

Short Communication

Stimulation of Seed Viability by Means of Dispersed Solutions  
of Copper and Silver Nanoparticles

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It was shown that water dispersed solutions at different concentration of copper (NPCu) and silver (NPAg) nanoparticles substantially influence germination of seeds of grain and vegetable crops. Stimulating effect was revealed after application of ultra-low concentrations NPCu (up to  $10^{-17}$  mg/l) and NPAg (up to  $10^{-13}$  mol/l). Treatment of seeds with sol boosts their resistance to fungal pathogens and low temperature and also increases productivity of plants in the field environment.

**Keywords:** Water dispersed solutions, Copper and silver nanoparticles, Germination of seeds, Stimulating effect, Ultra-low concentrations of nanoparticles.

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1. INTRODUCTION

Bactericidal and stimulating effects were observed after treatment of plant seeds with dispersed solutions of metal nanoparticles, at that, nanofactor acts as microelements for prolonged mineral nutrition improving productivity of plants [1-4]. Metal nanoparticles are far less toxic than salt solutions of said metals being effective when used in microdoses due to the great specific surface area [2-4]. Therefore it is relevant to use lower limits of these microdoses or microconcentrations [3, 4].

2. METHODS AND MATERIALS

Copper nanoparticles (NPCu) of 2-3 nm and silver nanoparticles (NPAg) of 35-40 nm were obtained chemically [6, 7]. To avoid fast oxidation NPCu were covered with polypyrrole film. It ensures prolonged action. Objects of research: seeds of 1) grain crops - winter triticale (Ingen-93 variety), winter wheat (H335 variety) and spring wheat (Arnautka-7 variety); 2) vegetable crop-tomato (Mikhaela variety). Seeds were soaked in water dispersed solutions of NPCu and NPAg. Then in order to assess bactericidal effect of nanofactor, grain seeds were inoculated with *Helminthosporium avenae*, the fungoid pathogen that causes root rots in sprouts [8]. Seeds were germinated in Petri dishes at 25 °C (300 seeds per embodiment). The following was recorded: seed germination energy (SGE), seed germinating ability (SGA), number of right sprouts (NRS, they have their first leaf turned clockwise [4]), sprout length (SL) and rootlet length (RL) of 6-day sprouts. Root Mean Square Error of object response to treatment did not exceed 3%. Triticale seeds activated by nanosilver were sown in the field plot in autumn 2012.

3. EXPERIMENTAL RESULTS AND DISCUSSION

3.1 Experiments with Water Dispersed Solutions of NPCu

3.1.1 Assessment of the Impact of Dispersed Solutions of NPCu at Different Concentrations

According to Fig. 1, substantial stimulation – by 12.3 % – as compared to the control was revealed with

regard to SGA of Arnautka-7 seeds after seed soaking with 6-hour exposition in the variant with ultra-low concentration of  $10^{-17}$  mg/l (corresponding values were  $70.0 \pm 1.90\%$  and  $57.7 \pm 4.12\%$ ). Next concentration of  $10^{-19}$  mg/l showed the result at the level of control ( $57.6 \pm 5.53\%$ ).

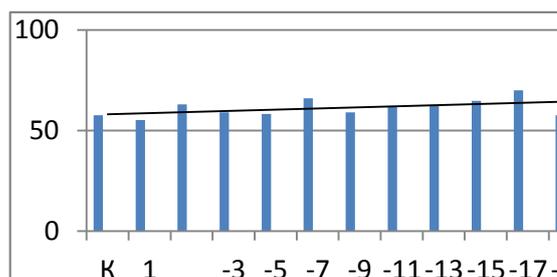


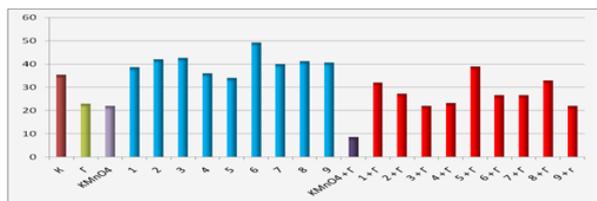
Fig. 1 – Seed germinating ability of Arnautka-7 wheat after treatment of seeds with dispersed solutions of NPCu at different concentrations. K – control, 1...-19 –  $10^{-1}$ ... $10^{-19}$  mg/l

3.1.2 Assessment of Bactericidal Effect of Dispersed Solutions of NPCu on Grain Seeds

General stimulation was revealed with regard to SGE after treatment of seeds with nanofactor.

