

Weight of Fruits in Self-Fertile, Male-Sterile, and Self-Sterile Diploid and Tetraploid Monogerm Beta Vulgaris L.

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Introduction

The replacement of multigerm sugar beet varieties by monogerm varieties is approaching completion in the USA (23)². The first phase of developing monogerm varieties, usually based on application of the backcross method for incorporation of the gene *m* (monogermity) into multigerm varieties is almost accomplished. At the present time, a second phase of breeding of monogerm varieties is starting. This phase is the routine improvement of monogerm sugar beets in different agronomic characters, including quality of the monogerm fruits themselves.

The weight of fruits and germs of monogerm beets has a practical importance in agriculture as one of the factors determining the quality of seed (18). During the past few years, utilization of differences in the size of fruits were used in breeding and seed production in the United States and in Europe for different purposes. In the USA male-sterile monogerm beets have been planted together with the multigerm pollinator to obtain commercial monogerm hybrids. The harvested monogerm seed (hybrid seed) has then been separated from the multigerm seed by screening (25,26). In Europe, seedballs of the multigerm sugar beets have been screened for size to increase the percent of triploids in the commercial polyploid (anisoploid) varieties (1,3,4,24).

Because the seedballs of tetraploids are larger, screening out the small seedballs does eliminate a portion of the diploids. In the variety Polybeta, for instance, seedballs 2 to 3 mm in diameter contained 39.4% diploids, whereas larger seedballs (5 mm in diameter) contained only 16.3% diploids (22). Schneider (21) reported that small seed contained 68% diploids and the large seed, 10.9%. According to Belgian data, small seed of Polybeta contained 49.06% diploids and the large seed (5 mm in diameter), 15.93% diploids (6,7). Therefore, retention of larger seedballs decreased the percent of diploid beets in the anisoploid varieties.

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² Numbers in parentheses refer to literature cited.

F_2 and F_3 generations of hybrids between monogerm beets and multigerm varieties give monogerm plants differing in the weight of fruits and germs. Some monogerm segregates develop small fruits (15,16). This is a natural consequence of inability to control the genes responsible for the weight of fruits in multigerm populations.

In the first phase of backcross selections for monogerm beets, the variability of stored genes in multigerm varieties responsible for the weight of fruits was used. The value of such stored genes in determining weight of monogerm fruits is based on the homeostatic mechanism which controls the interaction of these genes with the gene *m* in monogerm sugar beets (8).

Selection for desirable characters in monogerm fruits is necessary for further seed improvement. Selection for weight of fruits and germs has not been attempted in multigerm populations. Monogermity has made possible selection for improvement of the many characters of fruits and germs (2,15,16,18). No multigerm sugar beet population of proven high quality of fruits (size of fruits and germs, shape of fruits, etc.) was available which could be used as a recurrent parent for improvement of monogerm fruits. Further improvement of monogerm fruits is possible only by hybridization and selection of monogerm beets, and not by finding stabilized valuable characters of fruits and germs in the multigerm populations, which could be used for incorporation in the monogerm beets.

Information concerning the variability of weight of the monogerm fruits and germs under different environment and genetic factors, types of matings, selection, and ploidy levels is presented in this report.

Materials and Methods

The following monogerm and multigerm breeding materials developed by the authors at the Salt Lake City station were used for a study of the weight of fruits: 10 diploid monogerm self-sterile backcross populations; 12 diploid monogerm self-fertile inbreds; 18 tetraploid monogerm self-sterile brother-sister lines; 18 tetraploid monogerm self-fertile inbreds; 1 diploid and 1 tetraploid male-sterile equivalent of the monogerm inbred SLC 91; 1 tetraploid multigerm open-pollinated population of US 104; 1 tetraploid monogerm population US 35/2; and some other populations.

All diploid self-fertile inbred lines were derived from hybridization of the original monogerm SLC 101 with different American multigerm varieties (15,19). The diploid monogerm self-sterile stocks were obtained after three backcrosses of monogerm

SLC 3 to different self-sterile multigerm open-pollinated varieties (11,12,15). In every backcross the multigerm pollinator was represented by 30 to 100 plants. The male-sterile monogerm equivalents of SLC 91 carried male-sterile cytoplasm of the variety Janasz (19).

The tetraploid monogerm beets were obtained in two ways. Some of them, as SLC 15 mm self-sterile, SLC 91 mm self-fertile and its male-sterile equivalent SLC MS 91 mm, were obtained by colchicine treatment of the corresponding diploid stocks (10). All other monogerm self-fertile and self-sterile stocks were obtained by selection of monogerm tetraploid plants from hybrids between monogerm tetraploids SLC 15 and SLC 91, with the tetraploid multigerm population derived from US 35/2 (19,20).

All experiments were conducted near Salt Lake City, Utah, during 1958, 1959, and 1960. Seed beets were grown in 1-row plots 30 to 50 ft long in overwinter plantings, or by planting of stecklings in spring. The seed plants were not damaged by curly top, leaf spot, or virus yellows. Stecklings destined for each experiment were harvested the same day from plots grown in one field.

One sample for determination of the weight of 1000 fruits³ was taken from each plot, except in two experiments where the weight of 1000 fruits was studied in tetraploid self-sterile and self-fertile monogerm lines. In each of these plots, seed of 10 plants was harvested individually (Tables 9,10).

The number of replications in the experiments is indicated in tables giving the analysis of variances. To study the weight of germs³, fruits were treated by 4-5% hydrochloric acid (HCl) and dried to a constant weight in the oven at 50° C (13).

Experimental Results

Weight of fruits of multigerm beets

The fruit (seedball) of a multigerm sugar beet is a phylogenetically compound structure which accomplished several functions in the evolution of *B. vulgaris* L. In wild *Beta* species the multigerm seedball provided certain advantages of survival value. The presence of several germs in one fruit increased the number of seedlings. The presence of several flowers in a cluster extended the time of flowering of a cluster and increased the amount of pollen produced, which is of importance for the cross pollination of species.

