Hydrogen Ion Effects on the Early Growth of Sugar Beet Plants in Culture Solution

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Sugar beet plants grow well in soils of pH 6.0 to 8.0 (12)[2], but from pH 7.8 to 9.3 growth and pH have a negative correlation coefficient (3). Tomatoes and lettuce grow well in culture solutions kept at pH 5.0, 6.0, and 7.0 and Bermuda grass from pH 4 through 8.0, providing iron is supplied as a humate and all other nutrients are kept relatively constant (2). Root growth fails completely at pH 3 and top growth is greatly reduced for all plants at pH 9 (2).

Sugar beets in greenhouses make excellent growth in culture solutions supplied with iron sulfate, iron citrate or iron tartrate as a source of iron, providing the pH of the culture solution remains below 6.8 to 7.0 (17). Above pH 7.0 daily additions of iron salts are necessary to prevent iron deficiency, or alternatively, the culture solutions have to be acidified with sulfuric acid from time to time. The latter course of action is usually the simplest but even here the taking of pH readings and the addition of acid is time consuming. Care also has to be taken not to acidify the culture solutions much below pH 5.0, since at pH 4.0 injury has been observed at times. Fortunately, these precautions now appear to be unnecessary with the use of iron chelates as a source of iron.

With chelated iron in culture solutions it has been found that sugar beet plants do not become chlorotic at a pH of 7.0, and in fact, they remain quite green at a pH of 7.4 and 7.6 (17). This observation suggested that the entire matter of pH and its effect upon the growth of sugar beet plants in culture solutions should be subjected to further experimentation. The present paper gives the findings for sugar beet plants cultured in solutions kept at a pH of 4.0, 5.0, 6.0, 7.0, 8.0 and 9.0.

Methods and Materials

The seeds of U. S. 22/3 and GW 304 A were planted on October 27, 1953. Twenty-five grams of seeds were treated with 0.25 grams of Phygon XL and planted 2.0 cm. deep in a 31 x 76 x 20 cm. germinating tray filled with vermiculite No. 2. The seedlings were watered daily with one-half strength Hoagland's culture solution No. 1 (5). When the seedlings were in the early 2-leaf stage, they were carefully removed and the roots washed nearly free of vermiculite by dipping in tap water. The individual plants were supported in a cork ring with non-absorbent cotton. Three seedlings prepared in this manner were taken at random for each 20-liter tank. Five tanks were used for a variety and pH treatment, giving a total of 60 tanks for the 2 varieties and 6 pH values. The initial composition of the nutrient solution, in millimoles per liter, was as follows: 1.0 KH_2PO_4, 2.5 KNO_3, 0.25 K_2SO_4, 1.0 $MgSO_4$, 2.5 $Ca(NO_3)_2$, 0.25 $CaCl_2$, and 0.25 Na_2SiO_3. The microelements in p.p.m. were added as follows: 0.25 B, 0.25 Mn, 0.025 Zn, 0.01 Cu, and 0.005 Mo. Iron was added one time only at the

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[2] Numbers in parentheses refer to literature cited.

rate of 4.9 p.p.m. as the ferric potassium ethylene diamine tetra-acetate complex (7). After the salts were added, the pH value of the solution was 6.7. The pH values of the solutions were adjusted to 4.0, 5.0, 6.0, 7.0, 8.0 and 9.0 with either 1.0 N H_2SO_4 or 1.0 N NaOH. All pH measurements were made with the Beckman Model M pH meter and each 20-liter tank was adjusted individually with the appropriate reagent. Measurements for pH were taken every other day for the first 16 days and daily thereafter. At these times the solutions at pH 4.0 and 5.0 tended to increase by 0.1 to 0.3 pH units, those at pH 6.0 and 7.0 by 0.2 pH units and those at pH 8.0 and 9.0 varied from 7.7 to 8.0 and 8.5 to 9.0, respectively. The culture solutions were aerated continuously starting the 10th day after transplanting. Distilled water was added during the experiment. The plants were harvested on January 5, 1955, and fresh weights of the tops and beet roots were recorded. The tops and fibrous roots were dried in a 70° C. forced draft oven and the storage roots were frozen in a 2-ounce glass jar with dry ice. The dried tops and fibrous roots were ground to pass a 40-mesh screen by means of an intermediate model, Wiley Laboratory Mill.

