INSTITUTE OF GENETICS AND PLANT BREEDING
Academy of Agriculture
Wojska Polskiego St. 71 c
60-625 Poznań, POLAND

Poland

1) Identification and evaluation for mutation of agricultural characters in soybean.

In genetic and breeding work for soybean, it is essential to establish a variety adaptable to the climatic conditions of Poland. Induction of mutations is likely to broaden the variation range and help select traits favorable from the breeders' viewpoint. An attempt to induce mutations was made on material genetically differentiated. Careful attention was paid to the prospect of selecting mutant variation in the M_2 and M_3, relating to the length of growing period, morphotype and yield capacity.

Materials and methods: Seeds of 14 soybean varieties were treated with various mutagenic agents in three combinations: (1) radiation with 10 kR; (2) radiation with 10 kR in addition to 10^{-3}M NaN_3 solution against 0.1M phosphate buffer at pH 3 for 3 hr; (3) radiation with 10 kR, presoaking in H_2O_2 for 3 hr, treatment with NaN_3 solution. In the M_1, the number of well-developed seeds recorded for the three combinations were as follows: 2766, 875, and 173, respectively. Totally, 3814 seeds were obtained from gamma radiation combinations (Table 1). Seed of 7 varieties were treated with chemicals for 18 hr. From these combinations, 4112 seeds were obtained in the M_2 (Table 1). Seed of the M_2 generation and M_3 lines, derived the following year from the M_2 mutants, were grown in the field in rows spaced 10 x 30 cm. Observations were made on developmental stages, morphological traits, and yield capacity of single plants.

Results: A comparison between controls and M_2 plants revealed conspicuous differences in the emergence rate and number of emerging plants. The M_2 plants emerged approximately two weeks later than the controls. The highest number of late-emerging plants were recorded for the varieties 'Herb 622' and 'Nordia', despite their high emergence percentage (Table 1). Poor and uneven emergence was noted for the M_2 of 'Altona', 'Mazowiecka II', 'Vansoy', 'Ik-9', and 'Woronożeska'. Totally, 69.1% plants emerged from gamma radiation in the M_2 and 80.7% from chemical mutagen treatment (Table 1). At the beginning of the growing period, remarkable morphological abnormalities were observed. They affected primarily the onset of single leaflets at cotyledon angles and above the first pair of true leaves, atrophy of the top of main stem in plants 10-20 cm high, the size and shape of leaf-blades. Deformations of leaflets at the true leaf were observed over the whole growing period and were even visible at apical leaves. The abundance of abnormal leaves was largest in the M_2 of the following varieties: Mazowiecka II, Herb 622, Altona, and Vansoy. The high number of deformations, in addition to chlorophyll aberrations, manifest an effective action of mutagens on M_2 plants. All plants showing any chlorophyll aberrations were recorded. In some combinations (e.g., the M_2 of Altona, IInd combination) the number of aberrations reached 43.9% (Table 1). No regularity was observed between the frequency of chlorophyll aberrations and that of induced mutations.

As far as the phenotypic traits are concerned, the uneven emergence seems to have caused the differentiated growth rate and additionally influence the

Table 1. Emergence of plants and frequency of chlorophyll aberrations and mutants in M_2 generation

Variety/ combination of treatment	No. grown seeds	Emergence		Frequency of chlorophyll aberrations		Frequency of mutants	
		No.	%	No.	%	No.	%
Altona	50	50	100.0				
I	170	111	65.3	44	39.6	4	3.6
II	88	57	64.8	25	43.9	–	–
III	141	131	92.9	36	27.5	7	5.3
American Yellow	50	36	72.0				
I	147	91	61.9	18	19.8	6	6.6
Amurska	50	44	88.0				
I	373	304	81.5	46	15.1	19	6.2
II	85	69	81.2	11	15.9	4	5.8
Fiskeby V	50	37	74.0				
I	183	140	76.5	31	22.1	9	6.4
II	91	74	81.3	15	20.3	1	1.3
III	32	31	96.9	10	32.3	3	9.7
Flambeau	50	40	80.0				
I	282	246	87.2	61	24.8	12	4.9
II	158	119	75.3	47	39.5	4	3.4
Mazowiecka II	50	43	86.0				
I	644	414	64.3	48	11.6	19	4.6
II	78	52	66.7	15	28.8	–	–
Vansoy	50	43	86.0				
I	967	551	57.0	78	14.2	10	1.8
II	375	245	65.3	26	10.6	2	0.8
Total from combinations after radiation	3814	2635	69.1	511	19.4	100	3.8
Bydgoska	50	35	70.0				
0.025% MMS	402	325	80.8	42	12.9	12	3.7
Herb 622	50	40	80.0				
0.025% EES	984	756	76.8	59	7.8	14	1.8
IK-8	50	41	82.0				
0.025% MMS	198	155	78.3	24	15.4	5	3.2
IK-9	50	42	84.0				
0.025% EMS	296	168	56.7	29	17.3	9	5.3

