Nitrogen Constituents Associated with Reduction of Sucrose Percentage and Purity of Sugar Beets[1]

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The role of nitrogen fertilizer is associated intimately with the efficient production of sugar beets. Early in 1955 the senior author circulated a questionnaire to determine the fertilizer practices being followed in the principal sugar beet growing areas of western United States. Prominent among the fertilizer problems in need of solution in all areas was the proper use of nitrogen.

This general interest in nitrogen stems from the realization that nitrogen deficiencies result in relatively low yields, while excesses of nitrogen frequently give high tonnages of beets relatively low in sugar content and purity. Factory operators have become increasingly conscious of the nitrogen problem as they struggle with ways of increasing sugar recoveries from sugar beet extractions.

Studies by Afanasiev et al. (1)[3], Gardner and Robertson (4), Haddock (5), Hirst and Greaves (7), Larson (8), Walker and Hac (12), Walker and Hac (12), and Ulrich 10), among others, have shown a close inverse relation between nitrogen fertilization and sugar beet quality. The relation between these two factors is partially explained by Walker and Hac (12) on the basis of growth and storage potential and by Ulrich (10) on the basis of environmental factors such as intensity of sunlight, night and day temperatures, and nitrogen supply.

It should not be assumed that nitrogen fertilization is the only factor which may modify the sucrose and purity of sugar beet extracts. It is suggested by Brown and Wood (2) and Doxtator and Bauserman (3) that sodium and potassium may be melassigenic substances.

Since it has been shown that nitrogen fertilization and nitrogen content of plant tissue are related to depression of sugar and purity of the extract juice of beet roots, it seems desirable to determine which of the various nitrogen constituents in beet roots appears to be related directly to these quality factors.

Procedure

The experimental material used in this study was obtained from an extensive field experiment on Millville loam at Logan, Utah. Two methods of irrigation, sprinkler and furrow, with four soil moisture conditions superimposed on each method, were studied (available soil moisture remaining in root zone allowed to approximate 90 (W_4), 70 (W_3), 40 (W_2), and 20 (W_1) percent between irrigations). Soil moisture plots were further subdivided for nitrogen and phosphorus fertilizer treatments. Nitrogen was applied at the rate of 80 pounds N per acre as ammonium sulfate.

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[3] Numbers in parentheses refer to literature cited.

The pulp samples were dried for 48 hours in a forced draft oven at 70° centigrade. All analyses, with the exception of sucrose, purity, and glutamine, (made on fresh pulp) were made on dried pulp. Glutamic acid was determined by the biological assay procedure on frozen fresh pulp and calculated to dry basis. Glutamine nitrogen was calculated from glutamic acid data. The analytical chemical procedures were carried out as follows:

1. Total organic and ammoniacal nitrogen were determined by a slight modification of the method of Hillebrand et al. (6). Four grams of digestion mixture (500 parts Na_2SO_4 50 parts $CuSO_4$ and 5 parts Se were added to solution containing 0.5 gram of dry beet pulp. Fifteen ml. H_2SO_4 were added and the mixture boiled until clear plus 5 minutes. Solution was made alkaline and distilled.

For all other determinations 4 grams of the dried pulp was extracted with 140 ml. 2 percent acetic acid solution and 50 ml. aliquots were taken for the different analyses as follows:

2. Soluble organic and ammonical nitrogen were made as in 1 above.

3. Ammonia nitrogen was determined by direct Nesslerization of an aliquot of extract (14).

4. Soluble organic nitrogen = procedure 2 minus 3 above.

5. Ammonia plus amide nitrogen was determined by distilling an aliquot from a 20 percent NaOH solution and titrating (13).

6. Nitrate nitrogen was determined by adding Devardas Alloy to the residue from procedure 5 and distilling (6).

7. Amide nitrogen = procedure 5 minus 3.

8. Amine nitrogen (this may also include nitrogen from pyridine type compounds) = procedure 2 minus 5.

9. Soluble nitrogen = procedure 2 plus 6.

Table 1.—Yield and Quality of Sugar Beet Roots as Affected by Various Treatments (1950).

