

Chelated micronutrient fertilizers as effective antioxidants applied for foliar plant treatment

Valentina M. Pakhomova* and Anisa I. Daminova

Kazan State Agrarian University, Kazan, 420015, Russia

Abstract. The antioxidant status of spring wheat cells was studied for foliar treatment with a 0.1 % solution of copper, molybdenum-containing chelated micronutrient grade ZhUSS-2 in field experiments when cultivating plants on soil with a low content of these trace elements. The activation of the antioxidant enzymes of superoxide dismutase and peroxidase, as well as a decrease in the activity of lipid peroxidation (according to the content of malondialdehyde) under the action of this drug in all phases of plant vegetation was identified. A similar effect of iron-, manganese- and zinc-containing ZhUSS micronutrient fertilizers is analyzed. The conclusion is made about the antioxidant effect of these chelated micronutrient fertilizers.

1 Introduction

In recent years, attention of plant physiologists and molecular biologists has been focused on the functional role of active (“aggressive”) oxygen forms (AOFs) in plants. They have extreme reactivity and can oxidize almost all cell biomolecules, causing, in particular, lipid peroxidation (LP), and, as a result, damage to the membranes. The AOFs include $O_2^{\bullet-}$, HO^{\bullet} , H_2O_2 and 1O_2 [1].

Under the physiological norms, the content of AOFs is low due to their elimination by antioxidant (AO) protection enzymes: superoxide dismutase (SOD), catalase, peroxidase, glutathione S-transferase, phospholipidhydroperoxide oxidase, dehydrate, glutathione peroxidase, and dehydrate. Low molecular weight antioxidants (AO) are vitamins C and E, carotenoids, glutathione tripeptide, amino acids cysteine and methionine, etc. [1–4].

The intracellular concentration of AOFs is very small: $H_2O_2 - 10^{-2}$ micromol/l; $O_2^{\bullet-}$ and $HO^{\bullet} - 10^{-5}$ micromol/l [1]. They perform a signaling function and participate in the plant growth and a development program.

It is important that lipid peroxidation is also necessary in conditions of the physiological norm of cells. In particular, fatty acid hydroperoxides can be involved in cell differentiation. Low LP intensity is needed for autophagy of endoplasmic reticulum membranes after the termination of hydroxylation of foreign compounds.

LP also leads to a change and renewal of membrane lipids and, ultimately, can regulate the activity of enzymes and receptors of membranes. The formation of hydroperoxides can change the permeability of membranes, leading to the activation of hydrolases, the release of calcium ions into the cytosol from intracellular

depots and an increase in the activity of cell metabolism as a whole.

Under conditions of insignificant activation of lipid peroxidation, when regulating its intensity with the help of AO, the described processes are reversible and play a regulatory role.

Finally, AOFs, although capable of causing mutational processes in cells, can play a positive role in the adaptation of biosystems when a change in external environment occurs [5, 6].

However, various environmental stress factors in plant cells provoke overproduction of AOFs and, as a result, the development of oxidative stress, which is one of the indicators of the non-specific adaptation syndrome of all biosystems.

Oxidative stress is caused by drought, salinization, low and high temperatures, atmospheric pollutants, heavy soil metals, herbicides, ultraviolet radiation, starvation, anoxia, infectious pathogenesis, phytophage insects, physical damage to plants by herbivores, etc. [7–9].

Even under normal conditions, the AO systems do not provide plants with complete protection against the AOFs: some proteins and membranes are always damaged and replaced with new ones. Under the influence of external stress factors, a significant imbalance occurs between the formation of AOFs, possibilities of their elimination and the speed of repair processes. As a result, oxidative stress develops. It can be caused by overproduction of AOFs and a decrease in the AO activity [10].

It has been established that the violation of the mineral nutrition of plants, including microelements, violates the activity of AO-enzymes, which decreases the stress resistance of plants, their productivity and quality of crop production. It is also important that many

* Corresponding author: pahomovav@mail.ru

protective AO-enzymes contain such trace elements as copper, zinc, manganese, iron [1, 11, 12].

It is necessary to study various methods of correcting the activity of plant AO enzymes both under stressful environmental conditions and with an imbalance of mineral nutrition elements (including a lack of trace elements).

Kazan State Agrarian University has developed new liquid chelated micronutrient fertilizers (ZhUSSy), which include different combinations of trace elements.

