A STUDY OF INTERCHANGE HETEROZYGOSITY IN A POPULATION OF Datura meteloides

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Received March 6, 1961

CYTOLOGICAL studies of natural populations are revealing an increasing number of species, both plant and animal, whose populations are characterized by polymorphism for chromosomal rearrangements. Recent examples, for instance, have been found in the marine snail Purpura lapillus by Staiger (1954), in the grasshopper Moraba scurra by White (1956), in two species of cockroaches (Lewis and John 1957; John and Lewis 1958, 1959), and in several species of the genus Clarkia (refs. in Lewis and Lewis 1955). The studies on Drosophila populations, too numerous to cite individually, are of course the prime examples. Whatever may be the particular chromosomal basis of the polymorphism, whether interchange or inversion, the genetic effect in a balanced system is apparently the same: the protection from recombination of favorable combinations of genes which confer an adaptive advantage on the heterozygote compared with the homozygotes. In the case of inversion heterozygotes, the protected region lies within the inversion; in interchange heterozygotes in the interstitial regions between the centromere and the breakpoints of the interchange. Crossing over in these places leads to chromatids with duplications and/or deficiencies, hence non-crossover gametes are the only functional ones.

The genus Datura has been the subject of many studies of racial and specific differentiation with respect to interchanges (summarized in Avery, Satina and Rietsema 1959). However, a study of a population containing interchange heterozygotes with the aim of ascertaining whether they are favored by natural selection has not yet been carried out. It was of some interest when cytological examination of several plants of Datura meteloides growing wild in California showed them to be interchange heterozygotes (Snow 1959). From the associations observed it is obvious that several different chromosome arrangements are present in this species, as was reported by Satina (1953). She stated that at least four different chromosomal races exist, and worked out the end arrangements of chromosomes of two of them with respect to the standard strain of Datura stramonium (Avery et al. 1959). We have studied a population on the Davis campus which contains two independent interchanges in relatively high frequency, and have attempted a comparison of observed and expected frequencies of heterozygotes for one of them.

Natural history

D. meteloides DC. is a common perennial summer weed in the lower Sacra-
mento and San Joaquin Valleys and in Southern California. It occurs in dry creek bottoms, abandoned fields and waste areas, along roadsides and railroad rights-of-way. The species was probably introduced into California from northern Mexico, where it is native, before the American occupation of the state and was characteristically found around old Spanish settlements and Mexican ranches (JEPSON 1943). Active growth starts in late spring and flowering occurs in the first year from June to as late as October, depending upon local conditions. In favorable situations the plants may become 6–8 feet in diameter, and open a dozen flowers or more per day. The large, white, trumpet-shaped flowers open about sunset and wither in the heat of the next day. The original pollinators were undoubtedly sphinx moths. Two species, Phegetontius (Protoparce) sexta Johanson and P. quinquimaculata Haworth, have been captured while visiting our population. Bumblebees, solitary bees, and especially honeybees also serve as pollinators. Although the large size of the flower and the habits of the pollinators suggest a high degree of outcrossing, self-pollination was nevertheless frequent, since about half the plants in the population had short styles which placed the stigma amongst the stamens. These plants must have been almost entirely self-pollinated, since the anthers dehisced about noon of the day the flower was to open. The floral tube at this time was tightly folded around the five stamens, with the stigma in the center. Hence pollen from these natural selfings had about a nine hour lead over pollen carried to the stigma in the evening by insects. Controlled pollinations showed that the species is fully self-compatible. Furthermore, the long-styled plants were not entirely outcrossed, because the insects often visited several flowers on a plant successively. There was no correlation between style length and chromosomal type.

The population is located in waste ground which was at one time the bed of a creek. About 1935 a levee was built which diverted the creek into a new channel, and from that time until 1955 the place remained undisturbed. The area has been plowed for weed control every year since then, except for 1957 and 1958. A part of the population extends into a protected region where conditions are essentially the same as they were in the 20 years from 1935–1955. The Datura plants here occupy the banks and bottom of the old creek bed.

When we sampled the population in the summer of 1958, it contained an estimated 125 large mature plants, plus scores of seedlings. We have no certain knowledge of the usual life span, but it must be at least 5–10 years in favorable circumstances. However, when the creek flowed through the area much of the population may have been in effect annual, since its flooding in winter would probably wash out a great many plants. Also the weed control plowing done in the two years previous to our study, in effect, made the population reproduce annually. These two disturbances have undoubtedly greatly decreased the average generation time.