Potassium and sodium were determined with the Perkin Elmer Model 52A flame photometer; nitrate-nitrogen with the phenol-disulfonic acid method (9); phosphate-phosphorus soluble in 2 percent acetic acid with the phosphomolybdate method (14); and calcium with the calcium oxalate precipitation method.

Table 1.—Effects of pH on the Growth of Sugar Beets in Culture Solution.

pH culture solution	Fresh weight		Dry weight					
	Tops	Beets	Tops		Fibrous roots	Living leaves	Total leaves	Plant height
	grams	grams	grams	Percent	grams	number	number	centimeter
				pH Means[1]				
4.0	107	7.0	7.1	6.69	0.86	34.5	41.7	27.2
5.0	170	13.6	10.9	6.42	1.02	37.8	45.3	32.8
6.0	213	16.6	13.7	6.45	1.26	39.2	47.1	31.8
7.0	240	22.9	15.6	6.50	1.42	41.7	49.8	34.2
8.0	215	18.8	13.8	6.43	1.36	42.8	50.6	33.9
9.0	174	15.7	11.8	6.83	1.53	42.0	49.6	31.6
L.S.D.[2]	31	3.0	1.9	0.25	0.20	3.1	3.7	1.9
F-value[3]	18.7	25.1	19.4	3.79	13.0	8.3	6.8	14.6
				Variety Means[4]				
U.S. 22/3	177	14.8	11.6	6.55	1.15	39.9	47.9	31.0
GW 304	196	16.7	12.8	6.56	1.34	39.4	46.8	32.9
L.S.D.[2]	18	1.8	1.1	n.s.	0.12	n.s.	n.s.	1.1
F-value[5]	4.89	4.77	4.93	0.04	11.19	0.25	1.24	11.83

[1] Means of 10 values, varieties pooled.
[2] L.S.D. = least significant difference at the 5% level. n.s. = not significant.
[3] Required F-value at the 5% level is 2.43, at the 1% level, 3.46.
[4] Means of 20 values, treatments pooled.
[5] Required F-value at the 5% level is 4.06, at the 1% level, 7.24.

Table 2.—Effect of pH on the Mineral Composition of Tops, Fibrous Roots and Beet Pulp of Sugar Beet Plants.

pH culture solution	Tops[1]					Fibrous roots[1]		Beet pulp[2]	
	NO_3-N p.p.m.	PO_4-P p.p.m.	K Percent	Na Percent	Ca Percent	K Percent	Na Percent	K Percent	Na Percent
	pH Means[3]								
4.0	14,430	9440	7.52	1.83	0.67	4.26	0.08	.673	.058
5.0	18,190	4030	9.03	1.39	0.83	6.25	0.17	.724	.033
6.0	18,100	3090	9.60	0.95	0.89	7.55	0.20	.731	.024
7.0	18,290	2850	9.75	1.02	0.88	7.09	0.24	.715	.024
8.0	18,010	2550	9.79	1.14	0.67	7.16	0.27	.739	.026
9.0	16,970	2190	9.46	1.55	0.61	6.21	0.24	.756	.033
L.S.D.[4]	1,010	540	0.43	0.17	0.06	0.69	0.08	.042	.009
F-value[5]	17.9	210.3	33.5	32.1	30.9	24.3	6.5	3.6	18.2
	Variety Means[6]								
U.S. 22/3	17,270	4070	9.24	1.27	0.76	6.16	0.197	0.727	.031
GW 304	17,400	3980	9.14	1.36	0.76	6.68	0.203	0.719	.035
L.S.D.[4]	n.s.	n.s.	n.s.	n.s.	n.s.	0.40	n.s.	n.s.	n.s.
F-value[7]	0.20	0.32	0.65	3.48	<0.01	6.89	0.07	0.35	3.04

[1] Dry basis
[2] Fresh basis
[3] Means of 10 values, varieties pooled
[4] Least significant difference at the 5% level; n.s. = not significant
[5] Required F-value at the 5% level is 2.43, at the 1% level, 3.46
[6] Means of 20 values, treatments pooled.
[7] Required F-value at the 5% level is 4.06, at the 1% level, 7.24.

