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# Antioxidant enzyme activities are upregulated in response to cadmium in sensitive, but not in tolerant, rice (*Oryza sativa* L.) seedlings

Mei Chun KUO and Ching Huei KAO*

*Department of Agronomy, National Taiwan University, Taipei, Taiwan, Republic of China*

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**Abstract.** Changes in $H_2O_2$ and malondialdehyde (MDA) contents and antioxidant enzyme activities in Cd-treated rice (*Oryza sativa* L.) seedlings of two cultivars were investigated. On treatment with $CdCl_2$, increases in $H_2O_2$ and MDA contents and antioxidant enzyme activities [speroxide dismutase (SOD), ascorbate peroxidase, glutathione reductase, catalase, and peroxidase (POX)] were observed in the leaves of Cd-sensitive cultivar (cv. Taichung Native 1, TN1) but not in Cd-tolerant cultivar (cv. Tainung 67, TNG67). The increased content of MDA and activities of SOD and POX preceded the occurrence of toxicity in $CdCl_2$-treated TN1 leaves. Pretreatment with abscisic acid (ABA) enhanced Cd tolerance and reduced Cd-induced increase in the content of MDA and increase in the activities of SOD and POX in TN1 leaves. Exogenous application of ABA biosynthesis inhibitor, fluridone, decreased Cd tolerance, increased the content of MDA, and increased the activities of SOD and POX in Cd-treated TNG67 leaves. Furthermore, fluridone's effects on toxicity, the content of MDA, and the activities of SOD and POX in Cd-treated TNG67 leaves were reversed by the application of ABA. In conclusion, the oxidative stress is differently expressed in TN1 and TNG67 rice seedlings in response to $CdCl_2$. Results also suggest that $CdCl_2$ causes an oxidative stress and $CdCl_2$-induced toxicity is mediated through oxidative stress in TN1 leaves.

**Keywords:** Abscisic acid; Cadmium; *Oryza sativa* L.; Oxidative stress.

**Abbreviations: ABA,** abscisic acid; **APX,** ascorbate peroxidase; **CAT,** catalase; **Flu,** fluridone; **FW,** fresh weight; **GR,** glutathione reductase; **MDA,** malondialdehyde; **POX,** peroxidase; **SOD,** superoxide dismutase; **TN1,** Taichung Native 1; **TNG67,** Tainung 67.

## Introduction

Oxygen is essential for the existence of aerobic life, but toxic active oxygen species (AOS), which include the superoxide anion ($O_2^{\cdot-}$), hydroxyl radical ($OH^{\cdot}$) and hydrogen peroxide ($H_2O_2$), are generated in all aerobic cells during metabolic processes (Foyer et al., 1994; Asada, 1999). Injury caused by these AOS, known as oxidative stress, is one of the major damaging factors in plants exposed to environmental stress. Plants cope with oxidative stress by using antioxidant enzymes such as superoxide dismutase (SOD), ascorbate peroxidase (APX), glutathione reductase (GR), peroxidase (POX), catalase (CAT), and the low molecular weight antioxidants, ascorbic acid and glutathione (Noctor and Foyer, 1998; Asada, 1999).

Cadmium (Cd), a heavy metal toxic to humans, animals, and plants is a widespread pollutant with a long biological half-life (Wagner, 1993). Three lines of evidence indicate that one mechanism of Cd toxicity is related to oxidative stress in plant cells. First, Cd can promote the generation of AOS (Piqueras et al., 1999; Romero-Puertas et al., 1999; Chen et al., 2000; Shah et al., 2001; Sandalio et al., 2001; Schützendübel et al., 2001; Olmos et al., 2003). Second, Cd can inhibit or stimulate the activities of antioxidant enzymes (Shaw, 1995; Gallego et al., 1996; Chaoui et al., 1997; Dixit et al., 2001; Shah et al., 2001; Iannelli et al., 2002; León et al., 2002). Third, treatment with Cd results in cellular oxidative damage or lipid peroxidation (Shaw, 1995; Gallego et al., 1996; Chaoui et al., 1997; Lozano-Rodríguez et al., 1997; Dixit et al., 2001; Shah et al., 2001; Chien et al., 2002).

This study examines $H_2O_2$ content, lipid peroxidation, and antioxidant enzyme activities in two cultivars of rice (*Oryza sativa* L.) seedlings in response to Cd. One cultivar (cv. Taichung Native 1, TN1) is known to be sensitive to Cd, and the other (cv. Tainung 67, TNG67) is known to show significant tolerance to Cd (Hsu and Kao, 2003a). If a mechanism related to oxidative stress is involved in Cd toxicity, this mechanism should then be differently expressed in plants tolerant and sensitive to Cd. Therefore, the aim of this study was to investigate whether the oxidative stress mechanism is differently expressed in TN1 and TNG67 rice seedlings in response to Cd.