The purpose of the article is to study some indicators of the antioxidant status of plants under the influence of foliar treatment with chelated micronutrients (Cu, Mo-containing ZhUSS-2). The objective is: 1. to study the activity of lipid peroxidation (LP); 2. to study the activity of certain antioxidant enzymes.

2 Materials and methods

The studies were conducted on spring wheat (grade MiS) cultivated on the experimental fields of Kazan State Agrarian University in different years of research (2013–2015). The cultivation technology is used for spring wheat in this agroclimatic zone. The soil was characterized by a poor content of copper and molybdenum [13].

The field experiment design:

- option 1 – plants without treatment (control);
- option 2 – treatment of plants with chelated micronutrient fertilizers was carried out when tillering;
- option 3 – treatment of plants with chelated micronutrient fertilizers was carried out when shooting;
- option 4 – treatment of plants with chelated microfertilizers was carried out when tillering, shooting and earing – flowering.

The composition of chelated microfertilizers includes (g/l): Cu – 32...40, Mo – 14...22, ethanolamine – 170...200; pH 10...11; TU 2189-002-OP-2789377698; the registration number in the State catalog of pesticides and agrochemicals of the Russian Federation is 19-8002 (9333)–0309–1.

2.1 Research methods

The content of malondialdehyde (MDA) was determined photometrically during interaction with thiobarbiturate [14]; the SOD activity was determined by inhibiting the reaction of nitrosine blue tetrazolium reduction [14]; the peroxidase activity – by the benzidine oxidation reaction [15]; the amount of soluble protein – by interaction with the Coomassie dye G-250 [14].

The figures show the average of four replicates of the most repetitive experiment with standard deviations. The experiments were carried out 3-10 times. Differences in the variants were evaluated by the Student's criterion at $P_{0.05}$.

3 Results and discussions

It is known that the main targets of the AOFs are cell membranes that are easily damaged. AOFs trigger chain reactions of membrane destruction associated with lipoperoxidation (LPO). This process is a free radical reaction with the formation of intermediate compounds, including malondialdehyde (MDA). Some of them can be toxic and modify the structure of proteins and nucleic acids. Under the normal conditions, LPO is a normal metabolic process, but under certain conditions, its intensity goes beyond the physiological norm, which leads to the pathological changes in the cell [16].

In the field experiments, the MDA content in the leaves of spring wheat plants decreased at any rate of treatment with ZhUSS-2 and in all phases of vegetation (Fig. 1). The effect of reducing the formation of MDA was observed 4 days after the treatment.

The effect of ZhUSS-2 activated superoxide dismutase (SOD) (Fig. 2). The action of an inhibitor of copper-dependent enzymes of sodium diethyldithiocarbamate (DDC) decreases the activity of SOD [17], which is the basis to believe that an increase in its activity is due to the microelement of copper (Cu) of the drug used.

It is known that the elimination of hydrogen peroxide resulting from the SOD activity occurs with the participation of catalase and peroxidases.

Therefore, an increase in the SOD activity was accompanied by activation of peroxidase in all experimental variants (Fig. 3). Copper contributes to the synthesis of Fe-containing enzymes, in particular, peroxidase [18].

The SOD activation was observed during non-root treatment of wheat with chelated micronutrients which contain zinc, iron and manganese [13]. Different forms of this enzyme were described: Cu, Zn-containing SOD, Fe-containing SOD and Mn-containing SOD [1]. Under the action of Fe-containing micronutrient fertilizers, an increase in the activity and iron-containing protective enzymes of catalase and peroxidase was observed. This was observed under the combined stress of plant growth [13]. However, SOD is a key element in maintaining the constant level of $O_2^{\bullet-}$ [1].