A few plants of D. stramonium were also found in the vicinity. No evidence of hybridization was found, and indeed none would be expected, since these two species are among the most incompatible in the genus. The hybrids which SATINA (1953) studied had to be obtained by embryo culture.
METHODS

Buds for cytological examination were fixed in Carnoy's solution (6 parts absolute alcohol, 3 chloroform, 1 glacial acetic acid) since this causes less swelling of the chromosomes than acetic alcohol. Staining was done with acetocarmine. Pollen fertility was estimated by staining mature pollen grains with cotton blue in lactophenol. Anthers of flowers to be pollinated were removed the day before the flower would normally open and the pollination was made immediately. A length of drinking straw with the end folded was then slipped over the style, and the corolla was fastened shut by a band of scotch tape to prevent the moths from knocking off the straw. No difficulty was experienced in getting abundant seed set, except for certain pollinations made during a few days of extremely hot weather.

RESULTS

Cytology of the interchanges: Two interchanges, distinguishable by the meiotic configurations they produced at metaphase I in heterozygous individuals, were found in the population. Different chromosomes are involved in each case because plants simultaneously heterozygous for both interchanges occurred. The "standard" sequence of ends of the chromosomes involved in the A interchange may be designated 1-2, 3-4, while the interchanged sequence A' would be 1-3, 2-4. In similar fashion, the standard B sequence would be 5-6, 7-8; the interchanged B' sequence, 5-7, 6-8. One plant which formed 12 bivalents was arbitrarily designated as being homozygous for the standard sequence of both arrangements, and was given the formula AABB. If cytological examination showed that a certain plant was heterozygous for both the A and B interchanges, it was indicated as AA'B'B'. However, a plant heterozygous for only one interchange may have been homozygous for either the standard or the interchanged arrangement of the other (for instance, an AA' heterozygote could have been of the type AA'BB or AA'B'B'). A plant forming 12 bivalents could have been any one of four different chromosomal types. The complete chromosomal formula of plants other than the double heterozygotes was therefore determined by crossing to the standard plant and examining a plant of the progeny. A 12 bivalent plant, for example, which gave offspring heterozygous for the B ring would have the formula AAB'B'. In analogy with the F2 genotypic distribution of a dihybrid cross, nine chromosomal types were expected in the population, and all but one (A'A'B'B') were found.

Plants heterozygous for the A translocation characteristically formed a ring of four chromosomes at metaphase I (Plate I, 1). The orientation of this ring was almost always disjunctural, so little gametic abortion was expected. The relevant data on meiotic behavior and pollen abortion are given in Table 1. Judging from the regularity of ring formation, the interchange must have involved large portions of the chromosome arms concerned.

In contrast to the AA' heterozygotes, BB' heterozygotes formed a variety of configurations, the most frequent being a branched association of four chromo-
PLATE I.—Photographs of interchange heterozygotes at metaphase I. In all cases the interchange associations of four chromosomes lie next to the rightmost bivalent. All figures × 1500. Figure 1, AA' heterozygote, showing the characteristic disjunctural orientation of the ring (the connection between the upper left and lower right chromosomes is out of focus); Figures 2, 3, 4, BB' heterozygotes, showing three types of association. Compare these figures with those of Plate II, Figures 3, 5, and 8, respectively.

TABLE 1

Meiotic behavior and pollen abortion in AA' and BB' heterozygotes

<table>
<thead>
<tr>
<th>Orientation of interchange complex at metaphase I</th>
<th>Distribution of chromosomes at anaphase I</th>
<th>Percent pollen abortion†</th>
</tr>
</thead>
<tbody>
<tr>
<td>Alternate</td>
<td>Adjacent</td>
<td>12 + 12</td>
</tr>
<tr>
<td>AA' heterozygotes</td>
<td>92</td>
<td>8</td>
</tr>
<tr>
<td>BB' heterozygotes</td>
<td>See plate II</td>
<td>94</td>
</tr>
</tbody>
</table>

* Based on 100 cells.
† Based on three anthers from each of five plants, about 1000 pollen grains scored per anther. Pollen abortion of AABB homozygotes, 2.38 ± 1.30 percent; of AA'BB' heterozygotes, 41.10 ± 2.40 percent.

