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# Uptake, Assimilation and Translocation of Ammonium or Nitrate in Italian Ryegrass

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**ABSTRACT : To investigate the partitioning of newly absorbed N derived from $NO_3^-$ and $NH_4^+$, 6 mM $K^{15}NO_3$ or 3 mM $(^{15}NH_4)_2SO_4$ was fed continuously in Italian ryegrass (*Lolium multiflrum* L.) for 7 days. Nitrogen metabolites (nitrate, amino acid, soluble- and insoluble protein) were analyzed at the end of $^{15}N$ feeding. Dry weight in shoot, stubble and root was not significantly different between $NO_3^-$ and $NH_4^+$ feeding. Total nitrogen content in all three organs was significantly higher in $NH_4^+$ than $NO_3^-$ feeding. Sum on N content in reduced N fractions (amino acids + proteins) in shoot, stubble and roots in $NH_4^+$ feeding increased by 13.3, 12.5 and 35.4%, respectively, compared to $NO_3^-$ feeding. The Relative Specific Activity (RSA, percentage of newly absorbed $^{15}N$ relative to total N in a sample) values of amino acids and insoluble proteins were significantly higher in $NH_4^+$ feeding. Total amount of newly absorbed $^{15}N$ in $NH_4^+$ and $NO_3^-$ feeding was 52.3 and 69.5 mg /plant on dry matter basis, respectively. In both $NH_4^+$- and $NO_3^-$-grown plants, most of the N was allocated to the shoot, 67.5% in $NH_4^+$ feeding and 58.8% in $NO_3^-$ feeding, respectively. The $^{15}N$ amount incorporated in the reduced N compounds (amino acids and proteins) in $NH_4^+$-grown plants significantly increased by 74.8% compared to $NO_3^-$-grown plants. The increase of the $^{15}N$ amount assimilated to amino acids in $NH_4^+$-grown plants was remarkably higher in roots as more than 7.25 times compared to $NO_3^-$ feeding. These results indicated that Italian ryegrass was much efficiently utilized $NH_4^+$-N for the synthesis of reduced N compounds.**

***Keywords*** **:** Italian ryegrass, $^{15}N$ labeling, Uptake of $NH_4^+$ and $NO_3^-$, Assimilation, Translocation

Nitrogen is a major limiting nutrient for plants in most ecosystems. It is taken up from soils mainly as nitrate ($NO_3^-$) and/or ammonium ($NH_4^+$) by the roots of higher plants (Marschner, 1995). Nitrogen demands of higher plants are usually met by the net release of $NH_4^+$ and $NO_3^-$ into the soil solution via mineralization and nitrification. Absorption of $NO_3^-$ and $NH_4^+$ by plants allows them to form numerous N compounds, mainly proteins.

Many plant species absorbed most nitrogen as $NO_3^-$, because $NH_4^+$ is so readily oxidized to $NO_3^-$ by nitrifying bacteria. In the N metabolism studies, nitrate assimilation has been focused because of its aboundance in most soils. Nitrogen assimilation is an energy-intensive process, requiring the transfer of two electrons per $NO_3^-$ converted to $NO_2^-$, six electrons per $NO_2^-$ converted to $NH_4^+$ (Bloom *et al.*, 1988). Nitrogen assimilation is tightly linked to C-metabolism as energy and C-skeletons are needed to convert inorganic nitrogen to organic compounds. Whether nitrogen assimilation occurs in root or shoot, there is a high demand for carbon, independent of the N source. Uptake of $NO_3^-$ into root symplasm from the rhizosphere against an electrochemical gradient (Clarkson, 1986) and $NO_3^-$ transport out of the root symplasm into the xylem requires metabolic energy (Peuke and Jeschke, 1995; Cooper and Clarkson, 1989). Plants assimilate in root virtually all of the $NH_4^+$, from 5 to 95% of the $NO_3^-$ absorbed from the rhizosphere (Andrews, 1986; Oaks and Hirel, 1985). Estimates of root nitrogen acquisition and the partitioning have been limited, and these could not distinguish among expenditures for tissue maintenance, root growth and $NH_4^+$ and $NO_3^-$ assimilation.

