Growth and development of lettuce plants at high NH₄⁺:NO₃⁻ ratios in the nutrient solution

Jerônimo L Andriolo; Rodrigo dos S Godoi¹; Clarissa M Cogo; Orcial C Bortolotto; Gean L da Luz; José Carlos Madaloz²

UFSM-CCR-Dep^{to}. Fitotecnia, 97105-900 Santa Maria-RS; E-mail: andriolo@smail.ufsm.br

ABSTRACT

Lettuce plants, cv. Vera, were grown at five NH₄+:NO₃- ratios in the nutrient solution in a 0.15 m deep sand growing bed. A standard nutrient solution was used, with the composition of, in mmol L-1, 11.0 NO_3^- ; 1.5 $H_2PO_4^-$; 6.5 SO_4^- = 7.5 Ca^{++} ; 10.0 K^+ and 1.5 Mg^{++} , and, in mg L-1, 0.42 Mn; 0.26 Zn; 0.05 Cu; 0.50 B; 0.04 Mo, and 4.82 chelate Fe. Ammonium nitrate was used to supply NH₄ concentrations of 2.5; 5; 7.5 and 10 mmol L⁻¹, reaching NH₄⁺:NO₃ ratios of 0:11 (T1); 2.5:13.5 (T2); 5:16 (T3); 7.5:18.5 (T4) and 10:21 mmol L⁻¹ (T5) as treatments. A completely randomized experimental design was used, with four replications and 20 plants per plot. Four plants of each plot were harvested at 25 days after planting, to determine shoot and root dry mass, shoot fresh weight and number of leaves per plant. Number of leaves, shoot fresh and shoot and root dry mass decreased 25.5%; 52.5% and 68.5% from T1 to T5, respectively, following polynomial models. Root dry mass was 1.7 g/plant for T1 and T2, and 0.82 g/plant for T3, T4 and T5, decreasing in a proportion of 51.8%. For lettuce crop production in hydroponical facilities, the NH₄ threshold limit of about 9-12% of the total N should be retained.

Keywords: Lactuca sativa, fertigation, hydroponics, ammonium.

RESUMO

Crescimento e desenvolvimento da alface sob altas proporções $\mathrm{NH_4}^+{:}\mathrm{NO_3}^-$ na solução nutritiva

Plantas de alface, cv. Vera, foram cultivadas com cinco concentrações de NH, ha solução nutritiva, em uma camada de areia de 0,15 m de profundidade. Foi empregada uma solução nutritiva padrão, com a composição de, em mmol L-1, 11,0 de NO₃; 1,5 de H₂PO₄; 6,5 de SO₄—; 7,5 de Ca⁺⁺; 10,0 de K⁺ e 1,5 de Mg⁺⁺ e, em mg L⁻¹, 0,42 de Mn; 0,26 de Zn; 0,05 de Cu; 0,50 de B; 0,04 de Mo, e 4,82 de Fe quelatizado. Nitrato de amônio foi empregado para adicionar concentrações de NH₄ de 2,5; 5; 7,5 e 10 mmol L¹ atingindo proporções NH₄+:NO₃ de 0:11 (T1); 2,5:13,5 (T2); 5:16 (T3); 7,5:18,5 (T4) e 10:21 mmol L-1 (T5) como tratamentos. O delineamento experimental inteiramente casualizado foi empregado, com quatro repetições e 20 plantas por parcela. Quatro plantas foram coletadas aos 25 dias após o plantio, para determinar a massa fresca e seca da parte aérea e o número de folhas por planta. O número de folhas, a massa fresca da parte aérea e a massa seca da parte aérea e das raízes decresceram 25,5%; 52,5% e 68,5% de T1 para T5, respectivamente, seguindo modelos polinomiais. A massa seca de raízes foi de 1,7 g/planta em T1 e T2 e de 0,82 g/planta em T3, T4 e T5, decrescendo na proporção de 51,8%. Concluiu-se que o limite de concentração de NH, entre 9-12% do N total deve ser observado na produção hidropônica de alface.

Palavras-chave: Lactuca sativa, fertirrigação, hidroponia, amônio.

