PHOTOSYNTHETICA 36 (4): 597-599, 1999

BRIEF COMMUNICATION

Uniconazole (S-3307) induced protection of *Abelmoschus esculentus* L. against cadmium stress

S. PUROHIT and V.P. SINGH

Institute of Environment Management and Plant Sciences, Vikram University, Ujjain - 456 010, India

Abstract

In *Abelmoschus esculentus* L. uniconazole brought about a marked decrease in cadmium-induced loss of chlorophyll and Hill reaction activity, but it did not completely prevent cadmium toxicity.

Additional key words: biomass; chlorophyll; detoxification; growth; Hill reaction activity.

Cadmium severely inhibits several growth processes and chlorophyll (Chl) synthesis (Ernst *et al.* 1992, Singh 1993, Fodor *et al.* 1995, Thomas and Singh 1996, Vassilev *et al.* 1997, Ernst 1998). The triazoles belong to a class of compounds that have been developed for use as either fungicides or plant growth regulators. They also protect plants from various environmental stresses including chilling, drought, air pollutants (Fletcher and Hofstra 1988), and heavy metals (Singh 1993). Hence, the triazoles have been considered to be "multiplant protectant". The objective of this study was to assess the effect of uniconazole (S-3307), a triazole derivative, on *Abelmoschus esculentus* L. with particular emphasis on fresh and dry masses, Chl content, and Hill reaction activity.

In the present study, the response of *A. esculentus* to toxic levels of Cd was studied in plants growing in thermocol cups. Uniconazole was applied as a seed treatment for which the seeds were soaked in 5 g m^{-3} uniconazole solution for 5 h and subsequently dried. Both treated and non-treated seeds were raised in 300 cm^3 thermocol cups with perforated bottoms, each of which contained 200 g oven-dried soil. These cups were placed in second non-perforated cups which served as a reservoir for excess water. Prior to sowing, seeds were treated with different Cd concentrations (0, 50, 100, 200, and 400 mg kg^{-1}) supplied as $CdCl_2$. The plants were raised in a growth room with photon flux rate of 500 μmol m^{-2} s^{-1} and 16/8 light/dark cycles. Fresh and dry masses were determined 30 d after the date of sowing. The

Received 21 September 1998, *accepted* 13 May 1999.
Acknowledgements: Authors are thankful to ICAR, New Delhi for financial support.

Table 1. The effect of different concentrations of cadmium on growth [mg plant^{-1}], chlorophyll (a+b) [g(Chl) kg^{-1}(f.m.)], and Hill reaction activity [mol(DCPIP red.) s^{-1} kg^{-1}(Chl)] in non-treated and uniconazole-treated 30-d-old plants. Values in parentheses show percent reduction relative to 0 Cd or 0 Cd + uniconazole treatments. Means ± S.E. of 3 replicates from each treatment.

Cd concentration [mg(Cd) kg^{-1}]	Fresh mass	Dry mass	Chlorophyll (a+b)	Hill activity
0	792 ± 1 (0)	93.3 ± 15 (0)	297.9 ± 1.3 (0)	26.1 ± 0.2 (0)
50	774 ± 3 (2)	90.3 ± 5 (3)	245.0 ± 0.8 (17)	21.9 ± 0.1 (15)
100	705 ± 3 (10)	82.2 ± 3 (11)	219.5 ± 1.5 (26)	20.0 ± 0.2 (23)
200	632 ± 1 (20)	73.4 ± 21 (21)	195.1 ± 1.1 (34)	18.2 ± 0.1 (30)
400	515 ± 2 (34)	56.3 ± 4 (39)	148.6 ± 1.8 (50)	12.9 ± 0.1 (50)
0 + uniconazole	708 ± 1 (0)	83.3 ± 6 (0)	580.9 ± 1.1 (0)	56.0 ± 0.2 (0)
50 + uniconazole	690 ± 2 (2)	82.6 ± 11 (0)	551.0 ± 1.1 (5)	52.2 ± 0.3 (6)
100 + uniconazole	651 ± 8 (8)	77.3 ± 13 (7)	517.0 ± 0.3 (11)	49.1 ± 0.1 (12)
200 + uniconazole	618 ± 4 (12)	72.3 ± 7 (13)	470.8 ± 0.9 (18)	44.0 ± 0.1 (21)
400 + uniconazole	501 ± 2 (29)	57.3 ± 7 (31)	442.5 ± 0.6 (23)	38.6 ± 0.1 (31)

pigments were extracted in 80 % acetone from 100 mg leaf tissues and Chl content was determined as described by Strain and Svec (1966) by measuring the absorbances at 665 and 649 nm using a UV-Vis spectrophotometer. Hill reaction activity (HA) was determined following the DCPIP reduction method.

The plants grown from seeds treated with different concentrations of Cd^{2+} had reduced fresh and dry masses, Chl contents, and HA. The reduction increased with increasing concentration of Cd (Table 1). Plants grown from seeds treated with uniconazole also had lower fresh and dry masses than controls. The reduction of shoot height caused by uniconazole treatment was accompained by a reduction in plant mass. The percent protection gained by uniconazole treatment against Cd toxicity was between 5.00 and 5.75 % as determined by effects on Cd-induced fresh and dry masses, respectively. A shift in balance of gibberellin may account for this plant growth regulatory activity of uniconazole (Fletcher and Hofstra 1988).

