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BRIEF COMMUNICATION

Protection of photosynthesis and antioxidative system by 24-epibrassinolide in *Solanum melongena* under cold stress

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Abstract

This study was carried out to understand the mechanism of protection of plants under cold stress by exogenous 24-epibrassinolide (EBR). The eggplant (*Solanum melongena* L.) seedlings were pretreated with five concentrations of EBR (0, 0.05, 0.1, 0.2 and 0.4 μM) and then exposed to day/night temperatures of 10/5 °C for 8 d. The results show that EBR, especially 0.1 μM EBR, dramatically alleviated growth suppression and a decrease in chlorophyll content and photosynthetic rate caused by the cold stress. In addition, EBR also decreased malondialdehyde content and $O_2^{\cdot-}$ production rate induced by the cold stress, and increased the activities of superoxide dismutase, guaiacol peroxidase, catalase, and ascorbate peroxidase, and proline content. The results of the present study suggest that exogenous EBR could improve cold tolerance of eggplant by regulating photosynthesis and antioxidative systems.

Additional key words: ascorbate peroxidase, catalase, brassinosteroids, low temperature, malondialdehyde, oxidative stress, peroxidase, *Solanum melongena*, superoxide dismutase.

Low temperature is one of the most important environmental factors affecting plant growth and productivity (Thakur *et al*. 2010, Zhang *et al*. 2013). Among the primary processes affected by cold stress, photosynthesis belongs to the first ones (Allen and Ort 2001, Goh *et al*. 2012). Cold stress decreases capacity and efficiency of photosynthesis through changes in gas exchange, pigment content and composition, and chloroplast development, and also through a decline in chlorophyll fluorescence (Farooq *et al*. 2009). Cold stress also causes oxidative stress due to the overproduction of reactive oxygen species (ROS) which can cause oxidative damage to lipids, proteins, and nucleic acids (Mittler 2002). However, plants have evolved an efficient antioxidant system which includes enzymes, such as superoxide dismutase (SOD), peroxidase (POD), catalase (CAT), and ascorbate peroxidase (APX), and non-enzymatic antioxidants, such as ascorbic acid (AsA) and glutathione (GSH) (Mittler 2002). In a number of studies, cold-tolerant plants have been correlated with a higher photosynthetic efficiency and more effective antioxidant systems (Bajguz and Hayat 2009, Fariduddin *et al*. 2011, Mutlu *et al*. 2013). Thus, the development of methods to maintain high photosynthetic rate and enhanced anti-oxidant systems in cold-stressed plants is of vital importance.

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Abbrevations: AsA - ascorbic acid; BRs - brassinosteroids; CAT - catalase; Chl - chlorophyll; c_i - intercellular CO_2 concentration; E - transpiration rate; EBR - 24-epibrassinolide; F_v/F_m - variable to maximum chlorophyll fluorescence ratio [maximum quantum yield of photosystem (PS) II photochemistry]; F_v'/F_m' - efficiency of excitation energy capture by open PS II center; F_v/F_o - potential photochemical efficiency of PS II; g_s - stomatal conductance; GSH - reduced glutathione; MDA - malondialdehyde; NBT - nitroblue tetrazolium; NPQ - non-photochemical quenching; PS II - photosystem II; Φ_{PSII} - effective quantum yield of PS II photochemistry; POD - peroxidase; qP - photochemical quenching ; ROS - reactive oxygen species; SOD - superoxide dismutase.
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Brassinosteroids (BRs) play an essential role in plant growth and development (Kartal *et al*. 2009). Moreover, BRs are recognized to confer the resistance of plants against various abiotic and biotic stresses (Nakashita *et al*. 2003, Fariduddin *et al*. 2009, Hayat *et al*. 2010, Ding *et al*. 2012, Li *et al*. 2012, Ahammed *et al*. 2013). The involvement of BRs in cold tolerance has also attracted much attention in the last decade (Bajguz and Hayat 2009, Liu *et al*. 2009, Fariduddin *et al*. 2011). However, our knowledge of the mechanisms of changes in photosynthesis and antioxidant systems induced by BRs under cold stress are still far from complete. Eggplant (*Solanum melongena* L.), an important horticulture crop worldwide, is very sensitive to low temperature. Nonetheless, scarcely any studies have been conducted on the influence of BRs on eggplants under cold stress. Therefore, the purpose of this study was to clarify, whether exogenous 24-epibrassinolide (EBR) could alleviate the cold-induced growth suppression by regulating photosynthesis and antioxidant capacity.

