VARIATION IN MALE MATING ABILITY IN *Drosophila melanogaster*
AND ITS
GENETIC CONSEQUENCES

By

J. M. ROSADO

Lic.Vet., Lisbon

B.Sc., Lisbon

Submitted to the University of Edinburgh as a Thesis in fulfilment of the requirements for the degree of Doctor of Philosophy.

Institute of Animal Genetics
University of Edinburgh
October, 1959

LIST OF CONTENTS

Page

INTRODUCTION 1

REVIEW OF LITERATURE 5

METHODS

1. Theoretical Approach 14

1.1. The Components of Fitness 14

1.2. The Measurement of Fitness Components 15

1.3. The Combination of Fitness Components 24

1.4. Equilibrium of Gene Frequencies for a sex-linked gene 24

2. Experimental 29

2.1. Material 29

2.2. Results 30

2.2.1. Viability and Fertility 30

2.2.2. Male Mating Ability 39

2.2.3. Observed and Expected Evolution of Genotypic Frequencies 46

2.2.4. Change of Fitness by Inbreeding 53

SUMMARY 60

REFERENCES 62

ACKNOWLEDGEMENTS 65

INTRODUCTION

The importance of the mating system as an evolutionary factor has been recognised by many workers, including Darwin (1871), Haldane (1932), and Huxley (1942).

In fact, when genetic differences in mating ability exist among individuals, the genes leading to more successful matings will tend to spread in the population.

In *Drosophila melanogaster*, a variety of mutant male genotypes which are less successful in mating than the wild type is known, e.g. cut, raspberry, vestigial, white and yellow.

The aim of the present work is to predict changes of gene frequencies in experimental populations due to the sex-linked gene, yellow.

Since mating ability is only one of the forces of selection, effects caused by all other forces should be taken into account, hence total fitness should be measured.

By fitness we mean the "average ability to pass on the genes to the next generation" (Knight and Robertson, 1957). Any measure of fitness is then a measure of the representation/

representation of genes in the next generation.

If a population consists of different classes, their frequency distribution will change with time unless the relative increase in numbers of all classes is the same. This relative increase was proposed by Fisher (1930) as a measure of fitness - which he called the "Malthusian parameter of population increase". When the constituting classes are genotypes, each defined by a Malthusian parameter, m, the distribution of gene frequencies in populations can be predicted, (Crow and Kimura, 1955).

In what follows, an alternative measure of fitness will be used, the quantity w defined by Sewall Wright (1949) as the selective value of a given type of zygote, (fertilised egg), assumed to be measured by its average contribution under the prevailing conditions to the array of zygotes produced a generation later in such a way that w is the ratio of the effective size of the population in the following generation to that under consideration.

The Malthusian parameter, m, and the Sewall Wright, w, are related as $w = e^m$ (Haldane, 1932).

For a random mating population the change in frequency/

frequency of autosomal genes, q, and the average fitness, w, are, for diploids, related by Sewall Wright's (1942) formula,

$$\Delta q = \frac{q(1-q)}{2\bar{w}} \frac{d\bar{w}}{dq} \qquad (1)$$

This formula assumes that the selective values are independent of the sex, or, in other words, the same genotype has the same relative fitness whether male or female. This can be true for the case in which the viability to maturity is the unique fitness component (see later) and it can be fairly assumed that for autosomal genes it will be so. But if selection acts through differential fertility, this cannot be assumed "a priori", as fertility in the two sexes is a completely different biological process. In this case the above formula constitutes only a good approximation when the above coefficients are small. In the case of sex-linked genes two formulae should be used, one for each sex. But if the selective coefficients are small, the formula (Wright, 1942),

$$\Delta q = \frac{q(1-q)}{\frac{2}{3}\bar{w}} \frac{d\bar{w}}{dq} \qquad (2)$$

where w is the geometric mean of the average fitness in the/

the two sexes gives a good approximation.

In an experiment where changes in gene frequencies can be observed, it is possible to calculate the w's (see Reed and Reed, 1942; Kerr and Wright, 1954^{a}, 1954^{b}; Wright and Kerr, 1954).

The measurement of total fitness can be attempted by breaking it down into several components, measuring each separately and assembling them in proper order. In the present work the measurement of components, namely, viability, fertility and mating ability is attempted. Also, inbreeding is used to modify the genetic background hoping to cause changes in the selective coefficients.

REVIEW OF LITERATURE

It has been shown in Drosophila that yellow males mate more frequently with yellow than with wild type females. Also, the wild type males are more successful than the yellow ones in mating with either yellow or wild type females (Sturtevant, 1915; Diedrich, 1941; Merrell, 1949; Koref-Santibanez and Waddington, 1958).

Merrell (1949) conducted "female preference" experiments where wild type and yellow males competed for mating with wild type or yellow females. He showed that no difference existed in the proportions of females inseminated by yellow and wild type males when the experiments were performed either in the darkness or in light. Bastock, (1956), in similar experiments, was unable to show that the females, after the removal of their antennae, received the yellow males more readily. Rather it was found that the vibration of the wings is given by the yellow males in shorter bouts and at longer intervals. Bastock suggested that feeble courtship behaviour of the yellow males fails to induce mating response in the females.

The lesser success of the yellow males in mating when in competition with wild type has the immediate result of reducing the frequency of the yellow gene in a population, other things being equal.

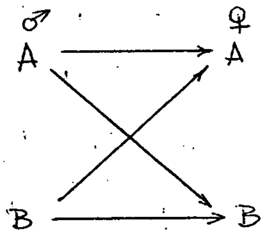
An attempt to measure the degree of selective mating and to use it to predict the change in gene frequency due to this effect was made by Merrell (1949, 1950, 1953). This author (1950) defined as "female mating ratio"the ratio of matings performed by one kind of female to the matings performed by the other kind; a "male mating ratio" is defined analogously. Using these ratios, with the Drosophila melanogaster mutants cut, raspberry, forked and yellow against wild type, he was able to predict the change in frequencies of experimental populations. The experimental values for cut and yellow obtained by Ludwin (1951) agree fairly well with the values calculated by Merrell (1953).

A similar attempt was made by Reed and Reed (1950) using the white mutant of Drosophila melanogaster. They found that the white mutants mated 25 per cent less frequently than the wild type. This was in agreement with the predicted change in gene frequency.

The female and the male mating ratios of Merrell, and their relationship with other measures of preferential mating will be discussed. In particular, the so called "isolation indices" are of interest, since these refer to the degree of sexual isolation between populations.

Consider/

Consider a situation in which two populations, A and B, coexist in the same area; the frequency of the four kinds of matings, two homogamics (AA and BB) and two heterogamics (AB and BA) determine the degree of isolation between the two groups. These matings are shown diagramatically (after Merrell, 1950),



Five kinds of experiments have been made to estimate the degree of isolation.

1. *Direct Observation*: Sturtevant (1915) observed the frequencies of mating between the two groups when put together.
2. *Pair Matings*: Patterson, Stone and Griffin, (1940), observed the proportion of matings when the four types of pairs are isolated from each other.
3. *Female Choice*: Diedrich, (1941) and Merrell, (1949) placed females of one kind with two kinds of males - holding the three classes in equal numbers, such that the males outnumber the females two to one.
4. *Male Choice*: Mayr and Dobzhansky (1945), Dobzhansky and Streisinger (1948), Patterson, McDonald and Stone (1947) and Merrell (1949) placed one kind of male with two/

two types of females. Records were kept of the first female to mate.

5. Multiple Choice: Reed and Reed (1950) placed all four types - two male and the two female types together. The proportions of the four types of matings were estimated from an examination of the the progeny.

For the measurement of sexual isolation, various indices have been proposed. Stalker (1942) proposed an "Isolation Index" which is defined as the ratio of the difference between homogamic and heterogamic matings to their sum in the "male choice" experiments. He used it to measure the sexual isolation between strains of Drosophila virilis.

Levene and Dobzhansky (1945), defined an "Isolation Ratio" which is the ratio of the probabilities (assumed constant) of homogamic and heterogamic inseminations in "male choice" experiments.

Bateman (1949), believing that the choice in mating is made by the female, distinguished between "sexual isolation" (dependent on the ability of the female to recognise the males of their own species) and "mating propensity" of the female, (the facility with which females become sexually excited). He proposed a measure of/

of "true" sexual isolation, (joint isolation indices), given by the average of the two complementary indices of Stalker, one for the mating (A + B)♀ x A♂ and the other for (A + B)♀ x B♂. The mating propensities are measured by half the difference of these indices.

Levene (1949), discussed Stalker's index and the Bateman "joint isolation index" derived from it, pointing out that neither takes account of the time taken before mating ocurred. In fact, if p_{11} and p_{12} are the progeny of females belonging respectively to the strains 1 and 2 inseminated by males of the strain 1, as time goes on both p_{11} and p_{12} tend to 1 and thus the mating propensity coefficient tends to zero. The disadvantage is still more marked if the joint coefficient is used. In this case, there are two complementary (in Bateman terms) or reciprocal (in Levene terms) coefficients whose average gives the joint index of Bateman. Suppose a situation where no isolation exists; the joint coefficient should then be computed as zero, but as the rate of insemination in the two reciprocal experiments may be different, a non-zero index could occur.

Levene then proposed a further index of isolation which was less affected by the time factor. He found with Levene and Dobzhansky (1945) that the probability of insemination in male choice experiments was constant and/

and was thus able to develop "coefficients of isolation" K_{12} and K_{21} for the two reciprocal crosses and a "joint coefficient" as the average of the two: both are functions of the probabilities only.

$$K_{12} = \frac{1-h_{12}}{1+h_{12}} \quad \text{and} \quad K_{21} = \frac{1-h_{21}}{1+h_{21}} \qquad (3 \text{ and } 3')$$

where h_{12} and h_{21} are respectively the ratios of the two probabilities for each reciprocal cross. From these Levene derived also a coefficient of "excess of insemination" of one strain over another. This was given by one half the difference between K_{12} and K_{21} (analogous to Bateman's "mating propensity" coefficient).

