

## 学位論文要旨 Dissertation Abstract

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学位論文題目 :  
Title of Dissertation Physiological basis of phenolics-induced salt stress recovery in tomato: Relevance of antioxidant defense associated systems  
(トマトにおけるフェノール類により誘導される塩ストレスからの回復の生理学的基礎：抗酸化防御に係るシステムの関連性)

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Salinity is a severe threat to world crop production and food sustainability. Salt toxicity hampers the seed germination, growth, development and physiology of plants which cause the yield loss. In addition, these devastating effects make plants being prone to death before going reproduction as most of the crops plants are sensitive to higher salinity. Here, four experiments had been conducted to evaluate the self-recovery approaches of tomato along with using plant phenolics as phytoprotectant. The main focus was on incorporating the plant antioxidant defense, glyoxalase system and nutrient homeostasis to increase the salt tolerance in tomato.

In the first experiment, salinity toxicity and the post-stress restorative process were examined to identify the salt tolerance mechanism in tomato. Hydroponically grown 10 day-old tomato plants (*Solanum lycopersicum* L.) were treated with 150 and 250 mM NaCl for 4 days and subsequently grown in nutrient solution for a further 2 days to observe the post-stress responses. Salt stress decreased seedlings growth, biomass contents, leaf relative water content (RWC), chlorophyll (Chl) and carotenoids (Car) content whereas reactive oxygen species ( $H_2O_2$  and  $O_2^{\cdot-}$ ), methylglyoxal (MG), proline (Pro), malondialdehyde (MDA) content,  $Na^+ : K^+$  ratio and lipoxygenase (LOX) activity increased. But, after 2 days of recovery treatment (grown in standard nutrient solution), stressed seedlings showed improvements in antioxidant and glyoxalase enzymes activities, followed by improvements in seedlings growth, water balance, and Chl synthesis, lowered accumulation of ROS, MG, MDA and membrane damage (as electrolyte leakage; EL). Considering this rapid recovery of tomato, three further experiments were undertaken to investigate the protective role of quercetin (Qu, as flavonoid: 15 and 25  $\mu M$ ), vanillic acid (VA; as hydroxybenzoic acid: 40 and 50  $\mu M$ ) and coumarin (COU; as hydroxycinnamic acid: 20 and 30  $\mu M$ ) to mitigate salt toxicity by

enhancement of nutrient homeostasis, antioxidant defense and glyoxalase systems.

Both Qu and VA applied as co-treatment with salinity (150 mM NaCl). Both Qu and VA application under salt stress resulted in lower  $\text{Na}^+:\text{K}^+$ , higher leaf relative water content (LRWC) with elevated Pro, and reduction of  $\text{H}_2\text{O}_2$  and MDA content, and LOX activity, which indicated alleviation of ionic, osmotic, and oxidative stress respectively. In addition, salt-stressed seedlings supplemented with Qu and VA up-regulated the components of non-enzymatic antioxidants (ascorbate, AsA; glutathione, GSH) along with strengthening of enzymatic components. Exogenous Qu and VA caused the higher activities of glyoxalase enzymes in stressed seedlings which reversed the excess MG production. The VA and Qu mediated coordinated strengthening actions of antioxidant defense and glyoxalase system resulted better growth performance in stressed seedlings. Both Qu and VA also insisted on better plant growth and photosynthetic pigments synthesis in without saline media.

But, COU has been applied as pretreatment to tomato seedling. Hence, osmotic and ionic toxicity had been disclosed in salt stressed tomato seedlings which led to oxidative stress indicated by higher  $\text{H}_2\text{O}_2$ , MG, MDA, EL along with upsetting of antioxidant defense and glyoxalase systems. The salt-induced damage increased with the increasing of stress level. The COU pretreated seedlings showed the protective role of it in stress mitigation through improving osmotic, and ionic balance along with higher growth performance upon both salt stresses (100 and 160 mM). The detoxification of ROS observed in COU pretreated seedlings upon stresses by up regulating the redox balance of AsA and GSH with higher activities of their enzymatic antioxidants (ascorbate peroxidase, APX; monodehydroascorbate reductase, MDHAR; glutathione reductase, GR, and catalase, (CAT); glutathione *S*-transferase, (GST). In addition COU-mediated higher performance of glyoxalase systems resulted in MG detoxification in stressed seedlings. These stress alleviation performance of COU-pretreatment led to increased Chl, Car contents, growth and biomass accumulation in tomato under saline toxicity. So, it is suggested that COU could be an actor in conferring salt stress by the coordinated action of ion homeostasis, antioxidant defense and glyoxalase systems.

Therefore, the possible mechanisms of salt stress alleviation by Qu, VA and COU are closely associated with the synchronized performances of nutrient homeostasis, antioxidant defense and glyoxalase systems. In addition, all these three phenolics caused the increment in GST activity upon salt stress which plays significant roles in mitigating salt stress in tomato. It is suggested that plant phenolics enhance salt tolerance in tomato at seedlings stage by reinforcing the plant antioxidative responses.