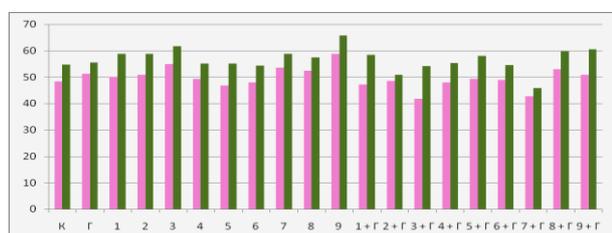
The highest effect was obtained with dispersed solution of NPCu at concentration of  $32 \cdot 10^{-8}$  mg/l (increase by 39.7%). Fungus and  $KMnO_4$  resulted in dramatic inhibition of SGE (by 51.5%). Combined influence of nanofactor and fungus had stimulating effect as compared to “Fungus” variant at almost all concentrations, especially at  $16 \cdot 10^{-7}$  mg/l (increase by 67.4%). Combined influence of  $KMnO_4$  and fungus resulted in dramatic (several-fold) decrease of SGE as compared to the variants of combined influence of nanofactor and fungus (Fig. 2).

In this experiment stimulation with regard to RL was revealed in the most variants (Fig. 3). The highest stimulating effect was obtained at concentration of  $2.56 \cdot 10^{-13}$  mg/l (increase by 20.3%). The same variant with concentration of  $2.56 \cdot 10^{-13}$  mg/l was also noted for SL (increase by 21.6%). Combined influence of nanofactor at this concentration and fungus also had stimulating effect as compared to sole fungus effect



**Fig. 2** – Seed germination energy of triticale seeds (Ingen-93 variety) after separate and combined treatment of seeds with dispersed solutions of nanocopper, potassium permanganate and fungus, %. K – control; Γ – fungus;  $KMnO_4$  – potassium permanganate; 1-9 – concentrations of NPCu solutions, mg/l:  $1 \cdot 10^{-3}$ ;  $2 \cdot 10^{-4}$ ;  $4 \cdot 10^{-5}$ ;  $8 \cdot 10^{-6}$ ;  $16 \cdot 10^{-7}$ ;  $32 \cdot 10^{-8}$ ;  $64 \cdot 10^{-9}$ ;  $128 \cdot 10^{-10}$ ;  $256 \cdot 10^{-11}$

(increase by 9.3 %). Therefore the highest positive effect on growth and bactericidal activity of sprouts was obtained at the lowest concentration of nanofactor ( $256 \cdot 10^{-11}$  mg/l) used in the experiment.



**Fig. 3** – Triticale rootlet and sprout length (Ingen-93 variety) after separate and combined treatment of seeds with solutions of copper nanopowder, potassium permanganate and fungus, %. K – control; Γ – fungus;  $KMnO_4$  – potassium permanganate; 1-3 – concentrations of NPCu, mg/l:  $1 \cdot 10^{-3}$ ;  $2 \cdot 10^{-4}$ ;  $4 \cdot 10^{-5}$ ; 8- $10^{-6}$ ,  $16 \cdot 10^{-7}$ ,  $32 \cdot 10^{-8}$ ;  $64 \cdot 10^{-9}$ ;  $128 \cdot 10^{-10}$ ;  $256 \cdot 10^{-11}$ . Bar 1 – sprout length, Bar 2 – rootlet length

Stimulating effect of dispersed solution of NPCu depends on object genotype (it was manifested only in Arnautka-7 wheat) and on concentration of nanofactor solution (Fig. 4). Increase in genotype resistance to fungal disease under the influence of nanofactor also depends on genotype (in triticale resistance increases at NPCu concentration of  $256 \cdot 10^{-11}$  mg/l, in H335 wheat and Arnautka-7 wheat – at  $128 \cdot 10^{-10}$  mg/l).