somes (Plate I, 2–4). Camera lucida drawings of most of the configurations found are shown in Plate II. All may be accounted for on the assumption that the B translocation involved the reciprocal exchange of relatively small segments,
PLATE II.—Drawings and chromatid diagrams of the interchange associations of BB' heterozygotes. Figures 1–8, camera lucida drawings of types of metaphase I associations. A, chromatid diagram of prophase interchange association (the numerals indicate regions where chiasmata may form); B, C, diagrams of two associations with chiasma formation in regions 1, 2, 3, 6, and 1, 2, 3, 4, 5, 6, respectively (compare B and C with Figures 1 and 8, above). The lower two lines of the plate show the frequency of types of metaphase I associations found in 118 cells of a BB' heterozygote.
so that chiasmata were frequently formed between the centromeres and the point of interchange (Plate II, A). The frequencies of the various associations observed in a preparation where the PMC's were exceptionally well fixed and stained are also given in this plate. The great majority of these associations should lead to chromatid nondisjunction even with a 2-2 distribution to the poles. Pollen abortion should therefore be considerable, and this was found to be so (Table 1). This table also shows that telophase distribution was essentially as regular as that of AA' heterozygotes. Thus, little pollen abortion was caused by lagging chromosomes.

Several plants were found in the wild population and in the garden cultures which were heterozygous for both translocations. Unexpectedly, the pollen abortion was no higher in these plants than in plants heterozygous for the B translocation alone. It should be pointed out that pollen abortion is quite variable even among plants of the same chromosomal type, apparently in response to environmental factors. A few days of exceptionally hot weather in the summer of 1958 caused a great increase in abortion, even in homozygotes, and most of the pollinations made during these days failed. On the basis of chromatid diagrams one would expect about 50 percent pollen abortion in BB' plants. It is entirely possible that some pollen grains from these heterozygotes which looked normal were in fact nonfunctional.

Analysis of data: Cytological determinations were made on 62 plants from the campus population. In addition, 109 small seedlings with 2–3 leaves were transplanted into the experimental field, of which 102 survived and 35 were studied cytologically. The frequencies of the various cytologically recognizable types found in the two samples are given in Table 2, in addition to the cytological type of 24 double homozygotes from the campus population which were progeny tested. It will be seen that the two samples do not differ significantly with respect to all classes considered together nor as to the A and B arrangements considered.

<table>
<thead>
<tr>
<th>Cytological type</th>
<th>12(2)</th>
<th>A ring + 10(2)</th>
<th>B ring + 10(2)</th>
<th>A+B rings + 8(2)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Campus population*</td>
<td>37</td>
<td>15</td>
<td>8</td>
<td>2</td>
</tr>
<tr>
<td>Field transplants</td>
<td>24</td>
<td>4</td>
<td>7</td>
<td>0</td>
</tr>
<tr>
<td>Totals</td>
<td>61</td>
<td>19</td>
<td>15</td>
<td>2</td>
</tr>
</tbody>
</table>

Tested double homozygotes:

<table>
<thead>
<tr>
<th>Type</th>
<th>12(2)</th>
<th>Comparison</th>
<th>P</th>
</tr>
</thead>
<tbody>
<tr>
<td>AABB</td>
<td>13</td>
<td>1. Homogeneity between samples</td>
<td>.2–.3</td>
</tr>
<tr>
<td>AAB'B'</td>
<td>10</td>
<td>2. Homogeneity of A arrangement homozygotes/heterozygotes between samples</td>
<td>.1–.2</td>
</tr>
<tr>
<td>A'A'BB</td>
<td>1</td>
<td>3. Homogeneity of B arrangement homozygotes/heterozygotes between samples</td>
<td>.5–.7</td>
</tr>
<tr>
<td>A'A'B'B'</td>
<td>0</td>
<td>4. Independence of A and B arrangements in total sample</td>
<td>.3–.5</td>
</tr>
</tbody>
</table>

* Two tetraploids were also found.
INTERCHANGE HETEROZYGOSITY

separately (comparisons 1, 2, and 3). Neither is there any evidence of an interaction or preferential association of any A and B arrangement (comparison 4). This latter evidence is supported by results from the 24 double homozygotes tested, in which essential equality was found between AABB and AAB'B' plants (13:10), and between A'A'BB and A'A'B'B' plants (1:0).

