The Influence of Cu and Co Nanoparticles on Growth Characteristics and Biochemical Structure of *Mentha Longifolia in Vitro*

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Abstract The aim of the given work was to investigate the influence of copper and cobalt nanoparticles on clonal microreproduction of *Mentha longifolia* plants received by method of isolated cells and tissues culture. Copper and cobalt nanoparticles were used in the form of colloidal solutions of high viscosity (gels). As object of research were *Mentha longifolia* plants. During experiments the optimum concentration of copper (0,5 mg/l) and cobalt (0,8 mg/l) nanoparticles colloidal solutions have been defined. These solutions were added in Murasige and Skug nutrient medium and have led to increase of microplant height and growth index on 45-48,4 %, the quantity of internodes on 29,4-33,9 % and quantity of shoots on 55,6-66,2 %, reproduction coefficient on 30-40 %.

Keywords Simultaneous Electron Beam Evaporation and Deposition, Co Cu Nanosized Powders, Zeta Potential, Particle Size, Colloidal Solution, Clonal Microreproduction, *Mentha Longifolia* Plants, In Vitro, Chromatography, Cultivation, Growth Index

enzymatic systems by direct participation in structure of enzymes molecules or by their activation. Enzymes, which contain copper, influence on carbohydrate and nitric metabolism. Thanks to the influence on phenolic nature growth inhibitors, copper raises plants stability to lodging and to other stress factors. Cobalt in plants can be found in the ionic form and as a part of vitamin B₁₂. It activates glycolysis enzyme phosphoglucomutase and enzyme arginase, which hydrolyzes arginine, participates in biosynthesis of leghemoglobin, and also in oxidising processes, activates enolase and kinase enzymes in the process of pyruvic acid transformation.

It is known from references that metals nanoparticles influence on seeds sprouting and plants development in an open ground [1, 2], and also on tylosis tissues [3, 4], but we did not find any accessible information about nanometals influence on growth and biochemical indices at medicinal essential oil plants microreproduction.

Thereupon, the investigation of copper and cobalt nanoparticles influence on *Mentha longifolia* explants growth characteristics depending on nanoparticles concentration at clonal reproduction (cultivation) *in vitro* and changes of basic biochemical indices of essential oils is actual.

1. Introduction

Despite the big achievements in biotechnology of plants, there are certain difficulties in definition of necessary components and concentration of macro-, microelements and growth regulators in nutrient medium for various kinds and grades of plants, purposely to receive the greater quantities of plants-regenerants.

It is known, that microelements activate definite

2. Method to Produce the Object of Research

As the object of research *Mentha longifolia* plants from the collection of Research station of medicinal plants of Agroecology and Nature Management Institute have been used. 100 explants with Cu and 100 explants with Co were

used for investigation, and all experiment was repeated three times.

Explants sterilization and all further work were spent in aseptic conditions according to standard techniques [5, 6].

Copper and cobalt nanoparticles were received by joint sedimentation from a steam phase of metal and alkaline metal halogenide (NaCl) [7, 8]. Stable colloidal solutions of high viscosity (gels) simultaneously with two and more amides were received further on the basis of nanoparticles data [9, 10]. The properties of received colloidal solutions were investigated by means of device Malvern Zetasizer Nano S (Malvern Instruments Ltd., the Great Britain) Ver. 6.20 in which particles size is defined on their moving with use of Einstein-Stoks relation.

Distribution of particles on sizes in colloidal solutions is resulted on fig. 1 (a – for copper, b – for cobalt). Distribution is monomial with a mode, which lays in the range of sizes from 0,5 to 2 nm.

Figure 1. Distribution of copper (a) and cobalt (b) particles on sizes in colloidal solutions on the basis of amines

Given colloidal solutions concentrations for addition into nutrient medium were 0,4 mg/l, 0,8 mg/l, 1,2 mg/l and 0,5 mg/l, 1,0 mg/l, 1,5 mg/l.

Sterile explants were placed on modified Murasige and Skug (MS) nutrient medium [11] (pH 5,6-5,8) which contains 0,75 mg/l of 6-benzylaminopurine, 0,1 mg/l of adenine, 0,5 mg/l of gibberellin acid and separately copper and cobalt nanoparticles in concentration specified above. The control variant of explants was placed on MS nutrient medium with the same structure, except nanometals.

Explants were cultivated in cultural room at temperature 23-25°C, air relative humidity 70 % and illumination 2500-3000 luxes with the photoperiod 16 hours.

