THE EFFECT OF COORDINATIVE COMPOUNDS OF COBALT WITH MONOETHANOLAMINE ON PHYSIOLOGICAL INDICATORS OF *PHASEOLUS VULGARIS*

EFECTUL COMPUŞILOR COMPLECŞI AI COBALTULUI CU MONOETANOLAMINA ASUPRA INDICATORILOR FIZIOLOGICI LA *PHASEOLUS VULGARIS*

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Abstract. *The present paper has as main objective testing the influence of some complex compounds of cobalt with monoetanolamina, obtained at the Chemistry Department of State University in the Republic of Moldova, on the seed germination capacity, biomass accumulation and biosynthesis capacity of assimilating pigments in Phaseolus vulgaris. These could eventually demonstrate their positive effects on physiological processes in plants. 16 complex compounds were tested, the results proved that a same stimulating effect on the studied indexes exists.*

Rezumat. *Lucrarea de fata a avut ca scop testarea influentei unor compusi complecsi ai cobaltului cu monoetanolamina, obtinuti la Facultatea de Chimie a Universitatii se Stat din Moldova, asupra capacitatii de germinare a semintelor, acumularii de biomasa si capacitatii de biosinteza a pigmentilor asimilatori la Phaseolus vulgaris pentru evidentierea eventualelor efecte benefice ale acestora asupra proceselor fiziologice la plante. Au fost testati 16 compusi complecsi iar rezultatele au demonstrat un efect stimulator a acestora asupra indicatorilor studiati.*

Cobalt, a transition element, is an essential component of several enzymes and co-enzymes. It has been shown to affect growth and metabolism of plants, in different degrees, depending on the concentration and status of cobalt in rhizosphere and soil. The distribution of cobalt in plants is entirely species-dependent. The uptake is controlled by different mechanisms in different species. In higher plants, absorption of Co^{2+} by roots involves active transport. Transport through the cortical cells is operated by both passive diffusion and active process. In the xylem, the metal is mainly transported by the transpirational flow. Distribution through the sieve tubes is acropetal by complexing with organic compounds. The lower mobility of Co^{2+} in plants restricts its transport to leaves from stems.

Cobalt is not found at the active site of any respiratory chain enzymes. Two sites of action of Co^{2+} are found in mitochondrial respiration since it induces different responses toward different substrates like α-keto glutarate and succinate.

Exogenously added metal causes morphological damage in plastids and changes in the chlorophyll contents. It also inhibits starch grain differentiation and alters the structure and number of chloroplasts per unit area of leaf. Co^{2+} reduces the export of photoassimilates and dark fixation of CO_2. Low concentration of Co^{2+} in medium stimulates growth from simple algae to complex higher plants. Relatively higher concentrations are toxic. A similar relationship is seen with crop yield when the metal is used in the form of fertilizer, pre-seeding, and pre-sowing chemicals. Toxic effect of cobalt on morphology includes leaf fall, inhibition of greening, discolored veins, premature leaf closure, and reduced shoot weight.

Being a component of vitamin B_{12} and cobamide coenzyme, Co^{2+} helps in the fixation of molecular nitrogen in root nodules of leguminous plants.

The beneficial effects of cobalt include retardation of senescence of leaf, increase in drought resistance in seeds, regulation of alkaloid accumulation in medicinal plants, and inhibition of ethylene biosynthesis. The aim of this paper is testing the biological effect of some coordinative compounds of cobalt with monoetanolamina, obtained at the Chemistry departament of State University in the Republic of Moldova, on *Phaseolus vulgaris* specie.

MATERIAL AND METHODS

Phaseolus vulgaris L. seeds were cultivated on filter paper dampened by 0.1 % coordinative compound solution. Control samples were cultivated on demineralisated dampened filter paper. The germination capacity was evaluated at 11 days and was determined in percentage according to the control. This one is considered to be 100%. Vegetative mass was determined at 21 day–old plants by weighing 10 random selected plants. Dry material was determined using the same material as vegetative mass by repetitive drying at 105°C for two hours until a constant weight was obtain, calculated with the formula

S.U. ((m4-m1) / (m3-m2)) x 100%

where: m1- empty capsule mass at 105 °C;m2 – empty capsule mass at room temperature; m3- capsule mass with fresh material at room temperature ,m4- capsule mass after drying at 105°C.

Assimilatory pigments were determined using a spectrophotometrical method. The vegetal samples extract was obtained by extraction with 85% acetone and filtered. The readings was made with UV-VIS spectrophotometer using E_{662}, E_{664} and $E_{440,5}$.