To test for agreement between observed and expected frequencies of the cytological types we must first determine if the population is in equilibrium. With no inbreeding, equilibrium with respect to two independent arrangements will be reached in about five generations (LI 1955, pp. 89–90), inbreeding serving to delay its attainment (LI 1955, p. 142). Although we cannot be sure that equilibrium has been reached, as we studied only one generation, we deduce on the basis of the history of the population that there has probably been enough time.

The estimates of the frequencies of the chromosomal types \( p_A \) and \( p_B \) are best obtained by maximum likelihood, using Wright's equilibrium expression, the generalized form of the familiar Hardy-Weinberg formula:

\[
(p^2 + pqF) + 2pq(1-F) + (q^2 + pqF).
\]

The use of this expression also involves estimation of \( F \), the inbreeding coefficient. Since the A and B arrangements seem to be independent of one another, estimates for \( p \) and \( F \) may be obtained separately, which greatly simplifies the calculations and, in this case, allows us to make a statistical comparison. The data are set out in Table 3.

Taking partial derivatives with respect to \( p \) and \( F \) of the logarithm likelihood expression

\[
L = a_1 \log m_1 + a_2 \log m_2 + a_3 \log m_3 + a_4 \log m_4,
\]

substituting \( q \) for \( 1 - p \), and equating to 0, we have

\[
\frac{\partial L}{\partial p} = A \left( \frac{2(2p-1)(1-F)}{1-2pq(1-F)} + a_2 \frac{1-2p}{pq} + a_3 \frac{(2p-1)(1-F)+1}{p^2 + pqF} + a_4 \frac{(2p-1)(1-F)-1}{q^2 + pqF} \right) = 0,
\]

\[
\frac{\partial L}{\partial F} = A \left( \frac{2pq}{1-2pq(1-F)} + a_2 \frac{-1}{1-F} + a_3 \frac{pq}{p^2 + pqF} + a_4 \frac{pq}{q^2 + pqF} \right) = 0,
\]

where \( A = a_1 - a_2 - a_4 \). These simultaneous equations may now be solved by iteration.

There is, however, an easier way to obtain the estimates. Bailey (1951) has shown that when the number of parameters being estimated is equal to the degrees of freedom, two each in this case, then the maximum likelihood estimates are those obtained by equating the observations to their expectations. Thus we may write.

\[
a_2 = 2n_1 p(q(1-F))
\]

\[
a_3 = n_2 (p^2 + pqF)/(1-2pq(1-F))
\]

\[
a_4 = n_4 (q^2 + pqF)/(1-2pq(1-F)).
\]
TABLE 3
Organization of data for maximum likelihood estimation of \( p \) and \( F \)

<table>
<thead>
<tr>
<th>Class</th>
<th>Frequency</th>
<th>Expectation</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>A data</td>
<td>B data</td>
</tr>
<tr>
<td>Both homozygotes ((a_1))</td>
<td>76</td>
<td>80</td>
</tr>
<tr>
<td>Heterozygotes ((a_2))</td>
<td>21</td>
<td>17</td>
</tr>
<tr>
<td>Standard homozygotes ((a_3))</td>
<td>23</td>
<td>14</td>
</tr>
<tr>
<td>Interchange homozygotes ((a_4))</td>
<td>1</td>
<td>10</td>
</tr>
</tbody>
</table>

\( p = 2n_1a_1a_3 - a_2(a_3 - a_4)/2n_1(a_3 + a_4), \)

\( F = 1 - 2n_1a_2(a_3 + a_4)^2/[2n_1a_3 - a_2(a_3 - a_4)] [2n_2a_4 + a_2(a_3 - a_4)]. \)

These values of \( p \) and \( F \) satisfy the partial derivatives above, and hence are maximum likelihood estimates.

The variances are given as

\( V_p = I_{pp}/\delta \)

\( V_F = I_{pp}/\delta \)

where

\( \delta = \begin{vmatrix} I_{pp} & I_{pf} \\ I_{pf} & I_{ff} \end{vmatrix} \)

\( I_{pp} = n \sum [\frac{1}{m} (\frac{\delta m}{\delta p})^2], \)

\( I_{ff} = n \sum [\frac{1}{m} (\frac{\delta m}{\delta F})^2], \)

\( I_{pf} = n \sum [\frac{1}{m} (\frac{\delta m}{\delta p}) (\frac{\delta m}{\delta F})], \)

in which \( n = 97 \) for expectations \( m_1 \) and \( m_2 \), and 24 for expectations \( m_3 \) and \( m_4 \).