*Corresponding author. Tel: 886-2-23698159; Fax: 886-2-23620879; E-mail: kaoch@ntu.edu.tw

## Materials and Methods

### *Plant Cultivation and Treatment*

Two rice cultivars, an Indica type cultivar, TN1 and a Japonica cultivar, TNG67, obtained from Taiwan Agricultural Research Institute, Taichung, Taiwan, were used in this study. Seeds were sterilized with 2.5% (v/v) sodium hypochlorite for 15 min and washed extensively with distilled water. These seeds were then germinated in Petri dishes with wetted filter papers at 37°C in the dark. After 48 h incubation, uniformly germinated seeds were selected and cultivated in a 250 ml beaker containing half-strength Kimura B solution without aeration as described previously (Chu and Lee, 1989). The concentrations of N, P, K, S, Ca and Mg in half strength Kimura B solution were 11.5, 2.9, 7.2, 15.0, 7.4, and 8.7 p.p.m., respectively. The hydroponically cultivated seedlings were grown in a Phytotron (Agricultural Experimental Station, National Taiwan university, Taipei, Taiwan) with natural light at 30°C day (12 h)/25°C night (12 h) and 90% relative humidity. Twelve-day-old seedlings with three leaves were used in all experiments.

For Cd, abscisic acid (ABA), and fluridone (Flu) treatment, $CdCl_2$ (0.5 or 1.5 mM), ABA (5 μM), and Flu (0.2 mM) were added directly to the culture solution during experiment.

### *Cd Determination*

For determination of Cd, leaves or roots were dried at 65°C for 48 h. Dried material was ashed at 550°C for 20 h. The ash residue was incubated with 31% $HNO_3$ and 17.5 % (v/v) $H_2O_2$ at 72°C for 2 h, and dissolved in 0.1 N HCl. Cd was then quantified using an atomic absorption spectrophotometer (Model AA-6800; Shimadzu, Kyoto, Japan). The amount of Cd was expressed on the basis of dry weight (DW).

### *Determination of Chlorophyll, Protein, $H_2O_2$, and Lipid Peroxidation*

Chlorophyll content was determined according to Wintermans and De Mots (1965) after extraction in 96% (v/v) ethanol. For protein determination, leaves were homogenized in a 50 mM sodium phosphate buffer (pH 6.8). The extracts were centrifuged at 17,600 *g* for 20 min, and the supernatants were used for determination of protein by the method of Bradford (1976) and antioxidant enzyme activities. The $H_2O_2$ content was measured colorometrically as described by Jana and Choudhuri (1981). $H_2O_2$ was extracted by homogenizing leaf tissue with phosphate buffer (50 mM, pH 6.5) containing 1 mM hydroxylamine. The homogenate was centrifuged at 6,000 *g* for 25 min. To determine $H_2O_2$ content, extracted solution was mixed with 0.1% titanium chloride in 20% (v/v) $H_2SO_4$. The mixture was then centrifuged at 6,000 *g* for 25 min. The absorbance was measured at 410 nm. The $H_2O_2$ content was calculated using the extinction coefficient 0.28 $\mu mol^{-1}\ cm^{-1}$. Malondialdehyde (MDA), routinely used as an indicator of lipid peroxidation, was extracted with 5% trichloroacetic acid and determined according Heath and Packer (1968). Chlorophyll, protein, $H_2O_2$, and MDA contents were expressed on the basis of initial fresh weight (FW).

### *Enzyme Assays*

CAT activity was assayed by measuring the initial rate of disappearance of $H_2O_2$ (Kato and Shimizu, 1987). The decrease in $H_2O_2$ was followed as the decline in absorbance at 240 nm, and activity was calculated using the extinction coefficient (40 $mM^{-1}\ cm^{-1}$ for at 240 nm) for $H_2O_2$ (Kato and Shimizu, 1987). POX activity was measured using a modification of the procedure of MacAdam et al. (1992). Activity was calculated using the extinction coefficient (26.2 $mM^{-1}\ cm^{-1}$ at 470 nm) for tetraguaiacol. SOD was determined according to Paoletti et al. (1986). APX was determined according to Nakano and Asada (1981). The decrease in ascorbate concentration was followed as the decline in absorbance at 290 nm and activity was calculated using the extinction coefficient (2.8 $mM^{-1}\ cm^{-1}$ at 290 nm) for ascorbate. GR was determined by the method Foster and Hess (1980). One unit of activity for CAT, POX, SOD, APX, and GR was defined as the amount of enzyme which degraded 1 μmol tetraguaiacol per min, inhibited by 50% the rate of NADH oxidation observed in control, degraded 1 μmol of ascorbate per min, and decreased 1 $A_{340}$ per min, respectively. Enzyme activities are expressed on the basis of mg protein.

### *Statistical Analysis*

Statistical differences between measurements (n = 4) on different treatments or on different times were analyzed following Duncan's multiple range test or Student's *t*-test.

## Results

### *TNG67 Is More Tolerant to Cd Than TN1*

In plants, the most apparent symptom of Cd toxicity is chlorosis of the leaves (Das et al., 1997). When rice seedlings of TN1 and TNG67 were treated with 0.5 mM $CdCl_2$ for 7 d, leaf chlorosis was observed in TN1 seedlings, but not in TNG67 seedlings (Figure 1). In short term (3 d) experiments, chlorosis was first observed in the second leaf of TN1 seedlings. In the present investigation, unless otherwise indicated, Cd toxicity in the second leaf caused by 0.5 mM $CdCl_2$ was assessed by a decrease in chlorophyll and protein contents. Figure 2 shows the time courses of chlorophyll and protein contents in the second leaf of TN1 and TNG67 seedlings treated with or without 0.5 mM $CdCl_2$. It is clear that the decrease in chlorophyll and protein contents in TN1 leaves is more pronounced than in TNG 67 leaves, indicating that TNG67 seedlings are a Cd-tolerant cultivar while TN1 seedlings are Cd-sensitive. Figure 3 shows the changes in Cd content in rice seedling treated with 0.5 mM $CdCl_2$. Cd content in the second leaf and roots of TNG67 seedlings remained unchanged and slightly increased, respectively, after Cd treatment. In contrast, a marked increase in Cd content in Cd-treated TN1 leaves and roots was observed.