EXPERIMENTAL RESULTS

Effects Upon Growth

The hydrogen ion concentrations of the culture solutions had a marked effect upon the over-all growth of the sugar beet plants. At pH 4 this effect was manifested by a smaller size of the leaves, a darker green color of the leaf blades and a less vigorous development of the fibrous roots. From pH 7 to 9 there were no visible changes in the growth of the plants and neither were there any signs of necrosis or of chlorosis. Without the use of iron chelates as a source of iron, chlorosis would have developed readily at pH 7, and at pH 9 growth would have been retarded greatly.

In general, the two varieties studied, U. S. 22/3 and GW 304, responded equally to changes in pH of the culture solution even though there were significant differences in growth (Table 1) and an occasional difference in mineral composition of the plants (Table 2). When the results were analyzed statistically (4), there were no significant pH-variety interactions and for this reason the values for the two varieties have been combined in Tables 1 and 2 and in Figures 1, 2 and 3.

Tops: The fresh weights of the tops for both varieties increased rapidly from pH 4 to pH 6 (Figure 1 and Table 1). From pH 6 to 8 top growth attained a maximum value at pH 7 but none of these differences are significant at the 5 percent level (Table 1). However, from pH 8 to 9 the decrease in top weight (Figure 1) is nearly significant to the 1 percent level (Table 1).

The dry weights of the tops (Table 1) in contrast to the fresh weights, differ from each other at approximately the 5 percent level or more at each pH interval. On this basis the optimum pH for dry weight production is at pH 7 (Table 1).

The dry weight percentages of the tops (Table 1) are surprisingly uniform over the entire pH range of values especially when one considers the large differences in fresh and dry weights of the tops observed over these pH values. The lowest average value is 6.42 percent and the highest value 6.83 percent or a difference of 0.41 percentage units. While this difference is relatively small, an increase of only 0.25 and 0.33 percentage units are required for significance at the 5 percent and 1 percent levels, respectively (Student's t-test) (4). On this basis the percentage dry weight increases are significant for pH 4 and for pH 9.

Beet Roots: The storage root weights differed significantly from each other at each pH interval (Table 1). Starting at pH 4 the roots gradually increased in size until they attained a maximum value at pH 7 (Figure 1). The decline from pH 7 to 9 while small, is significant statistically (Table 1).

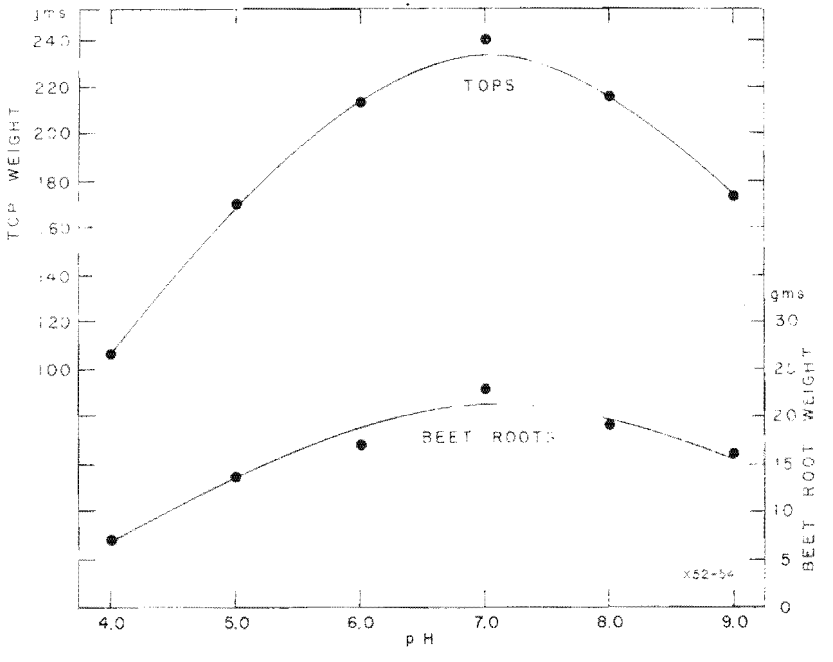

Figure 1.—Relation of top and root weight (fresh basis) of sugar beet plants to the pH of the culture solution.

Fibrous Roots: The dry weights of the fibrous roots (Table 1) increased gradually until pH 7, and thereafter, from pH 7 to 9, the pH of the culture solution had no effect upon the growth of the fibrous roots. Other than this reduction in growth, the fibrous roots remained filamentous in nature and appeared normal in all respects.

