

SELF-INCOMPATIBILITY IN *CREPIS FOETIDA* (L.) SUBSP.
RHOEADIFOLIA (BIEB.) SCHINZ ET KELLER

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SELF-INCOMPATIBILITY, or self-sterility caused by this condition, may be defined as the physiological inability of an organism possessing both functional male and female gametes to be self-fertilized. It refers only to instances in which the plant's own pollen and pistil come in contact with each other in the normal course of events, and excludes those cases in which self-fertilization is prevented by some natural device.

In the Compositae, a member of which is to be dealt with in this paper, very little work on self-sterility has been done. Only the work by STOUT on *Cichorium intybus* (1916, 1917, 1918), the observation by BABCOCK and HALL (1924) of intrasterile classes in *Hemizonia congesta*, a preliminary study of self-sterility in *Crepis foetida* by BABCOCK and CAVE (1938), and a brief study on *Cosmos bipinnatus* by LITTLE, KANTOR and ROBINSON (1940) were available when the work reported here was undertaken. In view of EAST's opinion (1940) that it is not unlikely that fully one-quarter of the approximately 14,000 species of the Compositae exhibit self-sterility to some degree, such paucity of data is surprising. The preliminary investigations of BABCOCK and CAVE had indicated that in *C. foetida* the situation in regard to self-sterility probably did not correspond to any of the previously known schemes. The present research was brought to a close in 1943, but the publication of a report has been unfortunately delayed. Meanwhile, GERSTEL (1950) has reported a similar genetic basis for self-incompatibility in guayule (*Parthenium argentalum*).

MATERIALS AND METHODS

The strain of *C. foetida rhoeadifolia* used in this study was grown from seed collected by DR. EDGAR ANDERSON in Yugoslavia in 1934. It was selected for this study from a number of other strains of this species because of its consistent and almost complete self-sterility, its short growing season and its abundance of large flowers.

Because of the nature of the flower and the large number of crosses involved, emasculation was impracticable. Preliminary study indicated that selfed heads very rarely set any seed and never over five percent of their potential capacity. Hence cross-pollination was carried on by bagging the heads prior to opening and massaging them together gently once each day from the time the outer circle of florets opened until the inner circle had opened. Thus, in the event of unfavorable weather a floret had more than one exposure to pollen. Flowers were kept bagged until the corollas had fallen. The possibility that compatible pollen on the stigmas might stimulate the plant's own pollen to

function is pretty well eliminated both by the experience of other workers on self-incompatibility and by a test carried on with *Crepis*. Since it was known that incompatible pollen fails to germinate, small amounts of compatible pollen were placed on stigmas and counted after application, with and without large quantities of the plant's own pollen. Later counts of the numbers of germinated pollen grains showed that addition of large amounts of the plant's own pollen to the small number of compatible grains did not increase the amount of germinated pollen.

Since *Crepis* pollen does not germinate on an incompatible stigma, it is possible to determine cross-relations between plants by an examination of stigmas a short time after they have been cross-pollinated. The florets being arranged concentrically, a head was selected whose outer circle had not yet begun to open and certain florets were removed, leaving three or four well-separated groups of three florets each. The style branches were then tapped gently to cause them to open out and expose the receptive part of the stigma. Pollen of a different male was placed on the stigmas of each group of florets, a pin being stuck into the head as a marker and the forceps being dipped in seventy percent alcohol between each pollen change.

Since *Crepis* pollen germinates rapidly, the stigmas were removed in about fifteen minutes, dipped several times in alcohol and then mounted in cotton blue lactophenol. If the cross was compatible a number of pollen tubes would have penetrated the stigma, firmly anchoring the pollen grains to the stigmatic surface. Both short sections of the tubes and many pollen grains would be visible. If, however, the cross was incompatible the alcohol wash would remove all pollen from the stigma because there would be no pollen tubes to hold them on. Occasionally a grain or two would be wedged in between some papillate cells and in a few instances a very small number of germinated grains were present, indicating slow or weak pollen tube growth.

With those plants whose cross relations were determined by this cytological method all determinations were duplicated and often triplicated. The method was found to be quite reliable. The cross relations of 43 of the 122 plants tested were determined by the cytological method but, of each family so tested, representative plants of each incompatibility group were thoroughly checked by actually performing the necessary crosses.

It was desired to retain at least one member of each homogeneous incompatibility group for use as a testing plant or "tester" in order to determine the behavior of untested plants. Vegetative propagation was accomplished by excising basal shoots, dipping them in hormodin and placing them in a propagating bed maintained at 25.6°C. Good rooting was obtained within two or three weeks.

