GENETICS OF NATURAL POPULATIONS. XXI. CONCEALED VARIABILITY IN TWO SYMPATRIC SPECIES OF DROSOPHILA[1]

THEODOSIUS DOBZHANSKY AND BORIS SPASSKY
Columbia University, New York City

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NATURAL populations of several species of Drosophila examined in this respect carry great stores of concealed genetic variability. This variability may be brought to light, and its quality and quantity may be measured, by means of fairly simple genetic techniques. Individuals are obtained which are homozygous for, that is, carry in duplicate, certain chromosomes derived from known progenitors collected in the natural habitats of the species. Such homozygotes are often deficient in viability, or sterile, or show various structural or physiological abnormalities which distinguish them from "normal" or "wild" flies.

Concealed genetic variability exists probably in all sexually reproducing and cross-fertilizing organisms including man. Its biological function is, however, little understood. It may be important as a store of genetic raw materials from which new adaptive genotypes are built in the process of evolution. The concealed variability is also important as a source of hybrid vigor, or heterosis. On the other hand, it is a source of the poorly adapted variants and hereditary diseases which lower the immediate fitness of some members of natural populations. Little is known about the agencies which determine the quality and quantity of the store of concealed variability in a given species or population. Comparison of these stores in closely related species which differ in their ecology and reproductive biology is a promising method of investigation of the problem at hand. The present article reports such a comparison for two species, *Drosophila pseudoobscura* and *Drosophila persimilis*. These forms are sibling species, very similar in external morphology, and with similar chromosomes which carry presumably the same gene loci, although arranged in somewhat different linear orders. The two species occur together in some localities in the western United States, making it possible to collect samples of populations living in the same general environment.

MATERIAL

Samples of the populations of *D. pseudoobscura* and *D. persimilis* were collected in July and August of 1951 at Mather, Aspen Valley and Porcupine Flats in the Yosemite Park region of the Sierra Nevada Mountains in California. The flies were shipped to the laboratory in New York via air mail. The

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females were placed singly in culture bottles, and some of the larvae which appeared in these bottles were used to examine the chromosomes in their salivary gland cells. This cytological examination permits determination of the species to which the flies in a given culture belong. Males captured in their natural habitats were placed with virgin females of both species marked with suitable mutant genes. In most cases only conspecific females were inseminated and produced offspring.

THE CROSSES

Methods used to detect concealed recessive variants in the chromosomes of Drosophila are sufficiently well known (see DOBZHANSKY, HOLZ and SPASSKY 1942; PAVAN *et al.* 1951, and other papers for descriptions). In brief, a single wild male, or a son of a wild female, is crossed to a laboratory strain with suitable mutant markers, and a series of crosses are executed designed to obtain