Table 1. *Continued*

Variety/ combination of treatment	No. grown seeds	Emergence		Frequency of chlorophyll aberrations		Frequency of mutants	
		No.	%	No.	%	No.	%
Mari-czau-vida	50	45	90.0				
0.015% NMU	592	504	85.1	176	34.9	5	1.0
Nordia	50	44	88.0				
0.025% NMU	1268	1161	91.6	81	7.0	9	0.8
Woronożeska	50	41	82.0				
0.025% IPMS	372	248	66.7	32	12.9	9	3.6
Total from combinations with EMS and derivatives and NMU	4112	3317	80.7	443	13.4	63	1.9

variation in developmental stages and length of growing period, particularly in Amurska, Mazowiecka II, Vansoy, and IK-8. Therefore, it was difficult to assay the extent to which abnormalities in M_2 plants were of hereditary character or resulted from physiological action of mutagens on germination and development of these plants. There are different opinions on induction of mutations in soybeans for performance traits, such as shortening of growing period, in addition to increased yield and changeability of morphological characters (Johnson, 1959; Dżosan et al., 1974; Ala, 1976; Siczkar, 1981). In the M_2 generation, a conspicuous range of variation was noted for these traits (Table 2). Hence, a good chance was offered to select for mutants prospective for crossing programs. On the basis of observations throughout the growing period and biometric measurements taken after harvest, a number of plants were selected, initially assumed as mutants. The criteria for selection were: distinct alterations in the length of growing period, seed yield, and morphological traits. As a result, 163 M_2 plants were selected (3.8 and 1.9% of plants emerged) for further examination in the M_3. Due to high damage by diseases, var. Mari-czau-wida was excluded from the study. No differences between M_3 and control plants were noted in the emergence rate and number of emerging plants. Also, no single mutant line showed shortening of the phase from planting to emergence. The plants emerged some 19-20 days after sowing. During the growing period, observations were made of atypically shaped, wrinkled, and ill-colored leaflets. Also, records were taken of differences between lines and controls with respect to growth rate, plant habit, and color intensity (e.g., at the age of three weeks, plants of three mutant lines derived from Fiskeby V were 20.0 cm higher than the controls). On the other hand, the M_3 plants of 'Herb 622' were observed to have distinctly slower growth rate. These differences were recorded throughout the growing period. A single line of the M_3 of Fiskeby V exhibited severe chlorosis resulting in necrosis of 33 from 45 plants examined at seedling stage (73.3%). The highest variation in the M_3, as compared with the controls, was recorded for the length of flowering and growing period and seed yield per plant. In the present study, flowering occurred in dry weather, reported to determine seed yield at this stage of

Table 2. Differences between M_2 plants and initial varieties in the length of growing period, morphological traits and yielding capacity