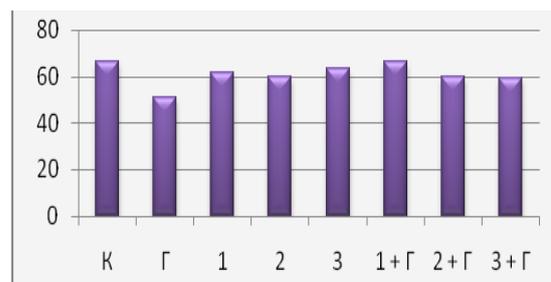
### 3.2 Experiments with Water Dispersed Solutions of NPAg

#### 3.2.1 Assessment of the Impact of Concentrations and Expositions of Treatment

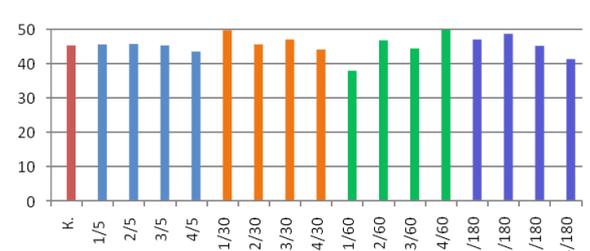
As it is shown in Fig. 5, RL substantially increases as compared to the control at concentrations of dispersed solutions of  $4 \cdot 10^{-5}$ ,  $16 \cdot 10^{-7}$ ,  $2 \cdot 10^{-4}$  mg/l after expositions of 3 h, 1 h and 30 min, respectively. These variants exceeded the control by 7.6; 10.0 and 9.8 %, respectively. In terms of technological effectiveness and profitability, NPAg concentration of  $16 \cdot 10^{-7}$  mg/l at 1 hour exposition may be considered the best variant.

#### 3.2.2. Assessment of Bactericidal Effect of NPAg on Seeds of Grain and Vegetable Crops

Influence of object genotype on bioeffect was revealed (Fig. 6). Stimulation was revealed with regard to SL in all variants for H335 wheat with maximum increase by



**Fig. 4** – Sprout length of winter triticale (Ingen-93 variety), winter wheat (H335), and spring wheat (Arnautka-7 wheat), respectively. K – control; Γ – fungus;  $KMnO_4$  – potassium permanganate; 1-3 – concentrations of NPCu solutions, mg/l,  $64 \cdot 10^{-9}$ ,  $128 \cdot 10^{-10}$ ,  $256 \cdot 10^{-11}$

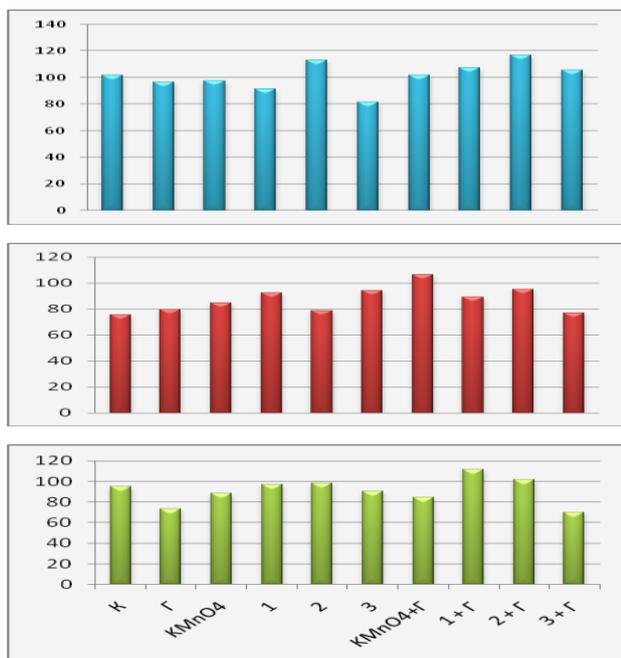


**Fig. 5** – Rootlet length of tomato sprout (Mikhaela variety) after treatment of seeds with nanosilver solutions at different concentrations and different expositions, mm. K – control; fraction – in numerator: 1-4 – concentrations of nanosilver solutions, mg/l,  $2 \cdot 10^{-4}$ ,  $4 \cdot 10^{-5}$ ,  $8 \cdot 10^{-6}$ ,  $16 \cdot 10^{-7}$ , respectively; in denominator: 5, 30, 60, 180 – expositions of treatment, respectively, min.

18.5 % at concentration of  $32 \cdot 10^{-8}$  mol/g (Fig. 6). Combined influence of factors (NPAg + Fungus) led to the increase by 15 % and 19 % in triticale and H335 wheat, respectively, at concentration  $16 \cdot 10^{-7}$  mol/g and by 7.0 % in Arnautka-7 wheat at concentration  $8 \cdot 10^{-6}$  mol/l. It is indicative of bactericidal effect of NPAg.