It seems that avoiding the build-up of Cd in the second leaf of TNG 67 prevents the toxic effect of Cd.

### *Oxidative Stress is Induced by Cd in TN1 but Not in TNG67 Leaves*

MDA content in the second leaf remained unchanged in TN1 seedlings treated without $CdCl_2$ (Figure 4). It is clear that increase in MDA content in TN1 leaves induced by $CdCl_2$ was evident at 1 d after $CdCl_2$ treatment (Figure 4).

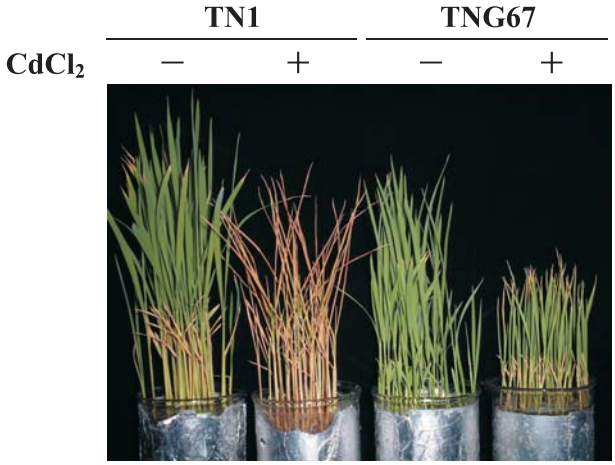


**Figure 1.** Effect of $CdCl_2$ (0.5 mM) on leaf chlorosis of Taichung Native 1 (TN1) and Tainung 67 (TNG67) rice seedlings. Picture was taken after 7 d of treatment.

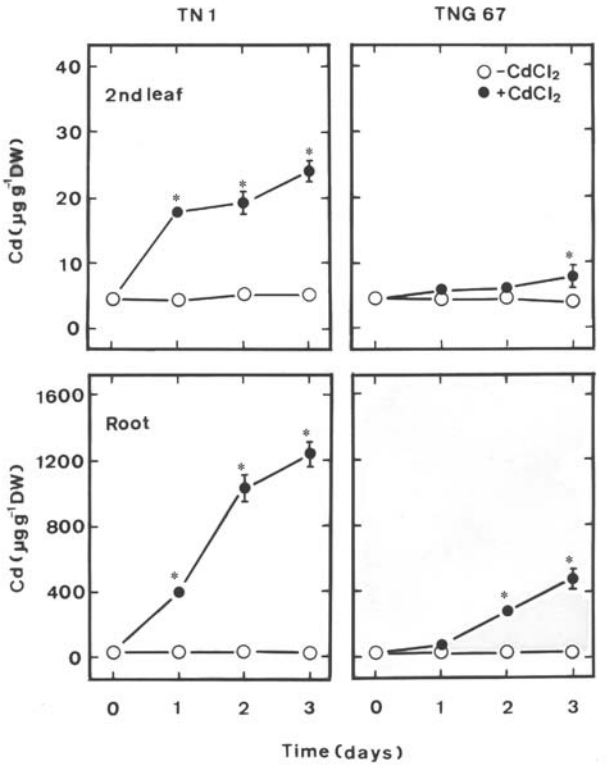


**Figure 3.** Changes in Cd content in the second leaf and roots of rice seedlings treated with or without $CdCl_2$ (0.5 mM). Bars represent standard errors (n = 4). Asterisks represent values significantly different at $P < 0.05$.

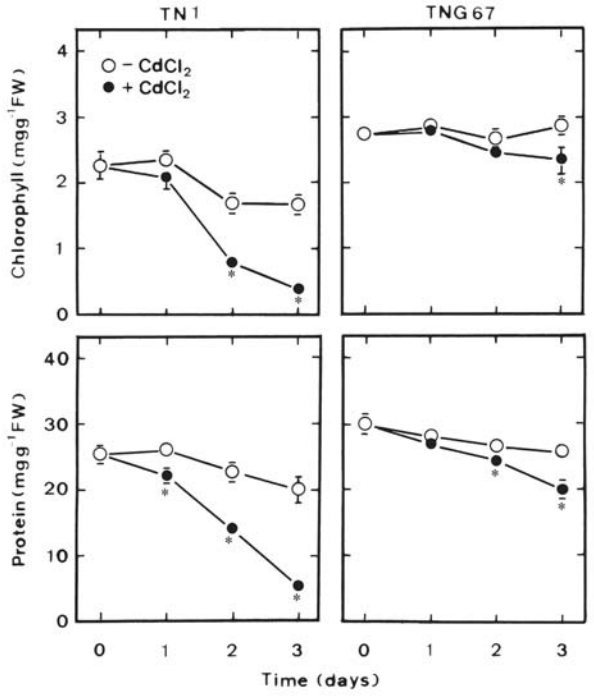


**Figure 2.** Changes in chlorophyll and protein contents in the second leaf of rice seedlings treated with or without $CdCl_2$ (0.5 mM). Bars represent standard errors (n = 4). Asterisks represent values significantly different at $P < 0.05$.

