THE SPECIES PROBLEM IN DATURA

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The discussion of our topic will be largely an explanation of the Datura exhibit. The exhibit, however, has an advantage in that the exhibitors who carried on the work are able to explain their own findings in person.

Our present summary is based on the results of a number of collaborators. A. D. BERGNER, with S. SATINA, is in charge of the work in cytology and J. L. CARTLEDGE of the work in pollen abortion; J. T. BUCHHOLZ is leader in the studies on pollen-tube behavior, E. W. SINNOTT in studies on internal anatomy and A. G. AVERY has charge of the care of the plants and the tabulation of the segregating cultures. BELLING early demonstrated that chromosomes can be matched up like dominoes and worked out the chromosomal basis of the "B" races. It is easy to play with dominoes after we have been taught the rules of the game.

The species problem in Datura has had an evolution. At first we attempted to analyze the differences between species by means of hybridization, selfing, and isolation of types through continued inbreeding, without knowing much about the genetic elements with which we were dealing. Our present program attempts to relate differences between species in terms of established standards. That so far our work has been more with our standards than with different species may not be a disadvantage to our ultimate objective. In triangulation, the surveyor finds it practical to first know accurately his base lines. To the geneticist pure living reagents of known composition are as important as are pure chemical reagents to the chemist.

The investigations in Datura may be logically classified under four main headings, although these divisions do not represent the actual order of research nor of our discussion. We have studied: (1) differences within a standard line or biotype, our line 1; (2) intraspecific differences within a single standard species, *Datura stramonium;* (3) interspecific differences between 7 of the species within the genus Datura; (4) synthesized or laboratory new "species."

DIFFERENCES WITHIN THE STANDARD LINE

Our standard line 1 of *D. stramonium* has been inbred by selfing for 19 generations and has been once passed through a haploid. Each season we grow a considerable number of plants of this standard race and scrutinize them closely for possible morphological variations. In addition the pollen of several hundred line 1 plants is examined microscopically each year for ab-

normalities that might not show in external appearance. Very few variants have been discovered during our experience with this line that could be attributed to gene mutations.

Chromosomal mutations, however, have been not infrequent. Out of 13,345 line 1 plants grown in our breeding plots from 2n parents, 22 or 0.17 percent have been identified as 2n+1 chromosomal abnormalities.

Since there are in Datura 12 different kinds of chromosomes each with two different ends, there are 24 different kinds of ends. In line 1 normal plants the ends of the chromosomes have been given consecutive numbers beginning with the odd number. The largest chromosome, therefore, is labeled [1 · 2] and the smallest [23 · 24]. They may be represented diagrammatically as dominoes with numbered ends. In assigning consecutive numbers to the two ends of the line 1 chromosomes we do not wish to imply that this race is the most primitive from the evolutionary standpoint. The numbering merely emphasizes again that we are interpreting Daturas in terms of this single standard.

As shown in table 1, the 2n+1 types have been classified as primary types with the extra chromosome unmodified and like the other two in the set, as secondaries with the extra chromosome composed of one half doubled, as tertiaries with the extra chromosome composed of parts of two normal chromosomes, and as fragment types with only a part of a chromosome extra. These types have been figured and discussed elsewhere (BLAKESLEE 1927, 1929, 1931, BLAKESLEE and BELLING 1924) and one of the primaries with its two secondaries is shown in figure 3, so we need not dwell on their peculiarities. Capsules are convenient parts with which to illustrate their diversity in external appearance, but probably all parts of the plant are more or less strongly affected by the presence of a particular extra chromosome. SINNOTT, for example, has found marked differences in their internal anatomy, CARTLEDGE in pollen grains, and BUCHHOLZ in the behavior of their pollen tubes as can be seen in their respective exhibits.

Table 1 shows types with extra chromosomes, all of which are in line 1 and have appeared spontaneously more than once with the exception of the tertiary 2n + [2 · 9] which has been found but a single time. Since they all belong to the same highly purified standard line, we can assume with a fair degree of assurance that the peculiarities of these types are due entirely to the extra chromosome. We have in these types, therefore, a means of learning something about the assemblage of factors in normal whole chromosomes and in major parts of chromosomes, at least of such factors as induce recognizable effects when present in an extra dose. This is especially true of

TABLE 1

Primary, secondary, and tertiary 2n + 1 types in standard line 1.
(Extra chromosomes are shown with numbered ends)

