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Role of Nanomaterials of Analcite, Tripoli and Silicon dioxide in Plants under Drought Conditions in Triticum aestivum L.

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Abstract. The effect of the application of nanoparticles of analcite, tripoli and silicon dioxide to gray podzolic soil at a dose of 250 mg per 100 g on drought resistance of wheat seedlings was studied under laboratory conditions. The dependence of the silicon effect on soil moisture level was also evaluated. For this, four levels of soil moisture were modeled: 20%, 40%, 60% and 80% field capacity. The positive effect has been established for all variants tested: seedlings growth criteria as well as content of photosynthetic pigments increased, while transpiration intensity decreased under water deficit. Application of nanoparticles caused accumulation of protective antioxidants. While proline, malondialdehyde, souble sugars content decreased. The dependence of analcite, tripoli and silicon dioxide effect on soil moisture level was discussed.

Key words: analcite, tripoli, silicon dioxide, nanoparticles, drought resistance, *Triticum aestivum* L.

Introduction

Drought is a worldwide issue that impacts seriously on the security of food production. Global climate change makes this even worse. Winter wheat is almost the most important crop in the world (GORNALL *et al.*, 2010; CURTIS & HALFORD, 2014). Studies have shown that drought has many adverse impacts on crops, inhibiting growth and photosynthesis, changing morphology, decreasing production and photosynthetic pigment content, affecting ion balance in plants, etc. (HONGBO *et al.*, 2005; SOFO et *al.*, 2005; TIAN & LEI, 2007; SINGH *et al.*, 2012; SHARMA *et al.*, 2012).

Silicon has not been considered as a generally essential element for higher plants, partly because its roles are poorly understood. However, numerous studies have demonstrated that silicon plays an important role in tolerance of plants to environmental stresses, especially to drought (GONG *et al.*, 2005; PEI *et al.*, 2010; SHEN *et al.*, 2010; GUNTZER *et al.*, 2012; ROSITSKA, 2012; ZAIMENKO *et al.*, 2014), but the defense mechanisms provided by Si are far from being understood. Silicon fertilization has the potential to mitigate environmental stresses and soil nutrient depletion and as a consequence is an alternative to the extensive use of phytosanitary and NPK fertilizers for maintaining sustainable agriculture (GUNTZER *et al.*, 2012). We suggest that silicon is useful for drought tolerance improvement of plants.

The objective of this study was to evaluate experimentally the effects of silicon application under drought conditions.

Materials and Methods

Wheat leaves were isolated from plants *Triticum aestivum* L., cv. Podil'ska 90 (drought–sensitive) grown during one month under

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laboratory conditions at 22-24 °C. Soil humidity of gray podzolic soils was maintained at 20%, 40%, 60%, 80% of field capacity. As a source of silicon was used natural minerals (analcite and tripoli) and silicon dioxide (SiO_2) at a dose of 250 mg per 100 g of soil. The concentration of silicon compounds was selected on the basis of our previous studies (ROSITSKA, 2014).

Lipid peroxidation was measured as malondialdehyde (MDA) in the leaves and was analyzed following KABASHNYKOVA *et al.* (2007) method. Proline was extracted from freshly cut leaves by 3% sulfosalicyl acid. Its quantitative content was determined using qualitative reaction with ninhydrin on spectrophotometer "Specord 200" (STATSENKO & BUTYLKIN, 1999). Catalase activity was determined by the method of PLESHKOV (1985).

The content of pigments was extracted from leaves by acetone. The solution mixture was analyzed for Chlorophyll-a, Chlorophyll-b and carotenoids content on spectrophotometer "Specord 200" (POCHINOK, 1976).

Soluble sugars were determined based on the method of Bertrand (KOLUSHEVA & MARTINOVA, 2011). The transpiration rate was determined by registering changes in weight of cut transpirating leaves for short time intervals (TRETYAKOV, 1990).

The results presented in the tables are the means of four replications. The data were statistically analyzed using the least significant difference (LSD) test ($p<0.05$). The effect of analcite dose, soil type, moisture level, and species of tested plants on their adaption to soil drought was assessed using analysis of variance (ANOVA) and correlation analysis with the help of Statistica 6.0 software (STATSOFT INC., 2001).

