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RESPONSES OF WHEAT AND CUCUMBER PLANTS ON THE TREATMENT WITH NEWLY DEVELOPED HUMIC SUBSTANCES CONTAINING FERTILIZER

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ABSTRACT

Humates are substances possessing high physiological activity, contributing to the photosynthetic yield in plants and having no carcinogenicity or mutagenicity. It is the combination of these unique properties that makes humates promising plant growth and development stimulators.

Within the framework of the present investigation the efficiency of application of two humate products has been assessed, and a comparative analysis of their influence on the growth and development of agricultural crops has been carried out. Commercial peat based products EDAGUM®SM and HUMAT K have been chosen as the subjects of research. To study their influence on the growth and development of plants, laboratory-induced germination, germination, plant survival and morphometric parameters were determined in the course of the experiments. To evaluate the photosynthetic rate, total plant leaf chlorophyll (a and b) content has been determined.

EDAGUM®SM has shown greater positive influence on the survival of both plants, compared to HUMAT K. The use of neither of the preparations influenced the germination, morphometric parameters or biomass of the plants. EDAGUM®SM influenced positively on total chlorophyll content in wheat leaves.

Keywords: humates, laboratory-induced germination of seeds, germination, survival, morphometric parameters, total chlorophyll

INTRODUCTION

Conservation and rehabilitation of soil fertility is one of major problems of modern agriculture. Fertilizers play an important role in increasing crop yield and agricultural product quality, in particular natural fertilizers, as exceptionally low concentrations of them can stimulate the growth and development of plants, and increase their resistance to stress conditions [1]. Humates belong to the group of natural high-molecular compounds possessing high physiological activity due to the peculiarities of their structure and physical and chemical properties. Absence of toxicity, carcinogenicity, or mutagenicity in humates paves the way to the output of ecologically clean products [2]. According to the literature data, one of the mechanisms of humic substance influence on plants is the stimulation of all the biological processes in the plant not only at the initial

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stages of seed germination and root system formation, but at the later stages of plant growth and development, as well [1,2,3].

The creation of humates is based on their ability to form water soluble salts of humic acids with sodium, potassium, and ammonium. The most common humic acid raw materials in the production of humates are peat and coal (sodium humate, potassium humate). Other sources of humic acids are organic wastes (lignohumates are obtained from pulp and paper industry wastes, and sapropels – from bottom settings). The main drawbacks of all the types of humates are the nonstoichiometry of their composition, the irregularity of their structure, and the heterogeneity of their structural elements [2]. When used as plant growth regulators, it is recommended to apply humates at all the stages of growth: presowing treatment is aimed to increase laboratory induced germination and germination; foliar spraying is aimed to increase crop yield and improves the resistance of plants to a wide range of unfavorable conditions. Besides, humates increase the biological activity of soils contributing to the growth of consumption of organic and mineral substrates [1,2,3]. The amounts of humates used depend both on the initial raw material and on the plant growth stage. The recommended concentrations may vary from 0.001 to 0.5% and are usually indicated in the product instructions. In some studies field tests of humates were carried out by the authors with the background application of different fertilizers: NPK fertilizers and NP fertilizers [3,4]. The present work is aimed to study the use of peat based humic substances produced by different manufacturers in the minimum recommended concentration in soil poor in organic matter without the use of background fertilizers.

Thus, the objective of the present work was to reveal the potential of using EDAGUM®SM and HUMAT K humic substances as plant growth and development regulators in a laboratory experiment in wheat and cucumber plants.

METHODS AND MATERIALS

Winter wheat *Hiems triticum*, variety Scepter, and cucumber *Cucumis sativus*, variety Balcony F1, have been used as test plants. Commercial products EDAGUM®SM (preparation A) and HUMAT K (preparation B) have been used as humic substances.

To evaluate the influence of the preparations on the growth of plants most fully sandy soil was modeled: soil (55°73'87"N 49°27'96"E) and river sand were mixed at the ratio 1:2. The soil was placed into 4 kg containers. The main soil parameters are presented in Table 1.

Table 1. Soil parameters

Sand (%)	Clay (%)	Silt (%)	TOC mg*g-1	Cmic mg*kg-1
10	2.5	81	20	448

Then, according to the instruction, presowing treatment of seeds was carried out by means of soaking for 15h in 0.005% aqueous solutions, after which the seeds were sown into the soil. Then, on days 1, 7, 14, 21, 28, 42, 56, and 70, plant-root fertilization with 0.015% aqueous solutions was realized. In the control (with no humates) the seeds were soaked in distilled water for 15h, sown into the soil, and no fertilization was carried out. The experiment was realized in a greenhouse with the light regime of 16:8h at the temperature of 22^{0}C, and the duration of the experiment of 70 days. Variants of the test are presented in Table 2.