PROCEDURE

From the seeds of *C. foetida rhoeadifolia* which were sown in the spring of 1939 only two plants reached maturity. These were self-incompatible but cross-compatible. Twenty plants were grown from this cross in 1940; all these plants were selfed and all possible crosses between sister plants were made and

TABLE 1
Cross Relations of H837

		MALE																				
		I					II				III					IV			UNCLASSIFIED			
PLANT NOS.		1	5	8	12	15	2	3	16	20	6	7	10	13	19	11*	17	18	4	9	14	
I	1	S	S	S	S	S	S	S	S	S	F	F	F	F	F	S	S	S	S	F	S	
		S	S	S	S	S	S	S	S	S	F	F	F	F	F	S	S	S	S	F	S	
	5	S	S	S	S	S	S	S	S	S	F	F	F	F	F	S	S	S	f	F	S	
		S	S	S	S	S	S	S	S	S	F	F	F	F	F	S	S	S	f	F	S	
	8	S	S	S	S	S	S	S	S	S	F	F	F	F	F	S	S	S	f	F	S	
		S	S	S	S	S	S	f	S	S	F	F	F	F	F	S	S	S	S	F	S	
	12	S	S	S	S	S	S	S	S	S	f	f	F	F	f	S	S	S	f	F	S	
		S	S	S	S	S	S	S	S	S	f	f	F	f	f	S	S	S	S	f	S	
	15	S	S	S	S	S	S	S	S	S	F	f	F	F	F	S	S	S	F	F	S	
		S	S	S	S	S	S	S	S	S	F	f	F	F	F	S	S	S	S	F	—	
	II	2	S	f	F	f	F	S	S	S	S	F	F	F	F	F	S	F	F	f	F	S
			F	S	S	f	f	S	S	S	S	F	F	F	F	F	S	f	F	f	F	S
		3	F	F	F	F	F	S	S	S	f	F	F	F	F	F	F	F	F	f	F	S
			F	f	f	F	F	S	S	S	S	f	F	F	f	F	S	f	F	S	F	S
		16	F	F	f	F	F	S	f	S	S	F	F	F	F	F	f	F	F	S	F	—
		F	f	f	F	F	S	S	S	S	F	F	F	F	F	S	F	F	S	F	—	
20		F	F	F	F	F	S	S	S	S	F	F	F	F	F	S	F	F	S	f	—	
		f	f	S	F	F	S	S	S	S	F	f	F	F	f	f	F	F	S	F	—	
III		6	F	F	S	F	F	F	F	F	F	S	S	S	S	S	S	S	S	S	S	—
			F	f	S	F	F	F	F	F	F	S	S	S	S	S	S	S	S	S	S	—
		7	F	f	F	F	F	F	F	F	F	S	—	S	S	S	S	S	S	S	S	S
			F	S	f	F	F	F	F	F	F	S	—	S	S	S	S	S	S	S	S	S
		10	F	f	F	F	F	F	F	F	F	S	S	S	S	S	S	S	S	S	S	S
			F	f	F	F	F	F	F	F	F	S	f	S	S	S	S	S	S	S	S	S
		13	F	F	f	F	F	F	F	F	F	S	S	S	—	S	S	S	f	S	S	—
		F	F	S	F	F	F	F	F	F	S	S	S	—	S	S	S	S	S	S	—	
	19	F	f	F	F	F	F	F	F	F	S	S	S	S	S	S	S	f	S	f	—	
		F	f	F	F	F	F	F	F	F	S	S	f	S	S	S	S	S	S	S	—	
	IV	11	S	S	S	S	S	F	F	F	F	S	S	S	S	S	S	S	S	S	S	S
			S	S	S	S	S	F	F	F	F	S	S	S	S	S	S	S	S	S	S	S
		17	S	S	S	S	S	F	F	F	F	S	S	S	S	S	S	S	S	S	S	S
			S	S	S	S	S	F	F	F	F	S	S	S	S	S	S	S	S	S	S	—
		18	S	S	S	S	S	F	F	F	F	S	S	S	f	S	S	S	S	S	S	S
		S	S	S	S	S	F	F	F	F	S	S	S	S	S	S	S	S	S	S	S	
Unclassified	4	F	F	S	F	F	f	f	S	S	f	S	S	S	S	S	S	S	—	S	S	
		F	F	f	S	F	S	f	f	f	f	S	S	f	f	S	S	S	—	S	S	
	9	F	f	F	F	F	S	S	S	S	S	S	S	S	S	S	S	—	S	S	S	
		F	S	F	F	F	S	S	S	S	S	S	S	S	S	S	S	S	S	S	—	
	14	F	F	F	F	S	f	F	—	—	—	S	S	—	—	S	—	S	—	—	S	
	F	S	S	—	—	—	S	—	—	—	S	S	—	—	—	—	—	S	—	S		

S: 0-10 seeds; f: 11-35 seeds; F: 36 or more seeds; —: no cross made.

The two letters in each section represent duplicate crosses.

* More than 97 per cent bad pollen.

duplicated. When it was found that these plants fell into homogeneous groups with regard to their cross relations, at least one family was grown from every cross between compatible groups. The individuals in each family were then crossed with testers representing each incompatibility group and classified according to their reactions.

Incompatibility was not always absolute so that a few seeds were occa-

sionally obtained from crosses that were obviously incompatible. All such seeds were grown and the plants were tested as to their cross relations.