Treatments		Net Water Applied	Yield T/A	Dry Matter Percent	Sucrose Percent	Purity Percent
Soil Moisture Condition						
Dry	W_1	16.3	14.74	22.90	16.05	88.38
Mod-dry	W_2	18.3	17.75	23.62	16.73	89.80
Mod-moist	W_3	26.3	19.20	23.91	17.01	90.28
Moist	W_4	50.9	18.38	23.99	17.06	90.80
s.e. of mean N = 32			.99	.21	.24	.48
Method						
Sprinkle		24.5	17.93	23.34	16.43	89.03
Furrow		31.4	17.10	23.87	16.99	90.60
L.S.D. @ .05			N.S.	0.36	0.36	0.71
Fertilizer						
Nitrogen		27.9	18.35	23.46	16.45	88.69
No Nitrogen		27.9	16.68	23.75	16.98	90.95
L.S.D. @ .05			0.69	0.22	0.04	0.35

Sugar and purity determinations were made by cold water extraction procedure.

Experimental Results

Data on yield and quality of sugar beets are presented in Table 1. It will be observed that yield, dry matter, sucrose, and purity are all affected to a significant extent by the variation in soil moisture condition used in the experiment. It will be noted that while yield is not affected significantly by method of irrigation the three quality factors are affected significantly. In agreement with work cited previously nitrogen fertilizers do affect the quality of the beets as well as the yield.

It will be well to keep in mind that quality (sucrose and purity) tends to increase as soil moisture is increased; that quality is higher under furrow than sprinkle irrigation; and that quality is lower under nitrogen fertilization than without it.

Table 2.—Nitrogen Constituents of Sugar Beet Roots as Affected by Various Treatments (1950).

Parts Per Million, Dry Basis

		Nitrogen Constituents in 2 Percent Acetic Acid Extract						Nitrogen in Whole Pulp	
Treatments		Nitrate	Ammonia	Amine	Amide	Organic	Soluble	Glutamine[1]	Total
Soil Moisture Condition:									
Dry	W_1	762	145	3089	796	3885	4789	1738	7968
Mod-dry	W_2	531	119	2676	685	3361	4006	1448	6932
Mod-moist	W_3	571	106	2525	645	3103	3787	1246	7169
Moist	W_4	521	82	2262	532	2776	3378	1018	6455
s.e. of mean n	32	91	16	193	N.S.	235	86	126	344
Method:									
Sprinkle		626	132	2868	724	3593	4343	1602	7505
Furrow		566	94	2409	605	2969	3637	1114	6757
L.S.D. @ .05		N.S.	24	284	N.S.	345	126	186	505
Fertilizer:									
Nitrogen		657	157	3194	799	3988	4804	1790	8067
No Nitrogen		535	69	2082	530	2575	3176	926	6195
L.S.D. @ .05		85	12	215	84	202	73	93	293

[1] Data used here by couresy of International Minerals and Chemical Corporation.

One should notice the general trends of various nitrogen constituents as affected by the irrigation and fertilizer treatments shown in Table 2, keeping in mind the trends in quality observed in Table 1. Here it is seen that all the nitrogen constituents with one minor exception are inversely related to the quality factors shown in Table 1. Also with the exception of amide as affected by irrigation regimes and nitrate nitrogen concentration as modified by method of irrigation, it will be noted that the treatments affect nitrogen content of beet roots significantly.

The data in Tables 1 and 2 mean that any essential change in irrigation or fertilizer practice may modify the nitrogen nutritional status of sugar beets with respect to all the nitrogen constituents, and further, that this change in nutritional status may be associated with changes in sugar beet quality.

The authors recognize the possibility of sugar beets absorbing part of their nitrogen as ammonia, nitrates, and even organic nitrogen from the soil, but assume with Nightingale (9) that under usual field conditions the principal source of nitrogen is nitrates. Apparently, when nitrates are plentiful in the substrate and concentrations become high in beet roots, the reduced form of inorganic nitrogen or ammonia increases correspondingly. When the ammonia concentration increases in the root the ammonia detoxication agents such as glutamine similarly increase. This statement is substantiated by the data in Table 2 and is in agreement with the findings of Vickery et al. (11).

It might be assumed from what has been said that the ratio of one nitrogen constituent in the sugar beet root to any other should be relatively constant. If this were true and if sugar beet quality were closely related to soluble nitrogen constituents, it may be possible to establish a relation of association between beet quality and any convenient nitrogen constituent. It does not appear possible from a cursory examination of the data in Tables 1 and 2 to determine which of the nitrogen constituents is most closely associated with or responsible for changes in sugar beet quality. This may be done by comparing the ratios of one constituent to another and by comparing this ratio with changes in quality observed in Table 1.

Although insoluble protein nitrogen is not listed in Table 2, it can be obtained by deducting soluble nitrogen in the acid extract from total

Table 3.—Ratios Among Some Nitrogen Constituents in Sugar Beet Roots as Affected by Various Treatments (1950).