In the present work, experiments were designed to investigate directly the flux and partitioning of $NO_3^-$ and $NH_4^+$ within the whole plant. Isotopes labeled were used to estimate the quantitative significance of $NO_3^-$ and $NH_4^+$ utilization in the full vegetative Italian ryegrass that is well supplied with nutrients.

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## MATERIALS AND METHODS

### Plant culture and experiment procedure

Seeds of Italian ryegrass (*Lolium multiflorum* L.) were germinated on a sand beach. Two-weeks old seedlings were transplanted to 3 L pots; ϕ200 mm, depth 150 mm (5 plants per pot) and grown hydroponically on a nutrient solution containing 1 mM $NH_4NO_3$, 0.4 mM $KH_2PO_4$, 0.25 mM KCl, 0.25 mM $CaCl_2$, 0.2 mM $MgSO_4$, 0.15 mM $K_2HPO_4$, micro-nutrient (14 μM $H_3BO_3$, 5 μM $MnSO_4 \cdot H_2O$, 3 μM $ZnSO_4 \cdot 7H_2O$, 3 μM $CuSO_4 \cdot 5H_2O$, 0.7 μM$(NH_4)6Mo_7O_{24}$, 0.1 μM $CoCl_2$). $CaCO_3$ was then given in excess at a final concentration of 2 mM to maintain the solution pH at 6.2 ± 0.3. The nutrient solution was continuously aerated and renewed every 7 days. Plants were grown in the complete nutrient solution containing 1.0 mM $NH_4NO_3$ for 6 months, and allowed two regrowth cycles to give enough tillering and root growth. For the treatment of $NO_3^-$ and $NH_4^+$ feeding, continuous $^{15}N$ labeling was carried out by replacing $NH_4NO_3$ with 6 mM $K^{15}NO_3$ (10 atom % $^{15}N$ excess) and 3 mM $(^{15}NH_4)_2SO_4$ (10 atom % $^{15}N$ excess) for 7 days. Plants in each treatment were harvest at the end of $^{15}N$ feeding. Plants were cut leaving a stubble of 6 cm above root base and separated with 3 organs (root, stubble and shoot). Samples were immediately frozen in liquid nitrogen. Freeze-dried samples were finely ground and stored under vacuum for further analysis.

### Chemical fractionation and isotope analysis

About 200 mg of freeze-dried sample was extracted with 25 ml of 80% ethanol. The ethanol-soluble fraction was filtered, centrifuged, and passed through a Dowex 50W (200–400 mesh, $H^+$) column. The collected solutions was concentrated to 5.0 ml (nitrate fraction). Amino acids were eluted with 25 ml of 0.5N HCl from the Dowex 50W column and concentrated to 3.0 ml. The residues of ethanol extraction were dried for 24 h to obtain dry weight. The resulting dried samples were designated as insoluble protein.

About 25 mg of finely ground freeze-dried sample for soluble protein was extracted with 1 ml of 100 mM $NaPO_4$ buffer (pH 6.8). Tubes were vortexed for 30 sec and placed on ice for 5 min, and centrifuged on 14,000 rpm at 4°C for 10 min. This procedure repeated four times. The supernatant was freeze-dried and residue was dissolved with distilled water to obtain the final volume of 0.2 ml.

The solid samples (total N and residues) were precisely measured into tin capsules. For the solution samples, an samples usually of 0.1 ml, was dropped into tin capsules to give more than minimum sample size (20 μg for $^{15}N$ abundance). The tin capsules containing solution were quickly cooled with liquid nitrogen, and then dried in a freeze-dryer. The residues in tin capsules were employed for analysis of N content and $^{15}N$ abundance. The N content and $^{15}N$ abundance were determined by an ANCA mass spectrometer (Europe Scientific, Crewe, UK). $^{15}N$-enriched L-glutamic acid (0.366 $^{15}N$ atom %) were used as the references for N analysis. The $^{15}N$ abundance obtained was converted to the relative specific activity (RSA, percentage of recently incorporated atoms relative to the total atoms in the sample) using equation (1). The amounts of newly absorbed $^{15}N$ (NAN) incorporated in the N compound were calculated per plant organ by equation (2).

$$RSA = (^{15}N \text{ atom \% measured} - \text{Natural } ^{15}N \text{ atom \%}) / (^{15}N \text{ atom \% nutrient solution fed} - \text{Natural } ^{15}N \text{ atom \%}) \times 100 \quad (1)$$

$$NAN = (RSA \times \text{N content measured in a compound})/100 \quad (2)$$

## RESULTS

### Dry weight

Dry weight as affected by 7 days of $NO_3^-$ and $NH_4^+$ feeding is presented in Table 1. Dry weight of three organs was slightly increased in both $NH_4^+$ and $NO_3^-$ feeding. However, there was no significant difference.

