Turk J Biol
29 (2005) 233-236

Exogenous Application of Gibberellic Acid Counteracts the Ill Effect of Sodium Chloride in Mustard

Shaheena AFROZ, Firoz MOHAMMAD, Shamsul HAYAT, Manzer H. SIDDIQUI
Plant Physiology Section, Department of Botany, Aligarh Muslim University, Aligarh, 202002 - INDIA

Received: 16.03.2005

Abstract: The response of mustard (*Brassica juncea* (L.) Czern & Coss cv. Varuna) to presowing seed treatment with sodium chloride (NaCl) was investigated. The plants raised from treated seeds were sprayed with water or 10^{-6} M GA_3 at the 30- day stage. The seeds imbibed in 1 or 10 mM of NaCl resulted in a decrease in dry mass, leaf chlorophyll content, carbonic anhydrase activity (E.C. 4.2.1.1), nitrate reductase activity (E.C. 1.6.6.1) and net photosynthetic rate at the 60-day stage, and pod number and seed yield at harvest. However, spray application of GA_3 neutralized the ill effect of soaking treatment in NaCl (1 or 10 mM).

Key Words: *Brassica juncea*, carbonic anhydrase, chlorophyll, nitrate reductase, salinity, seed yield.

Abbreviations: CA-carbonic anhydrase; DAS-days after sowing; DH- dry mass; NR-nitrate reductase; P_N-net photosynthetic rate.

Introduction

Brassica juncea (Indian mustard or brown sarson) is an important oil yielding crop, accounting for approximately 80% of the total production of rape seed and mustard in India (1). India is second in the world with regard to the production of Brassicas. The susceptibility of different varieties to salinity, drought, diseases and pests is the major cause of their low productivity. Salinity affects at least 20% of the world's arable land and more than 40% of irrigated land to various degrees (2). Salinity influences adversely several aspects of reproductive growth, including flowering, pollination, fruit development, yield and quality and seed production (3-6). It also inhibits the activity of many enzymes (7-10), photosynthesis (10,11), absorption of minerals (12) and respiration (13).

Plant growth regulators are known to affect growth, and assimilate translocation, flowering on transport (14-16). Although the effect of gibberellic acid on growth, photosynthesis, enzyme activities and productivity have been well studied (16-18) and its effect on salinity-induced inhibition was studied on various crops (6,19-21) but pertinent information with regard to mustard is not available. Therefore, the present experiment was carried out to investigate whether GA_3 could ameliorate the influence of salt stress on mustard, thereby increasing its salt tolerance.

Materials and Methods

Seeds of *Brassica juncea* (L.) Czern & Coss cv. Varuna were purchased from the National Seed Corporation Ltd., New Delhi, India. Healthy seeds of uniform size after surface sterilization with 5% NaOCl solution for 5 min were soaked in 0 (water), 1 and 10 mM NaCl for 6 h (durations were selected on the basis of a preliminary experiment). The treated seeds were then sown in earthen pots (25 cm diameter) filled with soil plus farmyard manure in the ratio of 9:1. The pots were kept in rows in a net house and irrigated regularly with tap water as and when required. The raised plants at 30 days after sowing (vegetative stage) were sprayed with water or 10^{-6} M of aqueous solution of GA_3. The concentration of GA_3 is based on our earlier experiment (16). A constant volume (50 ml) was sprayed in all cases with a manual pump. The plants were divided into 4 groups: (i) control (water soaked), (ii) plants received NaCl solution at 1 and 10 mM, (iii) plants treated with GA_3 (10^{-6}M), (iv) plants treated with GA_3 plus each of the salinity levels. Each treatment was replicated 5 times. DM (at 80 °C), photosynthetic rate (P_N), leaf chlorophyll (a+b) content, and carbonic anhydrase (CA) and nitrate reductase (NR) activities were assessed in the leaves of 60-day-old plants (flowering stage). The pod number and seed yield were recorded at harvest Chlorophyll content was estimated following the method described by Mackinney (22). The CA and NR activities were determined by adopting the standard procedures (23,24). P_N in intact leaves was measured with the help of an LI-6200 portable photosynthesis system (LI-COR, Lincoln, NE, USA). The data were statistically analyzed using the least significance difference (LSD) test (25).

Results and Discussion

The application of NaCl to *Brassica* plants adversely influenced the dry mass, chlorophyll, NR, CA and P_N compared with the control plants. However, GA_3-treated plants exhibited an increase in tolerance to salt treatment (Table). This improvement in salt tolerance was reflected in the above parameters where all the parameters have higher values compared with plants receiving only NaCl treatment (Table).

