AMMONIUM AND NITRATE AS SOURCES OF NITROGEN FOR SUGARBEETS

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INTRODUCTION

The reduction of nitrate to ammonium and the coupling of ammonium to carbon structures to form amino acids for protein synthesis are energy consuming processes (1) essential for the growth and development of the sugarbeet. If the sugarbeet plant could utilize ammonium exclusively, it would eliminate the large energy-requiring, sugar-using step of nitrate reduction, and the sugar saved could be used for plant growth or storage and thereby increase sugar production. Assuming this could be done, the oxidation of ammonium to nitrate in the soil would have to be prevented or at least reduced greatly. Furthermore, the rate of ammonium absorption would have to equal its rate of utilization or it would have to be stored, like nitrate, without toxicity until metabolized within the plant (3, 4, 9, 11, 13).

A comparison of NH_4^+ and of NO_3^- can be made with plants in soil under field or pot conditions with and without a nitrification inhibitor (e.g. with nitrapyrin) (5, 10). This approach has the advantage that the results will provide a practical answer to a practical problem. It will, however, fail to answer the question of whether NH_4^+ or nitrate are utilized preferentially for plant growth, for even with a nitrification inhibitor, NH_4^+ is oxidized in the soil to nitrate at an uncontrolled rate dependent upon the temperature, pH, moisture, and oxygen level of the soil.

An alternative technique is to grow plants in a complete nutrient solution with either ammonium or nitrate as the source of nitrogen. Again complications may arise. The absorption of NH_4^+ from a nutrient solution usually results in an increase in acidity,

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since ammonium is not absorbed simultaneously with an anion and the ionic balance of the nutrient solution is not maintained. Cations other than H^+ are not ordinarily exchanged for NH_4^+ and damage from NH_4^+ can thus be complicated by an increase in acidity of the nutrient solution. To prevent this acidification, alkali can be added and recorded automatically to a preset pH (2), or powdered $CaCO_3$ can be added to the nutrient solution to maintain the pH at 6.5. At this pH, there is no detectable loss of NH_3 by aeration (8). Iron chlorosis, which occurs for sugarbeets in solutions at pH 7.0 and above with nitrate as a nitrogen source can be prevented by the use of Fe-EDTA and adjusting the pH to 6.5 with 1.0 N H_2SO_4 as required.

The objective of this research was to determine the effects of ammonium and of nitrate, singly and in combination, on the growth, sucrose concentration, sucrose yield, and juice purity of sugarbeets when grown with calcium carbonate in a complete nutrient solution in a controlled climate under plant growth chamber conditions.

MATERIALS AND METHODS

Sugarbeet seed (Beta vulgaris L. cv. F 58-544 Hl) was planted in vermiculite and watered daily with one-half strength modified Hoagland's nutrient solution, H/2, (solution C, next page) for a 2-week period. Germination and growth of the experimental plants took place in smog-free air in growth chambers set at 25°C for photoperiods of 16 hours at an illumination of 43,000 lux (4,000 ft-c, 650 $\mu E\ m^{-2}\ s^{-1}$, over a wavelength of 400-700 nm) emitted by a combination of 30, 2.4m, 215-watt, cool-white fluorescent lamps, four 1.22m, 100-watt fluorescent lamps, and 22, 60-watt incandescent lamps (6). Shading of leaves by plants in adjacent pots was kept at a minimum by re-spacing the pots equidistant after each harvest.

Two weeks after planting, the seedlings were at the late cotyledon to early one-leaf stage. These were selected as to size and quality, wrapped around at the hypocotyl with non-absorbent cotton wool and then set upright into a one-hole cork ring and floated in trays containing distilled water. They were transferred at random several hours later to covered, aerated 20-liter pots

filled with modified H/2 nutrient solution, prepared to contain 7.5 me l^{-1} of nitrate from calcium nitrate as the sole source of nitrogen. Each cover held three seedlings. After 10 days, they were thinned to one seedling per plot. The pH value of the solution for the first 6 weeks of growth was adjusted as required with NaOH or H_2SO_4 to pH 6.0.

Six weeks from planting (4 weeks from transplanting) one set of plants was harvested, and the nutrient solutions in the remaining sets were replaced by a modified half strength Hoagland's solution (see below) containing one of the following nitrogen sources: a. minus N, b. 3.75 mmole l^{-1} (millimoles per liter) of ammonium sulfate, c. 3.75 mmole l^{-1} of calcium nitrate, and d. 3.75 mmole l^{-1} of ammonium nitrate. To minimize the possible toxic effect of ammonium, it was added at 1/8 H strength (0.94 mmole l^{-1}) initially, followed by 1/8 and 1/4 additions 4 and 5 days later, respectively, to give a total addition of 3.75 mmole l^{-1} for each nitrogen source.

