

to these are the primary exylem and pith. The periderm forms beneath elongated cracks in the epidermis. After secondary growth commences, transient radially arranged dilation meristems first occur in a poorly developed state in the primary body, and later more highly developed ones occur through previously formed dilation tissues and the phloem rays. They produce wedges of new tissue

(dilation tissue) which cause the older rings of tissues to be fragmented, and in the trunk and large limbs the fragments of the primary body are so widely dispersed that they are only occasionally encountered in a given section.

DEPARTMENT OF PLANT PATHOLOGY,
UNIVERSITY OF CALIFORNIA CITRUS EXPERIMENT STATION,
RIVERSIDE, CALIFORNIA

LITERATURE CITED

- BARTHOLOMEW, E. T., AND H. S. REED. 1943. Chapter VI (general morphology, histology, and physiology) In: H. J. WEBBER AND L. D. BATCHELOR. The citrus industry. Univ. California Press. Berkeley and Los Angeles.
- DE BARY, A. 1877. Vergleichende Anatomie der Vegetationsorgane der Phanerogamen und Farne. Leipzig.
- HAYWARD, H. E., AND E. M. LONG. 1942. The anatomy of the seedling and roots of the Valencia orange. U. S. Dept. Agric. Bull. 786.
- HOLDHEIDE, W. 1951. Anatomie mitteleuropäischer Gehölzrinden. Handbuch der Mikroskopie in der Technik 5: 195-367.
- MÖELLER, J. 1882. Anatomie der Baumrinden. Berlin.
- SCHNEIDER, H. 1952. The phloem of the sweet orange tree trunk and the seasonal production of xylem and phloem. *Hilgardia* 21(12): 331-366.
- . 1954. Condition of phloem of sour orange tree trunk in winter. *Hilgardia* 22(16): 583-591.
- SCOTT, FLORA M., AND KATHERINE C. BAKER. 1947. Anatomy of Washington Navel orange rind in relation to water spot. *Bot. Gaz.* 108: 459-475.
- , MARY R. SCHROEDER, AND F. M. TURRELL. 1948. Development, cell shape, suberization of internal surface and abscission in the leaf of the Valencia orange, *Citrus sinensis*. *Bot. Gaz.* 109: 381-411.
- SCHROEDER, C. A. 1953. Spirality in *Citrus*. *Bot. Gaz.* 114: 350-352.
- WEBBER, IRMA E., AND H. S. FAWCETT. 1935. Comparative histology of healthy and psorosis-affected tissues of *Citrus sinensis*. *Hilgardia* 9(2): 71-109.

PHYLOGENETIC STUDIES IN OENOTHERA: FURTHER ANALYSIS OF PLANTS FROM THE SOUTHEASTERN UNITED STATES¹

Harry T. Stinson² and Erich Steiner³

AN EARLIER ANALYSIS (Steiner, 1952) of oenotheras from the southeastern United States indicated the presence in this region of three phylogenetic groupings. One of these, the *grandiflora* group, is found as a relict population and consists of structural homozygotes with large open-pollinated flowers. The remaining collections which have been analyzed are structural heterozygotes, some of which belong to the *biennis* group 1 of the midwestern United States, while others, although phenotypically narrow-leaved *biennis* types, carry beta complexes with the distinctive *truncans* arrangement of chromosome ends and alpha complexes with certain *grandiflora* characters. Extension of the original study was undertaken to determine which of the latter types predominate in the southeastern populations and to obtain additional evidence which might clarify the relationships among the three types occurring in this area. Since both structural

homozygotes with conventional cytogenetic behavior and specialized, balanced heterozygotes occur in the Southeast, it is felt that a more intensive analysis of populations of this region may reveal how structural heterozygotes have evolved from their homozygous ancestors. With these considerations in mind a cytogenetic study of recently acquired collections from Virginia and North Carolina was carried out.

MATERIALS AND METHODS.—The races included in the present analysis are shown in table 1. With the exception of the strain from Washington, North Carolina, all are true-breeding complex-heterozygotes showing circles of fourteen chromosomes at meiosis. The eight races under consideration may all be classified phenotypically as *biennis* types, although they are by no means identical in appearance; each race can be distinguished from any other with little difficulty. Even the two *Camp Peary* strains, which were collected in the same field, are distinctive.