CHROMOSOME SIZE CLASS	SECONDARY CHROMOSOMES	PRIMARY CHROMOSOMES	SECONDARY CHROMOSOMES	TERTIARY CHROMOSOMES	GENES LOCATED IN PARTICULAR CHROMOSOME
L°	[°1·1°] (Py)	[°1·2] (Rl)	[2·2] (Sg)	[2·9] (ES)	p_{l7}; R_{f4} in 1·2 or 11·12
l	[3·3] (Sm)	[3·4] (Gs)	.. ..	[4·6] (Mp)	f_w, B_z in ·4; Q_S and p_{a_4} in 3·4, half not determined
l	[5·5] (St)	[5·6] (Bk)	[6·6] (At)	[4·6] (Mp)	
M°	[7·7] (Un)	[7·8°] (El)	..	..	r_{f_6} in 7·8 or 15·16
M°	[9·9] (Mt)	[9·10°] (Ec)	[°10·10°] (Th)	[2·9] (ES)	M_{S_1}
M°	[11·11] (Wd)	[11·12°] (Ck)	.. ..	.. ..	i_n, t_f, e, s_{kv}, in ·12; R_{f_4} in 11·12 or 1·2
M	[13·13] (Mb)	[13·14] (Mc)	[14·14] (Ml)	[13·18] (X)	a_{l_2}
M°	[15·15] (Sc)	[15·16°] (Rd)	.. ..	.. ..	t_c in ·16; p_{l_8} in 15·16, half not yet determined; r_{f_5} in 15·16 or 7·8
m	[17·17] (Df)	[17·18] (Pn)	.. ..	[13·18] (X)	c, w_t in ·17; p in ·18
m°	[19·19] (Dv)	[19·20°] (Sp)	..	..	s_h, p_{a_1}
S°	..	[°21·22] (Gl)	..	..	s_c, b_b, p_{l_3}
s°	..	[23·24°] (Ix)	..	..	s_w; z_g

The ends 1°, 8°, 10°, 12°, 16°, 20°, 21° and 24° are characterized by terminal humps.

primaries which may be secured by segregation from 3n parents where no break has been necessary in their formation as is the case for secondaries or tertiaries. These 12 chromosomes of line 1, together with parts of some of them, are our genetic standards for the genus Datura. They are not as precise standards as we might wish since they are obviously blocks of many

interacting units, but they have the advantage we believe of representing more surely the normal chromosomal constitution than standards made up of allelomorphs inferred from single gene mutations. We have in our standard race a rough genetic scale or balance with which we can weigh any chromosome that can be added to this race in terms of the unbalance which it exerts over the standard 2n chromosomes. How individual chromosomes of other species may be isolated and weighed as extras in our standard line 1 balance will be discussed later.

The tertiary forms that have arisen spontaneously in line 1 show that chromosomes may be formed with different end arrangements from those of our standard line 1 chromosomes which are used as the ultimate testers for ends of chromosomes. The standard chromosomes are separable into different size classes and in the majority a terminal hump distinguishes one end from the other. A humped chromosome is capable of identifying the end of a tertiary chromosome to which it is attached at reduction division. The double half chromosomes in secondary 2n+1 types are of especial value as chromosomal testers since both ends are identical. It will be noted from table 1 that we have secondaries for all the primaries except for the smallest two chromosomes, [21 · 22] and [23 · 24]. The [21 · 22] chromosome and the [23 · 24] chromosome are marked by a terminal hump. For both these two chromosomes other testers for their ends are also available in good tertiaries not listed in the table since they did not arise spontaneously within line 1.

A consideration of other line 1 types will be more profitable after a discussion of intraspecific differences.

INTRASPECIFIC DIFFERENCES

It was early seen that in *D. stramonium* there were cryptic races in nature in respect to the breeding behavior. Thus only the 2n + [17 · 18] primary gave trisomic ratios when heterozygous for "A" whites whereas both the primaries 2n + [1 · 2] and 2n + [17 · 18] threw abnormal ratios when heterozygous for "B" whites. By means of ratios from a more convenient type, "Nubbin," a considerable number of our white and purple races from nature were classified into A's and B's. BELLING (BELLING AND BLAKESLEE 1926) showed that the basis for this classification was segmental interchange in the origin of the "B" race such that instead of the [1 · 2] and the [17 · 18] chromosomes of line 1, which was an "A" race, we had in the "B" race the chromosomes [1 · 18] and [2 · 17]. These two chromosomes we had already had as extras in 2n+1 tertiary types which appeared to be related to both the [1 · 2] and the [17 · 18] primary 2n+1 forms.

Instead of using breeding tests for the identification of "A" and "B" races, we studied the configurations in the F_1 generations between line 1 and various races from nature. As might have been expected "B" races induced a large circle of 4 when combined with line 1 chromosomes having the formula

$$\begin{array}{ccc} 1 \cdot 2 & - & 2 \cdot 17 \\ | & & | \\ 1 \cdot 18 & - & 18 \cdot 17 \end{array}$$

BERGNER with the help of SATINA and others (BLAKESLEE 1929) has studied about 550 races from nature by means of the configurations which they induce in F_1 generations with our standard line 1. As seen by the table, only 5 cryptic chromosomal types have surely been discovered in nature in *D. stramonium*. Three of them differ from line 1 by interchange of segments of non-homologous chromosomes and one apparently by interchange of humps. A rather wide range of countries is represented and it seems unlikely therefore that a further search would greatly increase this number. Types such as these which are homozygous for modified chromosomes are called prime types. Each of the prime types from nature has been repeatedly backcrossed to line 1, chiefly through compensating types, until each resembles line 1 in appearance and presumably has all line 1 chromosomes except the two modified by segmental interchange. Presumably they now have practically the same assortment of genes since they are alike in appearance, but the grouping of the genes is different due to the interchanged chromosomes. Since the chromosome peculiarities are not obvious they have been called "cryptic types." Modified chromosomes from each prime type have been added to line 1 to form 2n+1 tertiary forms. The 2n + [1 · 18] and the 2n + [2 · 17] types, for example, shown in the exhibit and on the screen (BLAKESLEE 1927, plate 10) are to be classed as tertiary forms in terms of line 1. They are primary 2n+1 types so far as the "B" race is concerned and have both been secured in the offspring of "B" triploids.