The standard errors are obtained by taking the square root of the variances.

As a result of these calculations we obtained the following estimates and standard errors:

\( p_A = .8591 \pm .0372 \quad p_B = .5687 \pm .0830 \)

\( F_A = .1058 \pm .1835 \quad F_B = .6427 \pm .0798 \)

Both values of \( p \) and \( F \) are significantly different from each other, while \( F_A \) is not significantly different from 0.

The significant difference between the \( F \) values for the A and B data is unexpected, since the same sample of plants was used to obtain both. On the basis of our natural history observations the \( F_B \) value would seem to be the more likely inbreeding coefficient of the population. If this is so, and under the assumption of equilibrium, the low \( F_A \) value can be explained by selection in favor of \( AA' \)
heterozygotes, which would have the statistical effect of lowering this inbreeding coefficient.

We have already given our reasons for thinking that the population is at equilibrium. The adoption of \( F = .6427 \) as the population inbreeding coefficient is supported by the following reasoning. FYFE and BAILEY (1951) have given the relation between \( F \) and \( \alpha \), the proportion of successful pollinations in which the source of pollen was equally likely to have been from any plant in the population, as \( \alpha = 1 - \frac{F}{1 + F} \). Using \( F = .6427 \), \( \alpha = .217 \), i.e., about 78 percent of the total number of pollinations were selfs. This figure is somewhat of an overestimate, since the above relationship is not strictly applicable to a population such as ours where the inbreeding coefficient will be increased merely as a result of small population size. As an approximation we may take the proportion of self-pollinations to be about 70 percent, in which case \( F \) would be about .54, a figure still within the 99 percent confidence interval of the maximum likelihood estimate. We therefore think that the \( F_b \) value is characteristic of the population and that the low value of \( F_A \) is best explained by attributing a selective advantage to AA' heterozygotes.

We can test the A data by \( x^2 \) using .6427 as the population \( F \) value, since in doing so we gain a degree of freedom. This necessitates a recalculation of \( p_A \) (since \( p \) and \( F \) are correlated when estimated as we have done), which now turns out to be \( .7518 \pm .0617 \). The \( x^2 \) comparison for the A data may then be made as follows:

Expected AA' heterozygotes = 12.93370 (deviation \( d_1 \) = + 8.06630)
Expected AA homozygotes = 18.97375 (deviation \( d_2 \) = + 4.02643)
\[ x^2 = \frac{d_1^2}{97}(E_{AA'}) \left(1-E_{AA'}\right) + \frac{d_2^2}{24}(E_{AA}) \left(1-E_{AA}\right) = 9.88 \]
where \( E_{AA'} \) and \( E_{AA} \) are the expected proportions of AA' heterozygotes and AA homozygotes, respectively. AA' heterozygotes (and AA homozygotes) were in excess of expectation, \( P_{(1)} \) being \( < .005 \). This method of analysis does not allow us to say anything about the adaptive significance of the B arrangements, since we have estimated both \( p_B \) and \( F_b \) from the data, and have no degrees of freedom available for a \( x^2 \) test.

DISCUSSION

We have postulated adaptive superiority of the AA' heterozygotes after comparing observed and calculated zygotic frequencies. Both WALLACE (1958) and NOVITSKI and DEMPSTER (1958) have cautioned against improper use of this procedure. Their analyses, however, apply principally to populations which are not in equilibrium. LEWONTIN and COCKERHAM (1959) have shown that comparisons of the type we have made are essentially tests of whether the square of the adaptive value of the heterozygous class is equal to the product of the adaptive values of two homozygous classes. This is a much less restrictive condition to test for than that the adaptive values of the three classes be equal. Nevertheless, in an equilibrium population a comparison of observed and expected zygotic frequencies is still a valid procedure, and, in such a population, an excess of heterozygotes
is good *prima facie* evidence of their adaptive superiority (i.e., that a balanced polymorphic system exists). Actually, trial calculations will show that in a hypothetical population with heterozygote superiority a significant excess of heterozygotes can often be demonstrated with rather small samples of adults (50–100) even when the population is not yet in equilibrium, if the selective coefficients of the homozygotes are rather larger (say 0.2–0.5).