Levene's "coefficient of isolation" seems to give the most accurate estimate of a situation.

Nevertheless, as Merrell (1950) pointed out, any analysis of data from mating experiments should separate the effects of sexual isolation from those of selective mating, which the averaging of two coefficients obscures. He then proposed two measurements, one for the "sexual isolation" and another for the "mating propensity" As the latter measures the strength of selection for one type or another, it can be used to predict the changes in gene frequency due to selective mating. They are the "female mating ratio" and the "male mating ratio" referred

to/

to above.

He defined an "isolation estimate", which is the ratio of the heterogamic over the homogamic matings. Since the mating frequencies of both types of females and males appear in the numerator and in the denominator, the "mating propensities" cancel out, thus the "isolation estimate" is independent of the "mating propensity". On the other hand, assuming again with Levene (1949) that the probability of insemination of a particular cross is constant, the "isolation estimate" remains independent of the time lapse to mating.

The female and male mating ratios referred to above and used by Merrell as a measure of mating propensities and in a later paper (1953) to predict the change in gene frequency due to selective mating are defined respectively by

$$M_f = \frac{AA + BA}{BB + AB} \quad \text{and} \quad M_m = \frac{AA + AB}{BB + BA} \qquad (4 \text{ and } 4')$$

where AA, BB, AB and BA are the mating types defined above.

M_f and M_m are, then, respectively the ratios of matings performed by one type over the matings performed by/

by the other, for females and males, when the types to be tested are in equal numbers. Or in other words, they represent the actual selective mating proportions of the two types of females and males; they can thus be used to predict changes in gene frequency if the two types differ by one allele responsible for the mating differences.

Nevertheless, the mating ratios, when used to predict changes in gene frequency have the following dificiences:-

1. They must be calculated when the relative proportions of the two types to be tested are equal.
2. They assume the ratio to be constant for all genotypic frequencies in a population. This is not obviously so.
3. They assume a constant probability of mating for each cross, a fact which was not verified experimentally.

The most recent approach to this problem was that of Petit (1959). She proposed a "coefficient de selection sexuelle", K, given by

$$K = \frac{A/a}{B/b} \qquad (5)$$

where A and B are respectively the number of females inseminated by respectively <u>a</u> males of one kind and <u>b</u> males of the other.

This/

This "coefficient" was not proposed to predict changes in gene frequency, but for the study of the variations of the coefficient itself with changes in the genetic background.

Nevertheless, it overcomes the first of the objections raised above. On the other hand, the variations in K Petit observed might still be explained, at least in part, by the "one sided mating propensities" referred to by Dobzhansky (1944). An example is the situation where the A males mate more readily with A than with B females but where B males mate at random with both female types.

It can be seen from the above discussion that the measurement of fitness has been an extremely complex problem and that the introduction of many systems of measurement, each with a different terminology, has made any study in this field very tedious.

Nevertheless, since no system proposed to date is completely adequate, a further system will be introduced below in an attempt to overcome the remaining deficiences referred to above.

METHODS

1. THEORETICAL APPROACH

1.1. The Components of Fitness

The life cycle of a dioecious organism suggests the breaking down of total fitness into Viability and Fertility. Since a particular genotype can be less fertile than another, either because it mates less and/or because it produces less gametes, it is convenient to divide the fertility component into zygotic fertility (mating ability) and gametic fertility. Thus, viability is concerned with the survival of genotypes after formation of the zygotes (developmental component), whereas zygotic fertility deals with the ability of genotypes to secure mating (behaviour component) and gametic fertility measures the production of gametes (physiological component). Each of these components represents a different biological process. It is possible that the gametes produced by the same parent but with different gene content could differ in their ability to fertilise.

If this were so, the component would be included in the estimates of viabilities and those of gametic fertilities and thus counted twice. But, as only one case of this type of selection is known in Drosophila/

Drosophila (Wallace, 1948) it will be assumed it does not exist at all.

1.2. The Measurement of Fitness Components

Differential viability between two genotypes is measured as the proportion of mature individuals of one genotype in relation to that of the other, whereby each genotype is weighted by its zygotic frequency; if p and q are the zygotic frequencies and p' and q' the frequencies of genotypes at maturity, the viability component, w, is given by,

$$p'/q' = w\,p/q \qquad (6)$$

in accordance with Wright's (1949) definition. Similarly, differential gametic fertility is measured as the proportion of gametes produced by one genotype in relation to that of the other, weighted by respective gene frequencies at mating.

Maximum likelihood estimates of w coefficients are obtained simultaneously for viability and fertility by attributing coefficients of fertility to parents and of viability to the progeny. Tables 1 and 2 were set up respectively, for female and for male progeny, as follows:-

TABLE 1. Parent-Offspring Table for Female Progeny

		Male Parents				
		A 1		a $(1-\varepsilon)$		
	Female Progeny	AA $(1-\omega)$	Aa 1	Aa 1	aa $(1-\theta)$	
Female Parents	AA $(1-\phi)$	$(1-\phi)(1-\omega)$ a_{11}		$(1-\phi)(1-\varepsilon)$ a_{21}		$a_{11}+a_{21}=a$
	Aa 1	$\frac{1}{2}(1-\omega)$ b_{11}	$\frac{1}{2}$ b_{12}	$\frac{1}{2}(1-\varepsilon)$ b_{21}	$\frac{1}{2}(1-\varepsilon)(1-\theta)$ b_{22}	$b_{11}+b_{12}+b_{21}+b_{22}=b$
	aa $(1-\psi)$		$(1-\psi)$ c_{12}		$(1-\varepsilon)(1-\psi)(1-\theta$ c_{22}	$c_{12}+c_{22}=c$
	Totals	$a_{11}+b_{11}=n_{11}$	$b_{12}+c_{12}=n_{12}$	$a_{21}+b_{21}=n_{21}$	$b_{22}+c_{22}=n_{22}$	$a+b+c=n$

$(1-\phi)$: coefficient of fertility of the AA females

$(1-\psi)$: " " " " " aa "

$(1-\varepsilon)$: " " " " " a males

$(1-\omega)$: " " viability " " AA females

$(1-\theta)$: " " " " " aa "

$(1-\phi)(1-\omega)$, $(1-\phi)(1-\varepsilon)$, etc. : expected frequencies of daughters genotypes

a_{11}, a_{12}, etc. : numbers of each genotype observed in an experiment with the same number of replicates per type of mating

TABLE 2. Parent-Offspring Table for Male Progeny

		Male Parents				
		A 1		a $(1-\varepsilon)$		
	Male Progeny	A 1	a $(1-\eta)$	A 1	a $(1-\eta)$	
Female Parents	AA $(1-\phi)$	$(1-\phi)$ a_{11}		$(1-\phi)(1-\varepsilon)$ a_{22}		$a_{11}+a_{21}=a$
	Aa 1	$\frac{1}{2}$ b_{11}	$\frac{1}{2}(1-\eta)$ b_{12}	$\frac{1}{2}(1-\varepsilon)$ b_{21}	$\frac{1}{2}(1-\varepsilon)(1-\eta)$ b_{22}	$b_{11}+b_{12}+b_{21}$ $b_{22}=b$
	aa $(1-\psi)$		$(1-\psi)(1-\eta)$ c_{12}		$(1-\psi)(1-\varepsilon)(1-\eta)$ c_{22}	$c_{12}+c_{22}=c$
	Totals	$a_{11}+b_{12}=h_{11}$	$b_{12}+c_{12}=h_{12}$	$a_{21}+b_{21}=h_{21}$	$b_{22}+c_{22}=n_{22}$	$a+b+c=n$

$$\left.\begin{array}{l}(1-\phi)\\(1-\psi)\\(1-\varepsilon)\end{array}\right\} : \text{ as in Table 1}$$

$(1-\eta)$: coefficient of viability of the a males

$(1-\phi)$ $(1-\eta)$, $(1-\varepsilon)$, etc., and a_{11}, a_{12}, etc. : as in Table 1

The likelihood equation for female progeny is:-

$$L = a\log(1-\phi) + C\log(1-\psi) + n_{11}\log(1-\omega) + n_{22}\log(1-\theta) + (n_{21}+n_{22})\log(1-\varepsilon) - n\log\left\{\left(\tfrac{3}{2}-\phi\right)\left[(1-\omega) + (1-\varepsilon)\right] + \left(\tfrac{3}{2}-\psi\right)\left[1+(1-\theta)(1-\varepsilon)\right]\right\}. \qquad (7)$$

with the solution, (8)

$$(1-\phi) = \frac{a}{2(b_{11}+b_{21})}, \qquad (1-\psi) = \frac{C}{2(b_{12}+b_{22})},$$

$$(1-\varepsilon) = \frac{n_{21}\left(\frac{3}{2}-\psi\right)}{n_{12}\left(\frac{3}{2}-\phi\right)}, \qquad (1-\theta) = \frac{n_{22}\left(\frac{3}{2}-\phi\right)}{n_{21}\left(\frac{3}{2}-\psi\right)},$$

and $$(1-\omega) = \frac{n_{11}\left(\frac{3}{2}-\psi\right)}{n_{12}\left(\frac{3}{2}-\phi\right)}.$$

The likelihood equation for the male progeny is,

$$L = a\log(1-\phi) + b\log(1-\psi) + (n_{12}+n_{22})\log(1-\gamma) + (n_{21}+n_{22})\log(1-\varepsilon) - \eta\log(2-\varepsilon)\left[\left(\frac{3}{2}-\phi\right) + \left(\frac{3}{2}-\psi\right)(2-\gamma)\right]. \qquad (9)$$

with the solution, (10)

$$(1-\phi) = \frac{a}{2(b_{11}+b_{21})}, \qquad (1-\psi) = \frac{c}{2(b_{12}+b_{22})},$$

$$(1-\varepsilon) = \frac{n_{21}+n_{22}}{n_{11}+n_{12}} \quad \text{and} \quad (1-\gamma) = \frac{b_{12}+b_{22}}{b_{11}+b_{21}}$$

Both of these two components, viability and fertility are by their own nature independent of the presence of other genotypes in the population. Mating ability, on the contrary, may depend on genotypic composition. Furthermore, while viability is the same biological process in both sexes, mating ability in Drosophila is a completely different one and different statistical treatment is thus required. Mating ability for females means the attainment of sufficient nervous excitation, and for the males consists among/

among other things in inducing this excitation in the females.

