Inheritance of Raffinose Production in the Sugar Beet

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Introduction

The sugar beet is known to contain many substances other than the sucrose which is commonly recovered as a commercial product. In general these substances tend to be melassigenic in the process of sucrose recovery from the beet. Usually, in factory operations these currently unwanted chemical compounds are referred to by the one term "impurities," regardless of their individual nature. It seems, however, that in order to make the most effective study of such a problem it might be well to separate these impurities into component parts if the methods are available to effect such a separation.

One of the principal carbonaceous compounds in this general classification is the tri-saccharide, raffinose, with the chemical make-up $C_{18}H_{32}O_{16}$. At the present time no commercial use is made of this sugar and the total United States consumption is about 100 pounds yearly for laboratory use. Consequently, it would be desirable to eliminate or reduce this nuisance from the sugar beet unless some further use of it can be found.

The study of the inheritance of raffinose production in the beet was made possible; first, by the development of a rapid chromatographic method for raffinose determination by Brown (1)[2] in 1952; second, the development by Powers (2) of biometrical methods for study of quantitative characters; and third, by the discovery of inbred lines of sugar beets in the "Cooperative Inbred Indexing Program" (3) having extremes of raffinose content.

Progeny test results from one mass selection for both high and low raffinose content of beets have been reported by Wood (4).

Materials and Methods

The biometrical analysis of the data given here follows rather closely the methods developed and/or described by Powers (2). Chemical analysis followed the method of Brown (1) with slight modification for large scale laboratory operation. Two inbred lines from the Inbred Indexing Program (3) were selected as parental material. These were (a) number 50-415 developed by the late G. W. Deming of the USDA and (b) number 410701 from H. L. Kohls at Michigan State University. Hybrids in the F_1 generation were identified by the use of hypocotyl color as a marker gene. Backcross generations were produced by similar technique. Six populations were studied; high raffinose parent P_1 (410701), low raffinose parent P_2 (50-415) F_1, F_2, B_1P_1, and B_1P_2. All the above six populations were grown in a randomized complete block experiment with twenty replicates for each population. Twelve, fully competitive (12-inch by 22-inch spacing) beets were taken consecutively from the center row of each three-row plot and analyzed individually; thus, a total of 240 roots constituted each population studied.

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[2] Numbers in parentheses refer to literature cited.

Determination of raffinose is given as percent on dry substance in the beet juice and assumes a constant marc. Calculations are on obtained data without transformation.

Variability of Population

The variances of the parental and F_1 populations may be used to measure environmental variability if the parents are homozygous for the genetic factors under consideration. The total variance of the segregating generations is the sum of the genetic and environmental variability. In this study, the variances provide an estimate of the homozygosity of the parents. Inasmuch as the mean squares of the two parents and the F_1 (Table 1) do not differ significantly as measured by the standard errors of the variance as described by Fisher (5), it may be assumed that the parents are homozygous for genes conditioning raffinose production.

Table 1.—Variances and Their Standard Error for Percent Raffinose on Dry Substance for Six Populations Derived from Two Inbred Lines of Sugar Beets.

Population	Variance (Mean square)
410701 (P_1)	.120364 ± .00470
B_1 to 410701	.133621 ± .00579
F_1	.111687 ± .00405
F_2	.251375 ± .02050
B_1 to 50-415	.163038 ± .00862
50-415 (P_2)	.122431 ± .00486

Population Means

The obtained means of the various populations differ significantly in all comparisons except F_1 and F_2 as shown in Table 2. The means of the F_1 and F_2 populations are statistically equal and are equal to the average of the means of the parental populations. The means of the backcross populations are also equal to the average of the means of the F_1 and their respective parents. These relationships can exist only in the absence of measurable dominance and/or genic interactions or linkage. Comparison of obtained population means with calculated theoretical means on the arithmetic scale gives a good fit by both standard error and X^2 tests. The X^2 value for comparison obtained with geometric means eliminates the possibility that gene action in this case can be following the geometric scale. The scale thus can be considered as additive and not multiplicative.

Table 2.—Obtained Arithmetic and Theoretical Arithmetic Means Compared with Geometric Means for Percent Raffinose in Sugar Beets Based on Dry Substance.