Variability of the weight of germs within a seedball is characteristic of multigerm fruits. Up to a certain optimum, an in-

³ Weight of fruits and/or germs always designate weight of 1000 fruits or/and 1000 germs.

crease in the number of flowers in a cluster not only increases the variability of the weight of germs, but as shown in our experiments, one particularly large germ appears.

Natural and artificial selection controls the weight of seedballs in multigerm beets by affecting the number and the size of individual fruits in the seedball. This in turn influences the weight of germs in the seedballs.

The size and number of individual fruits within a seedball essentially are controlled by different genetic factors. This is indicated by the fact that the weight of seedballs and the number of individual fruits in them are only partially correlated genetically and environmentally. The environment coefficient of correlation for these characters equals -0.42 , while the genetic coefficient of correlation is lower and equals $+0.24$ (14). The weight of seedballs and number of individual fruits within a seedball correlate differently with other characters of seed beets (14). Also, genetic and environment variability is different for these 2 characters (14). A joint action of these 2 different groups of genes may increase the weight of the seedballs by increasing the number or the weight of individual fruits and sometimes by simultaneous augmentation of both.

Doubling of chromosomes increases the weight of seedballs (Table 1) but influences differently the two elements condition-

Table 1.—Weight of fruits (1000) in diploid and tetraploid open-pollinated sugar beet populations.

Variety	Diploid	Tetraploid
	grams	grams
US 35/2 multigerm	15.70	34.00
US 401 multigerm	26.60	36.20
SLC 15 monogerm	11.17	19.70

ing them. Increase of the weight of seedballs in multigerm tetraploids is caused by the increased weight of individual fruits within a seedball rather than by the greater number of flowers per cluster. The tetraploids usually develop fewer flowers per cluster than the original diploids (Table 2).

Table 2.—Percent of seedballs with different number of individual fruits in diploid and tetraploid sugar beet populations of US 401.

Level of ploidy	Number of fruits in the seedball			
	1	2	3	4 and more
	Percent of seedballs			
Diploid US 401	—	48	47	5
Tetraploid US 401	11	67	22	

Thus, the effect of these two characters on the weight of seedballs cannot be attributed solely to the simple pleiotropic action of the same set of genes on the number and size of individual fruits in the seedball of the multigerm beets.

Variability of weight of germs within a multigerm seedball and in monogerm fruits

Weight of germs in the individual fruits within a seedball is usually very different. In a Russian sugar beet variety "Uladovka" the large multigerm seedballs sifted on 3 mm sieves contained large, medium, and small germs. The weight of 1000 large germs equaled 3.7 gms, of 1000 medium germs 2.6 gms, and a 1000 small germs weighed 2.2 gms (27,28).

We studied the variability of the weight of germs within the seedballs taken at random in an open pollinated population of the variety US 35/2. The ratio of the weight of the largest and the smallest germs within three germ seedballs equaled 7:1 (Table 3). Such a big variation in the weight of germs was caused by a high reduction of the weight of the small germ, as well as by the poor development of some small germs. A considerable number of the small germs is inviable, although other small germs develop normally within a seedball and are able to germinate.

Table 3.—Weight of large, medium and small germs (1000 germs) in the seedballs with 2 and 3 fruits in the diploid open-pollinated sugar beet population US 35/2.

Number of flowers per cluster	Size of germs		
	Large	Medium	Small
	grams	grams	grams
3	4.75	2.50	0.65
2	4.26	—	3.45

In the population of US 35/2 the range of variation of the weight of germs was greater for seedballs with 3 fruits than for seedballs with 2 fruits (Table 3). This type of variability in the weight of germs within a seedball, which is caused by different number of fruits in seedballs, is more or less similar for different sugar beet varieties and for seed grown in different environments (years, soil, location). Therefore, seedballs containing 3 or 4 fruits usually have 1 germ of greater weight and 1 germ lower in weight than the weight of the largest and smallest germ in the double-fruited seedballs of the same seed sample.

Since there can be no variability of the weight of germs within a fruit of monogerm beets, the genes responsible for

monogerm and multigerm characters exhibit different pleiotropic effect on the variability of the weight of germs. Screening of seedballs of the multigerm beets according to size, or segmentation of seedballs, does not alter the variability in weight of germs within a seedball. Segmentation of seedballs leads to separation or to destruction of germs and results only in production of segments which contain one germ of any weight.

In monogerm beets the variability of weight of germs is determined by the variability of weight of fruits only. The coefficient of correlation between weight of fruit and germ equals 0.949 (16). Therefore, in monogerm beets the weight of germs may be controlled by separation of seeds of different size (18,19). Because of the absence of variability within a fruit, the range of variability of weight of germs in monogerm beets is less than in the multigerm beets (Table 4). Weight per 1000 germs in small and in large monogerm fruits varied from 2.9 to 4.70 gms. In the multigerm parental variety US 35/2 the range of variation in the weight of small and large germs was 0.65 to 4.75 gms (Table 3). The effect of transition from the multigerm to the monogerm condition is manifested in a sharp decline of variability in the weight of germs in the new monogerm population as compared to the previous multigerm populations.

Table 4.—Weight of fruits (1000) and germs (1000) in the diploid open-pollinated self-sterile monogerm sugar beet population SLC 15 derived from backcrosses of monogerm beets to multigerm variety US 35/2.

Size of monogerm fruits	Fruits	Germs
	grams	grams
Large	20.10	4.70
Small	12.59	2.90

The monogerm character acts as a stabilizing agent in the genetic composition of populations in M-m allele and in the corresponding linkage group (17,19). The different influence of individual genes members of the multiple allele M-m on weight of germs peculiar to multigerm beets disappears in the monogerm beets. In connection with this bias of all multigerm alleles in estimation of weight of fruits and germs also is eliminated. Accumulation of genes favorable for the weight of fruits and germs becomes simpler. A new stable interaction between gene *m* and all other genes causing variability of the weight of fruits is established. The former gene pool for the weight of fruits in the multigerm populations acquires a new level of expression in the monogerm populations.

