

Solution Electrical Conductivity and Ratio of Nitrate to Other Nutrients Affect Accumulation of Nitrate in Hydroponic Lettuce

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Abstract. Solution electrical conductivity (EC) and the supply of nitrate in proportion to other elements (nitrate supply ratio) should effect tissue composition of lettuce (*Lactuca sativa* L.) grown in hydroponic solution. These parameters were varied in several series of successive plantings in greenhouses in the northeast United States. In 1996, when the treatments differed only in EC, 0.65 and 0.9 dS·m⁻¹, but not in nitrate supply ratio, leaf tissue had more nitrate and total reduced-N and lettuce grew faster in the solution with higher EC. Over four series of plantings in 1997 and 1998, the nitrate supply ratio of a low-N treatment was only 60% of that for a high-N treatment, and EC was varied from 1.2 to 2.0 dS·m⁻¹. In 1997 and 1998, tissue nitrate was lower in the low-N treatment only when EC was less than in the high-N treatment. However, under irradiance greater than 10 MJ m⁻² per day, the lower EC also slowed growth. Stepwise regression over data from all experiments showed leaf nitrate was primarily a function of EC, and a term that described the interaction between irradiance and EC. Due to selective uptake by the plants, the ratio of elements in the recirculating solution differed from the ratio in which they were supplied. Under irradiance less than 10 MJ m⁻² per day and solution EC greater than 1.5 dS·m⁻¹, nitrate accumulated in solution to a concentration greater than expected from simple dilution of the concentrates. Tissue nitrate was also related to solution nitrate, increasing by 0.08–0.09 mg·g⁻¹ dry weight per 1 mg·L⁻¹ increase in solution nitrate. To prevent a rise in tissue and solution nitrate under low irradiance, both solution EC and nitrate supply ratio had to be reduced by about one-third, compared to the conditions required for rapid growth under high irradiance.

Some species of vegetables, such as beet (*Beta vulgaris* L.), celery [*Apium graveolens* L. var. *dulce* (Mill.) Pers.], lettuce (*Lactuca sativa* L.), and spinach (*Spinacia oleracea* L.), can accumulate more than 2500 mg nitrate per kg fresh weight of leaf tissue (Blom-Zandstra, 1989). Leaves of lettuce grown in hydroponics in northern Europe in winter can accumulate nitrate up to 6000 mg·kg⁻¹ (Van der Boon et al., 1990). This concentration is above the limits of 4500 and 3500 set by the European Economic Community, for winter and summer production, respectively, in view of the possible hazard to human health.

The most obvious strategy to lower nitrate in leafy vegetables is to reduce availability of nitrate to the plant. The nitrate in lettuce leaves grown in a greenhouse in the Netherlands in winter could be lowered by applying little or no fertilizer to the soil, but the level remained above that found when lettuce was grown with ample fertilizer in summer (Rooda van Eysinga and Meijs, 1985). Hydroponics allow complete withdrawal of nitrate from the nutrient solution. Transfer of hydroponic spinach to N-free solu-

tion for 2–3 d lowered leaf nitrate and increased ascorbic acid (Mozafar, 1996). However, temporary nitrate withdrawal lowered fresh weight and quality of mature lettuce (Van der Boon et al., 1990), and would starve younger plants of nitrate. Replacement of nitrate by ammonium fertilizer can lower tissue nitrate. Lettuce grown in peat with ammonium as the only N source had one-fourth the nitrate found in leaf tissue grown in nitrate-supplied media (Scaife et al., 1986). Unfortunately, hydroponic lettuce was of poor quality when ammonium accounted for >20% of the total N in solution (Van der Boon et al., 1990). Replacing 20% of the nitrate in solution with ammonium (Van der Boon et al., 1990) or proteinate supplement, a crude mixture of amino acids (Gunes et al., 1994), decreased the nitrate in lettuce leaves by only 30%, compared to lettuce grown with nitrate alone. The limited effectiveness of these strategies to lower tissue nitrate may be due to feedback inhibition of nitrate reductase activity by reduced forms of N (Tischner, 2000). Genetic modification to increase nitrate reductase may also fail to lower tissue nitrate. Tissue nitrate concentration was not directly related to nitrate reductase activity measured in vitro in a survey of lettuce cultivars (Blom-Zandstra and Enick, 1986).

Plants have one or more uptake systems that are specific for nitrate (Tischner, 2000).

In general, nitrate uptake is controlled by the requirement for plant growth, rather than being limited by the affinity of the transport system for uptake (Samuelson et al., 1995). However, nitrate may play a role in regulation of cell water potential. Blom-Zandstra and Lampe (1985) suggested nitrate replaced organic acids plus sugars in regulation of tissue water potential of lettuce grown under controlled conditions under different light intensities. Nitrate in leaves of greenhouse-grown lettuce also varies seasonally and is inversely related to the concentration of sugars (Drews et al., 1995). Plant sap must have negative water potential relative to the external solution to maintain cell turgor and to transport nutrients from the roots. If tissue nitrate is a major component of the regulation of water potential within plant cells, it should vary with solution electrical conductivity (EC). We hypothesize a low EC should lower the concentration of tissue nitrate.