Leaf Formation and Plant Height: The total number of leaves produced by the beet plants at each hydrogen ion concentration was affected adversely only at pH 4 and 5 (Table 1). These decreases in total leaf production, while significant, were nevertheless rather small in magnitude. From pH 6 to 9, hydrogen ion concentration had no effect on the number of leaves produced by the beet plants.

The number of living leaves at the time of harvest was related to the hydrogen ion concentration of the culture solution in the same manner as the total number of leaves produced during the course of the experiment (Table 1). Apparently, leaf formation and the number of living leaves on a plant are affected less by the pH of the culture solution than the top and beet root growth.

Similarly, plant height (Table 1) is affected only slightly at the extreme pH values of the culture solutions used within this experiment. As with

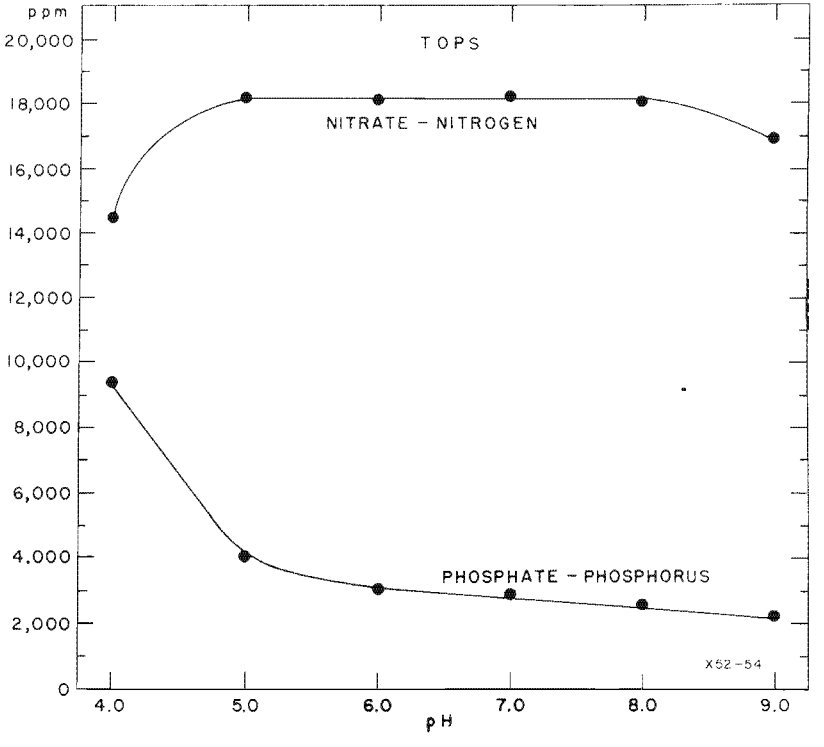

Figure 2.—Relation of nitrate-nitrogen and phosphate-phosphorus of the tops (dry basis) to the pH of the culture solution.