Results and Discussion

Plant growth and development

Water stress depressed the growth of shoot and root. There is a general agreement on the positive effect of Si application on the biomass yield under water stress (ENEJI *et al.*, 2008). Indeed, increases of biomass and grain yields have been observed on a large set of crops (ENEJI *et al.*, 2008; SHEN *et al.*, 2010; PEI *et al.*, 2010). We report the results of the study of natural minerals' effects on wheat leaves growth rate in Fig. 1 and 2. It appears that the values of the parameters in experimental plants exceed the control ones. After adding silicon in different forms to the substrate, we observed a significant increase in growth rate in comparison to control plants. Particularly, the growth rate of aboveground part of wheat seedling in experimental plants exceeded growth rate in the control plants at the level 34−73% (analcite), 10−94% (tripoli) and 30−77% (silicon dioxide) depending on the soil humidity. The growth rate of underground part of wheat seedling was increased by 12−66% (analcite), by 7−91% (tripoli) and by 5−66% (silicon dioxide) in comparison to control.

The content of chlorophyll and carotenoids

Along with determining the growth rate of wheat leaves, we studied distribution of photosynthetic pigments in leaves under different moisture conditions (Table 1). It is well established that drought inhibits photosynthesis, induces changes in chlorophyll content and composition, and also damages the photosynthetic apparatus (NAYYAR & GUPTA, 2006). SAIRAM *et al.* (1997/98) reported an increase in lipid peroxidation and a decrease in the level of total chlorophyll and carotenoids. Increased MDA accumulation has been correlated with a reduction in the relative water content and photosynthetic pigment content of leaves subjected to prolonged water deficit (JIANG & HUANG, 2001). Furthermore, dehydration of tissue inhibits photochemical activities and brings about a reduction in the activity of Calvin–Benson–Basshamn cycle enzymes (MONAKHOVA & CHERNYADEV, 2002). GONG *et al.* (2005) observed that Si increased antioxidant defenses and therefore maintained physiological processes such as photosynthesis.

The detailed analysis reveals increasing of the total amount of chlorophyll. That is to say the chlorophyll content from 20 to 60% soil humidity in experimental plants increased by 1.4–2.7 times (analcite), by 1.6–2.8 times (tripoli) and by 1.8–3.3 times (silicon dioxide). But at 80% humidity, these figures, in contrast, decreased, except for analcite. Our experiments showed a positive effect of silicon compounds in the biosynthesis of carotenoids.

Soluble sugars

The concentration of soluble sugars in general increases or at least remains constant under water deficit stress. Recent studies report the accumulation of simple sugars such as glucose and fructose following an increase in the invertase activity in the leaves of the drought challenged plants (PINHEIRO *et al.*, 2001; TROUVERIE *et al.*, 2003).

Analysis of the results showed (Table 2) reduction of soluble sugars in all treatments of the experiment compared to the control plants. The most significant decrease fixed in 20% soil humidity. In particular, analcite reduced concentration of mono- and disaccharides by 1.2–2.1 times, tripoli and silicon dioxide – by 1.1–2.7 and by 1.1–1.5 times respectively.

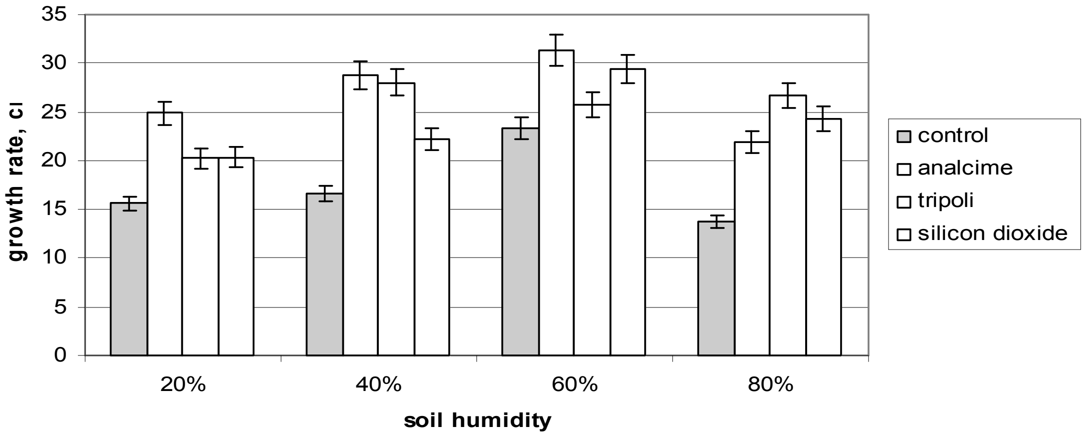

Fig. 1. The impact of different forms of silicon on the growth rate of aboveground part of wheat seedling. Vertical bars – LSD (least significant difference).