Seeds of eggplant, *Solanum melongena* L. cv. Huqie 9 from the Shanghai Academy of Agricultural Sciences, were rinsed thoroughly with distilled water and germinated on moist filter paper in an incubator at 28 °C. Uniformly germinated seeds were selected, sown in plastic pots filled with a 1:1 mixture of peat and *Vermiculite*, one plant per pot, and grown in a greenhouse under a 12-h photoperiod, irradiance of 600 μmol m^{-2} s^{-1}, temperatures of 25/20 °C (day/night) and relative humidity of 65 - 70 %. All plants were irrigated with a half strength Hoagland nutrient solution every 2 d. When the eggplant seedlings were at the fourth or fifth true leaf stage, EBR solutions at five concentrations (0, 0.05, 0.1, 0.2, and 0.4 μM) were sprayed onto the leaves. One day after spraying, the plants were transferred to a cold chamber (10/5 °C) and the cold stress was performed. Controls were grown at 25/20 °C. After the 8-d treatment, 24 plants were harvested, and shoots were rinsed three times in distilled water after their disinfecting with a non-ionic detergent, then blotted to dryness on filter paper and weighed.

Chl *a* and *b* were extracted by 95 % (v/v) ethanol on ice and determined according to Knudson *et al*. (1977) using a spectrophotometer *DU730* (*Beckman*, Coulter, USA). Gas exchange parameters, such as net photosynthetic rate (P_N), stomatal conductance (g_s), transpiration rate (E), and intercellular CO_2 concentration (c_i) were measured in five intact plants per cultivar per treatment by a photosynthesis system (*LI-6400*, *LI-COR*, Lincoln, NE, USA). During the measurements, irradiance was 600 μmol m^{-2} s^{-1}, relative humidity about 80 %, leaf temperature 25 °C, and the ambient CO_2 concentration about 400 μmol mol^{-1}. Chlorophyll fluorescence was measured with a *PAM-2100* pulse modulated fluorometer (*Walz*, Effeltrich, Germany). The minimal fluorescence (F_0) was determined under weak modulated irradiance on dark-adapted leaves. Then, a 0.8 s saturating pulse of 8 000 μmol m^{-2} s^{-1} was used to determine the maximum fluorescence (F_m). Quantum efficiency of PS II (Φ_{PSII}), the excitation energy capture efficiency of open centers (F_v'/F_m'), and non-photochemical quenching (NPQ) were calculated as $(F_m' - F_s)/F_m'$, F_v'/F_m', and $F_m/F_m' - 1$, respectively, according to Yu *et al*. (2004), where F_m' is maximum fluorescence of irradiance-adapted leaves, and F_s is steady-state fluorescence.

The malondialdehyde (MDA) content and superoxide production were measured as described by Jiang and Zhang (2001). The total superoxide dismutase (SOD) activity was assayed by the NBT method of Giannopolitis and Ries (1977). One unit of the SOD activity was defined as the amount of the enzyme required to cause a 50 % inhibition of the reduction of nitroblue tetrazolium (NBT) monitored at 560 nm. The peroxidase (POD) activity was measured according to Hammerschmidt *et al*. (1982) by monitoring the rate of guaiacol oxidation at 470 nm. The catalase (CAT) activity was assayed as described by Durner and Klessing (1996) and the activity was determined as a decrease in the absorbance at 240 nm following the decomposition of H_2O_2. The ascorbate peroxidase activity (APX) was measured according to Nakano and Asada (1981) by monitoring the rate of ascorbate oxidation at 290 nm. One unit (U) of POD, CAT, and APX activity was defined as the amount of enzyme that caused an increase or decrease in absorbance of 0.01 per min. All the measurements were done using the above mentioned spectrophotometer. Proline was measured according to the method of Bates *et al*. (1973) after its extraction with a 3 % (m/v) 5-sulfosalicylic acid solution at room temperature. All data presented are the means ± SD of at least three independent experiments. Differences between treatments were evaluated by the least significant difference (LSD) test and considered statistically significant when $P \leq 0.05$.