Weight of fruits in diploid backcross monogerm open-pollinated populations and in monogerm self-fertile inbred lines

Backcrossing is the usual method for breeding of the monogerm beets (15,19). For the evaluation of the effect of backcross method on the variability of the weight of monogerm fruits, the monogerm self-sterile populations obtained after repeated hybridization of self-sterile monogerm beets with different multigerm varieties (US 35/2, US 216, US 22/3, Klein E, Klein Z etc.) were used. In the following generations, which were segregating for the monogerm character, the multigerm segregates were discarded and the monogerm plants were crossed again with the corresponding recurrent parents in open-pollinated populations. When the backcross pollination was discontinued, the monogerm plants were intercrossed. In 10 such open-pollinated backcross populations the weight of 1000 monogerm fruits varied from 16.3 to 19.0 gms, i.e., the difference was 2.7 gms. The difference in weight of monogerm fruits in the backcrossed populations, however, was not statistically significant, because the standard error obtained on the basis of analysis of variances in 10 populations in 3 replications almost equaled this value (2.7882 gms). The calculated F-value was 1.09, whereas the tabulated F-value should be a minimum of 2.46 (Table 5).

Table 5.—Weight of fruits (in grams) in 10 sugar beet diploid monogerm self-sterile populations derived from backcrosses of monogerm beets to different multigerm varieties. Analysis of variance.

Factors	Degree of freedom	Variance	F		
			Calculated	5% Level	1% Level
Total	29				
Between populations	9	3.0533	1.0951	2.46	3.60
Between reps.	2	1.3295	0.4768	3.55	6.01
Experimental error	18	2.7882			

Absence of significant differences in the weight of monogerm fruits in backcrossed self-sterile populations is caused by the absence of differences in weight of fruits in different parental sugar beet multigerm populations. Multigerm populations have not been exposed to selection for this character. Therefore, the multigerm sugar beet varieties do not differ in the average weight of the fruits (14). This is one of the reasons why the weight, or the size of fruits, were never used as distinctive characteristics of the multigerm sugar beet varieties. There are no indications in the literature concerning hereditary varietal differences in the weight of fruits in multigerm sugar beets. At

the same time the individual seed samples, even the seed samples of the same variety, may differ in the weight of fruits, but these differences are usually caused by the locations, or by the conditions of seed growing.

Other studies of variability were made of monogerm fruits using self-fertile inbred lines obtained from selfing of backcross hybrids between multigerm US 35/2 and the self-fertile monogerm inbred SLC 101. It was found that inbred monogerm lines isolated from the backcross progeny of repeated backcrosses to the same multigerm population differed in weight of fruits.

Presence of monogerm inbreds with different fruit weights indicates that the original multigerm populations were heterogeneous in the genes determining fruit weight. Multigerm open-pollinated varieties carry an intravarietal storage of genetic variability in genes determining weight of fruits. These genes are maintained in a certain equilibrium in open-pollinated multigerm populations (14). By backcrossing of monogerm beets to the multigerm populations this system of stored variability is recovered in the same equilibrium in the backcross monogerm populations.

Inbreeding applied to such backcross populations destroys their genetic equilibrium and leads to isolation of genetic combinations and to the production of inbred lines differing in the weight of fruits. The grade of differences in the weight of monogerm fruits, fixed by inbreeding, will be proportional to the heritability of multigerm pollinators.

Comparison of the weight of fruits in 12 monogerm diploid inbreds shows that the value of variance among inbreds equals 19.46, whereas the variance between the blocks is only 2.37 (Table 6). The calculated value of F among inbreds is 9.25, which is statistically significant. A high value of variance among inbreds indicates the presence of significant stored heritability in the diploid multigerm populations and the genetic differences between monogerm inbreds isolated from these populations.

Table 6.—Weight of fruits (in gram) in 12 sugar beet diploid monogerm self-fertile inbreds. Analysis of variance.

Factors	Degree of freedom	Variance	F		
			Calculated	5% Level	1% Level
Total	59				
Between populations	11	19.4601	9.2553*	2.01	2.68
Between reps.	4	2.3656	1.1251	2.58	3.78
Experimental error	44	2.1026			

* significant

Weight of fruits of 12 diploid inbreds studied varied from 12 to 18 gms. Of these 12 lines, only 3 significantly exceeded (in the weight of fruits) the standard monogerm inbred SLC 91, by Duncan's multiple range test. These 3 lines were obtained as a result of selecting for the weight of fruits, whereas the remaining lines were selected for other characters. Some of these lines developed small fruits. Thus, breeding between monogerm diploid stocks for control of fruit weight was effective.

Environmental variability of the weight of monogerm diploid and tetraploid fruits

Effectiveness of breeding for the weight of monogerm fruits and germs depends upon the degree of variability of this character; it depends upon the effect of action of genes and the effect of ploidy levels, as well as on influence of the environmental factors that affect variability.

Influence of the environment on the variability of weight of monogerm fruits was studied in diploid and tetraploid male-sterile equivalents of the monogerm inbred SLC 91 during 3 years (1958, 1959, 1960) on 10 different isolated plots (Table 7,8).

Table 7.—Variability of the weight of fruits (in grams) in diploid and tetraploid sugar beet monogerm inbred SLC 91 during 3 years in 10 different locations.

Factors	Degree of freedom	Variance	F		
			Calculated	5% Level	1% Level
Total	179				
Reps.	2	33.28	8.88*	3.06	4.75
Populations (Ploidy Level)	1	3281.60	875.70*	3.91	6.81
Isolations	9	22.48	6.00*	1.94	2.53
Years	2	171.12	45.66*	3.06	4.75
Ploidy - Isolations	9	6.60	1.76	1.94	2.53
Ploidy - Years	2	6.70	1.78	3.06	4.75
Error	154	3.75			

* significant

Variability in different years. Analysis of variances showed that the value of calculated F exceeded by 1% the value of the tabulated F. In the 3 years the average weight of 1000 monogerm fruits ranged from 13.88 to 17.23 gms; in other words, difference between weight of fruits in separate years reached 24.14% and was significant (Table 8).

Variability in different locations. Fluctuations in the weight of fruits in different locations during the 3 years were also significant. The value of the experimental F equaled 6.00 and of tabulated F at 0.01 point, 2.53. The different isolations were

Table 8.—Range of variation in weight of fruits (1000) in the sugar beet monogerm male-sterile equivalent of SLC 91 in different years, locations, and at different ploidy levels.