RESULTS AND DISCUSSION

Table 1 shows the interrelations of the 20 original plants grown from the cross of the two self-incompatible, cross-compatible parents. Table 2 is a condensed record of the results of testing all of the individuals grown from crosses between members of the 20 offspring of the original two plants. In condensing this table some of the plants tested in Groups II, V, VI and VII were omitted. Their behavior was essentially the same as that of the others. The tested plants are grouped according to their compatibilities rather than their derivation. Table 2 will be found at the end of the paper.

Table 1 shows that the 20 plants from the original cross could be divided into four intra-sterile groups of homogeneous behavior. In the 15 families grown from crosses between members of this first family these same four intrasterile groups were found and, in addition, three other distinct and homogeneous incompatibility groups. This amounts to a total of seven homogeneous incompatibility groups arising from various combinations of the self-incompatibility genes present in the original two plants. The interrelations of these seven groups is as follows:

		MALE						
		I	II	III	IV	V	VI	VII
Female	I	S	S	F	S	S	F	F
	II	F	S	F	F	F	F	F
	III	F	F	S	S	F	S	F
	IV	S	F	S	S	S	S	F
	V	S	F	F	S	S	F	F
	VI	F	S	S	S	F	S	F
	VII	F	F	F	F	F	F	S

S: sterile; F: fertile. Reciprocal differences are shown in bold face.

The families grown during the course of the work, their parentage and the incompatibility groups found in their progenies are listed below:

FAMILY	PARENTS	GROUPS FOUND
H837	from H821-1 × H821-2	I, II, III, IV
H893	from H837-15 (I) × H837-19 (III)	II, IV, V, VI
H880	from H837-16 (II) × H837-15 (I)	I, II, V
H892	from H837-16 (II) × H837-19 (III)	III, VI, VII
H891	from H837-16 (II) × H837-11 (IV)	I, V
H894	from H837-2 (II) × H837-18 (IV)	I
H895	from H837-2 (II) × H837-17 (IV)	V
H870	from H837-19 (III) × H837-15 (I)	II, IV, V, VI
H875	from H837-19 (III) × H837-16 (II)	II, III, VI, VII
H876	from H837-19 (III) × H837-17 (II)	II, III, VI, VII
H874	from H837-11 (IV) × H837-16 (II)	I, III, V, VI

The families grown from "incompatible" combinations were as follows:

FAMILY	PARENTS	GROUPS FOUND
H890	from H837-8 (I) × H837-15 (I)	I
H889	from H837-8 (I) × H837-3 (II)	II
H888	from H837-3 (II) × H837-16 (II)	II, VII
H886	from H837-16 (II) × H837-3 (II)	II, VII
H885	from H837-10 (III) × H837-7 (III)	III, VII

Since the "oppositional factor" scheme of inheritance for self- and cross-incompatibilities is widely applicable in the plant kingdom it may be well to compare it with the behavior found in *Crepis*. In this scheme, as it operates in *Nicotiana* (EAST and MANGELSDORF 1926) and a great many other plants, there is a single series of alleles conditioning self- and cross-incompatibilities. The presence of any one of these alleles in the style of a plant inhibits the functioning of all pollen possessing the same allele. This causes all plants to be self-incompatible and gives reciprocal differences only when homozygotes are obtained by some device to break down the incompatibility barriers such as bud-pollination. If two plants are reciprocally compatible only two situations are possible among their progeny: either four inter-fertile, intra-sterile groups are obtained from the cross, all four of which are compatible with both parents; or (if the two parents have one incompatibility allele in common) only two inter-fertile, intra-sterile groups will be obtained, one of which will be fertile with both parents, the other fertile with only the female. All pollen behavior is gametophytic in that its behavior is determined by the incompatibility allele it is carrying.

Comparing the typical reaction between offspring and parents in *Nicotiana* with that found in two typical *Crepis* families:

Nicotiana—four intra-sterile, inter-fertile groups, all compatible with both male and female parents

Crepis —four intra-sterile groups, but compatibility with parents is as follows:

H870	H875
Group VI— incompatible with female compatible with male	Group III — incompatible with female compatible with male
Group V — compatible with female incompatible with male	Group II — compatible with female incompatible with male
Group IV— incompatible with female incompatible with male	Group VII— compatible with female compatible with male
Group II — compatible with female reciprocally different with male	Group VI — incompatible with female reciprocally different with male

The difference in the behavior of these two progenies from each other, as well as from *Nicotiana*, stresses the fact that in *Crepis* all families do not react

in the same way toward their parents. It is also apparent from the cross relations between the seven groups that, unlike *Nicotiana* where any group is compatible with all other groups and incompatible only within the group, in *Crepis* a group may be incompatible with one or several other groups besides its own.