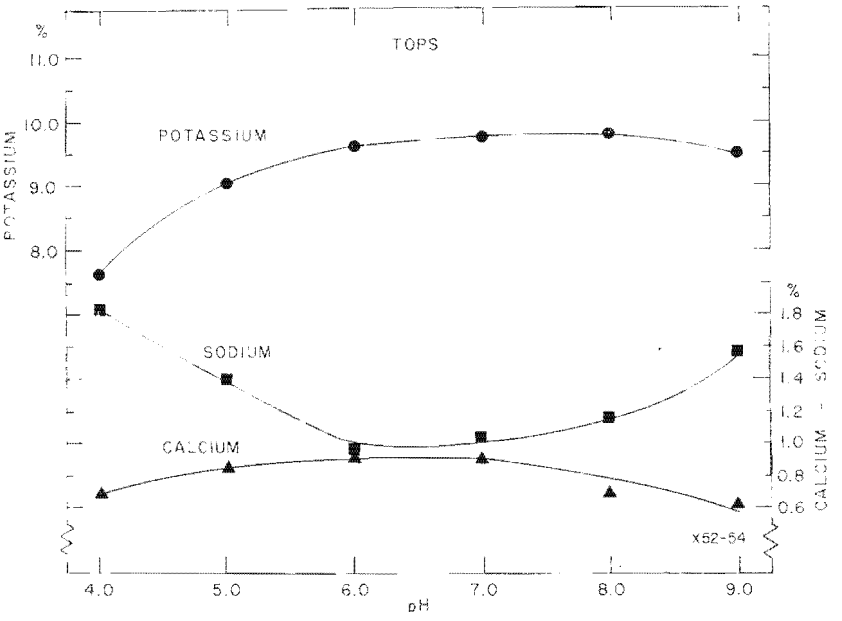

Figure 3.—Relation of potassium, sodium and calcium of tops (dry basis) to the pH of the culture solution.

the other measurements made on growth, the effects on plant height would have been greater had a wider range of hydrogen ion concentrations been used experimentally.

Effects Upon Mineral Composition

Even though a pH of 4 to 9 had no visible effect on the leaf color of the sugar beet plants, there still remained the possibility of a nutrient deficiency that could be revealed only by a chemical analysis of the leaves and the roots themselves. For example, quite often the only sign of a phosphorus deficiency in sugar beet plants is a reduction of leaf size and an intensified green color of the leaves. Under these conditions, the plants appear perfectly normal and frequently the deficiency is not detected, even by an experienced observer, unless plants with ample amounts of phosphorus are adjacent to the deficient plants. At low pH values cations, such as potassium (8, 10) and calcium (1), fail to be absorbed readily and the symptoms as ordinarily manifested at pH values from 6 to 7 could conceivably be absent at pH 4. If the hydrogen ion concentrations that cause decreases in growth are not associated with nutrient deficiencies, then the alternative possibility of a direct effect of the hydrogen ion or hydroxyl ion on growth reduction would thus appear quite likely.

Nitrate-nitrogen: The nitrate-nitrogen concentrations of the tops of the beet plants were relatively constant from pH 5 to 8 (Table 2 and Figure 2). Only at pH 4 and again at pH 9, were there significant decreases in nitrate-nitrogen concentrations but these decreases were far from indicating a

nitrogen deficiency of the plants. A deficiency would be indicated by values of 1,000 p.p.m. nitrate-nitrogen or less (15) and therefore, the cause of the growth reduction must be sought elsewhere.

As to the reason for the decrease in nitrate-nitrogen concentration of the tops at pH 4 and 9, it does not seem to be a matter of dilution either by an increase in percent dry matter of the tops or by an increased growth of the plants (Table 1). Thus, at pH 4 and 9 the grams of dry matter produced are less than at the intermediate pH values and also the dry matter percentages are relatively constant for the entire pH range study (Table 1). At pH 9 the competitive action of bicarbonate ions (8, 11) or a decreased capacity of the nitrate carrier system (6) may account for the decrease in absorption of nitrate but at pH 4 the nature of the decrease is not at all clear at present.

Phosphate-phosphorus: The phosphate-phosphorus concentrations within the tops of the plants formed an interesting pattern relative to the hydrogen ion concentration of the culture solution. At pH 4 the phosphate-phosphorus concentrations of the tops are exceedingly high (Table 2). This is followed by a precipitous decline at pH 5 and thereafter, the decreases are rather moderate in magnitude, ending at pH 9 with an average value of 2,190 p.p.m. of phosphate-phosphorus. This value is still well above the critical phosphorus value set for either petioles or blades of sugar beet plants (13) and accordingly, the differences in growth associated with pH of the culture solution cannot be directly attributable to a phosphorus deficiency within the sugar beet plant unless one is to assume that the plant requires more phosphate at pH 9 and that the phosphorus value of 2,190 is critical, whereas the value of 2,550 at pH 8 is not. At pH 4 there is the possibility that phosphate-phosphorus concentrations as high as 9,440 p.p.m. may be inhibitory to growth but as yet no definitive experiments have been run covering this situation.

Potassium: The changes in potassium content of the sugar beet plant relative to pH (Table 2 and Figure 3) offer no clue as to the cause of the decreases in growth observed at the extremes in hydrogen ion concentration of the culture solutions. Even the lowest potassium value at pH 4 is exceedingly high in comparison to the critical value of 1.0 percent potassium (16) and of course none of the values can be considered toxic because the potassium values from pH 6 to 9 are approximately identical and this includes the region of best growth. Similar conclusions can be drawn from the potassium values of the fibrous roots and of the beet pulp (Table 2).