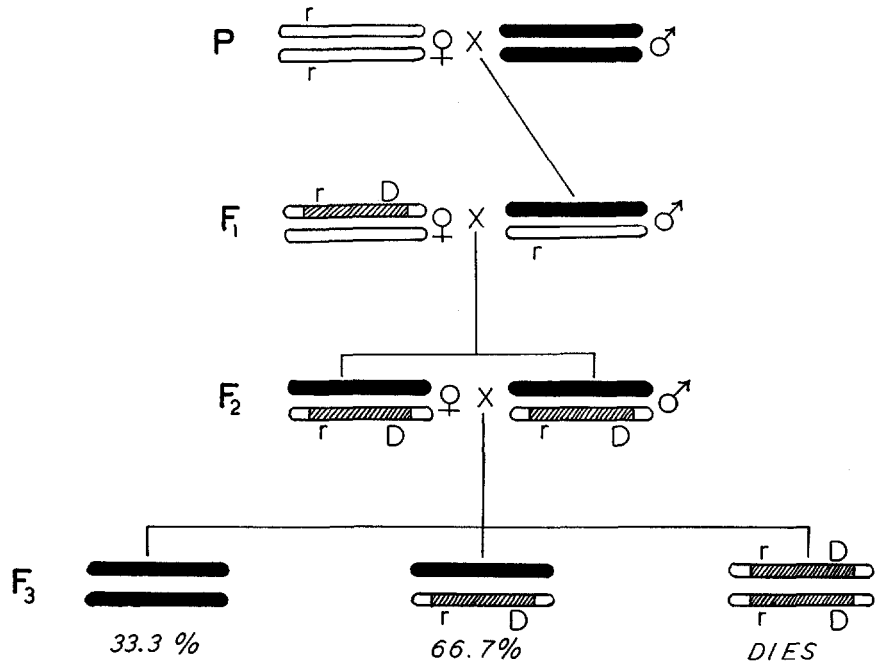

FIGURE 1.—The series of crosses used to obtain homo- and heterozygotes for wild chromosomes of Drosophila. White—chromosomes of laboratory strains with dominant (D) and recessive (r) markers; black—chromosomes derived from natural populations; cross-hatched—inverted sections which suppress recombination in females.

individuals homozygous for a given wild chromosome in the same culture bottles with individuals heterozygous for this chromosome and a chromosome carrying a dominant mutant marker (fig. 1).

In the experiments dealing with the second chromosomes of *D. pseudoobscura* the mutant gene glass, *gl,* was used as the recessive marker (*r,* in fig. 1). The analyzer chromosome (*D* in fig. 1) contained glass, the dominant

Bare (*Ba*), and an inversion which suppressed virtually all recombination in the second chromosomes. This analyzer chromosome was transferred by means of six consecutive backcrosses in *D. persimilis* also. But instead of outcrossing to glass as in *D. pseudoobscura,* the wild *D. persimilis* flies were outcrossed to a dominant mutant Delta. Single Delta/wild males from the progeny were then crossed to females which carried the analyzer chromosome, and Bare non-Delta females and males selected in the next generation were inbred.

The third chromosome was controlled by means of the recessives orange and purple (*or pr*), and an analyzer chromosome which contained these recessives and also the dominants Blade and Scute. The *or Bl Sc pr* analyzer chromosome had the standard gene arrangement, and accordingly permitted crossing over to occur when placed opposite a wild standard chromosome. Fortunately, the standard gene arrangement is relatively infrequent in the material studied (DOBZHANSKY 1952). The standard chromosomes were ignored. The *or pr* and the *or Bl Sc pr* chromosomes were transferred from *D. pseudoobscura* to *D. persimilis* by six backcrosses.

The fourth chromosome recessive incomplete (*inc*), and an analyzer chromosome containing *inc,* the dominant Curly (*Cy*), and an inversion which suppressed recombination were used in both *D. pseudoobscura* and *D. persimilis.* These markers were obtained originally in the former species and transferred to the latter by six backcrosses.

The cultures which produced the flies to be counted were kept in a constant temperature room at 25°C. All other cultures were kept at room temperature. In *D. pseudoobscura* the crosses *Ba* female × *Ba* male, *Bl Sc* female × *Bl Sc* male, and *Cy* female × *Cy* male were made with 6 females and 6 males, which were transferred three times at two-day intervals to fresh culture bottles. In *D. persimilis* about 10 females and 10 males were used, and the flies were permitted to oviposit for three days in each culture (four days for the *Ba* flies). The counts of the flies which hatched in the cultures were made at three-day intervals after the beginning of ecclosion from the pupae.

CONTROL EXPERIMENTS

The dominant marker in the analyzer chromosome (*D* in fig. 1) may reduce the viability of its carriers below normal. Normal viability is defined as that of flies which have the two chromosomes of a pair taken at random from the population of a given locality (DOBZHANSKY, HOLZ and SPASSKY 1942; DOBZHANSKY and SPASSKY 1944; PAVAN *et al.* 1951; and others). The effects of *D* on the viability and the standard of the "normal" viability are determined by means of control experiments. These consist in intercrossing wild/*D r* Inv females and males from different cultures. In the offspring, the wild/wild flies now have two wild chromosomes of a pair derived from *different* wild progenitors, instead of having the same wild chromosome in duplicate. Since the average viability of such heterozygous flies is normal by definition, any departure from the ratio 33.3% wild : 66.7% *D* must be ascribed to the analyzer chromosome carried in the *D* flies.