The growth indices analysis was spent on 28th day. Shoots height, growth index, internode quantity, shoots quantity, reproduction coefficient were measured and estimated for this purpose.

In 28-30 days plants-regenerants were grafted and placed on MS nutrient medium for rooting: half concentration of macro- and microsalts, 2 % sucrose, indolil-3-acetic acid and indolebutyric acid in concentration 0,5 mg/l.

In 30 days *Mentha longifolia* plants with well developed root system were taken out from flasks, washed from agar rests and bedded out in sterile substratum: peat: sand: vermiculite in ratio 2:1:1. In 40-45 days strike root plants were bedded out in open ground conditions and covered with an agricultural fibre for 10-12 days.

Dry raw materials were used for essential oil receipt: they were prepared in mass flowering period and distilled by Ginsberg's method (distillation with water steam) without cohobation [12-15].

Biochemical screens were spent by chromatography-mass spectrometry method on chromatograph AgilentTechnology 6890N with mass spectrometer detector 5973N at National institute of grapes and wine «Magarach» of the Ukrainian academy of agricultural sciences according to standard technique [16-19].

The library base of NIST05 and WILEY 2007 mass spectrometers with total quantity of spectrums more than 470000 by means of the program for identification AMDIS and NIST was used for components identification [14, 20, 21].

Mathematical analysis of researches results was spent with use of mathematical statistics methods by means of program Microsoft Office Excel 2007.

3. Results and Discussion

Growth processes initiation in the form of leaves first pair unfolding was observed in 4th-5th day. In the process of research it has been defined, that explants development on 28th day was more intensive on nutrient medium with copper in concentration 0,5 mg/l and on nutrient medium with cobalt in concentration 0,8 mg/l. This was confirmed by growth indices, which are performed in tab. 1.

Table 1. Growth indices of *Mentha longifolia* on 28th day

Shoot height, sm	Growth index	Shoots quantity, pcs.	Internode quantity, pcs.	Reproduction coefficient
Control				
4,6±0,48	11,5	5,48±1,28	10,26±1,47	1:10
MS with Cu (0,5 mg/l)				
6,83±0,74***	17,1	8,53±1,12***	13,74±1,86***	1:14
48,5%		55,6%	33,9%	
MS with Co (0,8 mg/l)				
6,67±0,50***	16,7	9,11±1,76***	13,28±1,68***	1:13
45%		66,2%	29,4%	

Note: *** (at $p \leq 0,001$) – significant difference between the group and control group

According to results in tab. 1, it is possible to draw a conclusion that copper and cobalt nanoparticles positively influence on growth indices. So, *Mentha longifolia* shoot height on 28th day in control samples was 4,6±0,48 sm, on nutrient medium with copper – 6,83±0,74 sm and has increased on 48,4 %, and shoot height on nutrient medium with cobalt was 6,67±0,50 sm, and was on 45 % bigger than in control samples. Shoots average quantity from one explant on 28th day in control samples was 5,48±1,28 pcs. while on nutrient medium with copper – 8,53±1,12 pcs. and has increased on 55,6 %, and shoot height on nutrient medium with cobalt was 9,11±1,76 pcs. and that was on 66,2 % bigger than in control samples (Fig. 2). Internode average quantity at control explants was 10,26±1,47 pcs., at explants on nutrient medium with copper nanoparticles – 13,74 pcs. and was on 33,9 % bigger than in control, and on nutrient medium with cobalt – 13,28 pcs., on 29,4 % bigger than in control. Also such indices as growth index, which at control plants was 11,5 and at experimental plants on nutrient medium with copper nanoparticles – 17,1, on nutrient medium with cobalt nanoparticles – 16,7, and reproduction coefficient, which was 1:10 on 28th day at control plants, 1:14 at experimental plants on nutrient medium with copper nanoparticles and 1:13 on nutrient medium with cobalt nanoparticles, have accordingly increased.

Figure 2. *Mentha longifolia* on 18th day of cultivation with cobalt (0,8 mg/l)

Figure 3. Plant-regenerant is ready to be planted in substratum

On 28th-30th day plants regenerants were grafted on MS nutrient medium with half concentration of macro- and microsalts for rooting.

The first signs of roots formation appeared on 8th-10th days, and in 30 days microplants with well-developed root system were bedded in substratum (Fig. 3). Strike root regenerates to substratum, which consisted of peat: sand: vermiculite in ratio 2:1:1. was 96 %.

