Pak. J. Bot., 38(5): 1703-1708, 2006.

ION TRANSPORT IN FOUR CANOLA CULTIVARS AS INFLUENCED BY SALT STRESS

QASIM ALI[1*], HABIB-UR-REHMAN ATHAR[2] AND MUHAMMAD ASHRAF[1]

Department of Botany, University of Agriculture, Faisalabad, Pakistan
Institute of Pure and Applied Biology, Bahauddin Zakariya University, Multan, Pakistan

Abstract

In order to assess relationship between ion transport and variation in growth responses of four canola cultivars under salt stress, four canola cultivars were grown at 0 and 150 mM NaCl for six weeks in hydroponics. Salt stress reduced the shoot and root fresh and dry weights of all canola cultivars. Maximum shoot fresh and dry weights were recorded in Dunkeld under both non-saline and saline conditions, while cv. Cyclone had the minimum shoot and root fresh and dry weights of all cultivars examined under both stress and non-stress conditions. On the basis of growth of canola cultivars under salt stress Dunkeld was found to be salt tolerant, CON-III and Rainbow intermediate and Cyclone as salt sensitive cultivar. Transport of Na^+ and Cl^- were increased due to salt stress with a decrease in K^+ and Ca^{2+} transport in all canola cultivars but transport of cations (Na^+, K^+, and Ca^{2+}) decreased consistently over time. Furthermore, salt tolerant Dunkeld had the highest K^+ and Ca^{2+} transports with a minimum Na^+ transport to the leaves under saline conditions. Cl^- transport remained almost unchanged over time under both non-saline and saline conditions. These results suggested that salt tolerant Dunkeld might have a key mechanism of ion exclusion and/or transport-restriction between the shoot and root to depress the transport of Na^+ and Cl^- to the upper plant parts, enabling a higher tolerance to NaCl.
Keywords: *Brassica napus*, ion selectivity, screening and selection, salt tolerance

Introduction

Selection of improved and high yielding cultivars having wide range adaptations to soil and environmental conditions has been an essential role of crop breeders to increase the crop yield per hectare (Ashraf & Foolad, 2007). Plant performance usually expressed as crop yield, plant biomass or crop quality, which adversely effected by the salinity induced nutritional disorders (Ashraf, 1994; 2004). These disorders may result from the effect of salinity on nutrient availability, competitive uptake, transport or partitioning within the plant (Munns, 2005). The capacity of plants to tolerate high levels of salinity depends on the ability to exclude salt from the shoot, or to tolerate high concentrations of salt in the leaf (tissue tolerance). It is widely held that a major component of tissue tolerance is the capacity to compartmentalize salt into safe storage places such as vacuoles (Ashraf, 2004; Munns, 2007).

It is evident from different reports that glycophyte can use both ion exlusion and inclusion mechanism in response to different saline substrates. These two mechanisms depend on pattern of ion distribution between leaves and on ion compartmentation within the cells (Tester & Davenport, 2003; Munns, 2005). As a further complication time courses of ion accumulation can be different in an organ specific way (Ashraf et al., 2003). Therefore, only with a full understanding of the ion response mechanism of particular species would ion content measurements *per se* serve as selection indicators.

[*] Corresponding author: Qasim Ali; qasimbot_uaf@yahoo.com

Of the different oilseed crops, canola is an important source of vegetable oil (Ashraf & McNeilly, 2004), which has been primarily developed for its low erucic acid contents (< 2% of total fatty acids) in the seed oil and low glucosinolates in the seed meal (≤ 30 µmole g^{-1} seed meal) (Francoise, 1994). The crop has a great potential to grow in salt affected areas. Although a large number of reports are available in the literature on salinity tolerance of canola, there is little information on the ion transport.

The primary objective of the present study was to investigate the transport of different ions from root to shoot in some differentially adapted canola cultivars to salinity stress, which will be useful in understanding the salt tolerance mechanism in different cultivars of canola at early growth stages.