The tertiary forms shown in table 1 have arisen spontaneously in our cultures. Since they appear to have been formed through segmental interchange they raise the question whether segmental interchange leading to homozygous prime types is not frequent in our material of *D. stramonium*. This seems not to be the case for several reasons. The spontaneous occurrence of these tertiaries is relatively rare. One, 2n + [13 · 18], arose twice from a 2n + [17 · 17] parent heterozygous for a particular gene which may have been responsible in some way for its appearance. Of the remaining two, the 2n + [4 · 6] occurred twice and the 2n + [2 · 9] occurred once. These three

tertiaries are not related to the tertiaries in the relatively few prime types in nature. Furthermore a test by BERGNER of 22 sublines of line 1 derived from continued selfing of the various 2n+1 types failed to disclose any which induced abnormal configurations in F_1 generations with a single line 1 plant. We can conclude that neither in our cultures nor in nature has segmental interchange leading to prime types been a frequent phenomenon in *D. stramonium*.

Fragmentations, translocations and segmental interchanges as well as gene mutations have been common in this species, however, following radiation treatment. Together with the 5 prime types from nature and the results of radiation we now have isolated in the homozygous condition a total of over 75 prime types. Twelve of these are shown in the accompanying table (table 2) which is part of a larger table that is published elsewhere (BERGNER, SATINA, and BLAKESLEE 1933). The ends of the modified chromosomes in about 40 prime types have been identified by attachments with tester chromosomes at meiosis.

In our collection we have at least one homozygous prime type tester for the ends of each of the 12 chromosomes except the [5 · 6] chromosome and for this chromosome we have a prime type in the heterozygous condition which we are attempting to render homozygous. Possibly among those with an 1 chromosome involved, which have not yet had their ends determined, we may already have isolated a homozygous prime type for this last chromosome. (Since the paper was read the prime type involving the [5 · 6] chromosome has been rendered homozygous.)

This collection of prime types is being found of value as a source of chromosomal testers for various purposes and as material for the synthesis of compensating types and pure-breeding morphological types or synthetic "species." It would take us too far to recount all the uses of prime types. It should be mentioned, however, that prime type testers have replaced 2n+1 forms in the identification of the ends of chromosomes from other prime types and from other species, by means of chromosome attachments.

Prime types contain tertiary chromosomes and therefore are a source of compensating types (BLAKESLEE 1931) which we are beginning to use in interspecific studies. "Nubbin" (BLAKESLEE 1927), which was secured from radiation treatment in 1921, may be used as an illustration of a compensating type. Its formula showing chromosome attachments may be written as follows:

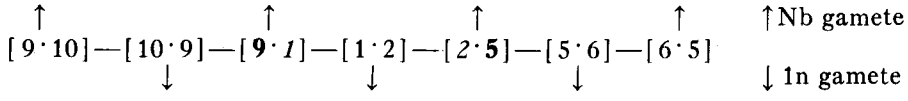

TABLE 2

Prime types (races homozygous for modified chromosomes).

DESIGNATION	CONFIGURATION IN F_1 WITH STANDARD LINE 1	PERCENT POLLEN ABORTION IN F_1	CHROMOSOMAL SIZE CLASSES	LINE 1 CHROMOSOMES INVOLVED	MODIFIED CHROMOSOMES	ORIGIN
PT 1	12 Bivalents	OK				Nature
PT 2	⊙4	OK	L= 1·2 m=17·18		1·18 2·17	Nature
PT 3	⊙4	±25	M=11·12 S=21·22		11·21 12·22	Nature
PT 4	⊙4	OK	l= 3·4 S=*21·22*		3·21 *4·22*	Nature
PT 5	Chain 4	OK	L= 1·2 M=11·12		. . 2·11·12	Radium treatment
PT 6	Chain 4	OK	L= 1·2 M=11·12		·1 2·11·12	Radium treatment
PT 7	∞4	±50	M= 9·10 m=19·20		$9 \cdot 10^{20}$ $19 \cdot 20^{10}$	Nature
PT 8	∞4	±50	M= 9·10 M=11·12		$9 \cdot 10^{12}$ $11 \cdot 12^{10}$	Radium treatment
PT 9	Chain 4	OK-25	m=19·20 s=23·24		20·19·23 ·24	X-ray treatment
PT 10	⊙4	OK	L= 1·2 M=13·14		1·13 2·14	X-ray treatment
PT 11	⊙6	±25	M=11·12 M=13·14 m=17·18		11·13 12·17 14·18	Radium treatment
PT 12	⊙4	OK	M=13·14 s=23·24		13·23 14·24	Radium treatment