There is strong evidence (Merrell, 1949) that Drosophila males have no preference in mating with different females and that deviations from random mating in "male choice" experiments are due to different responses of the females. Males apparently have the urge to mate indescriminately and special "stimuli" from females are not necessary to start the courting activities. The females will respond if the pattern and intensity of the courtship are adequate (Bastock, 1956). It is a well known fact that males attempt to mate with males.

In the light of these observations it may be justified to eliminate the mating ability component from the fitness of females.

If the mating ability of different male genotypes is to be measured and used to predict gene frequency changes in a population, the sex ratio of parents in the corresponding experiments should be the same.

An example will clarify this. Suppose two kinds of males a and b are used. Assume t_a and t_b to be the time necessary for each kind to stimulate the females and for t_b to be greater than t_a. If the number of females is equal to the number of males a, the females will/

will mate with these irrespective of the values of t_b. But if the number of females is greater than the number of more successful males the less successful may mate with the surplus females left over by the more successful ones if the time t_b is not greater than $2t_a$ plus Δt_a which is the time necessary for the a males to complete the mating. If $t_b > 2t_a + \Delta t_a$, the a males will mate with the second group, crowding out the b males, and so on for a third and fourth group of females. An important conclusion is that the two kinds of males with only slightly different stimulating times t_a and t_b may show no differences in mating index (random mating). It is not surprising to find the "one-sided mating frequencies" described above, because if one of the two kinds of females is more easily sexually stimulated, it will mate indiscriminately and the other, more discriminate, will mate first with the males which are able to induce the mating response in a shorter time.

It was logical to compare the mating processes with a catalytic reaction, where the virgin females represent the substrate to be irreversibly converted into inseminated females and the males represent the catalytic surface where the reaction takes place. In a situation where different kinds of females and one kind of males are present/

present, the end result will be the transformation of the females by order of the "affinities" and so one may speak of competition of the substrates and saturation of the catalytic surface. In a situation where one kind of females and different kinds of males are present, the proportion of end products (females fecundated by different male types) will depend on the affinities between the substrates and the different catalytic surfaces.

In the latter case the proportion of females fecundated by the first, second and third kind of males is given by the solution of a system of differential equations of the type,

$$\frac{dF_j}{dt} = m_j(1-F_1-F_2- \ldots F_j \ldots)M_j \qquad (11)$$

where F_1, F_2 ... F_j are the frequencies of females fecundated by the first, second and ... j^{th} kind of males, M_j is the frequencies of the different kinds of males and m_j the rate constant of each type of mating.

If at $t = 0$ all the females were virgins, the solution is,

$$F_j = \frac{m_j \; M_j}{\sum m_j \; M_j} \qquad (12)$$

But/

But noting that $\frac{m_j}{m_i} = w_j$, where m_i is the rate constant for the i kind of male and w_j the Sewall Wright selective coefficient of the j males relative to the former, F_j can be expressed in terms of relative fitness and male frequencies,

$$F_j = \frac{w_j \ M_j}{\bar{w}} \tag{13}$$

When females with different thresholds of response coexist, a different w_j should be attributed to each. In the case of a sex-linked gene there are for each female genotype two m's and thus only one w. These can be estimated as coefficients of regression, as in this case the solutions are of the type,

$$\frac{F_1}{F_2} = w \frac{M_1}{M_2} \tag{14}$$

The linearity of the regressions tests the independence of the thresholds from the genotypic frequencies in the population. For if the experimental values fit linear regressions when the genotypic frequencies of females are different in individual experiments, the constancy of the thresholds of response (the probability of insemination of Levene) is indicated.

1.3. The Combination of the Components of Fitness/

1.3. The Combination of the Components of Fitness

The proper method of combining the three components of fitness in a single expression which gives the expected change in gene frequency, depends on the point in the life cycle where the scoring of the genotype is made. If the scoring were possible at the formation of the zygotes, the expression for q would be simple as the coefficients for each component could be multiplied by each other, within genotypes. If the scoring is made at mating, the expression turns out to be more complex as total fitness, evaluated by multiplication of the coefficients, can only be used within the definition of Sewall Wright (1949). It can be derived by writing down the expected genotypic frequencies for each step of the cycle.

1.4. Equilibrium of Gene Frequencies for a Sex-linked Gene

It is interesting to know if a non-trivial stable equilibrium of gene frequency is possible for a particular set of fitness components; in this case, the components can be combined by multiplication within genotypes and the product treated as a coefficient of selection for each. The solution can be found as follows.

Because/

Because of the existence of three female AA, Aa, and aa and only two male A and a sex-linked genotypes, selection at sex-linked loci will involve different rates of change in gene frequency, Δq, for females and males. Let $(1-s_1):1:(1-s_2)$ and $1:(1-t)$ be the selective coefficients for the genotypes above, the changes in frequency are respectively,

$$\Delta q_f = -sq(1-q)\frac{s_1 q - s_2(1-q)}{1-s_1 q^2 - s_2(1-q)^2} \tag{15}$$

and

$$\Delta q_m = q(1-q)\frac{t}{1-t(1-q)} \tag{15'}$$

Random mating tends to equalise gene frequencies in both sexes, selection to establish equilibrium frequencies specific for each sex. A non-trivial equilibrium point with selection is possible for sex-linked genes if the change in gene frequencies due to selection, is exactly balanced by the change due to mating.

This implies different gene frequencies in each sex at equilibrium, except for the case where over-dominance in females coincides with no selection in males.

Around/

Around the equilibrium point let <u>f</u> and <u>m</u> be the respective gene frequencies of A in females and males, after selection has operated and before mating has taken place. Mating will change them to $\frac{1}{2}(f+m)$ and f respectively. Equilibrium is possible if selection of genotypes will restore to f and m the gene frequencies changed by mating,

$$\Delta f = f-\tfrac{1}{2}(f+m) \qquad \text{(selection } \Delta f \text{ of female genotypes)}$$

and

$$\Delta m = (m-f) \qquad \text{(selection } \Delta m \text{ of male genotypes)}$$

That is, if the changes of gene frequency due to selection among females (Δf) and males (Δm) satisfy the condition

$$2\Delta f + \Delta m = 0^{*} \tag{16}$$

It is easily seen that the difference between female and male frequencies is $-m$ after selection (before mating) and $\frac{1}{2}m$ after mating but before selection.

In order to calculate the frequencies at equilibrium which satisfy the conditions above, let x, 2y and z be the frequencies of the AA, Aa and aa female genotypes at mating. The progeny of the A and <u>a</u> genotypes in males are/

---

[*] I am indebted to Dr. Alan Robertson for this step in the derivation.

are thus $(x+y)+\Delta m$ and $(y+z)-\Delta m$. After mating, the genotypic frequencies are, by puting $(x+y) = q$,

$$\begin{array}{ll} AA & q^2+q\Delta m \\ Aa & 2q(1-q)-q\Delta m+(1-q)\Delta m \\ aa & (1-q)^2-(1-q)\Delta m \end{array}$$

Let $(1-s_1):1:(1-s_2)$ and $1:(1-t)$ be the fitness of female and male genotypes respectively. After selection the changes in frequency of the A gene among females and males are,

$$\Delta f = \frac{s_1q^2+q-(s_1q-\frac{1}{2})\Delta m}{1-s_1q^2-s_2(1-q)^2-\left[s_1q-s_2(1-q)\right]\Delta m} - (q+\tfrac{1}{2}\Delta m) \qquad (17)$$

and

$$\Delta m = \frac{q}{1-t(1-q)} - q \qquad (17')$$

By manipulation of these two expressions the change in gene frequency, Δq, in the whole population of males and females is obtained.

$$\begin{aligned} \Delta q &= \frac{1}{3}(2\Delta f+\Delta m) \\ &= \frac{2}{3}q(1-q)\frac{s_2(1-t)+\frac{1}{2}t-(s_1+s_2-s_2t)q}{\bar{w}_f\bar{w}_m} \end{aligned} \qquad (18)$$

where/

where $\bar{w}_f$ and $\bar{w}_m$ are respectively the average fitness in female and male sub-populations,

$$\bar{w}_f = 1-s_1q^2-s_2(1-q)^2+\left[s_2-(s_1+s_2)q\right]\Delta m$$

and

$$\bar{w}_m = 1-t(1-q)$$

Noting that the numerator of the above expression for Δq is equal to $\frac{d}{2dq}\,\bar{w}_f\bar{w}_m$, it becomes (compare with the approximate formula (2) page No. 3),

$$\Delta q = \frac{q(1-q)}{3\bar{w}_f\bar{w}_m}\,\frac{d}{dq}\,\bar{w}_f\bar{w}_m \qquad (19)$$

The equilibrium point is then

$$\hat{q} = \frac{s_2(1-t)+\frac{1}{2}t}{s_1+s_2-s_2t} \qquad (20)$$

and its stability can be investigated through the behaviour of the second derivatives (Li, 1955), of $\bar{w}_f\bar{w}_m$. The formula for equilibrium agrees with that of Bennett, (1957) who used a different approach for its derivation.