Materials	During 3 years				In 10 locations				Average for years and locations	
	Minimum		Maximum		Minimum		Maximum			
	grams	%	grams	%	grams	%	grams	%	grams	%
Diploid MS 91	9.9067	100.00	12.5267	126.45	9.5111	100.00	12.4889	131.31	11.1689	100.00
Tetraploid MS 91	17.8467	100.00	21.9300	122.88	18.7111	100.00	22.8889	122.33	19.7044	176.42
For all strains	13.88	100.00	17.2300	124.14	14.09	100.00	17.6900	125.55	-----	-----

grown on soils of different types and the weight of fruits ranged from 14.09 to 17.69 gms (Tables 7,8).

Four of the 10 isolations were always overwinter plantings. The remaining 6 isolations were grown from stecklings planted in spring. The weight of fruits was 15.4 gms for both types of plantings.

The F-values for the interaction ploidy level and years, and for the interaction ploidy level and isolations was not significant.

Effect of colchicine-induced tetraploidy on the weight of monogerm fruits produced in different environments

Analysis of variance showed that variability in weight of fruits was caused by the differences between the two populations studied—male-sterile diploid SLC 91 and male-sterile tetraploid SLC 91—rather than by the environmental factors associated with years of production or location of plots (Table 7).

The value of variance between diploid and tetraploid populations was 3281, whereas the value of variance between years was 171 and between locations only 22.

The differences in weight were as follows: the weight of fruits in the diploid monogerm male-sterile equivalent of SLC 91 during 3 years in 10 different locations was 11.1689 gms and the weight of fruits in the tetraploid monogerm male-sterile equivalent of SLC 91 under the same conditions was 19.7044 gms, or an increase of 76.42% for the tetraploid (Table 8). Thus, colchicine-induced tetraploidy greatly increased the weight of monogerm fruits in the inbred line SLC 91.

To verify the universality of this appearance, a comparative study of the weight of fruits in different diploid and tetraploid self-fertile and self-sterile lines was made.

Weight of fruits in tetraploid monogerm self-sterile lines

Eighteen tetraploid self-sterile lines were planted in overwinter planting in 2 replications together with the diploid check and tetraploid male-sterile equivalent of monogerm inbred SLC 91. The 18 self-sterile lines were derived from individual tetraploid plants isolated in an open-pollinated monogerm population and propagated by brother-sister mating for 2 generations.

Weight of fruits was studied on 10 plants of each line in each replication. Analysis of variance of the weight of fruits shows that the main factor of variability is the difference in weight between diploid and tetraploid populations (Figure 1). The calculated value of F highly exceeds its tabulated value at the 1% point (Table 9).

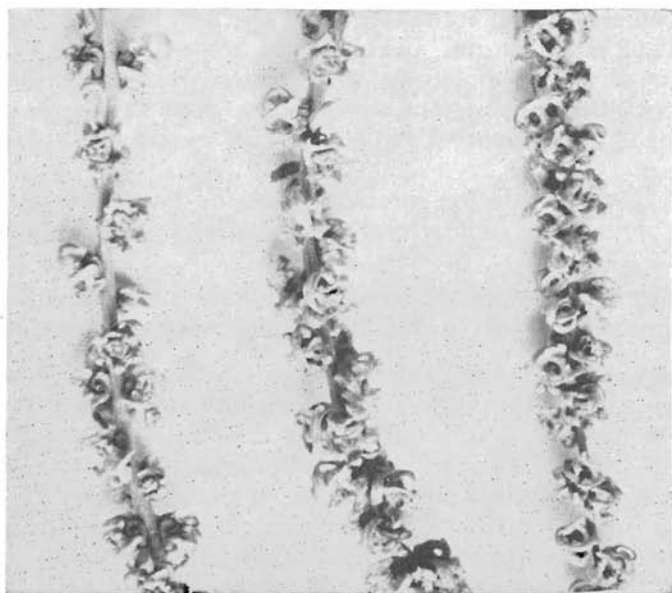


Figure 1a.—Monogerm fruits of a diploid self-sterile population. $\times 1.3$



Figure 1b.—Monogerm fruits of a tetraploid self-sterile population. $\times 1.3$

Table 9.—Weight in fruits (in grams) in 18 sugar beet tetraploid monogerm self-sterile lines and in diploid and tetraploid SLC 91. Analysis of variance.

Factors	Degree of freedom	Variance	F		
			Calculated	5% Level	1% Level
Total	399				
Between populations	19	530.91	18.96*	2.18	3.06
Between blocks	1	21.07	Not significant		
Experimental error (Interaction: blocks-population)	19	27.9950			
Grouping of population comparison					
Monogerm: 2n versus 4n	1	6488.33	231.77*	4.38	8.18
Among tetraploids	18	199.95	7.14*	2.21	3.12

* significant

Grouping of population comparison shows that the presence of a diploid population among the tetraploids is the main factor causing variability. The value of F is 231.77. Differences in the weight of fruits among separate tetraploid lines are also significant; the value of the calculated F is 7.14 whereas the value of the tabulated F is only 3.12.

For evaluation of differences in the weight of fruits among many populations, the principle of Duncan's new multiple range test was used. In 18 tetraploid self-sterile monogerm lines the weight of fruits fluctuated from 20 to 31 gms. The weight of fruits in the diploid male-sterile equivalent of SLC 91 was 8 gms, and in the tetraploid male-sterile equivalent of SLC 91 it was 18 gms.

In this experiment, the weight of fruits of all tetraploid lines significantly exceeded the weight of fruits in the diploid male-sterile equivalent of SLC 91. Among the tetraploids, only 2 lines with the largest fruits significantly exceeded in weight the tetraploid male-sterile equivalent of SLC 91.

Weight of fruits in the tetraploid monogerm self-fertile inbreds

Eighteen tetraploid inbred lines derived from the hybridization of tetraploid inbred SLC 91 monogerm with tetraploid multigerm population US 35/2, were studied for variability of the weight of fruits.

