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CHANGES OF SUPEROXIDE DISMUTASE ACTIVITY IN WHEAT SEEDLINGS EXPOSED TO NATURAL ENVIRONMENTAL STRESSES

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ABSTRACT

The effects of natural environmental stresses on the superoxide dismutase (SOD) enzyme activity were investigated in *Triticum aestivum* L. (cv. Bezostaya-1, cv. Seri-82, cv. Kıraç-66) and *Triticum durum* Desf. (cv. Kızıltan-91, cv. Kunduru 414-44, cv. Ç.1252). The seedlings that grown at normal (24/16°C), low (5/-5°C) and high (40/30°C) temperature conditions were exposed to waterlogging, drought and salinity (0.7 % NaCl) stresses. Changes of SOD activity in seedlings were investigated against to cross interactions between temperature and water-salt stresses. Activity of total SOD, Mn SOD and Cu/Zn SOD at low and high temperatures were slightly decreased. SOD activity was increased under drought conditions and salt stress. The highest activity of SOD was measured under isochronal effects of high temperature and drought-salinity stress. The results show that antioxidant defence system is important in plants that exposed to heavy water and salt stressses.

Key words: Stress, wheat, cross tolerance, superoxide dismutase.

INTRODUCTION

Oxidative stress arises from the deleterious reactions of oxygen. These reactions are mediated by reduced oxygen species such as superoxide radicals and hydrogen peroxide (Bowler *et al.* 1992). Superoxide dismutases (SOD; EC 1.15.1.1) have been identified as an essential component in defence mechanism of plants againts oxidative stress (Fridovich 1986). SODs are metal containing enzymes that catalyze the dismutation reaction of superoxide (O_2^{-}) into H_2O_2 and O_2. SODs are generally occur in three different molecular forms as Cu/Zn SOD, Mn SOD and Fe SOD (Bowler *et al.* 1992).

Plants are expose to temperature and water stress at different levels in natural conditions. Generally plants are effected isochronically more stress factors than one. Plants may improve to a degree the physiological and morphological adaptations to these stress conditions to a degree. Free radical generation is increase in plants under environmental stresses (Gillham and Dodge 1987). In this case an antioxidant defence system that includes enzymic and non-enzymic components is induced (Schöner and Krause 1990).

In this study, we tested SOD enzymes activities at low and high temperature conditions and under water-salt stresses.

MATERIALS AND METHODS

In this study, *Triticum aestivum* L. (cv. Bezostaya-1, cv. Seri-82, cv. Kıraç-66) and *Triticum durum* Desf. (cv. Kızıltan-91, cv. Kunduru 414-44, cv. Ç.1252) seedlings were used. Seedlings were grown at 24/16 °C (day / night) for six days in growth chamber. After 6^{th} days groups of seedlings were transferred to low (5/-5 °C), normal (24/16 °C), and high (40/30 °C) temperature conditions and the sub-groups, were isochronically exposed to waterlogging, drought and salt (0.7 %) stresses for six days.

0.5 g fresh leaf material were homogenized in 5 ml of 0.1 M potassium phosphate buffer with 0.1 mM EDTA (pH: 7). The homogenate centrifuged at 7000xg for 5 min, and the supernatant was vacuum filtered through Whatman GF/A glass microfibre discs. Filtrats were stored at –70 °C (Schöner and Krause 1990).

The activity of SOD was assayed as described in Beyer and Fridovich (1987). The reaction mixture (3 ml) contained 50 mM potassium phosphate buffer, including 0.025% triton X-100 and 0.1 mM EDTA (pH: 8), enzyme aliquots, 12 mM L-metionine, 75 μM nitro blue tetrazolium chloride (NBT) and 2 μM riboflavin. Reactions were carried out at 25°C and under a fluorescent tube for ten min in an incubator. One unit of SOD activity was defined as the amount of enzyme required to cause 50% inhibition of the rate of NBT reduction measured at 560 nm.

Activity of Mn SOD was measured after addition of potassium cyanide (2 mM) to the assay solution. Activity of Cu/Zn SOD was calculated by subtracting Mn SOD activity from the total SOD activity. Activity of Fe SOD was not measurable (Çakmak *et al.* 1997).

Spesific activity of SOD was calculated by dividing total activity to soluble protein amount. Soluble protein was determined according to Lowry *et al.* (1951).

Data obtained from the analyses and calculations were evaluated statistically by balanced anova.