TABLE 1

Frequency, in percent, of the wild-type class in the control experiments.

Species	Chromosome	Culture means	Total flies
pseudoobscura	Second	35.94 ± 0.57	35.88 ± 0.49
persimilis	"	34.46 ± 0.56	34.47 ± 0.48
pseudoobscura	Third	37.62 ± 0.80	37.39 ± 0.54
persimilis	"	39.00 ± 0.63	39.47 ± 0.47
pseudoobscura	Fourth	34.33 ± 0.83	32.82 ± 0.48
persimilis	"	33.87 ± 0.54	33.45 ± 0.43

A summary of the results of control experiments is presented in tables 1 and 2. Table 1 shows the over-all percentages of wild type flies in the crosses in which the wild chromosomes were derived from two different wild progenitors. These percentages can be calculated either as means of the frequencies of the wild-type class in separate crosses, or as the proportions of wild-type flies among the total numbers of the flies counted in tests of a given chromosome. These two modes of calculation give very similar results.

In the experiments testing the second chromosomes the wild-type class is slightly but significantly more frequent than the expected 33.3%. The *gl Ba* Inv analyzer chromosome evidently causes some reduction of the viability of its carriers. The same is true to a greater extent for the *or Bl Sc pr* analyzer chromosome used in the experiment in the third chromosome. No perceptible deleterious effects are however produced by the *inc Cy* Inv fourth chromosome, the ratios here being well within the expected sampling error limits for the ideal 33.3%.

Table 2 shows the percentages of the wild-type class in the different counts in the control cultures. If the analyzer chromosomes slow down the development of their carriers, wild-type flies should be more frequent in the early than in the late counts. A slight effect of this sort is observed in the second chromosome tests (it is significant statistically for the *D. persimilis* cultures and does not quite reach the conventional level of significance in the *D. pseudoobscura* ones). The *or Bl Sc pr* third chromosome causes a significant delay on the

TABLE 2

Percentages of wild-type flies on different days of hatching in the control experiments. The numbers of the flies are indicated in parentheses.

Species	Chromosomes			
	Counts	Second	Third	Fourth
pseudoobscura	1	37.6 (1637)	40.4 (1496)	35.8 (1527)
"	2	36.8 (2467)	36.8 (1732)	36.0 (1869)
"	3	34.4 (2738)	37.5 (2016)	31.0 (2531)
"	4	36.6 (1844)	36.8 (1748)	34.9 (2575)
"	5+	33.8 (940)	37.0 (963)	32.3 (2357)
persimilis	1	37.3 (2569)	43.6 (3026)	35.0 (3382)
"	2	35.0 (2599)	38.9 (3234)	33.0 (3358)
"	3	34.1 (2029)	36.9 (2228)	31.7 (3165)
"	4	32.2 (1828)	39.1 (1574)	35.1 (2466)
"	5+	29.8 (631)	33.4 (736)	36.6 (1124)

TABLE 3

Viability of homozygotes for certain chromosomes, expressed in per cent of the normal viability; the figures indicate numbers of chromosomes that fall in various viability classes.

Species	Chromosome	Percent normal viability															Total
		0	10	20	30	40	50	60	70	80	90	100	110	120	130	140	
pseudo-obscura	Second	21	2	3	3	7	7	15	27	19	5	....	....	....	....		109
persimilis	"	18	3	3	1	3	3	6	14	22	17	10	4	1	1		106
pseudo-obscura	Third	18	3	2	2	4	11	19	21	21	9	5	....	1	....		116
persimilis	"	28	3	3	2	3	9	11	22	54	24	13	....	....	....		172
pseudo-obscura	Fourth	11	3	5	4	5	2	7	17	26	19	5	3	1	....		108
persimilis	"	27	8	4	4	4	17	18	27	38	13	5	2	....	....		167

TABLE 4

Percentages of the chromosomes which are lethal or semilethal when homozygous.

Chromosome	*pseudoobscura*	*persimilis*
Second	33.0 ± 4.5	25.5 ± 4.2
Third	25.0 ± 4.0	22.7 ± 3.2
Fourth	25.9 ± 4.2	28.1 ± 3.5

D. persimilis but not on the *D. pseudoobscura* genetic background. Finally, the *inc Cy* Inv fourth chromosome does not affect the development rate of its carriers in any way.