### Content of nitrogen compounds in plant organ

Nitrogen content in various nitrogen metabolites in response to $NO_3^-$ and $NH_4^+$ feeding are shown in Table 2. Total nitrogen content in three organs was significantly higher in $NH_4^+$ feeding than $NO_3^-$. In $NH_4^+$ feeding, the N content in reduced N fractions (amino acids, soluble and insoluble proteins) in all three organs increased compared to those of $NO_3^-$ feeding (13.3, 12.5 and 35.4% increase in shoot, stubble and roots). In shoot and stubble, N contents in amino acid and soluble proteins in $NH_4^+$ feeding increased by on average 19.1% and 17.4%, while those in insoluble

**Table 1.** Dry weight at day 0 (mean before N treatment) and 7 days after $NO_3^-$ or $NH_4^+$ feeding. Each value is the mean ± S.E. for n=5.

| Treatment | Shoot | Stubble | Root |
|---|---|---|---|
| | D. W (g plant$^{-1}$) | | |
| Day (0) | 3.12 ± 0.21 | 1.12 ± 0.09 | 0.98 ± 0.01 |
| Day (7) | | | |
| $NO_3^-$-N | 3.25 ± 0.25 | 1.30 ± 0.13 | 1.02 ± 0.08 |
| $NH_4^+$-N | 3.30 ± 0.30 | 1.46 ± 0.12 | 1.21 ± 0.11 |

**Table 2.** Nitrogen content in various N metabolites after 7 days of $NO_3^-$ or $NH_4^+$ feeding.

| Organs | N compounds | Mineral N form | | Significant level |
|---|---|---|---|---|
| | | $NO_3^-$-N | $NH_4^+$-N | |
| | | --------(mg N/g, dry wt.)------- | | |
| Shoot | Total N | 41.79 | 45.29 | * |
| | Nitrate | 6.85 | 5.70 | * |
| | Amino acid | 2.11 | 2.69 | * |
| | Insoluble protein | 23.00 | 25.70 | ns |
| | Soluble protein | 9.83 | 11.20 | ** |
| Stubble | Total N | 30.68 | 33.76 | * |
| | Nitrate | 6.33 | 5.68 | ns |
| | Amino acid | 3.52 | 3.91 | * |
| | Insoluble protein | 12.53 | 13.46 | ns |
| | Soluble protein | 8.28 | 10.00 | * |
| Root | Total N | 19.45 | 23.99 | ** |
| | Nitrate | 3.78 | 2.77 | * |
| | Amino acid | 1.10 | 3.45 | ** |
| | Insoluble protein | 9.92 | 11.75 | * |
| | Soluble protein | 4.65 | 6.02 | ** |

Significant level of difference : n.s., non significant, *p<0.05, **p<0.01.

proteins were not significantly different between $NO_3^-$ and $NH_4^+$ feeding. In roots fed with $NH_4^+$, the increase of the N contents in all biochemical compounds was much higher than shoot and stubble. Especially, N content in amino acids was remarkably higher in $NH_4^+$ feeding (3.1 fold higher than $NO_3^-$ feeding). The N content in nitrate in all three organs was lower in $NH_4^+$ feeding than $NO_3^-$ feeding. In roots, the N content of all metabolites examined was found to be much sensitively responded to N supply forms.

### Percentage of $^{15}N$ derived from $^{15}NO_3^-$ and $^{15}NH_4^+$

Fig. 1 shows the changes in percentage of $^{15}N$ derived from $^{15}NO_3^-$ and $^{15}NH_4^+$ (Relative specific activity, RSA) in each organ after 7 days of $^{15}N$ feeding. In all three organs, RSA value of nitrate was significantly higher in $NO_3^-$ feeding, whereas RSA value of amino acids remarkably higher in $NH_4^+$ feeding. The RSA value of nitrate in shoot, stubble and root in $NO_3^-$ feeding was 48.6, 41.6 and 64.43%, respectively. The value in $NH_4^+$ feeding largely decreased in three organs. The value of amino acids in shoot, stubble and root in $NH_4^+$ feeding was 10.5, 20.5 and 30.4% higher than $NO_3^-$ feeding. The value of insoluble proteins in $NO_3^-$ feeding remained at about half of $NH_4^+$ feeding in three organs. The value of soluble proteins in shoot was significantly higher in $NO_3^-$ feeding (35.4%) than $NH_4^+$ feeding (27.9 %), but it was conversed in stubble and roots.