2. EXPERIMENTAL/

2. EXPERIMENTAL

2.1. Material

For an investigation of this kind, accurate scoring of all genotypes is crucial. The two closely linked recessive genes "Scute" (sc) and "yellow" (y) were chosen as suitable markers of the sex chromosome. The three female genotypes $\frac{sc+}{sc+}$, $\frac{sc+}{+y}$ and $\frac{+y}{+y}$ which appear phenotypically as *scute*, *wild type* and *yellow* respectively, and the two male $\underline{sc+}$ and $\underline{+y}$ which appear phenotypically as *scute* and yellow respectively constituted the experimental material. Both the *sc* and the *y* genes were introduced into the *Kaduna* stocks of the laboratory from the *Oregon K* stock. The crosses were then allowed to reproduce in a population cage for 21 weeks at a population size of 5,000 flies to allow recombination to take place. After this, scute and yellow genotypes were separated and maintained throughout the experiment in large populations. From these stocks, individuals were sampled for investigations. All experiments were carried out at a constant temperature of 25^{0}C and under usual laboratory conditions.

During the 21 week stabilising period, the population/

population was sampled at 3 week intervals and gene frequencies scored. It was observed that there was a steady increase in the frequency of *sc*. Experimental observations on changes in gene frequency were carried out in population bottles. This made possible a greater number of replicates than with cages. Also the generation interval was shorter and complications due to overlapping generations and differential longevity were avoided.

2.2. Results

2.2.1. Viability and Fertility

The standard proceedure was to pick 60 scute males and 60 yellow virgin females out of the scute and yellow stocks, to distribute the 60 pairs into 3 population bottles of 20 pairs each, and to divide the F_1 wild type, $\frac{sc+}{+y}$ daughters into two groups. One of these was then crossed with *sc+* and the other with *+y* males; the flies used to measure viability and fertility components were the F_2 progenies of these matings.

Two procedures were followed. In a first series of experiments, pairs of flies constituting six possible types of matings were put in vials with food during

24 hours/

24 hours. They were then transfered daily to fresh ones during 3 - 6 days. When all the progeny had emerged, they were scored for genotypes and counted. It was observed that a great number of matings with the yellow males did not take. Hence, in a second series of experiments, it was decided to let all flies from each type of cross mate together *en masse* for one day, (in some experiments the males outnumbered the females). Each female was then transfered to a single vial.

Unfortunately, maximum likelihood estimates of fertility and viability, given on pages 18 and 19, can only be applied when the numbers of individuals a, b, and c observed are each obtained from the same number of mated females. In this case the estimates will not include mating effects. Since a great majority of matings with yellow males did not take, it was necessary to employ a simple method of analysis as will be described below.

Estimation of Differential Viabilities

Over 150 individual segregations observed in 7 experiments were used to estimate viabilities. Results of $\frac{sc+}{+y}$ x $\underline{sc+}$ matings are shown in *Table 3*, and those of $\frac{sc+}{+y}$ x $\underline{+y}$ matings in *Table 4*.

It/

TABLE 3. Total Numbers of Each Genotype Resulting from $\frac{sc+}{+y} \times \underline{sc+}$ Matings

The upper and the lower χ^2 are respectively the χ^2 for heterogeneity within experiments and that for deviation from 1:1. When significant to 5% level marked with *

		Males			Females		
Experiment No.	No. of females	$\underline{sc+}$	$\underline{+y}$	χ^2	$\frac{sc+}{sc+}$	$\frac{sc+}{+y}$	χ^2
1	5	768	755	10.329* —	984	986	2.898 .002
2	10	731	725	4.384 .025	730	777	6.097 1.466
3	15	619	604	11.430 .184	596	590	18.412 .030
4	20	642	661	11.790 .277	637	721	19.000 5.196*
5	20	995	1050	14.519 1.479	1085	1079	13.410 .017
6	18	328	305	15.533 .836	334	362	10.298 1.126
7	19	801	778	24.222 .335	792	772	13.842 .143
Totals	107	4884	4878		5158	5287	

TABLE 4. Total Numbers of Each Genotype Resulting from $\frac{sc+}{+y} \times \underline{+y}$ Matings

The upper and lower χ^2 are respectively the χ^2 for heterogeneity within experiments and the χ^2 for the deviation from 1:1 ratio. When significant at the 5% level the χ^2's are marked with *

		Males			Females		
Experiment No.	No. of females	$\underline{sc+}$	$\underline{+y}$	χ^2	$\frac{sc+}{+y}$	$\frac{+y}{+y}$	χ^2
1	2	320	357	.088 .415	370	340	.154 1.268
2	3	245	264	.118 .709	225	257	1.514 2.124
3	9	374	388	9.985 .257	385	364	13.059 .389
4	16	704	623	10.392 4.944*	662	654	19.689 .049
5	14	482	455	8.333 .778	482	484	17.439 .004
6	13	281	267	6.505 1.552	233	245	8.119 .018
7	8	351	372	9.952 1.430	354	357	10.757 .013
Totals	65	2757	2726		2711	2701	

It was concluded that there was no differential viability between genotypes, male or female.

From Tables 3 and 4 it can be seen that significant heterogeneity of results only appeared in one experiment, and there were two cases of significant derivation from a 1:1 ratio.

Estimation of Differential Fertilities

As no differential viabilities were found among genotypes or sexes, no correction is needed for the fertilities calculated from the total number of offspring per female.

Fertility of female genotypes was estimated from the relative numbers of offspring. The calculations were based on experiments where the number of offspring produced by each pair was not limited by shortage of space or food. To check this, controls with 5 - 10 pairs per vial were run parallel to the experiment; they provided an estimate of the maximum number of flies possible within vials in a given experiment.

The result of six experiments are shown in Table 5 Obviously no conclusions about differential fertilities among the female genotypes can be drawn from these experiments. In two experiments (2 and 6) the wild type was the more fertile genotype; in the others it was about/

TABLE 5. Total Numbers of Offspring Produced by $\frac{sc+}{sc+}$, $\frac{sc+}{+y}$ and $\frac{+y}{+y}$ Females Mated with $\underline{sc+}$ Males

The% below the numbers of offspring produced by $\frac{sc+}{sc+}$ and $\frac{+y}{+y}$ females are the deviations from the 1:1 ratio, when those numbers are compared with the number of offspring produced by $\frac{sc+}{+y}$ females.

Experiment No.	No. of replicates	$\frac{sc+}{sc+}$	$\frac{sc+}{+y}$	$\frac{+y}{+y}$	χ^2 for the deviation from 1:1:1 ratio
1	10	2882 -3%	2963	3164 +7%	14
2	15	2331 -7%	2503	2060 -18%	44
3	20	3529 +34%	2641	1866 -29%	>100
4	20	3350 -20%	4197	4930 +17%	>100
5	18	1214 +6%	1145	1487 +30%	51
6	19	2805 -13%	3215	2492 -22%	92
Totals	102	16111	16664	15999	

TABLE 6. Total Number of Offspring Produced by $\frac{sc+}{sc+}$, $\frac{sc+}{+y}$ and $\frac{+y}{+y}$ Females, Mated With $\underline{sc+}$ Males, in the 1st and 2nd day of Production

The % below the numbers of offspring produced by $\frac{sc+}{sc+}$ and $\frac{+y}{+y}$ females are the deviations from the 1:1 ratio, when those numbers are compared with the number of offspring produced by $\frac{sc+}{+y}$ females.

Experiment No.	No. of replicates	$\frac{sc+}{sc+}$	$\frac{sc+}{+y}$	$\frac{+y}{+y}$	χ^2 for the deviation from 1:1:1 ratio
1	10	976 -2%	979	1077 +10%	6.534
2	15	2331 -7%	2503	2060 -18%	44
3	20	1934 +19%	1623	959 -59%	> 100
4	20	1459 -16%	1740	2370 +36%	> 100
5	18	598 +15%	518	798 +54%	65
6	19	2805 -13%	3215	2492 -22%	92
Totals	102	10103	10578	9756	

TABLE 7. Total Numbers of Offspring Produced by $\frac{sc+}{sc+}$, $\frac{sc+}{+y}$ and $\frac{+y}{+y}$ Females, Mated with $\underline{sc+}$ Males, After the 2nd day of production

The % below the numbers of offspring produced by $\frac{sc+}{sc+}$ and $\frac{+y}{+y}$ females are the deviations from the 1:1 ratio, when those numbers are compared with the number of offspring produced by $\frac{sc+}{+y}$ females.

Experiment No.	No. of replicates	$\frac{sc+}{sc+}$	$\frac{sc+}{+y}$	$\frac{+y}{+y}$	χ^2 for the deviation from 1:1:1 ratio
1	10	1906 -4%	1984	2087 +5%	8.274
3	20	1595 +57%	1018	907 -11%	>100
4	20	1891 -23%	2457	2560 +4%	>100
5	18	616 -2%	627	689 +10%	4.810
Totals	68	6008	6086	6243	

about intermediate in fertility. Scute females showed higher fecundity in experiment 3 and the yellow females in experiment 4. The χ^2 for heterogeneity between experiments was very highly significant.

The above results suggest that the difference in fertility and their variation from experiment to experiment were mainly non-genetic. The age of the flies at the beginning of observation could have been an important factor. For, if the flies of one genotype were younger than the other, not only would the average number of offspring produced by the former be less, but also the coefficient of variation would exceed that of the older flies. However, after the third day of laying, the number of offspring per female is fairly constant up to the 6th day and then declines, (Robertson and Sang, 1944). Thus it should be permissible to compare females between 3 - 6 days of age. The difference in age between flies in these experiments were never greater than 12 hours, and when progenies of the first days' egg production were discarded, the pattern of results was not altered. A breakdown of the first three days of lay versus those produced during the 4 - 6 days is given in Tables 6 and 7.

