Testing Hybrid Blends to Determine General Combining Ability of Parental Lines¹

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The efficiency with which superior genotypes may be sorted from a population generally dictates the amount of germplasm which may be examined and is related, ultimately, to the success of a breeding program. More efficient sorting methods are always being sought. This report describes a technique for identifying superior inbred lines of sugarbeets and presents the results of an experiment to test the method.

Inbred lines in the Great Western sugarbeet (*Beta vulgaris* L.) breeding program are first screened for general combining ability in tests of top-cross hybrids (3).³ These tests are considered adequate for preliminary screening; more critical testing is required for positive identification of superior strains.

For more precise testing, inbreds that perform well in topcross tests are intercrossed so that each inbred is involved in several single cross combinations. The resulting single cross hybrids are tested primarily to evaluate general combining ability of parental lines with less regard for specific combining ability. The mean performance of several hybrids having one parent in common is a reliable estimate of general combining ability of the inbred particularly when adjustments are made for combining ability of the other parents involved (2).

To obtain a realiable estimate of the general combining ability of an inbred line, evidence would indicate that the line should be tested in a minimum of six hybrid combinations and preferably 10 or more. Frequently, single cross seed quantities of individual hybrids are too small for inclusion in a test. Therefore, the number of testable hybrids involving a line is often too few to provide adequate information. Cost and labor limitations may restrict the number of

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³Numbers in parentheses refer to literature cited.

lines tested where adequate facilities exist. Only limited testing can be accomplished in outlying areas where personnel and facilities are limited.

An alternative method to testing several single cross hybrids to determine general combining ability of an inbred line would be to test as a single entity a composite or blend of seed of the same hybrids. The blend of seed would contain an equal proportion of sprouts of each constituent hybrid based on germination. To obtain reasonably accurate data, the trial plots should be at least 15 square meters planted in as many replications as are used in commercial variety trials.

A test was designed to gather evidence for using the results of trials of a blend of seed as a reliable measure of general combining ability.

Materials and Methods

Inbred lines that have been extensively tested and differ considerably in combining ability were selected as test parents. Remnant seed of many of the single cross hybrids involving these lines was available for this test. Each production year involved different sets of crosses with the tested lines. The four selected inbred lines are known to combine with other lines to produce hybrids having the following characteristics:

	Inbred			
	Α	В	С	D
Root Weight	High	Very High	Low	Low
Sugar	High	Low	Medium	Very High
Thin Juice Purity	High	Low	Medium	High
Recoverable Sugar/Acre	High	Very High	Low	Low

For each inbred a "general blend" was made up of seed from all crosses available involving that inbred. The numbers of crosses making up the general blends follows:

Blend No.	Inbred	No. Crosses
73HB30	А	66
73HB39	в	44
73HB52	С	26
73HB46	D	49

In addition to the general blends, hybrids within each seed production year were blended to determine the effect of different sets of tester parents on the performance of the blends. The number of hybrids making up the additional blends varied from five to twenty so performance of blends with varying numbers of constituent hybrids could be compared. The blends were tested in 1973 on the Great Western Research Center farm at Longmont, Colorado. The experimental design was a 5×5 triple lattice with plots, as harvested, consisting of four rows, each 18 feet in length, replicated six times. Root weight determinations and chemical analyses were made on the entire sample from each of the four rows.

Thin juice apparent purity was determined by the method developed by Brown and Serro (1) except phosphoric acid was used for pH adjustment instead of oxalic acid. Calculations of recoverable sugar assume a standard factory loss of 0.3% and a standard molasses purity of 62.5%.

Results and Discussion

A summary of all data from the test is presented in Table 1. The blends are listed in descending order of production of recoverable sugar per acre. The reliability of the test is good as indicated by the relatively low C.V. values.

(Results in percent of test mean)						
Acc. no.	Inbred tested	No. ¹ hybrids	Yield of rec. sug.	Yield of roots	% sugar	% purity
73HB41	В	10	116.2	122.3	95.7	99.6
73HB40	В	14	115.2	122.8	94.6	99.5
73HB42	В	5	112.5	119.3	94.2	99.8
73HB43	в	5	108.5	110.1	97.9	100.2
73HB45	в	5	108.1	110.5	98.5	99.5
73HB39	В	44	107.6	114.1	95.3	99.5
73HB34	A	6	104.7	102.6	101.6	100.0
73HB37	Α	5	104.4	105.1	99.7	99.7
73HB31	Α	5	103.4	101.4	100.7	100.4
73HB32	A	13	103.1	101.7	100.4	100.3
73HB33	Α	13	100.4	98.1	101.8.	100.0
73HB36	A	9	99.4	97.3	100.3	100.7
73HB30	А	66	98.8	96.2	101.5	100.4
73HB35	A	7	97.8	96.8	100.8	100.0
73HB88	С	5	96.8	95.9	100.8	99.7
73HB50	D	7	92.5	88.9	103.2	100.2
73HB46	D	49	92.0	86.3	104.8	100.7
73HB89	С	11	91.6	93.1	98.0	100.1
73HB47	D	9	91.2	86.5	104.6	100.2
73HB52	С	26	90.6	91.4	99.5	99.7
73HB49	D	7	90.3	86.7	103.8	100.1
73HB48	D	20	89.0	85.6	103.5	100.0
73HB90	С	11	86.0	87.4	98.8	99.6
Means			7757.7	26.2	17.3	92.8
LSD (P.05)			7.1	7.3	2.3	.7
C.V. %			5.8	5.9	1.9	. 6