Material and Methods

The present study was conducted to investigate the influence of salinity on growth, and different ions transport to the shoots in four canola (*Brassica napus* L.) cultivars, Dunkeld, CON-III, Rainbow and Cyclone. The seeds of four canola cultivars were obtained from the Ayub Agricultural Research Institute, Faisalabad. Before experimentation, all the seed samples were surface sterilized with 10% sodium hypochlorite solution for five minutes and washed three times with sterilized distilled water. The experiment was conducted in the Botanic Garden of the University of Agriculture Faisalabad during 2005-2006, where the average PAR measured at noon ranged from 848 to 1254 $\mu mol/m^2.s^{-1}$, day/night relative humidity 58/74 %, and temperature 24/8 ^oC. Sodium chloride was used as a source of salt.

Hundred seeds of each of four canola cultivars were sown in Petri plates supplied with Hoagland's nutrient solution or nutrient solution containing 150 m*M* NaCl. The experiment comprised two treatments with four replications and four canola cultivars. After the establishment of seedlings, ten seedlings of each cultivar per replicate were exposed to aerated Hoagland's nutrient solution with or without 150 m*M* NaCl for five weeks. The plants were harvested after five weeks of the initiation of salt treatment and separated into shoots and roots. After measuring fresh masses of shoots and roots, the samples were dried at 65°C for one week so as to measure dry masses.

Determination of Na^+, K^+, Ca^{2+} and Cl^- in leaves and roots: The oven-dried ground material (0.1 g) of leaves and roots was digested with 2 ml of sulfuric acid-hydrogen peroxide mixture according to the method of Wolf (1982). Potassium, sodium and calcium in the digests were determined with a flame photometer (PFP-7, Jenway Ltd. Felsted, Dunmow, Essex, U.K). Cl^- was determined with a chloride analyzer (Sherwood, Model 926).

Transport of ions: Transport of different ions to the shoots was calculated following Hunt (1982). Ion transport to the shoot was determined as the amount of the ion transported to the shoot on a root dry basis per day

$$\text{Transport} = \frac{M_2 - M_1}{T_2 - T_1} \times \frac{\ln W_2 - \ln W_1}{W_2 - W_1}$$

Where *M* is the amount of the ion in the shoot, *T* is the time and *W* is the root weight

Statistical analysis: Analysis of variance technique was employed for carrying out statistical analysis of the data collected using a Costat 6.33 computer package (Cohort Software, California, USA). The mean values were compared with the least significant difference test (LSD) following Snedecor & Cochran (1980).

Results

Salt treatment significantly reduced the shoot and root fresh weights ($P < 0.001$) of all four cultivars. Maximum shoot fresh weights were recorded in Dunkeld and CON-III under both non-saline and saline conditions. Cultivar Cyclone had the minimum shoot and root fresh weights of all cultivars examined under both stressed and non-stressed conditions (Fig. 1) significant reduction in dry weights of shoots and roots were observed in all the cultivars under salt stress ($P < 0.001$). Cultivar Dunkeld had the maximum shoot dry weight under both saline and control conditions, whereas the minimum dry weight was observed in cv. Cyclone under both saline and control conditions (Fig. 1).