Laboratory-induced germination (IG) was determined according to the Russian State standard GOST 10968-88. Grain. Methods for determination of germinating energy and germinating property. Germination was determined according to the Russian State standard GOST 12038-84. Agricultural seeds. Methods for determining the germination on the 7th day of the experiment. Plant survival was registered over the whole time of the experiment on days 14, 21, 28, 42, 56, and 70. On day 70 morphometric parameters and the biomass of the plants under investigation were determined: average stem length, average root length, and average biomass of plants. Besides, on the 70th day of the experiment total chlorophyll a and b (Cha+b) content was determined with the help of photometric method using spectrophotometer LEKI SS1104; 96% ethanol solution was used as the extractant [5].

Table 2. Variants of the test aimed to estimate the influence of humates on the plants

Sample	Application of product A	Application of product B	Wheat seed sowing	Cucumber seed sowing
WhA	-	-	+	-
CuA	-	-	-	+
WhB	+	-	+	-
CuB	+	-	-	+
WhC	-	+	+	-
CuC	-	+	-	+

The measurement of all the parameters was carried out at least in triplicate. Average values and standard deviations are presented in the Tables. The reliability of the difference in average values was estimated according to Student's criterion ($P<0.05$). Statistical processing was carried out in Microsoft Excel program.

Laboratory-induced germination (IG) of seeds characterizes their ability to produce good and even sprouts in the field, meaning good uniformity and plant survival [2, 3]. Laboratory-induced germination is an important parameter used to calculate seed application rates. Thus, when IG is low, seed application rates should be increased, which correspondingly leads to the decrease of the product cost effectiveness. As the use of humates supposes presowing seed soaking, we've made an assumption that it can influence this value. After presowing seed soaking in the solutions of humates for 15 h laboratory-induced germination was determined during 3 days.

So, according to our observations (Table 3), laboratory-induced germination of wheat and cucumber control samples is lower than that of samples treated with the preparations. IG of cucumber seeds for both products was 100%, which is 11% higher than control. IG of wheat seeds treated with EDAGUM®SM was 90%, and IG wheat seeds treated with was 92%, which is 12% and 14% higher than the control value, correspondingly.

Investigations of the influence of humates on laboratory-induced germination carried out by other authors show ambiguous results. Thus, in the studies of brown coal based humates by Borisenko et al (2015) the results were relatively low: after the treatment of cucumber seeds, Palchik variety, laboratory-induced germination after 3 days was

62.8%. However, the work by Sheudzhen et al describes positive results received after the use of different types of humates concerning laboratory-induced germination of rice seeds [2,3].

Table 3. Influence of humates on the laboratory-induced germination of seeds in test plants – winter wheat (*Hiems triticum*) and cucumber (*Cucumis sativus*)

Sample	Laboratory-induced germination, %
WhA	78.0±6
WhB	90.0±7
WhC	92.0±5
CuA	88.9±8
CuB	100.0±0.2
CuC	100.0±0.2

In the course of the following experiment the influence of the products under study on the growth and development of test plants was determined. Germination, survival, and morphometric parameters of test plants were evaluated. Field germination is the number of sprouts emerged, expressed as a percentage of the number of viable seeds sown into the soil. During the experiment germination was determined on the 7th day after sowing. Presowing seed soaking was carried out according to the product instructions for 15 h, control sample was soaked in distilled water (Table 4).

Table 4. Wheat and cucumber plant germination under the influence of humic preparations

Sample	WhA	WhB	WhC	CuA	CuB	CuC
Germination, %	70.00±7.14	70.00±7.14	73.33±7.87	80.00±12.5	70.00±7.14	80.00±12.5

The treatment of seeds and the introduction of preparations into soil on the 1st day of the experiment did not have any significant influence on the germination determined in laboratory conditions. Besides, the germination of wheat was low in all the samples. According to GOST R 52325-2005 (Seeds of agricultural plants. Varietal and sowing characteristics. General specifications), standard germination of reproduction seeds aimed to be used in the growth of commercial products is 87%. The best germination value determined for Class 1 wheat is 95%, and for Class 2 wheat is 90.