There is only a single [1 · 2] chromosome but the italicized parts · *1* and · *2* "compensate" to form the equivalent of a [1 · 2] chromosome leaving the · 9 and · 5 portions shown in bold-faced type as excess material to affect the appearance of the plant. Each 1n gamete, indicated by the arrows pointing down in the formula, contains the single [1 · 2] chromosome. The gamete with the chromosomes [1 · 9] and [2 · 5], indicated by the arrows

pointing up in the formula, will lack the intact [1 · 2] chromosome but have its equivalent. The excess · 9 and · 5 material will prevent this latter gamete from going through the pollen. If now the intact [1 · 2] chromosome contains a particular gene, all the 1n female gametes, as well as all the effective pollen, will have this gene. A particular [1 · 2] chromosome, therefore, may be retained and a highly heterozygous race purified and rendered homozygous for the original [1 · 2] chromosome by continued male backcrossing of the heterozygous compensating type to a line 1 type that compensates for the same chromosome. Similarly, if a prime type such as a "B" race which involves the [1 · 2] chromosome is crossed onto our compensating type Nubbin, all the 1n gametes will be B's and the 2n offspring from selfing in consequence will all be B's as is shown by the following formula:

↑ ↑ ↑ ↑ ↑ ↑ Nb gamete
[9·10]—[10·9]—[**9**·*1*]—[1·18]—[18·17]—[17·2]—[*2*·**5**]—[5·6]—[6·5]
↓ ↓ ↓ ↓ ↓ 1n gamete

The method of continued backcrossing to appropriate line 1 compensating types has been used in retaining the interchanged chromosomes in cryptic types in nature while at the same time replacing the other chromosomes by those of line 1. The method is also being used in isolation of prime types and the retention of particular chromosomes from other species. The method outlined is fully successful only when crossing over does not occur. The compensating type, however, does not of itself prevent crossing over and hence a single chromosome passed through a series of compensating types will tend ultimately to resemble the homologous chromosome of line 1 so far as the genes which cross over are concerned. The method is more effective in the isolation and purification of prime types than of single genes since the end arrangement of chromosomes in the prime type is not changed, so far as is known, in passing through a compensating type. However, crossing over between the "B" race and the chromosomes of Nubbin is known to occur, and continued use of a compensating type should tend to render the genic content of the cryptic type similar to that of line 1 without, however, changing the order of the loci in the interchanged chromosomes. Compensating types have now been secured for 7 out of the 12 chromosomes.

INTERSPECIFIC DIFFERENCES

Datura is not a large genus. Its species are shown in table 3. The chromosomes of all the species listed have been studied by BELLING or by BERGNER with the exception of some of the species of tree Daturas. All examined have

12 pairs of chromosomes which show size classes somewhat similar to those in line 1 of *D. stramonium*. The evolution within the genus, therefore, has not been by changes in chromosome number. The chromosomes of the first 7 have been brought directly or indirectly into contact with those of line 1, *D. stramonium*, with results which show that there are only 24 different kinds of ends in these 7 species. Evolution apparently has been accompanied by changes in the arrangement of the ends rather than in the ends themselves as attachment organs. The nature and origin of these terminal organs of attachment are a major problem of chromosome evolution.

TABLE 3

Species of Datura under cultivation.

I. Stramonium group
  (1) *D. stramonium* (species used as standard)
  (2) *D. ferox*
  (3) *D. quercifolia*
II. Meteloides group
  (4) *D. leichardtii*
  (5) *D. meteloides*
  (6) *D. innoxia*
  (7) *D. pruinosa*
  (8) *D. discolor*
  (9) *D. metel* (several horticultural varieties)
III. (10) *D. ceratocaula*
IV. Brugmansia group (tree Daturas).

As a preliminary to an analysis of these chromosomal changes, there has been established for each species a tester race which has been used in the same way in which line 1 has been used as a standard for *D. stramonium* in terms of which its cryptic chromosomal races are interpreted. In no other species has the chromosomal analysis been carried so far as in *D. stramonium*. In all species in which more than two races have been tested, however, cryptic types have been discovered. They are now known to exist in *D. stramonium, D. quercifolia, D. leichardtii, D. meteloides, D. innoxia* and *D. metel*.

Considerable progress has been made in the chromosomal analysis of the 3 species in the *D. stramonium* group (*D. stramonium, D. ferox* and *D. quercifolia*). This species triangle is illustrated in the Datura exhibit and in a previous publication (BERGNER and BLAKESLEE 1932) and need not be discussed in detail here. The chromosomes of *D. stramonium* that have taken part in the chromosomal evolution of this group are given below the label. The numbered dominoes between the 3 species in figure 1 repre-

