He = heterokaryotype; I = isolation index.)

			All matings							Early matings				
			Males			Females		Males			Females			
		Runs	Ho	He	χ^2	Ho	He	Ho	He	χ^2	Ho	He	χ^2
AR/AR : AR/CH			AR/AR	AR/CH		AR/AR	AR/CH	AR/AR	AR/CH		AR/AR	AR/CH	
1.	10 : 10	6	63	47	2·33	58	52	41	16	10·96	26	31	0·44
2.	18 : 2	6	83	17	5·44	89	11	42	8	2·00	42	8	2·00
3.	2 : 18	6	19	86	7·65	12	93	9	44	2·87	6	47	0·10
AR/AR : CH/AR			AR/AR	CH/AR		AR/AR	CH/AR	AR/AR	CH/AR		AR/AR	CH/AR	
4.	10 : 10	6	50	53	0·09	49	54	26	27	0·02	26	27	0·02
5.	18 : 2	6	84	27	25·31	99	12	43	13	10·87	45	11	5·79
6.	2 : 18	7	25	85	19·80	14	96	12	46	7·36	9	49	1·96
CH/CH : AR/CH			CH/CH	AR/CH		CH/CH	AR/CH	CH/CH	AR/CH		CH/CH	AR/CH	
7.	10 : 10	7	41	68	6·69	56	53	19	38	6·33	21	36	3·95
8.	18 : 2	8	75	26	27·81	87	14	39	14	15·87	40	13	12·43
9.	2 : 18	9	18	84	6·63	15	87	8	44	1·68	10	42	4·92
CH/CH : CH/AR			CH/CH	CH/AR		CH/CH	CH/AR	CH/CH	CH/AR		CH/CH	CH/AR	
10.	10 : 10	7	49	53	1·75	55	57	24	32	1·14	28	28	0·00
11.	18 : 2	7	72	49	125·03	105	16	38	24	56·78	48	14	10·90
12.	2 : 18	7	16	97	2·17	13	100	6	53	0·00	7	52	0·23

1, I = $-0{\cdot}11 \pm 0{\cdot}095$; 4, I = $+0{\cdot}09 \pm 0{\cdot}098$; 7, I = $+0{\cdot}06 \pm 0{\cdot}095$; 10, I = $+0{\cdot}04 \pm 0{\cdot}094$; N = 1287.

Table 2. *Matings observed with different proportions of homokaryotypic AR or CH and heterokaryotypic AR/CH (from an AR ♀ × CH ♂ cross) or CH/AR (from a CH ♀ × AR ♂ cross) flies per observation chamber, P strains*

(Ho = homokaryotype; He = heterokaryotype; I = isolation index.)

		All matings					Early matings					
		Males			Females		Males			Females		
	Runs	Ho	He	χ^2	Ho	He	Ho	He	χ^2	Ho	He	χ^2
AR/AR:AR/CH		AR/AR	AR/CH		AR/AR	AR/CH	AR/AR	AR/CH		AR/AR	AR/CH	
1. 10:10	6	28	74	20·75	45	57	15	36	8·65	17	34	5·67
2. 18:2	9	75	25	25·00	84	16	42	11	6·81	42	11	6·81
3. 2:18	6	21	82	12·35	12	91	10	43	4·63	8	45	1·53
AR/AR:CH/AR		AR/AR	CH/AR		AR/AR	CH/AR	AR/AR	CH/AR		AR/AR	CH/AR	
4. 10:10	9	33	68	12·13	49	52	16	36	7·69	26	26	0·00
5. 18:2	7	82	24	18·82	93	13	44	10	4·35	49	5	0·03
6. 2:18	6	24	86	17·07	11	99	11	45	5·79	4	52	0·51
CH/CH:AR/CH		CH/CH	AR/CH		CH/CH	AR/CH	CH/CH	AR/CH		CH/CH	AR/CH	
7. 10:10	6	32	80	20·57	53	59	16	41	10·96	23	34	2·12
8. 18:2	6	82	31	38·16	101	12	41	16	20·68	49	8	1·03
9. 2:18	4*	14	52	9·22	7	59						
CH/CH:CH/AR		CH/CH	CH/AR		CH/CH	CH/AR	CH/CH	CH/AR		CH/CH	CH/AR	
10. 10:10	8	40	77	11·70	53	64	21	39	5·40	15	45	15·00
11. 18:2	6	83	30	34·38	102	11	44	14	12·88	49	9	1·96
12. 2:18	7	23	81	16·96	13	91	13	40	12·43	9	44	2·87

1, I = $+0{\cdot}16 \pm 0{\cdot}098$; 4, I = $+0{\cdot}09 \pm 0{\cdot}099$; 7, I = $+0{\cdot}05 \pm 0{\cdot}094$; 10, I = $+0{\cdot}13 \pm 0{\cdot}092$; $N = 1247$.

* This experiment could not be completed because the AR + ♀ × CH + ♂ crosses stopped producing females; an attempt is now being made to analyse the reason for this aberrant sex ratio. The statistics presented in row 9 therefore are based upon about half as many observed matings as in all the other rows, and no tallies for 'early matings' were included.

dependent results. The males representing the rare genotype enjoy an advantage in matings.

The magnitude and time of expression of this advantage, however, is influenced by a maternal factor. Note that rows 7 and 10 differ only in the source of cytoplasm —that is, in the mother—of the heterozygote. Yet the chi-squares calculated for both the early and the total matings for the males are significant in one instance (row 7), and not for the other (row 10). There are similar differences to be seen if rows 9 and 12 are compared, as well as if early matings are considered, for instance, in rows 1 and 4, 2 and 5, and 3 and 6.

'I', the isolation index, and its standard error were calculated for all the 10:10 ratios, the only ratio to which it is applicable according to the recommendation of Malogolowkin-Cohen, Simmons & Levene (1965). Random mating would give a zero index, and complete isolation one of unity. A negative isolation index would indicate a preference for mating between unlikes. However, none of the indices calculated for these strains shows a strong isolation. The sexual isolation between strains used here is not an important factor influencing the mating performance.

The data for the set of P strains show similar frequency-dependent results (Table 2). However, here the four tests of equal numbers (rows 1, 4, 7 and 10) indicate in all cases advantage held by the heterozygous males, AR/CH and CH/AR. This difference in behaviour between the P and N strains is presumably due to their different selection histories; they have been maintained separately for seven years. All chi-squares are significant as they also are when only 'early matings' are considered.

SUMMARY

Matings of *Drosophila pseudoobscura* were observed in Elens-Wattiaux chambers, using individuals of three karyotypes: AR/AR, AR/CH and CH/CH. In each chamber two karyotypes were represented, with frequencies 10:10, 2:18 or 18:2. The males of the heterokaryotype AR/CH tend to have an advantage in mating compared to the homokaryotypes, provided that both kinds of males are equally frequent, i.e. the ratio 10:10. This advantage is further increased when the heterokaryotype is a minority, the ratio 2:18. When the homokaryotype, AR/AR or CH/CH, is a minority (18:2) it is equally or more successful than the heterokaryotype in securing mates. Among females, the mating success is independent of frequency, or the minority females have sometimes only a slight advantage.

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