RESULTS

Total SOD activity showed no correlation with temperature stress unless it was accompanied by other stress parameters studied. However total SOD activity was significantly ($P < 0.01$) increased by combined high temperature-water deficiency stresses, but not by water and salt stresses at low temperatures. Highest total SOD activity was measured under drought and salinity stresses at high temperature conditions (Figure 1 and Figure 4).

Cu/Zn SOD activity was induced by drought and salinity stresses. Water deficiency caused by directly drought or by salinity, significantly increased the Cu/Zn SOD activity at high temperature conditions. Cu/Zn SOD activity increased up to 10 times in all *T. durum* genotype groups treated, as compared to the control groups at combined high temperature-salt stress conditions (Figure 2).

Mn SOD activity decreased by both low and high temperature stresses, the activity slightly increased by water stress on the other hand (Figure 3). Heavy water stress caused by high temperature and soil salinity increased cytosolic and chloroplastic Cu/Zn SOD activity more than mitocondrial Mn SOD activity.

Total SOD activity of Kıraç-66 and Ç-1252 genotypes significantly increased under high temperature-drought stress ($P<0.01$). In this condition, the source of total SOD activity increase was Cu/Zn SOD. High temperature-waterlogging interaction increased total SOD activity approximately 50% in all genotypes.

DISCUSSION

SOD enzymes activities increase under low and high temperature stresses (Schoner and Krause 1990, Rabinowitch and Sklan 1980). Tsang *et al.* (1991) reported that only cytosolic Cu/Zn SOD mRNA level significantly increase in leaves of *Nicotiana plumbaginifolia* that exposed to heat shock. mRNA level of chloroplastic Fe SOD rises at combined effects of chilling and intense light conditions (Tsang *et al.* 1991).

Many plants are able to survive at a period of waterlogging only to die upon subsequent re-exposure to air, this suggests that oxidative damage may occur during the recovery phase (Bowler *et al.* 1992). A dramatic increase in total SOD activity coincides with the anoxic phase in the resistant variety but not in the sensitive one (Monk *et al.* 1987). The author also suggested that the increase in SOD activity was vital in the protection of plants against the oxidant stress generated upon re-exposure to air.

Drought conditions induce the antioxidant defence system. Cytosolic Cu/Zn SOD was induced significantly by drought while the chloroplastic Cu/Zn SOD remaind uneffected (Perl-Treves and Galun 1993).

Tolerance to all environmental stresses can increased by several possible mechanisms. SOD is one of the components that can determine the level of this cross tolerance (Bowler *et al.* 1992). This study shows that isochronic effect of temperature and water-salt stresses lead to significant differences.

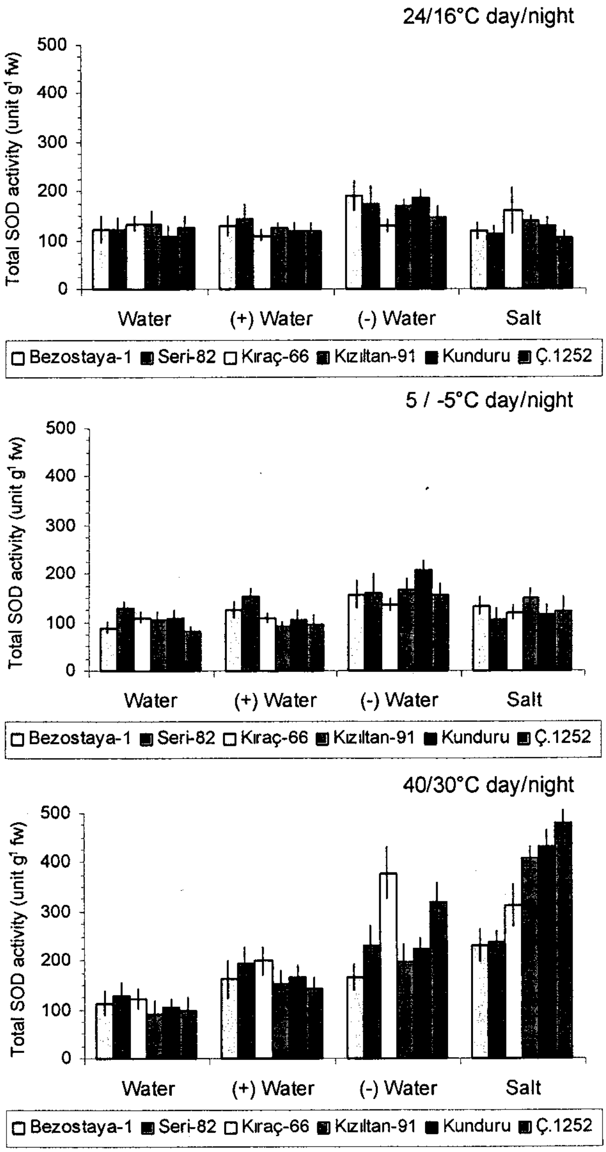

Figure 1. Changes of total SOD activity in wheat seedlings (*T. aestivum* cv. Bezostaya-1, Seri-82, Kıraç-66, and *T. durum* cv. Kızıltan-91, Kunduru 414-44, Ç-1252) that grown under temperature and water-salt stresses (N=3).