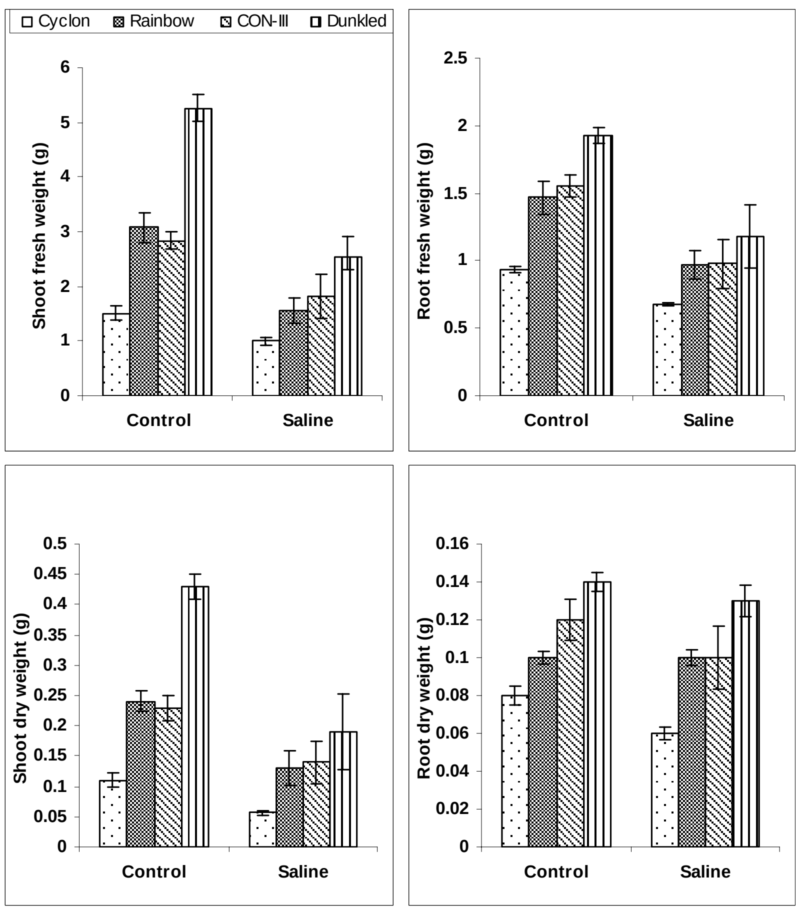

Fig. 1. Fresh and dry weights of shoots of four canola (*Brassica napus* L.) cultivars when six day old seedlings were subsequently allowed to grow for 30 days under normal or saline conditions.

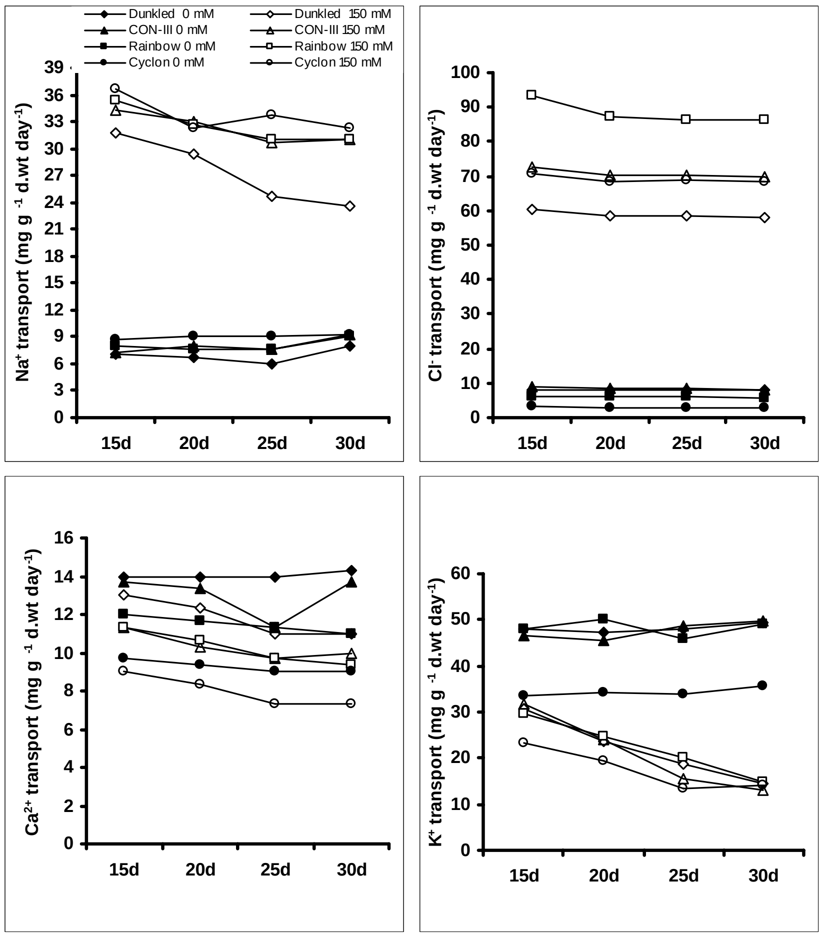

Fig. 2. Transport of Na^+ Ca^{2+}, K^+ and Cl^- to shoots of four canola (*Brassica napus* L.) cultivars when six day old seedlings were subsequently allowed to grow for 30 days under normal and saline conditions.