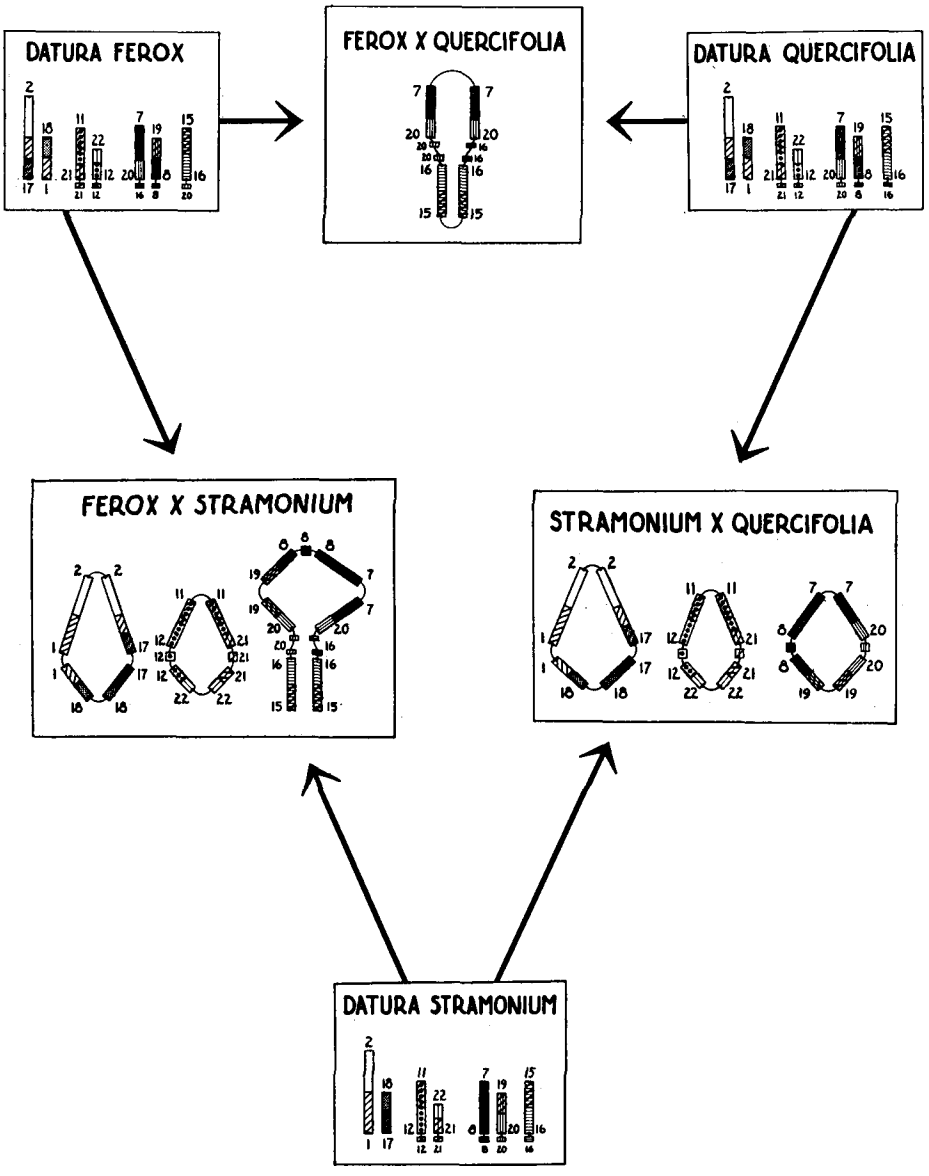

FIGURE 1.—Diagram to show chromosomal differences between the three species *Datura stramonium*, *D. ferox* and *D. quercifolia* shown at the corners of the triangle. The chromosomes which are not the same in all three species are represented by models. Those along the sides of the triangle represent the configurations which they form in hybrids between the species.

sent the chromosomes and the configurations which they form in hybrids between these species. What the chromosome ends are in these configurations has been determined by crossing to appropriate prime type testers and noting the attachments of the chromosomes at metaphase in reduction division. In the 2 hybrids with line 1, *D. stramonium,* we recognize the "B" circle. Both *D. ferox* and *D. quercifolia* are "B"s (or PT 2). They are both also prime type 3, the cryptic prime type from Peru. If races from Peru, which are at the same time "B" and the prime type 3, were used as the *D. stramonium* tester, there would be only 1 circle of 4 in F_1 with *D. quercifolia* and a single configuration of 6 with *D. ferox.* These remaining configurations would then represent interchanges which have not yet been found within *D. stramonium.* So far as investigated in this and also in other groups, species hybrids always show configurations due apparently to segmental interchange. There does not seem, however, to be a close connection between the degree of relationship of the species and the number of attached chromosomes in their hybrids. Thus *D. ferox* and *D. quercifolia* are obviously more closely related to each other than either to *D. stramonium.* It depends upon what tester race is used in *D. stramonium* whether *D. quercifolia* gives the same number of attached configurations with *D. ferox* as with *D. stramonium* or fewer.

The modified chromosomes of *D. ferox* and *D. quercifolia* have been freed from the other chromosomes by continued backcrossing to line 1, *D. stramonium.* The different prime types have been isolated or are being isolated by selfing and identifying the prime types by crossing to testers or by the use of compensating types. Thus through the use of the compensating type Nubbin we have isolated the B races of both *ferox* and of *quercifolia* and thus have now the B races of these 3 species to compare in otherwise line 1 standards. As a further test of these "B" chromosomes we shall isolate the interchanged B chromosomes in 2n + [1 · 18] and 2n + [2 · 17] types and compare the morphology of the plants which result.

It should be stated that in the process of purification by backcrossing to a standard, the end arrangements may be altered by crossing over. Thus from backcrossing *D. ferox,* two chromosomes ($[19 \cdot 20]^{16}$ and $[7 \cdot 20]^{20}$) are formed which are unknown in this species.