Calcium: The calcium concentrations at pH 4, 8 and 9 decreased very little in comparison to the best plants (Table 2 and Figure 3). Since the decreases were rather small, it is unlikely that the low calcium values were associated with a calcium deficiency. Apparently, the calcium absorbed was ample to meet the needs of the plant even though the hydrogen ions of the culture solution at pH 4 may have reduced the absorption of cations and the hydroxyl ions at pH 8 and 9 tend to precipitate the calcium as calcium phosphate. Thus, it seems that the reasons for the reduction in growth associated with pH must be sought elsewhere.

Sodium: The sodium concentrations of the tops of the sugar beet plants are affected by the pH of the culture solution (Table 2 and Figure 3). Strangely enough, the highest sodium values (1.83 percent) were observed for the plants grown in solutions with the highest hydrogen ion concentration. At pH 7 to 9 the sodium concentrations of the tops increased again. Most likely these increases are the result of adding the sodium hydroxide needed to maintain the pH of the culture solution at the specified value. Sodium addition is not a factor at pH 4 and 5, where sulfuric acid was added to maintain the solutions at the specified pH values. As with potassium, the accumulation of sodium was not great enough to be toxic to the growth of the sugar beet plant.

The sodium concentrations of the pulp for each pH level are in accord with those of the tops but in the fibrous roots the concentrations follow those of potassium (Table 2).

Discussion

The hydrogen ion concentration of the culture solution has been found to have an important effect upon the growth of the sugar beet plant (Table 1). Significant reductions in top growth took place at pH 4, and contrary to expectations, at pH 5. Growth was excellent at pH 6, 7 and 8 and only at pH 9 did a significant reduction in top growth again take place.

Storage root growth for young plants differed significantly at each pH level of the culture solution. If similar effects carry over to older plants, then slight deviations from pH 7 may diminish storage root weights significantly. Experiments with larger sugar beet plants with larger storage roots would add much to our knowledge concerning the long term effects of pH upon the growth and sugar concentration of plants in culture solutions.

The chemical analysis of the tops, fibrous roots and pulp (Table 2 and Figures 2 and 3), revealed no important factor that could account for the decreases in growth observed at pH 4, 5 and 9. At pH 4 the hydrogen ions of the culture solution did not prevent the absorption of adequate amounts of potassium and calcium, and in fact, appeared to enhance the accumulation of sodium in the tops of the plants. At pH 4 phosphate accumulated to a very high concentration within the tops of the plants and it is possible that this may have interfered with the metabolism of the plant. Nitrate concentrations were reduced only slightly and could not have caused a deficiency of nitrogen in the plants. Unless further analyses reveal a specific cause of the decrease in plant growth a direct effect of the hydrogen ions at pH 4 and of the hydroxyl ions of pH 9 appears most likely to have been the cause for the decrease in growth of the plants at these pH levels.

Summary

The effect of the pH of the culture solution on the growth and mineral composition of sugar beet plants was investigated from pH 4.0 to pH 9.0 for two beet varieties, U. S. 22/3 and GW 304. Chelated iron, instead of ferrous sulfate, tartrate or citrate, was used as a source of iron.

Over the pH range studied there were no visual deficiency symptoms of iron nor of any other nutrient. In fact, the plants appeared perfectly normal except for a reduction in size of tops at pH 4, 5 and 9. Maximum top growth took place at pH 6.0, 7.0 and 8.0. Storage root growth differed significantly at all pH values, with an indicated optimum at pH 7.0. Plant height and the total number of leaves formed or living at the time of harvest were reduced mainly at pH 4.0.

Potassium, sodium, calcium, nitrate and phosphate ions were present in the tops of the plants in amounts sufficient to maintain good growth. None of these ions were below the critical level, and accordingly, the reduction in top and root growth that took place mainly at pH 4, 5 and 9 appears to be associated with a "direct" effect of hydrogen or hydroxyl ions on the metabolism of the plant rather than with an "indirect" effect upon ion absorption or upon the availability of nutrients from the culture solution.

Grants-in-aid received from the Beet Sugar Development Foundation, Fort Collins, Colorado, in support of the present study are gratefully acknowledged.

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