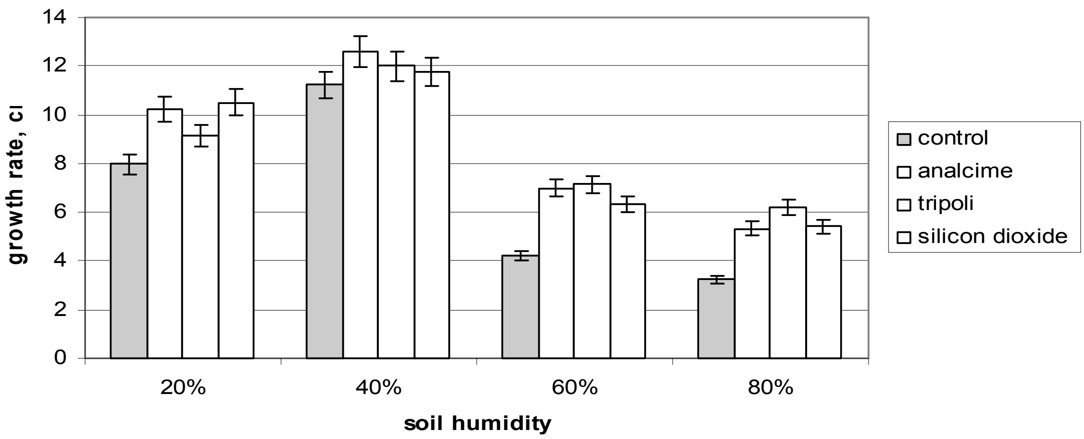

Fig. 2. The impact of different forms of silicon on the growth rate of underground part of wheat seedling. Vertical bars – LSD (least significant difference).

Table 1. The impact of silicon on photosynthetic pigments content in wheat leaves.

Soil humidity	Chlorophyll, mg/g fresh weight		Carotenoids, mg/g fresh weight
	a	b	
	Control		
20%	0.20	0.13	0.06
40%	0.33	0.14	0.06
60%	0.19	0.08	0.05
80%	0.58	0.23	0.11
	Analcite		
20%	0.63	0.25	0.14
40%	0.45	0.20	0.09
60%	0.43	0.18	0.09
80%	0.60	0.25	0.13
	Tripoli		
20%	0.54	0.20	0.10
40%	0.55	0.23	0.11
60%	0.54	0.23	0.10
80%	0.50	0.20	0.09
	Silicon dioxide		
20%	0.77	0.32	0.14
40%	0.72	0.28	0.13
60%	0.35	0.15	0.08
80%	0.56	0.23	0.10
LSD	*0.021*	*0.007*	*0.004*

Table 2. The impact of silicon on soluble sugars content in wheat leaves, %.

Soil humidity	Control	Analcite	Tripoli	Silicon dioxide
		Monosaccharides		
20%	0.57	0.27	0.21	0.40
40%	0.21	0.15	0.10	0.15
60%	0.17	0.10	0.10	0.16
80%	0.10	0.10	0.10	0.04
		Disaccharides		
20%	0.89	0.74	0.77	0.59
40%	0.95	0.87	0.88	0.82
60%	0.94	0.90	0.88	0.78
80%	0.89	0.79	0.79	0.83
LSD	*0.02*	*0.02*	*0.02*	*0.02*

Transpiration intensity

Wheat plants subjected to drought and treated with Si maintained higher stomatal conductance, relative water content, and water potential than non-treated plants. Besides, leaves were larger and thicker, thereby limiting the loss of water through transpiration (GONG *et al.*, 2003; HATTORI *et al.*, 2005) and reducing water consumption (ENEJI *et al.*, 2005). It was established decreasing in the intensity of transpiration after adding analcite by 2.0–8.0 times, tripoli – by 1.5–4.0 times, and silicon dioxide – by 1.4–3.2 times compared to the control (Fig. 3).

Lipid peroxidation

It has been shown that under stress conditions, MDA accumulation takes place in plants due to membrane lipid peroxidation (HONGBO *et al.*, 2005). It is an effective means of assessing oxidative stress induced membrane damage including changes to the intrinsic properties of the membrane, such as fluidity, ion transport, loss of enzyme activity and protein cross–linking. These changes eventually result in cell death (MELONI *et al.*, 2003; SAIRAM *et al.*, 2005; SHARMA *et. al.*, 2012). It is possible that the detrimental effect of water deficit in soil is associated with levels of lipid peroxidation in tissues. SINGH *et al.* (2012) showed that instability of biological membranes, as reflected by lipid peroxidation, was greater in drought–sensitive than in drought–tolerant wheat (*Triticum aestivum* L.) genotypes.