The methods employed for the cytogenetic analysis of these races are those which have been commonly used in studies of *Oenothera* phylogeny (for details, see Cleland et al., 1950).

CHARACTERIZATION OF THE COMPLEXES.—*Genetic*.—The alpha complexes of these races all produce

¹ Received for publication May 16, 1955.

² Some of the data in this paper were taken from a thesis presented by the first named author to the Botany Department of Indiana University in partial fulfillment of the requirements for the degree of Doctor of Philosophy.

³ Work by the later author supported by the Faculty Research Fund of the Horace Rackham School of Graduate Studies, University of Michigan. This project was carried out in part during tenure of a Summer Faculty Fellowship.

TABLE 1. Races analyzed in the current study

Race	Source	Collector	Chromosome Configuration
Beaufort	Washington, N. C.	B. Mikula	⊙ 10, ⊙ 4
Camp Peary E	Williamsburg, Va.	J. T. Baldwin, Jr.	⊙ 14
Camp Peary L	Williamsburg, Va.	J. T. Baldwin, Jr.	⊙ 14
Newport News B	Newport News, Va.	H. Stinson	⊙ 14
Oxford	Oxford, N. C.	B. Mikula	⊙ 14
Roanoke B	Roanoke, Va.	E. Steiner	⊙ 14
Warwick A	Warwick, Va.	H. Stinson	⊙ 14
Williamston	Williamston, N. C.	B. Mikula	⊙ 14

similar phenotypic effects, namely those associated with the *biennis* phenotype (see further, Cleland et al., 1950). There are, however, in the alpha complexes of *Beaufort* and the *Camp Peary* strains leanings toward the *grandiflora* type, especially in the glabrous, clear-green, long and tapered bud cones, clasping bracts, and narrower leaves (as compared with typical *biennis* complexes). Thus, these complexes tend to be intermediate in phenotype between the alpha *biennis* group 1 and the alethal *grandiflora* complexes.

The beta complexes produce *strigosa* characters—thick, gray-green leaves, appressed pubescence, and an open habit with limited branching. The beta *Camp Peary* complexes depart somewhat from the typical *strigosa* features in yielding darker leaves and thinner bud cones. They are probably not sufficiently distinct, however, to consider them other than beta *biennis* 1 complexes.

Cytological.—The chromosomal end arrangements which have been determined in the present study are shown in table 2. With the exception of alpha *Beaufort*, all alpha complexes possess the arrangement found among approximately 80 per cent of the alpha *biennis* group 1 complexes so far studied. The alpha *Beaufort* arrangement differs from the typical alpha *biennis* 1 arrangement by two interchanges; it is of interest to note that the alpha *Beaufort* arrangement is identical with that

of *acuens*, a complex which was first encountered in a structurally heterozygous race collected by deVries, near Dixie Landing, Alabama, and showing the *grandiflora* phenotype (Cleland, 1935).

The beta complexes of three races, *Newport News B*, *Warwick A*, and *Roanoke B*, are also typical of the *biennis* 1 phylogenetic grouping, since the two arrangements occurring here are those found most commonly among the beta *biennis* group 1. In contrast, the beta complexes of *Camp Peary E* and *Camp Peary L* are far removed segmentally from either of the common beta *biennis* 1 arrangements. The *Camp Peary* betas do, nevertheless, show some affinity to certain beta *biennis* 1 complexes (see table 3). Thus, beta *Camp Peary L* is but two interchanges removed from beta *Richmond*, a *biennis* 1 race collected approximately sixty miles inland from the Camp Peary locale. Beta *Citronelle*, a race from southeastern Alabama, is three interchanges distant from beta *Camp Peary L*. Beta *Camp Peary L* is separated from every other beta *biennis* 1 complex by at least four interchanges.