The small number of groups obtained, even though the attempt was made to combine the incompatibility genes of the two original parents in as many ways as possible, suggests that incompatibility in *Crepis* is conditioned by a single series of alleles. Since members of different groups can be cross-incompatible, it seems probable that possessing only one allele in common is sufficient to cause incompatibility. But they behave differently because of the allele in which they differ, even to the extent of overcoming the incompatibility that might have been caused by the allele they have in common. Furthermore, if all of the pollen of a plant is inhibited on the stigmas of another plant which possesses only one allele in common, then that half of the pollen not carrying said allele must be inhibited. This is sporophytic determination; that is, the pollen behaves according to the genotype of the plant from which it came rather than according to its own genotype.

Therefore, the theoretical cause of reciprocal differences in *Nicotiana* and other plants, namely gametophytic determination of pollen behavior, will not explain reciprocal differences in *Crepis*. Another reason for its rejection is the doubt as to whether homozygotes are involved in the reciprocal differences in *Crepis*. The group which functions with another group as a female but not as a male should, according to the oppositional theory, be homozygous. However, group II, which might be considered as homozygous, is present in family H837 in the frequency of four out of a total of twenty plants, or twenty percent. It is also present in other families at a high frequency. Homozygosity, however, represents a breakdown or failure in the incompatibility mechanism and is expected to occur quite rarely except where special techniques are used to eliminate the barrier such as bud-pollination. Furthermore, if group II plants are homozygotes only group II plants would appear in their offspring, assuming that it was possible to get any offspring from crossing such plants. Progenies H886 and H888, however, originated in this way and each possesses group VII as well as group II plants. It appears, therefore, that group II is not homozygous and that homozygotes are probably not essential for the occurrence of reciprocal differences in *Crepis*.

It has been assumed that two incompatibility groups would be cross-incompatible if they possessed one allele in common. If the situation were so simple, then any group would be incompatible with both parents and with all but one of its three sister groups. Such regularity as this, or as that which occurs in *Nicotiana*, is not found in *Crepis*. Each group is not incompatible with the same number of other groups. Each homogeneous group or genotype appears to have its own particular strength or potency, which, since it is the same irrespective of the source of the particular plant, appears to reflect the different strengths of the individual alleles making up that genotype.

An explanation of the incompatibility behavior of the *Crepis* family de-

scribed above must include the following provisions: (1) The mechanism by which two plants of different genotypes can be cross-incompatible; (2) the means by which reciprocal differences occur; (3) the cause of the different strengths or potencies of the different genotypes. To meet these requirements the following hypothesis is proposed.

HYPOTHESIS

1. The reaction of the pollen is determined by the genotype of the plant from which it came rather than by its own genotype.

2. Four alleles of a single allelic series are responsible for the self- and cross-incompatibilities which have been found. They will be designated S_1 , S_2 , S_3 and S_4 .

3. S_1 is recessive to the other three alleles. It is able to inhibit fertilization only when it is homozygous in both the male and the female involved. Therefore, any cross between two plants carrying it, if compatible, will produce some homozygotes among the offspring.

4. S_2 is dominant over S_1 but recessive to S_3 and S_4 . Therefore, when present in a plant in combination with either of the latter two alleles the plant's reaction is determined by the dominant allele present. When it is present in combination with S_1 the plant's reaction is determined entirely by the S_2 allele.

The dominance relations or difference in potencies of the several alleles accounts for the reciprocal differences. For example, if two plants are both heterozygous for S_2 but in one it is associated with an allele to which it is recessive and in the other it is associated with S_1 the following result will be obtained: $S_1S_2 \text{♀} \times S_2S_3 \text{♂}$ —compatible; $S_2S_3 \text{♀} \times S_1S_2 \text{♂}$ —incompatible. This is because S_3 dominates in determining the pollen reaction in the first instance but S_2 dominates in the reciprocal cross.

5. S_3 and S_4 are "strong" alleles, both dominant over S_1 and S_2 but neither dominant over the other. Hence the presence of either or both in a plant causes its pollen to fail on plants containing like alleles.

GENOTYPES

In terms of the above hypothesis the cross relations between the seven groups and their hypothetical genotypes appear as follows:

		MALE						
		I (S_2S_4)	II (S_1S_2)	III (S_1S_3)	IV (S_3S_4)	V (S_1S_4)	VI (S_2S_3)	VII (S_1S_1)
	I(S_2S_4)	S	S	F	S	S	F	F
F	II(S_1S_2)	F	S	F	F	F	F	F
e	III(S_1S_3)	F	F	S	S	F	S	F
m	IV(S_3S_4)	S	F	S	S	S	S	F
a	V(S_1S_4)	S	F	F	S	S	F	F
l	VI(S_2S_3)	F	S	S	S	F	S	F
e	VII(S_1S_1)	F	F	F	F	F	F	S

Reciprocal differences are shown in bold face.