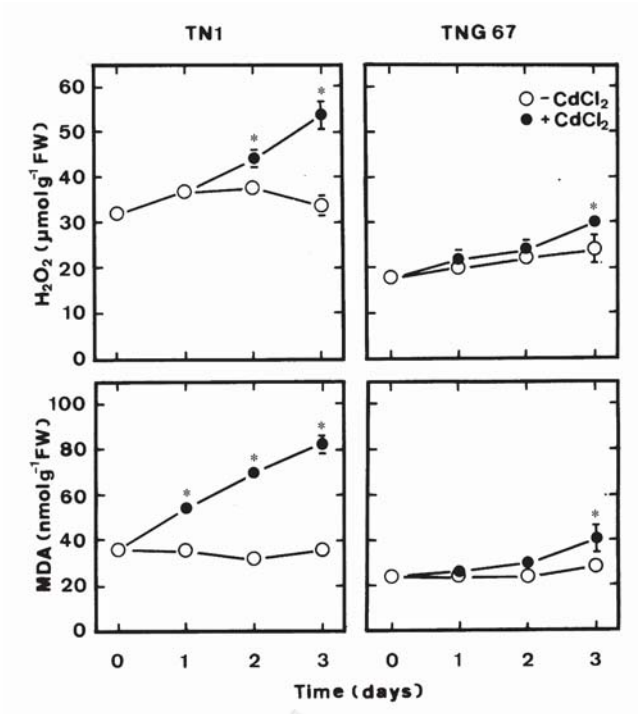


**Figure 4.** Changes in $H_2O_2$ and MDA contents in the second leaf of rice seedlings treated with or without $CdCl_2$ (0.5 mM). Bars represent standard errors (n = 4). Asterisks represent values significantly different at $P < 0.05$.

However, no increase in MDA content in TNG 67 leaves was observed throughout the $CdCl_2$ treatment (Figure 4). This showed that the Cd toxicity in TN1 leaves was linked to lipid peroxidation. Lipid peroxidation is caused by AOS (Thompson et al., 1987). $CdCl_2$ treatment also caused an increase in $H_2O_2$ content in TN1 but not in TNG67 leaves (Figure 4). These results all support the involvement of AOS as the chemical species inducing Cd toxicity in TN1 leaves.

The striking increase in lipid peroxidation seen in TN1 leaves treated with $CdCl_2$ may be a reflection of the changes in antioxidant enzyme activities. As shown in Figures 5 and 6, $CdCl_2$-treated TN1 leaves had higher activities of SOD and POX than the controls at 1 d after treatment. Higher activities of GR, APX, and CAT activities of TN1 leaves was observed at 2 and 3 d, respectively, after treatment. However, no or a slight increase in antioxidant enzyme activities in Cd-treated TNG67 leaves was observed (Figures 5 and 6).

### *Oxidative Stress in TN1 Leaves Induced by Cd Precedes the Occurrence of Cd Toxicity*

In order to know if increase in MDA content and antioxidant enzyme activities caused by $CdCl_2$ in TN1 leaves is a cause or a consequence of Cd toxicity, a short-term experiment (24 h) was conducted. As shown in Figure 7, increase in the content of MDA and the activities of SOD

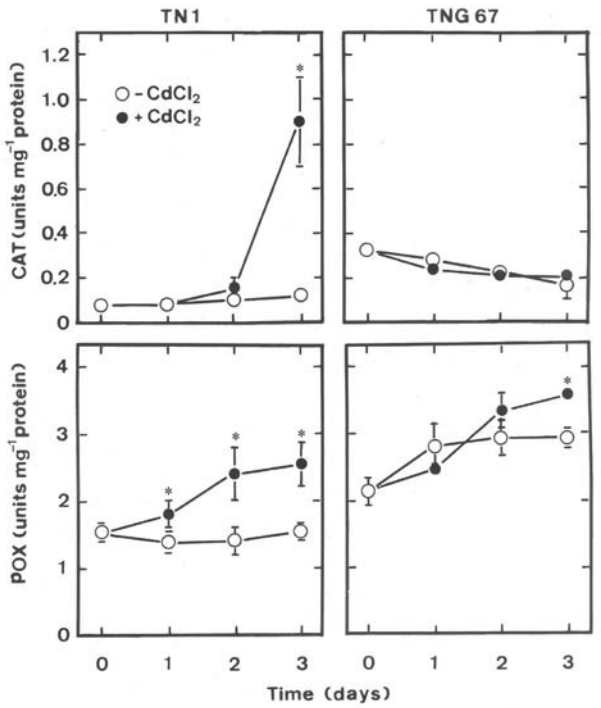


**Figure 6.** Changes in the activities of CAT and POX in the second leaf of rice seedlings treated with or without $CdCl_2$ (0.5 mM). Bars represent standard errors (n = 4). Asterisks represent values significantly different at $P < 0.05$.