Variety M_2 generation	Length of flowering period	Length of growing period	Height of plants (cm)	No. of lateral branches	No. of pods up to 15 cm	No. of seeds
Altona	25.0	142.0	62.3	1.3	6.1	46.1
	+7.0	-7+16.0	-40.3+24.0	-1.3+2.7	-6.1+5.9	-45.1+59.0
American Yellow	20.0	154.0	70.4	3.7	5.8	49.9
	--	--	-17.0+18.0	-2.7+3.3	-5.8+19.2	-44.9+108.1
Amurska	20.0	154.0	103.0	3.2	0.1	54.1
	+21.0	-10.0+23.0	-57.0+37.0	-3.2+3.8	+11.9	-53.1+219.0
Fiskeby V	16.0	128.0	41.3	1.8	10.3	35.5
	+5.0	-9+15.0	-24.0+17.0	-1.8+6.2	-9.3+17.7	-34.5+54.3
Flambeau	22.0	156.0	70.9	2.9	2.3	36.6
	--	--	-24.0+32.1	-2.2+5.1	-2.3+10.7	-35.6+136.4
Mazowiecka II	21.0	147.0	59.7	3.8	12.3	93.9
	+18.0	-7.0+16.0	-42.7+17.3	-3.8+3.2	-12.3+27.7	-92.9+134.1
Vansoy	33.0	153.0	87.1	4.3	0.4	52.0
	-4.0+18.0	+24.0	-35.1+42.9	-4.3+4.8	-0.4+6.6	-51.0+183.0
Bydgoska	30.0	147.0	69.4	3.8	5.2	28.4
	--	-5.0	-32.4+11.1	-3.8+2.2	-5.2+18.8	-26.4+110.6
Herb 622	26.0	148.0	57.3	3.7	1.9	33.0
	--	-12.0	-38.8+29.7	-3.7+1.3	-1.9+10.1	-32.0+147.0
IK-8	30.0	150.0	69.6	2.4	1.1	31.2
	+4.0	-10.0+12.0	-31.6+45.4	-2.4+4.6	-1.1+8.9	-30.2+156.8
IK-9	19.0	148.0	58.6	3.1	2.5	28.9
	+14.0	-6.0	-35.6+21.4	-3.1+2.9	-2.5+9.5	-27.9+63.1
Mari-czau-vida	23.0	142.0	47.8	1.9	4.9	27.5
	-4.0	--	-18.8+28.2	-0.9+3.1	-4.9+9.1	-24.5+62.5
Nordia	25.0	140.0	50.5	2.4	5.9	24.2
	+5.0	+7.0	-25.5+25.0	-2.4+2.6	-5.9+11.1	-22.2+68.8
Woronożeska	19.0	142.0	52.3	4.0	7.0	45.9
	--	-11.0	-20.3+13.7	-3.0+4.0	-7.0+24.0	-43.9+107.1

Table 3. Characteristics of selected mutants of M_3 generation

Variety/ Combination treatment	Traits differing mutants from the initial variety
Altona	Onset of flowering belated by 5 days; length of flowering period shortened by 3 days.
10 kR	Growing period the same as Altona. Plants higher by 10.3cm. First pod set higher. Seed yield higher by 13.5%
Fiskeby V	End of flowering 6 days later growing period 5 days longer than Fiskeby. Plants higher by 7.1cm; first pod set higher by 2.1cm. Seed yield higher by 10.0%
10 kR + H_2O_2 + NaN_3	The same type of mutants within three mutant lines. Onset of flowering 7 days later and end of flowering 3 days later than Fiskeby. Growing period longer by 3 days. Plants higher by 15.7cm; first pod set higher by 3.2cm. Seed yield lower by 30.7%
Amurska	Flowering period longer by 9 days; growing period the same as Amurska. Plants lower by 8.3cm; first pod set 4.1cm lower, more lateral branches. No. of seeds higher by 28.6%; seeds markedly smaller in size; 100-seed weight lower by 2.6g
10 kR + NaN_3	Onset of flowering belated by 15 days; flowering period shorter than Amurska by 7 days. Growing period the same as controls. Plants lower by 5.9cm. Seed yield higher by 14.0%; seeds large and plump
Mazowiecka II 10 kR	Mutant lines showing very wide variation in the length of flowering and growing periods. Within three mutant lines growing period longer by 12 days. Plants higher by approx. 10.6cm. Seed yield higher by 46.0%
	Seven mutant lines with growing period and variation range similar to Mazowiecka II. Also, seed yield higher and ranging from 11.0 to 23.0%. One mutant line exhibited the same growing period and plant height as the control but seed yield higher by 43.0%
Woronożeska 0.25% IPMS	Two mutant lines with growing period shorter than the control by 7 days. Plant height lower by approx. 10.0%. Seed yield lower by 24.5%.
	One line with the same growing period as the control; plant height lower by 10.0cm and seed yield lower by 25.0%. One mutant line higher by approx. 6.7cm; no differences in growing length and yielding capacity
Herb 622 0.025% EES	Three mutant lines with growing period shorter by 15-25 days. Height of two lines lower by approx. 11.0cm and seed yield lower by 10.0%. The highest decrease in seed yield (41.0%) was noted for the earliest mutant line whose plants were lower by 33.3cm
Bydgoska 0.025% MMS	Growing period and seed yield the same as controls; plant height lower by 24.9%

