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NITRATE UPTAKE BY EXCISED ROOTS OF PLANT AND RATOON CROP OF SUGARCANE

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SUMMARY

Nitrate (NO_3^-) uptake by excised roots of sugarcane cultivars was studied by the method of depletion of NO_3^- from the nutrient medium. Excised roots of late maturing sugarcane cultivar CoS767 took up about 50% higher NO_3^- as compared to that of early maturing cv. CoJ64 during formative phase and 20% higher during grand growth phase of plant crop. Excised roots of ratoon crop of cv. CoS767 and CoJ64 showed 39% and 20% lower NO_3^- uptake rate respectively than similar roots of plant crop. Tissue NO_3^- content in top visible dewlap leaf of plant crop was marginally higher in late maturing cultivar (CoS767) as compared to that in early maturing type (CoJ64). The difference in tissue NO_3^- content vanished during grand growth phase of plant crop and was not observed during either stage in ratoon crop of the two cultivars. The difference in NO_3^- uptake rate and tissue NO_3^- content is discussed with respect to growth and maturity of sugarcane crop.

Key words **: Maturity group, nitrate uptake, sugarcane.**

INTRODUCTION

Sugarcane cultivars differ with respect to cane yield and maturity. CoJ64, a high sugar and early maturing cultivar is poor in cane yield as compared to CoS767, a late maturing type. Ratoon crop yields relatively less and mature early as compared to similar plant crop (Van Dillewijin 1952, Kidder 1977). Experiments at Hawaii indicated that ratoons required nearly 25% more nitrogen compared to plant crop for the same level of yield (Borden 1944). Studies conducted later emphasized requirement of more nitrogen (Plucknett *et al.* 1970) and lower nitrate reductase activity in the leaves of ratoon crop as compared to that of plant crop (Rai *et al.* 1989). NO_3^- is not only a nutrient, but it also acts as a signal for the initiation of various processes (Tischner *et al.* 1993, Tischner 2000). It has been shown that some of the genes involved in photosynthesis, cell cycling and translation machinery are regulated, at least in part, by NO_3^- (Takei *et al.* 2002). Carbohydrate metabolism is also affected by the presence of NO_3^-, which shifts the starch synthesis in favor of sucrose synthesis (Crawford 1995) and may thus affect growth and sucrose accumulation in sugarcane. However, investigations on NO_3^- uptake in sugarcane cultivars differing in growth rate and maturity group are rare. The present work aims at comparing the NO_3^- uptake rate by excised roots of plant and ratoon crop and of variable maturity group of sugarcane.

MATERIALS AND METHODS

Sugarcane hybrids (*Saccharum spontaneum* L. x S. *Officinarum* L.), CoJ64 (early maturing) and CoS767 (late maturing) were planted in the field during 1st week of March at experimental farm of CCS HAU regional research station at Karnal, following recommended cultural practices. Roots were excised from the settlings so raised during 1st week of June (Formative phase) and August (Growth phase), washed under running tap water and rinsed with distilled water. NO_3^- uptake by the root

samples so prepared was determined. In some experiments roots from clumps of ratoon crop were excised where the plant crop was harvested during 3rd week of January.

Nitrate uptake

Nitrate uptake was measured as the amount disappeared from the nutrient solution. About 5 g of roots excised from settlings of plant/ratoon crop raised as explained above were incubated in 15 ml of nutrient medium containing 0.7 mM KNO_3 for 6 h. The nutrient medium was one-quarter strength Hoagland solution (Hoagland and Arnon 1950) buffered with 0.5 mM HEPES (N-2 Hydroxyethyl dipiperazine N'-2 ethane sulphonic acid) at pH 5.75 ± 0.1. No loss of NO_3^- was noticed from controls without roots. All uptake assays were performed after an initial exposure of roots to 0.7 mM KNO_3 for 10 minutes. The nitrate uptake was expressed as nmol NO_3^- g^{-1} root fw h^{-1}. Nitrate from the nutrient medium before and after the experiment was assayed spectrophotometrically by taking direct absorbance of the nutrient medium at 210 nm (Thayer and Huffaker 1980). Quarter strength Hoagland solution lacking NO_3^- (showing negligible absorbance at 210 nm) was used as blank and the values computed from a standard curve having 10-50 µM NO_3^-.