LETHALS AND SEMILETHALS

The data on the viability of the homozygotes for the different chromosomes are summarized in table 3 in terms of percentages of the normal viability. These are obtained by dividing the percentage of the wild-type class observed in the cultures testing a given chromosome by the frequency of the same class in the corresponding control experiment (table 1) and multiplying the result by 100.

In all 778 chromosomes have been tested. Some of them produced either no or very few wild-type flies in the cultures. These chromosomes may be said to be lethal when homozygous (the 0–10 percent class in table 3). Those which produced some wild-type flies, but less than half in the percentage obtained in the corresponding control experiments, are semilethal to homozygotes (the 10–50 percent classes in table 3). The remainder of the chromosomes permit the survival of most of the homozygotes. The modal classes are either 70–80% or 80–90% of the normal viability. Several chromosomes gave greater proportions of wild-type flies than obtained in the control cultures. These may be the " super-vitals," i.e., genotypes which, in at least some environments, survive more frequently than do normal heterozygotes.

Table 4 shows that roughly from one-quarter to one-third of all chromosomes tested were lethal or semilethal in homozygotes. No significant differences between the two species are apparent. Similarly there are no significant differences between the frequencies of lethals and semilethals in the second, third and fourth chromosomes of the same species. This last result is, in a way, surprising. The second chromosomes are longer than the third and the fourth in the salivary gland cells; DOBZHANSKY, HOLZ and SPASSKY (1942) found

TABLE 5

Mean viability of homozygotes for certain chromosomes.

Species	Chromosome	All chromosomes	Non-lethals
pseudoobscura	Second	55.22 ± 3.12	75.00 ± 1.22
persimilis	"	67.64 ± 2.90	87.94 ± 1.77
pseudoobscura	Third	61.25 ± 2.95	77.06 ± 1.50
persimilis	"	66.54 ± 2.61	83.42 ± 1.11
pseudoobscura	Fourth	68.28 ± 2.90	85.62 ± 1.49
persimilis	"	59.64 ± 2.50	77.91 ± 1.40

greater frequencies of lethals and semilethals in the second and the fourth chromosomes of *D. pseudoobscura* than obtained in the third chromosome of the same species by WRIGHT, DOBZHANSKY and HOVANITZ (1942). It must however not be forgotten that the second, third and fourth chromosomes studied by the above authors came from samples collected in different localities. In fact the data reported in the present article are the first in which the genetic variability of the different chromosomes and different species is examined in population samples collected at the same time and in the same localities. Furthermore, the experimental errors in table 4 are too large to exclude the possibility that the third chromosomes have actually fewer lethals than do the others.

MEAN VIABILITY OF THE HOMOZYGOTES

The data summarized in table 3 permit calculation of the mean viabilities of individuals homozygous for the second, third or fourth chromosomes found in natural populations of the two species. Such calculations are shown in table 5. They have been made in two ways, namely taking into account either all chromosomes, or only the quasi-normal ones (i.e., those which do not contain either lethals or semilethals). The first way has a serious defect, because variances computed from grossly abnormal distributions of the kind shown in table 3 are not reliable. The quasi-normal chromosomes, although defined arbitrarily as those giving more than 50 percent of the normal viability in the homozygotes, form fairly regular bell-shaped distributions.

Fortunately, both ways yield rather similar results. The depression of the viability caused by homozygosis for the second and the third chromosomes is significantly smaller in *D. persimilis* than it is in *D. pseudoobscura*. The relationship is reversed in the fourth chromosomes.

SUBVITALS AND SUPERVITALS

Inspection of tables 3 and 5 clearly shows that the mean viability of homozygotes for quasi-normal chromosomes is below 100, i.e., below the mean viability of the heterozygotes. It is evident that at least some of the chromosomes which are free of lethals and semilethals are nevertheless subvital in homozygotes. And yet, table 3 shows that a minority of the quasi-normal chromosomes have produced homozygotes the viability of which appears to be above 100. These chromosomes may be supervital in homozygotes. (In the earlier papers of the present series the subvitals and supervitals were referred to as " minus modifiers " and " plus modifiers " of the viability.)