Analysis of variances for 18 monogerm tetraploid self-fertile inbreds and for the diploid and tetraploid male-sterile equivalent of SLC 91 (Table 10) indicated that in the inbreds, as in self-sterile lines, the main factor of variability is the difference in weight of fruits among different populations. The calculated value of F is 19.48 and the tabulated value of F is 2.18 or 3.06.

Grouping of population comparison also shows that the main

Table 10.—Weight of fruits (in grams) in 18 sugar beet tetraploid monogerm self-fertile inbred lines and in diploid and tetraploid SLC 91. Analysis of variance.

Factors	Degree of freedom	Variance	F		
			Calculated	5% Level	1% Level
Total	399				
Between populations	19	237.4123	19.4830*	2.18	3.06
Between blocks	1	1.4641	N o t s i g n i f i c a n t		
Experimental error (Interaction: blocks-population)	19	12.1856			
Grouping of population comparison					
Monogerm: 2n versus 4n	1	1787.1280	146.6590*	4.38	8.18
Among tetraploids	18	151.3170	12.4177*	2.21	3.12

* significant

factor of variability is the difference in weight of fruits between a diploid SLC 91 and tetraploid inbreds. The value of the mean squares between a diploid and tetraploids is 1787, whereas the value of the mean squares among tetraploid lines is 151. In spite of this, the calculated value of F among tetraploids is 12.42, while the tabulated value of F is 2.21 or 3.12.

The monogerm tetraploid inbred lines differed in the weight of fruits and these differences were statistically significant. In the individual lines, the weight of fruits fluctuated from 7 to 21 gms.

All tetraploid inbreds had larger fruits than the diploid inbred SLC 91. But, according to Duncan's new multiple range test, only 13 tetraploid inbreds significantly exceeded the diploid inbred SLC 91 in fruit weight. One tetraploid inbred line had significantly larger fruits than 7 other lines.

Thus, genetic variability in the weight of fruits was recorded for tetraploid inbreds. Tetraploidy not only increased the weight of fruits, but a new genetic variability of this character arose at this new level.

Variability of the weight of fruits, estimated for the diploid strains only, will be highly increased if diploid and tetraploid strains are studied together in one experiment, even if these strains are closely related.

Coefficient of variation for weight of fruits estimated in this experiment in diploid self-sterile monogerm populations was only 10%, while coefficient of variation for tetraploid self-sterile monogerm populations and 1 diploid line rose to 90.2%. The same was recorded for the inbreds. When only diploid inbreds were studied, the coefficient of variation was 28.74%. For tetraploid inbreds studied together with 1 diploid line, the coefficient of variation increased to 96.55%.

Weight of germs in diploid and tetraploid monogerm sugar beets

Weight of fruits and weight of germs are highly correlated in diploid monogerm beets (16). The present investigation was conducted to study the relationship of these correlations in related diploids and tetraploids. Increased weight of fruits in monogerm tetraploids may be important only if it is correlated with the increased weight of germs. To study the correlation between weight of fruits and germs, samples were taken from 29 monogerm diploid and 29 monogerm tetraploid strains (Table 11).

Table 11.—Correlation between weight of fruits and germs in monogerm diploid and tetraploid sugar beet strains.

Weight of 1000 fruits in grams	Weight of 1000 germs in grams							Number of strains	
		1.9	2.5	3.1	3.7	4.3	4.9	Diploid	Tetraploid
	to	to	to	to	to	to	to		
5-6	3							3	
7-10	5	2						7	
11-14		9	<i>1*</i>	<i>1</i>				9	2
15-18		1	1	<i>4</i>	<i>1</i>			2	5
19-22			4	<i>3</i> <i>7</i>	<i>1</i>	<i>1</i>		7	9
23-26				1	6	2		1	8
27-30						4	<i>1</i>		5
Diploid	8	12	5	4	—	—	—	29	—
Tetraploid	—	—	1	12	8	7	1	—	29

* The tetraploids are in blackface italic type.

The average weight of fruits of the diploid monogerm strains was 13.12 ± 0.78 gms, and in tetraploid monogerm strains, 21.43 ± 0.64 gms, or an increase of 63.3%. Difference in weight of fruits between diploid and tetraploid strains was significant.

Average weight of 1000 germs in the diploid monogerm strains was 2.169 ± 0.073 gms and in the tetraploid monogerm strains, 3.631 ± 0.078 gms, or an increase of 67.4%. This difference is significant. Thus, tetraploid monogerm beets developed larger fruits and larger germs than diploids (Fig. 2).

Weight of 1000 germs in percent of weight of fruits was 16.53% in diploids and 16.94% in tetraploids; therefore, in monogerm tetraploid strains with different weight of fruits the increase in germ weight is proportional to the increase in fruit weight.

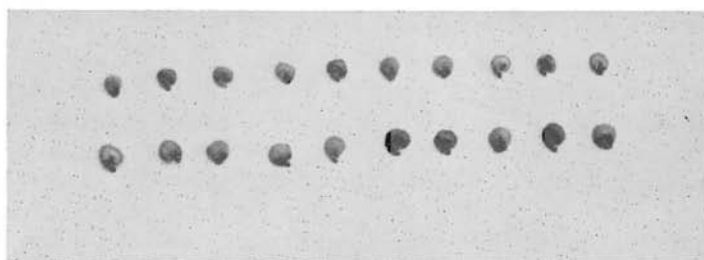


Figure 2.—Germs excised from: a) a monogerm diploid strain (upper row), b) a monogerm tetraploid strain (lower row) $\times 1.5$.

Tetraploids exhibit the same correlation between weight of fruits and weight of germs as the diploids. For 29 diploid strains studied, this correlation equaled 0.9500; and for 29 tetraploid strains, 0.9244. The t -value for the diploids was 31.03, and for tetraploids, 23.79. Value of t for diploids and for tetraploids is significant at a probability of above the 1% level; therefore, both correlations are significant.

Hence, selection for the larger fruits in monogerm diploids and in tetraploids automatically increases the weight of germs. We have not observed monogerm diploid or tetraploid strains with large fruits containing small germs nor strains with small fruits and large germs.