Previous studies have shown that the effects of BRs on different cells are either protective or toxic, depending on their concentrations (Kartal *et al*. 2009). In this study, the cold stress significantly decreased the shoot fresh mass. However, the inhibitory effects were significantly alleviated by the 0.05 - 0.2 μM EBR applications. Furthermore, 0.1 μM EBR was found to be the best in enhancing eggplant growth under the cold stress (Table 1). The growth promoting effects of BRs on plants under stress conditions may be through cell elongation by promoting the transverse orientation of microtubules (Sasse 2003).

The present study also shows that P_N, g_s, and E less decreased by the cold stress after the application of 0.05 - 0.2 μM EBR, and 0.4 μM EBR ameliorated only a stress induced decrease in E (Table 1). The increase in c_i due to the cold stress and due to the EBR application suggests that g_s was not the sole factor limiting photosynthesis. Non-stomatal limitations to photosynthetic rate may include changes in photosynthetic pigments,

Table 1. The shoot fresh mass (FM) [g], content of Chl *a*, Chl *b*, and Chl (*a*+*b*) [mg g^{-1}(f.m.)], P_N [μmol(CO_2) m^{-2} s^{-1}], g_s [mol m^{-2} s^{-1}], c_i [μmol mol^{-1}], E [mmol m^{-2} s^{-1}], parameters of Chl fluorescence F_v/F_m, F_v/F_o, F_v'/F_m', qP, Φ_{PSII}, NPQ, content of MDA [μmol g^{-1} (f.m.)], $O_2^{\cdot-}$ production rate [nmol g^{-1}(f.m.) min^{-1}], activities of SOD [U g^{-1} (f.m.)], POD [U g^{-1}(f.m.)], CAT [U g^{-1} (f.m.)], APX [U g^{-1}(f.m.)], and proline content [μg g^{-1}(f.m.)] in control eggplant seedlings and seedlings subjected to cold stress (10/5 °C for 8 d) and pre-treated with different concentrations of EBR (0 - 0.4 μM). Means ± SD of at least three independent experiments. Different letters within each row indicate significant differences ($P \leq 0.05$) according to the LSD test.