(Recebido para publicação em 20 de outubro de 2005; aceito em 30 de agosto de 2006)

Titrogen is one of the most important mineral nutrients determining plant growth and crop yield. Its effects are associated with leaf area growth and photosynthetic rate (Pinheiro Henriques & Marcellis, 2000; Pons & Westbeek, 2004). Linear relationships have been demonstrated between canopy nitrogen content, in g m⁻², and leaf area index (LAI) in early developmental stages of several species (Yin et al., 2003). Carbon, nitrogen and water uptakes rates are the key processes determining fresh yield of vegetables (Schenk, 1996; Yeo, 1999; Shanon & Grieve, 1999). In hidroponically horticultural production,

nutrient solutions at high N concentration and low electrical conductivity were used as a tool to maximize leaf growth and crop yield.

Plants can absorb NH₄⁺ only in a limited proportion of the nitrogen, because of its toxicity (Salsac *et al.*, 1987). Nevertheless, in hydroponical production of horticultural crops, a fraction of the total N is supplied as NH₄⁺ to better control the pH of the nutrient solution. For the lettuce crop, a proportion between about 9% and 12% of the total N has been reported in the literature (Faquin & Furlani, 1999). The tolerance of plants to NH₄⁺ varies with

species, environmental factors and plant stage development (Castro, 1999; Kotsiras *et al.*, 2002).

One of the most criticizable characteristics of vegetable soilless culture is the drainage in the environment of large amounts of used nutrient solutions. Recycling and reusing of such solutions is a current trend in searching for sustainable agricultural production systems. However, this practice might increase the NO₃ concentration in the nutrient solution, leading to its accumulation in leaves. Replacing NO₃⁺ by NH₄⁺ could be a mean to minimize such risk,

¹ Acadêmicos do Curso de Agronomia da UFSM, bolsistas de iniciação científica do Programa PIBIC-CNPq e da FAPERGS, respectivamente.

provided toxic levels were not reached.

Data describing the effect of N concentration and NH₄⁺:NO₃ ratios on hidroponically grown lettuce crops are scarce in the literature. Effects of electrical conductivity of the nutrient solution on plant growth and yield have been previously demonstrated (Ayers *et al.*, 1951; Andriolo *et al.*, 2005), but results were from experiments in which the concentration of all nutrients was modified in a similar way. In such condition, results represent salinity effects and could not be attributed to any nutrient in particular.

The goal of this work was to determine effects of increasing NH₄⁺:NO₃ ratios in the nutrient solution on growth, development and yield of hydroponically grown lettuce plants.

MATERIAL AND METHODS

The experiment was carried out inside a 150 m² polyethylene tunnel at UFSM, between February 25 and March 22, 2005. Sand of 0.0015-0.003 m particle size was used to make up a 0.15 m depth growing bed over fibber cement tiles, 3.05 m length and 1.10 m width, placed over concrete blocks in a slope of 3%. The tile gullies were previously filled with 0.015-0.025 m particle size gravel and covered with a 1,5 x 10⁻³ m polyethylene screen. The nutrient solution was pumped from a 500 L polyethylene reservoir to the upper side of the tile and drained down, flowing back to the same reservoir. Before planting, the growing bed was covered by a white polyethylene sheet (For more details about the experimental set-up, see Andriolo et al., 2004).

At February 25, 2005, six-leaved lettuce plantlets, cv. Vera, were transferred to the growing bed through cuttings made on the polyethylene sheet, in a plant density of 25 plants m⁻². Water and nutrients were supplied by means of a standard nutrient solution with the following composition, in mmol L⁻¹ (modified from Castellane & Araújo, 1995): 11.0 NO₃; 1.5 H₂PO₄; 6.5 SO₄—; 7.5 Ca⁺⁺; 10.0 K⁺ and 1.5 Mg⁺⁺, and, in mg L⁻¹, 0.42 Mn; 0.26 Zn; 0.05 Cu; 0.50 B; 0.04 Mo, and 4.82 chelate Fe. Ammonium nitrate was used to supply