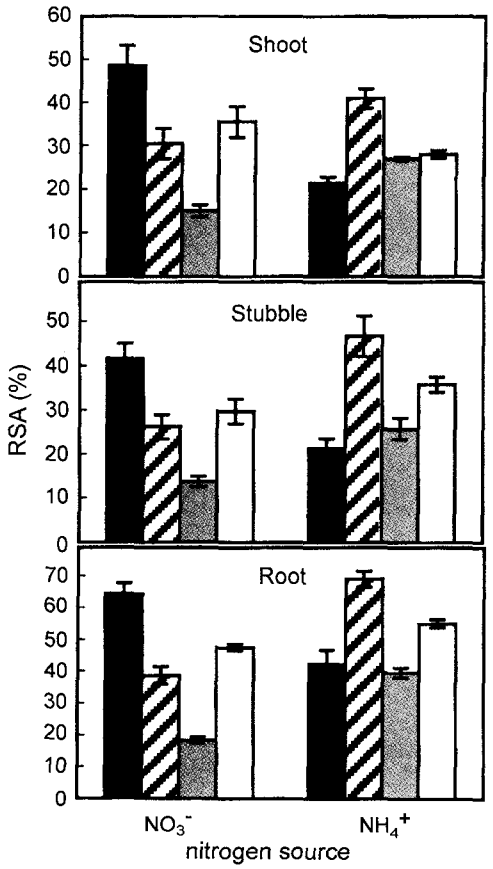


**Fig. 1.** The relative specific activity (RSA) in various N metabolites in each organ 7 days after $NO_3^-$ or $NH_4^+$ feeding. ■ : nitrate, ▨: amino acid, ▩: insoluble protein, □ : soluble protein. Each value is the mean ± S.E. for n=5.

### $^{15}N$ content

$^{15}N$ content of four N metabolites in each organ after 7 days of $^{15}NO_3^-$ or $^{15}NH_4^+$ feeding is presented in Fig 2. In shoot, $^{15}N$ contents of nitrate in shoot, stubble and root in $NO_3^-$ feeding was 270, 230 and 230 μg/g, dry wt., while those of $NH_4^+$ feeding decreased by 44.4, 26.1 and 47.8%, respectively. In $NH_4^+$ feeding, $^{15}N$ content of amino acids was about 2-fold higher in shoot and stubble, and 6-fold higher in roots compared $NO_3^-$ feeding. $^{15}N$ content in insoluble protein in all plant organ was also significantly increased in $NH_4^+$ feeding, showing the highest content among N compounds examined. $^{15}N$ content in soluble protein in stubble and roots was 1.9 and 1.6 fold higher in $NH_4^+$ feeding, while no significant difference was observed in shoot between two N supply forms.

### Partitioning of newly absorbed $^{15}N$

Flow sheet of absorbed $^{15}N$ derived from $^{15}NO_3^-$ or $^{15}NH_4^+$ in whole plant is summarized in Fig. 3. Total amount