The data presented in the Table show that CA, NR and P_N were significantly reduced in salinized mustard plants compared with the control plants. As it is well known that salinity-stressed plants have a consistently lower photosynthetic rate in other plants (10,11,26). The reduction in photosynthesis of plants raised from NaCl-soaked seeds might have resulted from the inhibition of leaf formation and expansion as well as early leaf abscission or due to ion toxicity, membrane disruption and complete stomatal closure (27,28). The decrease in CA and NR activities and P_N was maximum in 10 mM of NaCl. However, GA_3 alone or in combination with NaCl levels significantly stimulated NR, CA and P_N. The increase in CA might be due to its *de novo* synthesis by involving transcription and/or translation (29). CA is found in abundance in the photosynthesizing tissue of both C_3 and C_4 plants and regulates the availability of CO_2 to RuBPCO by catalyzing the reversible hydration of CO_2 (30). Khan et al. (31) and Hayat et al. (16) also demonstrated that GA_3 enhanced the P_N and CA activity in mustard leaves.

The elevated leaf NR activity by GA_3 alone or in combination with NaCl might additionally support its photosynthetic efficiency. NR is responsible for the initiation of the nitrate metabolism and consequently for the protein synthesis at various levels of the plant body. The activity of NR is very much unsteady and depends on the presence or absence of irradiation (32), and the presence of hormones (16,33). In the present investigation also NR is increased by the application of GA_3 (Table).

The total chlorophyll content of NaCl-treated plants was significantly decreased below that of the control plants (Table). Similarly, Dela-Rosa and Maiti (34) and Khodary (10) also reported inhibition in chlorophyll biosynthesis and reduced chlorophyll content in salt-stressed sorghum and maize plants, respectively. GA_3-treated plants exhibited higher values of chlorophyll than did the control or salinity-treated samples (Table). The enhancing effect of GA_3 on P_N could be attributed to its stimulatory effect on CA activity and chlorophyll content

Table. Effect of foliar spray of GA_3 on various parameters of mustard grown from NaCl treated seeds (f.m. = fresh mass).

Gibberellic Acid (M)	NaCl (mM)	DM plant^{-1} [g]	Chl (a+b) [g kg^{-1} f.m.]	NR [n mol NO_2 h^{-1}g^{-1}f.m.]	CA [mol (CO_2) kg^{-1} f.m. s^{-1}]	P_N [μ mol (CO_2)m^{-2}s^{-1}]	Pods plant^{-1}	Seed yield plant^{-1}[g]
0	0	2.35	1.316	292.54	1.68	14.92	205.10	7.25
	1	2.04	1.113	268.50	1.32	13.10	196.80	6.73
	10	1.83	0.965	249.68	1.15	11.86	189.41	6.56
10^{-6}	0	2.65	1.429	329.30	1.95	18.31	219.17	7.70
	1	2.41	1.330	297.31	1.71	15.73	207.05	7.29
	10	2.13	1.169	283.45	1.43	14.65	201.35	7.10
L.S.D. at P = 0.05		0.22	0.13	16.20	0.12	1.14	8.71	0.31

in this study. An increase in chlorophyll content with GA_3 was also reported earlier (16,31). The decrease in photosynthesis in the NaCl-stressed plants of mustard was further reflected in reduced vegetative growth (data not included) and dry matter production (Table). Therefore, the availability of photosynthates decreased during the reproductive phase, which finally resulted in decreased pod number and seed yield at harvest. However, this decrease was partially overcome by the application of GA_3 in stressed plants.

In summary, it might be concluded that GA_3 treatment of salt-stressed mustard plants could stimulate their salt tolerance by accelerating their photosynthetic performance and nitrogen metabolism.

Acknowledgment

The authors thank the Chairman of the Department of Botany, Aligarh Muslim University, Aligarh, India, for providing the necessary facilities in the department.

Corresponding author:
Firoz MOHAMMAD
Plant Physiology Section,
Department of Botany,
Aligarh Muslim University,
Aligarh, 202002, India