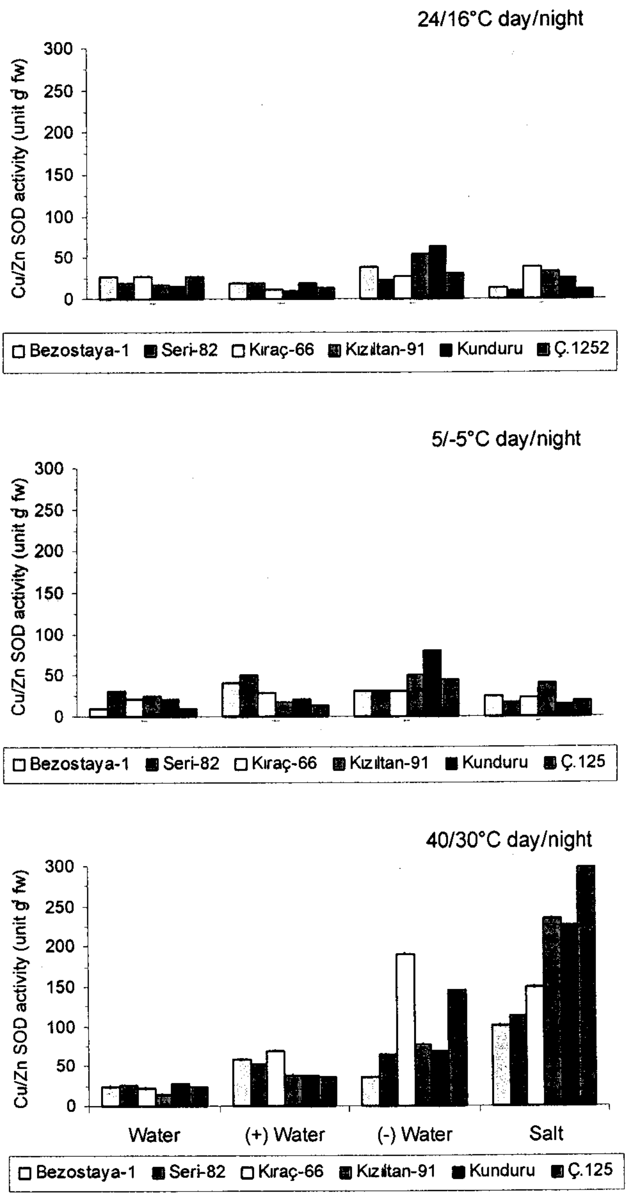

Figure 2. Changes of Cu / Zn SOD activity in wheat seedlings (*T. aestivum* cv. Bezostaya-1, Seri-82, Kıraç-66, and *T. durum* cv. Kızıltan-91, Kunduru 414-44, Ç-1252) that grown under temperature and water-salt stresses.