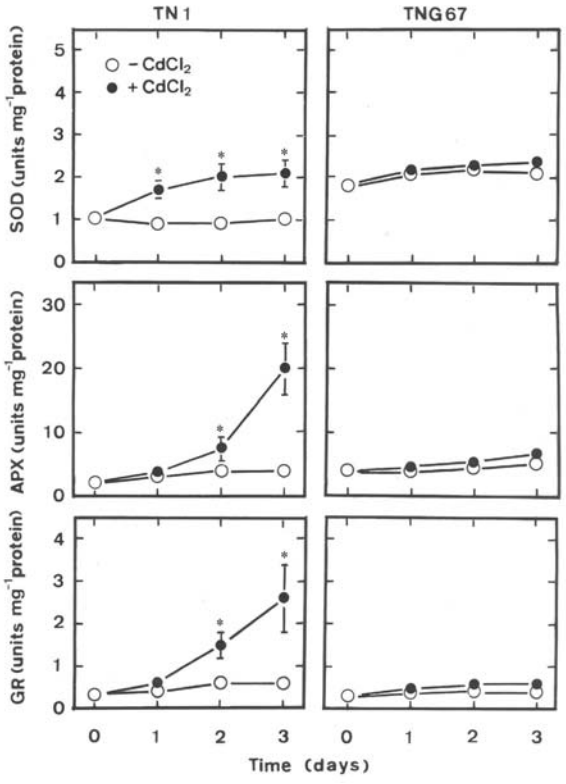


**Figure 5.** Changes in the activities of SOD, APX, and GR in the second leaf of rice seedlings treated with or without $CdCl_2$ (0.5 mM). Bars represent standard errors (n = 4). Asterisks represent values significantly different at $P < 0.05$.

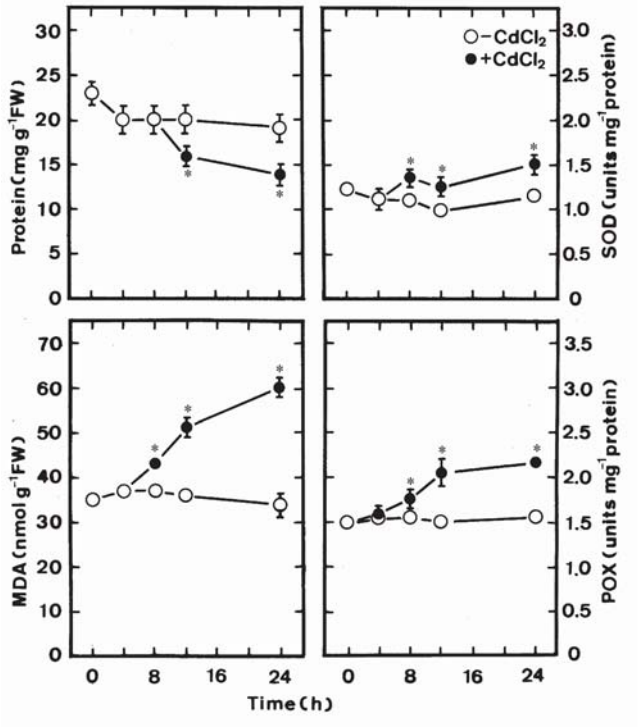


**Figure 7.** Changes in the contents of protein and MDA and the activities of SOD and POX in the second leaf of TN1 rice seedlings treated with or without $CdCl_2$ (0.5 mM) for 24 h. Bars represent standard errors (n = 4). Asterisks represent values significantly different at $P < 0.05$.

and POX in TN1 seedlings treated with $CdCl_2$ was observed prior to the occurrence of Cd toxicity (decrease in protein content), indicating that the induction of oxidative stress in Cd-treated TN1 leaves is a cause of Cd toxicity.

### *Pretreatment with Abscisic Acid (ABA) Reduces Cd Toxicity and Cd-induced Oxidative Stress in TN1 Leaves*

In a more recent work, we reported that ABA plays an important role in Cd tolerance and pretreatment of TN1 seedlings with ABA counteracting Cd-induced toxicity (Hsu and Kao, 2003a; b). If oxidative stress is an important mechanism in regulating Cd toxicity in TN1 leaves, then pretreatment with ABA is expected to reduce Cd toxicity and Cd-increased content of MDA and activities of SOD and POX in TN1 leaves. Since some chlorosis was observed in the second leaf of TN1 seedlings treated with ABA for 2 d, Cd toxicity was evaluated by using the third leaf and 1.5 mM $CdCl_2$. As expected, ABA pretreatment reduced Cd toxicity and reduced Cd-induced increase in the content of MDA and increase in the activities of SOD and POX in the third leaf of TN1 seedlings (Figure 8 and 9).

### *Fluridone (Flu) Treatment Enhances Toxicity and Oxidative Stress in Cd-treated TNG67 Leaves*

Previously, we have shown that treatment with Flu, an inhibitor of ABA biosynthesis, inhibited the increase in ABA content and enhanced toxicity in the second leaf of TNG67 seedlings treated with $CdCl_2$ (Hsu and Kao, 2003a; b). Here, we also observed that Flu enhanced toxicity, increased the content of MDA, and increased the activities of SOD and POX in Cd-treated TNG 67 leaves (Figures 10 and 11). It is interesting to note that the effect of Flu on Cd toxicity and oxidative stress in TNG67 can be reversed by the application of ABA (Figures 10 and 11).