In commercial practice, a concentrated mixture of nutrients is added to hydroponic solution to maintain a certain EC. In general these nutrients are not adjusted individually, but they are added in fixed proportion. If plants do not absorb a nutrient at the rate in which it is supplied, it will either accumulate or become depleted in the solution, as more nutrient concentrate is added to maintain EC. Nitrate will accumulate to a high concentration if this manner of nutrient addition results in a supply in excess of growth requirements. We hypothesize that decreasing the ratio of nitrate to other elements in the concentrate should lower the nitrate supply to less than the growth requirements, and alleviate any accumulation of nitrate in solution. This should lower tissue nitrate in solution when solution composition is adjusted on the basis of EC.

Hydroponic lettuce was grown in a greenhouse to see if high tissue nitrate is a problem in the northeast United States. The injection of nutrient concentrates into the re-circulating solution was governed by feedback based on EC. In an attempt to lower tissue nitrate, the supply of nutrients was modified by changing the EC set point, the supply of nitrate as a ratio of the other elements, or both.

Materials and Methods

Growth conditions. Lettuce was grown in greenhouses in Hamden, Conn. (lat. 42°N, long. 73°W). The greenhouses had a double-glazing of 0.1-mm clear-polyethylene film. They were heated by forced-air heaters, and horizontal air flow equalized temperature throughout each house. The minimum air temperature was 14 °C. Ventilation cooled the air when it was warmer than 18 °C. A shielded thermocouple measured air temperature at 1.5-m height in the center of each house. Temperature in troughs and in nutrient reservoirs of the hydroponic systems was measured with thermocouples insulated in glass tubes. Solar radiation outside the greenhouse was measured by a pyranometer (LI 190S; LI-COR, Lincoln, Nebr.). The output of these sensors was read every minute and recorded hourly by a data logger (CR10; Campbell Scientific, Logan,

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Utah). The greenhouses were covered with 50% shade cloth after 28 May 1997 to achieve a wider range of irradiance. After this date, the reported irradiance is one-half the value measured outside the greenhouse.

There were two independent re-circulating hydroponics systems within each greenhouse. Construction of these systems was described elsewhere (Gent, 2000). Nutrient solution was circulated past the plant roots and returned to a solution reservoir, where continuous feedback control was used to replenish the solution. A vertical liquid-level switch (model M800B; Madison Controls, Branford, Conn.) was set in the reservoir to maintain 100 L of solution. A conductivity cell was immersed in the solution (model 13-620-160; Fisher Scientific, Pittsburgh) and readings were corrected for temperature. Relays for injection of water or nutrients were activated for a 1-min interval whenever readings from the float switch or conductivity cell were below their set points. Water was injected through a solenoid at 120 mL/min. Nutrient concentrates were injected through peristaltic pumps. All concentrates were injected at the same time and at a fixed rate of 3 mL/min.

Two different nutrient solutions were compared at any one time. Within each planting, a high-N treatment was compared to a low-N treatment. Except for nitrate, all essential elements were supplied at the same ratio to both treatments. In 1996, two nutrient concentrates were injected when conductivity fell below set points of 0.65 and 0.90 dS·m⁻¹, for low- and high-N treatments, respectively. The composition of concentrate #1 was 595 mM KNO₃, 100 mM K₂HPO₄, 211 mM KH₂PO₄, 114 mM MgSO₄, 4.6 mM H₃BO₃, 1.8 mM MnSO₄·H₂O, 0.74 mM ZnSO₄·7H₂O, 0.21 mM CuCl₂·2H₂O, and 0.03 mM MoO₃. Concentrate #2 contained 300 mM Ca(NO₃)₂ and 9.8 mM Fe chelate.

In 1997 and 1998, the two treatments differed in the supply of nitrate as a ratio of the other elements. Concentrate #1 contained 297 mM KNO₃, half the amount of used in 1996. Concentrate #2 contained 300 mM CaCO₃ titrated with 305 mM HNO₃ and 280 mM HCl, and 9.8 mM Fe chelate. A concentrate #3 containing 417 mM HNO₃ was injected only in the high-N supply. Thus, the nitrate supplied in the low-N treatment was only 60% of the high-N treatment, as a ratio of the other nutrients supplied. In 1997, the conductivity set point was 1.5 dS·m⁻¹ for both treatments. Conductivity varied among plantings in 1998. The set point was 2.0 dS·m⁻¹ in both treatments for two plantings harvested in mid-winter. The conductivity set points were 2.0 dS·m⁻¹ and 1.2 dS·m⁻¹ in the next three plantings, and 1.5 dS·m⁻¹ and 1.2 dS·m⁻¹, for three plantings harvested in Spring 1998, for high- and low-N treatments, respectively.