Beta *Camp Peary E*, which is a minimum of six interchanges removed from beta *Camp Peary L*, appears to be intermediate between certain *biennis* 1 complexes and complexes with the *truncans* arrangement. It is three interchanges from the *biennis* 1 complexes, beta *Tuscaloosa A* and beta *Harrisonburg*, and three interchanges distant from

TABLE 2. Newly determined segmental arrangements

<i>alpha complexes</i>								
alpha Beaufort	1.4	3.2	5.6	9.8	7.10	11.12	13.14	
alpha Oxford	1.2	3.4	5.14	9.8	7.10	11.12	13.6	
alpha Williamston	1.2	3.4	5.14	9.8	7.10	11.12	13.6	
alpha Newport News B	1.2	3.4	5.14	9.8	7.10	11.12	13.6	
alpha Warwick A	1.2	3.4	5.14	9.8	7.10	11.12	13.6	
alpha Roanoke B	1.2	3.4	5.14	9.8	7.10	11.12	13.6	
alpha Camp Peary E	1.2	3.4	5.14	9.8	7.10	11.12	13.6	
alpha Camp Peary L	1.2	3.4	5.14	9.8	7.10	11.12	13.6	
<i>beta complexes</i>								
beta Warwick A	1.14	3.2	5.9	7.8	6.12	11.10	13.4	
beta Roanoke B	1.14	3.2	5.9	7.8	6.12	11.10	13.4	
beta Newport News B	1.4	3.9	5.2	7.8	6.12	11.10	13.14	
beta Camp Peary E	1.13	3.8	5.12	7.6	9.14	11.10	2.4	
beta Camp Peary L	1.10	3.9	5.13	7.11	8.12	2.6	4.14	
beta Williamston	1.7	3.2	5.4	6.10	9.12	11.14	13.8	

TABLE 3. *Previously determined complexes showing relationships segmentally to the newly determined arrangements*

acuens	1.4	3.2	5.6	7.10	9.8	11.12	13.14
beta Citronelle	1.4	3.13	5.9	7.2	11.6	8.12	10.14
beta Harrisonburg	1.3	4.8	5.7	6.12	9.14	11.10	13.2
beta Richmond	1.5	3.9	8.12	7.2	4.14	11.6	13.10
truncans	1.13	3.7	5.2	4.6	9.14	11.10	8.12
beta Tuscaloosa A	1.13	3.2	5.7	4.8	9.12	11.10	14.6
beta Tuscaloosa B	1.4	3.7	5.11	6.10	9.2	8.12	13.14
beta Iowa 1	1.11	3.2	5.7	6.10	9.14	4.12	13.8
beta Iowa 6	1.4	3.2	5.7	6.10	9.14	11.12	13.8
ancestral arrangement	1.2	3.4	5.6	7.10	9.8	11.12	13.14

truncans. The beta of *Camp Peary E* is similar in this respect to beta *Tuscaloosa B* which also occupies an intermediate position between the more typical beta *biennis* group 1 complexes and *truncans*.

Beta *Williamston* shows relationship segmentally to only one other complex found in the southeastern area. It is three interchanges removed from beta *Tuscaloosa A*. The only other beta *biennis* 1 complex showing as close a segmental relationship to beta *Williamston* is beta *Iowa 1*. Beta *Williamston* also shows affinities segmentally to the beta *strigosa* group; it is only two interchanges distant from beta *Iowa 6*, and three interchanges removed from five other beta *strigosa* complexes.

The Beaufort collection.—The unusual nature of the *Beaufort* race was first suspected when plants derived from the original seed collection showed $\odot 10$, $\odot 4$ at meiosis. Intermediate circles have been found only rarely in samples of natural populations. Cytological determinations were obtained for four of the twelve plants in the culture. Since the twelve plants were phenotypically identical, it was assumed that all plants possessed the same chromosome configuration. That this assumption was warranted is supported by the fact that in subsequent generations the same phenotype has always been associated with $\odot 10$, $\odot 4$ configuration.

Hybrid seed was readily obtained using *Beaufort* both as male and female parent in crosses with a number of our standard races. Controlled self-pollinations for the maintenance of the *Beaufort* collection were also made.

The hybrids of *Beaufort* with the standard races revealed that *Beaufort* possesses an alpha complex producing *grandiflora*-like characters and a beta complex yielding the *strigosa* phenotype. The alpha complex was transmitted to 220 of the 246 F_1 plants obtained from ten crosses using *Beaufort* as female parent. The remaining 26 plants, which occurred among five of the crosses, received the beta complex. When *Beaufort* was used as male parent, the beta complex was transmitted exclusively to the offspring in only six out of sixteen crosses. Of the 231 F_1 progeny from these sixteen crosses, 168 plants carried alpha *Beaufort*.