For further clarification of the segregation which occurred in the tested families, the following arrangement shows the probable genotypes of parent and offspring groups together with the number of plants of each genotype found in the progeny:

	P				F ₁						
H837	V	×	VI	4	II	5	III	5	I	3	IV
	(S ₁ S ₄)		(S ₂ S ₃)		(S ₁ S ₂)		(S ₁ S ₃)		(S ₂ S ₄)		(S ₃ S ₄)
H893	I	×	III	2	II	3	VI	4	V	2	IV
	(S ₂ S ₄)		(S ₁ S ₃)		(S ₁ S ₂)		(S ₂ S ₃)		(S ₁ S ₄)		(S ₂ S ₄)
H880	II	×	I	4	II	2	V	?		2	I
	(S ₁ S ₂)		(S ₂ S ₄)		(S ₁ S ₂)		(S ₁ S ₄)		(S ₂ S ₂)		(S ₂ S ₄)
H892	II	×	III	2	VII	1	III	0	II	5	VI
	(S ₁ S ₂)		(S ₁ S ₃)		(S ₁ S ₁)		(S ₁ S ₃)		(S ₁ S ₂)		(S ₂ S ₃)
H891	II	×	IV	0	III	1	V	0	VI	4	I
H894	(S ₁ S ₂)		(S ₃ S ₄)		(S ₁ S ₃)		(S ₁ S ₄)		(S ₂ S ₃)		(S ₂ S ₄)
H895											
H870	III	×	I	9	II	5	V	4	VI	10	IV
	(S ₁ S ₃)		(S ₂ S ₄)		(S ₁ S ₂)		(S ₁ S ₄)		(S ₂ S ₃)		(S ₃ S ₄)
H875	III	×	II	10	VII	7	II	2	III	5	VI
H876	(S ₁ S ₃)		(S ₁ S ₂)		(S ₁ S ₁)		(S ₁ S ₂)		(S ₁ S ₃)		(S ₂ S ₃)
H874	IV	×	II	2	III	2	VI	3	V	2	I
	(S ₃ S ₄)		(S ₁ S ₂)		(S ₁ S ₃)		(S ₂ S ₃)		(S ₁ S ₄)		(S ₂ S ₄)

Those from "incompatible" combinations were:

H890	I	×	I	0		1	I	1	V?		
	(S ₂ S ₄)		(S ₂ S ₄)		(S ₂ S ₂)		(S ₂ S ₄)		(S ₄ S ₄)		
H889	I	×	II	1	II	0		0	V	0	I
	(S ₂ S ₄)		(S ₁ S ₂)		(S ₁ S ₂)		(S ₂ S ₂)		(S ₁ S ₄)		(S ₂ S ₄)
H886	II	×	II	3	VII	7	II	?			
H888	(S ₁ S ₂)		(S ₁ S ₂)		(S ₁ S ₁)		(S ₁ S ₂)		(S ₂ S ₂)		
H885	III	×	III	2	VII	2	III	?			
	(S ₁ S ₃)		(S ₁ S ₃)		(S ₁ S ₁)		(S ₁ S ₃)		(S ₃ S ₃)		

It should be noted that the progeny of H891 also included one group II and one group VII plant. Since the male parent (H837-11) of this family possessed over 97 percent bad pollen both plants may have arisen through self-fertilization.

It is apparent that in those instances when S₂S₂, S₃S₃, or S₄S₄ plants were obtained they could not be distinguished by their compatibilities from S₁S₂, S₁S₃ or S₁S₄, respectively.

The hypothesis proposed and elaborated above includes the three essential provisions called for (p. 576) as shown below:

1. Two plants belonging to different groups can be cross-incompatible because they have an incompatibility allele in common. Since determination of pollen behavior is sporophytic, all of the pollen of a plant is inhibited on another's style if there is one allele in common.

2. Reciprocal differences are caused not by gametophytic determination and homozygosis, but by unequal potency of the alleles or dominance in the

sporophytic determination of pollen behavior. The dominance relations do not appear to hold in the stilar tissue or stigma, since the presence in such tissue of S_2 together with S_4 is capable of inhibiting the germination of S_2 pollen from an S_1S_2 plant (see H889, p. 577).

3. Intra-incompatible groups are incompatible with various other groups because of the difference in potencies of the alleles present in the incompatibility genotype. Six heterozygotes are possible when dealing with one series of four alleles and when all plants originate from the same two original parents directly or indirectly.

Two of these heterozygotes, groups III and V, which possess a strong allele plus the weak one, will each be incompatible with the other two groups possessing the same strong allele.

The two groups, I and VI, which possess a strong allele plus S_2 , will likewise be incompatible with the two groups possessing the same strong allele but reciprocally different with group II.

Group IV, which has both strong alleles, will be incompatible with all four groups which have either one.

Group II, the remaining heterozygote, will be fertile as a female with all other heterozygous groups because only its own pollen is dominated by the S_2 allele. For the same reason it will be incompatible as a male on all other groups possessing the S_2 allele.

Group VII will be fertile with all other groups because it is homozygous for S_1 , the weakest of the four alleles, a double dose being needed in both male and female to cause inhibition.

Group VII is the only homozygote found among the plants tested, but homozygous S_2 plants should be possible, since plants containing S_2 and differing in their strong allele are compatible. Homozygous S_2 plants are not distinguishable from group II in their behavior and so would have to be identified by their offspring.