These results suggest that there was no clear relationship between age and the differential fertilities of genotypes: the results are, in fact, impossible to interpret.

2.2.2. Male Mating Ability/

2.2.2. Male Mating Ability

The standard procedure was to pick 60 scute males and 60 virgin females from the stocks; to distribute them in 3 bottles with 20 pairs each. The F_1 wild type ($\frac{sc+}{+y}$) daughters were divided into two groups. One was mated with scute males and the other with yellow males. The groups were then placed in population bottles with 10 - 15 pairs per bottle. The females were allowed to lay eggs for 4 - 5 days and were then discarded. When the F_2 emerged and their numbers approached 300 per bottle, the flies were scored and transferred to fresh bottles where they were allowed to continue mating overnight. The next day each individual female was placed into a vial with food, and allowed to produce offspring. The genotype of F_3 daughters permitted a diagnosis of the father's genotype. Thus in these experiments the two male genotypes were competing with each other for female mates. These female mates were scute and wild type in one group, and wild type and yellow in the other group. In order to observe simultaneously the competition between males for mating with all three female genotypes, in given proportions, an appropriate mixture of F_1 females previously mated to scute and yellow males respectively was made.

For/

For each resulting F_2 bottle, the number of females of each genotype inseminated by scute or yellow males were recorded as well as the numbers of double-matings and that of females without progeny. The results of these tests are given in Table 8.

From these tables, the following conclusions were drawn:-

(1) The numbers of scute females inseminated by scute and yellow males are not different from those of the wild type females, for a given proportion of scute and yellow males.

(2) The numbers of wild type females inseminated by scute and yellow males differed from that of the yellow females within experiments.

(3) The percentage of double-matings measured over all experiments was less than 1 per cent and the number of females without progeny less than 6 per cent.

Since there was no difference in mating preferences between scute and wild type females, they can be considered jointly in this analysis as "non-yellow" females. Thus, only two regressions of the ratio of the number of females inseminated by yellow males over the number of females inseminated by scute males on the ratio of the number of yellow over the number of scute males, according to formula (14), page No. 23 were calculated/

TABLE 8. Numbers of Each Genotype, Obtained in Population Bottles, Resulting from $\frac{sc+}{+y} \times \underline{sc+}$ or $\frac{sc+}{+y} \times \underline{+y}$ Matings or From a Mixture of Both

The figures below the numbers of female genotypes represent respectively the number of inseminations by scute males, by yellow males, the number of double-matings and that of the females without progeny

Males		Females		
$\underline{sc+}$	$\underline{+y}$	$\frac{sc+}{sc+}$	$\frac{sc+}{+y}$	$\frac{+y}{+y}$
87	102		120 100:3:0:17	99 45:44:1:9
56	77		62 50:2:0:4	66 27:33:3:3
50	37	43 40:2:0:1	48 44:3:0:1	
99	92	92 84:6:0:2	100 88:8:0:4	
41	43		36 33:3:0:0	47 18:29:0:0
58	45		53 52:1:0:0	44 29:15:0:0
56	48		49 45:3:0:1	42 29:10:2:1
40	145	43 36:7:0:0	109 82:22:0:5	36 9:20:0:7
37	100	30 22:6:0:2	47 31:8:0:8	47 3:36:0:8
31	99	22 12:0:0:0	78 59:14:0:5	31 10:21:0:0
46	126	54 44:9:0:1	90 71:15:0:4	30 11:26:0:0
59	161	43 39:2:0:2	108 84:16:0:8	21 6:12:0:3
27	116	25 17:8:0:0	77 49:14:0:28	36 5:30:0:1
59	128		65	113 31:78:2:2

Contd.

CONTINUATION OF TABLE 8

Males		Females		
sc+	+y	sc+/sc+	sc+/+y	+y/+y
43	114		56	115 44:62:6:3
52	93		34	101 32:66:2:1
18	124		39	73 11:53:0:6
29	102		17	93 17:70:0:6
37	99		24	103 29:69:0:7
73	71	59 54:2:0:3	69 62:5:0:2	
40	48		52 46:5:0:1	41 18:22:0:1
24	23	7 6:1:0:0	14 14:0:0:0	
30	33	9 9:0:0:0	22 21:0:1:0	
111	122		115 97:5:0:13	103 43:43:0:17
70	80		68 61:6:0:1	71 37:32:0:12
86	94		80 72:8:0:0	79 50:29:0:0
94	93		90 77:6:0:7	85 45:38:0:2
94	92		64 53:10:0:1	83 51:30:0:2
27	77		66 52:12:0:0	21 3:17:0:0
32	93		81 57:17:0:7	13 5:5:0:8
54	132		69 49:9:0:1	85 36:47:0:2

Contd.

CONTINUATION OF TABLE 8

Males		Females		
sc+	+y	sc+/sc+	sc+/+y	+y/+y
25	118		25	106 10:67:0:29
30	114		37	100 15:79:1:5
22	87		25	103 18:67:0:10
29	102		26	108 13:72:0:23
35	88		35	89 18:63:0:7
39	88		35	102 28:65:0:7
37	119		30	106 34:65:0:7
49	113		41	105 29:70:0:6
33	88		32	93 32:59:2:0
33	95		29	94 27:61:2:4
22	99		39	102 14:82:0:6

calculated, one for the yellow and the other for the non-yellow females.

It should be noted that when the errors of the proportion of scute to yellow inseminations were greater than 6 per cent, the data were not used for the computation of regressions.

The following example will illustrate the method. Taking the experiment No. 8 of <u>Table 8</u> where the observed number of genotypes in F_2, is,

$\underline{sc+}$	$\underline{+y}$	$\frac{sc+}{sc+}$	$\frac{sc+}{+y}$	$\frac{+y}{+y}$
40	145	43	109	36
		36:7:0:0	82:22:0:5	9:20:07

Respectively 36 and 7 scute, 82 and 22 wild type, and 9 and 20 yellow females have been inseminated by scute and yellow males; double matings were not observed; 5 wild type and 7 yellow did not produce progeny. The χ^2 for difference in proportions between scute and wild type is .46, between scute and yellow 19.2, and between wild type and yellow 19.2.

The proportions of scute-yellow inseminations for the non-yellow females was then 111:29 with an error of 3.2 per cent, and for the yellows 9.20 with an error of 8.6 per cent. The data from the yellow females were not taken into consideration and that of the non-yellow females were entered for the computation of the regressions/

regressions as,

$$y = \frac{\text{yellow inseminations}}{\text{scute inseminations}} = \frac{29}{111} = .261$$

$$x = \frac{\text{number of yellow males}}{\text{number of scute males}} = \frac{145}{40} = 3.625$$

$$y = wx$$

It should be noted that when the numbers of inseminations observed were very small, which was particularly frequent for non-yellow females inseminated by yellow males, the exact test for heterogeneity (Fisher, 1948) was applied, instead of χ^2.

The independence of the mating abilities of the males with respect to each group of females was reflected in a linear regression, w, of y on x, (w = constant). This was shown to be the case for all experiments conducted as shown in Fig. 1.

Calculated regression estimates are shown in Table 9.

TABLE 9. Coefficients of Regression, w, Calculated with the Data of Table 8.

	w	Deviation from 1	p%
Yellow Females	$1.060 \pm .144$	$t_{23} = .413$	70-60
Non-yellow Females	$.079 \pm .008$	$t_{24} = 100$	$< .1$

Summarising/

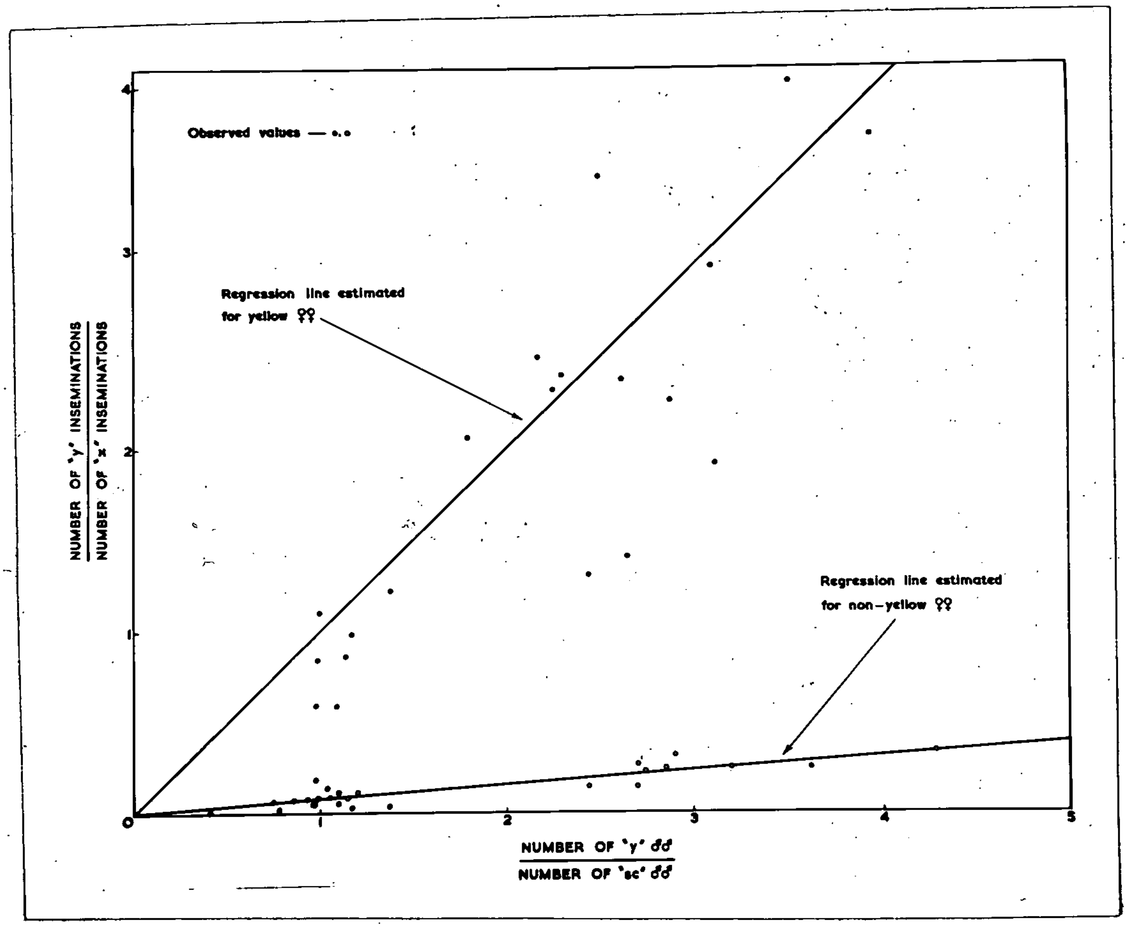


Fig. 1. Regressions of the ratio of the number of females inseminated by yellow males over the number of females inseminated by scute males on the ratio of the number of yellow over the number of scute males.