Thus, the concentration of MDA in all variants of the experiment decreased with increasing of soil humidity (Fig. 4). In particular, analcite and tripoli reduced concentration of MDA by 1.2–2.4 times, silicon dioxide – by 1.2–3.6 times. These results suggest that stress-induced membrane lipid peroxidation could be partly alleviated by added silicon.

Proline content

Proline is one of the minor amino acids in non-stressed plants, but accumulates very strongly during adaptation to water stress. Special functions proposed for proline are the stabilization of membranes under water deficit or role as small compatible molecule (BÜSSIS & HEINEKE, 1998). Some of the crop plants for instance wheat is marked by the accumulation and mobilization of proline was found to increase tolerance towards water deficit stress (NAYYAR & WALIA, 2003). The analysis revealed that adding analcite reduced proline concentration by 1.1–1.5 times, tripoli – by 1.1–2.1 times and silicon dioxide – by 1.3–1.5 times (Fig. 5).

Catalase activity

It was reported that catalase is highly expressed enzyme, particularly in plant cell, and is thus an integral part of the plant antioxidant system. Catalase functions as one of the key enzymes in the scavenging of reactive oxygen species and affects on toxic H_2O_2 levels in a cell (SOFO *et al.*, 2005). Furthermore, SINGH *et al.* (2012) concluded that the progressive increase in MDA during plant development may have resulted from greater levels of hydrogen peroxide. Moreover, a very strong and positive correlation was reported to exist between levels of hydrogen peroxide and MDA in the leaves of wheat plants grown under irrigated and rain–fed conditions. Conversely, the correlation between H_2O_2 and MDA was positive (TIAN & LEI, 2007). The analysis of the data showed a positive effect of silicon compounds on the activity of catalase (Fig. 6). In experiments observed a direct correlation between the level of soil humidity and the activity of the enzyme.

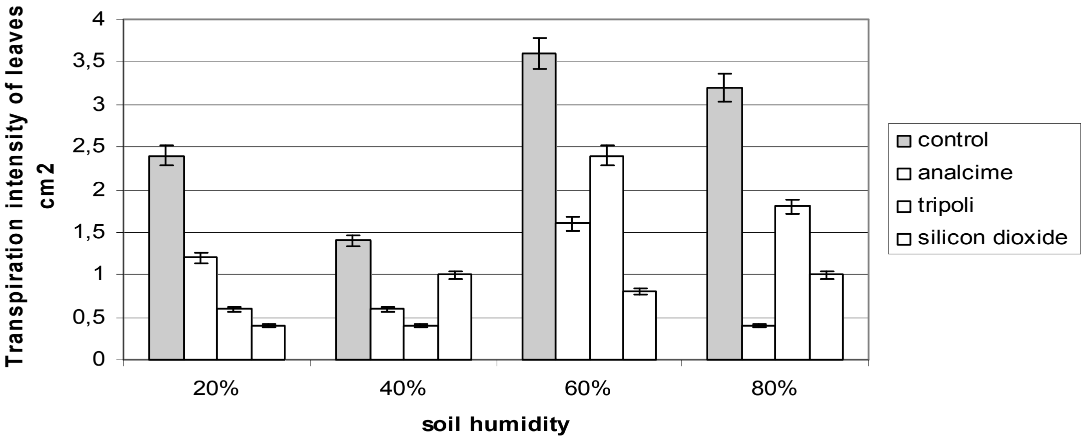

Fig. 3. Transpiration intensity of wheat seedling under different sources of silicon. Vertical bars – LSD (least significant difference).

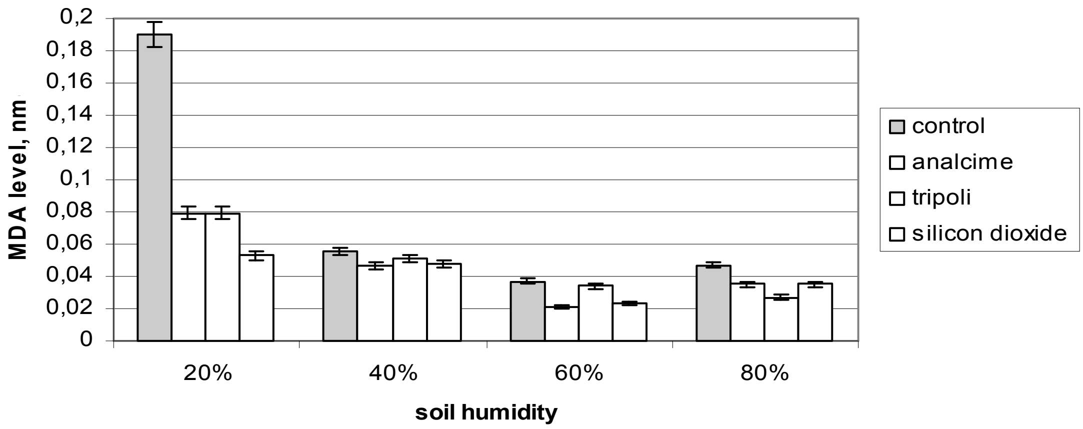

Fig.4. The impact of silicon on MDA content in wheat leaves. Vertical bars – LSD (least significant difference).