SUPPORTING EVIDENCE

The assumptions made to explain the data are in line with ideas advanced from time to time by other workers in this field. RILEY (1932, 1934, 1936) working on *Capsella grandiflora*, found it necessary to assume sporophytic determination of pollen behavior in order to explain the incompatibility phenomena in that species. LEWIS (1947) has shown that some incompatibility alleles in *Oenothera organensis* exert dominance over others. Self- and cross-incompatibilities in this species are conditioned by a single set of alleles according to the oppositional-factor scheme. Since determination of pollen behavior is gametophytic, dominance is possible only after tetraploidy has been induced and two alleles are thus present in the pollen grains. LEWIS showed that S_4S_6 pollen functioned on styles containing S_4 , but not on styles containing S_6 . Since as much seed was obtained when this pollen was used on styles containing S_4 as when used on styles containing neither allele, dominance was believed to be complete. LEWIS also found other instances in which one allele is domi-

nant over another in the pollen grain. He does not, however, find dominance being exerted in the style and points out that this would not be expected, since dominance in the style would cause the whole incompatibility mechanism to break down. If dominance occurs in a plant whose pollen behavior is gametophytically determined, even though it can manifest itself only after the chromosome number is doubled, it seems legitimate to assume that it can also occur in *Crepis*, whose pollen behavior is sporophytically determined. Likewise, it is in line with LEWIS' data on *Oenothera* to assume that dominance occurs in the pollen but not in the style.

The concept that some *S* alleles are weaker in potency than others is also in agreement with observations on other plants. For example, EAST (1929), in commenting on STOUR's failure to get regular behavior in *Cichorium intybus*, suggested that STOUR was working with alleles which were on the borderline between self-compatibility and self-incompatibility and could often be tipped in one direction or the other by environmental influences within or without the plant. Probably S_1 and S_2 of the present study come more or less within the category of borderline factors. In an evolutionary sense they might be thought of as intermediate steps between the primitive condition of self-compatibility and the advanced one of self-incompatibility.

ANALYSIS OF CERTAIN FAILURES OF THE DATA TO CONFORM TO THE HYPOTHESIS

Table 1, representing the interrelationships of 20 sister plants, conforms fairly well to the expected behavior except for three plants. Plants 4 and 14 were apparently somewhat male sterile (a phenomenon frequently encountered in the subsequent generation) and plant 9 deviated from group III behavior by being incompatible with group II. It is possible that all three of these plants could be placed in group III but, since there is some doubt about them, they are classified separately.

Table 2 is a compilation on the basis of behavior rather than family lines. In assembling the data on which this table is based certain deviations from expectation were noted and some accounting for these is in order. In the results of 2037 pollinations it was found that 356 deviate from the expected behavior. These deviations may be classified as follows:

1. FS—expected to be compatible but in one or more tests incompatible.
2. Ff—expected to be compatible but in one or more tests only feebly so.
3. SF—expected to be incompatible but in one or more tests compatible.
4. Sf—expected to be incompatible but in one or more tests feebly compatible.

It is apparent that a certain number of the first two types of deviation from expected may be due to fluctuations in the environment. Although the study of self- and cross-incompatibilities should properly be carried on under reasonably constant environmental conditions this ideal is rarely attainable in practice. In the present study it was customary to carry on pollinations nearly every day and at any season of the year. Although all of the work was

done in the greenhouse it was not possible to control light and humidity at all and the temperature varied within a fairly wide range. Another factor of importance was the physiological condition of the plant. On many occasions one plant needed for a cross was just beginning to bloom while the other was approaching senility. It is therefore to be expected that a combination which should have been fully compatible might occasionally fail completely or show only weak fertility due to the external or internal environment.

As noted in table 2, there were several plants which failed to function as males and whose pollen was later found to be bad. H893-5 and -9 apparently belong in this group, although their pollen was not examined. Plant H837-11, however, had over 97 percent bad pollen and yet it behaved erratically in the 36 crosses attempted with plants of groups II and VII with which it would be expected to be fertile. In two of these crosses enough seeds were produced to call for classification as F; but most of the remainder yielded from 0 to nine seeds, there being only eight that produced from 12 to 34 seeds and so would be classified as f. This plant was completely sterile when used as a male with plants of all the other groups. Since it shows partial fertility with groups II and VII it was placed in group IV. The two instances of high fertility in spite of the low percentage of good pollen grains remain unexplained.

FS and Ff. Although 260 of the 356 deviations from expected were FS or Ff, only 210 of these represented different plant combinations. 102 of these behaved according to expected when duplicated or, in a few cases, reduplicated. These can therefore largely be attributed to unfavorable internal or external environmental conditions at the time of the first pollination. Of the remaining 108 cases 81 were not duplicated. Many of these likewise were caused, no doubt, by environmental conditions.

The 27 cases of FS and Ff which persisted after duplications are probably genetic rather than environmental. Of the 54 parents involved, group VII contributed 12, group II contributed 11 and group V, 11. The other groups contributed five parents each. Not only were these deviations from expected more closely associated with certain groups, but also with certain individual plants. Plant H870-21 was a parent in ten of these combinations, H875-21 in five, and H876-17 in three others. In other words, a genetic difference in three plants could account for 18 of the 27 deviations. Of the 26 individual plants involved in these 27 deviations, 13 were involved more than once.