Parameters	Control	Cold stress	0.05 μM EBR	0.1 μM EBR	0.2 μM EBR	0.4 μM EBR
FM	2.81 ± 0.17a	1.46 ± 0.01d	1.71 ± 0.04c	2.04 ± 0.01b	1.64 ± 0.01c	1.51 ± 0.01d
Chl *a*	1.22 ± 0.00a	0.83 ± 0.00e	0.95 ± 0.03bc	0.98 ± 0.00b	0.93 ± 0.01cd	0.93 ± 0.01d
Chl *b*	0.54 ± 0.03a	0.23 ± 0.00e	0.38 ± 0.01c	0.42 ± 0.01b	0.36 ± 0.03cd	0.33 ± 0.04d
Chl (*a*+*b*)	1.76 ± 0.03a	1.06 ± 0.00e	1.33 ± 0.03c	1.40 ± 0.00b	1.29 ± 0.03cd	1.25 ± 0.04d
P_N	14.49 ± 0.79a	6.51 ± 0.13d	8.08 ± 0.26c	9.22 ± 0.34b	7.72 ± 0.36c	6.98 ± 0.17d
g_s	0.69 ± 0.02a	0.21 ± 0.01e	0.41 ± 0.01c	0.45 ± 0.01b	0.33 ± 0.02d	0.25 ± 0.01e
c_i	281.40 ± 17.7e	320.80 ± 21.8d	386.40 ± 9.29b	420.40 ± 10.6a	352.20 ± 15.1c	336.60 ± 7.57cd
E	5.92 ± 0.28a	2.20 ± 0.16e	3.32 ± 0.12c	3.97 ± 0.17b	3.10 ± 0.06c	2.66 ± 0.18d
F_v/F_m	0.82 ± 0.01a	0.75 ± 0.02e	0.79 ± 0.00cd	0.81 ± 0.07b	0.80 ± 0.01c	0.78 ± 0.00d
F_v/F_o	4.46 ± 0.28a	3.02 ± 0.25e	3.81 ± 0.20cd	4.16 ± 0.15b	3.88 ± 0.12c	3.61 ± 0.08d
qP	0.79 ± 0.02a	0.68 ± 0.02d	0.74 ± 0.02c	0.76 ± 0.02b	0.74 ± 0.01c	0.72 ± 0.02c
Φ_{PSII}	0.59 ± 0.01a	0.49 ± 0.01e	0.53 ± 0.01c	0.55 ± 0.02b	0.52 ± 0.01cd	0.51 ± 0.01d
F_v'/F_m'	0.76 ± 0.01a	0.69 ± 0.01d	0.71 ± 0.02c	0.74 ± 0.01b	0.71 ± 0.01c	0.70 ± 0.02cd
NPQ	0.30 ± 0.01c	0.58 ± 0.4a	0.53 ± 0.03b	0.50 ± 0.03b	0.58 ± 0.03a	0.57 ± 0.04a
MDA	0.31 ± 0.03f	0.83 ± 0.04a	0.67 ± 0.02d	0.62 ± 0.00e	0.71 ± 0.03c	0.78 ± 0.02b
$O_2^{\cdot-}$	0.90 ± 0.03d	1.54 ± 0.02a	1.44 ± 0.02b	1.38 ± 0.05c	1.47 ± 0.02b	1.47 ± 0.03b
SOD	23.41 ± 1.88e	33.12 ± 0.64c	41.60 ± 0.32a	43.47 ± 0.09a	37.55 ± 0.934b	29.55 ± 2.00d
POD	15.10 ± 0.92f	18.10 ± 0.35e	28.50 ± 0.60c	39.90 ± 0.30a	32.10 ± 0.52b	26.60 ± 1.05d
CAT	21.84 ± 0.73d	24.36 ± 0.69c	33.08 ± 2.64b	39.96 ± 1.68a	35.00 ± 0.84b	25.16 ± 0.37c
APX	266.16 ± 9.56d	351.00 ± 5.30c	442.32 ± 1.85a	466.36 ± 1.63a	417.84 ± 31.1b	358.08 ± 2.65c
Proline	3.93 ± 0.41d	35.10±1.07zc	48.11 ± 2.29b	65.46 ± 4.59a	47.06 ± 1.59b	45.77 ± 0.89b

Rubisco content and activity, and use of assimilates. The application of EBR mitigated a cold-induced decline in the Chl *a*, Chl *b*, and Chl *a*+*b* content (Table 1). The most likely reason for supporting the increase in Chl content is that BRs might directly or indirectly stimulate Chl biosynthesis or inhibit chlorophyllase activity (Hayat *et al*. 2011). In this work, the application of EBR significantly alleviated a cold stress-induced decrease in F_v/F_m and F_v/F_o (Table 1) indicating that EBR could alleviate the damage of PS II reaction centers (Yuan *et al*. 2012). Correspondingly, 0.05 - 0.2 μM EBR showed similar effects on Φ_{PSII}, qP, and F_v'/F_m', suggesting that application of EBR increased the photochemical quenching and the efficiency of energy capture by open PS II reaction centres in cold-stressed seedlings. A significant decrease in NPQ in 0.05 and 0.1 μM EBR-treated seedlingss implies that EBR resulted in less thermal dissipation of excitation energy in PS II antennae.