Fruits and germs in monogerm beets containing twin germs

Some monogerm fruits produce double, triple, or multiple seedlings. Presence of two or more germs in a locule complicates the relation between weight of fruits and germs in monogerm beets, because the increase in the number of germs generally is not followed by a corresponding enlargement of a fruit. The total weight of 2 or 3 germs developed in 1 fruit almost corresponds to the weight of a single germ in a fruit of a given size. The weight of the individual germs is naturally much reduced.

The appearance of twins is caused by an abnormal flower development. H. Savitsky observed different deviations from the normal embryological development of flowers and ovules in multigerm beets which lead to formation of twins. These abnormalities consisted of 1) development of 2 ovaries in the same flower with 1 ovule in each (Figure 3); 2) development of 2 ovules in the same cavity of the ovary (Figure 4)—sometimes in the flower cluster 2 ovaries contained double ovules; and 3) development in 1 ovule of 2 nucelli surrounded by the internal integuments, each nucellus containing a normally developed

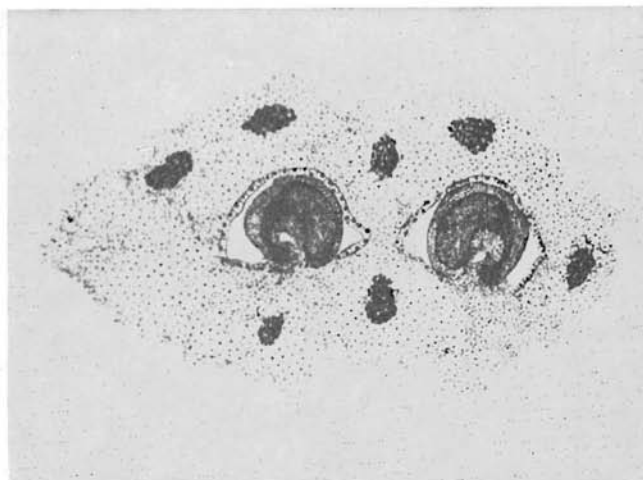


Figure 3.—Two ovaries in 1 flower, each containing 1 ovule, $\times 58$.



Figure 4.—Two ovules in 1 cavity of the ovary, $\times 58$.

embryo sac (Figure 5). Each of these types of deviations produces non-identical twins.

The identical twins are formed as a result of a splitting of a porembryo at an early stage of development, or after a cleavage of a zygote and formation of a group of embryogenic cells from which several embryos develop. Other manifestations of polyembryony—development of adventive embryos from other nuclei of the same embryo sac (mostly from cynergids), or from the cells outside the embryo sac (from nucellus, or integument) cannot result in formation of identical twins because such embryos

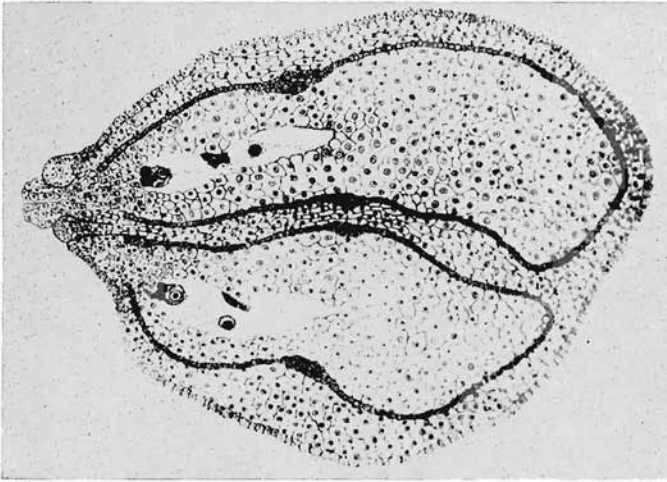


Figure 5.—Ovule with 2 nucelli, each containing an embryo sac, $\times 125$.

will be genetically unlike the embryo developed from the fertilized egg cell. Only if a twin pair will arise from somatic cells outside the embryo sac (nucellus, integument) in a result of partogenesis, such twins will be identical. Polyembryony resulting from splitting of an embryo, or from a cleavage of a fertilized egg cell is a rare appearance in angiosperms. A development of a twin pair from somatic cells is also extremely rare. An intensive embryological study of sugar beets indicates that occurrence of identical twins in sugar beets should be extremely rare.

In his intensive study of twins in beets, Fisher (3), using the morphological analysis, found that twin seedlings grew from 2 ovules of the same flower as well as from a single ovule. Triple seedlings were also observed. Some twin pairs grew from the ovules which contained 2 separate perisperms, and some of the ovules with a common perisperm. The latter twin pairs may be represented by the identical twins. However, the majority of twin pairs, even in this group, did not appear to be identical, although the morphological resemblance of some twin plants lead the author to the conclusion that identical twins are also produced in *Beta*.

In monogerm beets the appearance of twin seedlings is caused mainly by the development of 2 ovules in the ovary of a flower. Some twins grew from the same ovule. In the majority of monogerm strains the percent of twins is not higher than in the multi-germ strains. In self-sterile monogerm populations twin seedlings

are rare. But in some monogerm inbred lines, up to 30% of fruits produce twin seedlings. Variation in percent of twins produced in the individual inbred lines indicates that formation of twins is hereditarily controlled. In fruits containing many small germs, the germs are of a low and variable weight. It is important that the increase in weight of the monogerm fruits in tetraploid strains is not followed by an increase in the number of fruits that produce twin or triple germs. In all breeding materials investigated, tetraploid beets developed fewer twin germs than the diploid beets. It is self-evident that the problem of twins is much more important for the monogerm than for the multigerm beets. The genetic nature of their formation and the grade of influence of the environment on their production are not known at present. The supposition can be made, however, that in addition to genetic effects, environmental factors greatly influence the development of surplus ovules and twin seedlings in monogerm beets.

Discussion and Conclusion

In multigerm sugar beets, the number of flowers in clusters is controlled by the genes which are members of one multiple allele *M-m* (partially also by modifying genes); therefore, segregation for multigerm and monogerm plants is evident in the F_2 and b_1 generations (15,17,19).