Beta *Beaufort* has as yet not been obtained in enough hybrid combinations to allow analysis of its segmental arrangement.

Beaufort does not breed true upon selfing. Although the original planting was uniform, the following generation obtained by selfing showed a striking segregation into two classes, which only became apparent shortly after the plants were placed in the field. In the seedling and rosette stages the two classes were phenotypically indistinguishable. One class proved to be the parental type showing $\odot 10$, $\odot 4$ at meiosis. Plants of the second class were only two-thirds as tall as the parental type and were much less vigorous. Early in the season the leaves of this segregate were severely attacked by a leaf-eating insect, while the parental type remained unmolested. The segregate showed the alpha *Beaufort* phenotype, possessing thin, dark-to clear-green, fairly narrow leaves and glabrous, tapered, twisted bud cones with clasping bracts. These plants were pollen sterile. Numerous anther smears were made, but even the best slides showed only a few pollen mother cells. In those cells which could be interpreted, five pairs of chromosomes were invariably present. The remaining four chromosomes were always observed in chains of two's. It was not possible to establish with certainty whether these represented $\odot 4$ in which chiasmata had failed to form, or whether they were bivalents. Therefore, the chromosome configuration of the segregate must have been either 7 pairs or $\odot 4$, 5 pairs. Nevertheless, the phenotype clearly indicated that the segregate was homozygous (if 7-paired) or nearly so (if $\odot 4$, 5 pairs) for the alpha *Beaufort* complex.

The *Beaufort* race has been carried through four generations with essentially the same behavior as described above. Table 4 summarizes the pertinent data of each generation.

In all cases, the parental type has shown $\odot 10$, $\odot 4$ at meiosis. In spite of the several hundred slides which have been prepared, it has been possible to establish with certainty the chromosome configuration of only five plants of the segregate type. These were all plants of the third generation. Four possessed 7 pairs of chromosomes, and one showed $\odot 4$, 5 pairs. The latter plant was phenotypically identical with the others, however. An-

TABLE 4. Segregation in the selfed line of *Beaufort*^a

Year	Parental type	alpha	
		<i>Beaufort</i> segregate	<i>strigosa</i> -type segregate
1951 ^b	12	0	0
1952	2	13	0
1953: 1st planting ^c	11	21	2
2nd planting	28	110	0
1954 ^d	27	29	0

^a Plants were grown in test plots at Ann Arbor, Michigan.

^b Plants grown from original seed collection.

^c As a result of unusually wet conditions in the field, 35 rosettes died before classes could be recognized.

^d 11 plants died before classes could be distinguished.

other four plants were interpreted as having 7 pairs, although not entirely without question.

One of the segregates of the second generation was hybridized as the female parent with a number of standard races. Seed was obtained from these crosses but in reduced quantities as compared with crosses made with the parental type. The cytological configurations of the hybrids were those which the alpha *Beaufort* arrangement gives with the standard complexes. There were no exceptions. Therefore, the specific *Beaufort* segregate used in these crosses, if 7-paired, must have been homozygous for alpha *Beaufort* or, if $\odot 4$ were present, must have transmitted only alpha *Beaufort* chromosomes to its offspring.

Two plants appearing in the third generation differed from the ordinary segregate in possessing gray-green, thick, stiff leaves and in remaining unbranched. Both plants died before producing buds which could be used for cytological preparations. It is possible that these represented $\odot 10$, 2 pairs segregates which theoretically should appear on the basis of the behavior outlined above.

DISCUSSION.—If the phenotypes as well as the segmental arrangements of their complexes are considered, at least five of the eight races here analyzed belong without question to the *biennis* group 1. These races show essentially the *biennis* 1 phenotype; their alpha complexes possess the typical alpha *biennis* 1 end arrangement and produce *biennis* characters. The beta complexes produce a *strigosa* phenotype in every case, and those segmental arrangements which have been determined are identical with or closely related to the betas of *biennis* group 1.

