Genotypic Control of Mating Times in *Drosophila melanogaster*

Many experiments have been carried out on behavioural differences between strains of mammals and insects[1,2], but few of these studies lend themselves to a complete analysis of the components of variance (additive genetic, dominance and environmental) controlling the character under analysis, which is necessary to accurately assess the heritability of the character[3]. For this reason, it was decided to take a behavioural character in *Drosophila* of obvious selective significance, namely the time taken for pairs of previously unmated flies to mate.

The experimental procedure consisted of storing unmated males and females singly in tubes until they were 6 days old, then a single male was shaken in with a single female and observed until copulation began. The time in minutes for this to occur was then recorded. Those not mating in 40 min were recorded as unmated. The stocks used consisted of 6 inbred lines built up by brother-sister mating for at least 100 generations. Data were collected from the 6 inbred lines, the 15 possible crosses between the inbred lines and the 15 reciprocal crosses, thus making up a complete diallel cross. For each of the 36 crosses, 14 trials were carried out split into two replicates of 7.

Table I(a) gives the mean number of matings out of 7 occurring in <11 min for the 36 crosses. In Table I(b), the means for the *hybrids* of a given inbred line as female parent crossed with all other inbred lines, and as male parent crossed with all other inbred lines, are compared with the means for the appropriate inbred lines. Except for line N2, there is considerable hybrid vigour in the frequency of mating in <11 min especially for the OR line. The overall difference between the hybrids and inbreds is significant ($P < 0.01$). Since the data consist of proportions of matings out of a fixed total, it is more correct to apply the angular transformation[4] before carrying out tests of significance, since then the variances are independent of the means and merely depend on the sample size (7) on which each observation is based. After transformation, the difference between the hybrids and inbreds remains significant ($P < 0.02$).

An analysis of variance (Table II) can be done on the data. In this analysis, which was carried out by the methods of GRIFFING[5,6] after applying the angular transformation, the inbred lines were omitted, so that unbiassed estimates of the population components of variance could be obtained. The analysis shows the general combining ability component to be highly significant. Inspection of the means in Table I(b) shows this to be mainly due to the line OR having a better, and N1 a poorer combining ability than the other lines. The variance for specific combining ability is also significant. In particular, the hybrids between lines N1 and N2 mated less frequently than would be expected on the basis of their general combining abilities. There were also significant reciprocal differences. Hybrids between N2 and Y1, and between G5 and Y2 showed large reciprocal differences.

Based on the mean squares it is possible to obtain variances for the general combining ability ($\sigma^2_{g.c.a.}$) and specific combining ability ($\sigma^2_{s.c.a.}$), and hence the additive genetic variance (σ^2_A) and dominance variance (σ^2_D)[5,6]. The analysis gave $\sigma^2_A = 513$, $\sigma^2_D = 169$, and the environmental variance $\sigma^2_E = 166$ (Table II), so giving the heritability $h^2 = (\sigma^2_A/\sigma^2_A + \sigma^2_D + \sigma^2_E) = 0.605$, showing the very great importance of independently acting genes in determining the mating frequency. It is interesting that when the analysis was repeated on the results for <21 min, $h^2 = 0.510$ and for <41 min, $h^2 = 0.342$. This progressive reduction in h^2 with time was due to σ^2_A decreasing as σ^2_D increased, the value for σ^2_E not changing much. Thus initially, $\sigma^2_{g.c.a.}$ and σ^2_A are high, so that indepen-

Table I. The numbers of successful matings out of 7 occurring in less than 11 min

(a) Mean numbers mating (based on two replicates)

Female parent	Male parent N1	N2	Y1	Y2	G5	OR
N1	1.5	2.5	3	0	2	4.5
N2	3	4.5	1	2.5	2.5	5.5
Y1	2.5	4.5	2	3.5	3.5	6.5
Y2	1	5	2.5	0.5	4	7
G5	4.5	1.5	2.5	0.5	1.5	4.5
OR	6.5	4	4	5.5	6	1

(b) Mean numbers for hybrids and inbred lines

Inbred line	Hybrids: Female parent crossed with the 5 other stocks	Hybrids: Male parent crossed with the 5 other stocks	Inbred lines
N1	2.4	3.5	1.5
N2	2.9	3.5	4.5
Y1	4.1	2.6	2
Y2	3.9	2.4	0.5
G5	2.7	3.6	1.5
OR	5.2	5.6	1
Overall mean	3.53	3.53	1.83

Table II. Analysis of variance of the diallel cross

	d.f.	Mean square	F	P
General combining ability (g.c.a.)	5	2554.805	13.42	<0.001
Specific combining ability (s.c.a.)	9	504.129	3.04	<0.05
Reciprocal effects	15	499.582	3.02	<0.01
Error	30	165.670		

$\sigma^2_{g.c.a.} = 256.334$, $\sigma^2_{s.c.a.} = \sigma^2_D = 169.230$, $2\sigma^2_{g.c.a.} = \sigma^2_A = 512.668$, $\sigma^2_E = 165.670$.