It was established that the embryological development of the inflorescence is different in multigerm and monogerm beets. In the monogerm beets only one flower develops on the peduncle. Subsequent flowers which develop in multigerm beets on the same peduncle are absent in the monogerm beets. Their absence is not caused by the arrested development, or degeneration of the rudimental flowers, but because they are not formed in the ontogenetic development of the homozygous monogerm plants (10). This explains why the monogerm character is not subject to a wide range of variability under different genetic or environmental conditions.

Segregation for weight of fruits or germs is more complicated than segregation for the number of flowers in a cluster (monogermity or multigermity). The limits between large and small fruits are easily changeable under different environment.

The availability of monogerm self-sterile and self-fertile populations made it possible to obtain information concerning environmental (years, soils, locations) and genetic variability of the weight of fruits and germs and also permitted a study of the variability of this character caused by polyploidy. An adequate study of the variability of the weight of fruits and germs

is impossible in the multigerm races of *B. vulgaris* L., because of the bias action of genes determining the number of flowers in the clusters.

Environment is an important factor which causes variability of the weight of monogerm fruits. Variability of weight of 1000 monogerm fruits was proved by cultivation of a diploid and a tetraploid monogerm inbred line during 3 years in 10 different locations. At the same time, interaction between level of ploidy and years, and between level of ploidy and locations was statistically insignificant.

Mutation which led to the development of a single flower on a peduncle (monogerm character) was not accompanied by increase in fruit size; therefore, improvement of seed size must be accomplished by selection.

In spite of their different origins, the monogerm diploid populations obtained from repeated backcrosses of monogerm beets to the open-pollinated multigerm varieties, did not differ from each other in the weight of monogerm fruits. But if inbreeding or selection for the weight of fruits is started within such monogerm population, the monogerm lines with different weight of fruits and germs may be isolated. Inbreeding or selection fixes the genetic variability in the weight of fruits derived from heterogeneous multigerm populations. Stored heterogeneity for weight in fruit may be transmitted to the monogerm populations from the multigerm populations used in backcrosses. Every one of these populations carried diverse genes determining the weight of fruits, but these genes were not expressed in the multigerm fruits and, therefore, the multigerm populations did not differ greatly among themselves in the weight of fruits.

Open-pollinated populations of sugar beet are heterogeneous in the multiple allele *M-m* (17). A multiple allelic system of a locus may give rise to genotypes of different viability in the population. If these genes remain in equilibrium in the population, the population will exhibit a balanced polymorphism in the number of flowers per cluster. A stable equilibrium is reached when the viability of homozygotes is less than the mean viability of a population (a kind of generalized heterosis). Under such conditions no heterozygote will be less viable than either of its associated homozygotes (9).

The new monogerm panmictic populations did not receive a ready system of variability of the weight of fruits based on the gene pool obtained as a result of previous selection for the breeding for the weight of monogerm fruits is based on the weight of fruits in the multigerm populations. The temporary

breeding for the weight of monogerm fruit is based on the genes that were present in the multigerm populations. These genes could not be controlled by the natural or artificial selection in the interaction with the gene *m* in the homozygote. Thus, the stored genetic variability of multigerm varieties furnishes the genes which produce variations in weight of fruits in monogerm beets. Variability in the weight of fruits may occur in different, and sometimes undesirable, directions. To obtain the optimal weight of fruits and germs, selection of monogerm lines with desirable expression of this character is necessary, and such selection was successful in our experiments.

Heritability in the direction of increased weight of fruits and germs appears to be low in different monogerm open-pollinated populations; but because of heterogeneity of populations in the genes controlling the weight of fruits and the absence of bias caused by the variation of the different number of flowers in the clusters, selection for increased weight of monogerm fruits is effective. Such selection results in a rapid change of the frequency of genes controlling the weight of fruits and germs in the monogerm populations. A former type of variability of weight of fruits and germs, which was created in the process of evolution of the multigerm populations on the basis of 2 hereditarily different characters (size and number of individual fruits within a seedball), is completely replaced in the monogerm populations by the variability of genes controlling the weight of fruits. This determines the selective advantage of monogerm beets for the improvement of the weight of fruits.

Occurrence of twins is a factor which increases the variability of the weight of germs, without changing the variability of the weight of fruits. Twin germs reduce the weight of individual germs within a fruit. Twin seedlings appear in multigerm, as well as in monogerm sugar beets. Their appearance is caused by the abnormal development of flowers and ovules. In monogerm sugar beets twin seedlings grow mainly from 2 ovules which developed in the same cavity of the ovary. As indicated by embryological study appearance of identical twins should be extremely rare in beets. The tetraploid strains develop fewer twins than their diploid ancestors.

The effectiveness of breeding for larger fruits in the monogerm populations may be increased if the tetraploid monogerm strains or the hybrids between tetraploid monogerm and tetraploid multigerm beets are used. Doubling of chromosomes increases the weight of fruits and also the weight of germs about 60 to 70%.

Presence of tetraploid monogerm strains raises the comparative genetic variability of monogerm diploid beets in the weight of fruits and germs. This phenomenon was observed in self-sterile open-pollinated populations and in self-fertile inbred lines.

Tetraploidy results in an increase in weight of fruits in populations, regardless of their origin. A new genetic variability arises within autotetraploid populations. Therefore, tetraploid lines with different weight of fruits were selected from the same population.

Of the 3 factors—environment (years, locations), genetics, and ploidy level—which determine the degree of variability in weight of fruits in monogerm beets, ploidy level appeared to be the most significant and universal. Increase in weight of fruits in tetraploid beets may be caused by an increase in the volume of nuclei and cells, a phenomenon manifested in all tetraploid crops. Universality of the increase in the weight of fruits and germs in tetraploid sugar beets may be caused by this reason.

However, a new genetic variability of weight of fruits and germs in monogerm tetraploid sugar beets is caused as directly by chromosome doubling, as also by the shift from diploid to tetraploid heredity (new mode of gene action, changes in equilibrium of genes in populations, in segregation of hybrids, in linkage and inbreeding effect).

Self-fertile and self-sterile tetraploid monogerm strains permit breeding for the weight of fruits on a higher level than is possible for the diploid monogerm beets. Diploid monogerm populations did not provide as wide a range of genetic variability as tetraploid monogerm populations in breeding for the weight of fruits.