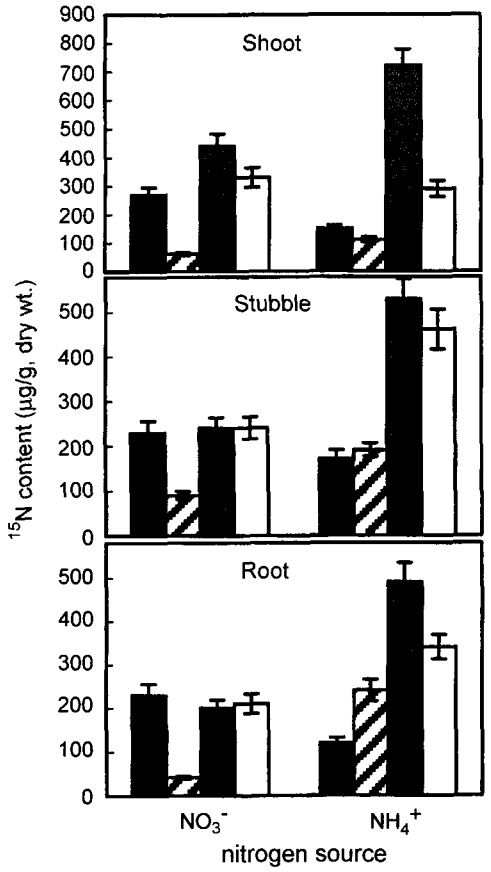


**Fig. 2.** $^{15}$N content in various N metabolites in each organ 7 days after $NO_3^-$ or $NH_4^+$ feeding.
■ : nitrate, ▨: amino acid, ■: insoluble protein, □ : soluble protein. Each value is the mean ± S.E. for n=5

of absorbed $^{15}$N during 7 days of feeding was 52.3 and 69.5 mg/plant. A large portion of absorbed $^{15}$N (67.4% in $NH_4^+$ feeding and 58.8% in $NO_3^-$ feeding) translocated into shoot. About 30.6% of $^{15}$N inflow to this organ was found in nitrate fraction in $NO_3^-$ feeding, while only 9.9% in $NH_4^+$ feeding. The sum of $^{15}$N incorporated into reduced N compounds (especially into the insoluble protein) in $NH_4^+$ feeding increased by about 50% comparing to $NO_3^-$ feeding. The $^{15}$N amount transferred to stubble in $NH_4^+$ and $NO_3^-$ feeding was 10.0 and 14.7 mg/plant, respectively. In $NO_3^-$ feeding, 34.1% of $^{15}$N inflow to this organ was remained at nitrate without further assimilation, while the percentage in $NH_4^+$ feeding largely decreased to 12.0%. The $^{15}$N amount incorporated to amino acids and insoluble proteins was more than 2-times in $NH_4^+$. The incorporation to soluble proteins was also significantly higher in $NH_4^+$ (5.2 mg/plant) than $NO_3^-$ feeding (3.2 mg/plant). The $^{15}$N amount transferred to roots in $NH_4^+$ and $NO_3^-$ feeding was 7.0 and 13.9 mg/plant, respectively. The incorporation to nitrate was 2.5 and 1.4 mg/plant, respectively, in $NO_3^-$ and $NH_4^+$ feeding, representing the lowest level of 3 organs in both two N supply forms. The $^{15}$N amount assimilated to amino acids in $NO_3^-$ feeding was only 0.4 mg/plant, while in $NH_4^+$ feeding remarkably increased to 2.9 mg/plant. The incorporation to insoluble and soluble proteins was also 3.1-fold and 1.7-fold higher, respectively, in $NH_4^+$ feeding