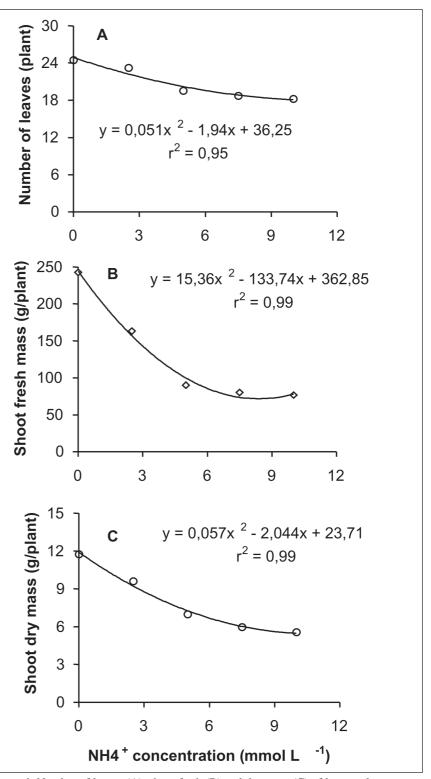


Figure 1. Number of leaves (A), shoot fresh (B) and dry mass (C) of lettuce plants grown hydroponically at NH₄⁺ concentrations of 0; 2.5; 5; 7.5 and 10 mmol L⁻¹, reaching NH₄⁺:NO₃⁻¹ ratios of 0:11; 2.5:13.5; 5:16; 7.5:18.5 and 10:21 mmol L⁻¹. Santa Maria, UFSM, 2005.

 ${
m NH_4^+}$ concentrations of 2.5; 5; 7.5 and 10 mmol ${
m L^{-1}}$, reaching ${
m NH_4^+}$: ${
m NO_3^-}$ ratios of 0:11 (T1); 2.5:13.5 (T2); 5:16 (T3); 7.5:18.5 (T4) and 10:21 mmol ${
m L^{-1}}$ (T5) as treatments. Values of pH and

electrical conductivity (EC) were maintained between 5.1 and 5.9 and 1.7 and 2.8 dS m¹, respectively, by adding KOH, water or aliquots of new nutrient solution when necessary. The nutrient

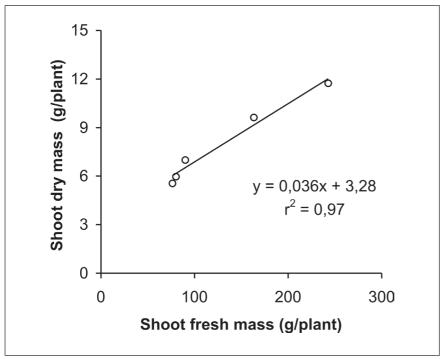


Figure 2. Relationship between shoot fresh and dry mass accumulation of lettuce plants grown hydroponically at NH₄⁺ concentrations of 0; 2.5; 5; 7.5 and 10 mmol L⁻¹, reaching NH₄⁺:NO₃ ratios of 0:11; 2.5:13.5; 5:16; 7.5:18.5 and 10:21 mmol L⁻¹. Santa Maria, UFSM, 2005.

solution was not replaced till the end of the experiment. The nutrient solution was delivered four times a day, at 8 h AM, 11 h AM, 14 h PM and 17 h PM, by means of a pump controlled by a timer. At each fertigation, a volume of 130 L of nutrient solution was delivered in each growing bed. The lateral sides of the tunnel were opened during the day. A randomised experimental design was used, with four replications with 20 plants.

Plants were harvested at 25 days after planting (DAP), when the first signs of senescence were identified on fully expanded older leaves. The number of leaves longer than 0.01 m was counted. Shoot fresh weight was determined and roots were carefully removed out from the sand and washed in water. Dry mass of leaves, stem and roots were determined after drying at 60°C until constant dry weight was reached. The effect of treatments was tested by analysis of variance (ANOVA) and statistical models were fitted by regression analysis for variables differing by the F test (P=0.05).

RESULTS AND DISCUSSION

Average number of leaves per plant was 24.5 and 23.2 in T1 and T2, without

significant difference, and decreased to 18.2 in T5, following a polynomial model (Figure 1A). Fresh mass of shoot organs decreased significantly by a proportion of 32.6% from T1 to T2 and by 53.2% from T2 to T5. The decrease from T4 to T5 was of about 4.4% and was not significant. Data from this variable pooled together follow a polynomial model (Figure 1B). Similar trend was observed for shoot dry mass, with 18.2% decrease from T1 to T2 and 42.2% thereafter (Figure 1C). Root dry mass did not differ among treatments and the average was 1.2 g/plant.