Table 1.—Performance of hybrid blends, Longmont, Colorado, 1973.

Some individual tester parents are in several blends but no 2 blends have identical sets of tester parents.

Yield of Recoverable Sugar

This data clearly differentiates the combining ability of lines B and A from each other and from lines C and D. Lines C and D were known to be about equal in combining ability for yield of sugar and the results confirm this.

For both inbreds B and C, the lowest and highest yielding blends differed from each other by more than the LSD for this test at the .05 level of probability (Table 2) indicating effects of different tester parents. On the other hand, no sugar yield of the individual blends deviated significantly from the mean of the blends which tested that particular line (colums 3 & 4). If it is assumed that the mean of the blends approaches the true estimate of general combining ability for that line, all blends were satisfactory for evaluating general combining for yield of sugar.

Table 2.—Comparison of blend ranges and extremes for each inbred (yield of recoverable sugar in % of test mean)

Inbred tested	Mean of blends	High minus low	High minus mean	Mean minus low
A	101.5	6.9	3.2	3.7
В	111.4	8.6*	4.9	3.8
С	91.3	10.8*	5.6	5.3
D	91.0	3.5	1.5	2.0

*Significant at the 5% level. LSD (P.05) 7.1.

The data from the general blends made up of a large number of hybrids should be more accurate than from blends constituted from fewer hybrids. The apparent deviation from this assumption for three of the four inbred lines tested could be attributed to experimental error or possibly to heritable productivity differences of the uncommon parental lines. An increase in the number of tester parents above some level should have little or no effect on measuring general combining ability of a parent. The performance of only one general blend, 73HB39, however, differed significantly from the performance of the blend showing the greatest deviation testing that same inbred.

Root Yield

The results for root yield paralleled those for sugar yield. The deviation between the high performing and low performing blends was significant for Inbred C as well as Inbreds A and B (Table 3). Large differences in combining ability for root yield were clearly delineated but certainly more precise testing will be required for small differences.

Inbred tested	Means of blends	High minus low	High minus mean	Mean minus low
A	99.9	8.9*	5.2	3.7
В	116.5	12.7*	6,3	6.4
С	92.0	8.5*	3.9	4.6
D	86.8	3.3	2.1	1.2

Table 3.—Comparison of blend ranges and extremes for each inbred root yield (in % of test mean)

*Significant at the 5% level. LSD (P.05) = 7.3.

Sugar Percent

This test reflected the known large differences in sugar content between the lines tested. In order, high to low, were lines D, A, C, and B. The ranges between the high and low blend were narrowest for the high-sugar-content lines D and A and much wider for lines B and C (Table 4). The ranges for B and D exceeded the LSD for the test at the 5% level of probability and the deviation to the high side from the mean exceeded the LSD for line B, the very low sugar content line. Significant tester parent effects on the low sugar lines suggest less stability in combining ability than in the high sugar lines. Perhaps there is some degree of dominance for high sugar in the 2 lines.

Table 4.—Comparison of blend ranges and extremes for each inbred for sucrose percent (in % of test mean).

Inbred tested	Means of blends	High minus low	High minus mean	Mean minus low
A	100.9	2.1	0.9	1.2
В	96.0	4.3*	2.5*	1.8
С	99.3	2.8*	1.5	1.3
D	104.0	1.6	0.8	0.8

*Significant at the 5% level. LSD (P.05) = 2.3

Laboratory Thin Juice Purity

Differences in thin juice purity among varieties are generally numerically quite small because the ratio "purity" represents percent sugar in total solids in a solution where sugar always predominates. Nonetheless, it is a very important characteristic because it is a principal factor in percent extraction. Significant differences are not always obtained for purity in sugarbeet variety trials although the ranking of varieties is relatively consistent from trial to trial.

This trial, Table 1, using the LSD, failed to differentiate the blends. However, with this number of entries per line, it is obvious that Inbreds B and C produced lower purity hybrids while Inbreds A and D produced higher purity hybrids. There was a significant

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deviation between the high and low purities of blends testing lines A, B, and D (Table 5) which would have resulted in mischaracterizing lines as to their purity, based on the performance of certain blends of seed.