Imposition of salt stress caused an increase in Na^+ translocation to the shoots of all the canola cultivars. The cultivars did not differ significantly under non-saline conditions in Na^+ transport to the shoots. However, under saline conditions cultivars differed in Na^+ transport to the shoots and the minimum transport was recorded in cv. Dunkeld which was further decreased with the passage of time. This time interval decrease was more prominent in salt tolerant cv. Dunkeld (Fig. 2)

Rooting medium salinity increased Cl^- transport in the shoots of all four cultivars. Cultivars differed significantly in Cl^- transport under salt stress. The maximum transport was observed in cv. Rainbow and cv. Dunkeld was the minimum in Cl^- transport under salt stress. However, the increase or decrease in Cl^- transport with time remained unchanged in all the cultivars under saline or non-saline conditions (Fig. 2)

Ca^{2+} transport to the shoots of all the cultivars decreased with the imposition of salt stress in the rooting medium. The maximum decrease in Ca^{2+} transport to the shoots under salt stress was recorded in salt sensitive cv. Cyclone and minimum in salt tolerant cv. Dunkeld. However, the decrease in Ca^{2+} transport to the shoots with increasing time remained unchanged in all cultivars under both saline and non-saline conditions (Fig. 2).

Imposition of salt stress caused reduction in K^+ transport to the shoots of all cultivars. Under saline or non-saline conditions the minimum K^+ transport was recorded in cv. Cyclone, but the other cultivars did not differ in K^+ transport under saline or non-saline conditions. However, K^+ transport was further decreased at varying time intervals, particularly 30 day after the application of salinity (Fig. 2).

Discussion

Salinity has long been identified as one of the most pervasive environmental hazards, limiting crop production mostly in arid regions of the world (Ashraf 1994; 2004; Munns, 2007). Although plants have the ability to adapt the salt stress, it depends on the type of a species or even a cultivar (Ashraf, 2004). A great magnitude of variation for salt tolerance at inter-specific and intra-specific levels has earlier been reported in a number of crop species e.g., canola (Ashraf & Sharif 1998), wheat (Sairam *et al*. 2002), tomato (Cuartero *et al*., 2002). From the results of the present study, the inter-cultivar variation for salt tolerance was also found in canola cultivars (Dunkeld being salt tolerant, CON-III and Rainbow intermediate and Cyclone salt sensitive). This variation in salt tolerance among canola cultivars might have been due to differential effects of salt stress on physiological and biochemical processes of these canola cultivars.

Plants used various mechanisms to cope with the deleterious effects of salinity stress. Of various determinants of salt tolerance, maintenance of ion homeostasis under salt stress is the most important one, which is generally achieved by exclusion of toxic ions and higher uptake of K^+ or Ca^{2+}. In the present study, Na^+ and Cl^- were more accumulated in the roots and leaves of the salt stressed plants of all cultivars, and cultivars also differed in accumulation of Na^+ and Cl^- in the leaves (data not shown). For example, salt tolerant cv. Dunkeld had lower Na^+ but higher K content in their shoots and roots in comparison with other canola cultivars. These results suggest that salt tolerant Dunkeld might have a key mechanism of ion exclusion and/or transport-restriction between the shoot and root to depress the transport of Na^+ and Cl^- to the upper plant parts, enabling a higher tolerance to NaCl. However, in the present study, transport of Na^+ and Cl^- were increased due to salt stress with a decrease in K^+ and Ca^{2+} transport in all canola cultivars, but transport of cations (Na^+, K^+, and Ca^{2+}) decreased consistently over time. Furthermore, salt tolerant Dunkeld had the highest K^+ and Ca^{2+} transports with a minimum Na^+ transport to the leaves under saline conditions. These results are similar to those of Ashraf & Khanum (2000) in which they found that salt tolerance of wheat cultivars was associated with low transport of Na^+. It is well evident that plant adopted different strategies to limit Na^+ transport to the leaves to protect photosynthesizing tissues, a very little is known about mechanisms regulating the Na^+ transport. Recently, Hussain *et al*. (2004) reported that limited Na^+ transport to shoot in the salt tolerant wheat cultivar was due to a regulation of xylem loading transporters. While working with rice, Anil *et al*. (2005) found that restricted uptake of Na^+ depends on Ca^{2+} and regulated by transpirational bypass flow in salt sensitive and salt tolerant rice cultivars, but with differing efficiencies and set points. They also found that nutrient transport from the roots to the shoots is also restricted by the reduced transpiration rates (Anil *et al*., 2005). However, the ability of plant cells to maintain ion homeostasis, plants use a number of transporters (Tester & Davenport, 2003). Therefore, differences in limited Na^+ transport among canola cultivars observed in the present study might have been due to differences in cation transport systems at different stages and it needs to be further elucidated.