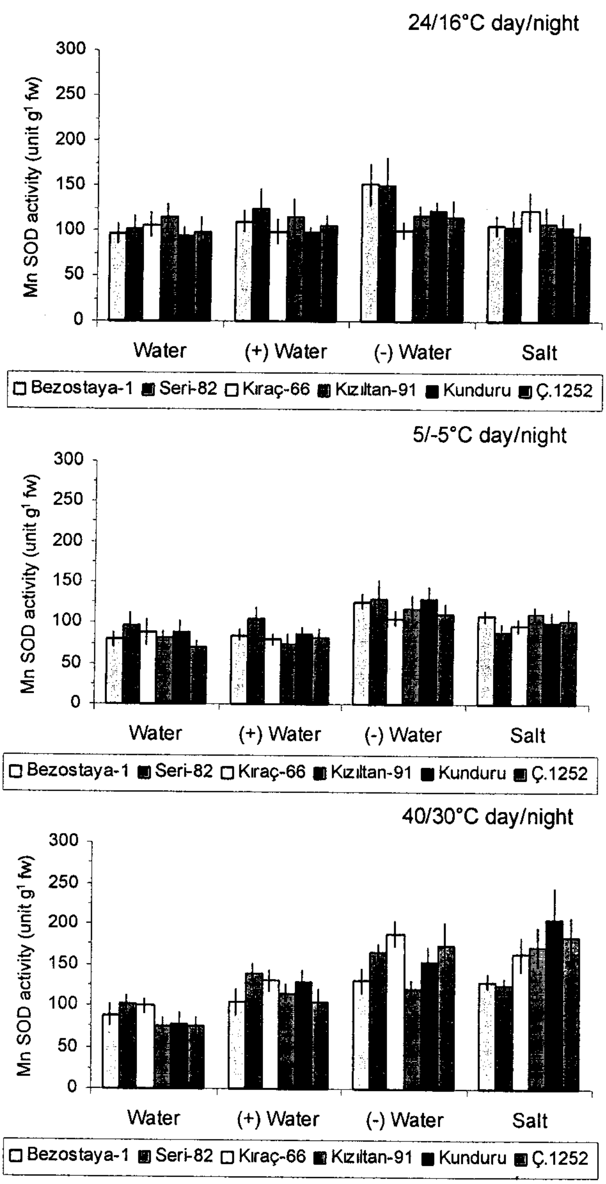

Figure 3. Changes of Mn SOD activity in wheat seedlings (*T. aestivum* cv. Bezostaya-1, Seri-82, Kıraç-66, and *T. durum* cv. Kızıltan-91, Kunduru 414-44, Ç-1252) that grown under temperature and water-salt stresses (N=3).

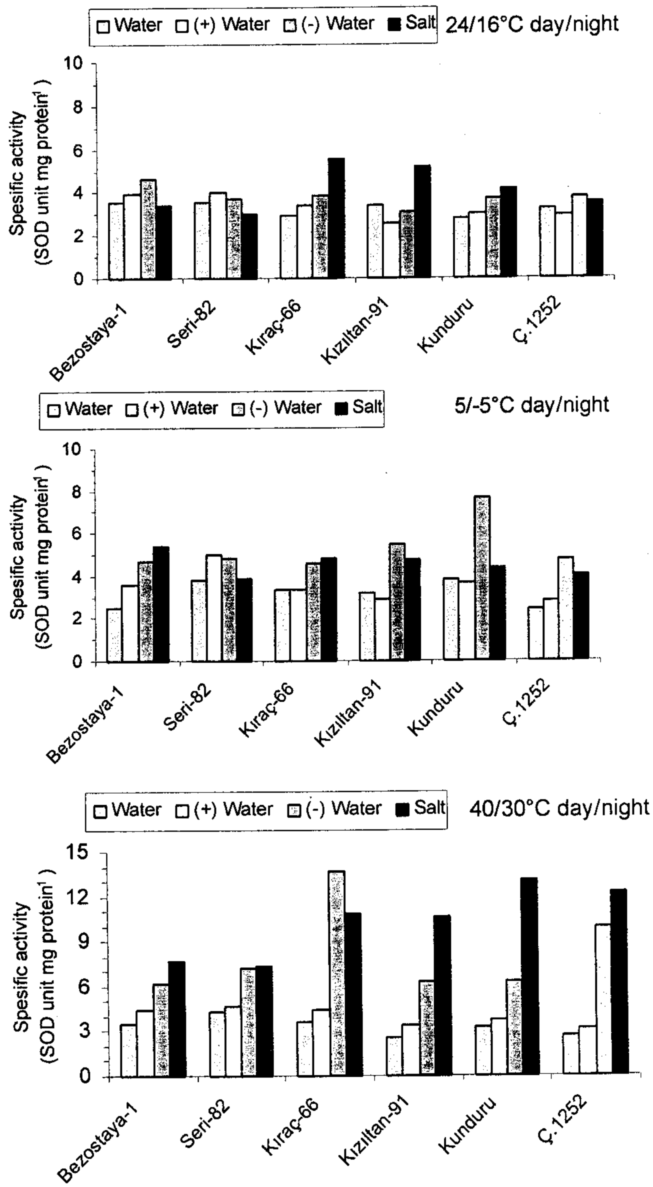

Figure 4. Changes of SOD spesific activity in wheat seedlings (*T. aestivum* cv. Bezostaya-1, Seri-82, Kıraç-66, and *T. durum* cv. Kızıltan-91, Kunduru 414-44, Ç-1252) that grown under temperature and water-salt stresses.

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