The modified half strength Hoagland's solution also contained the following salts, expressed in mmole l^{-1}: 1.0 KH_2PO_4, 0.5 $MgSO_4 \cdot 7H_2O$, 1.0 NaCl, 1.5 K_2SO_4. Micronutrients, expressed in mg l^{-1}, were: 2.5 Fe as ferric ammonium ethylene diamine tetraacetate complex 0.25 B as H_3BO_3; 0.25 Mn as $MnSO_4 \cdot 4H_2O$; 0.25 Zn as $ZnSO_4 \cdot 7H_2O$; 0.01 Cu as $CuSO_4 \cdot 5H_2O$; and 0.005 Mo as MoO_3. Calcium was supplied from 3.75 mmole l^{-1} of $Ca(NO_3)_2$ for the nitrate series and from 2.5 mmole l^{-1} of $CaSO_4 \cdot 2H_2O$ for the $(NH_4)_2SO_4$ and NH_4NO_3 series. Also, two teaspoonfuls of powdered $CaCO_3$ (5-6 g or 0.25 g l^{-1}) were added to all pots to maintain the pH at about 6.5. Consequently, the calcium in solution with nitrate as the sole source of nitrogen was somewhat higher initially in calcium (3.75 versus 2.5 mmole l^{-1}) and somewhat lower in sulfate (2.0 versus 4.5 mmole l^{-1}) than for the solutions containing $(NH_4)_2SO_4$ or NH_4NO_3 as a nitrogen source. Later, these solutions, because of the uptake of NH_4^+, were higher in calcium than with nitrate alone. The small differences in calcium or sulfate, however, do not effect plant growth.

All solutions were aerated continuously with smogfree carbon-filtered air. Distilled water was added as needed. All treatments

were arranged in a randomized complete block design and were replicated three times for the 6, 9 and 12 week harvests and four times for the 15 week harvest. At the 9 and 12 week harvest intervals, the stock solutions and the required nitrogen salts were added at the H/2 level to the solutions of the remaining pots.

Until the first harvest, all nutrient solutions were adjusted to pH 6.0 with NaOH or H_2SO_4 and thereafter they were buffered at about pH 6.5 by the addition of more $CaCO_3$ powder or H_2SO_4 as required. At each harvest the plants were separated into tops, fibrous roots and storage roots. Fresh weights were recorded immediately and then dry weights determined following a 3-day drying period in a forced draft at 80°C. Brei from the storage roots was frozen with dry ice and analyzed later by standard commercial procedures for sucrose and juice purity by the Spreckels Sugar Company, Woodland, California. Nitrate and ammonia electrodes (7) were used for the determination of nitrate and ammonium in the nutrient solutions and in dried, ground plant material.

RESULTS AND DISCUSSION

GROWTH

The plants reached the late cotyledon, early two-leaf stage of development 2 weeks after seeding in vermiculite, with nitrate as the sole source of nitrogen. The seedlings after transplanting to the 20-liter pots reached the 4 to 5 leaf stage of development 10 days later. These plants were thinned from 3 to 1 plant per pot. Eighteen days later, the plants, now six weeks from planting, had developed 15 to 17 leaves. The harvest results for this set of plants and for three other sets, harvested at three week intervals, are given in Tables 1 to 3 and Figures 1 to 4.

The results show that the sugarbeet plants grew much better with nitrate than with ammonium even though special precautions were taken to prevent possible harmful effects of ammonium on plant growth (Tables 1A and 1B and Figures 1A and 1B). These precautions included the addition of powdered calcium carbonate to prevent the acidification of the culture solution, the addition of ammonium in small doses over a 9-day period and by adding

Table 1. Influence of ammonium and nitrate on sugarbeets.**

Age	Deficient Series				Sufficient Series			LSD
Wks*	N_o	NH_4-N_D	NO_3-N_D	AN-N_D	NH_4-N_S	NO_3-N_S	AN-N_S	5%#
			A. Total weight, g/plant, fresh basis					
6	466	466	466	466	466	466	466	---
9	820a	940a	1560b	1300b	940a	1560b	1300b	290
12	770a	1270bc	1530c	1510c	1030ab	2680e	2360d	290
15	-	1130a	1250a	1100a	1690b	3490d	2670c	200
			B. Total weight, g/plant, dry basis					
6	37	37	37	37	37	37	37	---
9	105ab	85a	139b	128b	85a	139b	128b	35
12	150a	203bc	200b	233bcd	146a	271d	244cd	40
15	-	248a	289a	264a	250a	583c	466d	40
			C. Sucrose, percent, fresh basis					
6	5.4+	5.4	5.4	5.4	5.4	5.4	5.4	---
9	10.6c	7.1ab	6.6a	7.9b	7.1ab	6.6a	7.9b	1.0
12	13.5c	11.8bc	10.5b	11.4b	8.2a	7.4a	7.1a	1.9
15	-	12.3c	12.8c	12.9c	7.3a	10.9b	10.5b	1.2
			D. Purity, percent					
9	79	75	76	74	75	76	74	NS
12	83.9c	83.3c	83.1c	83.9c	71.6c	80.2bc	74.9ab	5.6
15	-	83.0b	84.6b	83.2b	72.9a	82.7b	81.3b	3.4
			E. Petiole nitrate-N, µg/g, dry basis					
6	21000	21000	21000	21000	21000	21000	21000	---
9	1200a	3500b	6800c	6300c	3500b	6800c	6300c	#
12	390a	320a	1500c	800b	840b	11200d	9400d	#
15	-	370a	290a	270a	6200c	950d	870b	#
			F. Blade ammonium-N, µg/g, dry basis					
6	200	200	200	200	200	200	200	---
9	270a	920b	180a	340a	920b	180a	340a	#
12	280a	390a	230a	230a	2730c	230a	560b	#
15	-	240a	190a	200a	860b	190a	210a	#

*Age in weeks from planting.