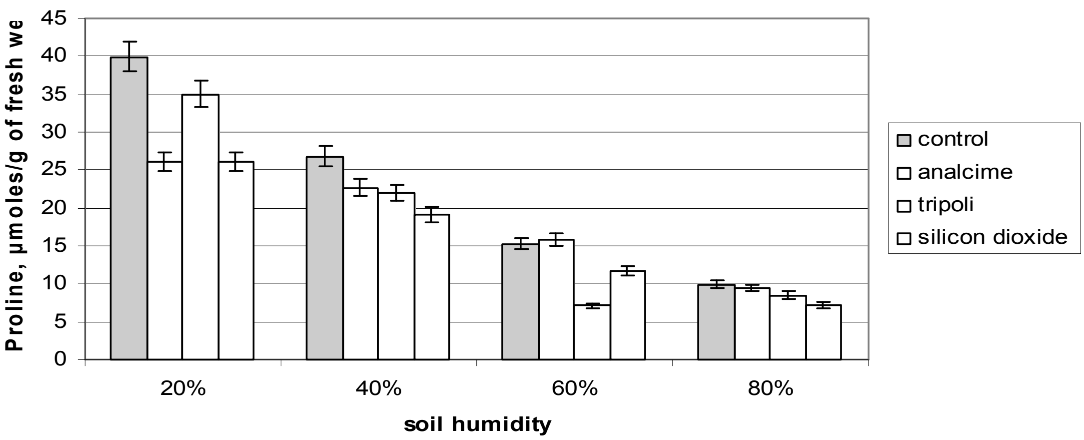

Fig.5. The impact of silicon on proline content in wheat leaves. Vertical bars – LSD (least significant difference).

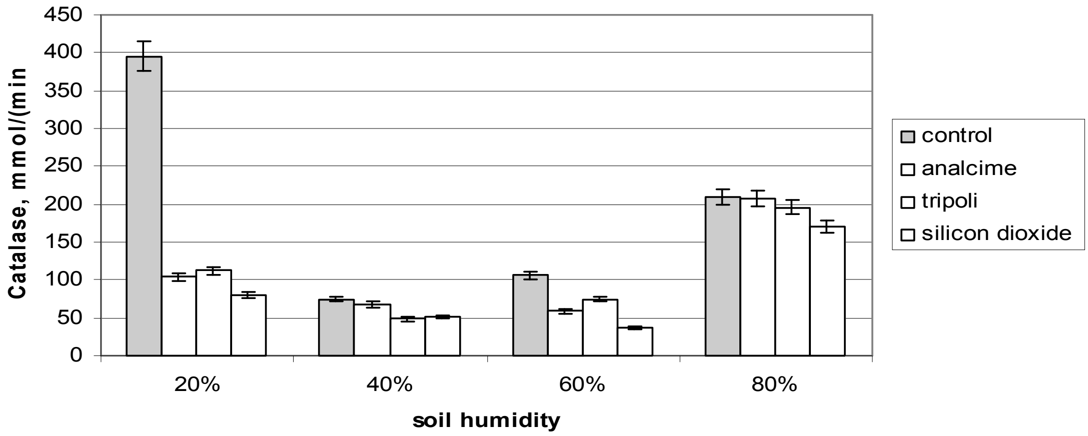

Fig. 6. The impact of silicon on catalase activity in wheat leaves. Vertical bars – LSD (least significant difference).

Conclusions

The data presented indicate that the application of nanoparticles of analcite, tripoli and silicon dioxide to gray podzolic soil contributes to the increase in the resistance of wheat plants to drought stress. The positive effect has been established for all variants tested: seedlings growth criteria as well as content of photosynthetic pigments increased, while transpiration intensity decreased under water deficit. Application of nanoparticles also caused a decrease in the content of proline, souble sugars and malondialdehyde.

Thus, the physiological and biochemical parameters measured indicated that Si could alleviate seedling damage under water stress conditions, improve mineral nutrition, increase drought resistance of plants *Triticum aestivum* L., and stimulate growth processes by enhancing antioxidant system.

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