References

1. Anand IJ. Plant breeding challenges and constraints. Suggested areas of tissue culture relevance in oleifferous. In: Scope and potential of tissue culture for crop improvement. IARI, New Delhi, 1989.
2. Rhoades JD, Loveday J. Salinity in irrigated agriculture. In: Steward BA, Nielsen, DR (ed.), American Society of Civil Engineeres, Irrigation of Agricultural Crops American Society of Agronomists, USA. 30: 1089-1142, 1990.
3. Lumis GP, Hofstra G, Hall R. Sensitivity of roadside trees and shrubs to aerial drift of deicing salt. Hort Sci 8: 475-477, 1973.
4. Waisel Y. Adaptation to salinity. In: Raghavendra AS (ed.), Physiology of Trees, Wiley, New York, 1991, pp. 359-383.
5. Shannon MC, Grieve CM, Francois LE. Whole plant response to salinity. In: Wilkinson RE (ed.), Plant Environment Interactions, Marcel Dekker, New York, 1994. pp. 199-244.
6. Kozlowski TT. Response of woody plants to flooding and salinity. Tree Physiology Monograph No.1, Heron publishing, Victoria, Canada, 1997.
7. Greenway H, Munns R. Mechanism of salt tolerance in non halophytes. Ann Rev Plant Physiol 31: 149-194, 1980.
8. Blum A. Crop responses of drought and the interpretation of adaptation. Plant Growth Regul 20: 135-148, 1996.
9. El-Shihaby OA, Alla MMN, Younis ME et al. Effect of kinetin on photosynthetic activity and carbohydrate content in waterlogged or sea water treated *Vigna sinensis* and *Zea mays* plants. Plant Biosystem 136: 277-290, 2002.
10. Khodary SEA. Effect of salicylic acid on the growth, photosynthesis and carbohydrate metabolism in salt stressed maize plants. Int J Agri Biol 6: 5-8, 2004.
11. Ziska LH, Seeman LH, DeJong TM. Salinity induced limitations on photosynthesis in *Prunus salicina*, a decidous tree species. Plant Physiol 93: 864-870, 1990.
12. Dutt SK, Ball AR, Bandyopadhay AK. Salinity induced chemical changes in Casuarina equisetifolia Forst. Egyp J Soil Sci 31: 57-63, 1991.
13. Kalir A, Poljakoff-Mayber A. Effect of salinity on respiratory pathways in root tips of *Tamarix tetragyna*. Plant Physiol 57: 167-170, 1976.
14. Crozier A, Turnball C. Gibberellins: Biochemistry and action in extension growth, What's New. Plant Physiol 15: 9-12, 1984.
15. Arteca RN. Plant Growth Substances, CBS Publishers, New Delhi. 1997. pp 112-120.
16. Hayat S, Ahmad A, Mobin M et al. Carbonic anhydrase, photosynthesis and seed yield in mustard plants treated with phytohormones. Photosynthetica 39: 111-114, 2001.
17. Saxena OP, Pandey AV. Hormonal modulation for crop productivity of low quality seeds. In: Khan NA, Samiullah (ed.), Phytohormones in Crop Productivity under Different Environments, Scientific Publishers, Jodhpur, India, 2001, pp. 135-166.
18. Khan NA, Samiullah. Comparative effect of modes of gibberellic acid application on photosynthetic biomass distribution and productivity of rapeseed-mustard. Physiology & Molecular Biology of Plants 9: 141-145, 2003.
19. Angrish A, Kumar B, Datta KS. Effect of gibberellic acid and kinetin on nitrogen content and nitrate reductase activity in wheat under saline conditions. Ind J Plant Physiol 6: 172-177, 2001.
20. Aldesuquy HS, Ibrahim AH. Interactive effects of seawater and growth bioregulators on water relations abscisic acid concentration and yield of wheat plants. J Agro Crop Sci 187: 185-189, 2001
21. Chakrabarti N, Mukherji S. Effect of phytohormones pretreatment on metabolic changes in *Vigna radiata* under salt stress. J Env Biol 23: 295-300, 2002.

22. Mackinney G. Absorption of light by chlorophyll solutions. J Boil Chem 140: 315-322, 1941.

23. Dwivedi RS, Randhawa NS. Evaluation of a rapid test for the hidden hunger of Zinc in plants. Plant Soil 40: 445-451, 1974.

24. Jaworski EG. Nitrate reductase assay in intact plant tissue. Biochem Biophy Res Comm 43: 1274-1279, 1971.

25. Gomez KA, Gomez AA. Statistical procedure for agricultural research. Wiley International Publishers, New York, 1984. pp 156-174.

26. Wattana P, Monica AM. Water deficit on raffinose family oligosaccharides metabolism in coleus. Plant Physiol 121: 987-993, 1999.

27. Kozlowski TT, Pallardy SG. Physiology of woody plants. Academic Press, San Diego, 1997.

28. Pezeshki SR, Chambers JL. Effect of soil salinity on stomatal conductance and photosynthesis of green ash (*Fraxinus pennsylvanica*). Can J Forest Res 16: 569-573, 1986.

29. Okabe K, Lindlar A, Tsuzuki M et al. Effects of carbinic anhydrase on ribulose 1, 5-bisphosphate carboxylase and oxygenase. FEBS Lett 114: 142-144, 1980.

30. Badger MR, Price GD. The role of carbonic anhydrase in photosynthesis. Ann Rev Plant Physiol Plant Mol Biol 45: 369-392, 1994.

31. Khan NA, Ansari HR, Mobin M. Effect of gibberellic acid on carbonic anhydrase, photosynthesis, growth and yield of mustard. Biol Plant 38: 145-147, 1996.

32. Knypl JS, Krystyna M. Light and molybdenum requirements for substrate induction of nitrate reductase in cotyledons of *Lactuca sativa*. Biol Plant 21: 214-219, 1979.

33. Hayat S, Ahmad A. Soaking seeds of *Lens culinaris* with 28-homobrassinolide increased nitrate reductase activity and grain yield in the field in India. Ann Appl Biol 143: 121-124, 2003.

34. Dela-Rosa IM, Maiti RK. Biochemical mechanism in glossy sorghum lines for resistance to salinity stress. J Plant Physiol 146: 515-519, 1995.