The three remaining races, *Beaufort* and the two *Camp Peary* forms, give evidence of phylogenetic connections with the other groups previously identified in the southeastern United States. Thus, the alpha complex of *Beaufort* is phenotypically and segmentally intermediate between the alethal *grandiflora* complexes and the alpha *biennis* group 1 complexes. The alpha complexes of the two *Camp Peary* strains show tendencies toward the *grandi-*

flora phenotype. The *Camp Peary* betas display some phenotypic effects suggestive of those produced by the *truncans* complex. Moreover, the segmental arrangement of beta *Camp Peary E*, like that of beta *Tuscaloosa B*, appears related to certain beta *biennis* complexes on the one hand, and to the *truncans* arrangement on the other hand. These affinities point to phylogenetic relationship between the *biennis* group 1 betas and complexes with the *truncans* arrangement, a connection already suggested by Steiner (1952).

Although they have not been worked out segmentally, the beta complexes of *Beaufort* and *Oxford* are certainly not closely related to *truncans*, since they give $\odot 14$ and $\odot 12$, 1 pair, respectively, when combined in hybrids with *truncans*. They, like the betas of *biennis* 1, also produce *strigosa* phenotypes. It thus seems likely that these beta complexes will turn out to possess one of the beta *biennis* group 1 arrangements.

With the analysis of these races from the Southeast, it seems increasingly apparent that the characteristic type found in these populations is identical with or very similar to the *biennis* group 1 forms. Of all the groups of oenotheras which have so far been analyzed, the *biennis* group 1 occupies one of the most extensive ranges. Its success, at least in part, must result from a pronounced and permanent heterosis, since its members carry two complexes which are genetically and segmentally very different.

One of the interesting features of the *biennis* group 1 is the widespread occurrence of the 1-2 3-4 5-14 7-10 9-8 11-12 13-6 arrangement which is characteristic of 34 of the 44 alpha *biennis* 1 complexes so far analyzed. In the southeastern area two arrangements closely related to the typical alpha *biennis* 1 arrangement are found among the *grandiflora* complexes. One of these is the arrangement considered to be ancestral for the subgenus, i.e., 1-2 3-4 5-6 7-10 9-8 11-12 13-14; the other is the *acuens* arrangement (see table 3) also occurring in alpha *Beaufort*. The close relationship phenotypically and segmentally of the *grandiflora* to the alpha *biennis* 1 complexes has led to the hypothesis that both these types have been derived from a common, structurally homozygous ancestral population, the members of which carried alethal genomes with the ancestral chromosome end arrangement (Steiner, 1952). The alpha *biennis* 1 arrangement is only one interchange removed from the ancestral arrangement and two interchanges from the *acuens* arrangement. Why is the alpha *biennis* 1 arrangement, and not the ancestral or the *acuens* arrangement so widely distributed throughout the alpha *biennis* 1 complexes? An answer may be found in table 5. In the first column are shown the configurations which are obtained when each of these three arrangements is combined with thirty-seven different beta *biennis* 1 com-

plexes. The striking fact is that the alpha *biennis* 1 arrangement gives $\odot 14$ with the beta *biennis* 1 arrangements in thirty-five of the combinations, whereas the ancestral arrangement gives $\odot 14$ in but seventeen, and the *acuens* arrangement in only five. Since it is generally agreed that $\odot 14$ in *Oenothera* possesses survival value, one may conclude that the typical alpha *biennis* 1 arrangement is widely distributed because of its versatility in producing $\odot 14$ with many different beta *biennis* 1 arrangements. Here, therefore, is evidence of the selective advantage of a specific chromosome end arrangement.

Table 5 also reveals some interesting facts pertinent to the problem of the origin of the beta *biennis* 1 complexes. Cleland (1954) has recently made a detailed study of the *strigosa* races of the western United States. He postulates that the beta *strigosas* covered the western half of the continent and spread eastward into the area occupied by the ancestral structural homozygotes which were the progenitors of the alpha *biennis* 1 complexes. Crossing occurred between members of these two populations and hybrids with \odot 's 14 were produced. These later developed balanced lethals and the self-pollinating habit. As mentioned above, the evidence indicates that the ancestral *biennis*-type structural homozygotes possessed the ancestral chromosome end arrangement, from which later arose the alpha *biennis* 1 arrangement. If either of these arrangements is combined with the alpha or beta *strigosa* arrangements which have been analyzed by Cleland, the configurations shown in the last two columns of table 5 are obtained. Not in a single case is $\odot 14$ formed. The number of small circles among these configurations is quite surprising, if one thinks of the beta *strigosas* as