In a paper published in the present issue, WALLACE and MADDEN describe a statistical technique of estimation of the frequencies of subvital and supervital chromosomes. They show that the observed variance of the viability effects of different chromosomes and chromosome combinations (σ_{res}^2) has at least three components. Indeed, the proportions of the wild-type flies observed in the test cultures in our experiments depend, in part, upon the genotypic differences between the different chromosomes found in a natural population. A part of

the observed variance is, thus, " real," or genetic, variance, σ_r^2. But the proportions of the wild-type flies depend also upon the environmental variations in the different test cultures (σ_e^2); finally, a part of the observed variance is due to sampling errors, σ_s^2.

PROFESSOR HOWARD LEVENE has very kindly indicated to us methods for the estimation of the observed and the real variance most suitable for our type of data, which differ somewhat from the data of WALLACE and MADDEN. The method is as follows. Let x_{ij} be the observed frequency of the wild type class in the j-th culture of the i-th chromosomes (or a combination of chromosomes for the heterozygotes).

$$\bar{x}_i = (\Sigma x_{ij})/r_i$$

is the unweighted mean of the frequencies in the r_i cultures of the i-th chromosome or combination. Let $\bar{x}$ be the unweighted average of the x_i. The total observed variance is given by:

$$\sigma_{res}^2 = \Sigma(\bar{x}_i - \bar{x})^2/(k-1),$$

where k is the number of different chromosomes or combinations studied. If the number of cultures, r_i, is always equal to r, one can estimate the variance between replicate cultures due to environmental effects and to sampling errors as:

$$\sigma_{es}^2 = \Sigma\Sigma(x_{ij} - \bar{x}_i)^2/(r-1)k.$$

In the absence of real (genotypic) variance, the observed variance of the means $\bar{x}_i$ would be due entirely to environment and to sampling, and would be 1/r times this. It could then be estimated as:

$$\sigma_{es}^2 = \Sigma\Sigma(x_{ij} - \bar{x}_i)^2/r(r-1)k.$$

The σ_r^2 could then be estimated easily as the difference between σ_{res}^2 and σ_{es}^2. When r_i is variable from one cross to another, as happens to be the case in the present data, it can be shown that σ_{es}^2 should be estimated as:

$$\sigma_{es}^2 = \Sigma\Sigma(x_{ij} - \bar{x}_i)^2/r_i(r_i-1)k.$$

The σ_r^2 is then equal to $\sigma_{res}^2 - \sigma_{es}^2$. The results of the calculations are summarized in table 6. The units used in these calculations are percentages of the normal viability (see pages 473 and 474). It can be seen that both the observed and the genetic variances are invariably higher in the homozygous than in the heterozygous cultures. The conclusion which obviously follows is that the viabilities of the heterozygotes are more nearly uniform than those of the homozygotes (DOBZHANSKY and WALLACE 1953).

Following WALLACE and MADDEN, we define as subvital the homozygotes which, under the conditions of our experiments, exhibit viabilities which deviate by more than two σ_r below the average viability of the heterozygotes. Similarly, a supervital is a homozygote the viability of which is more than two σ_r above the average for the heterozygotes. Since the viabilities of the heterozygotes, and of the homozygotes for the quasi-normal chromosomes, are normally distributed, one may, again following WALLACE and MADDEN, estimate

TABLE 6

Variance of the viability of homozygotes and heterozygotes for certain chromosomes.