Lettuce seeds (*Lactuca sativa* L. 'Buttercrunch'; Hart Seed Co., Wethersfield, Conn.) were germinated on wet paper and grown under controlled conditions of 25 to 30 °C and 18-h photoperiod of 60 W/m² irradiance. After 5 d, seedlings were transplanted in a tray of half-strength modified Hoagland solution (Lorenz and Maynard, 1980). After 14 d, the

seedlings were moved to the greenhouses and transplanted into polyethylene troughs. The bare-root plants were supported by foam-rubber plugs placed in holes in the cover of the trough. Fifteen plants were spaced 20 cm apart within each trough, and six or eight troughs were planted in each hydroponic system. As the plants grew in the greenhouse, they were spaced further apart by sequential harvests and by separation of the troughs. Plant canopies never overlapped. Plants were grown for 4 to 6 weeks until they reached a size of 90 to 250 g fresh weight. Typically, three plantings separated in age by 2-week intervals grew simultaneously in one hydroponic system.

Measurement and analysis. Harvests of 8 to 12 plants per system were done at 1 or 2-week intervals. Fresh weight, leaf area, and dry weight were recorded. The total fresh weight and number of plants were recorded at the final harvest, at which time ≈40 plants remained in each system. One leaf from each plant was selected from the layer just under the outer leaves of the loose head. The aggregate fresh weight and area of these leaves was determined, and they were freeze dried, weighed, and ground to a fine powder. Duplicate sub-samples of this leaf tissue were analyzed for nutrients. Nutrient solutions were sampled weekly for an independent measure of nutrient concentrations, conductivity, and pH.

The crop growth rate was the final weight per lettuce divided by the number of days elapsed from 1-g fresh weight to harvest. Relative growth rate was calculated at 1–2 week intervals using the formula:

$$\text{Relative growth rate} = \frac{\ln(\text{weight}_2) - \ln(\text{weight}_1)}{(\text{day}_2 - \text{day}_1)} \quad [1]$$

Dry weight ratio or fraction of dry matter, and leaf area to dry weight ratio were calculated from the sub-sample of leaves taken at the final harvest.

Subsamples of leaf tissue were wet-ashed. Cations were determined using inductively coupled plasma spectrometry (Gent, 2002) and total reduced nitrogen and phosphorus were determined in the tissue digests by colorimetric procedures (Gent, 2002). Other sub-samples were extracted with water and anions were analyzed by high pressure liquid chromatography (Gent, 2002).

Statistical analysis. Pair-wise comparisons of low-N to high-N treatment effects were tested for each series of plantings grown under similar conditions. The significance of the treatment effect was determined for growth rate and the concentrations of nutrients in leaf tissue and in solution. One treatment comparison was replicated over seven sequential plantings in 1996, but not within plantings, as there were only two hydroponic systems in one greenhouse. In 1997 and 1998, the high- and low-N treatments was replicated within each planting in identical systems in two greenhouses. In addition, treatments were replicated over a series of two to five sequential plantings. Analysis of variance (ANOVA) used planting and treatment as factors. All interactions of planting, treatment and replicate greenhouse

were combined with the error term. Regression analysis was performed on the combined data over all plantings to determine the effect of EC set point, nitrate supply ratio, and irradiance or temperature as continuous variables. The natural logarithm of irradiance was found to adequately explain the nonlinear response to integrated solar radiation. All products of these variables were also entered in regression to account for interaction effects. These terms were added in stepwise regression to determine their predictive value, with the condition that F statistic to enter >4.0. All statistics were done using SYSTAT (version 10.2; SPSS, Richmond, Calif.). Because there was a high correlation between temperature and irradiance (Table 3), mean temperature was not included as an independent variable in stepwise regression.

Results and Discussion

Solution composition. In 1996, the N-treatments only differed in EC, and both treatments had a lower EC than in later comparisons (Table 1). In 1997, the conductivity set points were about 1.5 dS·m⁻¹ for the two treatments, but the nitrate supply ratio for the low-N treatment was only 60% of that for the high-N treatments. The irradiance for the last two plantings in 1997 was reduced by shading the greenhouse with 50% shade cloth on 28 May, about half way through growth for plants harvested on 12 June. In 1998, the greenhouses remained covered with 50% shade cloth, and EC was varied in three sets of plantings. The irradiance in mid- and late-Winter 1998 was much lower than in 1996 and 1997.

Fig. 1 shows the concentrations of nitrate-N, phosphate-P and potassium, and pH over a period of 2 months in 1997, as several plantings of lettuce were grown and harvested. The concentration of some nutrients changed, but no persistent accumulation or depletion of particular elements occurred (including those not shown in Fig. 1) as the solution was maintained by addition of water and nutrient concentrates. The low-N solution had less nitrate and phosphate than the high-N solution. More potassium remained in the solution with less nitrate to maintain a similar solution EC. Solution pH was relatively constant, although there was no explicit pH control. The low-N solution pH was about 1 unit higher than for the high-N solution, because more nitric acid was injected in the latter solution. The change in composition on 28 May 1997 was likely related to shading of the greenhouses beginning on this date. Nitrate was diminished in the low-N solution before the plants were shaded. The concentration of potassium in the low-N solution increased after shading. These changes illustrate that sunlight intensity can affect composition of the recirculating solution.