Tissue nitrate concentration

Tissue NO_3^- concentration was analyzed in top visible dewlap (TVD) leaf of plant as well as ratoon crop of two sugarcane cultivars following the method described by Cataldo *et al.* (1975). Dry leaf sample (500 mg) was homogenized with 5 ml distilled water, incubated at 45°C for 1 h and centrifuged at 5000 g for 15 min. The supernatant (0.5 ml) was mixed with 1.5 ml of 5% salicylic acid in conc H_2SO_4. After 20 min 7.6 ml of 10 N NaOH was added to raise the pH above 12 and absorbance recorded at 410 nm. For blank, H_2SO_4 substituted salicylic acid solution.

RESULTS

Nitrate uptake by excised roots of sugarcane

Excised roots from late maturing sugarcane cultivar CoS767 took up more NO_3^- from the nutrient bathing medium during formative as well as grand growth phase of plant as well as ratoon crop (Fig. 1). The effect being more pronounced during formative phase of plant crop

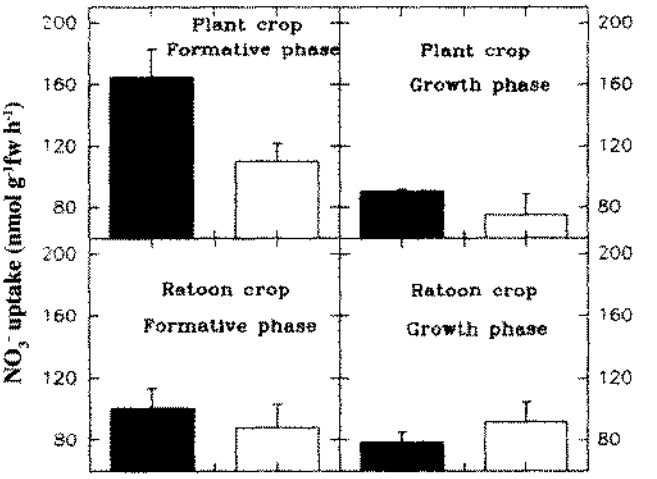

Fig. 1. NO_3^- uptake by excised roots of sugarcane cv. CoS767 (■) and CoJ64 (□)

(Excised roots from sugarcane plant/ratoon crop was incubated in quarter strength Hoagland solution having 0.7 mmolar KNO_3. Depletion of NO_3 from the bathing medium was assayed as described in material and methods. Values are mean of three independent assays ± S.E.).

when 164.7 nmol NO_3^- g^{-1}fw h^{-1} was recorded as compared to 110.1 nmol g^{-1}fw h^{-1} by roots of early maturing CoJ64. During growth phase an uptake of 90.4 nmol NO_3^- g^{-1}fw h^{-1} was recorded by roots of CoS767 as compared to 75.2 nmol g^{-1}fw h^{-1} observed by roots of CoJ64. Excised roots from ratoon crop of CoS767 as well as CoJ64 were less efficient in NO_3^- uptake as compared to similar roots from plant crops. About 39% lower uptake was recorded by roots of ratoon crop of CoS767 as compared to similar roots of plant crop during formative phase. Corresponding decrease in uptake was about 20% for roots of early maturing CoJ64. During growth stage, only 13% decrease in NO_3^- uptake was observed by roots of CoS767 as compared to no significant change in NO_3^- uptake by roots of CoJ64.

Tissue nitrate concentration

During formative phase, marginally higher NO_3^- content was observed in TVD leaf of plant crop of late maturing cv. CoS767 as compared to that in early maturing cv. CoJ64 (Fig. 2). About 1000 nmol NO_3^- g^{-1}dw was recorded in CoS767 as compared to 916 nmol g^{-1}dw found in early maturing CoJ64. However, in ratoon crop similar NO_3^- content (about 800 nmol g^{-1}dw) was observed in the leaves of two cultivars. During growth phase also, there

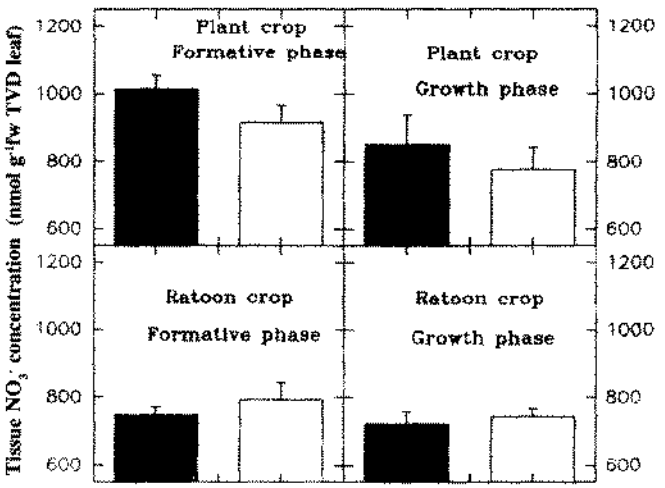

Fig. 2. Tissue NO_3^- concentration in TVD leaf of sugarcane cv. CoS767 (■) and CoJ64 (□) (Values are mean of three independent estimations ± S.E.).

was no difference in NO_3^- content of TVD leaf of two maturity groups when the concentration was about 700 nmol g^{-1}dw in ratoon crop and about 800 nmol g^{-1}dw in plant crop.