Species	Chromosome	σ_{res}^2	σ_{es}^2	σ_r^2	σ_r
		Heterozygotes			
pseudoobscura	Second	45.05	26.64	18.41	4.29
"	Third	168.32	115.47	52.85	7.27
"	Fourth	87.37	59.68	27.69	5.26
persimilis	Second	151.47	132.79	18.69	4.32
"	Third	179.77	168.92	10.85	3.29
"	Fourth	100.66	99.41	1.25	1.12
		Homozygotes			
pseudoobscura	Second	155.03	36.89	118.14	10.87
"	Third	248.10	132.80	115.30	10.74
"	Fourth	168.96	114.21	54.74	7.40
persimilis	Second	153.14	91.82	61.32	7.83
"	Third	271.13	127.49	143.63	11.99
"	Fourth	186.95	101.65	85.29	9.24

the frequencies of subvital and supervital chromosomes by computing t values for the viability distribution, as follows:

$$t_{subvitals} = [(m_{het} - 2\sigma_{r\,het}) - m_{hom}]/\sigma_{r\,hom}, \text{ and}$$
$$t_{supervitals} = [(m_{het} + 2\sigma_{r\,het}) - m_{hom}]/\sigma_{r\,hom}.$$

The mean viability of the heterozygotes, m_{het}, is, by definition, equal to 100. The mean viabilities of the homozygotes, m_{hom}, are shown in the right-hand column in table 5. The estimated σ_r for the heterozygotes and the homozygotes are found in the rightmost column in table 6. The frequencies of subvital and supervital chromosomes are, then, the probabilities that a standard normal variable will fall below the calculated t.

The frequencies of the subvitals and the supervitals are reported in table 7. It can be seen that a decided majority of the quasi-normal second and fourth chromosomes in the natural populations of both *D. pseudoobscura* and *D. persimilis* are, in fact, subvital in homozygotes. Homozygosis for wild third chromosomes seems to be somewhat less deleterious, but even for this chromosome the estimates given in table 7 are much higher than those obtained by DOBZHANSKY, HOLZ and SPASSKY (1942) and quoted by DOBZHANSKY (1951) and elsewhere. These authors defined the frequencies of the subvitals and supervitals in an entirely different way, so that the present estimates are not comparable to the old ones. The present method of estimation is considered to be much more satisfactory than the old one.

Chromosomes which are supervital when homozygous are, as expected, quite rare.

STERILITY

Females and males homozygous for the quasi-normal, and for the less extreme semilethal chromosomes, were tested for fertility. The technique of testing was as described by PAVAN *et al.* (1951). The frequencies of chromo-

TABLE 7

Estimated percentages of the quasi-normal chromosomes which are subvital or supervital when homozygous.

Chromosome	*pseudoobscura*		*persimilis*	
	Subvitals	Supervitals	Subvitals	Supervitals
Second	93.5	0.1	84.4	<0.1
Third	41.3	0.7	74.2	4.0
Fourth	95.4	<0.1	98.4	0.4

somes which cause the homozygotes to be sterile are indicated in table 8. As a rule, a chromosome causes sterility either of homozygous females or of homozygous males. Chromosomes that make homozygotes of both sexes sterile are relatively few. Most of them belong to the semilethal or extreme subvital classes; the sterility of both sexes is probably due to a low vitality of the flies rather than to any specific disturbance of the reproductive functions.

VARIATIONS IN THE DEVELOPMENT RATES

It is known that some of the homozygotes for chromosomes derived from natural populations of *Drosophila pseudoobscura* show modifications of the development rates (DOBZHANSKY, HOLZ and SPASSKY 1942, and others). As a rule, homozygotes develop more slowly than do the normal heterozygotes. Much less frequently homozygotes with abnormally rapid development are encountered. In the course of the present work variations in the development rates have also been found quite frequently, both in *D. pseudoobscura* and in *D. persimilis*. An attempt to determine the frequencies of these variations from our data would however be too hazardous, because our test cultures were obtained by allowing the parent flies to oviposit for several days.

ALLELISM OF LETHALS

Chromosomes which are lethal to homozygotes can be preserved indefinitely by keeping them in balanced strains. Some of these strains were intercrossed, and the offspring of the intercrosses were examined to determine how frequently the lethals involved were allelic. Table 9 shows that only two of the intercrosses happened to contain allelic lethals. A pair of alleles were found in

TABLE 8

Percentages of the chromosomes which cause sterility when homozygous.

