

PRELIMINARY OBSERVATIONS ON A NEW TRAIT, OPPOSITE FIRST COMPOUND LEAF, IN *GLYCINE MAX*, (SOYBEAN)<sup>1</sup>

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**Abstract.** A new trait in Strain T219 of *Glycine max* (L.) Merrill, opposite first compound leaves (Opp) followed by alternate subsequent compound leaves as opposed to the usual exclusively alternate compound leaves (NOpp) was found. This strain segregated in a 1:2:1 ratio for dark green: light green: yellow at the  $y_{11}$  locus. In the original sowing of 371 seeds, a 3:1 ratio of NOpp: Opp was observed. Following 4 generations of selection in which only Opps from the seed source with the highest frequency of Opp were raised to maturity, the frequency of Opp decreased from 42% Opp in the P1 generation to 2.2% Opp in the S4 generation. In a parallel selection with the same material, against Opp, the frequency of Opp consistently decreased from 25 to 0%. Water at 47 °C applies to dormant seeds for 2 hr before sowing modified dormant, but seed storage for 2 years had no noticeable effect on, the frequency of Opp.

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Numerous morphological alterations have been observed in soybean. Simple Mendelian loci have been shown to control lanceolate (*ln*) leaflets (Bernard and Weiss 1973), oval-shaped (*lo*) leaflets (Domingo 1945), and multiple-leaflets (*lf<sub>2</sub>*) (Fehr 1972). In Strain T219, we have observed opposite rather than alternate (NOpp) compound leaves. Opposite compound leaves have been ob-

velopmentally because the first compound leaf primordia are present in the dormant seed (Miksche 1961). We attempted to ascertain the mode of inheritance of Opp.

## MATERIALS AND METHODS

Plants were tested by sowing seed in greenhouse benches. Selected plants were raised to maturity and seed collected in a garden. The soybean self-pollinates with ca. 95% efficiency, so all plants were raised to maturity without crosses.

TABLE I  
Parallel Selection For and Against the Opposite First Compound (Opp) Leaf Trait.

Selection for Opp	% Opp	No.	Range (%)	Selection against Opp	% Opp	No.	Range (%)
P 1 (1 plant)	42.0	238	1 plant				
S1-20 Opps from P1	24.0	371	0-60				
I. Plant 1 of S1	44.0	18	1 plant				
S2A from 44 Opps in S1	24.2	752	0-60	S2B from 56 NOpps in S1	25.5	901	0-60
S3A from 9 Opps in S2A	19.6	280	8-32	S3B from 15 NOpps in S2B	9.2	260	0-25
S4A from 20 Opps in S3A	2.2	317	0-12.5	S4B from 11 NOpps in S3B	0.0	190	—
II. Plant 7 of S1	37.0	19	1 plant				
S2C from 34 Opps in S1	31.1	569	5-79	S2D from 15 NOpps in S1	35.0	257	9-60

served at the first (Opp), second (Opp2), and/or third (Opp3) compound leaf nodes. Traits affecting the first compound leaf are of greatest interest de-

In selection for Opp, the plant yielding the highest percentage of seedlings with Opp and with sufficient residue seed for the necessary additional progeny tests was selected. An aliquot of the residue seed from the selected plant was sown and only Opps from that plant were raised to maturity. The selected plants always yielded a high frequency of Opp but

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were not always the plant with the highest frequency because in some cases the highest frequency plant had too small a residue of seed. Selection for NOpp proceeded in the same fashion except that selected plants had the lowest frequency of Opp of all plants tested.

In heat treatment experiments, all seeds were first treated in a dessicator to obtain the desired 6-8% moisture content (Hoskins 1967). All seeds, in small glass bottles, were soaked in water at the 47 °C for 2 hr. Controls were soaked in water at room temperature (22 °C) during the same 2 hr.

#### RESULTS AND DISCUSSION

Strain T219 (Johnson and Bernard 1963) segregated for dark green, light green, and yellow at the  $y_{II}$  locus in a 1:2:1 ratio, respectively. The Opp trait was independent of the genotype at the  $y_{II}$  locus, based on parallel selection for Opp with dark green and light green seedlings. One Opp seedling and one NOpp seedling are depicted in figure 1 (both seedlings are light green).

25.5% in table 1). In subsequent generations, continuous selection against Opp was effective, as indicated by the reduction of Opp to 0% within 3 generations.