**All plants received nitrate-N as the only source of nitrogen for the first six weeks of growth. At six weeks the nutrient solutions were changed to a minus-N, half-strength modified Hoagland's solution, plus the different nitrogen treatments. At the 9- and 12-week harvests, the remaining plants in the N_D series received minus-N stock solution only, and in the N_S series the various nitrogen treatments were included.

#Mean separations within a row are made by Duncan's multiple-range test, 5% level, except for Tables E and F, which differ greatly as indicated.

+Calculated $\hat{Y} = -0.1935 + 0.6034X = 5.4\%$ sucrose when $X = 9.2\%$ dry weight.

Table 2. Influence of ammonium and nitrate on sugarbeets.*

Age Wks	Deficient Series				Sufficient Series			LSD 5%
	N_o	NH_4-N_D	NO_3-N_D	AN-N_D	NH_4-N_S	NO_3-N_S	AN-N_S	
	G. Storage root, g/root, fresh basis							
6	101	101	101	101	101	101	101	---
9	267	276	371	331	276	371	331	NS
12	393a	583b	546ab	618b	468ab	887c	792c	161
15	–	720a	789a	740a	746a	1731c	1475b	171
	H. Sucrose, g/root							
6	5	5	5	5	5	5	5	---
9	29	20	25	26	20	25	26	NS
12	53ab	69b	57ab	72b	38a	66b	56ab	20
15	–	89b	102b	96b	55a	189d	154c	24
	I. Tops, g/plant, fresh basis							
6	323	323	323	323	323	323	323	---
9	469a	594a	1070b	863b	594a	1070b	863b	241
12	281a	556bc	881d	774cd	460ab	1619e	1402e	232
15	–	299a	313a	243a	805b	1477c	998b	273
	J. Tops, g/plant, dry basis							
6	25	25	25	25	25	25	25	---
9	51a	45a	80b	73b	45a	80b	73b	15
12	55a	89bc	102c	98c	74ab	147d	131d	23
15	–	79a	99ab	84a	110b	216d	166c	23
	K. Storage root, g/root, dry basis							
6	9.2	9.2	9.2	9.2	9.2	9.2	9.2	---
9	46.4	35.8	47.5	45.6	35.8	47.5	45.6	NS
12	82ab	100b	86ab	113b	61a	103b	97b	30
15	–	152ab	171b	164b	119a	326d	271c	43
	L. Fibrous roots, g/plant, dry basis							
6	3.1	3.1	3.1	3.1	3.1	3.1	3.1	---
9	7.4ab	5.0a	11.8c	9.0bc	5.0a	11.8c	9.0bc	2.8
12	13.3ab	13.2ab	12.1a	12.4a	13.2ab	20.6c	16.8bc	4.1
15	–	16.5a	18.8a	18.8a	21.5a	41.1c	29.3b	5.3
	M. Tops, percent of total plant, fresh basis							
6	69	69	69	69	69	69	69	---
9	58	64	68	66	64	68	66	NS
12	36a	44ab	58c	52bc	43ab	60c	60c	10.6
15	–	26ab	25ab	22a	47c	42c	37bc	13.3

*See footnote for Table 1.

the ammonium when the plants were relatively large at the 15-17 leaf stage of development. In spite of these precautions the total growth with ammonium after 3 weeks was only 60% of that with nitrate (Tables 1A and 1B). Toxicity from ammonium was

also observed in the nitrogen-sufficient plants at the 12- and 15-week harvests (Tables 1A and 1B and Figures 1A and 1B).

In the nitrogen-deficient series, the initial toxicity from ammonium disappeared as the plants grew in size and depleted the