closely related to present day beta *biennis* 1 complexes, most of which, as shown above (table 5, first column) do form $\odot 14$ with the typical alpha *biennis* 1 arrangement. Thus, if Cleland is correct in his belief that the *biennis* group 1 arose from hybridization between the beta *strigosas* and an ancestral *biennis*-type population (the only plausible hypothesis on the basis of the currently-known facts), then the beta *strigosa* complexes which were the specific progenitors of the beta *biennis* 1 complexes do not exist today, or at least are not represented in our collections.

The *Beaufort* data are of interest to the phylogenetic study of *Oenothera* in several respects. Unlike other complex-heterozygotes with intermediate circles such as *O. biennis* (*albicans-rubens* — $\odot 8$, $\odot 6$) (Cleland, 1923) and *Manistique* ($\odot 10$, $\odot 4$) (Geckler, 1950) which breed true and thus possess a lethal in each circle, *Beaufort* throws alpha-alpha segregates. Therefore, alpha *Beaufort* has not yet acquired a lethal or has lost its lethal. Loss of lethals has been described by Renner (1941) and by Oehlkers and Harte (1944). However, since alpha *Beaufort* is closely related to the aethal *grandiflora* complexes, it seems less likely that loss of a lethal is here involved. Alpha *Beaufort* behaves much like another *grandiflora* complex, namely *acuens*, which segregates in the homozygous condition from the *grandiflora* race of deVries (*acuens-truncans*). Although lacking a lethal, alpha *Beaufort* clearly possesses deleterious recessive genes which affect both vegetative vigor and reproductive capacity. These genes must have been acquired while the complex existed in a heterozygote (i.e., a circle-bearing race), since a plant homozygous for alpha *Beaufort* could hardly persist as a distinct entity in a natural population.

TABLE 5. Configurations obtained when the *acuens*, the ancestral, and the typical alpha *biennis* 1 end arrangements are combined with the known beta *biennis* 1, beta *strigosa* and alpha *strigosa* complexes

	beta <i>biennis</i> 1		beta <i>strigosa</i>		alpha <i>strigosa</i>	
	Configuration	Number	Configuration	Number	Configuration	Number
With <i>acuens</i>	$\odot 8$, $\odot 4$, 1 pair	10	$\odot 4$, $\odot 4$, 3 pairs	22	$\odot 10$, 2 pairs	1
	$\odot 10$, 2 pairs	14	$\odot 6$, $\odot 4$, 2 pairs	2	$\odot 8$, $\odot 4$, 1 pair	10
	$\odot 4$, $\odot 4$, $\odot 4$, 1 pair	1	$\odot 4$, $\odot 4$, $\odot 4$, 1 pair	1	$\odot 6$, $\odot 6$, 1 pair	8
	$\odot 8$, $\odot 6$	4				
	$\odot 12$, 1 pair	2	$\odot 8$, 3 pairs	2	$\odot 6$, $\odot 4$, $\odot 4$	3
	$\odot 14$	5	$\odot 4$, 5 pairs	1	$\odot 12$, 1 pair	2
	$\odot 6$, $\odot 6$, 1 pair	1			$\odot 10$, $\odot 4$	3
With ancestral arrangement	$\odot 8$, $\odot 6$	13	$\odot 4$, $\odot 4$, $\odot 4$, 1 pair	20	$\odot 8$, 3 pairs	1
	$\odot 14$	17	$\odot 6$, $\odot 4$, 2 pairs	3	$\odot 6$, $\odot 4$, 2 pairs	15
	$\odot 8$, $\odot 4$, 1 pair	1	$\odot 6$, $\odot 4$, $\odot 4$	2	$\odot 8$, $\odot 4$, 1 pair	3
	$\odot 6$, $\odot 4$, $\odot 4$	1	$\odot 8$, $\odot 4$, 1 pair	2	$\odot 4$, $\odot 4$, $\odot 4$, 1 pair	5
	$\odot 12$, 1 pair	5	$\odot 4$, $\odot 4$, 3 pairs	1	$\odot 6$, $\odot 4$, $\odot 4$	2
					$\odot 10$, 2 pairs	1
With typical alpha <i>biennis</i> 1 arrangement	$\odot 14$	35	$\odot 8$, $\odot 4$, 1 pair	11	$\odot 10$, 2 pairs	4
	$\odot 8$, $\odot 6$	1	$\odot 10$, 2 pairs	3	$\odot 6$, $\odot 6$, 1 pair	5
	$\odot 8$, $\odot 4$, 1 pair	1	$\odot 4$, $\odot 4$, 3 pairs	10	$\odot 8$, $\odot 4$, 1 pair	10
			$\odot 10$, $\odot 4$	2	$\odot 6$, $\odot 4$, $\odot 4$	2
			$\odot 4$, $\odot 4$, $\odot 4$, 1 pair	1	$\odot 12$, 1 pair	4
			$\odot 6$, $\odot 4$, 2 pairs	1	$\odot 10$, $\odot 4$	2