The \( \chi^2 \) test of the A data showed that AA homozygotes were also in excess of expectation. This is not consonant with the assumption of a balanced polymorphic system, as it implies selection for them also, or in other words, that the population is not in equilibrium. This excess, which amounted to only about four plants of the 24 double homozygotes tested, may be due to sampling error. On the other hand, it may indicate that our assumption of equilibrium was wrong, a possibility we must always entertain owing to the shortcomings of our data.

In the case of a balanced polymorphic system, the high selective advantage which must be attributed to the AA′ heterozygotes to obtain a significant \( \chi^2 \) with a sample as small as ours may seem unlikely, but several cases are known where the heterozygotes must enjoy a very considerable advantage over the homozygous sibs. Thus, Dobzhansky and Pavlosky (1955) and Pavan, Dobzhansky and da Cunha (1957) have described cases in Drosophila populations where inversion heterozygotes occurred in significantly greater frequency than 50 percent, unequivocal evidence of strong selection in their favor. Jain and Allard (1960) calculate that in a composite barley variety cross, individuals heterozygous for certain chromosome segments identified by marker genes leave twice as many progeny as the corresponding homozygotes. It seems probable that the magnitude of the selective advantage enjoyed by a heterozygote in a balanced polymorphic system may be related in part to the life cycle and population size. In a Drosophila population, for example, where several generations are produced in a single year and population sizes may be very large, a relatively small heterozygote advantage may be sufficient to ensure the maintenance of a close adaptation to small temporal changes in the environment. A case in point is probably that of *D. pseudoobscura*, studied by Epling, Mitchell and Mattoni (1957), where marked seasonal fluctuations in inversion frequencies occurred which were regularly repeated year after year. On the other hand, organisms with much longer life cycles and smaller populations, such as some annual or perennial plants, must maintain a greater degree of tolerance to temporarily unfavorable conditions (which may extend over periods of years), and hence selective advantages possessed by heterozygotes might be rather large. An experimental demonstration of heterozygote superiority in a balanced polymorphic system could probably be made most easily with such organisms.

The cytological behavior of the A interchange is concordant with its presumed genetic effect. No configurations of the A ring were found which clearly indicated chiasma formation in the interstitial regions. In marked contrast, chiasmata in the B ring were formed in these regions in about 85 percent of the cells. Genetic recombination is thus prevented in the interstitial regions of AA′ heterozygotes but is frequent in the B interstitial segments, the more so in region 2 than in 5.
It might appear that a selective advantage should also be attributed to the B ring, offsetting the sterility which it causes. We do not think this assumption necessarily follows. Assuming ovule abortion to be about the same as pollen abortion in BB' plants, such plants must produce about 40 percent more ovules than will mature into seed if they are to produce approximately the same amount of seed as a homozygote. Examination of capsules of any plant always shows a number of tiny, abortive seeds, though we cannot say for sure whether their number reaches 40 percent of the normal set. This is not unlikely, however. In the case of BB' plants the 40 percent of abortive embryos would be the effect of their genetic unbalance which resulted from chromatid nondisjunction, while in the several homozygous types abortion would be determined by other factors that have their effect after fertilization and the beginning of embryo growth. A vast excess of pollen is produced (20,000 grains per anther (100,000 per flower) is certainly a conservative estimate), hence a 40 percent reduction of pollen fertility would probably be of little consequence.

The A interchange, which has resulted from the exchange of relatively large segments, is the type commonly found in nature. In most cases a necessary condition for the successful integration of such a translocation into a polymorphic system is the directed segregation of the ring, unless the sterility caused by nondisjunction is greatly overbalanced by the superiority of the heterozygote. Such is possibly the case in *Paeonia californica* (Walters 1942). In the genus Datura, many interchange rings undergo directed segregation (Bergner, Satina and Blakeslee 1933). This type of chromosomal preadaptation is also characteristic of cockroach chromosomes (John and Lewis 1959). The B type of ring is quite uncommon. We know of only two instances of the natural occurrence of this kind of translocation. One is in the marine snail, *Purpura lapillus*, where Staiger (1955) found less than one percent of heterozygous individuals. The other case concerns two primary spermatocytes of an individual of the grasshopper *Chorthippus brunneus*, found by John, Lewis and Henderson (1960). The interchange had obviously arisen in this individual, and the sterility it would cause in grasshoppers would no doubt rapidly lead to its disappearance from the population.

Artificially produced B-type interchanges have been reported for rye (Price 1959) and *D. stramonium* (Avery et al. 1959). Some of the configurations illustrated by Price are almost identical to those found in *D. meteloides*.