Table 3. Influence of ammonium and nitrate on sugarbeets.*

Age	Deficient Series				Sufficient Series			LSD
Wks	N_O	NH_4-N_D	NO_3-N_D	AN-N_D	NH_4-N_S	NO_3-N_S	AN-N_S	5%
	N. Storage roots, percent of total weight, fresh basis							
6	21.8	21.8	21.8	21.8	21.8	21.8	21.8	---
9	31.8	29.1	24.2	25.4	29.1	24.2	25.4	NS
12	51.1d	46.0cd	35.5ab	41.0abc	46.3cd	33.4a	33.2a	9.5
15	-	64.0b	62.9b	67.4b	44.6a	49.6a	55.4ab	12.3
	O. Fibrous roots, percent of total weight, fresh basis							
6	9.1	9.1	9.1	9.1	9.1	9.1	9.1	---
9	10.3b	7.1a	7.5a	8.4ab	7.1a	7.5a	8.4ab	2.4
12	12.4b	10.0ab	6.9a	7.6a	10.1ab	6.3a	7.2a	3.5
15	-	10.1abc	12.0c	10.6bc	8.1ab	8.0ab	7.5a	2.6
	P. Tops, percent of total plant, dry basis							
6	67	67	67	67	67	67	67	---
9	50	53	57	57	53	57	57	NS
12	37a	44ab	51bc	45ab	50bc	54c	54c	7.6
15	-	32a	35a	33a	44b	37a	36a	6.9
	Q. Storage root, percent of total weight, dry basis							
6	25	25	25	25	25	25	25	---
9	43b	41ab	34a	35ab	41ab	34a	35a	8
12	54c	50bc	43abc	50bc	42ab	38a	39ab	10
15	-	61b	59b	61b	47a	56b	58b	8
	R. Fibrous roots, percent of total weight, dry basis							
6	8.2	8.2	8.2	8.2	8.2	8.2	8.2	---
9	7.2	6.4	8.4	7.1	6.4	8.4	7.1	NS
12	9.0c	6.6ab	6.1a	5.6a	8.9bc	7.6abc	6.9abc	2.3
15	-	6.6	6.5	7.3	8.7	7.0	6.4	NS
	S. Blades, g/plant, dry basis							
6	15.7	15.7	15.7	15.7	15.7	15.7	15.7	---
9	28.0a	23.7a	38.3b	37.0b	23.7a	38.3b	37.0b	8.2
12	30.1a	46.8b	43.9ab	43.2ab	38.9ab	66.1c	60.6c	13.7
15	-	41.2a	53.1b	41.9a	54.1b	89.7d	73.3c	8.9
	T. Storage root (brei), percent dry weight							
6	9.2	9.2	9.2	9.2	9.2	9.2	9.2	---
9	17.2b	12.9a	12.8a	13.7a	12.9a	12.8a	13.7a	2.2
12	20.6d	17.2c	15.9bc	18.0cd	13.2ab	11.6a	12.3a	3.2
15	-	21.1c	21.6c	22.1c	15.9a	18.9b	18.3b	2.1

*See footnote for Table 1.

nitrogen supply of the culture solution (Tables 1A and 1B). Under these conditions nitrogen became limiting and all sources of nitrogen gave approximately the same total yield even though ammonium had been toxic initially (Figures 1A and 1B). Further-

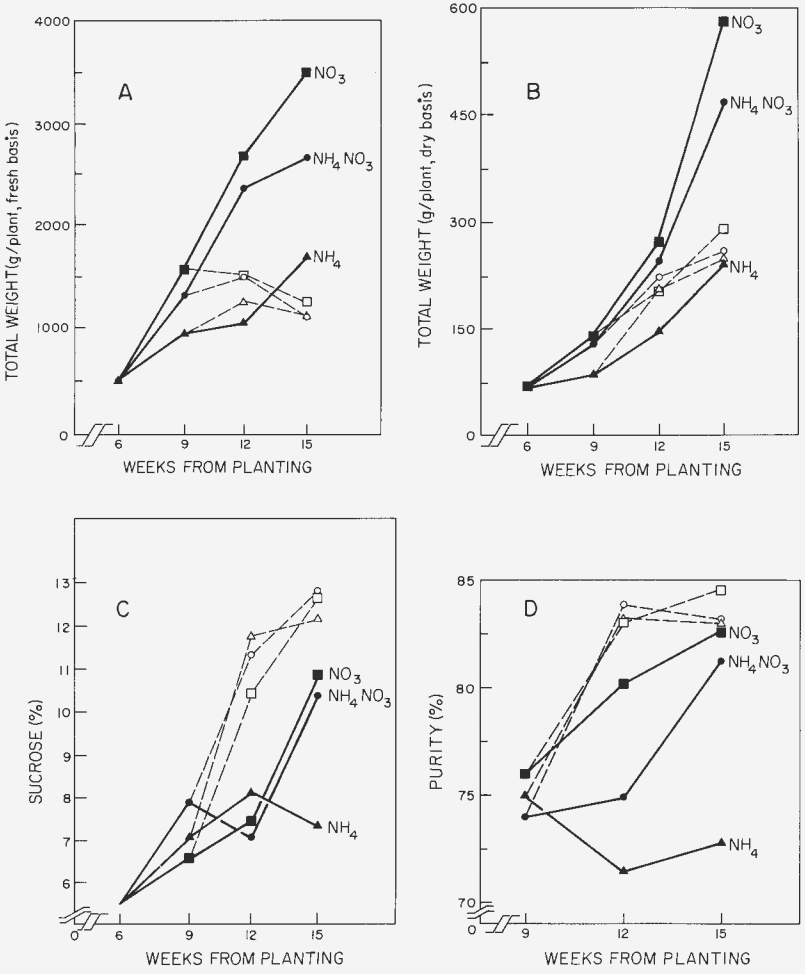

Figure 1. Influence of nitrogen source and time under nitrogen-sufficient (solid line) and nitrogen-deficient (broken line) conditions on fresh weight of plant (A); on dry weight of plant (B); on percent sucrose of storage root (C); and on percent purity of thin juice from brei of storage root (D); starting with nitrogen treatments six weeks from planting. Percent sucrose for 6 weeks was calculated from the percent dry weight of brei. See footnote Table 1.