The alpha *Beaufort* complex may, in fact, represent one method whereby lethals arise in *Oenothera*, namely through an accumulation of detrimental genes which eventually reach a lethal potency. Renner (1933) has given evidence for such an interpretation of the lethal effect in at least certain races. Following this line of reasoning, it is possible to give, in outline, a series of events which might lead to the establishment of lethals.

First, hybrids produced as a result of crossing between two alethal, structurally homozygous populations of different genotypes would be expected to manifest hybrid vigor. If such populations differed also in segmental arrangements, circles would be present in the hybrids. The hybrids might be favored over either parental type because of their hybrid vigor. In addition, as was indicated earlier, the greatest survival value would accrue to those hybrid individuals with large circles, since the larger the circle, the greater the degree of permanence of the heterozygosity in subsequent generations. The circles would, therefore, tend to be maintained in the population even though, because of the absence of lethals, 7-paired segregates identical or similar to the parental populations would continually arise.

Detrimental recessive mutations occurring within the chromosomes of the circles would, before the origin of lethals, suffer one of two fates: (1) such mutations appearing in the 7-paired segregates would be eliminated by selection; (2) those mutations remaining in the circle, to the extent they were completely recessive, would not be subjected to elimination by natural selection and hence would persist. Thus, prior to the occurrence of mutation the 7-paired segregates would be at a selective disadvantage with respect to the circle-bearing individuals because of the increased vigor of the latter. As mutations occurred in the circles, the 7-paired segregates would be burdened further by the presence of these deleterious mutations, while the same mutations would survive within the circle. The continued build-up of detrimental mutations in the circle would lead to even greater reductions in survival value in the 7-paired segregates. That is, a lethal situation would be approached in plants homozygous for a given complex of 7 chromosomes. This lethal action might at first affect the mature plant, e.g., pollen production, but selection might tend to favor those mutations which eliminated the plant at a much earlier stage until eventually embryo development was impaired. When the latter condition obtained, zygotic lethals as they actually occur in *Oenothera* would have been developed.

Gametophytic lethals could arise in much the same way, i.e., by the accumulation of recessive deleterious mutations which rendered pollen or egg cells non-functional. These incipient gametophytic lethals, when arising within the circle, would also

impair appreciably the success of the 7-paired segregates. In the segregates there is near absolute homozygosity, which means that any genes that affect the functioning of either kind of gamete would be carried by both members of the chromosome pair, or pairs, concerned, and hence no combination of 7 chromosomes would exist which would allow normal functioning of one or the other kind of gamete.

On the view advanced here a balanced lethal condition is not necessary for the accumulation of detrimental mutations. On the contrary, the lethal situation may arise as a result of the build-up of mutations occurring in the chromosomes of the circle. Nor are balanced lethals essential for the survival of circle-bearing races. The latter become established, initially, through the increased selective advantage imparted by their heterosis. Circle formation itself, therefore, assumes the major role in the establishment of complex-heterozygotes. Lethals are thus a more or less inevitable consequence once circles arise. And obviously lethals arising in the manner indicated would have to be "balanced" in the sense that both complexes of a plant cannot acquire the same lethal.