Other species have been studied by means of their chromosomal configurations in hybrids, but their analysis has not proceeded so far as with the species triangle just discussed. Figure 2 represents BERGNER'S preliminary findings in hybrids with 5 species. *D. leichardtii* acts as a bridging

CONFIGURATIONS IN SPECIES HYBRIDS

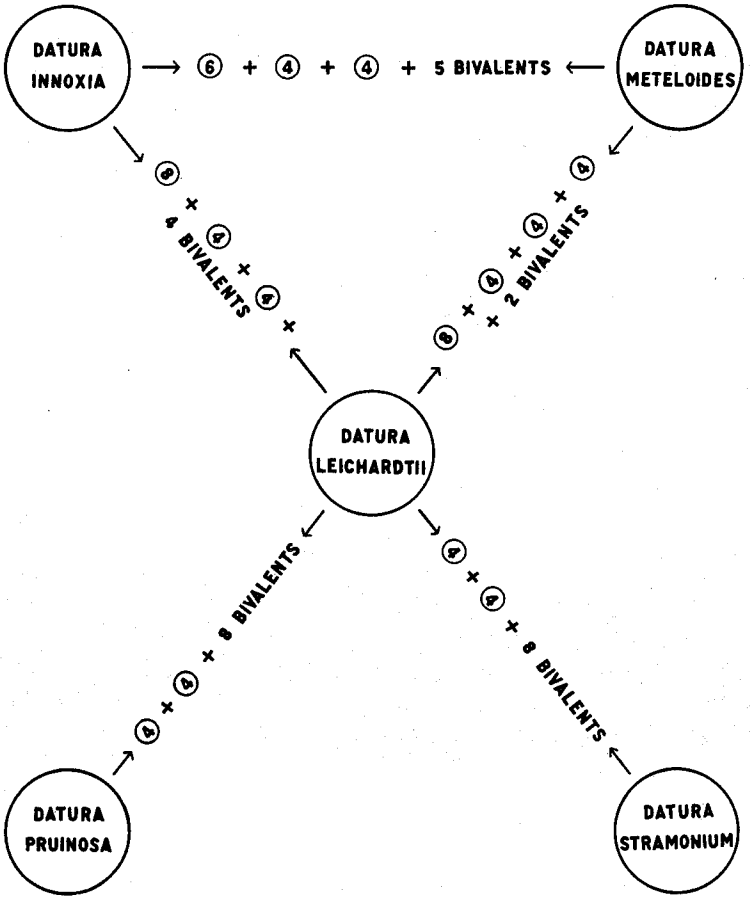

FIGURE 2.—Diagram of hybrids between 5 species of Datura. Chromosomal configurations in F_1 generations, consisting of circles of 4, 6, 8 and of pairs, are shown on lines which connect the species.

species between our standard species *D. stramonium* and the *meteloides* group with which *D. stramonium* will not cross directly. *D. leichardtii* differs from the line 1 *D. stramonium* tester by only two interchanges. If a "B" race had been used as a tester they would appear to differ by only a single interchange. Specifically, however, they are very dissimilar. We are in the process of getting *leichardtii* chromosomes into otherwise line 1 plants

in order to weigh the interchanged chromosomes as extras and to learn what we can about the genic content of the *leichardtii* chromosomes in terms of those of the *D. stramonium* standard. We are also getting line 1 testers into *leichardtii* to use in analyzing the chromosomes of the other species. The interchanged chromosomes of the other species we are freeing from their accompanying chromosomes by continued backcrossing to *D. leichardtii.* Difficulties are involved in crossability. BUCHHOLZ (BUCHHOLZ and BLAKESLEE 1927) has found that pollen-tube growth may be a factor in preventing certain hybrid combinations. Thus the pollen of *meteloides* grows

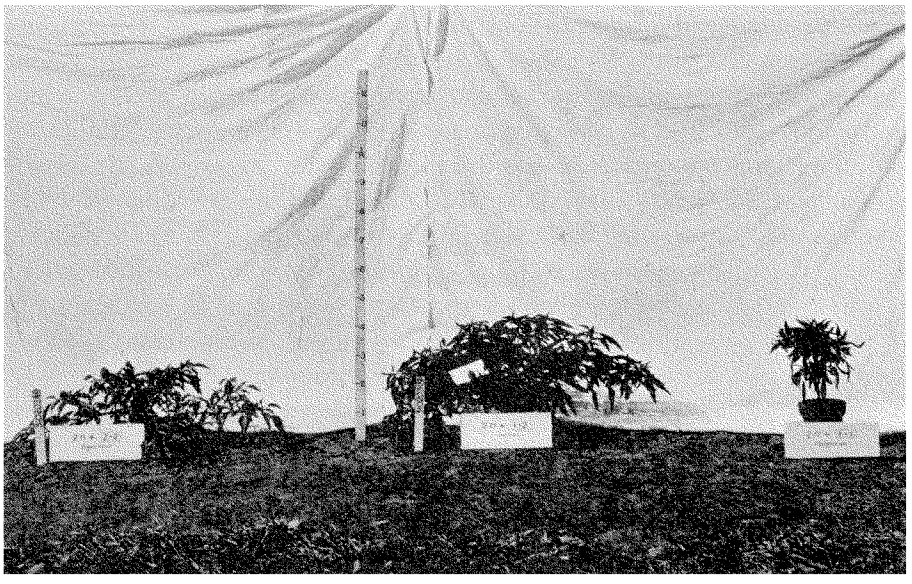

FIGURE 3.—In center, plant of the 2n + [1 · 2] primary "Rolled." At right its 2n + [1 · 1] secondary "Polycarpic" and at left its 2n + [2 · 2] secondary Sugarloaf."