Table 4. Frequency of selected mutant lines in M_3 generation

Variety/ combination of treatment	No. of selected M_3 lines	% selected M lines against: M_3 line of a given variety	M_3 line of the same combination of treatment	M_2 plants
Altona				
I	1	9.1	25.0	0.9
Amurska	2	8.7		
I	1	4.3	5.3	0.3
II	1	4.3	25.0	1.4
Fiskeby V	4	30.8		
I	1	7.7	11.1	0.7
III	3	23.1	100.0	9.7
Mazowiecka II				
I	11	57.9	57.9	2.7
Bydgoska 0.025% MMS	1	8.3	8.3	0.3
Herb 622 0.025% EES	3	21.4	21.4	0.4
Woronożeska 0.025% IPMS	4	44.4	44.4	1.6
TOTAL	26	15.9	--	1.1

plant development (Mackiewicz, 1959; Szyrmer, 1971). Despite the atypical conditions, attempts were made to find out an interdependence between the length of flowering and seed yield. No data to support this hypothesis were obtained, although 14 lines of the M_3 exhibited shorter or longer flowering periods than the controls. Considerable alterations in the growing period were noted for the M_3 of Mazowiecka II, Woronożeska and Herb 622 (Table 3). Not a single mutant was found to have distinctly shorter growing period and higher seed yield. It is worth mentioning that neither mutants of the M_2 and M_3 of very late maturation varieties exhibited such characters (Amurska, Flambeau, Vansoy). Of interest were lines which, despite growing periods very much like the initial line, were characteristic of higher seed yield (e.g., M_3 line of Altona, Amurska, Mazowiecka II). Despite favorable phenotypic traits of M_3 lines of Amurska and Mazowiecka II, their late maturation poses a serious handicap, prolonged growing likely to end in autumn and meet bad weather conditions known to produce difficulties at harvest. Most of the examined mutant lines produced from 4.1 to 5.7 g seeds per plant and had the same length of growing period and variation of morphological traits as the control plants (M_3 American Yellow, Flambeau, Vansoy, IK-8, IK-9, Nordia). Mutant lines of Vansoy had seed yield per plant reduced by half and those of American Yellow by 35.0% as compared with the initial variety. These mutants were eliminated from further studies. According to Malczeno (1970) there is little chance to improve traits of poor early mutants in the following generations. Successful selection for mutants stem from the early variety Fiskeby V, as well as for mutants with a considerably shortened growing period derived from late varieties (Herb 622, Woronożeska) is considered advantageous (Table 3). Differences in growing period, morphological traits, and yield were a good indicator of the tolerance level of mutant lines to growing conditions, as well as forming a good basis for selection of mutants.

For further studies, 26 mutant lines were selected, i.e., 15.9% of lines from the M_3 generation and only 1.1% of plant populations from the M_2 generation (Table 4). From an analysis of M_2 plants and one year observation of mutant lines of the M_3 generation, it can be assumed that the examined lines manifest a good variation, likely to contribute to crossing and selection work ahead.

References

Ala, A. 1976. Genetika 1:33.

Dżosan, N. and J. Nikolaja. 1974. Genetika 6:43.

Johnson, H. W. 1959. Soybean breeding. Handbuch d. Pflanzenzüchtung 29.

Mackiewicz, Z. 1959. Hodowla Roślin Aklimatyzacja i Nasiennictwo t. 3:24.

Malczenko, W. W. 1970. Genetika 6:11, 64.

Siczkar, W. J. 1981. Genetika 12:2184.

Szyrmer, J. 1971. Zeszyty Naukowe SGGW nr 112. Warszawa.

Halina Skorupska