SF and Sf. SF reactions are the most serious deviations from expectation, since it is hardly likely that environment would alter an incompatible reaction to that extent. In the 27 cases of SF only 17 different crosses are involved. Ten of these are between group VII plants, and seven involve combinations between groups III, IV, and VI. No reason is apparent why groups I, II, and V are not represented in this type of deviation from expectation. However, the aberrant behavior of the ten combinations between group VII plants is in keeping with our concept of the S_1 allele for which group VII is homozygous. It is considered to be a weak allele which acts as a barrier to fertility only when homozygous in both male and female. If it is a borderline allele, then it is probable that minor genes in the plant's genotype might be able to alter the

incompatibility response. If two plants alike in being homozygous for S_1 were sufficiently unlike in other components of their genotypes, this might move the threshold below the level needed for cross-incompatibility.

Of the 69 cases of Sf, involving 58 different plant combinations, 32 reacted according to theory when duplicated; 15 were not duplicated. Eleven persisted in the f reaction when the cross was repeated.

The deviations of the FS and Ff type are believed to have been caused in large part by environmental fluctuations, internal as well as external to the plants. The SF responses and part of the Sf are probably genetic in nature and exhibit the influence of minor genes which alter the incompatibility reaction, particularly when the S allele in the principal allelic series happens to be one of low potency.

THE CREPIS-PARTHENIUM SYSTEM OF SELF-STERILITY COMPARED WITH OTHER KNOWN SYSTEMS

Is the genetical system controlling self-sterility which has been discovered in *Crepis* and *Parthenium* unique? LEWIS (1944) made a helpful review of the known systems for self-sterility in plants. His treatment may be summarized as follows:

I. Heteromorphic (heterostyled) plants: *Primula sinensis* and many other investigated species.

1. One or two genes with two alleles.
2. Co-ordinated gene action in pistil and pollen.
3. Diploid pollen control.
4. Dominance.

II. Homomorphic (non-heterostyled) plants: *Nicotiana Sanderae* and all other investigated species, except *Capsella grandiflora*, *Crepis foetida* and *Parthenium argentatum*.

1. One gene with multiple alleles.
2. Independent gene action in the pistil but not in the pollen.
3. Haploid pollen control.
4. No dominance.

LEWIS points out that *Capsella grandiflora*, a non-heterostyled species, possesses all the genetical characteristics of the heterostyled system. Thus, a striking exception to the general rule has already come to light (RILEY 1936). Now, from investigations on two species, representing widely different tribes of the Compositae, the evidence calls for the following system:

III. *Crepis* and *Parthenium*.

1. One gene with multiple alleles.
2. Independent gene action in the style.
3. Diploid pollen control.
4. Dominance in the anther.

The genetical system controlling self-sterility in *Crepis* and *Parthenium*, therefore, is unique in that it is a combination of features from the two sys-

tems of homostyled and heterostyled plants. It may be noted that LEWIS (1944) dismissed the likelihood of the existence of such a system as the one proposed by GERSTEL and the present authors as hardly to be imagined. The only serious difficulty involved, however, concerns the second and fourth features: independent gene action in the pistil and dominance in the anther. In discussing the physiological control of incompatibility in plants, LEWIS states (p. 582) that "plants without heterostyly and heterostyled plants differ in a way which reflects the genetical differences. . . . Only one mechanism has been found in plants without heterostyly, and this appears to be an immunity type of reaction between specific proteins of the pollen and style. This alone has the possibility of variation necessary to meet the requirements of a large multiple allele series and the specificity necessary for the independent gene action in the style."

Since the specificity of proteins is practically unlimited, is it not conceivable that in the Compositae, the most highly evolved division of the Angiosperms, protein relations have developed which provide just the types of specificity required for independent gene action in the style or stigma and dominance in the anther? It should be remembered that, under normal conditions, no *Crepis* pollen will germinate on an incompatible stigma regardless of which allele a particular pollen grain carries. This fact suggests the possibility that in the Compositae the physiological incompatibility mechanism involves differences in osmotic pressure as in *Linum grandiflorum* (LEWIS 1943).

In this connection a statement of GERSTEL (1950) is worthy of repetition: "The reaction is one between cytoplasm produced in the male organ and a pistil. One cannot assume, however, that the same incompatibility substance, or substances, are diffused through the protoplasm of the entire plant, since an incompatibility *reaction* could then not take place; there must be two different substances which are able to react with each other, synthesized at two different places, the androecium and the gynoecium, by the same gene (or, as LEWIS [1949] recently postulated, by two self-reproducing parts between which no crossing over takes place)."