When plants grew under high irradiance in 1996 and 1997, solution nitrate was less than or equal to that expected from dilution of concentrates to the appropriate EC set point. However under low irradiance in Winter 1998, solution nitrate was greater than that expected from dilution, even in the low-N treatment with

Table 1. Effect of environment, and EC set point on composition of the solution used to grow hydroponic lettuce, and concentrations expected from simple dilution of concentrates to the EC set-point. Environment variables and concentrations are averaged over each series of plantings. The low nitrate supply ratio (low-N) was 60% of the high nitrate supply ratio (high-N), except in 1996 when they were equal.

Series	Irradiance (MJ·m ⁻² per day)	Temp (°C)	Solution EC		Nitrate-N		Phosphate-P		Potassium	
			High-N	Low-N	High-N	Low-N	High-N	Low-N	High-N	Low-N
			dS·m ⁻¹		mg·L ⁻¹		mg·L ⁻¹		mg·L ⁻¹	
Summer 1996	20.7	25.1	0.9	0.65	43**	24	17	17	81*	53
From dilution					51	37	35	25	99	72
Spring 1997	21.6	19.8	1.5	1.4	82*	56	55**	32	72**	138
From dilution					86	59	58	67	166	192
Mid-Winter 1998	6.0	17.1	2.0	2.0	147**	126	64*	34	181*	228
From dilution					114	78	77	89	221	256
Late Winter 1998	5.9	17.4	2.0	1.2	117***	57	70***	33	143*	111
From dilution					114	47	77	54	221	153
Spring 1998	10.8	18.5	1.5	1.2	80***	17	39***	14	86	70
From dilution					86	47	58	54	166	153

*, **, ***Significance difference between treatments within a series of plantings at $P < 0.05$, 0.01 , and 0.001 , respectively.

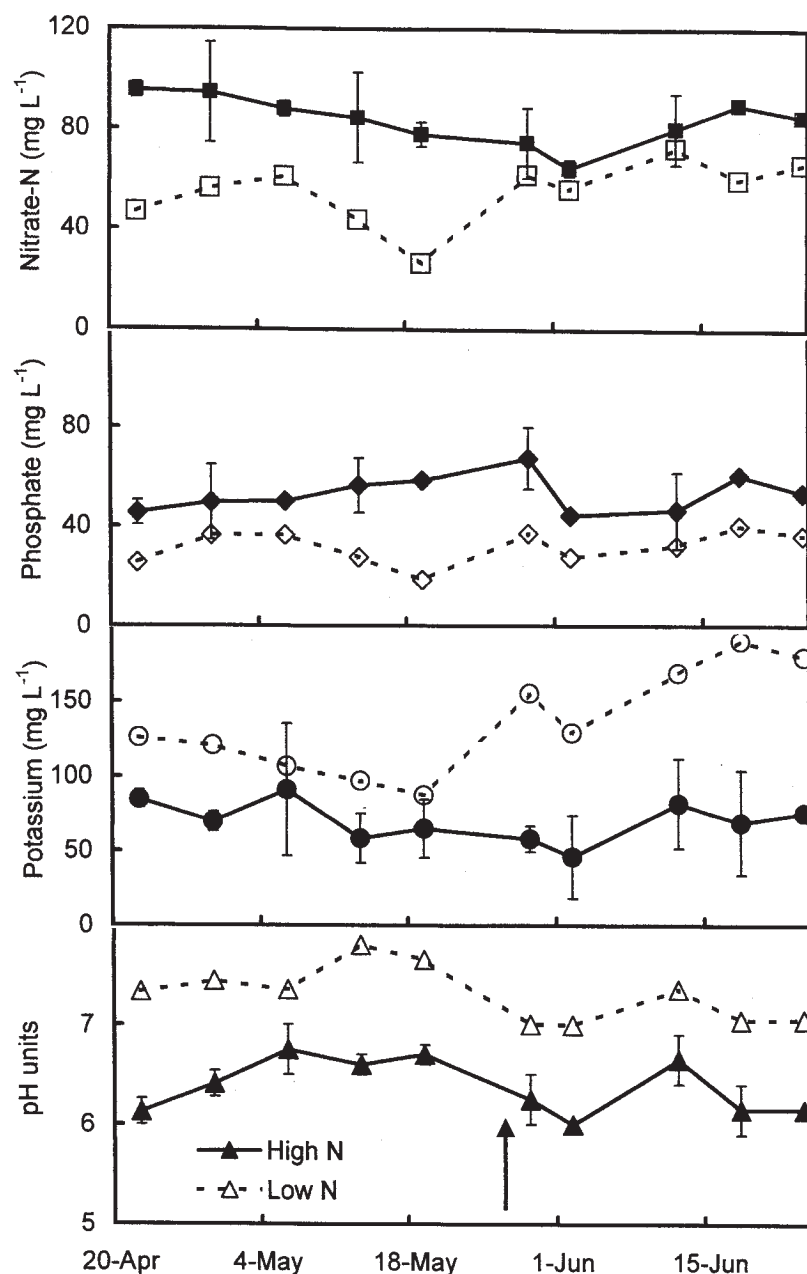


Fig. 1. The variation with time in concentrations of nitrate, phosphate and potassium, and pH in high-N and low-N solutions over 5 plantings in 1997. A 50% shade cloth was applied after the date shown by the arrow. Error bars indicate standard error of the estimate for both treatments.

a low nitrate supply ratio. Under a moderate irradiance in Spring 1998, plants depleted nitrate in the low-N solution far below that predicted from dilution of the concentrates. Irradiance had no such effect on the high-N solution. Stepwise regression of treatment and environment variables indicated a complex interaction of effects governing solution nitrate concentration (Table 4). The EC set point and nitrate supply ratio were significant independent variables, as was the interaction between these, and an interaction between EC and irradiance. Although both EC and nitrate supply ratio had to be lowered to deplete solution nitrate, the combined effect was less than additive.