#### 3.2.3. Assessment of Plant Productivity (2012-2013 Field Experiment)

Triticale seeds were treated with dispersed NPAg solution at concentration of  $16 \cdot 10^{-7}$  mol/l and 1 hour exposition. Substantial stimulation by 20.1 % was obtained in laboratory experiment with regard to NRS



**Fig. 6** – Sprout length of triticale (Ingen-93 variety), winter wheat (H335 variety), and spring wheat (Arnautka-7 variety), respectively, after separate and combined treatment of seeds with dispersed solutions of nanosilver (1 hour exposition), potassium permanganate and fungus, mm. K – control; Γ – fungus;  $\text{KMnO}_4$  – potassium permanganate; 1-3 – concentrations of nanosilver solutions, mol/l,  $8 \cdot 10^{-6}$ ,  $16 \cdot 10^{-7}$ ,  $32 \cdot 10^{-8}$ , respectively

(Table). Since NRS correlates with growth activity of grain crops [6], selected mode was used in field experiment. It was observed that yielding of grain was 1.56 times higher than that of control. As it follows from the table, the effect was determined mainly by

## REFERENCES

1. H.W. Yang, M.Y. Hua, H.L. Liu, C.Y. Huang, K.C. Wei, *Nanotechnol. Sci. Applicat.* **5**, 73 (2012).
2. A.P. Raykova, L.A. Panichkin, N.N. Raykova, *Materials of the International Scientific and Practical Conference "Nanotechnologies and Information Technologies are the Technologies of the XXI Century"*, 108 (Moscow: 2006).
3. G.E. Folmanis, N.V. Kovalenko, *Ultradisperse metals in agricultural production* (Moscow: 1999).
4. N.N. Glushchenko, *Abstract of the thesis of the Doctor of biological sciences* (Moscow: 1988).
5. Yu.A. Mirgorod, N.A. Borsch, V.G. Borodina, G.Yu. Yurkov *Chem. Industry* **89** No 6, 310 (2012).
6. Yu.A. Mirgorod, V.G. Borodina, *Inorg. Mat.* **49** No 10, 980 (2013).
7. *Methods of experimental mycology* (Kiev: Naukova Dumka: 1982).
8. S.N. Maslobrod, Yu.A. Mirgorod, G.A. Lupashku, N.A. Borsch, *Proc. 2nd Intern. Conf. on Nanotechnologies and Biomedical Engineering*, 455 (Chisinau: Moldova: 2013).
9. S.N. Maslobrod, Yu.A. Mirgorod, G.A. Lupashku, N.A. Borsch, *Proc. 2nd Intern. Conf. on Nanotechnologies and Biomedical Engineering*, 527 (Chisinau: Moldova: 2013).

**Table 1** – Structural elements of winter triticale (Ingen-93 variety) yield obtained in the field plot after pre-sowing treatment of seeds with water dispersed solution of NPAg

Variant	NRS, %	NS, %	MSL, cm	NPS, pcs	WTS, g	YP, g	I, %
Control	$41.5 \pm 3.7$	$65.9 \pm 1.2$	$95.6 \pm 1.03$	$2.05 \pm 0.21$	51.5	1551	100
Experiment	$60.2 \pm 2.7^{**}$	$75.4 \pm 2.0^{**}$	$96,3 \pm 1,23$	$2.21 \pm 0.20$	50.5	2426*	156.4

Note: NRS – number of right sprouts in laboratory experiment, NS – number of sprouts, MSL – main stem length, NPS – number of productive stems from one nest, WTS – weight of thousand seeds, YP – yield from plot of 4 m<sup>2</sup>, I – increase as compared to control

higher number of survived experimental plants and higher number of productive stems of plant grown from one seed.

## 4. CONCLUSIONS

1. For the first time it was shown that water dispersed solutions of nanocopper and nanosilver at ultra-low concentrations of NPCu (up to  $10^{-17}$  mg/l) and NPAg (up to  $10^{-11}$  mol/l) have stimulating effects on seed germination and sprout growth of some grain and vegetable crops;

2. Bactericidal effect of nanofactor was revealed. At that, there occurs almost full recovery of growth activity in experimental variants treated with fungal pathogen. Conventional method of seeds disinfection with potassium permanganate was less effective.

3. Substantial increase of plant yield was obtained in the field environment as a result of pre-sowing treatment of triticale seeds with water dispersed solution of nanosilver at concentration showed to be stimulating in laboratory setting.