Species	Chromosome	Females		Males	
		Sterile %	Chromosomes tested	Sterile %	Chromosomes tested
pseudoobscura	Second	10.6 ± 3.3	85	8.3 ± 3.0	84
persimilis	"	18.3 ± 4.6	71	13.2 ± 4.1	68
pseudoobscura	Third	13.6 ± 3.7	88	10.5 ± 3.3	86
persimilis	"	14.3 ± 3.1	133	15.7 ± 3.1	134
pseudoobscura	Fourth	4.3 ± 2.1	93	11.8 ± 3.5	93
persimilis	"	18.3 ± 3.4	126	8.4 ± 2.5	119

TABLE 9

Frequency of allelic lethals in intercrosses within and between species.

Species	Chromosome	Lethals tested	Intercrosses	Alleles
pseudoobscura	Second	16	120	0
persimilis	"	20	54	1
pseudoobscura × *persimilis*	"	16 × 15	174	1
pseudoobscura	Third	12	66	0
persimilis	"	20	190	0
pseudoobscura × *persimilis*	"	12 × 20	191	0

second chromosomes of *D. persimilis,* and another pair in a second chromosome of *D. persimilis* and one of *D. pseudoobscura.* The data are insufficient to decide whether the frequencies of allelism are the same in intra- and in interspecific crosses.

DISCUSSION

Studies on the genetics of natural populations have emphasized the importance of a hitherto vaguely known but scarcely appreciated fact. Namely, populations of sexual and cross-fertilizing organisms contain great arrays of genotypes, no one of which can be regarded " normal " or " typical " for the species or population. Natural populations of Drosophila, and probably of other sexual organisms including man, consist mostly of more or less complex heterozygotes. The gene pool of each population contains a variety of gene alleles and of linked gene complexes. Natural selection maintains and augments this variety, provided that the variants give rise, in combinations with other variants present in the same population, to highly adapted heterozygotes. Most of the genotypes formed in any one population, so long as the historically established breeding system is adhered to, are coherent enough to enable their carriers to survive and reproduce in the ecological niches which the population occupies.

The situation becomes altered when the normal breeding system is interfered with. Genotypes can be obtained, by inbreeding or with the aid of special genetic techniques, which carry in duplicate certain chromosomes or gene complexes which in nature occur usually in heterozygotes. By such techniques, we have found that the homozygotes are often lethal, semilethal, subvital, sterile, or show various physiological or structural abnormalities. In foregoing articles of this series, this fact was interpreted as meaning that natural populations carry great stores of concealed recessive mutants.

This interpretation is valid as far as it goes, but it begins to appear in the light of new data that it may not go far enough. Tables 4 and 7 show that almost all chromosomes, at least in the populations studied, are more or less deleterious when homozygous. It may be that even the rare chromosomes which give " normal " or supervital homozygotes in certain environments may become subvital in other environments, as demonstrated for some chromosmes by DOBZHANSKY and SPASSKY (1944). But many of the chromosomes which are deleterious when homozygous give rise to highly adapted " normal " heterozygotes.

The store of concealed variability carried in an outbred population arises from two sources. First, the mutation process generates mutants most of which are more or less deleterious to their possessors in the environments in which the species normally occurs. With the exception of dominant lethals, deleterious mutants may persist in the population for some generations; the "life expectancy" is greater for recessive than it is for semi-dominant or dominant mutants. This is because recessives are sheltered from natural selection in heterozygotes.

Secondly, natural selection retains in populations gene alleles and gene complexes which give rise to highly adapted, heterotic, heterozygotes, even if the corresponding homozygotes are poorly adapted. Mutants which are useful neither when homozygous nor when heterozygous occur in natural populations because natural selection is not absolutely efficient. Mutant genes and gene complexes which are deleterious when homozygous but favorable when heterozygous are multiplied by natural selection until they reach certain equilibrium frequencies.

There is, of course, no sharp dividing line between the two kinds of concealed variability. Genetic variants which are heterotic in some environments may be deleterious in other environments (DA CUNHA 1951). Nevertheless, it is useful to distinguish the two kinds, if only because the importance of the gene complexes which are useful in heterozygotes but harmful in homozygotes is often lost sight of, especially in writings dealing with eugenics and population problems.

