# **Broadening the Genetic Base of Sugarbeet**

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## ABSTRACT

The narrow base from which sugarbeet originated, the need for disease resistance and the negative relationship between root yield and sugar accumulation have all contributed to make the current gene pool from which most present-day sugarbeets originate narrow. Of the wild germplasm available, Beta vulgaris subspecies maritima offers the greatest promise of broadening the genetic base for future sugarbeet improvement. Crosses between B. maritima and sugarbeet male sterile inbreds have been advanced through four successive cycles of mass selection for root shape. Two of these crosses are approaching sugarbeet in root shape, root yield and sucrose concentration; however, they are still below commercial sugarbeet hybrids in root yield and sugar concentration. Even though these populations are inferior to commercial sugarbeet hybrids, it is the author's belief that superior combining germplasm exists in some of this material and that combining these with commercial germplasm will produce superior hybrids. Additional populations (crosses between sugarbeet and regional populations of B. maritima) are in the developmental stage. Sugarbeet inbreds segregating for mendelian male sterility were used in the initial crosses to insure crossing and recombination in each selection cycle.

The current gene pool from which most present-day sugarbeet hybrids originated is considered by many to be narrow. N. O. Bosemark stated that "... the genetic base of sugarbeet is probably narrower than that of most cross-pollinated crops" (Bosemark, 1979). This narrow genetic background is largely due to: 1) the narrow base from which sugarbeet originated, 2) the need for disease resistance, and 3) the negative relationship between root yield and sugar accumulation.

Sugarbeet, one of our youngest major crop plants, originated about 200 years ago when a German chemist, A. S. Marggraf, first discovered sugar (sucrose) in beets (Marggraf, 1747). However, it wasn't until the beginning of the 19th century that sugar was processed from beets on a commercial scale. This was largely made possible by the efforts of Franz Karl Achard, a student of Marggraf's, who found that white, cone-shaped roots deeply set in the soil were the highest in sugar content. The genetic types he identified and selected, which were a relatively limited range of fodder beet types, became the genetic basis of future sugarbeet breeding. The white Silesian beet developed by Achard and Koppy is reputed to be the "mother stock of all sugar beets in the world" (Lippmann and Iversen, 1925).

The first major breeding thrust in the U. S. was initiated in the 1920's utilizing European open-pollinated commercial varieties. The incorporation of additional European germplasm has been limited due in part to the proprietary nature of this germplasm. With the joining together of U. S. and European seed companies, more divergent germplasm has been made available. Occasional infusion of exotic germplasm has occurred for specific characters such as disease resistance. However, most breeders agree that the basic sugarbeet breeding germplasm is rather narrow.

Resistance to the major sugarbeet diseases (curly top, Cercospora leaf spot, Rhizoctonia root rot, beet western yellows, and Rhizomania) is complex and multi-genic in inheritance. In many cases only a few sources of resistance have been utilized commercially for many diseases. Developing disease-resistant germplasm requires a major breeding effort and has the effect of further narrowing the potential germplasm base in those areas where disease resistance is essential.

Efforts to increase root yield generally have resulted in reduced sucrose concentration and vice versa (Bergen, 1967; Oldemeyer, 1975; Powers, 1957; Rosielle and Hamblin, 1981). Dahlberg stated, "it has been extremely difficult to develop sugar beet varieties of high sugar content without losing the ability to produce good tonnage in the process" (Dahlberg, 1952). Because of this negative relation, early breeders developed varieties classified as Z types (high sugar) or E

types (high root yield). Doney et al. (1981) and Milford (1973) found a significant negative correlation between cell diameter and sucrose concentration. Further genetic studies identified the gene effects for cell size to be additive (Doney and Theurer, 1983). It was hypothesized that the additive genes affecting cell size may be the same genes responsible for the additive genetic variance of sucrose concentration. This would explain the negative correlation between sucrose concentration and root yield, i.e., large cells result in large roots and low sucrose concentration and vice versa. The importance placed recently on high sugar content beets has caused breeders to emphasize the concept of increasing sugar concentration while maintaining root yield. This negative relationship has, therefore, tended to decrease potential genetic variation.