DISCUSSION

These results indicate that roots of plant crop are more active in NO_3^- uptake than that of ratoon crop and take up more nitrogen during formative phase than during grand growth phase. Lower activity during growth phase and during both the phases in ratoon crop may be due to lignification and suberization of the roots at maturity affecting hydraulic conductivity of the roots. Though, development of new roots is a continuous process, yet fraction of new roots as compared to total root biomass may be less during growth stage as compared to that during formative phase, when almost all the roots are young and more active. The difference in uptake by early and late maturing cultivars indicate that it may have to do something with the growth and sucrose accumulation pattern of the two cultivars. NO_3^- is known to affect carbohydrate metabolism shifting from starch accumulation to sucrose accumulation leading to increase in the availability of organic acids to utilize available nitrogen for growth (Crawford 1995). Practically no difference in NO_3^- content in the leaves indicate that higher growth rate in late maturing CoS767 might have utilized the more available NO_3^-. High nitrate reductase activity reported in plant crop as compared to that in ratoon crop (Rai *et al.* 1989) further supports the hypothesis.

Another important aspect is the allocation of resources for growth or sucrose accumulation in sugarcane. It has been known for a long time that the allocation of resources during vegetative growth depends on the availability of nitrogen and other nutrients (Brower 1962). Moderate nitrogen deficiency inhibits shoot growth and even stimulates root growth (Agren and Ingestad 1987, Fichtner and Schulze 1992, Wagner and Beck 1993). It is more likely that relative allocation of metabolites for growth or sucrose accumulation are also modulated by signals related to the nitrogen status of sugarcane plants. Many studies (Huber 1983, Chu *et al.* 1992, Buysse *et al.* 1995, Ericsson 1995) have found co-relations between levels of starch, sugars, amino acids, and shoot root allocations when the nitrogen supply is altered. A similar change in allocation from growth to sucrose accumulation might be due to lower nitrogen availability in sugarcane. This hypothesis is supported by the following observations. (I) Early maturing and high sugar cv. CoJ64 recorded lower NO_3^- uptake activity as compared to late maturing cv. CoS767. (II) Roots of ratoon crop (that matures earlier than plant crop) of both the cvs. recorded lower NO_3^- uptake activity than corresponding roots of plant crop. (III) Tissue NO_3^- content in the TVD leaf of late maturing cv. CoS767 was marginally higher than that in early maturing cv. CoJ64. (IV) Tissue NO_3^- content in the leaves of ratoon crop was lower than that in the corresponding plant crop. Additionally, transition from low NO_3^- to high NO_3^- nutrition in wheat has been shown to affect sucrose production severely while enhancing phosphoenolpyruvate carboxylase activity (Van Quy *et al.* 1991a, b). In C_4 plants, like sugarcane, phosphoenolpyruvate carboxylase is of primary importance because it is involved in the direct pathway of conversion of HCO_3^- to carbohydrate in photosynthesis (Coombs 1979). Its activity has been shown to be modulated by NO_3^- in maize (Foyer *et al.* 1994) and sugarcane (Abellan *et al.* 1994) in a similar manner to that observed in wheat. Therefore, it appears likely that high NO_3^- uptake activity of sugarcane roots might be responsible for high growth and low sucrose accumulation activity perhaps via its effect on phosphoenolpyruvate carboxylase activity. The above observations and the

foregoing discussion support the hypothesis that allocation of metabolites for growth or sucrose accumulation is altered by availability and tissue concentration of NO_3^-.

REFERENCES

Abellan, I.P., Urguiza, R.D.A., Valadier, M.H. and Champigny, M.L. (1994). Short term effect of nitrate on carbon metabolism of two sugarcane cultivars differing in their biomass production. *Phytochemistry* **16**: 819-823.

Agen, G.I. and Ingested, T. (1987). Root : shoot ratio is a balance between nitrogen productivity and photosynthesis. *Plant Cell Environ.* **10** : 579-586.