Treatment		Protein / Soluble	Protein / Sol. Org.	Protein / Inorg.	Protein / Glutamine	Amine / Glutamine	Nitrate / Glutamine	Amide / Glutamine	Ammonia / Glutamine
Soil Moisture Condition									
Dry	W_1	.75	1.00	4.22	2.15	1.89	.472	.504	.082
Mod-dry	W_2	.92	1.09	7.04	3.08	2.24	.435	.541	.080
Mod-moist	W_3	1.01	1.22	7.20	3.57	2.46	.578	.611	.082
Moist	W_4	1.09	1.43	6.44	5.41	3.17	.819	.611	.101
s.e. of mean n = 32		.11	.18	.83	.73	.35	.11	N.S.	N.S.
Method									
Sprinkle		.87	1.09	5.83	2.68	2.09	.442	.473	.085
Furrow		1.01	1.28	6.62	4.41	2.78	.709	.661	.088
L.S.D. @ .05		N.S.	N.S.	N.S.	1.07	.52	.165	N.S.	N.S.
Fertilizer:									
Nitrogen		.78	.94	5.67	2.13	1.89	.377	.455	.084
No nitrogen		1.10	.143	6.78	4.06	2.99	.775	.679	.088
L.S.D. @ .05		.17	.27	N.S.	0.82	.35	.143	.104	N.S.

nitrogen. This is a relatively constant value of approximately 3140 p.p.m. under all conditions of the experiment. This form of nitrogen is relatively stable and it is probably the most satisfactory basis to use in studying the ratio changes in other constituents. Furthermore, since it is insoluble it probably has less influence on the quality factors of sucrose and purity than soluble constituents. Some of the ratios of protein to soluble nitrogen fractions are shown in Table 3. Although these ratios vary within relatively narrow limits they are regular and consistent with changes in sugar beet quality as affected by irrigation and fertilizer treatments (see Table 1). This increase in rotio with increase in quality indicates that the soluble constituents are varying inversely with changes in quality. Columns 5, 6, 7, and 8 in Table 3 indicate similarly that the concentration of glutamine in the beet root decreases more rapidly than does amine or amide nitrogen and that glutamine also decreases more rapidly than nitrate or ammonia nitrogen as quality increases. The ratio of ammonia to glutamine does not follow a consistent change as beet quality changes. This indicates a close relation between ammonia and glutamine concentrations in sugar beet roots.

If one were to decide from the data in Table 3 which of the nitrogen constituents in the sugar beet root were most closely associated with variations in quality, it appears that glutamine and ammonia nitrogen would be selected. In order to determine more specifically which of the nitrogen constituents were most closely associated with the quality of roots correlation coefficients were calculated. Simple correlations among the various nitrogen concentrations are shown in Table 4.

Table 4.—Simple Correlations Among Nitrogen Constituents of Sugar Beet Roots.

	Nitrate	Amine	Amide	Ammonia	Organic	Total	Total Soluble
Glutamine	.0956	.6337	.4173	.7284	.7758	.6271	.7547
Nitrate	-------	.1234	.0422	.0642	.1291	.1353	.3001
Amine	-------	-------	.1430	.4667	.9085	.4424	.8690
Amide	-------	-------	-------	.2678	.3589	.2184	.3459
Ammonia	-------	-------	-------		.5686	.5261	.5693
Organic	-------	-------	-------	-------	-------	.5137	.9518
Total	-------	-------	-------	-------	-------	-------	.5415
Total Soluble	-------	-------	-------	-------	-------	-------	-------

n = 128 Sig. @ .05 0.174
.01 0.228

The high correlations between soluble and organic as well as organic and amine is explainable on the ground that organic and amine constitute approximately eighty percent of the soluble and organic fractions respectively. Some of the other high correlations may be explained in a similar manner. The high correlation between glutamine and ammonia are associated on a different basis.

The data in Table 5 consist of simple regression coefficients indicating the association of change in nitrogen fractions with changes in yield and quality factors. There is a tendency for yields to increase as nitrogen constituents increase. The data show a highly significant inverse relation among quality factors and all nitrogen constituents.

Since it has been shown that all nitrogen constituents tend to vary inversely with quality of sugar beet roots when studied together, it was thought advisable to test the theoretical relation of each nitrogen constituent to quality, assuming that all other nitrogen constitutents were not permitted to vary. These partial regression coefficients are shown in Table 6. Under these theoretical conditions, it will be observed that glutamine is significantly related to dry matter and sucrose and approaching significance to purity. Ammonia nitrogen appears to be the only other fraction significantly related to quality.