In July, 2015 at the same time the raw materials have been gathered, from which was isolated essential oil (tab. 2).

Table 2. Characteristics of essential oil

Investigated samples	Essential oil output, %	Linalool content in essential oil, %	Linalyl acetate content in essential oil, %	Linalool and linalyl acetate total content, g/100 g of raw material
контроль	1,164	85,97	6,057	1,071
MS c Cu	2,226	91,33	4,689	2,137
MS c Co	2,190	93,00	0,169	2,04

Essential oil was analyzed on chromatograph Agilent Technology 6890N with the mass spectrometer detector 5973N. Results of research are shown in tab. 3-5.

Table 3. Component composition of *Mentha longifolia* essential oil (control plants)

№	Substance output time, min	Substance content, %	Substance
1	6,626	0,060	β -myrcene
2	6,829	0,033	isobutyl isovalerate
3	7,555	0,356	1,8-cineole
4	7,609	0,043	limonene
5	7,875	0,102	trans-ocimene
6	8,191	0,079	cis-ocimene
7	8,755	0,376	trans-linalool oxide
8	9,206	0,151	cis-linalool oxide
9	9,323	0,020	α -terpinolene
10	9,535	0,206	2,2,6- trimethyl-3-keto-6- vinyltetrahydropyran
11	9,905	85,970	linalool
12	10,049	0,137	oct-1-en-3-ol acetate
13	10,460	0,466	3-octylacetate
14	11,019	0,109	menthone
15	11,163	0,022	iso-menthone
16	11,392	0,027	menthofuran
17	12,187	0,041	menthol
18	12,377	0,912	p-menth-1-en-8-ol
19	13,842	0,013	pulegone
20	13,960	0,043	hexyl isobutyrate
21	14,036	0,093	piperitone
22	14,623	6,057	linalyl acetate
23	15,728	0,078	dihydroedulan
24	17,951	0,257	neryl acetate
25	18,560	0,378	geranyl acetate
26	18,795	0,060	β -bourbonen
27	19,598	0,076	α -gurjunen
28	19,796	0,744	trans-caryophyllen
29	20,608	0,264	α -cubebene
30	20,793	0,037	gumelen
31	21,059	0,443	β -farnesene
32	21,591	0,911	germacren D
33	22,047	0,071	bicyclegernacren
34	22,281	0,054	germacren A
35	22,561	0,036	γ -cadinene
36	22,660	0,080	cis-calamenene
37	22,863	0,054	δ -cadinene
38	23,273	0,055	α -muurolen
39	24,243	0,196	caryophyllen oxide
40	24,654	0,142	ledol
41	25,317	0,355	α -kubenol
42	25,980	0,104	tau-cadinol
43	26,273	0,288	α -cadinol

As follows from received data, there are 43 individual components in control plants essential oil, 22 of them are presented in content more than 0,1 % and 21 of them are presented in content less than 0,1 %. Linalool (85,97 %) and linalyl acetate (6,057 %) have the greatest content at control plants essential oil.

Table 4. Component composition of *Mentha longifolia* essential oil (on MS with Cu 0,5 mg/l)

№	Substance output time, min	Substance content, %	Substance
1	6,626	0,266	β -myrcene
2	6,901	0,016	isobutyl isovalerate
3	7,564	0,221	1,8-cineole
4	7,618	0,058	limonene
5	7,880	0,218	trans-ocimene
6	8,196	0,078	cis-ocimene
7	8,457	0,015	γ -terpinene
8	8,647	0,037	trans-sabinene hydrate
9	8,764	0,180	trans-linalool oxide
10	9,337	0,031	α -terpinolene
11	9,896	91,335	linalool
12	10,464	0,274	3-octylacetate
13	11,019	0,314	menthone
14	11,299	0,067	iso-menthone
15	11,461	0,031	menthofuran
16	11,975	0,030	terpinene-4-ol
17	12,169	0,062	menthol
18	12,377	0,781	p-menth-1-en-8-ol
19	13,215	0,051	nolil acetate
20	13,960	0,036	hexyl isobutyrate
21	14,627	4,689	linalyl acetate
22	15,042	0,211	geranial
23	15,737	0,074	menthyl acetate
24	17,956	0,221	neryl acetate
25	18,542	0,419	geranyl acetate
26	19,805	0,088	trans-caryophyllen
27	21,077	0,049	β -farnesene
28	21,604	0,084	germacren D
29	22,060	0,042	bicyclegernacren
30	24,252	0,021	caryophyllen oxide

At the same time in *Mentha longifolia* essential oil on MS with Cu 0,5 mg/l there are 30 individual components, 12 of them are presented in content more than 0,1 % and 18 of them are presented in content less than 0,1 %. Linalool (91,335 %) and linalyl acetate (4,689 %) have the greatest content at control plants essential oil.