One of the questions arising from the present experiment is the nature of the negative effect of high NH,+ concentrations on lettuce plant growth and development. These two plant physiological processes could be affected either by salinity or by ion toxicity. In a previous paper (Andriolo et al., 2005), it has been demonstrated that only fresh weight was reduced by salinity of the nutrient solution in a concentration range between about 1.0 and 5.0 dS m-1. This reduction was related to restrictions in plant water flux by effect of salinity. Other physiological processes seem to be involved in NH,+

toxicity, as number of leaves and shoot fresh weight, and shoot and root dry weights were affected. In fact, a linear relationship was found between shoot and fresh weight, indicating that plant water content was affected by NH,+ in a similar way in the five treatments (Figure 2). This implies restrictions in both dry mass synthesis and water uptake by roots. In the literature, effects of NH₄ toxicity have been attributed to reduction/inhibition of cation absorption, especially K+, as a consequence of ion imbalance (Salsac, 1989; Kotsiras et al., 2002). Nevertheless, reduction in dry mass became less pronounced at higher NH,+ concentrations of T4 and T5 (Figure 1C) and was not recorded in shoot fresh mass (Figure 1B). A saturation-type response was fitted by regressions, suggesting a threshold limit above its adaptative processes might take place and plants might survive at high NH,+ concentrations in the root media.

The reduction in number of leaves by increasing ratios of NH₄⁺ in the nutrient solution implies that plant development was affected by limitations growth. Growth affecting development has not been considered in models of horticultural crops (Streck et al., 2002, 2003). In such models, temperature is the key variable used to estimate the phyllochron index, but this formalization could only be valid under non-limiting growing conditions. These conditions were often not realistic in horticultural soilless crops, due to extreme fluctuations in environmental variables affecting shoot and root growth of plants. Presented data suggest that other approaches should be considered in developmental models for theses crops, like that proposed by the Hardwick's (1987) isometry hypothesis. Its theoretical basis states that growth and development of a plant follows a proportion between accumulation of metabolic surface area, represented by leaves, and biomass, disregarding fluctuations in the environment.

Although growth of roots was reduced at higher NH₄⁺ concentrations in the nutrient solution (Figure 1C), this observation might not be attributed to a direct effect of NH₄⁺ toxicity. Growth of roots has been considered as

dependent of the carbon/nitrogen balance between shoot and root compartments (Thornley, 1998). Current hypothesis underlying such models have considered that roots grow to search for water and nutrients, especially nitrogen. In this way, the negative effect of NH,⁺ recorded in root growth might also be explained by the higher nitrogen availability in the nutrient solution. This hypothesis derives from the higher values of shoot:root ratio in plants grown at higher NH₄⁺ concentrations (Figure 3). This implies that growth of roots was lower than that of shoot organs, but NH₄ toxicity affecting growth of roots more severely than that of shoot seems improbable.

Total nitrogen in the nutrient solution currently more used to grow the lettuce crop in NFT in Southern Brazil was of about 16.9 mmol L⁻¹ as NO₃ (Castellane & Araújo, 1995). This concentration is near to that of 16 mmol L⁻¹ in T2. Thus, the 32.6% reduction in fresh mass in plants from T1 to T2 can be attributed to the 16.6% NH₄⁺ in the nutrient solution. It might be concluded that for commercial purposes of lettuce crop production in hydroponical facilities, the NH₄⁺ threshold limit of about 9-12% of the total N reported in the literature should be retained.