In cucumber plants, treatment with the preparations did not influence significantly the germination, either. Thus, the germination of **CuC** sample was the same as that of the control sample and was 80%, and the germination of **CuB** sample was 10% lower than that of the control sample and was 70%. Like in the investigations carried out by Borisenko et al (2015), the germination of cucumber seeds treated with brown coal based humates was 80%, but that value was 17.2% higher than that of the control sample.

In the course of the work plant survival was evaluated. The value characterizes the ability of seeds to develop into fully grown plants participating in the formation of the yield. Plant survival is calculated as a percentage ratio of plants at harvest time to the number of sown viable seeds [3]. Plant survival was calculated in the experiment on days 14, 21, 28, 42, 56, and 70, the data are presented in Fig. 1.

The analysis of plant survival in the control samples showed that on day 14 the number of wheat sprouts was 100%, however, by the end of the experiment, on day 70, the percentage of lodging was 14.3%, and plant survival reduced to 85.7% (Fig.1). In the control samples of cucumber plants the number of sprouts on day 14 of the experiment was 87.5%, and by the end of the experiment, on day 70, the value did not change; the percentage of lodging between days 14 and 70 was 0%.

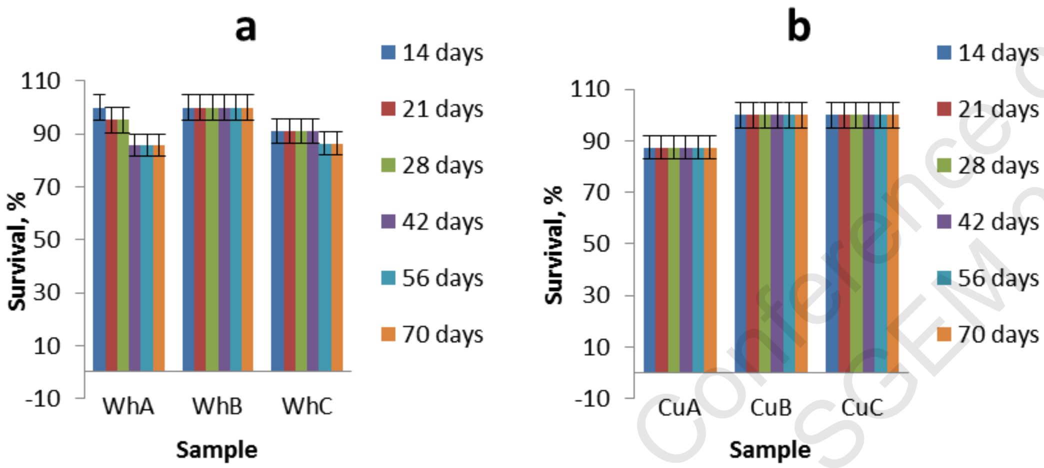

Figure 1. Survival of wheat (a) and cucumber (b) plants after the treatment with humic products A and B

Low values of germination and survival of wheat and cucumber control samples are most likely connected with high requirements of both plants to soil fertility and with the difference of their biological characteristics. Treatment with EDAGUN@SM lead to the growth of survival in both plants (up to 100%), and treatment with HUMAT K lead to the same effect in cucumber plants, but to lower positive effect (86.4%) in wheat plants.

One of the most informative features reflecting the influence of different factors is the integral parameter of plant growth. Growth is one of the most important indices reflecting the condition of a plant organism. The record of the growth parameters reflects to the fullest extent the influence of different factors on the development of plants. To determine the influence of the preparations on the growth and development of the plants their morphometric parameters were analyzed at the end of the experiment (Fig.2). The analysis of the data received revealed no significant differences from the corresponding control values for samples **WhB**, **WhC**, **CuB**, **CuC** in the stem length and the biomass. It is interesting to mention that in literature there are data both about the presence and the absence of the positive influence of humates on plants at the later stages of their development. Thus, in the work by Sheudzhen et al (2015), when the influence of presowing treatment of rice seeds and further foliar spraying of sprouts with humates was evaluated, positive influence of humates on the growth and development of plants was registered: those plants demonstrated more intensive growth, greater indices of dry matter, and greater leaf surface values than the control [3]. Along with that, in the work by Tereshchenko et al. (2009) devoted to the investigation of the influence of peat oxihumate on the biomass of plants no significant positive effect was determined after the use of 0.075% solution of the preparation [4].