Population	Obtained Arithmetic	Theoretical Arithmetic	Theoretical Geometric
50-145 (P_2)	0.58 ± .036	.....	.....
B_1 to P_2	0.76 ± .038	0.86 ± .025	0.81
F_1	1.13 ± .034	1.06 ± .025	0.94
F_2	1.16 ± .051	1.09 ± .028	1.03
B_1 to P_1	1.25 ± .041	1.34 ± .025	1.31
410701 (P_1)	1.53 ± .036	.....	.....

$X^2 = .273$ with P = between .95 and .98 for comparison between obtained and theoretical means.

Table 3.—Frequency Distributions as Percent for Six Populations for Percent Raffinose on Dry Substance.

	Upper Limit of Class														
Population	.24	.39	.54	.69	.84	.99	1.14	1.29	1.44	1.59	1.74	1.89	2.04	2.19	2.34
410701 (P_1)					.42	.83	6.67	10.83	18.33	17.92	20.00	18.33	2.92	2.50	1.25
B_1 to P_1		.42	.42	2.92	4.17	8.33	21.67	16.25	17.08	13.33	9.17	5.42	.42	.42	
F_1		.42	.83	2.92	6.67	17.92	24.16	19.58	14.58	7.08	3.75	1.67	.42		
F_2		.42	2.08	6.67	8.75	13.33	22.50	9.17	14.17	8.33	6.25	7.50	.42	.42	
B_1 to P_2	.83	5.00	15.00	16.67	25.83	17.50	12.92	2.92	1.25		.83	1.25			
50-415 (P_2)	11.25	12.92	21.67	17.08	21.25	9.17	3.33	2.50	.83						

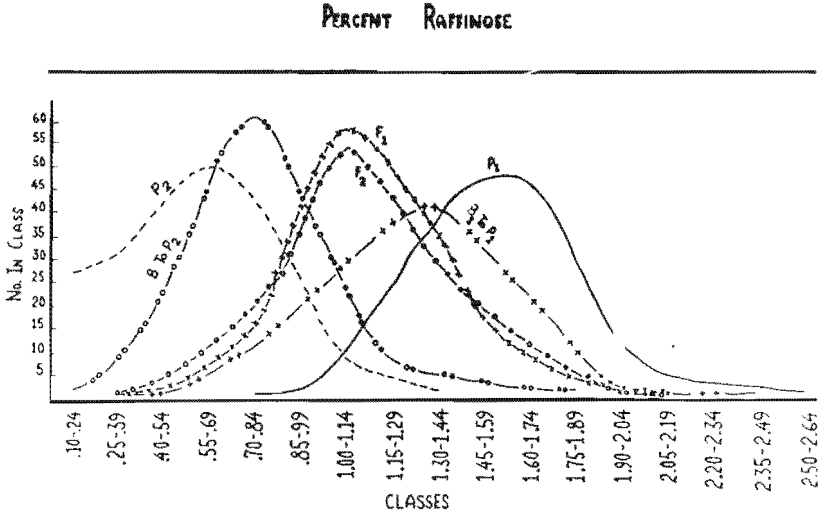

Figure 1.—Frequency distribution for six populations.

Genetic Analysis

Both Table 3 and Figure 1 indicate that the frequency distributions for the P_2 and B_1 to P_2 populations do not reach zero on the scale, indicating P_2 carries some genes for raffinose production.

Theoretical values for the ratios of the extreme genotypes each population has in common are given in Table 4. This table has been calculated from the expansion of the binomial for the backcross and F_2 populations, as demonstrated by Powers (2); the ratios are developed assuming no dominance, linkage, or gene interactions; and that effective factor pairs for the production of raffinose are equal in magnitude, and that one effective factor pair is non-isodirectional (or in other words, one effective factor pair for raffinose production enters the cross from the low raffinose parent).

One premise in the use of genotypic ratios for determination of effective factor pairs is that the frequency distributions should not be inhibited at the end of the curve from which the ratios are determined. It was indicated above (Table 3 and Figure 1) that both the P_2 and to a lesser extent the B_1 to P_2 populations, appear to be inhibited at the lower end of the population curve. The lower classes of the P_2 and B_1P_2, however, are probably accumulated rather than inhibited. In the chemical analysis used it is necessary to accumulate numbers in the lower classes due to the lack of refinement in measuring minimal values. Since arithmetic accumulation is involved in the determination of the ratios of the extremes of the populations, this analytical accumulation should not influence the obtained ratios unless there is accumulation beyond the classes which involve the genotypes under consideration. Assuming this to be an accumulation, rather than an inhibition, it is possible to utilize both extremes of the frequency distributions in estimating genotypic ratios.