Table 5. Component composition of *Mentha longifolia* essential oil (on MS with Co 0,8 mg/l)

№	Substance output time, min	Substance content, %	Substance
1	1,430	0,155	ethanol
2	6,134	0,046	sabinene
3	6,216	0,022	β -pinene
4	6,626	1,040	β -myrcene
5	6,897	0,100	octane-3-ol
6	7,551	1,568	1,8-cineole
7	7,605	0,116	limonene
8	7,875	0,107	trans-ocimene
9	8,187	0,023	cis-ocimene
10	8,444	0,026	γ -terpinene
11	8,624	0,850	trans-sabinene hydrate
12	8,759	0,217	trans-linalool oxide
13	9,201	0,254	cis-linalool oxide
14	9,332	0,034	α -terpinolene
15	9,909	93,003	linalool
16	10,469	0,043	3-octylacetate
17	11,023	0,041	menthone
18	11,163	0,031	iso-menthone
19	11,569	0,057	terpinil hydrate
20	11,966	0,139	terpinene-4-ol
21	12,386	0,262	p-menth-1-en-8-ol
22	13,838	0,124	neral
23	13,955	0,080	hexenyl isovalerate
24	14,618	0,169	linalyl acetate
25	14,862	0,169	geranial
26	15,209	0,059	citronellol
27	15,732	0,041	dihydroedulan
28	16,043	0,065	geranyl formate
29	17,965	0,054	neryl acetate
30	18,572	0,086	geranyl acetate
31	19,796	0,370	trans-caryophyllen
32	20,621	0,021	α -cubebene
33	20,793	0,033	gumelen
34	21,059	0,058	β -farnesene
35	21,586	0,292	germacren D
36	22,042	0,092	bicyclegernacren
37	22,281	0,020	germacren A
38	22,858	0,016	δ -cadinene
39	24,257	0,118	α -kubenol

Also in *Mentha longifolia* essential oil on MS with Co 0,8 mg/l there are 39 individual components, 18 of them are presented in content more than 0,1 % and 21 of them are presented in content less than 0,1 %. Linalool (93,003 %) and linalyl acetate (1,568 %) have the greatest content at control plants essential oil.

On the basis of the aforesaid, it is possible to draw a conclusion that copper and cobalt nanoparticles positively influence on *Mentha longifolia* development in vitro, however results depend on copper and cobalt nanoparticles concentration. Copper and cobalt nanoparticles use in nutrient mediums at clonal reproduction increases plant

height and growth index on 45-48,5 %, internode quantity on 29,4-33,9 % and shoots quantity on 55,6-66,2 %, reproduction coefficient on 30-40 %.

At chromatographic research of the essential oil, received from blossoming plants, distinction in number of the basic components (linalool and linalyl acetate) between control plants and the samples, which has grown up on nutrient mediums with addition of copper and cobalt nanoparticles, is observed. It is possible to draw a conclusion that changes in a metabolism, occurring under copper and cobalt nanoparticles action remains for the considerable period of growth and in some months continues to influence on formation of essential oil components.

In our further researches we are planning to investigate influence of various silicon and selenium nanoparticles concentrations in nutrient medium on clonal microreproduction and rhizogenesis process *in vitro* of medicinal essential oil plants of family *Lamiaceae*. Also we are planning to compare our received results with the results of other authors, who had investigated the influence of another nanoparticles, for example iron oxides nanoparticles, on plants. It is especially interesting, because there are some completed researches, which, in some way, reveal this question. There are studies, which show detailed evidence of impact of Fe₂O₃ magnetite nanoparticles (MNPs) on Chinese mung bean plants by measuring the physiological parameters such as germination, root activity, activity of catalase (CAT), peroxidase (POD), and superoxide dismutase (SOD), content of chlorophyll, soluble protein, and content of malondialdehyde (MDA) [22]; uptake and intracellular trafficking of superparamagnetic iron oxide nanoparticles (spions) in plants [23]; effects of nano-iron oxide particles on agronomic traits of Soybean [24]; iron oxide and titanium dioxide nanoparticle effects on plant performance and root associated microbes [25]; assessment of concentrations of nano and bulk iron oxide particles on early growth of wheat (*Triticum aestivum* L.) [26]; effects of superparamagnetic iron oxide nanoparticles on photosynthesis and growth of the aquatic plant *Lemna gibba* [27]; nanoparticles and their impact on plants [28]; developmental phytotoxicity of metal oxide nanoparticles to *Arabidopsis thaliana* [29]; nanoparticle penetration and transport in living pumpkin plants: *in situ* subcellular identification [30]; biosynthesis of stable iron oxide nanoparticles in aqueous extracts of *Hordeum vulgare* and *Rumex acetosa* plants [31]. These studies show the contribute of iron and another nanoparticles in different aspects of plant life, so, we have to make such detailed description of copper and cobalt nanoparticles contribution on family *Lamiaceae* essential oil plants life cycle, because they are absent for today.