In general, the solution concentrations of PO₄-P and K were substantially less than those expected from dilution of the concentrates to the appropriate EC set point (Table 1). Whenever treatments differed in nitrate supply ratio, there was significantly less phosphate in low-N compared to high-N solutions. This result was surprising, as dilution of concentrate to the EC set point should result in more phosphate in low- compared to high-N treatments, particularly in 1997 and mid-Winter 1998, when a low nitrate supply ratio required a greater amount of all other nutrients to achieve a given EC. In contrast, potassium was much greater in the low-N than in the high-N solution, to provide the soluble salts necessary to maintain the EC set point.

Tissue composition. The treatments affected concentrations of several elements in leaf tissue of hydroponic lettuce (Table 2). Nitrate-N was more sensitive than other elements to environment or solution composition. The nitrate concentration in leaf tissue of hydroponic lettuce grown in a greenhouse in Connecticut was never as high as the action level of 4500 mg·kg⁻¹ fresh weight developed by the European Economic Community. The range of tissue nitrate-N in individual plantings was 3 to 16 mg·g⁻¹ dry weight. The highest level, corresponding to 3500 mg·kg⁻¹ nitrate on a fresh weight basis, was observed when lettuce was grown in a shaded greenhouse in mid-winter with a solution in a shaded greenhouse in mid-winter with a solution EC of 2.0 dS·m⁻¹ and a high nitrate supply ratio.

Tissue nitrate responded more to EC set point than to nitrate supply ratio. Tissue nitrate differed among treatments in 1996 (Table 2),

Table 2. The effect of N-supply treatment on tissue composition and growth of hydroponic lettuce. Values for the final harvest are averaged over each series of plantings.

Series	Reduced N		Nitrate-N		Phosphorus		Potassium		Fresh wt		Growth rate	
	High-N	Low-N	High-N	Low-N	High-N	Low-N	High-N	Low-N	High-N	Low-N	High-N	Low-N
	mg·g ⁻¹		mg·g ⁻¹		mg·g ⁻¹		mg·g ⁻¹		g		g/d	
Summer 1996	47.0**	43.3	5.4*	5.0	7.4	7.2	71**	62	156**	97	8.1***	4.9
Spring 1997	40.4	39.8	6.6	6.3	5.0	5.3	96	97	224	189	8.2	6.8
Mid-Winter 1998	48.7	49.0	15.1	13.9	8.4	8.3	86	90	109	107	3.3	3.3
Late Winter 1998	46.0	43.1	11.7*	8.7	6.4	5.9	89*	84	122	126	4.0	4.0
Spring 1998	43.9	41.4	11.2*	6.0	5.1	6.5	89*	82	132*	110	5.6*	4.5

*, **, *** Significant difference between treatments within a series of plantings at $P < 0.05$, 0.01, and 0.001, respectively.

although nitrate supply ratio did not differ. Solution EC in 1996 was lower than in later comparisons, and tissue nitrate was also less. When EC differed among low- and high-N comparisons in later experiments, the low-N treatment lowered nitrate in leaf tissue significantly (Table 2). However, when treatments only differed in nitrate supply ratio, as in 1997, they had no significant effect on tissue nitrate. Some of the highest concentrations of nitrate in leaf tissue were observed with a low nitrate supply ratio, when plants were grown under low irradiance and high EC in midwinter 1998. Stepwise regression showed conductivity and the interaction of conductivity and irradiance accounted for most of the variance in tissue nitrate (Table 4). This relation suggests solution EC can be used to moderate accumulation of nitrate in leaf tissue of hydroponic lettuce. The success of this method likely depends on the role of nitrate as a regulator of plant water potential, as suggested previously (Blom-Zandstra et al., 1985; Steingrover et al., 1986). A high solution EC may require the plants to increase nitrate and other solutes in plant tissue to maintain a more negative water potential. It should be noted that EC was highly correlated with solution nitrate over all experiments, $r = 0.87$ (Table 3), and the importance of EC in this and the other stepwise regressions may be as a proxy for solution nitrate concentration.

When tissue nitrate was plotted as a function of solution nitrate for each planting and treatment, nitrate in leaves increased in proportion to solution nitrate, at least up to about 150 mg·L⁻¹ in solution (Fig. 2). Regression of tissue nitrate against solution nitrate, without regard for treatment, accounted for most of the variation in tissue nitrate, $r = 0.77$. Solution nitrate and sunlight were correlated because the treatments with high EC, resulting in highest solution nitrate, were applied under low irradiance. However, a relation similar to Fig 2 was observed when the regression was restricted to data in winter 1998, all under low irradiance. For this limited data, the regressions within each treatment gave $r > 0.77$. If data were restricted only to high irradiance, the correlation between solution and tissue nitrate was much lower, $r \approx 0.45$. Thus, accumulation of nitrate in the nutrient solution contributed to the high concentration in leaves of plants grown under low sunlight.