well on *D. stramonium* whereas the pollen of *D. stramonium* bursts in the style of *D. meteloides.*

Our survey of species has shown that segmental interchange has accompanied the formation of biotypes within a single species and also the differentiation of the larger groups called species. Since it has been shown that prime types homozygous for interchanged chromosomes do not necessarily differ from normals, it is not clear what if any role segmental interchange has played in bringing about the differences which characterize distinct species. It has been somewhat difficult to conceive of species being evolved by the gradual accumulation of gene mutations. A method of adding whole

blocks of genes at a single stroke, such as would be afforded by simple translocations, would be more satisfying. It must be confessed, however, that our studies of Datura species have as yet given no evidence that such methods have been used in nature.

SYNTHESIZED NEW "SPECIES"

Artificial new "species" can be synthesized by the addition of extra chromosomal material as is shown in more detail in another publication (BLAKESLEE, BERGNER and AVERY 1933). The photograph (figure 3) rep-

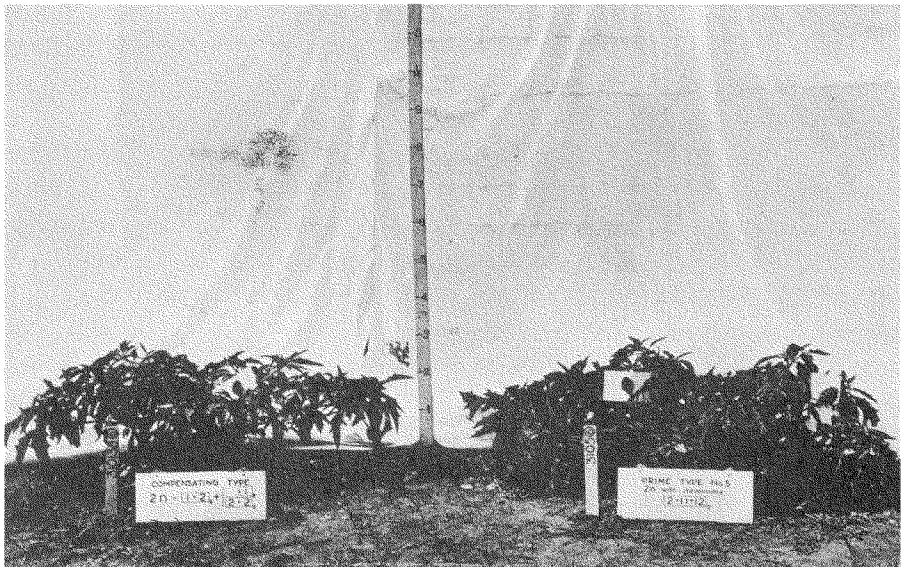

FIGURE 4.—Two synthesized new "species," morphologically similar but chromosomally different. Each has two extra doses of the · 2 half chromosome and breeds true. Capsules of these plants are shown in figure 5.

resents a 2n + [1 · 2] plant between its two secondaries, 2n + [1 · 1] and 2n + [2 · 2]. The latter type has 2 extra doses of the ·2 half chromosome. This extra [2 · 2] chromosome BUCHHOLZ (BUCHHOLZ and BLAKESLEE 1932) has shown can be transmitted through the pollen. In consequence, it has been possible to build up types homozygous for this excess ·2 material, as is shown by figures 4 and 5. So far we have four types that are morphologically similar but chromosomally different.

The first is the 2n + [2 · 2] secondary called "sugarloaf" which is shown in figures 3 and 5. It has 25 chromosomes and since the extra chromo-