Two plants with high seed yield and high frequency of Opp offspring (44% and 37% Opp) were used to compare selection for and against Opp in the first 2 generations. Based on contingency  $\chi^2$ -tests, we concluded that the phenotype of the offspring was independent of the parental phenotype in both plants 1 and 7 (table 1). For plant 1,  $\chi^2=0.317$ ,  $0.5 < P < 0.7$ . For plant 7,  $\chi^2=1.066$ ,  $P > 0.3$ , both with 1 d.f. Comparison of the frequency of Opp between the second generation offspring from plants 1 and 7 (I.S2A vs II.S2C and I.S2B vs II.S2D) indicated that significant differences were obtained following the 2 generations of selection. The grandparental source did not directly reflect

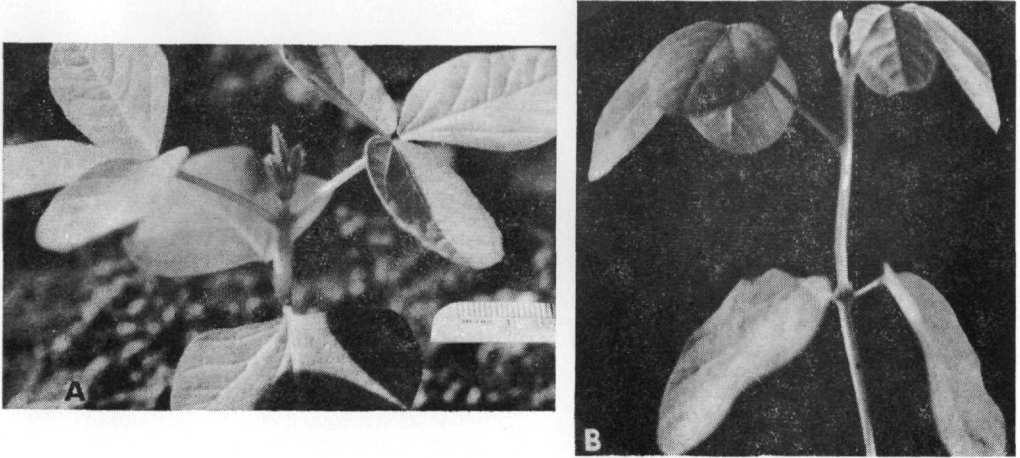


FIGURE 1. (A.) Example of an opposite first compound leaf of soybean strain T219.

(B.) Normal alternate compound leaves of soybean strain T219.

Opp was not a simple Mendelian recessive trait, as Opp did not breed true in the S2 or subsequent generations (table 1). In the P1 generation, the frequency of Opp was 42% while after 4 generations of selection for Opp, the frequency decreased to 2.2%, suggesting that a polygenic trait was not involved. From the same P1, we also selected against Opp. According to t-tests, the first generation of selection for and against Opp did not alter the frequency of Opp (24.2% versus

the frequency of Opp among the second generation offspring because plant 1 had a higher percentage of Opp than plant 7 in its immediate offspring but a lower percentage Opp in its S2 generation. Since plants 1 and 7 were themselves Opp, it is evident that the S2 offspring did not breed true for the phenotype of the grandparent. Similar results were obtained in other experiments using  $Y_{II}/Y_{II}$  seedlings instead of  $Y_{II}/y_{II}$  as in all of the above. In selection against Opp, the

frequency increased from 24% (n=371) to 35.1% (n=1103) in one generation, while selection for Opp in yet another case resulted in a decrease in the frequency of Opp from 53.2% (n=442) to 5.8% (n=498) in one generation.

Increased age of seed at the time of sowing decreases germination percentage (Paddock 1966), increases the frequency of mitotic crossing over, increases the frequency of seedlings that lack geotropism (Evans 1975), and increases the frequency of chromosomal aberrations (Sax and Sax 1961).

We observed in some early progeny tests that seeds sown immediately after harvest had a lower percentage of Opp than expected. A possible age effect on the frequency of Opp was therefore examined. As part of a larger group of progeny tests, data pooled from 3 of the  $Y_{11}/y_{11}$  Opp plants (No. 63, 64, and 91) arising from plant 7 were 75% NOpp and 25% Opp among 69 offspring when sown only 20 days after harvest (table 2).

TABLE 2

*Effect of Age on the Progeny of Three  $Y_{11}/y_{11}$  Opp Plants Whose Grandparent and Respective Parent Plants were Opp.*

From Plant 7 No. 63, 64, 91	NOpp	Opp	Total*	Germination (%)
1974 sowing	52	17	69	92.0
1976 sowing	99	47	146	99.3
Total*	151	64	215	

\*Contingency  $\chi^2=0.9431$ , 1 d.f., n.s.