Frota and Tucker (1972) found temperature had a marked influence on nitrate absorption. The rate of nitrate absorption by lettuce starved for nitrogen increased more than tenfold as the root and shoot temperature was warmed from 8

Table 3. Pairwise correlation coefficients between electrical conductivity (EC) set point, nitrate supply ratio, or environment variables, and tissue or solution composition or growth parameters for hydroponic lettuce.

Parameter	Treatment or environment variable			
	EC set point	Nitrate supply ratio	Ln (irradiance)	Temp
Nitrate supply ratio	---	0.059	---	---
Ln (irradiance)	0.671***	0.333**	---	---
Temperature	0.737***	0.430***	0.789***	---
Tissue reduced-N	0.218	0.217	-0.323**	0.020
Tissue nitrate-N	0.701***	0.021	-0.660***	-0.597***
Tissue phosphorus	0.060	0.045	-0.179	0.198
Tissue potassium	0.615***	-0.271	-0.153	-0.586***
Solution nitrate-N	0.870***	0.168	-0.531***	-0.536***
Solution phosphate-P	0.728***	0.282*	-0.388**	-0.508***
Solution potassium	0.690***	-0.321**	-0.521***	-0.432***
Relative growth rate	-0.609***	0.248*	0.830***	0.688***
Growth rate	-0.167	0.197	0.650***	0.335**
Dry matter fraction	0.266*	-0.252*	-0.294*	-0.393**
Leaf area ratio	0.227	0.041	-0.083	-0.081

*, **, *** Significant correlation over all plantings at $P < 0.05$, 0.01, and 0.001, respectively. N = 64.

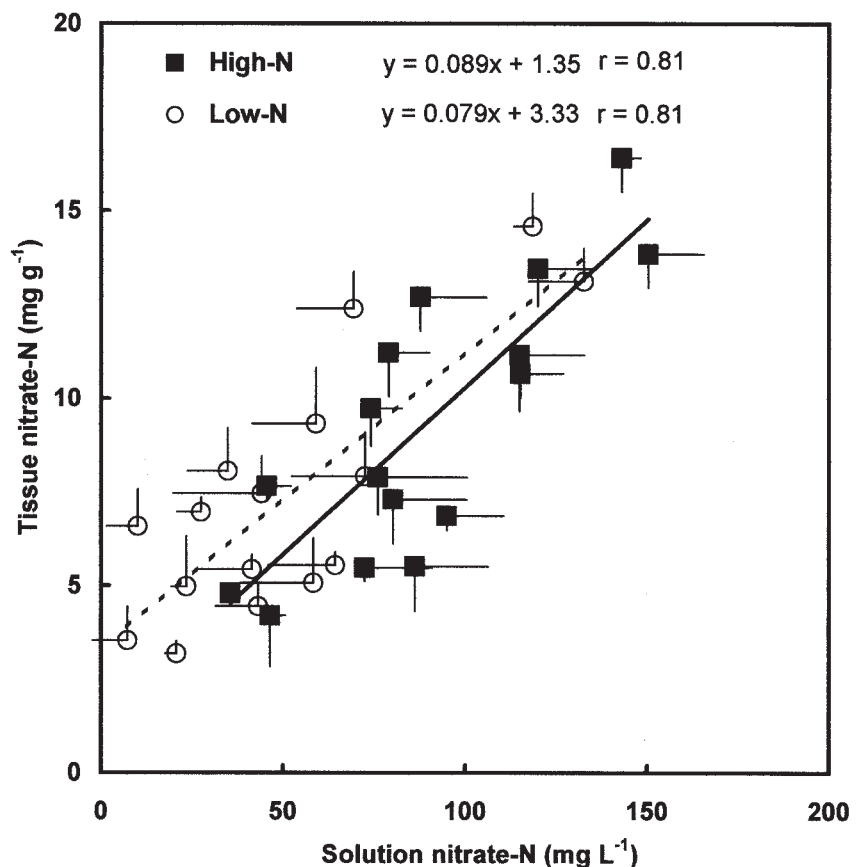


Fig. 2. Nitrate concentration in lettuce tissue as a function of solution nitrate. Symbols are measurements for each planting and lines are linear regressions of tissue against solution nitrate, for high-N and low-N treatments, separately. The bars indicate standard error of the estimate for both tissue and solution nitrate.

to 23 °C. However in our study, leaves had more nitrate when grown under low irradiance and cool temperature in winter than when grown under warm temperatures in summer. Thus, the temperature dependence of absorption did not appear to limit nitrate accumulation under the conditions in a heated greenhouse. It is likely that nitrate reductase activity in leaves was limited more by irradiance in winter than the uptake of nitrate by roots was limited by temperature.