The past half century of hybrid breeding in major crop plants suggests that broad genetic differences between parents tend to give the greatest heterosis (Frankel, 1983). Inheritance studies of cell-division rate (Doney and Theurer, 1985) concluded that cell-division rate is conditioned largely by non-additive type gene action and that heterosis for root size is due primarily to increases in cell-division rate rather than increases in cell size. They concluded that breeding and selection methods that capitalize on non-additive genetic variation (hybrid breeding), will, therefore, increase sugarbeet root yield by increasing cell-division rate and not affect cell size or sucrose concentration. This is an extremely important concept, since it explains a genetic mechanism for overcoming the frustration of breeding for root yield and sucrose concentration simultaneously. One of the first studies of hybrid sugarbeet development observed this phenomenon. Doxtator and Skuderna (1942) found that hybrids gave significantly higher sugar yields per acre than their parents and that these increases were due to increases in root size and not due to changes in sucrose concentration. Progress in developing superior hybrids is, therefore, dependent on the availability of genetic variation for cell-division rate, i.e., non-additive genetic variation for root yield. Since sugarbeet is known to possess a narrower genetic base than most cross-pollinated crops, future increases in heterosis may be small. It becomes imperative that significant efforts be made to broaden the base.

Of the wild germplasm available, *Beta vulgaris* subspecies *maritima* offers the greatest promise of broadening the genetic base for future sugarbeet improvement. In its native habitat, *B. maritima* exists over a wide range of environmental conditions (from Sweden in the north, throughout the Mediterranean, and east as far as India) (Zossimovitch, 1940). Its adaptation and survival to this wide range of hársh environments has accumulated many stress resistance and growth genes. Observations made by the author over the past 10 years suggest an

abundance of genetic variation (Doney et. al., 1990; Doney, 1991). The accumulation of these genes over the centuries has developed a gene pool different from our cultivated sugarbeet. Collection expeditions conducted the past 15 years by the International Board of Plant Genetic Resources (IBPGR), United States Department of Agriculture (USDA), etc., have been effective in preserving much of the native variation presently existing within the subspecies. Another desirable feature of this subspecies is that it produces fertile offspring when crossed with sugarbeet germplasm.

Breeding efforts by Italian sugarbeet breeder O. Munerati in the early part of this century succeeded in incorporating Cercospora leaf spot resistance from wild *B. maritima* into cultivated sugarbeet (Biancardi and Biaggi, 1979). The genes responsible for leaf spot resistance in today's leaf spot germplasm are believed by many to have originated from his efforts (Lewellen, 1992).

In the late 1930's, Dahlberg (Great Western Sugar Co.) began crossing experiments between sugarbeet germplasm and wild *B. maritima* types from the North Atlantic because, "this type looked more promising than any of the others" (Dahlberg, 1938). He stated that "considerable optimism is required to persevere in a breeding program which includes these lines . . . . the first generation hybrids are nearly always disappointing in both size and type of roots, these being small, scrawny and very much sprangled". His efforts continued through several cycles of back crossing to sugarbeet germplasm. In 1940, he reported that lines coming out of this program were higher in yield but lower in sugar than commercial cultivars (Dahlberg, 1940).

At the first World *Beta* Network meeting, N. O. Bosemark reported on research conducted several years ago utilizing wild *B. maritima* germplasm (Bosemark, 1989). He crossed wild *B. maritima* with a white-skinned fodder beet cultivar. After two cycles of mass selection for bolting resistance and root shape and three cycles of recurrent half-sib family selection, root yields and sucrose concentration were approaching acceptable levels for introduction into elite sugarbeet breeding pools. He suggests that the wild gene *Beta* pool may not be as inaccessible as might be thought.

This paper reports efforts to broaden the sugarbeet genetic base utilizing wild *Beta* germplasm.