In conclusion, the higher growth of salt tolerant Dunkeld under saline conditions as compared to other canola cultivars is associated with low transport of Na^+ and relatively higher K^+ and Ca^{2+} transport to the shoots.

Acknowledgement

The authors whish to acknowledge the financial support of Higher Education Commission to the principal author through a research grant No. 118.

References

Anil, V.S., P. Krishnamurthya, S. Kuruvillab, K. Sucharithaa, G. Thomasb and M. K. Mathew. 2005. Regulation of the uptake and distribution of Na^+ in shoots of rice (*Oryza sativa*) variety Pokkali: role of Ca^{2+} in salt tolerance response. *Physiol. Plant.,* 124: 451–464.

Ashraf, M, and M. R. Foolad. 2007. Roles of glycinebetaine and proline in improving plant abiotic stress resistance. *Env. Exp. Bot*., 59: 206-216.

Ashraf, M. and A.Khanum. 2000. Transport and accumulation of ions in two spring wheat lines differing in salt tolerance. *Acta Physiol. Plant*., 22(2):103-110

Ashraf, M. and T. McNeilly. 2004. Salinity tolerance in *Brassica* oilseeds. *Crit. Rev. Plant Sci*., 23(2): 157-174.

Ashraf, M. 1994. Breeding for salinity tolerance in plants. *Crit. Rev. Plant Sci*., 13: 17-42.

Ashraf, M. and R. Sharif. 1998. Does salt tolerance vary in a potential oilseed crop *Brassica carinata* at different growth stages? *J. Agron. Crop Sci*., 181: 103–115.

Ashraf, M., S. Hanif, M.Y. Ashraf, S. Rehman and E.S.Rha. 2003. Time course changes in nutrient composition of hexaploid wheat (*Triticum aestivum* L.) cultivars differing in yield potential. *Agrochimica*., 3-4

Ashraf, M. 2004. Some important physiological selection criteria for salt tolerance in plants. *Flora*, 199: 361-376.

Cuartero. J., R. Romero-Aranda, A. R. Yeo and T. J. Flowers. 2002.Variability for some physiological characters affecting salt tolerance in tomato. *Acta Hort*., 573: 435–441,

Francoise, L. E. 1994. Growth, seed yield and oil contents of canola grown under saline conditions. - *Agron. J.,* 86: 233-237.

Hunt, R.1982. Plant growth curves. An introduction to the functional approach to plant growth analysis. Edward Arnold, London.

Husain S., S.V. Caemmerer and R. Munns. 2004. Control of salt transport from roots to shoots of wheat in saline soil. *Func Plant Biol.,* 31: 1115–1126.

Munns, R. 2005. Genes and salt tolerance: bringing them together. *New Phytol*., 167: 645-663.

Munns, R. 2007. Utilising genetic resources to enhance productivity of salt-prone land. CAB Reviews: Perspectives in Agriculture, Veterinary Science, Nutrition and Natural Resources 2, No. 009. doi: 10.1079/PAVSNNR20072009

Sairam, R. K., K. V. Rao and G. C. Srivastava. 2002. Differential response of wheat genotypes to long-term salinity stress in relation to oxidative stress, antioxidant activity and osmolyte concentration. *Plant Sci*., 163: 1037-1046.

Snedecor, G. W. and W. G. Cochran. 1980. Statistical methods. 7^{th} Edition. Lowa Stat University Press, Ames, IOWA.

Tester, M. and R. Davenport. 2003. Na^+ tolerance and Na^+ transport in higher plants. *Ann. Bot.,* 91: 503-527.

Wolf, B. 1982. A comprehensive system of leaf analysis and its use for diagnosing crop nutrient status. *Commun. Soil Sci. Plant Anal*., 13: 1035-1059.

(Received for publication 18 September, 2006)