The data in Table 7 were prepared in order to determine the percent of the total variation in quality in beet roots associated with nitrogen constituents. It will be seen that the largest variation in root quality associated with nitrogen composition is attributable to the presence of

Table 5.—Simple Regression Coefficients Relating Yield and Quality Factors to Nitrogen Fractions in Sugar Beet Roots.

Nitrogen Constituents p.p.m.	Yield T/A	Dry Matter Percent	Sucrose Percent	Purity Percent
Glutamine	+.0051	—.0070[2]	—.0091[2]	—.0289[2]
Nitrate	—.0023	—.0103[2]	—.0103[2]	—.0237[2]
Amine	+.0043	—.0031[2]	—.0047[2]	—.0162[2]
Amide	+.0029	—.0104[2]	—.0126[2]	—.0391[2]
Ammonia	+.0029	—.0611[2]	—.0792[2]	—.2241[2]
Organic	+.0034	—.0032[2]	—.0045[2]	—.0150[2]
Total	+.0036	—.0020[2]	—.0028[2]	—.0098[2]
Total Sol.	+.0025	—.0030[2]	—.0041[2]	—.0129[2]

[1] Significant beyond .05 level n = 128
[2] Significant beyond .01 level

Table 6.—Partial Regression Coefficients relating Yield, Quality Factors, and Nitrogen Constituents in Sugar Beet Roots.

P.P.M.	Yield T/A	Dry Matter Percent	Sucrose Percent	Purity Percent
Glutamine	+.0047	—.0078[1]	—.0067[2]	—.0024
Nitrate	—.0883	—.0119	—.0066	+.0024
Amine	—.0082	+.0023	+.0014	—.0040
Amide	—.0147	—.0009	—.0008	—.0069
Ammonia	—.2485[1]	—.0326	—.0348[1]	+.0163
Organic	—.0642[2]	—.0039	—.0017	+.0085
Total	+.0065	+.0012	+.0009	—.0006
Total Sol.	+.0775	+.0038	+.0008	—.0068
R^2	.094[2]	.323[2]	.559[2]	.783[2]

n = 128 [1] Sig @ .05
[2] Sig. @ .01

glutamine. Associated with this constituent are 23 percent of the variation in dry matter, 50 percent of the sucrose, and 77 percent of the purity. While ammonia accounts for a high percent of variation, it is less closely associated with quality than is glutamine nitrogen. A comparison of variations in quality associated with all nitrogen constituents (R^2), in Table 7, shows that glutamine is associated with or can account for 73 percent of the total variation in dry matter, 89 percent in sucrose, and 98 percent in purity. It may be interesting to note that nitrate nitrogen is associated with a relatively low percentage of the total variation.

Table 7.—Simple Correlation Coefficients Relating Yield and Quality Factors to Nitrogen Fractions of Sugar Beet Roots (r^2).

P.P.M.	Yield T/A	Dry Matter Percent	Sucrose Percent	Purity Percent
Glutamine	.009	(—).235	(—).497	(—).769
Nitrate	(—).000	(—).117	(—).147	(—).119
Amine	.015	(—).102	(—).297	(—).541
Amide	.001	(—).134	(—).247	(—).361
Ammonia	.000	(—).210	(—).446	(—).545
Organic	.013	(—).153	(—).389	(—).651
Total	.027	(—).116	(—).285	(—).516
Total Sol.	.010	(—).189	(—).429	(—).654
R^2	.094[2]	.323[2]	.559[2]	.783[2]

n = 128 Sig. @ .05 0.0303
.01 0.0519

Summary and Conclusions

Numerous observations have shown that the problem of nitrogen fertilization is a prominent one in the sugar beet growing areas of the United States. Yields and quality of sugar beets are so markedly affected by the quantity of available soil and fertilizer nitrogen that neither a deficiency nor a large excess should be tolerated.

Although factors of climate and soil, other than nitrogen, may affect the quality of sugar beet roots, it cannot be denied that nitrogen fertilization and nitrogen plant composition are closely associated with sucrose storage in beet roots and sugar recoveries from extract juice.

The authors do not contend that nitrogen constituents per se prevent sugar crystallization, but there is strong evidence that soluble nitrogen constituents of the sugar beet root are highly associated with, if not responsible for, variations in purity and sucrose percentage as well as dry matter percentage.

The particular nitrogen components which appear to be most highly related to changes in sugar beet quality are the glutamine and ammonia fractions. It is the opinion of the authors that glutamine nitrogen is of greatest significance in quality variation, not only because of its high association with quality factors, but also because the concentration of nitrogen in this form is ten times that of ammonia nitrogen.

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