(Not all ratios are represented in the figure)

Summarising the results of the experiment on fitness component measurement, the following conclusions can be drawn:-

(1) Mating abilities are independent of the genotype frequency.

(2) Yellow males are at a disadvantage relative to scute ($w = .079$) when mating with non-yellow females and show no disadvantage when mating with yellow females ($w = 1$).

The only selection factor detected in these experiments was this partial male mating ability ("one-sided preference of Dobzhansky).

2.2.3. Observed and Expected Evolution of Genotypic Frequencies in a Population

To observe changes in gene frequencies, 20 replicates were set up in bottles with 20 pairs of flies each; the females were $\frac{sc+}{+y}$ and the males $\underline{+y}$. These flies were obtained in a manner similar to that used for the measurement of fitness components. After 5 days in the bottles the flies of the initial generation were shaken out and discarded. When the progeny emerged, a sample of at least 200 individuals, was taken from each population/

population, scored for genotypes, and placed for two days into fresh bottles as parents of the next generation; this was repeated for 13 generations. The harmonic mean number of males per generation per population was 124 and that of the females 118. The numbers of males and females in the first and second generations were greater than average, respectively 154 and 144 for males and 147 and 130 for females. The greatest number of individuals in any generation scored and used as parents were 231 males and 221 females (in the first generation); the most unbalanced sex ratio was 127 males to 54 females.

The average generation interval was 13 days.

Observed mean frequencies and the expected frequencies calculated with the estimate of partial mating ability, $w = .079$, are given in Table 10 and shown in Figs. No. 2 and 3.

Expected genotype frequencies were calculated as follows:-

Let, in generation n, at mating, x, 2y and z be respectively the frequencies of the $\frac{sc+}{sc+}$, $\frac{sc+}{+y}$ and $\frac{+y}{+y}$ female genotypes, and p and q respectively the frequencies of the $\underline{sc+}$ and $\underline{+y}$ male genotypes.

The expected frequencies of genotypes in the generation (n+1) are then:/

TABLE 10. Observed Mean Genotypic Frequencies (in %) in the Experimental Populations (upper figure) and Expected Frequencies Calculated from Fitness Estimates

Generation No.	No. of populations with the 2 genes segregating	Females				Males		
		sc+/sc+	sc+/+y	+y/+y	N_f^*	sc+	+y	N_m^{**}
0	20		100		20		100	20
1	20		50.0	50.0	147	48.6	51.4	157
			50.0±4	50.0±4		50.0±4	50.0	
2	20	22.5	53.0	24.5	130	26.4	73.6	144
		23.2±4	50.0±5	26.8±5		25.0±4	75.0	
3	20	35.4	40.8	23.8	114	46.4	53.6	117
		38.9±6	36.1±5	24.8±6		48.1±7	51.8	
4	20	51.9	35.6	12.5	122	57.5	42.5	107
		52.6±7	33.1±5	14.3±5		57.0±8	42.9	
5	20	64.2	28.4	7.4	116	68.5	31.5	114
		65.2±7	27.6±6	7.1±3		69.1±7	30.9	
6	20	69.6	28.4	2.0	99	78.2	21.8	117
		76.4±6	20.9±5	2.6±2		79.1±7	20.9	
7	19	84.7	14.3	1.0	123	86.2	13.8	117
		85.1	14.1	.8		86.9	13.1	
8	18	90.9	8.5	.6	119	93.1	6.9	117
		91.1	8.7	.2		92.2	7.8	
9	16	93.6	6.2	.2	118	94.6	5.4	120
		94.8	5.2	.0		95.4	4.6	
10	15	96.9	3.1	.0	124	96.5	3.5	126
		96.9	3.1	.0		97.4	2.6	
11	14	98.7	1.3	.0	120	98.7	2.3	124
		98.3	1.7	.0		98.5	1.5	
12	6	99.5	.5	.0	112	99.5	.5	134
		99.0	1.0	.0		99.1	.9	
13	6	99.7	.3	.0	116	99.8	.2	129
		99.4	.6	.0		99.5	.5	

N_f^* harmonic mean number of females
N_m^{**} " " " " males

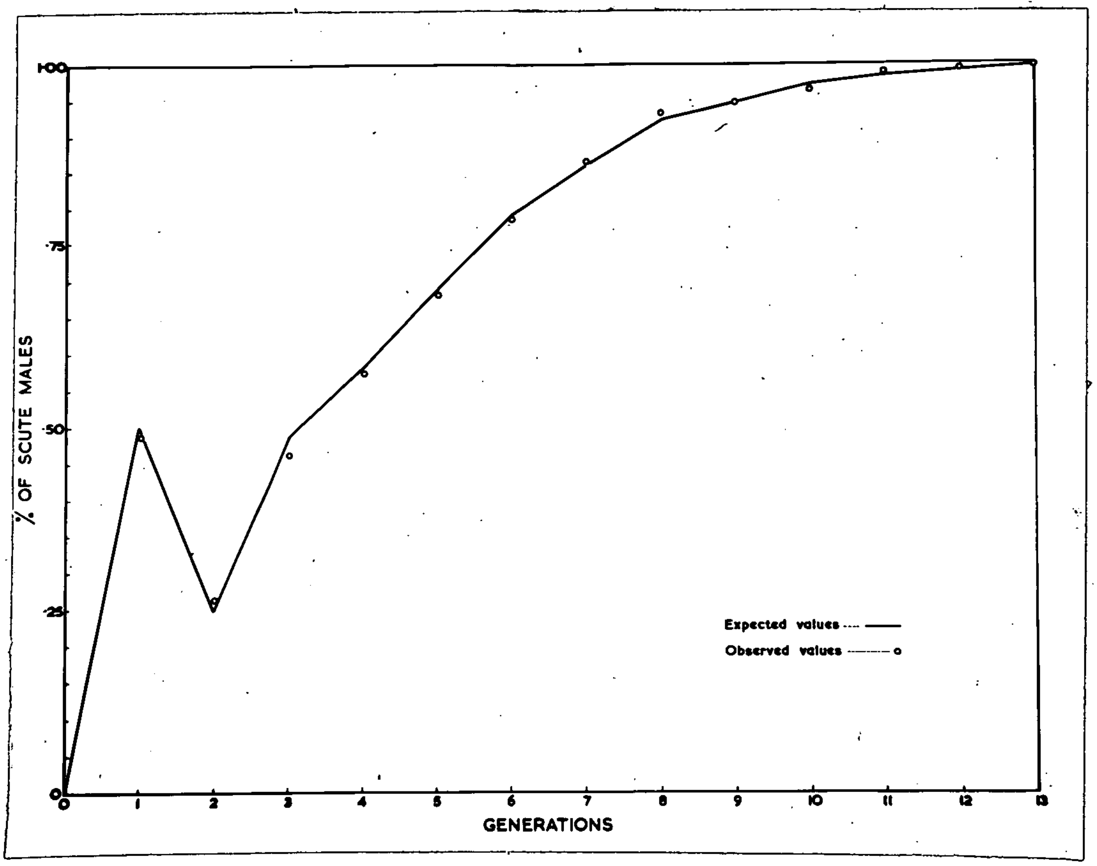
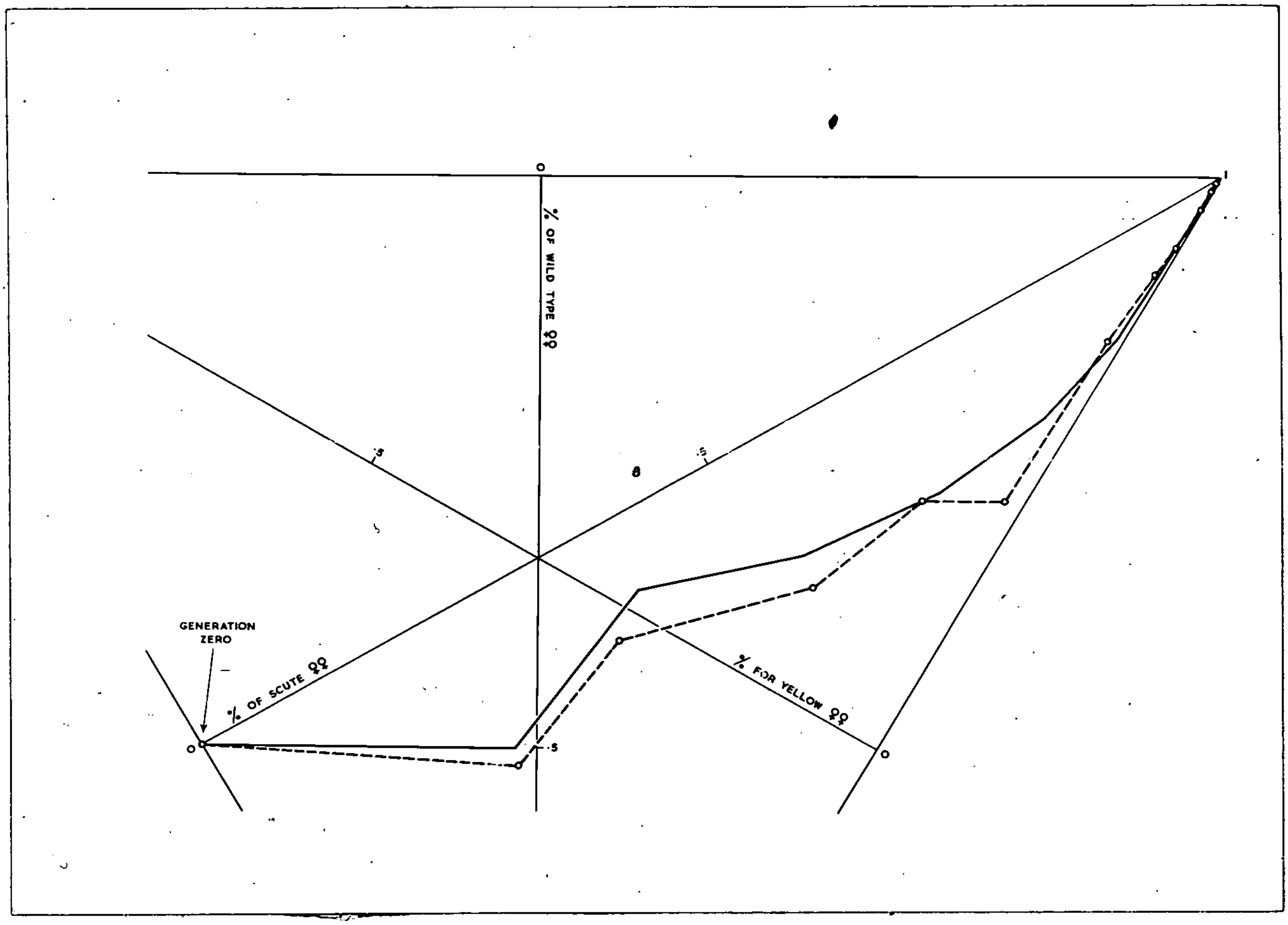