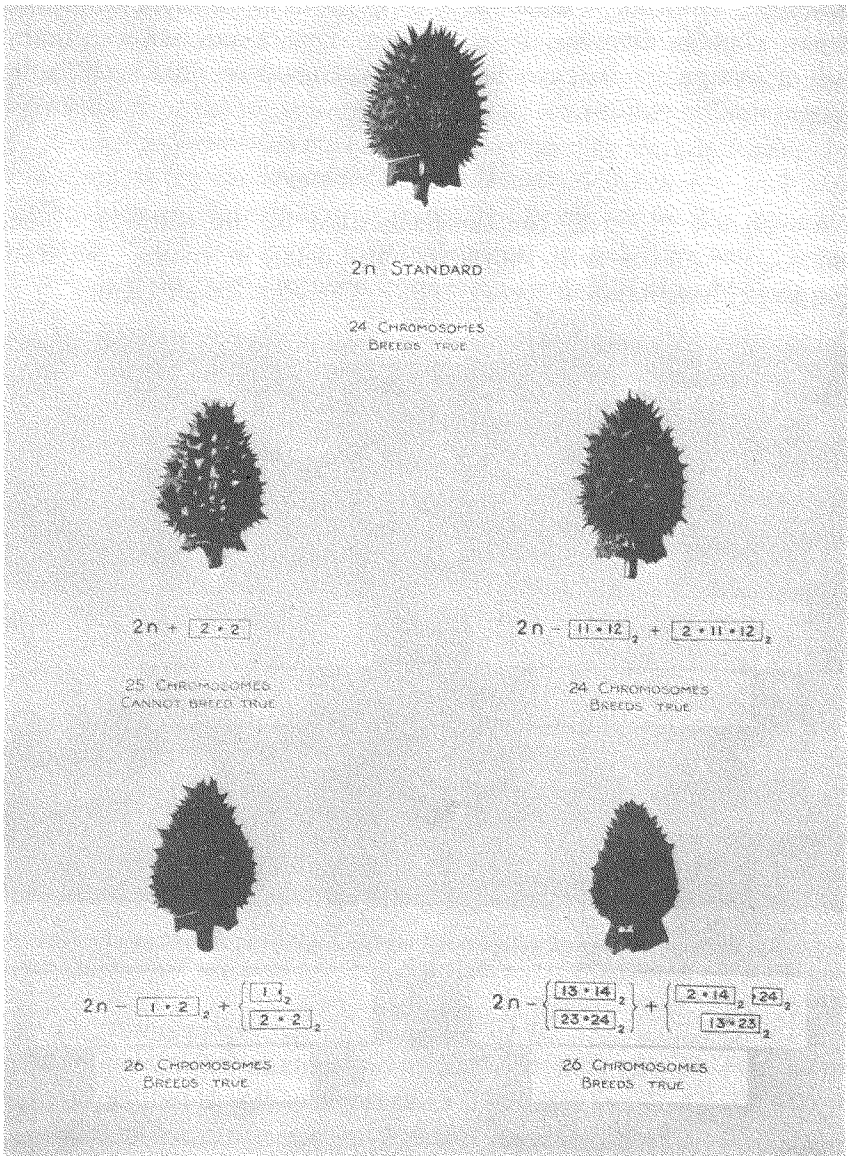

FIGURE 5.—Capsule of a normal 2n plant from line 1 in comparison with those of four types which are different in chromosomal constitution but alike in having two extra doses of the · 2 half chromosome. The 2n + [2 · 2] is the secondary "Sugarloaf" with 25 chromosomes and can not breed true. The other three have similar sugarloaf-shaped capsules and are true breeding.

some is carried by only part of the gametes, this type can not breed true.

The second type (figures 4 and 5) lacks the normal [11 · 12] chromosomes but is homozygous for a [11 · 12] chromosome to which is attached a ·2 fragment. It has two extra doses of the ·2 and breeds true.

The third type lacks any [1 · 2] chromosome. It is homozygous for the [2 · 2] chromosome and also for the ·1 fragment. The ·1 and the ·2 portions compensate to form the equivalent of the missing [1 · 2] chromosome leaving the other ·2 part in excess. Since the plant is homozygous it will accordingly have 2 extra doses of ·2 and breed true.

The fourth was synthesized by combining three prime types, PT 9, PT 10 and PT 12, which are listed in table 2. It has two extra doses of the ·2 half and breeds true. It was obtained too late this summer for a planting in the garden but capsules from offspring grown in the greenhouse, one of which is shown in figure 5, show the sugarloaf shape due to the extra ·2 material. The capsules of the four types with double doses of the ·2 half shown in figure 5 may appear slightly different, but as great differences in appearance could be secured in capsules taken from a single plant of any of the four types. At the time when the photograph was taken the choice of capsules was limited. The secondary 2n + [2 · 2] plant in figure 3 and the two types in figure 4 are indistinguishable in gross appearance. The last type shows certain characters found in the 2n + [23 · 24] primary that suggest that it contains excess material of ·23 or of ·24 in addition to that of the ·2 half.

It has been seen that we have produced two morphologically similar types that breed true, the one with 24 and the other with 26 chromosomes. If we had in our collection of prime types the proper translocations we should be able to secure a pure-breeding plant with 22 chromosomes. Thus if we had the ·1 half translocated to the [13 · 14] chromosome, for example, we should be able to obtain a plant with the following formula:

$$2n - \left\{ \begin{matrix} [1 \cdot 2]_2 \\ [11 \cdot 12]_2 \\ [13 \cdot 14]_2 \end{matrix} \right\} + \left\{ \begin{matrix} [1 \cdot 13 \cdot 14]_2 \\ [2 \cdot 11 \cdot 12]_2 \end{matrix} \right\}$$

Such a plant should be normal in appearance if no excess ·1 or ·2 material were present but would differ from normals if excess material were left over after the compensation.

The homozygous types that have been obtained differ from random gene mutations in that the characters that they show have been definitely planned for. They offer a new method of bringing about variations in economic

forms that are propagated by seed. In producing such pure-breeding races we may perhaps be said to have exercised a measure of control of speciation, if such synthetic forms are not eliminated from the category of species by definition, because their origin is known. Whether new species with the same or different chromosome numbers have been produced by such methods in nature is not known, but the ease of their formation artificially when the excess material can be transmitted through the pollen suggests that the same process may have been utilized in natural evolution. In this as in many other processes devised by man nature may be found to have had the priority.

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