Borden, R.J. (1944). Crop relationships with special reference to nitrogen efficiency. *Hawaiian Plant Rec.* **48**: 65-72.

Brower, R. (1982). Nutrient influences on the distribution of the dry matter in the plant. *Netherlands J. Agric. Sci.* **10** : 399-408.

Buysse, J., Smolders, E. and Merkx, R. (1995). The role of free sugars and amino acids and its regulation of biomass partitioning and plant growth. *Plant Soil* **155** : 191-194.

Chu, C.C., Coleman, J.S. and Mooney, H.A. (1992). Control of biomass partitioning between roots and shoots : atmospheric carbon dioxide enrichment and the acquisition and allocation of carbon and nitrogen in wild radish. *Oecologia* **89** : 580-587.

Coombs, J. (1979). Photosynthesis II. Photosynthetic carbon metabolism and related processes. In : M. Gibbs and E. Latzko (eds.), Encyclopedia of Plant Physiology. New series, Vol. 6, pp. 251. Springer, Berlin.

Crawford, N.M. (1995). Nitrate : nutrient and signal for plant growth. *Plant Cell* 7:859-868.

Cataldo, D.A., Haroon, M., Schrader, L.E. and Youngs, V.L. (1975). Rapid colorimetric determination of nitrate in plant tissue by nitration of salicylic acid. *Commun. Soil and Plant analysis* **6** : 71-80.

Ericson, T. (1995). Growth and shoot : root allocation of seedlings in relation to nutrient availability. *Plant Soil* **168** : 205-214.

Fichtner, K. and Schulze, E.D. (1992). The effect of nitrogen nutrition on annuals originating from habitats of different nitrogen availability. *Oecologia* **92** : 236-241.

Foyer, C., Noctor G., Lelandais M., Lescure, J.C., Valadier, M.H., Boutin, J.P. and Hortron, P. (1994). Short term effects of nitrate, nitrite and ammonium assimilation on photosynthesis, carbon partitioning and protein phosphorylation in maize. *Planta* **192** : 211-220.

Hoagland, D.R. and Arnon, D.I. (1950). The Water Culture Method for Growing Plants Without Soil. Cir. No. 347, California Agricultural Experiment Station.

Huber, S.C. (1983). Relation between photosynthetic starch formation and dry weight partitioning between the shoot and root. *Can J. Bot.* **61** : 2709-2716.

Kidder, G. (1977). Production differences between plant cane and ratoon crops of Florida sugarcane. *Proc. Amer. Sugarcane Technol.* 67-70.

Plucknett, D.L., Evenson, J.P. and Sanford, W.G. (1970). Ratoon cropping. *Adv. Agron.* **22** : 285-330.

Rai, R.K., Shrivastava, A.K., Ghosh, A.K. and Shukla, S.P. (1989). Nitrate reductase activity *in vivo* in plant and ratoon crop of sugarcane. *Indian J. Plant Physiol.* **4** : 320-324.

Takei, K., Takahashi, T., Sugiyama, T., Yamaya, T. and Hitoshi, S. (2002). Multiple routes communicating nitrogen availability from roots to shoots : a signal transduction pathway mediated by cytokinin. *J. Exp. Bot.* **53** : 971-977.

Thayer, J.R. and Huffaker, R.C. (1980). Determination of nitrate and nitrite by high pressure liquid chromatography : comparison with other methods for nitrate determination. *Anals Biochem.* **102** : 110-119.

Tischner, R. Waldeck, B., Goyal, S.S. and Rains, W.D. (1993). Effect of nitrate pulses on the nitrate-uptake rate, synthesis of mRNA coding for nitrate reductase, and nitrate reductase activity in the roots of barley seedlings. *Planta* **189** : 533-537.

Tischner, R. (2000). Nitrate uptake and reduction in higher and lower plants. *Plant Cell. Environ.* **23** : 1005-1024.

Van Dillewijn, C. (1952). Botany of Sugarcane. The Chronica Botanica Co., Waltham, Mass, USA.

Van Quy, L., Foyer, C. and Champigny, M.L. (1991a). Effect of light and NO_3^- on wheat leaf phosphoenolpyruvate carboxylase activity. *Plant Physiol.* **97** : 1476-1482.

Van Quy, L., Lamaze, T. and Champigny, M.L. (1991b). Short term effects of nitrate on sucrose synthesis in wheat leaves. *Planta* **185** : 53-57.

Wagner, B.M. and Beck, E. (1993). Cytokinins in the perennial herb *Urtica dioica* L. as influenced by its nitrogen status. *Planta* **190** : 511-518.