DOBZHANSKY and WALLACE (1953) found that the viability of chromosome homozygotes in *Drosophila pseudoobscura, D. persimilis, D. prosaltans,* and *D. melanogaster* is, on the average, subject to much greater environment variance (σ_e^2) than that of the corresponding heterozygotes. The developmental processes seem to be less adequately buffered against environmental disturbances in homozygotes than in heterozygotes. It is an attractive, though by no means proven, working hypothesis that heterosis in Drosophila is, at least in part, due to heterozygotes possessing, on the average, a more perfect homeostatic adjustment to the environment than is the case in homozygotes. A finding described in the present article is relevant at this point. Homozygotes for quasi-normal chromosomes exhibit a greater genetic variance (σ_r^2) than do heterozygotes (table 7). Now, this may mean that many wild chromosomes carry subvital mutant genes of varying strength. But the same result is expected also if most of the homozygotes possessed narrower environmental tolerances than most heterozygotes. These two explanations are certainly not mutually exclusive, and may, in fact, represent merely different ways of looking at the same phenomenon. What is, however, important is that the gene pools of natural populations are built in such ways that most of the genotypes which arise in every generation are sufficiently homeostatic to be "normal" under a certain gamut of environmental conditions.

The "normality" of heterozygous genotypes does not, however, mean that they give similar and adaptively equivalent phenotypes. "Normal" Drosophila

exists no more than a "normal" man. Appreciable genetic variance (σ_r^2) exists among heterozygotes for all the chromosomes of the two species tested (table 7). Genetic differences between heterozygotes for different chromosomes extracted from the same populations have been found also by CORDEIRO (1952) in *D. willistoni* and by WALLACE and KING (1952) in *D. melanogaster*. These findings are important: they show that the genetic variability in natural populations which we have studied is "concealed" and "potential" only in the sense that homozygosis brings out properties of the variants which could not be predicted from examination of the heterozygotes, and that crossing over and recombination of these variants yield a great abundance of new genotypes. Heterozygotes are less diversified than homozygotes because natural selection canalizes the developmental processes in the former, so that excessive deviations from the "normal" development characteristic for the species are avoided. But natural selection also maintains in populations sufficient genetic polymorphism to enable the species to exploit a variety of adaptive niches to which it has access.

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SUMMARY

Samples of populations of *Drosophila pseudoobscura* and *D. persimilis* were collected in some localities in California in which both species occur together. By means of a series of crosses (fig. 1), individuals were obtained which were homozygous for certain chromosomes derived from the ancestors collected outdoors. Control experiments yielded heterozygous individuals, in which the two chromosomes of a pair came from different wild ancestors.

Homozygotes, which carry pairs of identical chromosomes derived from natural populations, are often lethal, semilethal, subvital, sterile, or otherwise physiologically or structurally abnormal (tables 3, 7 and 8). *D. pseudoobscura* and *D. persimilis* do not differ significantly in the frequency of lethal and semilethal chromosomes (table 4). The mean viability of homozygotes for second and possibly for third chromosomes seems to be greater in the former than in the latter species, but in the fourth chromosomes the relationships seem to be reversed (table 5). In both species, a great majority of at least second and fourth chromosomes which are free of lethals and semilethals are subvital when homozygous (table 7).

It is concluded that the concealed, or potential, variability carried in populations of normally outbreeding species has two, probably overlapping, com-

ponents. It consists, first, of mutant genes which are deleterious to their carrier in homozygous and often also in heterozygous condition. The second, and perhaps more important component are genes and gene complexes which give rise to superior, heterotic, heterozygotes. Such genes are retained and, up to a certain point, multiplied by natural selection, even though they are more or less deleterious when homozygous. The gene pools of *D. pseudoobscura* and *D. persimilis* contain a great variety of gene complexes most of which, or perhaps all of which, are disadvantageous in homozygotes. Yet, the same gene complexes produce " normal " individuals when heterozygous in combination with other gene complexes from the same gene pool. The adaptive " norms " of these species are, then, arrays of complexly heterozygous genotypes. These genotypes condition developmental processes which are buffered against environmental disturbances which are recurrent in the habitats in which the species normally lives.

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