Stresses increase the production of ROS, and the accumulation of MDA is often used as an indicator of lipid peroxidation (Liu *et al*. 2013). The present study shows that the cold stress significantly elevated the MDA content and $O_2^{\cdot-}$ production rate relative to the control (Table 1). However, the EBR application resulted in a significant decline in the MDA content and $O_2^{\cdot-}$ production rate compared to the cold-stressed plants which is consistent with the observations of Liu *et al*. (2009) and Fariduddin *et al*. (2011). One of the mechanisms that may explain EBR protective action against oxidative damage lies in the enhancement of the activity of the antioxidant system. Our study found that the cold stress significantly increased the activities of SOD, POD, and APX but not of CAT. The application of 0.05 - 0.2 μM EBR induced a further increase of the activities of SOD, POD, and APX. The enhanced activities of these enzymes suggest a possible role of EBR in amelioration of oxidative stress generated by cold stress. However, it is still unclear whether BRs directly or indirectly modulate the responses of plants to oxidative stress. Proline is one of the important organic osmolytes in plant cells and its rapid accumulation has been shown to occur in plants as a consequence of cold stress (Liu *et al*. 2009, 2013). The present study shows that the content of proline in the plants exposed to the cold stress increased, and EBR further increased its content (Table 1). It might be due to the fact that BRs activated the enzymes of the proline biosynthesis (Fariduddin *et al*. 2009).

In summary, the dose-dependent effects of EBR application (the optimum concentration was 0.1 μM) could alleviate the cold stress-induced growth inhibition,

decrease in photosynthetic parameters, and increase in ROS production by elevating the activities of anti-oxidative enzymes. However, the detailed studies at the molecular level would be necessary to elucidate the mechanism by which endogenous and exogenous BRs regulate the stress response.

References

Ahammed, G.J., Zhou, Y.H., Xia, X.J., Mao, W.H., Shi, K., Yu, J.Q.: Brassinosteroid regulates secondary metabolism in tomato towards enhanced tolerance to phenanthrene. - Biol. Plant. **57**: 154-158, 2013.

Allen, D.J., Ort, D.R.: Impact of chilling temperatures on photosynthesis in warm-climate plants. - Trends Plant Sci. **6**: 36-42, 2001.

Bajguz, A., Hayat, S.: Effects of brassinosteroids on the plant responses to environmental stresses. - Plant Physiol. Biochem. **47**: 1-8, 2009.

Bates, L.S., Waldren, K.P., Teare, I.D.: Rapid determination of free proline for water stress studies. - Plant Soil **39**: 205, 1973.

Ding H.D., Zhu X.H., Zhu Z.W., Yang S.J., Zha D.S., Wu X.X.: Amelioration of salt-induced oxidative stress in *Solanum melongena* L. by application of 24-epibrassinolide. - Biol. Plant. **56**: 767-770, 2012.

Durner, J., Klessing, D.F.: Salicylic acid is a modulator of tobacco and mammalian catalases. - J. biol. Chem. **271**: 28492-28502, 1996.

Fariduddin, Q., Yusuf, M., Chalkoo, S., Hayat, S., Ahmad, A.: 28-homobrassinolide improves growth and photosynthesis in *Cucumis sativus* L. through an enhanced antioxidant system in the presence of chilling stress. - Photosynthetica **49**: 55-64, 2011.

Fariduddin, Q., Yusuf, M., Hayat, S., Ahmad, A.: Effect of 28-homobrassinolide on antioxidant capacity and photosynthesis in *Brassica juncea* plants exposed to different levels of copper. - Environ. exp. Bot. **66**: 418-424, 2009.

Farooq, M., Aziz, T., Wahid, A., Lee, D.J., Siddique, K.H.M.: Chilling tolerance in maize: agronomic and physiological approaches. - Crop Pasture Sci. **60**: 501-516, 2009.

Giannopolitis, C.N., Ries, S.K.: Superoxide dismutases. I. Occurrence in higher plants. - Plant Physiol. **59**: 309-314, 1977.

Goh, C.H., Ko, S.M., Koh, S., Kim, Y.J., Bae, H.J.: Photosynthesis and environments: photoinhibition and repair mechanisms in plants. - J. Plant Biol. **55**: 93-101, 2012.

Hammerschmidt, R., Nuckles, E.M., Kuc, J.: Association of enhanced peroxidase activity with induced systemic resistance of cucumber to *Colletotrichum lagenarium*. - Physiol. Plant Pathol. **20**: 73-82, 1982.

Hayat, S., Hasan, S.A., Yusuf, M., Hayat, Q., Ahmad, A.: Effect of 28-homobrassinolide on photosynthesis, fluorescence and antioxidant system in the presence or absence of salinity and temperature in *Vigna radiata*. - Environ. exp. Bot. **69**: 105-112, 2010.