It should be noted that the above sequence of events could also account for the origin of lethal systems dependent upon a single locus rather than many loci. In this case the circle would serve not so much to allow *accumulation* of detrimental mutations as to prevent elimination by natural selection of the locus involved.

The *Beaufort* case is instructive also in the light of a suggestion put forth that in the evolution of the *oenotheras*, lethal-bearing complex-heterozygotes have probably not given rise to alethal complex-homozygotes (Stinson, 1953). The argument advanced was based on the fact that although it may be possible for a complex to lose its lethal and thereby come to reside in a homozygous condition, one would hardly expect the homozygous individuals arising in this manner to survive in nature. It was reasoned on a priori grounds that a complex which had existed in a complex-heterozygote could have accumulated a number of deleterious genes because of the enforced heterozygosity, and that these genes, when allowed to express themselves fully, would cause a severe reduction in the survival capacity of the individual carrying them. The results obtained upon self-pollinating *Beaufort* offer convincing evidence in favor of this hypothesis, since the plants homozygous for either five or seven of the alpha chromosomes were vegetatively stunted and showed complete pollen sterility. When the same chromosomes are associated with the beta *Beaufort* complex, the latter must "cover" the detrimental genes and so prevent manifestation of their effects.

A number of questions about *Beaufort* remain unanswered. For example, the variable propor-

tion of segregates in successive generations is puzzling. Further studies are in progress with the hope of clarifying some of these problems.

SUMMARY

Eight new collections of *Oenothera* from the southeastern United States have been analyzed cytogenetically. Five of these races are structural heterozygotes which belong to the *biennis* group 1. This group, therefore, occurs not only throughout the Midwest, but extends into the Southeast. The three remaining races, although complex-heterozygotes, show relationships to the homozygous, alethal *grandiflora* forms known to occur in Alabama. One of the races, *Beaufort*, is of special interest because it possesses a chromosome configuration of intermediate circles, namely, $\odot 10$,

$\odot 4$, rarely found among samples of natural populations. A further distinction of *Beaufort* is its production in each generation of homozygous segregates which are reduced in vigor and are pollen sterile. This race is interpreted as having accumulated deleterious recessive genes, which are, however, not of lethal potency. It thus represents an intermediate form in the evolution of complex-heterozygotes. The manner in which complex-heterozygotes may have evolved from structurally homozygous populations is discussed.

DEPARTMENT OF GENETICS,
THE CONNECTICUT AGRICULTURAL EXPERIMENT STATION,
NEW HAVEN, CONNECTICUT

AND
DEPARTMENT OF BOTANY,
UNIVERSITY OF MICHIGAN,
ANN ARBOR, MICHIGAN

LITERATURE CITED

- CLELAND, R. E. 1923. Chromosome arrangements during meiosis in certain *Oenotheras*. *Amer. Nat.* 57: 562-566.
- . 1935. Chromosome configurations in *Oenothera* (*grandiflora* \times *lamarckiana*). *Amer. Nat.* 69: 466-468.
- . 1954. Evolution of the North American *euoenotheras*: the *strigosas*. *Proc. Amer. Phil. Soc.* 98: 189-203.
- , Ed. 1950. Studies in *Oenothera* cytogenetics and phylogeny. *Indiana Univ. Publ. Sci. Ser.* 16.
- GECKLER, L. B. H. 1950. The cytogenetics and phylogenetic relationships of certain races of *Euoenothera* from northeastern North America. *Indiana Univ. Publ. Sci. Ser.* 16: 160-217.
- OEHLKERS, F., AND C. HARTE. 1944. Über die Aufhebung des Gonen und Zygotenausfalls bei *Oenothera*. *Flora* 137: 106-124.
- RENNER, O. 1933. Zur Kenntnis der Letalfaktoren und des Koppelungswechsel der *Oenotheren*. *Flora* 27: 215-250.
- . 1941. Über die Entstehung homozygotischen Formen aus Komplexheterozygotischen *Oenotheren*. *Flora* 135: 201-238.
- STEINER, E. 1952. Phylogenetic studies in *Euoenothera*. *Evolution* 6: 69-80.
- STINSON, H. T. 1953. Cytogenetics and phylogeny of *Oenothera argillicola* MacKenz. *Genetics* 38: 389-406.