## MATERIALS AND METHODS

#### Single Crosses

In 1986, 10 separate *B. maritima* accessions were each crossed to two sugarbeet male sterile inbreds ('C3747' and 'L53'). 'C3747'

was segregating for the recessive mendelian male sterile gene. F<sub>2</sub> plants were interpollinated within each cross to allow for recombination before proceeding with selection. For each selection cycle, roots selected for cone-type shape and reduced sprangling were interpollinated in separate open-pollination isolation chambers. Six of the original crosses survived four successive cycles of mass selection for root shape. Selection for soluble solids was included in one cycle. The resulting populations were tested in a replicated field trial in 1991. The most promising populations, based on the 1991 field data, were reevaluated in replicated field trials in 1992.

## Population Development

Accessions that gave a broad representation of the germplasm for the *B. vulgaris* subspecies *macrocarpa*, *atriplicifolia* and *patula*, and three different geographic populations of *maritima* (Denmark, Belgium and Ireland) were selected from the NC-7 *Beta* collection. These were crossed to a sugarbeet line ('C3747') segregating for mendelian male sterility. In the initial crosses all plants within the 'C3747' line that produced pollen were discarded prior to crossing. Two cycles of random intercrossing were conducted prior to selection in order to achieve recombination between sugarbeet and wild germplasm. In each cycle, seed was harvested only from male sterile plants. This technique maintains the male sterile gene in the population and insures maximum intercrossing and recombination.

## Field Trials

Field trials were conducted on the North Dakota State University (NDSU) Prosper experiment station (10 miles west of Fargo, North Dakota). Each field trial was replicated six times in a randomized block design. Plots were two rows, 25 ft (7.6 m) long, and 22 in (55 cm) between rows. Plants were thinned to a 9-in (23 cm) spacing at the four leaf stage. At harvest, all beets from each plot were machine harvested and weighed for root yield. Duplicate samples of 10 beets were selected at random from each plot and placed in rubberized bags for sugar analysis. Sucrose concentration was determined polarimetrically on beet pulp by the cold digestion method (McGinnis, 1971).

## RESULTS

The 1991 field trial data for the six populations resulting from crosses between sugarbeet and wild beet followed by four cycles of mass selection for root shape are given in Table 1. Two commercial hybrids commonly grown in the Fargo, North Dakota area were included in the

**Table 1.** Root and sugar yield; sucrose, sodium, potassium, and amino nitrogen concentrations; root number and percent tare for six populations resulting from crossing wild times sugarbeet germplasm followed by four cycles of mass selection for root shape, and for two commercial sugarbeet hybrids (mean). 1991 field trial at the NDSU Prosper experiment station.

Entry	Description	Root Yield (t ha-1)	Sugar Yield (t ha-1)	Sucrose (g kg <sup>-1</sup> )	Na (ppm)	K (ppm)	AmN (ppm)	Root No.	Tare
x111	B. maritima* x 1.53	25.5	3.52	139	346	1843	1053	66	9.0
x112	WB 284 x L53	23.3	3.32	142	339	1986	1115	56	8.9
x113	WB 245 x L53	23.1	3.35	144	480	1848	942	64	8.7
x114	WB 31 x C3747	19.5	2.68	137	277	1870	1208	54	10.6
x115	WB 252 x L53	19.9	3.09	153	365	1600	822	46	6.8
x116	WB 172 x L53	22.6	3.14	138	345	1803	1106	62	8.7
Check (hybrids)		32.2	5.09	161	426	1818	873	80	7.3
LSD $p = 0.05$		6.9 •	0.81	7	88	111	41	16	4.4

Random mixture of F, plants from all 10 original crosses followed by four cycles of mass selection for root shape.

test. The means of the two hybrids are given in Table 1 for comparison purposes. Root maggot pressure was unusually high in the 1991 field trial even though the plot was treated with Lorsban<sup>†</sup> at planting. Stands appeared to be satisfactory at thinning; however, stand counts were not taken. At harvest only 46 to 80 plants per 100 ft of row were harvested (Table 1).