*D. stramonium* is the classic example of a species composed of structurally homozygous races differing from one another in their chromosome arrangements (Blakeslee, Bergner and Avery 1937). This species, unlike *D. meteloides*, is almost completely self-pollinated, since all plants have short styles. With such a breeding system an interchange heterozygote must have an extremely high selective advantage if it is to be retained in the population against the pressure of almost complete inbreeding. Though no direct studies have been made of interchange heterozygotes in natural populations of this species, it seems unlikely that their frequency would approach that found in our population of *D. meteloides*. This is not to say, however, that interchange heterozygosity cannot have
played an important role in racial differentiation in *D. stramonium*, but only that that role was probably a different one from the role played by heterozygotes in a balanced polymorphic system. Thus, it is possible that the establishment of new positional relationships of genes through interchange could at times have a selective advantage (internal intrachromosomal coadaptation, Lerner 1958). An example of such coadaptation in a lower organism is provided by the series of closely linked loci controlling related biochemical reactions in Salmonella, where where Demerec and Hartman (1959) have given reasons for believing these sequences are maintained by selection. If an advantageous sequence were established by interchange in Datura, the cytological polymorphism would be transitory since the direction of selection would be toward homozygosity for the new arrangement. Strong inbreeding would of course shorten the period of transition. This explanation has been invoked to account for the establishment of structurally homozygous races in *Clarkia dudleyana* (Snow 1960), a predominantly outcrossing (though self-compatible) species, and it can be equally well applied to a similar situation found in *Clarkia amoena* subsp. *caurina* (Hakansson 1942, as Godetia Whinney; Snow, unpublished), a predominantly self-pollinating subspecies. If the origin of homozygous chromosomal races has been due to the influence of natural selection acting upon favorable sequences of loci, the various chromosomal arrangements ought to be correlated with the adaptedness of the race to its particular habitat, an hypothesis which should be amenable to test by properly designed experiments.

**SUMMARY**

A wild population of *Datura meteloides* was studied which contained two different, independent types of interchange heterozygotes, distinguishable by their meiotic metaphase configurations. Heterozygotes for the A interchange regularly formed a ring of four chromosomes with highly directed disjunction; meiosis was very regular; pollen sterility was about ten percent compared with the two percent sterility of homozygotes. This interchange apparently resulted from the translocation of relatively large segments of chromosome arms. Heterozygotes for the B interchange formed a variety of configurations, all of which can be accounted for by assuming that this interchange involved relatively short segments, allowing chiasma formation in the interstitial regions. Pollen abortion in these heterozygotes was about 41 percent, owing to chromatid nondisjunction from the ring.

Cytological examination was made of 97 plants from both wild and field-grown samples. Maximum likelihood estimates of the frequency of the "standard" chromosomal arrangements were \( p_A = .8591 \pm .0372 \), and \( p_B = .5687 \pm .0830 \); estimates of the inbreeding coefficients were \( F_A = .1058 \pm .1835 \), and \( F_B = .6427 \pm .0798 \). The significant difference between the two \( F \) values is surprising since all the estimates were made from the same total sample of plants. Circumstances about the population incline us to believe that it is in equilibrium, while observations of the habits of the pollinators (sphinx moths, bees) and of
the proportion of plants with short styles support the assumption that the $F_B$ value is the inbreeding coefficient characteristic of the population. The low $F$ value for the A data can then be explained on the basis of a higher fitness of the AA' heterozygotes, i.e., that the A arrangements are components of a balanced polymorphic system. A $\chi^2$ comparison indicated as excess of both AA' heterozygotes and standard type (AA) homozygotes. The excess of AA plants is disturbing, since it is not compatible with the assumption of equilibrium and a balanced polymorphic system; however, the excess is relatively small and may be due to sampling error. Our method of analysis prevents us from drawing conclusions about the relative fitness of BB' heterozygotes. There was no evidence for any preferential association of the various arrangements.

ACKNOWLEDGMENTS

We are happy to express our appreciation to Drs. Everett Dempster and R. W. Allard, who instructed us in the method of maximum likelihood, patiently answered our many queries, and contributed much of value in discussions of the data. Our thanks also go to Dr. Henry Alder for statistical advice, and to Dr. Gerald A. Powell for identification of the sphinx moth pollinators.

LITERATURE CITED


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