Hayat, S., Yadav, S., Wani, A.S., Irfan, M., Ahmad, A.: Comparative effect of 28-homobrassinolide and 24-epi-brassinolide on the growth, carbonic anhydrase activity and photosynthetic efficiency of *Lycopersicon esculentum*. - Photosynthetica **49**: 397-404, 2011.

Jiang M.Y., Zhang J.H.: Effect of abscisic acid on active oxygen species, antioxidative defence system and oxidative damage in leaves of maize seedlings. - Plant Cell Physiol. **42**: 1265-1273, 2001.

Kartal, G., Temel, A., Arican, E., Gozukirmizi, N.: Effects of brassinosteroids on barley root growth, antioxidant system and cell division. - Plant Growth Regul. 5**8**: 261-267, 2009.

Knudson, L.L., Tibbits, T.W., Edwards, G.E.: Measurements of ozone injury by determination of leaf chlorophyll concentration. - Plant Physiol. **60**: 606-608, 1977.

Li, Y.H., Liu, Y.J., Xu, X.L., Jin, M., An, L.Z., Zhang, H.: Effect of 24-epibrassinolide on drought stress-induced changes in *Chorispora bungeana*. - Biol. Plant. **56**: 192-196, 2012.

Liu, T., Li, Y., Ren J., Zhang C., Kong M., Song X., Zhou J., Hou X.: Over-expression of *BcFLC1* from non-heading Chinese cabbage enhances cold tolerance in *Arabidopsis*. - Biol. Plant. **57**: 262-266, 2013.

Liu, Y., Zhao, Z., Si, J., Di, C., Han, J., An, L.: Brassinosteriods alleviate chilling induced oxidative damage by enhancing antioxidant defense system in suspension cultured cells of *Chorispora bungeana*. - Plant Growth Regul. **59**: 207-214, 2009.

Mittler, R.: Oxidative stress antioxideants and stress tolerance. - Trends Plant Sci. **7**: 405-410, 2002.

Mutlu, S., Karadağoğlu, Ö., Atici, Ö., Nalbantoğlu, B.: Protective role of salicylic acid applied before cold stress on antioxidative system and protein patterns in barley apoplast. - Biol. Plant. **57**: 507-513, 2013.

Nakano, Y., Asada, K.: Hydrogen peroxide is scavenged by ascorbate-specific peroxidase in spinach chloroplasts. - Plant Cell Physiol. **22**: 867-880, 1981.

Nakashita, H., Yasuda, M., Nitta, T., Asami, T., Fujioka, S., Arai, Y., Sekimata, K., Takatsuto, S., Yamaguchi, I., Yoshida, S.: Brassinosteroid functions in a broad range of disease resistance in tobacco and rice. - Plant J. **33**: 887-898, 2003.

Sasse, J.M.: Physiological actions of brassinosteroids: an update. - J. Plant Growth Regul. **22**: 276-288, 2003.

Thakur, P., Kumar, S., Malik, J.A., Berger, J.D., Nayyar, H.: Cold stress effects on reproductive development in grain crops: an overview. - Environ. exp. Bot. **67**: 429-443, 2010.

Yu, J.Q., Huang, L.F., Hu, W.H., Zhou, Y.H., Mao, W.H., Ye, S.F., Nogues, S.: A role of brassinosteroids in the regulation of photosynthesis in *Cucumis sativus*. - J. exp. Bot. **55**: 1135-1143, 2004.

Yuan, L.Y., Shu, S., Sun, J., Guo, S.R., Tezuka, T.: Effects of 24-epibrassinolide on the photosynthetic characteristics, antioxidant system, and chloroplast ultrastructure in *Cucumis sativus* L. under $Ca(NO_3)_2$ stress. - Photosynth. Res. **112**: 205-214, 2012.

Zhang, X.D., Wang, R.P., Zhang, F.J., Tao, F.Q., Li, W.Q.: Lipid profiling and tolerance to low-temperature stress in *Thellungiella salsuginea* in comparison with *Arabidopsis thaliana*. - Biol. Plant. **57**: 149-153, 2013.