Uniconazole treatment resulted in significantly higher Chl contents and HA in control plants and in all Cd-treatments (Table 1): even at high Cd-concentrations (400 mg kg^{-1}), Chl and HA were protected. The higher values of Chl and HA in the uniconazole-treated leaves, even in the presence of Cd, suggest that uniconazole prevents the symptoms of Cd phytotoxicity on these physiological parameters. According to Stobart *et al.* (1985), Cd inhibits biosynthesis of 5-aminolevulinic acid (ALA), a precursor of Chl. Cytokinins stimulate Chl biosynthesis by promoting ALA synthesis (Fletcher and McCullagh 1971). Uniconazole also increases cytokinin concentrations in rice (Izumi *et al.* 1988). Therefore, it is likely that the stimulation of Chl synthesis in uniconazole-treated plants is mediated through an effect on cytokinins.

Ernst (1998) reported that isolated chloroplasts could be affected by heavy metals, and metals such as Cd and Zn directly inhibited photosynthetic electron transport reactions, especially photosystem 2 (PS2), both at the oxidizing (donor) and reducing

(acceptor) sites. Another mechanism of metal injury is a direct effect on the Calvin cycle, which results in accumulation of ATP and NADPH and finally in loss of PS2 function (Ernst 1998). These effects would decrease HA in the presence of cadmium. Inhibition of HA by Cd is shown in the present study. Triazole-treated chloroplasts are larger than those in control (Deshpande *et al.* 1991). Leaves with more Chl and more chloroplasts may have a more effective free radical scavenging system. Damage to the chloroplast by Cd (Stoyanova and Tchakalova 1997) may be prevented or delayed by effective removal of free radicals. The triazoles are effective inhibitors of catalase and peroxidases as well as of other enzymes. This would lead to a decrease in free radical scavenging capacity, and thus it is difficult to explain our results in terms of increased antioxidant defences. The uniconazole-treatment must, however, change gene expression that enables the seedlings to tolerate high Cd concentrations.

References

Deshpande, S., Hofstra, G., Fletcher, R.A., Abrahamson, E.W.: Light scattering spectrophotometry from isolated control and triazole treated chloroplasts. - Photochem. Photobiol. **54**: 485-488, 1991.

Ernst, W.H.O.: Effects of heavy metals in plants at the cellular and organismic levels. - In: Schuurmann, G., Markert, B. (ed.): Ecotoxicology. Pp. 587-620. John Wiley and Sons, London, Spektrum Akademischer Verlag, Heidelberg - Berlin 1998.

Ernst, W.H.O., Verkleij, J.A.C., Schat, H.: Metal tolerance in plants. - Acta bot. neerland. **41**: 229-248, 1992.

Fodor, E., Szabo-Nagy, A., Erdei, L.: The effects of cadmium on the fluidity and H^+-ATPase activity of plasma membrane from sunflower and wheat roots. - J. Plant Physiol. **147**: 87-92, 1995.

Fletcher, R.A., Hofstra, G.: Triazoles as potential plant protectants. - In: Berg, D., Plempel, M. (ed.): Sterol Biosynthesis Inhibitors: Pharmaceutical and Agrochemical Aspects. Pp. 321-331. Ellis Horwood, London - Weinheim - Cambridge 1988.

Fletcher, R.A., McCullagh, D.: Benzyladenine as a regulator of chlorophyll synthesis in cucumber cotyledons. - Can. J. Bot. **49**: 2197-2201, 1971.

Izumi, K., Nakagawa, S., Kobayashi, M., Oshio, H., Sakurai, A., Takahashi, N.: Levels of IAA, cytokinins, ABA and ethylene in rice plants as affected by gibberellin biosynthesis inhibitor, uniconazole-P. - Plant Cell Physiol. **29**: 97-104, 1988.

Singh, V.P.: Uniconazole (S-3307) induced cadmium tolerance in wheat. - J. Plant Growth Regul. **12**: 1-3, 1993.

Stobart, A.K., Griffiths, W.T., Ameen-Bukhari, I., Sherwood, R.P.: The effect of Cd^{2+} on the biosynthesis of chlorophyll in leaves of barley. - Physiol. Plant. **63**: 293-298, 1985.

Stoyanova, D.P., Tchakalova, E.S.: Cadmium-induced ultrastructural changes in chloroplasts of the leaves and stems parenchyma in *Myriophyllum spicatum*. - Photosynthetica **34**: 241-248, 1997.

Strain, H.H., Svec, W.A.: Extraction, separation, estimation, and isolation of the chlorophylls. - In: Vernon, L.P., Seely, G.R. (ed.): The Chlorophylls. Pp. 21-26. Academic Press, New York - London 1966.

Thomas, R.M., Singh, V.P.: Reduction of cadmium-induced inhibition of chlorophyll and carotenoid accumulation in *Cucumis sativus* L. by uniconazole (S. 3307). - Photosynthetica **32**: 145-148, 1996.

Vassilev, A., Yordanov, I., Tsonev, T.: Effects of Cd^+ on the physiological state and photosynthetic activity of young barley plants. - Photosynthetica **34**: 293-302, 1997.