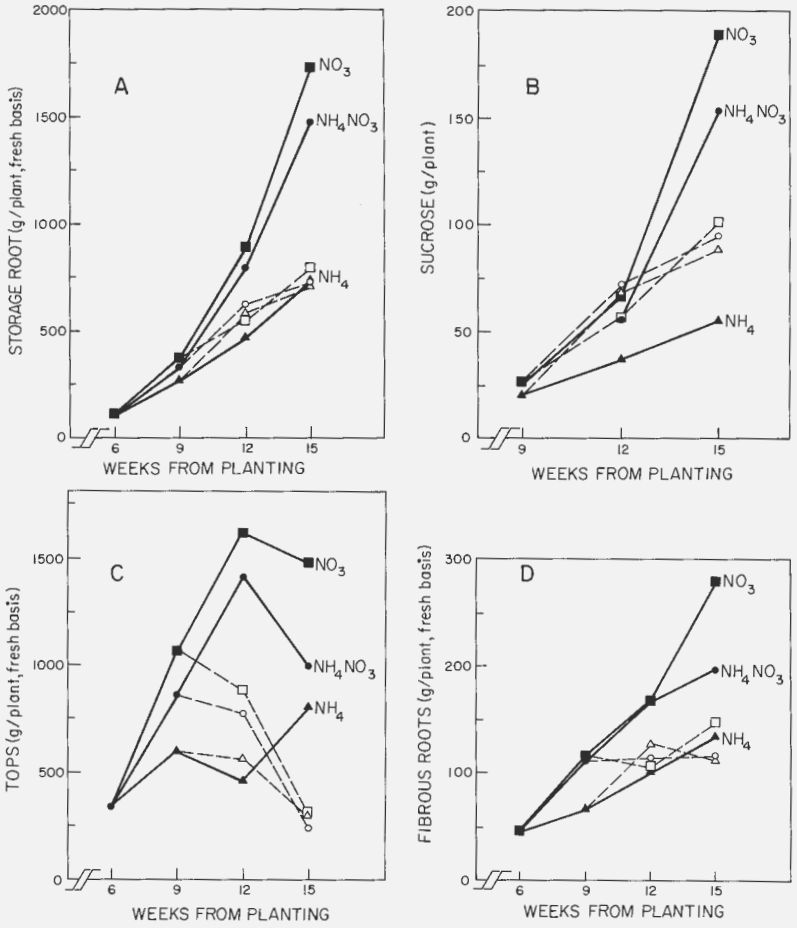

Figure 2. Influence of nitrogen source and time under nitrogen-sufficient (solid line) and nitrogen-deficient (broken line) conditions on storage root weight (A); on sucrose in storage roots (B); on fresh weight of tops (C); and on fresh weight of fibrous roots (D). Note the depressing effect of ammonium on fibrous root development 9 weeks from planting.

more, with prolonged nitrogen depletion, neither the sucrose concentrations nor the purities of the storage roots differed significantly at 15 weeks (Tables 1C and 1D, Figures 1C and 1D), indicating within the accuracy of the experiment that ammonium and nitrate were equally efficient as a nitrogen source, providing sufficient time had elapsed to overcome the initial toxicity of ammonium.

In the nitrogen-sufficient series all plants contained much unused nitrogen as measured either by petiole nitrate-N or blade ammonium-N (Tables 1E and 1F). At 15 weeks the storage roots were low in sucrose concentration and purity, with the lowest values occurring with ammonium as the sole source of nitrogen (Tables 1C and 1D). Here, root size was also the smallest (Table 2G and Figure 2A), and concequently the sucrose produced was still smaller (Table 2H and Figure 2B). Apparently ammonium is harmful to the sugarbeet plant, unless it becomes depleted or until nitrate becomes available to the plant either directly by absorption, or indirectly by the oxidation of ammonium to nitrate within the plant. Also, it appears that the mere presence of nitrate within the plant largely prevents the toxic effect of ammonia. This inter-relationship of ammonium and of nitrate may be seen in Tables 2G, 2I, 2J, 2K, 2L and 3S.

With ammonium the growth of tops, blades, storage root and fibrous roots was depressed, even though moisture and nutrients were readily available to the plants for growth. Among the plant parts, the decrease in fibrous root growth (Table 2L) could be

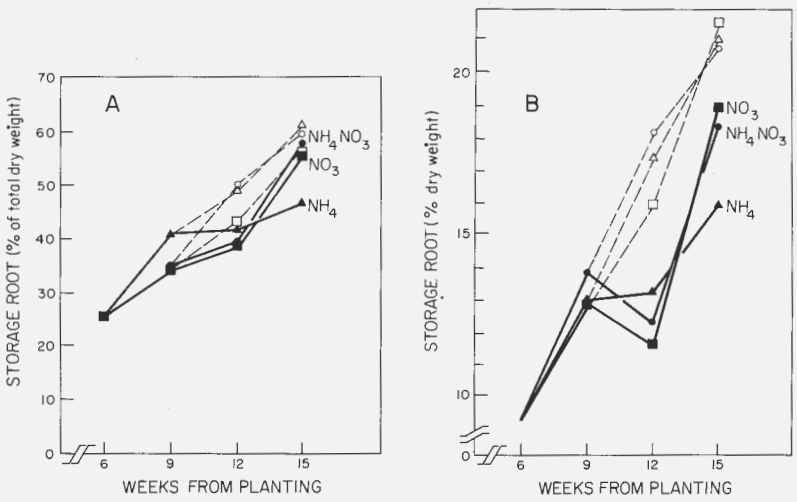

Figure 3. Influence of nitrogen source and time under nitrogen-sufficient (solid line) and nitrogen-deficient (broken line) conditions on dry weight of storage root as percent of total dry weight (A) and on percent dry weight of storage root brei (B).