Literature Cited

- (1) BARTL, K., P. CURTH, H. E. FISHER, and H. S. SCHNEIDER. 1957. Untersuchungen über die Keimfähigkeit von Polyploidem Zuckerrüben-saatgut. Zucker 7: 142-147.
- (2) DOXTATOR, C. W. and R. H. HELMERICK. 1962. Selection for seed size in monogerm varieties. J. Am. Soc. Sugar Beet Technol. 2 (3): 268-272.
- (3) FISHER, H. E. 1956. Untersuchungen an Zwillingen von *Beta vulgaris* L. Züchter 26: 136-152.
- (4) FURSTE, W. 1958. Massnahmen zur Erhöhung des Anteiles triploider Pflanzen in polyploiden Zuckerrüben. Deutsche Akademie der Landwirtschaftswissenschaften zu Berlin. Wissenschaftliche Abhandlungen 34: 28-36.
- (5) Knapp, E. 1958. Beta-Rüben bes. Zuckerrüben. In Roemer, T. and W. Rudolf. Handbuch der Pflanzenzüchtung. 2d ed. 3: 197-284.

- (6) IAZAR, O. 1961. Metabolisme respiratoire de la betterave sucriere, IX. Activite de la polyphenoloxydase et des decarboxylases au cours du reveil des plantules diploides et tetraploides. La Sucrierie Belge. 80(10) : 409-419.
- (7) IAZAR, O. 1962. Germination des glomerules de la betterave sucriere en fonction du lavage, du faconnage et des activitiés enzymatiques I. La Sucrierie Belge 81(5) : 175-196.
- (8) LERNER, I. M. 1954. Genetic homeostasis. Wiley, New York. 134 pp.
- (9) MANDEL, S. P. H. 1959. The stability of a multiple allelic system. Heredity 13: 289-302.
- (10) SAVITSKY, H. 1950. Embryology of mono- and multigerm fruits in the genus *Beta* L. Proc. Am. Soc. Sugar Beet Technol. 6: 160-164.
- (11) SAVITSKY, H. 1952. Selective fertilization studies and recovery of self-sterile from self-fertile races of monogerm sugar beets. Proc. Am. Soc. Sugar Beet Technol. 7: 339-343.
- (12) SAVITSKY, H. 1954. Self-sterility and self-fertility in monogerm beets. Proc. Am. Soc. Sugar Beet Technol. 8(2) : 29-33.
- (13) SAVITSKY, H. 1954. Obtaining tetraploid monogerm self fertile, self sterile, and male sterile beets. Proc. Am. Soc. Sugar Beet Technol. 8(2) : 50-58.
- (14) SAVITSKY, V. F. 1930. A study of the variability of characters of *Beta vulgaris* L. in the second year of life. Bull. Belaya Tserkov Plant-Breed. Sta. Bull. 5: 69 pp.
- (15) SAVITSKY, V. F. 1952. Methods and results of breeding work with monogerm beets. Proc. Am. Soc. Sugar Beet Technol. 7: 344-350.
- (16) SAVITSKY, V. F. 1954. Relation between the weight of fruit and weight of germ in mono- and multigerm beets. Proc. Am. Soc. Sugar Beet Technol. 8(2) : 16-22.
- (17) SAVITSKY, V. F. 1954. Inheritance of the number of flowers in flower clusters of *Beta vulgaris* L. Proc. Am. Soc. Sugar Beet Technol. 8(2) : 3-15.
- (18) SAVITSKY, V. F., G. K. RYSER, G. E. RUSH, and C. P. PARRISH. 1954. Interrelation between weight of seed and fruit and utilitarian characters in inbred lines and hybrids of monogerm sugar beets. Proc. Am. Soc. Sugar Beet Technol. 8(2) : 399-403.
- (19) SAVITSKY, V. F. 1958. Genetische Studien und Züchtungsmethoden bei monogermen Rüben. Zeitschr. für Pflanzenzüchtung. 40: 1-36.
- (20) SAVITSKY, V. F. 1962. Sucrose and weight of root in tetraploid monogerm and multigerm sugar beet populations under different mating systems. J. Am. Soc. Sugar Beet Technol. 11(8) : 676-711.
- (21) SCHNEIDER, H. 1960. Untersuchungen über die Beziehungen zwischen Knäuelgrösse, Reifegrad, Keimfähigkeit und Ploidiestufe bei Zuckerrübensaatgut. Deutsche Akademie der Landwirtschaftswissenschaften zu Berlin. Wissenschaftliche Abhandlungen. 48: 39-46.
- (22) SEDLEMAYR, T. 1961. Untersuchungen über den Einfluss verschiedener Faktoren auf die Ploidiestufenanteile einer anisoploiden Zuckerrübensorte. Züchter. 31(7) : 310-317.

- (23) STEWART, D. 1961. Development of monogerm varieties of sugar beets in North America. Report of first joint meeting Am. Soc. of Sugar Beet Technol. with Inst. Int. de Recherches Betteravieres. London. pp. 119-122.
- (24) STAUDE, H. 1959. Toleranzen bei Stichprobenprüfungen von Ploidiegradbedingungen im Handel mit anisoploidem Zuckerrübensaatzgut. Deutsche Akademie der Landwirtschaftswissenschaften zu Berlin. Wissenschaftliche Abhandlungen. 41: 29-50.
- (25) TOLMAN, B. 1961. Genetical monogerm seed and its practical application. Report of first joint meeting Am. Soc. of Sugar Beet Technol. with Inst. Int. de Recherches Betteravieres. London. pp. 115-119.
- (26) TOLMAN, B. and R. JOHNSON. 1956. Materials and methods used in producing commercial male-sterile hybrids. Proc. Am. Soc. Sugar Beet Technol. 9 (2): 153-160.
- (27) ZOSSIMOVITSCH, V. 1935. The weight and quality of sugar beet germs in different flowers of a seed ball. Scientific Notes for Sugar Industry. 5 (6): 128-131.
- (28) ZOSSIMOVITSCH, V. 1936. Variation in weight of seed from individual flowers of a sugar beet ball. Selektiv. i Semenovodstvo. 5: 90-92.
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