THE ROS-MEDITED COPPER TOXICITY IN RYE VARIETIES

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Introduction

The copper ore processing generates large amounts of wastes extremely difficult for revitalisation due to metal concentrations toxic to plants (Lottermoser 2014). The effects of environmental stress in plants are know to be mediated, at least partially, by an enhanced generation of the reactive oxygen species (ROS; Stepien and Klobus 2005). Entry of toxic ions into plant provokes inhibition of photosynthesis leading to generation of superoxide ($^{\bullet}O_2^-$) *via* photoreduction of O_2 or singlet oxygen through the interaction with triplet-excited chlorophyll (Asada, 2000; Foyer et al., 2002). ROS are highly reactive and can cause widespread damage to membranes, proteins and DNA. In addition, it is well known that transition metals like Cu catalyze formation of extremely cytotoxic hydroxyl radicals ($^{\bullet}OH$) in the Haber–Weiss reaction between superoxide and H_2O_2 (Stepien and Klobus, 2005).

Materials and Methods

The pot experiment with eleven rye (*Seceale Cereale*) varieties were grown on the post-flotation sediment from the copper ore processing. The plant material was harvested 28 DAS and analysed for Cu with AAS. The total Cu determined with ICP-MS in both the sediment and in the water extract (1:10) of the sediment were 1775 mg kg^{-1} and 0,052 mg dm^{-3}, respectively. The rate of superoxide ($^{\bullet}O_2^-$) generation was measured following formazan formation by nitroblue tetrazolium (NBT) dye with $^{\bullet}O_2^-$ (Omoto et al. 2013).Hydroxyl radical ($^{\bullet}OH$) content was measured as the accumulation of thiobarbituric acid-reactive degradation products from the reaction between 2-deoxyribose and $^{\bullet}OH$ (Tiedemann 1997). The level of lipid peroxidation was determined as a thiobarbituric acid-reactive substances (TBARS) according to Stepien and Klobus (2005),

Results

In the course of the experiment significant variety-dependent elevation in the tissue Cu level was observed in plants grown on the post-flotation sediment (Fig. 1). In spite of the high total Cu content in the sediment, due to high pH and carbonates the predominant Cu form in the sediment WE was $CuCO_3$ (aq), with Cu^{+2} being only 11% (Tab. 1).The rye varieties studied revealed an increased levels of $^{\bullet}O_2^-$; however, these were not significantly different amongst genotypes, except the two with the highest copper uptake

(Fig. 2). In contrast, the levels of $^{\bullet}OH$ were noticeably dependent on the Cu accumulation (Fig. 3). Free radicals provoke the degradation of membrane lipids; therefore, oxidative stress was expected to produce an increased level of lipid peroxidation. A significant increase in the TBARS level was observed in all the wheat varieties grown on the post-flotation sediment (Fig. 4), with this being Cu-dependent and following the extent of $^{\bullet}OH$ formation.

Conclusions

The results obtained clearly demonstrated the relationship between the Cu uptake by the rye genotypes and the level of oxidative stress, with the latter being demonstrated by an increased ROS formation. The harmful influence of the ROS, with particular reactivity of hydroxyl radicals, resulted in widespread damage to cellular compartments, as confirmed by increased lipid peroxidation.

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