The range of reduced-N in individual plantings was 38 to 49 mg·g⁻¹. The relative variation of reduced-N was small compared to that of nitrate. The N-treatments affected reduced-N significantly only in 1996. Environment and treatment variables accounted for only a small fraction of the variation in reduced-N (Table 3). High irradiance lowered the concentration of reduced nitrogen and an increase in nitrate supply ratio increased it.

The tissue potassium differed among N-treatments when they differed in EC (Table 2). Both tissue and solution potassium increased with solution conductivity and decreased with nitrate supply ratio (Table 3), but the relative changes in solution concentration were far greater than those in leaf tissue. The correlations between tissue and solution concentrations of potassium, $r = 0.36$, was much lower than for nitrate. The tissue concentrations of the other major elements (P, Ca, and Mg) were not correlated with any single treatment or environment variable. The depletion of solution phosphate that was related to a low nitrate supply ratio did not affect the concentration of phosphorus in leaf tissue. Thus nitrate in leaf tissue was unique in its responsiveness to the concentration in solution.

Growth. In 1996, growth rate per plant on a fresh weight basis was faster in solutions with an EC of 0.9 dS·m⁻¹ (high-N) than in solutions with an EC of 0.65 dS·m⁻¹ (low-N) (Table 2). Because low-N plants grew faster in 1997 than in 1996, solution composition did not affect the mean growth rate over the plantings in 1997. In mid- and late-Winter 1998, growth was slow, and N-treatments did not affect growth rate. As irradiance increased in spring 1998, plants grew faster in the high-N treatment (Table 1).

The growth rate was correlated primarily to sunlight (Table 3). Growth approached 10 g per day under irradiance of 25 MJ·m⁻² per day, compared to 3 g·d⁻¹ under the lowest irradiance of 5 MJ·m⁻² per day (Fig. 3). The effect of treatment depended on irradiance. Growth rate did not depend on N-treatment under low irradiance, but under high irradiance the low-N treatment slowed growth. N supply seemed to limit growth when irradiance was greater than 10 MJ·m⁻² per day (Fig. 3). Under continuous light conditions, lettuce seedlings required 400 mg·L⁻¹ nitrate for optimum growth (Mitchell et al., 1991), although 125 mg·L⁻¹ was sufficient for commercial production practices (Thompson et al., 1998). Thus, any changes used to lower the concentration of nitrate in solution should be applied with care, as they may inhibit rapid growth of lettuce under high light.

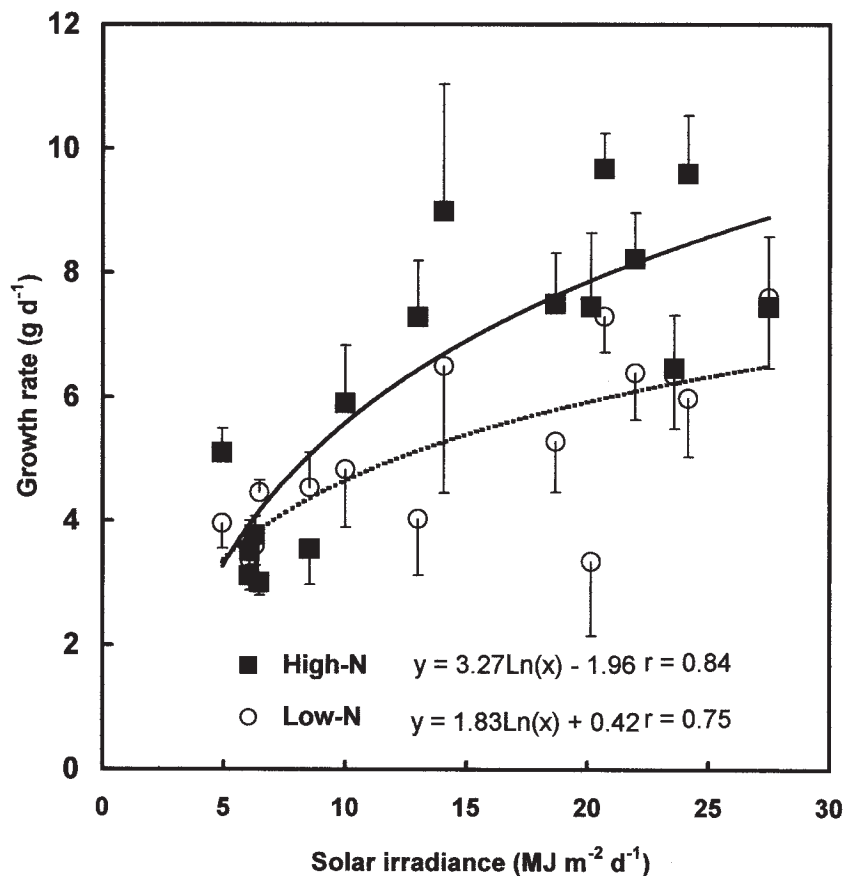


Fig. 3. Growth rate per plant for hydroponic lettuce as a function of solar irradiance. Symbols are growth rates for each planting and lines are regressions vs. logarithm of solar irradiance, for high-N and low-N treatments, separately. The bars indicate standard error of the estimate.