serious under certain field conditions. On soils relatively low in nutrients and water supplying power, root damage could induce a nutrient deficiency during periods of rapid growth or could cause wilting during periods of high transpiration or drought, which would not ordinarily occur with undamaged fibrous roots.

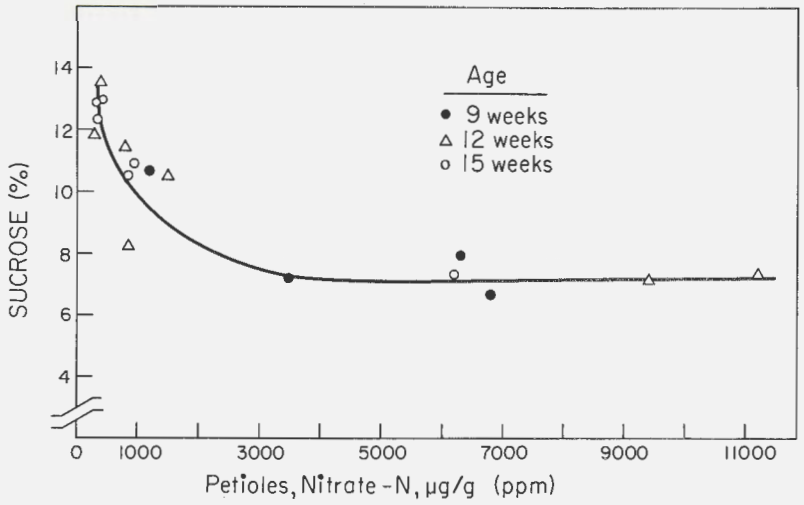

Figure 4. Relationship of petiole nitrate-N of young mature leaves to storage root sucrose concentration. Increases in sucrose concentration begin at a petiole nitrate-N concentration of about 3,000 ppm (dry basis), followed by rapid increases below 1,000 ppm, the critical petiole nitrate-N concentration.

PARTITIONING

The 6-week old plants averaged 466 and 37 g per plant on a fresh and dry weight basis, respectively (Tables 1A and 1B). Partitioned, the tops, storage roots and fibrous roots made up 69, 22 and 9% of the total fresh weight of the plant (Tables 2M, 3N, 3O). On a dry weight basis the corresponding values were 67, 25 and 8% (Tables 3P, 3Q, 3R). These percentages differed very little from those for fresh weight. During the next 9 weeks of growth with nitrate as the source of nitrogen the proportion of tops on a fresh weight basis decreased gradually from 69% to 25 and 42% for the low and high nitrogen plants, respectively (Table 2M), and on a dry weight basis, from 67% to 35 and 37% (Table 3P).

Simultaneously, the proportion of storage root increased on a fresh weight basis from 22% to 63 and 50% for the low and high nitrogen plants, respectively (Table 3N). On a dry weight basis the storage root values increased from 25 to approximately 57% for both the low and high nitrogen plants (Table 3Q and Figure 3A). These distributions of biomass were not affected consistently by substituting NH_4^+ for NO_3^- in the culture solution, starting when the plants were 6 weeks old (Table 2M-3R).

QUALITY

Quality, as measured by the sucrose concentration and percent purity of the storage root, was not affected primarily by the nitrogen source, either under low or high nitrogen conditions (Tables 1C and 1D, Figures 1C and 1D). Quality, however, was greatly affected by the nitrogen status of the plant. Since ammonium retards growth (Figures 1A and 1B), such plants remained higher in nitrogen longer and were lower in sucrose concentration and purity than those on nitrate alone (Figures 1C and 1D). Thus, quality was very low (Tables 1C and 1D) when either the petioles were high in nitrate-N (more than 1000 ppm) or the blades high in ammonium-N or the brei low in percent dry weight (Tables 1E, 1F and 3T and Figure 3B). Conversely, quality was high when the petioles were low in nitrate-N (less than 1000 $\mu g\ g^{-1}$, Figure 4), or low in blade ammonium-N (Table 1F) or high in brei dry weight percentage (Table 3T). For example, at the second harvest, when the plants were 9 weeks old, the petioles of the N_o treatment contained only 12000 $\mu g\ g^{-1}$ of nitrate-N (Table 1E), the blades had only 270 $\mu g\ g^{-1}$ of ammonium-N (Table 1F) and the brei had a high dry weight value of 17.2% (Table 3T), the sucrose concentration of these beets was 10.6% (Table 1C). The corresponding plants treated with nitrate contained 6800 $\mu g\ g^{-1}$ of petiole nitrate-N (Table 1E). These had only 6.6% of sucrose (Table 1C) and a low brei dry weight value of 12.8% (Table 3T). Similar relationships were observed with ammonium.