Table 4.—Theoretical Genetic Model for Number of Factor Pairs. One Effective Factor Pair Non-isodirectional but All of Equal Magnitude in Effect.

Number of Factor Pairs	Either B_1 to P_2 / F_1 or B_1 to P_1 / F_1	Either B_1 to P_1 / P_1 or B_1 to P_2 / P_2	Either F_2 / P_1 or F_2 / P_2	Either F_2 / B_1 to P_1 or F_2 / B_1 to P_2
2	75.000	75.000	68.750	125.000
3	50.000	50.000	34.375	87.500
4	31.250	31.250	14.453	56.250
5	18.750	18.750	5.469	34.374
6	10.9375	10.938	1.929	20.314
7	6.250	6.250	0.647	11.726

Table 5.—Ratios, as Percentages, Between Frequency Distribution Classes for Specified Populations Percent Raffinose on Dry Substance.

B_1 to P_1 / F_1	B_1 to P_2 / P_2	F_2 / P_2	F_2 / B_1 to P_2	B_1 to P_2 / F_1	B_1 to P_1 / P_1	F_2 / P_1	F_2 / B_1 to P_1
99.60	97.92	77.09	85.42	56.54	12.59	12.59	74.51
95.76	97.48	63.45	78.73	71.67	25.04	33.36	82.09
89.84	96.98	55.60	65.09	74.73	34.29	32.42	89.14
81.83	86.60	33.48	57.33	81.83	45.71	36.43	94.37
74.73	75.24	21.20	38.66	89.84	56.42	45.65	98.32
71.67	59.60	14.57	28.30	95.76	67.43	50.24	100.00
56.54	45.44	5.45	21.45	99.60	84.82	69.63	
	24.12		12.00		92.48	82.44	
	7.38		7.20		96.26	90.84	

Table 5A.—Indicated Number of Factor Pairs for Each of Eight Genotypic Ratios.

	B_1 to P_1 / F_1	B_1 to P_2 / P_2	F_2 / P_2	F_2 / B_1 to P_2	B_1 to P_2 / F_1	B_1 to P_1 / P_1	F_2 / P_1	F_2 / B_1 to P_1
Indicated No. of Factor Pairs	3	3-5	3-5	5-6	3	3-5	3-5	5-6

The obtained ratios for the various comparisons are listed in Table 5. The following number of factor differences between the two parents were estimated when the obtained values are compared to the theoretical values for different numbers of factor pairs (Table 4).

This information indicates that the two parents differ by about 5 effective factor pairs for raffinose production, one of which is non-isodirectional.

Inheritance in Relation to Results of Previous Mass Selection

It was reported by Wood (4) that progeny of one mass selection for low raffinose content had about 25 percent lower raffinose than the parent variety. This reduction is in line with the findings of this inheritance study. Assume the parental variety from which the selection was made had the same genetic characteristics as the F_2 population studied. 10 beets were

chosen from 150 in the selection. If these represented the lower 1/15 of the genotypic distribution, assuming 5 effective factor pairs of equal magnitude in effect, additive, and in equilibrium, the theoretical reduction in raffinose should have been 60 percent; the result of a reduction in gene frequency from 50 percent to 20 percent. However, with genetic and environmental variability confounded, it might be assumed that the roots selected had genotypes falling in the lower half of the genotypic distribution. On this basis the frequency in raffinose genes would be reduced from 50 percent to 37.7 percent, resulting in a reduction in raffinose of only 25 percent.

Summary

1. The inbred lines of sugar beets used as parental material for this study were apparently homozygous for the factors each carried for production of raffinose in the beet.

2. Quantitatively, the factors for raffinose production in the two parents followed an arithmetic scale and consequently are additive.

3. The number of effective factor pairs for production of raffinose between the two parents is about five and at least one is non-isodirectional and all are equal in magnitude.

4. Neither dominance, heterosis, nor linkage appeared to be involved in the cross studied.

References

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