processing of seeds by copper nanoparticles. Bulletin RGATU, No. 2 (18), pp. 104-106;

REFERENCES

- [1] Polishchuk S.D., Nazarova A.A., Kutsir M.V. (2013) Productivities and biochemical structure of sunflower at

- [2] Polishchuk S.D., Nazarova A.A., Stepanova I.A., Kutsir M.V., Churilov D.G. (2014) Biologically active preparations on a basis of metals nanodimensional particles in an agricultural production. Nanotechnology, No. 1 (37), pp. 72-81;
- [3] Bugara I.A., Maltseva O.A. (2011) *Mentha piperita* L. tylosis culture receiving and their cytological characteristic at growing on nutrient mediums with different selenium concentration, Scientific Notes of Taurida V.I. Vernadsky National University. Series: Biology, chemistry, Vol. 24 (63), No. 4, pp. 17-23;
- [4] Kopach O.V., Kuzovkova A.A., Azizbekyan S.G. (2013) Microelements nanoparticles use in biotechnology of medicinal plants: the copper nanoparticles influence on *Silybum marianum* L. cell structures, Proceedings of Byelorussian State University. Series: physiological, biochemical and molecular principles of biosystems functioning, Vol. 8, P.2, pp. 20-23;
- [5] Kushnir G.P., Sarnatskaya V.V. (2005) Plants microclonal reproduction. Scientific book, Kiev;
- [6] Bugara I.A. (2006) Induced morphogenesis and clonal microreproduction of perspective mint grades: the thesis for the candidate of biological sciences degree in specialty 03.00.20. NBS-NNTS, Yalta, 20 p.;
- [7] Ustinov A.I., Melnichenko T.V., Liapina K.V., Chaplyuk V.I. (2008) Method to produce encapsulated nanopowders and unit for its implementation. Patent of Ukraine #82448;
- [8] Ustinov A.I., Melnichenko T.V., Liapina K.V., Chaplyuk V.I. (2013) Method of producing encapsulated nanopowders and installation for its implement. US Patent #8491972B2;
- [9] Dulnev P.G., Liapina K.V., Davidova O.E., Ustinov A.I. (2014) Sposob dispergirovaniya i stabilizatsii nanochastich medii v vodnih sredah. Patent of Ukraine #91374;
- [10] Murashige T.A. (1962) Revised medium for rapid growth and bioassays with tobacco tissue culture. Physiol. Plant, Vol. 15, No. 13, pp. 473-497;
- [11] Sheludko L.O. (2004) *Mentha piperita* (selection and seed farming). Poltava, 204 p.;
- [12] Rudnik A.M. (2013) Gemma carboxylic acids of balsamic poplars. Pharmaceutical journal, No. 4, pp.34-36;
- [13] Sychoy C.S., Ilyin M.M., Davankov V.A., Sochilina K.O. (2004) Elucidation of retention mechanism on hypercrosslinked polystyrene use as column packing material for highperformance liquid chromatography. J.Chromatogr. A, No. 1, pp. 17-24;
- [14] Vlasjuk P.A. (1969) Biological elements in plants vital activity, Publishing house "Scientific thought", Kiev, 526 p.;
- [15] Kutsir M.V. (2014) Definition of nanomaterials ecological safety on the basis of morphophysiological and biochemical indices of agricultural crops: dissertation for the candidate of biological sciences degree in specialty 03.02.08. Ryazan, 133 p.;
- [16] Churilov G.I. (2009) Iron, copper, cobalt nanopowders influence in the system ground-plant, Herald of OSU, No. 12 (106), pp. 148-151;