LEWIS (1949) also reports that radiation experiments show that the *S* gene behaves toward X-rays as though it were two independent units, one determining pollen reaction and one determining style reaction. And he alludes to other evidence from polyploid plants which indicates the dual nature of the *S* gene. This concept that the *S* gene is a "dual" entity may prove to be a fruitful idea in future research on self-sterility in the Compositae. But of equal interest is an answer to the question as to whether the genetical system controlling self-incompatibility in *Crepis* and *Parthenium* exists in many other genera, representing all the various tribes, of this great family.

SUMMARY

1. A cross between two self-incompatible plants of *Crepis foetida rhoeadifolia* yielded four intra-sterile groups in its progeny.
2. Some groups were inter-sterile and some were not. Reciprocal differences were found.

3. Progenies were grown from all possible combinations of the four original groups. They were tested by crossing with representatives of all available groups.

4. Seven incompatibility groups in all were found in the F_1 and F_2 from the two original plants.

5. These groups ranged in cross relations from one group which was compatible with all groups but its own, to one which was incompatible with all groups but two.

6. The self- and cross-incompatibilities in this strain of *C. foetida rhoeadifolia* can be explained genetically on the assumption that a single series of oppositional alleles is responsible. They may be referred to as S_1 , S_2 , S_3 , and S_4 .

7. It is postulated that:

a. Pollen behavior is sporophytically or diploid determined. That is, the behavior of a pollen grain is dependent upon the genotype of its parental sporophyte.

b. S_1 is recessive to all three of the other alleles. S_2 is dominant to S_1 but recessive to S_3 and S_4 . S_3 and S_4 are both dominant to the weaker alleles but either is capable of complete expression in the presence of the other. Dominance is not expressed in the pistil.

c. Reciprocal differences are due to dominance, not, as in other known cases, except Parthenium, to homozygosity of one parent.

8. Other genes are believed to be able to exert some modifying effects, especially when associated with the S alleles of weaker potency.

9. The *Crepis*-Parthenium type of self-sterility is compared with other known types and is found to be unique.

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TABLE 2
Cross Relations of Derivatives of H837.

	TESTERS													
	I	II		III		IV		V		VI		VII		
	H837	H870	H837	H870	H837	H870	H837	H870	H870	H870	H870	H870	H870	H870
*H874-12	S	S	S	S	S	S	S	S	S	S	S	S	S	S
H874-24	S	S	S	S	S	S	S	S	S	S	S	S	S	S
*H874-7	SS	SS	SS	SS	SS	SS	SS	SS	SS	SS	SS	SS	SS	SS
H874-6	ff	Sf	Sf	Sf	Sf	Sf	Sf	Sf	Sf	Sf	Sf	Sf	Sf	Sf
*H880-4	SS	SS	SS	SS	SS	SS	SS	SS	SS	SS	SS	SS	SS	SS
*H880-5	S	S	S	S	S	S	S	S	S	S	S	S	S	S
H890-2	S	S	S	S	S	S	S	S	S	S	S	S	S	S
H890-5	SS	SS	SS	SS	SS	SS	SS	SS	SS	SS	SS	SS	SS	SS
*H891-6	S	S	S	S	S	S	S	S	S	S	S	S	S	S
H891-9	S	S	S	S	S	S	S	S	S	S	S	S	S	S
*H891-12	SSS	S	S	S	S	S	S	S	S	S	S	S	S	S
H894-2	S	S	S	S	S	S	S	S	S	S	S	S	S	S
II														
H870-11	FFFF	SSSS	FF	FF	SS	SS	FF	FF	FFF	F	FF	FF	FF	FF
H870-15	SSSS	fSS	Sf	Sf	Sf	Sf	Sf	Sf	F	F	F	F	F	F
H876-21	F	S	S	S	S	S	S	S	F	F	F	F	F	F
H876-16	FF	SS	Sf	Sf	Sf	Sf	Sf	Sf	ff	ff	ff	ff	ff	ff
H880-2	F	S	S	S	S	S	S	S	F	F	F	F	F	F
H880-24	F	S	S	S	S	S	S	S	F	F	F	F	F	F
H880-26	FF	SS	Sf	Sf	Sf	Sf	Sf	Sf	FF	FF	FF	FF	FF	FF
H886-5	FF	SS	Sf	Sf	Sf	Sf	Sf	Sf	FF	FF	FF	FF	FF	FF
H886-16	F	S	S	S	S	S	S	S	F	F	F	F	F	F
H886-17	F	S	S	S	S	S	S	S	F	F	F	F	F	F

Upper letters refer to results obtained when tester plants were used as males; lower letters, when testers were used as females; two or more contiguous letters indicate duplicate or reduplicate tests.
 S-0-10 seeds; f 11-34 seeds; F-35 or more seeds.
 * Male sterile.
 † H837-11 was largely male sterile but was sometimes erratic, in contrast with other male sterile plants.
 ‡ Results considered unreliable because the female parent died, or the branch containing the seed was broken or injured, shortly after pollination.
 § Flower very small which accounts for the small amount of seed, that is, the percent of florets which set seed was relatively high.
 Note: In condensing the above table 15 tested plants were omitted from Group II, four from Group V, nine from Group VI, and seven from Group VII. Their behavior was essentially the same as that of the others.