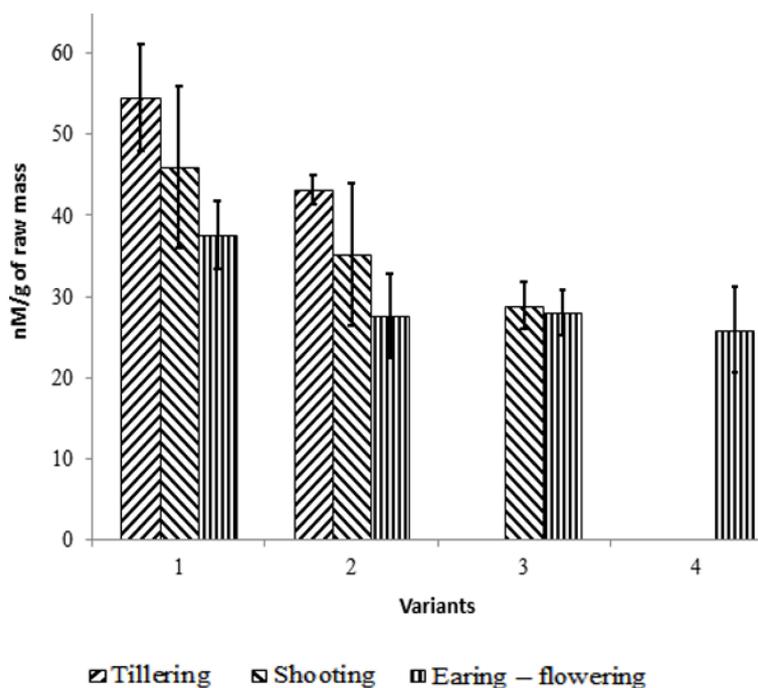


Fig. 1. The effect of ZhUSS-2 on the content of MDA during the growing season of wheat