At the 12-week harvest the petiole nitrate-N values for the N_o treatment decreased from 1200 $\mu g\ g^{-1}$ to 390 $\mu g\ g^{-1}$ and for the NO_3-N_D and NH_4NO_3-N_D treatments the values decreased from 6800 $\mu g\ g^{-1}$ and 6300 $\mu g\ g^{-1}$ to 1500 $\mu g\ g^{-1}$ and 800 $\mu g\ g^{-1}$, respec-

tively (Table 1E). The corresponding sucrose concentration for the N_o treatment increased from 10.6 to 13.5% and for the NO_3-N_D and NH_4NO_3-N_D treatments they increased from 6.6 and 7.9% to 10.5 and 11.4%, whereas the values for the nitrogen sufficient plants remained around 7.3%. The storage roots for plants with the highest petiole nitrate-N value of 1500 $\mu g\ g^{-1}$ in the nitrogen-deficient series at 12 weeks (Table 1E) also had the lowest sucrose concentration, 10.5% (Table 1C), and the lowest brei dry weight, 15.9% (Table 3T).

Even though the nitrogen-sufficient beets were much lower in sucrose concentration and in purity, the storage were much larger in size and consequently they produced much more sugar than those for the nitrogen-deficient series. In terms of nitrogen source, the beets from nitrogen-sufficient plants with nitrate were consistently higher in sucrose produced (Figure 2B) and in purity (Figure 1D) than those with ammonium alone or in combination with ammonium and nitrate. The marked increases in sucrose concentration and purity of the beets at the 12-week harvest (Tables 1C and 1D, Figures 1C and 1D) in the nitrogen-sufficient plants treated with nitrate or ammonium nitrate are due to a marked decline in petiole nitrate-N values (Table 1E), which occurred even though more nitrogen had been added 3 weeks earlier. Without a nitrate-N analysis of the petioles, the cause of the marked improvement in beet quality would not have been known.

At 15-weeks a pronounced increase in brei percent dry weight took place, which paralleled the increase in percent sucrose of the storage root. The regression equation, based on 57 pairs of values for harvests 2, 3 and 4 is $\hat{Y} = -0.194 + 0.603X$ with a coefficient of determination, r^2, equal to 0.827. The estimated percent sucrose concentration ($\hat{Y}$) for a 9.2% dry weight value (X) of the brei at 6 weeks (Table 3T) is 5.4% (Table 1C).

PETIOLE NITRATE-N

Petioles from the ammonium treated plants contained nitrate at a fairly high concentration 3 weeks after treatment, but the values were much lower than those treated with nitrate (Table 1E). The source of this nitrate in the ammonium plants could be from

the residual nitrate absorbed during the first 6 weeks of growth but thereafter the nitrate could have come from the nitrification of ammonium in the nutrient solution or the oxidation of ammonium to nitrate within the plant. This point needs further study.

At 6 weeks, the plants were clearly well supplied with nitrogen as indicated by the presence of 21,000 $\mu g\ g^{-1}$ of nitrate-N in the mature petioles, dry basis. Nevertheless, it was believed the plants would soon become deficient in nitrogen if the supply of nitrogen was not replenished. Three weeks later, the petioles of the plants without further nitrate addition contained only 1200 $\mu g\ g^{-1}$ of nitrate-N, whereas those with nitrate added contained 6800 $\mu g\ g^{-1}$ (Table 1E). These changes in petiole nitrate-N, as mentioned earlier were also related to the sucrose concentration of the storage roots (Table 1C and Figure 1C). With low petiole nitrate-N, the sucrose concentrations increased whereas those with ample nitrate-N (more than 1000 $\mu g\ g^{-1}$) the sucrose concentrations decreased rapidly (Figure 4). In terms of sugar recovery, nitrogen-deficient beets are undoubtedly superior to nitrogen-sufficient beets, at least until the increase in sugar concentration and purity fail to compensate for the loss in beet yield associated with nitrogen deficiency.

BLADE AMMONIUM-N

Mature blades of sugarbeets at the 15 to 17 leaf stage of development contained 200 $\mu g\ g^{-1}$ of ammonium-N (dry weight basis) even though the plants were exclusively on a nitrate regime during the first 6 weeks of growth (Table 1F). The ammonium-N present was apparently within the normal background range of 180 to 300 $\mu g\ g^{-1}$ for older plants fertilized with nitrate only. Three weeks after replacing nitrate by ammonium, the blades contained 920 $\mu g\ g^{-1}$ of ammonium-N, whereas those exclusively on nitrate-N were still low at only 180 $\mu g\ g^{-1}$. In replacing nitrate by ammonium nitrate, the ammonium-N value of the blades was reduced to 340 $\mu g\ g^{-1}$. In the 12-week harvest, the comparable values for ammonium-N in the nitrogen-deficient series were 390, 230 and 230 $\mu g\ g^{-1}$ with the highest value of 390 for the earlier ammonium ion treatment. However, in the nitrogen-sufficient series, the the blades of comparable plants contained 2730, 230 and 560 $\mu g\ g^{-1}$

of ammonium-N, with the highest value associated with the ammonium sulfate treatment. In the 15-week harvest only the plants treated with ammonium-N were above the background values of 180 to 300 μg g^{-1}. These had a value of 860 μg g^{-1}.