REFERENCES

- ANDRIOLO JL; WITTER M; ROSS T; GODOI RS. 2003. Crescimento e desenvolvimento do tomateiro cultivado em substrato com reutilização da solução nutritiva drenada. *Horticultura Brasileira* 21: 489-493.
- ANDRIOLO JL; LUZ GL; GIRALDI C; GODOI RS; BARROS GT. 2004. Cultivo hidropônico da alface empregando substratos: uma alternativa a NFT?. *Horticultura Brasileira* 22: 794-798.
- ANDRIOLO JL; LUZ GL; WITTER MH; GODOI RS; BARROS GT; BORTOLOTTO OC. 2005. Growth and yield of lettuce plants under salinity. *Horticultura Brasileira*. (Submited).
- AYERS AD; WADLEIGH CH; BERNSTEIN L. 1951. Salt tolerance of six varieties of lettuce. Proceedings of the American Society for Horticultural Science 57: 237-242.

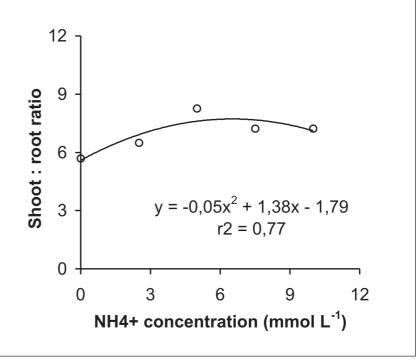


Figure 3. Shoot:root ratio of lettuce plants grown hydroponically at NH_4^+ concentrations of 0; 2.5; 5; 7.5 and 10 mmol L^- 1, reaching NH_4^+ : NO_3^- ratios of 0:11; 2.5:13.5; 5:16; 7.5:18.5 and 10:21 mmol L^- 1. Santa Maria, UFSM, 2005.

- CASTELLANE PD; ARAÚJO JAC. 1995. *Cultivo sem solo Hidroponia*. 4. ed. Jaboticabal: FUNEP. 43p.
- CASTROAA. 1999. Solución nutritiva: principios básicos. Comportamento e interacción de los distintos elementos. In: MILAGROS MF; GÓMEZ IMC (eds). Cultivos sin suelo. II. Curso Superior de Especialización. Almería: DGIFA-FIAPA-Caja Rural de Almería. 2. ed. p. 229-246.
- FAQUIN V; FURLANI PR. 1999. Cultivo de hortaliças de folhas em hidroponia em ambiente protegido. *Informe Agropecuário* 20: 99-104.
- HARDWICK RC. 1987. The nitrogen content of plants and the self-thining rule of plant ecology: a test of the core-skin hypothesis. *Annals of Botany* 60: 439-446.
- KOTSIRAS A; OLYMPIOS CM; DROSOPOULOS J; PASSAM HC. 2002. Effects of nitrogen form and concentration on the distribution of ions within cucumber fruits. *Scientia Horticulturae* 95: 175–183.
- PINHEIRO-HENRIQUES AR; MARCELIS FM. 2000. Regulation of growth at steady-state nitrogen nutrition in lettuce (*Lactuca sativa* L.): interactive effects of nitrogen and irradiance. *Annals of Botany* 86: 1073-1080.
- PONS TL; WESTBEEK MHM. 2004. Analysis of differences in photosynthetic nitrogen-use efficiency between four contrasting species. *Physiologia Plantarum* 122: 68-78.

- SALSAC L; CHAILLOU S; MOROT-GAUDRY JF; LESAINT C; JOLIVET E. 1987. Nitrate and ammonium nutrition in plants. *Plant Physiology and Biochemistry* 25: 805-812.
- SHANNON MC; GRIEVE CM. 1999. Tolerance of vegetable crops to salinity. *Scientia Horticulturae* 78: 5-38.
- SCHENK MK. 1996. Regulation of nitrogen uptake on the whole plant level. *Plant and Soil* 181: 131-137.
- STRECK NA. 2002. A generalized non linear air temperature response function for mode appearance rate in muskmelon (*Cucumis melo L.*). Revista Brasileira de Agrometeorologia 10: 105-111.
- STRECK NA. 2003b. A vernalization model in onion (*Allium cepa* L.). *Revista Brasileira de Agrociência* 10: 99-105.
- THORNLEY JHM. 1998. Modelling shoot: root relations: the only way forward? *Annals of Botany* 81: 165-171.
- YEO A. 1999. Predicting the interaction between the effects of salinity and climate change on crop plants. *Scientia Horticulturae* 78: 159-174.