- [17] Ampleeva L.E., Stepanova I.A., Nazarov A.A. (2009) Nanocrystalline metals influence on accumulation of biologically active compounds in plants, Herald of RGATU of P.A.Kostycheva, No. 2, pp. 34-36;
- [18] Nazarova A.A., Kutckir M.V. (2012) Peculiarities of nanodimensional copper particles action on physiological processes in vegetative organism, Collected theses of XXIV conference "Modern chemical physics", Tuapse, pp. 93-94;
- [19] Nazarova A.A., Polischuk S.D. (2011) Prospects of metals nanopowders use in plant growing, Works of the All-Russia council of young scientists and experts of agrarian educational and scientific institutions "Actual problems of development of agrarian and industrial complex in scientific researches of young scientists", Moscow, pp. 52-57;
- [20] Fedorenko V.F. (2008) Nanotechnology and nanomaterials in agriculture. Scientific publishing house, Moscow, "Rosinformagrotech", 148 p.;
- [21] Churilov G.I., Nazarova A.A., Ampleeva L.E., Polischuk S.D., Tcherkasov O.V. (2010) Biological action of nanodimensional metals on various groups of plants, monography, Ryazan, 153 p.
- [22] Hong-Xuan R., Ling L., Chong L., Shi-Ying H., Jin H., Jun-Li L., Yu Z., Xing-Jiu H., Ning G. (2011) Physiological investigation of magnetic iron oxide nanoparticles towards Chinese mung bean, Journal of Biomedical Nanotechnology, Vol. 7, pp. 677-684;
- [23] Ahmadov I.S., Ramazanov M.A., Sienkiewicz A., Forro L. (2014) Uptake and intracellular trafficking of superparamagnetic iron oxide nanoparticles (spions) in plants. Digest Journal of Nanomaterials and Biostructures, Vol. 9, No. 3, pp. 1149-1157;
- [24] Sheykhbaglou R., Sedghi M., Tajbakhsh Shishevan M., Seyed Sharifi R. (2010) Effects of nano-iron oxide particles on agronomic traits of Soybean. Not Sci Biol, No. 2 (2), pp. 112-113;
- [25] Burke D.J., Pietrasiak N., Situ S.F., Abenojar E.C., Porche M., Kraj P., Lakliang Y., Samia A.C.S. (2015) Iron oxide and titanium dioxide nanoparticle effects on plant performance and root associated microbes. Int. J. Mol. Sci., No. 16, pp. 23630-23650;
- [26] Feizi H., Moghaddam P.R., Shahtahmassebi N., Fotovat A. (2013) Assessment of concentrations of nano and bulk iron oxide particles on early growth of wheat (*Triticum aestivum* L.). Annual Review & Research in Biology, No. 3(4), pp. 752-761;
- [27] Barhoumi L., Oukarroum A., Taher L.B., Smiri L.S., Abdelmelek H., Dewez D. (2015) Effects of superparamagnetic iron oxide nanoparticles on photosynthesis and growth of the aquatic plant *Lemna gibba*. Arch. Environ. Contam. Toxicol., No. 68(3), pp. 510-20;
- [28] Bhattacharyya A., Mohammad F., Naika H.R., Epid S.T.T., Reddy J., Prakasham R.S. (2015) Nanoparticles and their impact on plants. Research Journal of Nanoscience and Nanotechnology, No. 5, pp. 27-43;
- [29] Lee C.W., Mahendra S., Zodrow K., Li D., Tsai Y.-C., Braam J., Alvarez P.J.J. (2010) Developmental phytotoxicity of metal oxide nanoparticles to *Arabidopsis thaliana*. Environmental Toxicology and Chemistry, Vol. 29, Issue 3, pp. 669-675;
- [30] Corredor E., Testillano P.S., Coronado M.-J., González-Melendi P., Fernández-Pacheco R., Marquina C., Ibarra M.R., de la Fuente J.M., Rubiales D., Pérez-de-Luque A., Risueño M.-C. (2009) Nanoparticle penetration and transport in living pumpkin plants: *in situ* subcellular identification. BMC Plant Biology, No. 9, pp. 9-45;
- [31] Makarov V.V., Makarova S.S., Love A.J., Sinitsyna O.V., Dudnik A.O., Yaminsky I.V., Talianky M.E., Kalinina N.O. (2014) Biosynthesis of stable iron oxide nanoparticles in aqueous extracts of *Hordeum vulgare* and *Rumex acetosa* plants. Langmuir, No. 30 (20), pp. 5982-5988.