## DISCUSSION

Seven days of $NH_4^+$ feeding significantly increased total

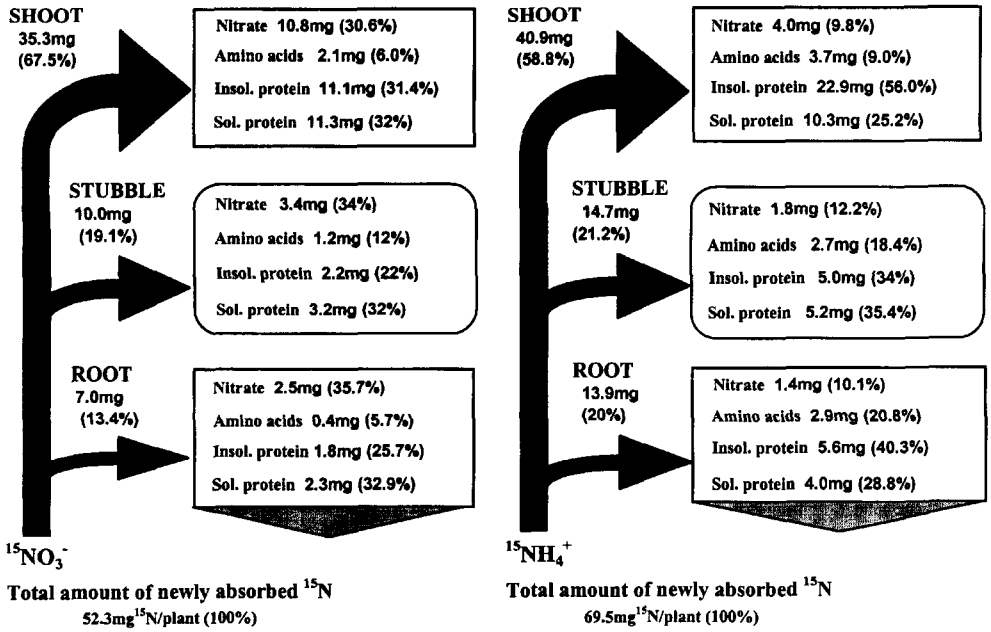


**Fig. 3.** Flow sheet of newly absorbed $^{15}$N into N compounds in three organs during 7 days of $^{15}NO_3^-$ or $^{15}NH_4^+$ feeding. Results are expressed in mg $^{15}$N/plant.

N content in three organ compared to $NO_3^-$ feeding (Table 2), although dry weight was not significantly different between two N supply forms (Table 1). The N content in amino acids and soluble proteins in all three organs was also significantly higher in $NH_4^+$ feeding. These indirectly indicated that the dynamics of N metabolism (uptake, assimilation and incorporation to organic N compounds) was significantly modified by the form of inorganic N source, as like $NH_4^+$ and $NO_3^-$ in this study. Uptake of the two inorganic N sources (anionic nitrate versus the cationic ammonium) differentially modify the uptake and accumulation of other inorganic cations and anions (Lang and Kaiser, 1994).

The $^{15}N$ amount found in nitrate fraction in of shoot and stubble fed with $NO_3^-$ was 2.1-fold higher than that of $NH_4^+$-grown plants (Fig. 2). $^{15}N$ distribution to nitrate was remarkable in root (Fig. 2). In agreement with Gojon *et al.* (1991) and Peuke *et al.* (1996) these results indicated that nitrate was largely transported to the actively growing tissues. Thus, the site of nitrate reduction within the whole plant seems to be under metabolic control, although the metabolic basis of this regulation is not yet known (Andrews, 1986). A simple explanation for the distribution of nitrate reduction between root and shoot may be the maximum capacity for nitrate reduction in the root. Once this capacity is exceeded, more nitrate will be transported to the shoot and stubble as revealed by the present data, and in consequence, should be reduced in these organs.

In contrast, under $NH_4^+$ nutrition, a significantly higher RSA value (Fig. 1) and a remarkable increase in $^{15}N$ content (Fig. 2) of amino acids were observed especially in roots. These suggested that the primary site of $NH_4^+$ assimilation was roots. A relative higher significant difference in roots for all reduced N compounds between two inorganic N supply treatments was also well consistent with this suggestion (Table 2). In several works of xylem sap analysis, very few $NH_4^+$ was found in the xylem sap of $NH_4^+$-grown plants, indicating that most of it was retained and assimilated in the root (Lewis et al., 1982; Murphy and Lewis, 1987; Van Beusichem et al., 1988; Allen et al., 1988).

The flow sheet of newly absorbed $^{15}N$ showed that total N uptake in $NH_4^+$-grown plants significantly increased by 32.8% compared to $NO_3^-$-grown plants (Fig. 3). This suggested that Italian ryegrass prefer $NH_4^+$ over $NO_3^-$ as source for inorganic nitrogen. It is well consistent with the results of Flaig and Mohr (1992) who reported that over the 21-day period, approximately three times more ammonium-N was taken up than nitrate-N. As both inorganic N sources, most of the N was allocated to the shoot (67.4% in $NH_4^+$ feeding and 58.8% in $NO_3^-$ feeding, respectively, Fig. 3), in agreement with data for *Lupinus* (71%, Jeschke et al., 1985), *Ricinus* (78%, Jeschke and Pate, 1991) and *Triticum* (80-95%, Lasson *et al.*, 1991).