Root and sucrose yields were very low due to the poor stands; therefore, not much reliance can be placed on these yield data. The sucrose percentages were all lower than the commercial hybrids; however, the 'x115' population approached the hybrids in sucrose concentration. The selection populations were not different from the commercial checks for the quality factors, Na and K, but were generally higher for amino nitrogen. The 'x115' population was lower for all three impurities. This population, although very low in yield, showed promise in quality factors. The 'x115' and 'x116' populations were much smoother and more sugarbeet-like in root shape than the other populations as indicated by the lower percent tare (Table 1). In other selection trials it appeared that populations 'x111', 'x112', 'x113' and 'x114' were still very sprangled and did not exhibit significant genetic variation for smoother, less sprangled roots. Based on these data and the selection observations, populations 'x112', 'x113' and 'x114' were discarded.

The 'x111', 'x115' and 'x116' populations along with the same commercial checks were again tested in a replicated field trial in 1992 (Table 2). Plot stands were at the desired level at thinning time; however, a significant number of roots in the selection populations died prior to harvest. Root and sugar yields and sucrose concentrations were significantly lower than the commercial hybrids. The 'x111' population was significantly lower in sucrose concentration and higher in the other quality factors than the other two new populations. The 'x115' and 'x116' populations were higher in sodium and amino nitrogen than the hybrid checks.

These new selection populations still exhibit significant genetic variation and might be expected to increase in quality and yield with continued selection. All the selection pressure to this point was from mass selection and, therefore, was due to additive gene action. Since root yield is conditioned by both additive and non-additive gene action, it was determined that future selection should be for non-additive gene action.

In 1992, 66 half-sib families of the 'x115' population and 31 half-sib families of the 'x116' population were crossed to the sugarbeet inbred

<sup>\*</sup> Mention of a specific product is for reference purposes and does not constitute preferential endorsement.

**Table 2.** Root and sugar yield; sucrose, sodium, potassium, and amino nitrogen concentrations; root number and percent tare for the x111, x115 and x116 populations and for two commercial sugarbeet hybrids (mean). 1992 field trial at the NDSU Prosper experiment station.

Entry	Description	Root Yield (t ha-1)	Sugar Yield (t h-1)	Sucrose (g kg <sup>-1</sup> )	Na	K (ppm)	AmN (ppm)	Root No.		Tare
					(ppm)			Thin	Har	0%
x111	B. mar. x L53	39.2	3.93	100	1449	1951	1118	144	100	1.7
x115	WB 252 x L53	38.3	4.69	123	1073	1833	1033	136	84	1.5
x116	WB 172 x L53	38.3	4.76	124	1022	1806	1080	130	1.6	
Check (hybrids)		47.0	6.62	141	842	1896	861	149	117	1.1
LSD $p = 0.05$ 6.7		0.81	6	165	149	96	18	18	1.1	

'L33'. These half-sib families will be evaluated for combining ability in replicated field trials in 1993 and 1994.

# **Population Development**

Crosses with the subspecies *macrocarpa*, *atriplicifolia* and *patula* and three regional populations of *maritima* to sugarbeet are in the development stage. All have advanced through two cycles of random intercrossing to allow for recombination to take place between the wild and sugarbeet germplasm.

Two characteristics of wild *Beta* germplasm that have discouraged its utilization in sugarbeet breeding are: 1) slow germination and 2) slow leaf initiation. If selection is practiced without attention to slow germination and slow leaf initiation, advanced populations may continue to carry these two undesirable characteristics.

These characteristics have been found to be highly heritable. Germination and leaf initiation have been significantly improved by employing growth chamber selection methods. Seedling selection for early germination and leaf initiation will be conducted in these populations for one to two cycles prior to field selection studies.

Our efforts thus far have not produced germplasm that shows superiority to our present sugarbeet hybrids. However, progress in incorporating wild germplasm into desirable sugarbeet germplasm utilizing subspecies of the *Beta* section has been successful and appears to be less difficult than might be expected. Those populations approaching near-sugarbeet type roots have maintained significant phenotypic variation, i.e., are still segregating for many foliage characteristics. It is the author's belief that superior combining germplasm exists in some of these populations, that proper evaluation such as testcross and combining ability analysis should be able to identify new growth genes arising from wild ancestors, and that combining these with commercial germplasm will produce superior hybrids.

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