1 P. L. BROADHURST, *Experiments in Personality*, Ed. H. J. Eysenck (Routledge and Kegan Paul, London 1960), vol. I, p. 1.

2 J. L. FULLER and W. R. THOMPSON, *Behavior Genetics* (John Wiley, New York and London 1960).

3 P. L. BROADHURST and J. L. JINKS, J. Hered. *54*, 170 (1963).

4 R. A. FISHER, Heredity *3*, 229 (1949).

5 B. GRIFFING, Heredity *10*, 31 (1956).

6 B. GRIFFING, Aust. J. biol. Sci. *9*, 463 (1956).

dently acting genes are important; however, with time $\sigma^2_{s.c.a.} = \sigma^2_D$ increases, so that *specific* crosses and hence specific gene combinations become progressively more important.

The other temporal variation observed was the progressive reduction in the discrepancy between the inbreds and hybrids with time, i.e. the inbreds, although taking longer to mate, eventually tend to catch up with the hybrids.

In conclusion, therefore, the genotype exerts an extremely important influence on the time flies take to mate. Since this must be an important component of fitness, any variations in times occurring in the wild must be of evolutionary significance.

Résumé. Chez quelques lignées pures et hybrides de *Drosophila melanogaster* l'accouplement sexuel s'effectue avec des vitesses variables. On peut conclure que la constitution génotypique exerce une action très importante sur la vitesse de l'accouplement.

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A Biologically Active Analogue of Oxytocin not Containing a Disulfide Group

A conspicuous structural feature in the molecule of oxytocin (I, $X = S$, $R = NH_2$) and related neurohypophysial peptides is the presence of a twenty-membered ring incorporating the disulfide bond of a cystine residue[1,2]. It early appeared that this structural feature was necessary for biological activity since desulfuration of oxytocin[3,4] or its reduction followed by alkylation[5,6] gave biologically inactive products[7]. Recent work[8] has confirmed early claims[9,10] that the reduced form of oxytocin, now known to be the acyclic cysteine peptide, has no uterotonic activity. The results of studies on a range of synthetic oxytocin analogues, as recently summarized and discussed by JARVIS and DU VIGNEAUD[11], have shown that any change in the size of the ring incorporating the disulfide group led to a sharp decrease, or complete disappearance, of biological activity. Such an effect might be ascribed either generally to a change in the topochemistry of the molecule, resulting from the change in ring size, or more specifically to its influence on the reactivity of the disulfide bond. SCHWARTZ, RASMUSSEN et al.[12], in a series of studies, established a correlation between the biological effects of oxytocin and vasopressin on the kidney and toad bladder, and the amount of radioactivity bound to the target tissue after administration of the labelled hormone and released from it by treatment with thiols. It was plausibly suggested[12] that at any rate one of the molecular events leading to the manifestation of biological activity involved reaction of the disulfide group of the hormone with sulfhydryl groups on the receptor with formation of a hormone-receptor disulfide bond.

In order to establish unequivocally whether the role of the disulfide bond in the biologically active molecules is essentially chemical, i.e. connected with its special reactivity, or purely structural, an analogue would be required which would closely resemble one of the active hormones in its steric and hence topochemical properties but would lack the reactive disulfide bond. A structure with one of the sulfur atoms of the disulfide group replaced by a methylene would meet this requirement since the change from S-S to CH_2-S can be regarded as approximately isosteric. We have now completed the synthesis of such a 'carba' analogue in the oxytocin series. For greater preparative convenience we chose in the first instance to synthesize a derivative lacking the terminal amino group (I, $X = CH_2$, $R = H$); this appeared justified as DU VIGNEAUD et al.[13–15] had demonstrated that this amino group is not essential for biological activity.

The protected octapeptide amide III, obtained as the hemihydrate, m.p. 214–217°, from the heptapeptide amide[14,16] II and benzyloxycarbonyltyrosine azide[17], was reduced with sodium in liquid ammonia and alkylated *in situ* with tert-butyl 4-iodobutyrate. The crude reaction product was treated with acid to split the tert-butyl ester grouping and the S-(3-carboxypropyl)octapeptide amide

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