Table 5.—Comparison of blend ranges and extremes for each inbred for thin juice apparent purity (in % of test mean)

Inbred tested	Mean of blends	High minus low	High minus mean	Mean minus low
A	100.2	1.0*	0.5	0.5
В	99.7	0.7*	0.5	0.2
С	99.8	0.5	0.3	0.2
D	100.2	0.7*	0.5	0.2

*Significant at the 5% level. LSD (P.05) = 0.7.

Conclusions

This experiment demonstrated that general combining ability for yield of roots, sugar content, and yield of recoverable sugar can be determined by hybrid blend tests. The blend or entity placed in trial to test an inbred is constituted from equal portions of seed, based on germination, of several hybrids having the line to be tested as the common parent. The inbreds involved in this experiment were selected on the basis of data from previous trials to represent a wide range of combining ability for all characters.

Performances of most of the blends included in this test characterized the inbreds they were intended to test. The only serious misclassification might have occurred for purity. Had this trial been less precise or had the differences between lines been smaller, differentiation would have been less pronounced.

In practice it will probably be necessary to conduct several trials of the same blend in different locations and years to obtain accurate data for characterization of an inbred line. Similar testing over locations and years is necessary with other types of general combining ability tests for reliable evaluation of lines.

The choice of a testing method demands the consideration of a number of factors. Certainly, with very small quantities of seed, the blend test might be the only method which can be used.

For testing in new locations and/or for new characters following elimination of many parents on the basis of other trials the blend test should be considerably more efficient than testing with partial diallel trials. On the other hand, for initial screening, much of the efficiency of using blends is lost because only one parent is tested by each blend whereas two parents are tested by each individual hybrid tested. Comparison of the land or plot requirements for the two methods leads to no definite conclusion. Normally, GW screens hybrids in two row plots with four replications, making a total of eight samples. A trial to test blends might have blends placed in four row plots in six replications for a total of 24 samples. The question remains whether 24 samples in a blend test evaluates an inbred as well as 24 samples testing three different hybrids. In practice, one would not rely on as few as 24 samples on which to make a decision using either method or on performance of only three hybrids unless the performance were exceptional.

The interpretation of data from blend trials is simpler than from data obtained by testing several hybrids, but some information is lost in testing blends. It could sometimes be important to know how individual hybrids fluctuate about the mean when hybrids having a common parent crossed to several other parents are evaluated. No specific combining ability data is obtained from testing blends but this loss can be considered small because the data from an individual hybrid are generally imprecise.

Without proper design and materials some errors could occur and should be considered when the blend test is used. The difference between the average combining ability of the non-parental lines used in testing two lines might be greater than the difference in the combining ability of the lines under trial, in which case an incorrect conclusion would be drawn. If care is taken in the selection of tester lines, this need not occur. In normal use of the blend method, hybrid productions should involve a given set of tester parents. Although similar to topcross testing the method can provide greater genetic diversity than is attainable with existing topcross parents. Including broad-based male sterile populations in seed multiplication isolations involves risk because male sterility is often unstable in genetically diverse populations. Both male and female parents can be tested by the blend method, which can result in more efficient use of seed isolations. Breeding and seed production methods and seed inventories will determine whether blend testing will be beneficial in a breeding program.

Another possible error could result from differential emergence of the blend components which could cause one component to be disproportionately represented in the harvested population. Such a case would result in a deviation in the performance of the blend from its true value. This possibility is more likely to happen if the number of blend components is small. The results from this test indicate this does not generally occur because blends with as few as five components performed in about the same manner as blends having far more component lines. Where individual hybrids are tested, weak emerging seed lots often result in poor stands which affect the performance of the line itself and provide less competition to neighboring plots. Blending will reduce or eliminate the problem resulting in improved variety tests.

Little or nothing is known about differential intraplot competition of hybrids. Neither is anything known whether pure stands of hybrids correctly reflect combining ability when the ultimate hybrid is likely to be a different combination altogether. The same arguments could apply to the generally accepted procedure of topcross testing.

The blend test may be considered another tool for sorting lines or strains for general ability if care is taken in its use.

Summary

A procedure to use the performance of blends of hybrids having a parent in common rather than using a series of individual hybrids for predicting general combining ability was examined. Using inbreds of known and widely varying combining ability for yield of roots, sugar content and juice purity, a trial of blends of hybrids resulted in differentiation of the inbred lines. Blends with as few as five component hybrids evaluated general combining ability. This method has proven to be another tool for sorting germplasm and could result in substantial saving when the seed supply of hybrids is low, when testing is required in areas having limited resources, and possibly when some screening of inferior lines has already occurred.

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