To assess the effect of seed age and to reduce variability due to weather-related greenhouse conditions, seed from the same 3 plants were sown on the same date 2 years later (27 October 1976) to ascertain if the frequency of Opp was altered. Germination was 99.3% (eliminating the possibility of differential germination selectivity) and 68% NOpp, 32% Opp was observed among 146 seedlings. A contingency  $\chi^2$ -test implied equality of the 2 sowings with regard to percentage of Opp and hence no effect of seed storage age.

Preliminary tests to measure possible distortion of phenotypic ratios at the  $y_{11}$

locus by heat treatments suggested that the frequency of Opp varied considerably. To test this frequency, two  $Y_{11}/y_{11}$  Opp seed source plants were used: plant 111 with 16% Opp among 25 offspring and 112 with 17.4% Opp among 23 offspring. Another 250 seeds of each plant were sown, 125 as controls and 125 treated with 47 °C for 2 hr (table 3). Germina-

TABLE 3

*Contingency  $\chi^2$ -tests for the Effect of Heat on the Expression of Opp in Progeny of Two  $Y_{11}/y_{11}$  Opp Plants.*

Plant No.	Treatment	NOpp	Opp	Total
111	Control	95	16	111
	47 °C, 2 hr	51	26	77
	Total*	146	42	188
112	Control	75	33	108
	47 °C, 2 hr	36	41	77
	Total**	111	74	185

\* $\chi^2=8.731$ , highly significant.

\*\* $\chi^2=8.721$ , highly significant.

tion percentages were 90.4 and 88 in controls, 62.4 and 63.2 in heated for plants 111 and 112, respectively. The percentage of Opp among controls was 14.4 among 111 offspring of plant 111, and 30.6 among 108 offspring of plant 112. Among seedlings from heat treated seeds, 33.8% Opp from plant 111 and 53.2% Opp from plant 112 were observed. For each plant, contingency  $\chi^2$ -tests for Opp resulted in values of 8.731 for plant 111 and 8.721 for plant 112, both significant with  $P<0.001$ . Since most other variables were held constant, we concluded that heat increased the frequency of Opp, making this an environmentally modifiable phenotype. We suggest two possible mechanisms: (1) heat may kill cells essential to internode elongation at the embryo shoot apex, (2) heat may inactivate chemicals which otherwise inhibit formation of a second leaf primordium at the first compound leaf position. Aside from these mechanisms, seedlings with Opp may inherently have greater heat resistance. In Opps, one leaf sometimes becomes fully expanded earlier than the other leaf, lending support to the first hypothesis which relies on internode elongation.

Our results on the selection for Opp were consistent with a nongenetic determination of the Opp trait within Strain T219. To account for the high frequency of Opp in this strain, it may be necessary to conclude that Opp is a result of a homozygous genetic system with a variable and easily modifiable expressivity and not sufficient in itself to result in Opp.

#### LITERATURE CITED

- Bernard, R. L. and M. G. Weiss 1973 Qualitative Genetics, pp. 117-186. In: Caldwell, B. E., (ed.) Soybeans. Amer. Soc. Agron.
- Domingo, W. E. 1945 Inheritance of number of seeds per pod and leaflet shape in the soybean. Jour. Agr. Res. 70: 251-268.
- Evans, D. A. 1975 Relationships of heat treatments of seeds in altering segregation ratios and frequencies of somatic crossovers in *Glycine max*. Unpubl. M.S. Thesis, Ohio State Univ., Columbus.
- Fehr, W. R. 1972 Genetic control of leaflet number in soybeans. Crop Sci. 12: 221-224.
- Hoskins, R. W. 1967 Alteration of spot frequency in heterozygous seedlings of *Glycine max* (L.) Merrill (soybean) by radiation. Unpubl. M.S. Thesis, Ohio State Univ., Columbus.
- Johnson, H. W. and R. L. Bernard 1963 Soybean genetics and breeding. Advances Agron. 14: 149-221.
- Miksche, J. P. 1961 Developmental vegetative morphology of *Glycine max*. Agron. Jour. 53: 121-128.
- Paddock, E. F. 1966 Mendel could have, your students can. Sci. Teacher 33: 21-23.
- Sax, K. and H. J. Sax 1961 The effect of age of seed on the frequency of spontaneous and gamma-ray induced chromosomal aberrations. Radiat. Bot. 1: 80-83.
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