

学位論文要旨 Dissertation Abstract

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学位論文題目 : Physiology and Metabolism in Heavy Metal Toxicity and
Title of Dissertation Tolerance of *Brassica* species: Roles of Different Exogenous
Phytoprotectants
(ブラシカ種の重金属毒性と耐性における生理と代謝 : 種々の植物保護剤の役割)

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Heavy metal pollution in soil is a terrible environmental threat which represents the major limiting factors to agricultural productivity worldwide and becomes serious health hazard for human and animal. Among the heavy metal cadmium (Cd) and chromium (Cr) are gaining more attention due to their extensive exploitation in crop production. Cadmium and Cr toxicity in soil hamper the seed germination, growth and developments of plants. Thus, remediation of Cd and Cr from soil or enhancing plants' tolerance to metal toxicity is urgent. Different phytoprotectants like organic acid, amino acid, metal chelator etc. are very effective against metal toxicity. Therefore, several studies were undertaken to investigate the biochemical and physiological basis of *Brassica* species against Cd and Cr stress; functions of citric acid (CA) and ethylenediaminetetraacetic acid (EDTA) under Cd stress; functions of γ -aminobutyric acid (GABA) and maleic acid (MA) under Cr stress. First study was carried out to find out the metal accumulation and tolerance abilities of three *Brassica* species (*B. napus*, *B. campestris*, and *B. juncea*) seedlings exposed to two levels of Cd (0.25 and 0.5 mM CdCl₂ for three days) in a semi hydroponic media. Of the *Brassica* species studied, *B. juncea* accumulated the highest amount of Cd in a dose-dependent manner and in every case, the Cd content was higher in the roots than the shoots. Cadmium stress reduced seedlings biomass, leaf relative water content (RWC), and chlorophyll (chl) content, whereas proline (Pro), malondialdehyde (MDA), and hydrogen peroxide (H₂O₂), superoxide anion (O₂^{•-}), methylglyoxal (MG) content and lipoxygenase (LOX) activity increased in all species. But, the rate of oxidative damage was lower in *B. juncea* than the other two species as antioxidant defense and glyoxalase system performed better. So,

B. juncea was relatively tolerant species to Cd toxicity because of its lower damage even though higher accumulation of Cd. Considering this nature, second and third studies were undertaken to investigate the possible role of EDTA (0.5 mM) and CA (0.5 mM and 1.0 mM) in mitigating Cd toxicity (0.5 and 1.0 mM CdCl₂, 3 d) in *B. juncea* seedlings. Without EDTA and CA, the roots and shoots of *B. juncea* accumulated Cd in a dose-dependent manner, which hampered growth and physiology of plant by disrupting antioxidant defense and glyoxalase system. Moreover, Cd-stressed seedlings supplemented with EDTA and CA upregulated the components of non-enzymatic antioxidants (Ascorbate, AsA; glutathione, GSH), enzymatic antioxidants (ascorbate peroxidase, APX; monodehydroascorbate reductase, MDHAR; dehydroascorbate reductase, DHAR; glutathione reductase, GR; glutathione peroxidase, GPX; superoxide dismutase, SOD; catalase, CAT) and glyoxalase system enzymes (Glyoxalase I, Gly I, glyoxalase II, Gly II), significantly or to some extent. Furthermore, adding EDTA to the Cd-treated plants limiting Cd uptake but CA (1.0 mM) increased the root and shoot Cd content as well as Cd translocation from root to shoot in a concentration-dependent fashion. But, EDTA and both dose of CA counteracted Cd stress considerably where the performance of 0.5 mM CA was conspicuous. Fourth and fifth experiment was carried out to explore the effects of GABA and MA in alleviating Cr toxicity. We treated eight-d-old *B. juncea* seedlings with Cr (0.15 mM and 0.3 mM K₂CrO₄, 5 days) alone and in combination with GABA (125 μM) or MA (0.25 mM). The roots and shoots of the seedlings accumulated Cr in a dose-dependent manner, which led to an increase in oxidative stress by upsetting antioxidant defense and glyoxalase systems. Chromium stress also disrupted the growth, water status and photosynthetic performance. Moreover, supplementing the Cr-treated seedlings with GABA reduced Cr uptake but MA increased Cr uptake of *B. juncea*. However both of them upregulated the most of the antioxidant enzymes of AsA-GSH cycle (MDHAR, DHAR, GR); increased activities of SOD, CAT and GR; and improve the performance of glyoxalase system which finally led to reduce oxidative damage and increased the chlorophyll content, water status as well the growth and biomass of the plants. Therefore, our findings suggest that CA (under Cd stress) and MA (under Cr stress) played dual role in *B. juncea* seedlings by increasing phytoremediation and enhancing stress tolerance through upregulating antioxidant defense and glyoxalase systems but EDTA (under Cd stress) and GABA (under Cr stress) limited metal accumulation and conferred stress tolerance.