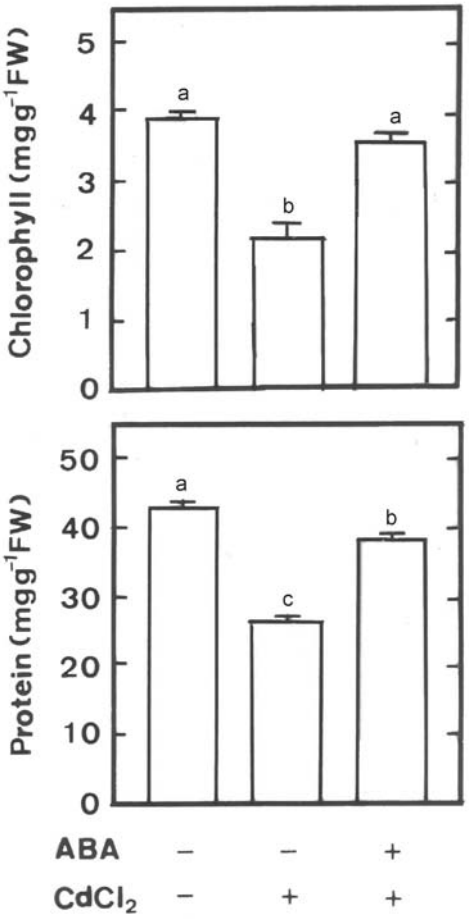


**Figure 8.** Effect of ABA-pretreatment on chlorophyll and protein contents in the third leaf of TN1 rice seedlings treated with or without $CdCl_2$ (1.5 mM). Taichung Native 1 (TN1) rice seedlings were pretreated with ABA (5 µM) for 2 d and then treated with or without $CdCl_2$ (1.5 mM) for 2 d. Bars represent standard errors (n = 4). Values with the same letter are not significantly different at $P < 0.05$.

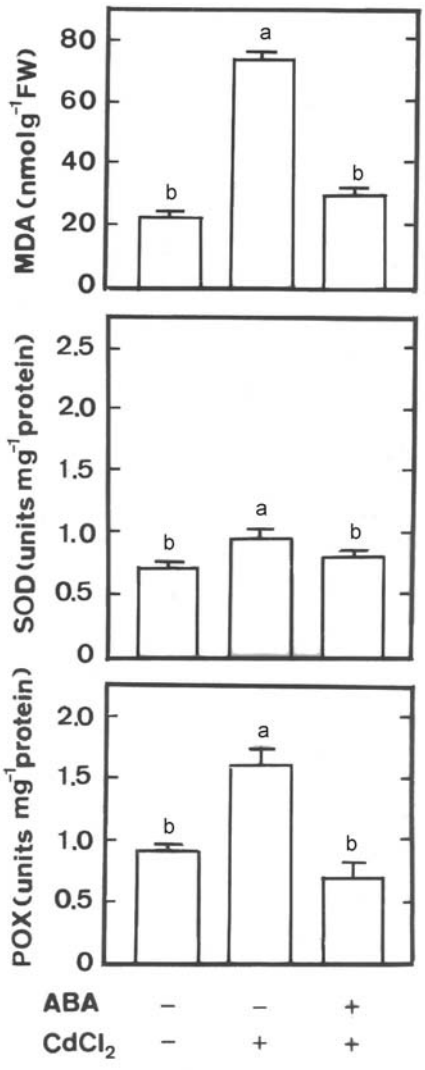


**Figure 9.** Effect of ABA-pretreatment on the content of MDA and the activities of SOD and POX in the third leaf of Taichung Native 1 (TN1) rice seedlings treated with or without $CdCl_2$ (1.5 mM). TN1 rice seedlings were pretreated with ABA (5 µM) for 2 d and then treated with or without $CdCl_2$ (1.5 mM) for 2 d. Bars represent standard errors (n = 4). Values with the same letter are not significantly different at $P < 0.05$.

## Discussion

Plants have a range of potential mechanisms at the cellular level that might be involved in detoxification and thus tolerance to heavy metals. These all appear to be involved primarily in avoiding the build-up of toxic concentrations at sensitive sites within the cell and thus preventing damage (Hall, 2002). Obviously Cd-tolerant plant must be able to prevent the absorption of excess Cd. Of the two rice cultivars used in the present study, more absorption of Cd was detected in Cd-sensitive TN1 seedlings than in Cd-tolerant TNG67 (Figure 3). Jarvis et al. (1976) examined the Cd distribution in 23 species and found that the higher up in the plant one checked, the lower the Cd concentration, which decreased in the following order; fibrous roots > storage roots > stems > leaves. We also observed that Cd content was higher in roots than leaves in both TN1 and TNG67 seedlings (Figure 3).

It has been documented that Cd can cause an increased production of AOS, including $H_2O_2$ (Piqueras et al., 1999; Romero-Puertas et al., 1999; Shah et al., 2001; Sandalio et al., 2001; Schützendübel et al., 2001; Olmos et al., 2003) and induce lipid peroxidation (Shaw, 1995; Gallego et al., 1996; Benavides and Tomaro, 1996; Chaoui et al., 1997; Lozano-Rodríquez et al., 1997; Dixit et al., 2001; Shah et al., 2001; Chien et al., 2002). This suggests that Cd treatment causes oxidative stress in plants. In the present study, five lines of evidence indicated that oxidative stress seems to be involved in toxicity of rice seedlings caused by $CdCl_2$. First, on treatment with $CdCl_2$, lipid peroxidation, $H_2O_2$ content, and antioxidant enzyme activities increased in the leaves of the Cd-sensitive TN1 but not in the Cd-tolerant TNG67 (Figures 4, 5, and 6). Second, Cd-increased lipid peroxidation, and the activities of SOD and POX preceded the appearance of Cd toxicity in TN1 leaves (Figure 7). Third, pretreatment with ABA enhanced Cd tolerance and reduced Cd-increased lipid peroxidation and the activities of SOD and POX in TN1 leaves (Figures 8 and 9). Fourth, exogenous application of ABA biosynthesis