The $^{15}N$ amount incorporated in the reduced N compounds (amino acids and proteins) in $NH_4^+$-grown plants significantly increased by 74.8% compared to $NO_3^-$-grown plants (Fig. 3). Plants grown with $NH_4^+$ have greater concentrations of free amino acids and protein-N in foliage than nitrate-fed plants (Clarkson *et al,* 1992; Geiger *et al.*, 1999) and the composition of total pool of free amino acids may vary with N-source (Barneix *et al.*, 1984; Lavoie *et al.*, 1992; Atilio and Causin, 1996). It is probably a consequence of differences in sites, patterns and rates of N assimilation, and associated with the interrelationships between the pools of soluble N and C in foliage. Assimilation of ammonium depends on the supply of C skeletons from TCA cycle, which may lead to a reduced concentration of soluble carbohydrates (Raab and Terry, 1995). The assimilation of $NH_4^+$ and the partitioning of assimilates over plant tissues are energetically less cost than those of $NO_3^-$ (Lewis *et al.*, 1986; Raven, 1985). Nitrate assimilation involves a similar anapleurotic synthesis of carboxylates, in addition to the synthesis of carboxylates that are required to maintain the cation-anion balance and intracellular pH, placing greater demands on C supply (Salsac *et al.*, 1987). In consequence, the concentration of carbohydrates is often lower in nitrate- than ammonium-fed plants (Chaillou *et al.*, 1991). Accompanied work of the present study showed that sugar concentration in $NH_4^+$-fed plants was higher than $NO_3^-$-fed plants (data not shown). These results give a direct evidence that $NH_4^+$ feeding is much more efficient and useful practice as a strategic for nitrogen fertilization.

## REFERENCES

Allen, S., J. A. Raven and J. I. Sprent. 1988. The role of long-distance transport in intracellular pH regulation in *Phaseolus vulgaris* grown with ammonium or nitrate as nitrogen source, or nodulated. *J. Exp. Bot.* 39 : 513-528.

Andrews, M. 1986. The partitioning of nitrate assimilation between root and shoot of higher plants. *Plant Cell Environ.* 9 : 511-519.

Atillio, B. and H. F. Causin. 1996. The central role of amino acids on nitrogen utilization and plant growth. *J. Plant Physiol.* 149 : 358-362.

Barneix, A. J., D. M. James, E. F. Watson and E. J. Hewitt. 1984. Some effects of nitrate abundance and starvation on metabolism and accumulation of nitrogen in barley (*Hordeum vulgare* L.) cv. Sonja. *Planta.* 162 : 465-476.

Bloom, A. J. 1988. Ammonium and nitrate as nitrogen sources for plant growth. *ISI atlas of science*, animal and plant science. Institute for Scientific Information, Philadelphia. PP. 55-59.

Chaillou, S., J. K. Vessey, J. F. Morot-Gaudry, C. D. Raper, L. T. Henry and J. P. Boutin. 1991. Expression of characteristics of ammonium nutrition as affected by pH of the root medium. *J. Exp. Bot.* 42 : 189-196.

Clarkson, D. T. 1986. Regulation of the absorption and release of nitrate by plant cells; a review of current ideas and methodology. *In* H Lambers, ed, Fundamental, Ecological and Agricultural Aspects of Nitrogen Metabolism in Higher Plants. Martinus Nijhoff, Dordrecht. PP. 3-27.

Clarkson, D. T., L. H. P. Jones and Purves, J.V. 1992. Absorption of nitrate and ammonium ions by Lolium perenne from flowing solution culture at low root temperatures. *Plant Cell Environ.* 15 : 99-106.

Copper, H. D. and D. T. Clarkson. 1989. Cycling of amino-nitrogen and other nutrients between shoots and roots in cereals-a possible mechanism integrating shoot and root in the regulation of nutrient uptake. *J. Exp. Bot.* 40 : 753-762

Flaig, H. and H. Mohr. 1992. Aaaimilation on nitrate and ammonium by the Scots pine (*Pinus sylvestris*) seedling under conditions of high nitrogen supply. *Physiol. Plant.* 84 : 568-576.

Geiger, M., V. Haake, F. Ludewig, U. Sonnewald and M. Stitt. 1999. The nitrte and ammonium nitrate supply have a major influence on the response of photosynthesis, carbon metabolism, nitrogen metabolism and growth to elevated carbon dioxide in tobacco. *Plant Cell Environ.* 22 : 1177-1199.