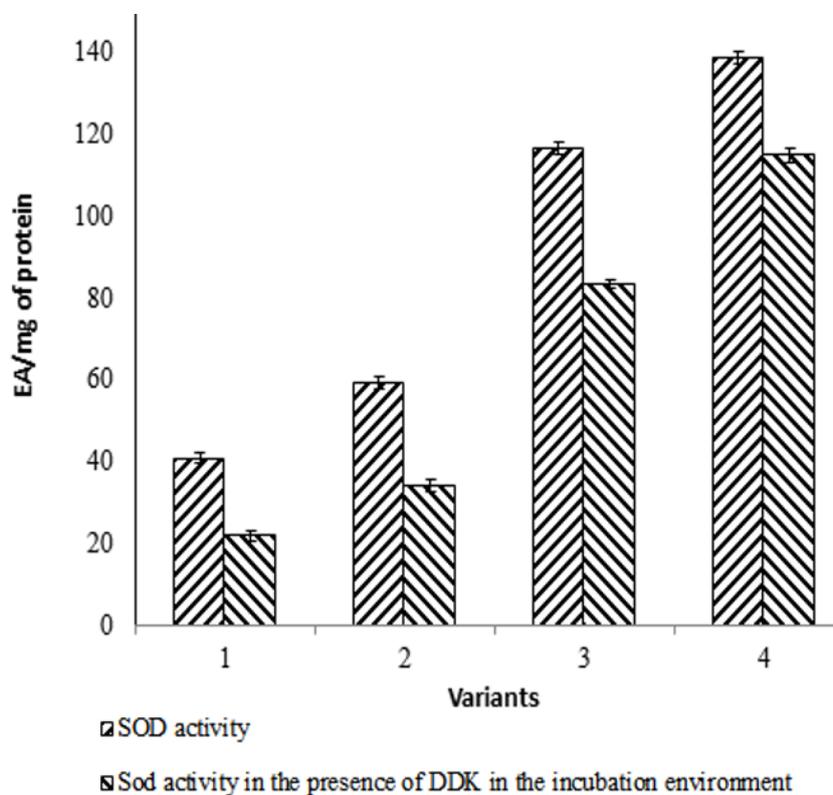


Fig. 2. The effect of ZhUSS-2 on the SOD activity

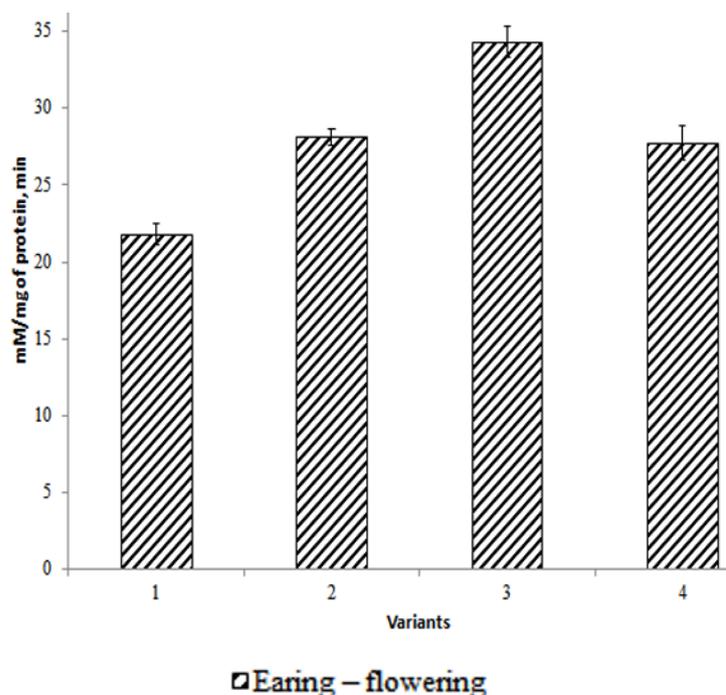


Fig. 3. The effect of ZhUSS-2 on the peroxidase activity

4 Conclusion

1. When cultivating spring wheat in the absence of trace elements (zinc, copper, manganese and iron), chelate micronutrients containing can be used to activate the SOD antioxidant protective enzymes and peroxidase;
2. Chelated micronutrient fertilizers have a pronounced protective antioxidant effect and can be used in crop production under stressful environmental factors.

References

1. G.N. Chupakhina, P.V. Maslennikov, L.N. Skrypnik, *Natural antioxidants (environmental aspect)* 111 (Publishing House of the Baltic Federal University named after I. Kant, Kaliningrad, 2011)
2. L. Liu, Y. Liu, J. Rao et al., *Molecular Biology* **47**, 591–601 (2013)
3. V. Demidchik, *Environ. Exp. Bot.* **109**, 212–228 (2015)
4. Yu.E. Kolupaev, T.O. Yastreb, A.I. Oboznyy, *Plant physiology* **63**, 346–359 (2016)
5. V.D. Kreslavsky, D.A. Los', S.I. Allahverdiyev et al., *Plant physiology* **59(2)**, 163–178 (2012)
6. H. Rogers, S. Munne-Bosch, *Plant Physiol.* **171(3)**, 1560–1568 (2016)
7. B. Barna, J. Fodor, B.D. Harrach et al., *Plant Physiol. Biochem.* **59**, 37–43 (2012)
8. S.V. Veselova, G.F. Burkhanova, I.V. Maximov, *Materials of the II Int. Symp. "Molecular Aspects of Plant Redox Metabolism" and the Int. Sci. School "The Role of Active Oxygen Forms in Plant Life"* 87-92 (First Printing House, Ufa, 2017)
9. Yu.E. Kolupaev, *Successes of modern biology* **136**, 181–198 (2016)
10. J. Bose, A. Rodrigo-Moreno, S. Shabala, *J. Exp. Bot.* **65**, 1241–1257 (2014)
11. N.P. Bityutsky, *Trace elements of higher plants* 368 (Publishing House of St. Petersburg University, St. Petersburg, 2011)
12. M. Kumar, P. Kumari, C.R.K. Reddy et al., *Adv. Bot. Res.* **71**, 91–123 (2014)
13. V.M. Pakhomova, A.I. Daminova, I.A. Gaysin, *Agrochemical Bull.* **6**, 29–31 (2015)
14. V.V. Kuznetsov, V.V. Kuznetsov, G.A. Romanov, eds. *Molecular genetic and biochemical methods in modern plant biology* 487 (BINOM. Knowledge laboratory, Moscow, 2012)
15. A.I. Ermakov, V.V. Arasimovich, N.P. Yarosh, Yu.V. Peruanskiy, G.A. Lukovnikova, M.I. Ikonnikova, *Methods of biochemical research of plants* 445 (Agropromizdat, Leningrad, 1987)
16. G.A. Abilova, P.Sh. Shagrudinova, *Materials of the II Int. Symp. "Molecular Aspects of Plant Redox Metabolism" and the Int. Sci. School "The Role of Active Oxygen Forms in Plant Life"* 43-46 (First Printing House, Ufa, 2017)
17. R. Dawson, D. Elliot, W. Elliot, C. Jones, *Handbook of biochemist* (Mir, Moscow, 1991)
18. I.G. Kirillova, *Materials of the II Int. Symp. "Molecular Aspects of Plant Redox Metabolism" and the Int. Sci. School "The Role of Active Oxygen Forms in Plant Life"* 129–132 (First Printing House, Ufa, 2017)