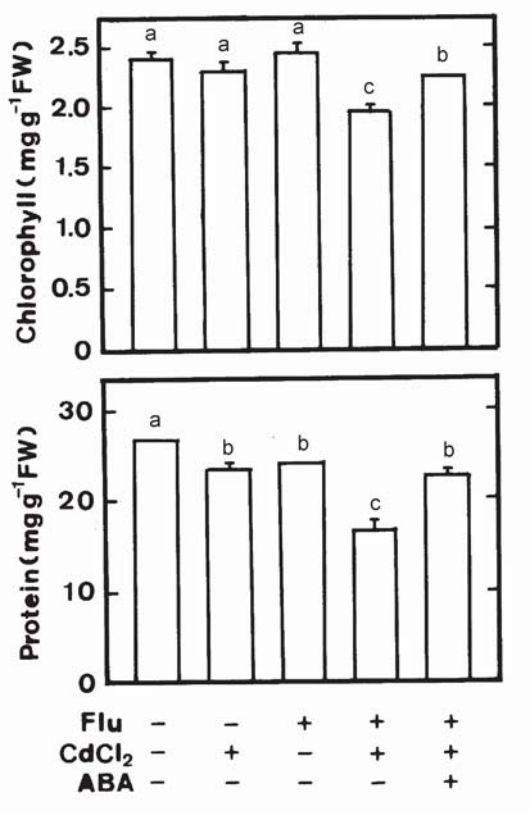


**Figure 10.** Effect of fluridone (Flu, 0.2 mM) on the contents of chlorophyll and protein in the second leaf of Tainung 67 (TNG67) rice seedlings treated with $CdCl_2$ (0.5 mM) and ABA (5 μM). Chlorophyll and protein contents were measured after 2 d of treatments. Bars represent standard errors (n = 4). Values with the same letter are not significantly different at $P<0.05$.

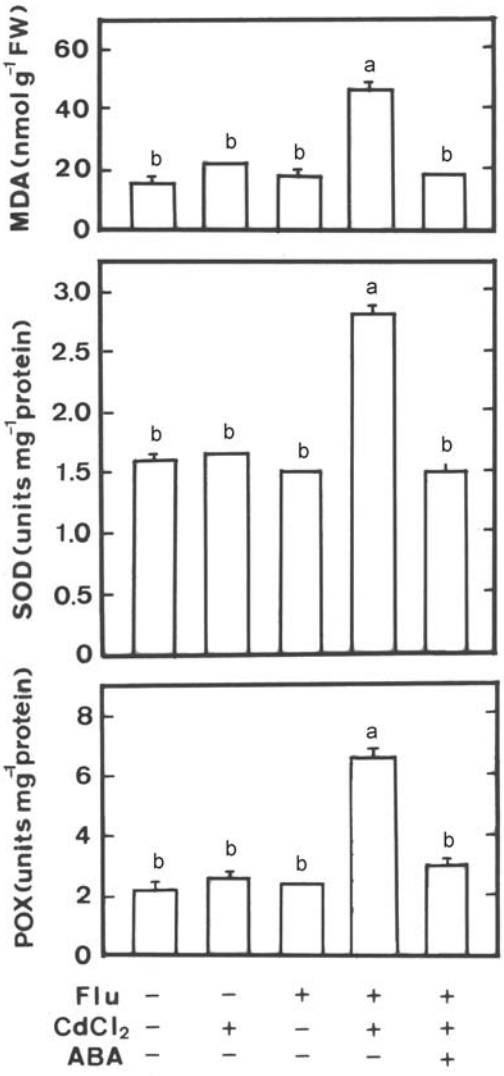


**Figure 11.** Effect of fluridone (Flu, 0.2 mM) on the content of MDA and the activities of SOD and POX in the second leaf of Tainung 67 (TNG67) rice seedlings treated with $CdCl_2$ (0.5 mM) and ABA (5 μM). Chlorophyll and protein contents were measured after 2 d of treatments. Bars represent standard errors (n = 4). Values with the same letter are not significantly different at $P < 0.05$.

inhibitor, Flu, decreased Cd tolerance, increased lipid peroxidation, and increased SOD and POX activities in Cd-treated TNG67 leaves (Figures 10 and 11). Fifth, Flu effect on Cd toxicity, lipid peroxidation, and the activities of SOD POX in Cd-treated TNG67 leaves was reversed by the application of ABA (Figures 10 and 11).