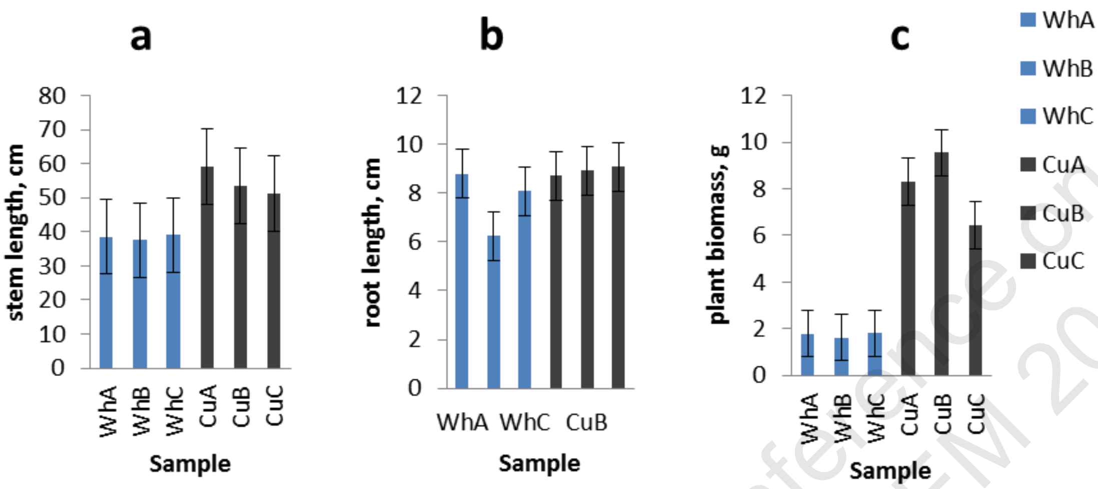

Figure 2. Morphometric parameters (a – stem length, b – root length) and biomass (c) of wheat and cucumber plants after the treatment with humic preparations A and B on day 70 of the experiment

The photosynthesis of green plants is the basis of all life on Earth, it involves the transformation of light energy into chemical energy. An important index of plant physiology is the content of the basic photosynthetic pigment, chlorophyll [6]. The light regime is one of the governing factors in the process of the synthesis of pigments in plant leaves, however, in case of equal light regime and humidity conditions it is possible to determine the influence of environmental factors on the photosynthesis [7]. In the present work the influence of introducing humates into soil on total chlorophyll content (Cha+b) in the leaves of the plants under investigation was evaluated on day 70 of the experiment with the following equal environmental conditions for all the samples: light regime (16 h of light + 8 h of darkness), soil humidity (60% of the maximum water holding capacity). The data are presented in Table 5.

Table 5. Total chlorophyll content in wheat and cucumber plants after the treatment with humic preparations A and B

Sample	Cha+b, mg/g of wet weight
WhA	4.30±5.21
WhB	6.55±6.15
WhC	3.47±3.45
CuA	2.01±6.36
CuB	1.18±7.85
CuC	0.98±6.28

The comparison of Cha+b content did not reveal significant differences from the corresponding controls in all the samples treated with the preparations under study, except for sample **WhB**. In the investigations carried out by other authors, Sheudzhen et al (2015) determined greater leaf surfaces and chlorophyll content in rice plants treated with humates, and Skudra et al (2017) described the increase of chlorophyll concentration (content) after the introduction of different fertilizers, including nitrogen fertilizers, into soil during 3 years, and the increase was 102 to 120% [8].

CONCLUSION

In the course of the investigation the efficiency of the use of two humate products as plant growth regulators in a 70 days laboratory experiment in winter wheat and cucumber plants has been assessed. Positive influence of the application of both products, EDAGUM®SM and HUMAT K, on the laboratory-induced germination in both plants has been registered.

Both products under investigation have influenced the plants positively at the first stages of their growth, and EDAGUM®SM has proved to be more effective. Thus, when EDAGUM®SM was used, sprout survival increased up to 100%, and laboratory-induced germination increased up to 90% and 100% compared to control wheat and cucumber plants, correspondingly. The application of HUMAT K helped to increase laboratory-induced germination and survival in cucumber plants up to 100% and laboratory-induced germination and survival in wheat plants up to 92%.

The degree of influence of the products on the plants at the later stages of their development was lower. Thus, no significant difference from reference values of germination and morphometric parameters has been found for both plant species and both products. However, weak positive influence of EDAGUM®SM on total chlorophyll (a and b) content in wheat leaves was detected.

In whole, the application of both preparations has had greater influence at the initial stages of the plant growth, and EDAGUM®SM has proved to be more effective. However, compared to the published data on the positive influence of humates, the products under investigation can be characterized as not effective enough.

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