Fig. 2. Observed and expected genotypic frequencies in males.



Observed frequencies ————

Expected frequencies --------

Fig. 3. Observed and expected genotypic frequencies in females.

The expected frequencies of genotypes in the generation (n+1) are then:-

Generation	$\frac{sc+}{sc+}$	$\frac{sc+}{+y}$	$\frac{+y}{+y}$	$\underline{sc+}$	$\underline{+y}$
n	x	2y	z	p	q
n+1	$p'(x+y)$	$y+q'x+pz$	$q'y+pz$	$x+y$	$y+z$

where $p' = \frac{p}{p+.079q}$ and $q' = 1-p'$

The frequency of the "sc" gene in females is,

$$\tfrac{1}{2}\left\{(x+y) + p' - \Delta p\, z\right\},$$

where $\Delta p = p'-p$.

These calculations were applied repeatedly to successive generations substituting the frequencies in the generation n for those in generation (n+1).

The variances of the expected frequencies were calculated numerically. Differentiating and squaring the expressions for the expected frequencies, the variance in an infinite large generation (n+1) is obtained as shown in <u>Table 11</u>.

The sampling variance due to finite numbers of selected/

TABLE 11. Variances of the Expected Genotypic Frequencies in an Infinitely Large $(n+1)^{th}$ Generation

	Frequencies		Variances (V')
Genotypes	in n^{th} generation	in $(n+1)^{th}$ generation	
$\frac{sc+}{sc+}$	x	$p'(x+y)$	$(x+y)^2 V_{p'} + p'^2 V_{(x+y)}$
$\frac{sc+}{+y}$	$2y$	$y+pz+q'x$	$V_y + q'^2 V_x + x^2 V_{q'} + p^2 V_z + z^2 V_p + 2q' Cov_{x.y} + 2p Cov_{y.z} + 2pq' Cov_{xz} + 2xz Cov_{pq'}$
$\frac{+y}{+y}$	z	$q'y+q^z$	$q'^2 V_y + y^2 V_z + z^2 V_q + 2qq' Cov_{y.z} + 2yz Cov_{qq'}$
$\underline{sc+}$	p	$x+y$	$V_{(x+y)}$
$\underline{+y}$	q	$y+z$	$V_{(y+z)}$

The variance of the frequency of the "sc" gene in females is:

$$\frac{1}{4} V_{(x-y)} + V_{p'} + z^2 V_p + (\ p)^2 V_z + 2p Cov_{(x-y)z} + 2z Cov_{p'p}$$

$V_x, V_y, \ldots V_p \ldots V_{q'}$, are respectively the variances of x, $y \ldots p \ldots p$ in zero generation, and,

$Cov_{x.y}, Cov_{x.z} \ldots Cov_{pq'}$ respectively the covariance between x and y, x and z, p and q' in the n^{th} generation selected male (Nm) and (Nf) female parents was then added to the expected variance of the infinitely large population.

An estimate of sampling variance for a finite number of parents of either sex was obtained as follows:-

(1) Assume parent genotype frequencies in the infinite population distributed normally with variance V'.

(2) Subdivide this parent population into K classes covering equal intervals of absissa equal to .02.

(3) Assume that genotype frequencies of a given interval, i, can be represented by the mid-point u_i between class limits.

(4) Determine from tables of the normal curve the sampling variance contributed by each class as $\frac{u_i(1-u_i)}{N} f_i$ where f_i is the area under the normal curve of the i^{th} interval.

(5) Sum over all intervals to obtain the total sampling variance

$$V'' = \sum_{i=1}^{i=K} \frac{u_i(1-u_i)}{N} f_i$$

(6) Obtain the total variance for a finite number of parents of a given sex as

$$V_1 = V' + V''$$

This/

TABLE 12. Observed and Expected Variances of the Frequency of "sc" in Experimental Populations

Generation	No. of replicates	In Females			In Males	
		Observed	Expected		Observed	Expected for N_m**
			for N_f^*	for .5 N_f		
1	20	2.5	4.2		8.8	15.9
2	20	40.8	22.7	40.8	12.8	17.3
3	20	62.6	36.7	62.2	45.1	49.1
4	20	103.8	34.6	54.6	150.3	59.6
5	20	105.8	25.5		136.5	53.4
6	20	33.8	19.6		138.2	46.4

N_f^* harmonic mean of females

N_m^{**} " " " males

This procedure was applied repeatedly to successive generations in order to obtain estimates of variance of expected gene frequencies up to generation 6.

The observed variances between populations and the expected variances calculated as indicated above are shown in *Table 12*. N_m and N_f were set equal to the actual numbers of parents mated. Thus the expected variances are almost certain to be underestimated.

Alternatively, the expected variances for females were calculated by using 0.6 x N_f in place of N_f. This second procedure appears to give improved predictions of variances. (see *Table 12*).

Faulty experimental technique may account for the large variance between experimental populations observed at the 4th generation.

2.2.4. *Change of Fitness by Inbreeding*

Five inbred lines were made by mating brother to sister for 30 generations using $\frac{sc+}{+y}$ females and *+y* males, in order to keep the two genes *sc* and *y* segregating within lines.

Only one line survived to the end; the others were lost respectively at the 7th, 8th, 10th and 21st generation. All lines but this last one were replaced/

replaced by replicates of the surviving line. Thus the four lines available at generation 30 were closely related to each other.

At generation 31, five replicates of each inbred line were expanded in size and set up in population bottles of about 250 flies each, to observe the evolution of genotypic frequencies for three generations. The same procedure was used as described earlier (page 46) for the outbred population.

Differential viabilities between scute and yellow males and between wild type and yellow females were estimated from the proportions of individuals of the first generation. These results are shown in Table 13. It appears that there is no differential viability between scute and yellow males; whereas yellow females are less viable than the double heterozygotes. The analysis of variance given below indicates this difference to be highly significant.

Analysis of Variance of Female Viabilities
(percentage survivors of the $\frac{sc\,+}{+\,y}$ genotype)

Source of Variation	D.F.	Mean Square
Deviation from 1:1 ratio	1	8521
Between lines	3	52
Between replicates within lines	15	29
Error	18	33

TABLE 13. Total Number of Each Genotype Resulting from $\frac{sc\,+}{+\,y}$ × $\underline{+\,y}$ Matings

		Males		Females	
Line	No. of replicates	$\underline{sc\,+}$	$\underline{+\,y}$	$\frac{sc\,+}{+\,y}$	$\frac{+\,y}{+\,y}$
1	5	298	300	286	238
2	5	269	288	270	267
3	5	327	249	317	266
4	5	170	195	272	197
Totals	20	1064	1032	1145	968

TABLE 14. Total Numbers of Each Genotype Resulting from $\frac{sc+}{+y}$ × $\underline{sc+}$ Matings

	Males		Females	
Line	$\underline{sc+}$	$\underline{+y}$	$\frac{sc+}{sc+}$	$\frac{sc+}{+y}$
1	166	126	140	152
2	187	196	177	202
3	234	244	303	335
4	181	132	203	269
Totals	768	698	823	959

Differential viabilities between wild type and the scute females were estimated from segregation in $\underline{\frac{sc+}{+y}} \times \underline{sc+}$ matings. The results are given in <u>Table 14</u>. They confirm those of <u>Table 13</u> in that no consistant differences were found between <u>sc+</u> and <u>+y</u> male genotypes. Heterozygous ($\frac{sc+}{+y}$) females were again more viable than the homozygous ($\frac{sc+}{sc+}$) females.