Cadmium produced an enhancement of MDA content in TN1 leaves (Figure 4), which is an indicator of lipid peroxidation and, therefore, of oxidative stress. Similar increases in MDA content by Cd treatment have been observed in *Phaseolus vulgaris* (Shaw, 1995; Chaoui et al., 1997), *Helianthus annuus* (Gallego et al., 1996), *Pisum sativum* (Lozano-Rodríguez et al., 1997; Dixit et al., 2001), and *Oryza sativa* plants (Shah et al., 2001). The peroxidation of cell membranes severely affects its functionality and integrity and can produce irreversible damage to cell function (Thompson et al., 1987). Lipid peroxidation can be initiated by AOS (Thompson et al., 1987). Unlike Cu and Fe, Cd is not a redox metal and therefore cannot catalyze Fenton-type reactions yielding AOS. However, Cd can induce oxidative stress indirectly by producing a disturbance in chloroplasts. Thus, Cd produced degradation of chlorophyll and carotenoids as well as an inhibition of their biosynthesis (Bazzaz et al., 1992), which can produce disturbances in electron transport rates of PSI and PSII, leading to generation of AOS. Ascorbate is a major antioxidant in photosynthetic and non-photosynthetic tissues which reacts directly with AOS and is utilized as a substrate for APX catalysed $H_2O_2$ detoxification (Noctor and Foyer, 1998). Reduced glutathione (GSH) is involved in ascorbate regeneration and functions also as a direct antioxidant of AOS (Noctor and Foyer, 1998). We have shown that $CdCl_2$ treatment significantly reduced the contents of GSH and ascorbate in rice leaves in TN1 leaves (Hsu and Kao, 2004). In a recent review, Schützendübel and Polle (2002) also suggest that the depletion of GSH is apparently a critical step in Cd-induced AOS.

Our results indicated an increase in antioxidant enzyme activities in Cd-treated TN1 leaves (Figures 4, 5 and 6). In line with our observations, a Cd-induced increase in antioxidant enzyme activities was also reported by Dixit et al. (2001), Schützendübel et al. (2001), Shah et al. (2001), and Iannelli et al. (2002). Nitric oxide, a bioactive free radical, is known to act as an AOS scavenger (Lamattina et al., 2003). In a recent report, we were able to show that nitric oxide effectively reduced Cd-increased MDA content and antioxidant enzyme activities in TN1 leaves (Hsu and Kao, 2004). It appears that Cd-induced increase in antioxidant enzyme activities is a consequence of AOS overproduction (Thompson et al., 1987).

Information is limited about the mechanism with which Cd leads to the generation of $H_2O_2$. In cultured tobacco cells (BY-2 line), diphenyleneiodonium and imidazole, both inhibitors of neutrophil NADPH oxidase, prevented the generation of $H_2O_2$ induced by Cd (Olmos et al., 2003). These data suggest the involvement of an NADPH oxidase-like enzyme leading to $H_2O_2$ production through $O_2^-$ dismutation by SOD. We have not investigated whether $H_2O_2$ production in Cd-treated TN1 leaves was augmented by the stimulation of NADPH oxidase-like enzyme. However, this is likely to occur because Cd treatment resulted in an increase in $H_2O_2$ content and in SOD activity in TN1 leaves (Figures 4 and 5).

The most novel aspects of this report are the findings that ABA reduces oxidative stress indicators in Cd sensitive cultivar (TN1) and Flu increases indicators in Cd tolerant cultivar (TNG67) (Figures 9 and 11). The simplest explanation of this finding is that ABA closed stomata and reduced Cd uptake while Flu opened stomata and increased Cd uptake. Indeed, in our previous work, we have shown that exogenous ABA application could result in the decrease in transpiration rate and Cd content of TN1 seedlings and Flu treatment caused not only the block of ABA biosynthesis but also the increase in transpiration rate and Cd content in TNG67 seedlings (Hsu and Kao, 2003a).

In conclusion, we provide evidence that oxidative stress is differently expressed in TN1 and TNG67 rice seedlings in response to $CdCl_2$. Results also suggest that $CdCl_2$ causes oxidative stress and $CdCl_2$-induced toxicity is mediated through oxidative stress in TN1 leaves.

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# 鎘所誘導之抗氧化酵素活性變化：耐鎘與不耐鎘水稻幼苗之比較

郭美君　高景輝

國立臺灣大學農藝學系

本研究探討鎘處理對水稻兩個品種過氧化氫與 malondialdehyde（MDA）含量以及抗氧化酵素活性變化之影響。氯化鎘處理增加不耐鎘品種台中在來一號（TN1）葉片之過氧化氫與 MDA 含量以及增加抗氧化酵素［superoxide dismutase (SOD), ascorbate peroxidase, glutathione reductase, catalase 與 peroxidase (POX)］之活性，但不影響耐鎘品種台農 67 號 (TNG67) 葉片過氧化氫與 MDA 含量以及抗氧化酵素之活性。氯化鎘處理之 TN1 葉片，其 MDA 含量與 SOD 及 POX 活性之增加早於毒害之發生。以離層酸前處理 TN1 幼苗，增加 TN1 對鎘的耐性，同時降低鎘所誘導之 MDA 含量與 SOD 及 POX 活性之增加。以離層酸合成抑制劑（fluridone）與鎘同時處理 TNG67 幼苗，可降低 TNG67 對鎘之耐性，增加 MDA 含量與 SOD 及 POX 之活性。同時，此種 fluridone 處理 TNG67 幼苗之效應又可被離層酸處理所恢復。本研究之結果似乎說明，氯化鎘處理 TN1 幼苗，其葉片可表現氧化逆境，然而鎘處理 TNG67 幼苗，其葉片不會表現氧化逆境，同時也說明鎘處理後 TN1 幼苗葉片所表現之鎘毒害是經由氧化逆境所造成。

**關鍵詞：**離層酸；鎘；水稻；氧化逆境。