Bierhuizen et al. (1973) noted a linear relation between irradiance and dry matter accumulation of lettuce grown in soil in glasshouses in Holland, but in the present study the low-N treatment was a factor limiting growth above 10 MJ·m⁻² per day. A similar seasonal effect of nitrate nutrition was seen for hydroponic lettuce in Holland. In winter, yield and growth rates were the same with nitrate concentrations of 2.5 or 10 mM in the nutrient solution, but in summer, the yield with 2.5 mM nitrate was only 75% of that with 10 mM nitrate (Van der Boon et al., 1990).

Relative growth rate increased from 0.08 g·g⁻¹ per day at low irradiance up to ≈0.20 g·g⁻¹ per day, at irradiance above 20 MJ·m⁻² per day (data not shown). Stepwise regression showed irradiance accounted for most of the variance, but there was a weak inverse relation to EC set point, because the highest EC was under the lowest irradiance. Both the dry matter ratio and leaf area to weight ratio could be predicted by a single independent variable describing the interaction of irradiance and N treatment (Table 4), but the variance accounted for by regression was small compared to that for growth rate or relative growth rate. The average dry to fresh weight ratio was 0.053, and the leaf area per unit dry weight was 375 cm²·g⁻¹.

Air and root-zone temperatures in the range of 20 to 30 °C influence growth of lettuce in nutrient flow systems (Hickleton and Wolyntz, 1987; Thompson et al., 1998). In our

experiments, daily average air and solution temperature ranged from 17 °C in winter to 25 °C in summer (Table 1). The cooler temperatures could have slowed growth. However, irradiance and temperature were highly correlated, so the influence of temperature could not be ascertained independently of that for irradiance. Logarithm of irradiance had a higher correlation with growth parameters than did temperature, except for the fraction of dry matter (Table 3). For lettuce grown in soil, the principal effect of temperature was on leaf area expansion, rather than dry weight gain (Bierhuizen et al., 1973).

Tissue nitrate was lowered by the low-N treatment, without affecting growth rate, only in late winter 1998. In this series of plantings, the nitrate concentrations measured in the recirculating solution were similar to those expected from dilution of nutrients (Table 1). When low-N treatments depleted solution nitrate below that expected from dilution of concentrates, in 1996 and Spring 1998, they also slowed growth with respect to the high-N treatment. In these instances nitrate in solution was <50 mg·L⁻¹, but far above the apparent affinity of the uptake system in lettuce, on the order of 15 μM or 0.2 mg·L⁻¹ (Swiader and Freiji, 1996). However, there may be a lower nitrate concentration in the solution in contact with roots than in the bulk solution. Any manipulations used to lower tissue nitrate should avoid depletion of nitrate below ≈50 mg·L⁻¹ in solution.

Table 4. Regression coefficients for treatment and environment variables, and their interaction terms, selected by stepwise regression for prediction of solution or tissue nitrate concentrations or growth parameters of hydroponic lettuce.

Independent variables	Solution nitrate-N (mg·L ⁻¹)	Tissue nitrate-N (mg·g ⁻¹)	Relative growth rate (g·g ⁻¹ ·d ⁻¹)	Growth rate (g·d ⁻¹)	Dry matter fraction (g·g ⁻¹)	Leaf area ratio (cm ² ·g ⁻¹)
Ln (irradiance)	---	---	0.052***	2.28***	---	---
EC set point	174.4***	8.85***	-0.019*	---	---	---
Nitrate supply ratio	185.1***	---	---	---	---	---
Ln (irradiance) × EC set point	-8.2**	-1.93***	---	0.61**	---	20.7*
Ln (irradiance) × Nitrate supply ratio	---	---	---	---	-0.003**	---
EC set point × Nitrate supply ratio	-89.3*	---	---	---	---	---
Multiple R	0.918***	0.792***	0.791***	0.716***	0.353**	0.299*

***, **, *Significant correlation over all plantings at $P < 0.05$, 0.01 , and 0.001 , respectively. $N = 64$.

The growth response may be related to the modest change in tissue reduced-N. For lettuce grown in sand, relative growth rate expressed in degree days was correlated with organic nitrogen content of leaves expressed on a fresh weight basis (Dapoigny et al., 1997). Our treatments had a significant effect on reduced-N only in 1996 (Table 2). Regression over all data under our growth conditions showed growth rate was only weakly correlated to reduced-N, $r = -0.26$, or to nitrate, $r = 0.29$, in leaf tissue on a dry weight basis.

Summary. When solution EC and nitrate supply ratio were used to control nutrient concentrations in the solution to grow hydroponic lettuce, there was a complex interaction between these variables and sunlight that defined the actual concentration of nitrate in solution. Tissue nitrate was affected more by EC set point than by nitrate supply ratio as a control variable. However, both solution EC and nitrate supply ratio had to be reduced by about one third, compared to the conditions for optimum growth under high light, to prevent accumulation of solution nitrate under low sunlight. There was a high correlation between concentrations of nitrate in solution and in leaves. Solution EC may affect leaf nitrate both through its effect on plant water potential, in which nitrate appears to play a role as an osmoticum, and through its effect on accumulation of nitrate in the nutrient solution.

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