In both tests (Tables 13 and 14) the homozygous females for either scute or yellow were about 15 per cent less viable than the double heterozygote. The greater viability of the double heterozygote in inbred lines may be attributed to a region of the x-chromosome adjoining the "sc" and "y" loci which has been retained in a heterozygous state.

The estimate of 15 per cent reduction in viability of the homozygous females and the estimate of partial mating ability of .079 for yellow males as calculated in previous experiments (page No. 45), were used to predict genotype frequencies in the inbred lines. The predicted frequencies and the observed mean frequencies of genotypes measured over all lines are shown in <u>Table 15</u>. There is good agreement between actual and predicted results.

From this, it may be concluded that inbreeding did not change the relative mating abilities of the yellow males. In other words, it appears that the effect of

of/

TABLE 15. Observed Mean Genotypic Frequencies (in %) of the Inbred Lines (upper figure) and Expected Frequencies Calculated from Estimates of Fitness (lower figure)

	Females				Males		
Generation	$\frac{sc+}{sc+}$	$\frac{sc+}{+y}$	$\frac{+y}{+y}$	N_f^*	$\frac{sc+}{}$	$\frac{+y}{}$	N_m^{**}
0	0	1	0	20	0	1	20
1	0	54.2	45.8	111	50.8	49.2	110
		54.0	46.0		50.0	50.0	
2	22.9	57.2	19.9	116	25.7	74.3	103
	23.0	54.0	23.0		27.1	72.9	
3	32.4	46.6	20.9	99	45.5	54.5	97
	38.9	36.9	24.2		50.0	50.0	

N_f^* mean number of females for population

N_m^{**} " " " males " "

of the gene "yellow on mating ability of the males is independent of its genetic background.

From experiments conducted by Merrell (1949), where yellow males competed with wild type males for mating with heterozygous females, it was estimated that the relative mating ability of the yellow males was about .062. The same figure can be calculated from identical experiments conducted by Koref-Santibanez and Waddington (1958), using stocks different from those of Merrell.

The mating ability of scute males relative to that of wild type can thus be predicted to be $\frac{.062}{.079}$ or .78.

SUMMARY

Methods of evaluation of the relative fitness of genotypes were studied to predict changes in the frequencies of the sex-linked genes *scute* and *yellow* in *Drosophila melanogaster* populations. The exact formula which relates the change in frequency of sex-linked genes and the average fitness of a population was derived.

Total fitness of the genotypes can be measured in one single operation or it may be broken down into components namely viability, fertility and mating ability which may be measured separately. This latter procedure was followed here since the components of fitness had to be reassembled in different ways depending on the point of the life cycle where the scoring of the genotypes is made.

Maximum likelihood solutions were obtained to estimate simultaneously relative viabilities and fertilities between sex-linked genotypes. The solutions apply when no selective mating ability exists between genotypes. A new system of measuring mating ability is presented. In this system the mating process is compared with a catalytic reaction and fitness is defined in terms of Sewall Wright's selective coefficients. The only selective force detected in the present experiments was the greater mating ability of/

of scute males relative to that of yellow males when mating with non-yellow females.

Good agreement was obtained between observed changes in genotypic frequencies of scute and yellow genotypes and the changes predicted using the components of fitness measured in independent experiments.

An attempt was made to change the relative fitness of the genotypes by inbreeding. Homozygous for sc or y females were 15 per cent less viable than heterozygotes.

REFERENCES

BASTOCK, M. 1956. A gene mutation which changes a behavior pattern. *Evolution, 10* : 421-439.

BATEMAN, A.J. 1949. Analysis of data on sexual isolation. *Evolution,* 3 : 174-177.

BENNETT, J.H. 1957. Selectively balanced polymorphism at sex-linked locus. *Nature, 180* : 1363.

CROW, J.F., and KIMURA, M. 1955. Some genetic problems in natural populations. *Proc. III Berkeley Symp. Math. Stat. & Probability.*

DARWIN, C. 1871. The descent of man and selection in relation to sex. *Random House, Modern Library Edition.*

DIEDRICH, G.W. 1941. Non-random mating between yellow-white and wild type of *D. melanogaster. Genetics, 26* : 148.

DOBZHANSKY, Th. 1944. Experiments on sexual isolation. I. Geographic strains of *D. willistoni. Proc. Nat. Acad. Sci., 30* : 238-244.

DOBZHANSKY, Th., and STREISINGER, G. 1944. Experiments on sexual isolation of Drosophila. II. Geographic strains of *Drosophila prosattans. Proc. Nat. Acad. Sci., 30* : 340-345.

FISHER, R.A. 1930. The genetical theory of natural selection. *Oxford University Press.*

FISHER, R.A. 1948. Statistical methods for research workers. *Oliver & Boyd.*

HALDANE, J.B.S. 1932. The causes of evolution. *Longmans, Green and Co.*

HUXLEY, J.S. 1942. Evolution, the modern synthesis. *George Allen & Unwin, Ltd.*

KERR, W.E., and WRIGHT, S. 1954a. Experimental studies in the distribution of gene frequencies in a very small population of *D. melanogaster.* I. Forked. *Evolution, 8* : 172-177.

KERR, W.E., and WRIGHT, S. 1954b. Experimental studies of the distribution of gene frequencies in very small populations of *D. melanogaster.* III. Aristopedia and spineless. *Evolution, 8* : 293-302.

KNIGHT, G.R., and ROBERTSON, A. 1957. Fitness as a measurable character in Drosophila. *Genetics, 42* : 524-530.

KOREF-SANTIBANEZ, S., and WADDINGTON, C.H. 1958. The origin of sexual isolation between different lines within a species. *Evolution, 4* : 485-493.

LEVENE, H. 1949. A new measure of isolation. *Evolution*, *3* : 315-321.

LEVENE, H., and DOBZHANSKY, Th. 1945. Experiments in sexual isolation in Drosophila. V. The effect of varying proportions of *D. pseudo-obscura* and *D. persimilis* on the frequency of insemination in mixed populations. *Proc. Nat. Acad. Sci.*, *31* : 274-281.

LI, C.C. 1955. The stability of an equilibrium and the average fitness of a population. *Amer. Nat.*, *89* : 281-295.

LUDWIN, I. 1951. Natural selection in *D. melanogaster* under laboratory conditions. *Evolution*, *5* : 231-242.

MAYR, E., and DOBZHANSKY, Th. 1945. Experiments on sexual isolation in Drosophila. IV. Modification of the degree of isolation between *D. pseudo-obscura* and *D. persimilis* and of sexual preferences in *D. prosattans*. *Proc. Nat. Acad. Sci.*, *31* : 75-82.

MERRELL, D.J. 1949. Selective mating in *Drosophila melanogaster*. *Genetics*, *34* : 370-389.

MERRELL, D.J. 1950. Measurement of sexual isolation and selective mating. *Evolution*, *4* : 326-331.

MERRELL, D.J. 1953. Selective mating as a cause of gene frequency changes in laboratory populations of *D. melanogaster*. *Evolution*, *7* : 287-296.

PATTERSON, J.T., STONE, W.S., and GRIFFEN, A.B. 1940. Evolution of the *virilis* group in Drosophila. *Univ. Texas Publ.* 4032 : 218-250.

PATTERSON, J.T., McDONALD, L.W., and STONE, W.S. 1947. Sexual isolation between members of the *virilis* group of species. *Univ. Texas Publ.* 4270 : 7-31. Cited by Merrell, 1949.

PETIT, C. 1959. Les facteurs genetiques de la competition sexuelle entre une forme mutante et son allelomorphe sauvage chez *D. melanogaster*. *Ann. Genetique*, *1* : G83.

REED, S.C., and REED, E.W. 1948. Natural selection in laboratory populations of Drosophila. *Evolution*, *2* : 176-186.

REED, S.C., and REED, E.W. 1950. Natural selection in laboratory populations of Drosophila. II. Competition between a white-eye gene and its wild type allele. *Evolution*, *4* : 34-42.

ROBERTSON, F.W., and SANG, J.H. 1944. The ecological determinants of population growth in a Drosophila culture. I. Fecundity of adult flies. *Proc. roy. Soc.*, *B*, *132* : 258-291.

STALKER, H.D. 1942. Sexual isolation studies in the species complex *Drosophila virilis*. *Genetics*, *27* : 238-257.

STURTEVANT, A.H. 1915. Experiments in sex recognition and the problem of sexual selection in Drosophila. _J. Anim. Behav._, _5_ : 351-366.

WALLACE, B. 1948. Studies in sex-ratio in _Drosophila_ _pseudo-obscura._ I. Selection and sex-ratio. _Evolution_, _2_ : 189-217.

WRIGHT, S. 1942. Statistical genetics and evolution. _Bull. Amer. Math. Soc._ 48 : 223-246.

WRIGHT, S. 1949. Adaptation and selection. In: _Genetics, Paleontology and Evolution._ Ed. : G.L. Jepson, G.G. Simpson, and E. Mayr.

WRIGHT, S., and KERR, W.E. 1954. Experimental studies of the distribution of genotypes in very small populations of _D._ _melanogaster._ II. Bar. _Evolution_, _8_ : 225-240.

ACKNOWLEDGEMENTS

It is a pleasure to acknowledge my indebtedness to:

The University of Lisbon for study leave to carry out this work.

The Foundation Calouste Gulbenkian for a grant.

Professor C. H. Waddington, F.R.S., for the provision of facilities in the Institute of Animal Genetics.

Dr. Alan Robertson for guidance throughout this investigation.

Miss. M. Wheeler and Mrs. M. Smith for assistance in the computations.

Dr. H. Abplanalp and Dr. G. McBride for reading and correcting the manuscript.

All those of this Institute who have contributed much in the way of discussion.