AMMONIUM-N VS. NITRATE-N

The absorption of ammonium by roots requires either an exchange of a cation or the simultaneous absorption of an anion to maintain the ionic balance in the culture solution and in the plant (11). Hydrogen as a proton (H^+) is apparently involved in the cation exchange process since the culture solution becomes acid as the ammonium is absorbed by the plant roots. In the presence of ammonium, but in the absence of nitrate, the pH values of the nutrient solution frequently fall as low as 3.0. At pH 4.0 there is a detrimental effect on sugarbeet growth as shown by Ulrich and Ohki (12), using nitrate as the source of nitrogen in nutrient solutions maintained at pH 4.0 with sulfuric acid. Best growth was obtained at pH 7.0, with slight reductions at either 6.0 or 8.0. In the present study, acidification was prevented with calcium carbonate as a pH-stat at about 6.5, but nevertheless the growth with ammonium was not nearly as good as with nitrate. Thus, it appears that the toxicity from ammonium is primarily internal and not external.

Internally, ammonium toxicity would most likely occur whenever the carbon structures (carboxylates) fail to combine with the unutilized ammonium in the plant cells. Evidently when ammonium accumulated and was stored in the plant tissues even at a fairly low concentration (Table 1F), damage occurred, whereas when nitrate accumulated and was stored at a fairly high concentration (Table 1E) there was no apparent damage to the plant (Tables 1B and 2G).

The absorption and utilization of ammonium or of nitrate by the sugarbeet plant is essentially a two-step process. One is the actual absorption of the nutrient from the soil by the fibrous roots and the other, after translocation, is its utilization within the plant. In the absence of nitrate the solutions become acid, with the pH decreasing quickly to 3.0 for rapidly growing plants. Injury from acidity was avoided in the present study by adding

powdered calcium carbonate to the culture solution. However, this corrective measure was apparently insufficient to prevent injury from the use of ammonium ions in the culture solution. Apparently, the carbohydrate supply for amino acid and organic acid formation, the source of the protons causing the increased acidity of the nutrient solution, became depleted, and therefore ammonium accumulated to upset the ionic or enzymatic balance of the plant cells. However, a small amount of nitrate within the plant, absorbed either directly from the nutrient solution or produced enzymatically within the plant, had a large ameliorating influence on the harmful effect of ammonium on plant growth. Results of this study suggest that nitrate is a "safening" agent for ammonium in sugarbeet plant growth--clearly a point in need of further study in sugarbeet nitrogen nutrition on poorly drained soils.

SUMMARY

Sugarbeet plants were grown for 6 weeks in plant growth chambers from seed to the 15 to 17 leaf stage in half strength modified Hoagland's nutrient solution containing nitrate as the sole source of nitrogen. At this time, one set of plants was harvested and the nutrient solution in the remaining pots was replaced by one containing nitrogen as follows: a. minus N, b. ammonium sulfate, c. calcium nitrate, and d. ammonium nitrate. Comparable plants were harvested at 9, 12 and 15 weeks. After each harvest the nitrogen-sufficient plants were re-fertilized with nitrogen, the others in the nitrogen-deficient series were not.

Three weeks after treatment, the plants with only ammonium were half the size of those with nitrate. This damage occurred even though acidification of the culture solution was prevented by the addition of powdered calcium carbonate, the ammonium was added in small doses over a nine day period and the plants were large at the time of the ammonium addition. This early damage by ammonium persisted in the nitrogen-sufficient plants at the 12- and 15-week harvests, but gradually decreased in the nitrogen-deficient plants until at the last harvest there was no difference in root size or quality of the beets among the nitrogen sources. Thus, indicating that with a prolonged nitrogen

deficiency, ammonium and nitrate were equally efficient as a nitrogen source, which was contrary to the prediction that ammonium should be a more efficient nitrogen source than nitrate.

In the nitrogen-sufficient series the beets that were supplied with nitrate, either alone or in combination with ammonium, were much larger than those with ammonium alone. These beets, because of their larger size, produced much more sucrose than those from the nitrogen-deficient plants even though their sucrose concentration and purity was much lower. Furthermore, the beets supplied with nitrate were consistently higher in sucrose concentration and purity than those supplied with ammonium alone. All in all, under nitrogen-sufficient conditions, nitrate was somehow a more effective source of nitrogen for sugar beets than ammonium alone, which was again contrary to prediction.

Agronomically, the early damage of ammonium could be important economically, since any damage to the fibrous roots could induce nutrient deficiencies and wilting on soils low in nutrients and in water supplying power. In this instance, nitrate could act as a "safening" agent for ammonium. Plants with petiole nitrate-N values of less than 1000 $\mu g\ g^{-1}$ (ppm) dry basis, were much higher in sucrose concentration and purity than those of plants above this value. Percent sucrose, $\hat{Y}$, can be estimated from the percent dry weight (X) of the brei by the regression equation $\hat{Y} = -0.194 + 0.603X$.

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