Gojon, A., C. Bussi, C. Grignon and L. Salsac. 1991. Distribution of $NO_3^-$ reduction between roots and shoots of peach-tree seedlings as affected by $NO_3^-$ uptake rate. *Physiol. Plant.* 82 : 505-512.

Jeschke, W. D., C. A. Atkins and J. S. Pate. 1985. Ion circulation via phloem and xylem between root and shoot of nodulated white lupin. *J. Plant Physiol.* 117 : 319-330.

Jeschke, W. D. and J. S. Pate. 1991. Modelling of the partitioning, assimilation and storage of nitrate within root and shoot organs of castor bean (*Ricinus communis* L.). *J. Exp. Bot.* 42 : 1091-1103.

Lang, B. and W. M. Kaiser. 1994. Solute content and energy status of roots of barley plants cultivated at pH on nitrate- or ammonium-nitrogen. *New Phytol.* 128 : 451-459.

Larsson, C-M., M. Larsson, J. V. Purves and D. T. Clarkson. 1991. Translocation and cycling through roots of recently absorbed nitrogen and sulphur in wheat (*Triticum aestivum*) during vegetative and generative growth. *Physiol. Plant.* 82 : 345-352.

Lavoie, N., L-P. V zina and H. A. Margolis. 1992. Absorption and assimilation of nitrate and ammonium ions by jack pine seedlings. *Tree Physiol.* 11 : 171-183.

Lewis, O. A. M., D. M. James and E. J. Hewitt. 1982. Nitrogen assimilation n barley (*Hordeum vulgare* L. *cv. Mazurka*) in response to nitrate and ammonium nutrition. *Ann. Bot.* 49 : 39-49.

Lewis, O. A. M., M. I. M. Soares and S. H. Lips. 1986. A photosynthetic and $^{15}N$ investigation of the differential growth response of barley to nitrate, ammonium and nitrate+ammonium nutrition. *In*: Lambers H et al., eds. *Fundamental, ecological and agricultural aspects of nitrogen metabolism in higher plants.* Dordrecht Martinus Nijhoff Publishers. PP. 295-300.

Marschner, H. 1995. Mineral Nutrition of Higher Plants, 2nd edn. Academic Press, London.

Murphy, A. T. and O. A. M. Lewis. 1987. Effect of nitrogen feeding source on the supply of nitrogen from root to shoot and the site of nitrogen assimilation in maize (*Zea mays* L. cv. R201). *New Phytol.* 107 : 327-333.

Oaks, A. and B. Hirel. 1985. Nitogen metabolism in roots. *Annu. Rev. Plant Physiol.* 36 : 345-365.

Peuke, A. D. and W. D. Jeschke. 1995. Effects of nitrogen source, nitrate concentration and salt stress on element and ion concentrations in transport fluids and on C and N flows in *Ricinus communis* L. *In*: Baluska F, Ciamporova M, Gasparikova O, Barlow P, eds. *Structure and function of root.* Dordrecht, Netherlands; Kluwer Academic Publishers, PP.229-236.

Peuke, A. D., J. Glaab, W. M. Kaiser and W. E. Jeschke. 1996. The uptake and flow of C, N and ions betwen roots and shoots in *Ricinus communis* L. . Flow and metabolism of inorganic nitrogen and malate depending on nitrogen nutrition and salt treatment. *J. Exp. Bot.* 47(296) : 377-385.

Raab, T. K. and N. Terry. 1995. Carbon, nitrogen and nutrient interactions in *Beta vulgaris* L. as influenced by nitrogen source, $NO_3^-$ versus $NH_4^+$. *Plant Physiol.* 107 : 575-584.

Raven, J. A. 1985. Regulation of pH and the generation of osmolarity in vascular plants: a cost-benefit analysis in relation to efficiency of use of energy, nitrogen and water. *N ew Phytol.* 101 : 25-77.

Salsac L., S. Chaillou, J-F. Morot-Gaudry, C. Lesaint and E. Jolivet. 1987. Nitrate and ammonium nutrition in plants. *Plant Cell Physiol.* 28 : 805-812.

Van Beusichem, M. L., E. A. Kirkby and R. Baas. 1988. Influence of nitrate and ammonium on the uptake, assimilation and distribution of nutrients in *Ricinus communis. Plant Physiol.* 86 : 914-921.