The quantity of assimilatory pigments was calculated by using the following formula:

chlorophyll *a* (mg) = ($E_{662} \cdot 9,78$) – ($E_{664} \cdot 0,99$);

chlorophyll *b* (mg) = ($E_{664} \cdot 21,4$) – ($E_{662} \cdot 4,65$);

carotenoids (mg) = ($E_{440,5} \cdot 4,69$) – (clorofilele *a* + *b* $\cdot$ 0,267).

The results were related to dry substance

RESULTS AND DISCUTIONS

The seed germination is a complex process starting when exogenous factors like humidity, light, temperature are favorable and endogenous dormancies factors are eliminated. In our experiments the seeds was cultivated in the climatic room at constant temperature (25°C) and dark conditions, therefore the only variable factor was humidity. The optimal hydration degree of seeds tissue is determined by the absorption capacity and water retention by tissue biocoloids and this process depends on nature of storage substances of seeds endosperm. The bean seeds have a high protein content of endosperm and require plenty of water for hydration.

In table no.1 experimental data are related. We can observe a weak diminution of germination seed capacity at almost all experimental variants. The water availability decrease cut out for osmotic pressure increasing and ionic concentration of solution. This fact can elongate the germination period. Exceptions were variants 1, 2 and 4. Nevertheless, the obtained value does not suggest a toxic or cytostatic action of studied substances bat rather a slowing of the germination process, by their action on the unleashing factors of germination. This conclusion was confirmed by the fact that, on some variants with a lower germination percent the biomass accumulation was bigger than the witness sample's.

Table no. 1.

The germination capacity of *Phaseolus vulgaris L.*seeds in presence of coordinative compounds of cobalt with monoethanolamine

Coordinative compound	Germ seed (%)	Biomass (g)	Dray subst %	Chl *a* (mg/g d.s.)	Chl *b* (mg/g d. s.)	Carotenoids (mg/g d. s.)
1,2,3-$[Co(etm)_3]\cdot 3H_2O$ (**1**)	105	38.82	7.3726	2.41	0.75	0.56
1,2,3-$[Co(etm)_3]\cdot 5H_2O$ (**2**)	103	37.25	7.1836	2.16	0.70	0.56
1,2,6-$[Co(etm)_3]\cdot 5H_2O$ (**3**)	100	34.29	7.1799	1.96	0.62	0.48
1,2,6-$[Co(etm)_3]\cdot 3H_2O$ (**4**)	102	35.87	7.2018	2.04	0.66	0.51
$[Co(Hetm)_3](HSO_4)(SO_4)\cdot H_2O$ (**5**)	94	31.68	7.7391	2.25	0.77	0.61
$[Co(Hetm)_3][Co(etm)_3](NO_3)_3$ (**6**)	98	38.54	6.4283	2.26	0.78	0.55
$[Co(Hetm)_3][Co(etm)_3](CH_3COO)_3\cdot 7H_2O$ (**7**)	95	27.39	8.2831	1.98	0.56	0.36
$\{[Co(Hetm)_3][Co(etm)_3]\}_2(SO_4)_3\cdot 8H_2O$ (**8**)	99	36.65	**8.9837**	**2.32**	0.76	**0.66**
$[Co^{II}\{Co^{III}(etm)_3\}_2]Cl_3\cdot H_3N(CH_2)_2OH\cdot 2H_2O$ (**9**)	97	35.28	5.9364	2.11	0.66	0.42
$[Co^{II}\{Co^{III}(etm)_3\}_2]Br_3\cdot H_3N(CH_2)_2OH\cdot 4H_2O$ **(10)**	98	35.22	6.8926	2.02	0.79	0.48
$[Co^{II}\{Co^{III}(etm)_3\}_2]I_2\cdot 4H_2O$ (**11**)	97	36.01	7.2162	2.13	**0.84**	0.38
$[Co(Hetm)_3]_2(SO_4)_3\cdot 4H_2O$ (**12**)	100	37.01	8.8394	2.24	0.69	0.64
$[Co(Hetm)(etm)_2]NO_3$ (**13**)	96	**41.53**	3.2116	2.29	0.72	0.54
$[Co(Hetm)_2(etm)](NO_3)_2$ (**14**)	96	40.09	3.9114	2.33	0.69	0.52
$[Co(Hetm)_3](NO_3)_3$ (**15**)	98	39.42	4.2861	2.31	0.71	0.51
$[Co(Hetm)_3]Cl_3\cdot 2H_2O$ (**18**)	96	37.28	6.1162	2.19	0.72	0.41
Control	**100**	**36.02**	**7.0127**	**2.18**	**0.72**	**0.54**

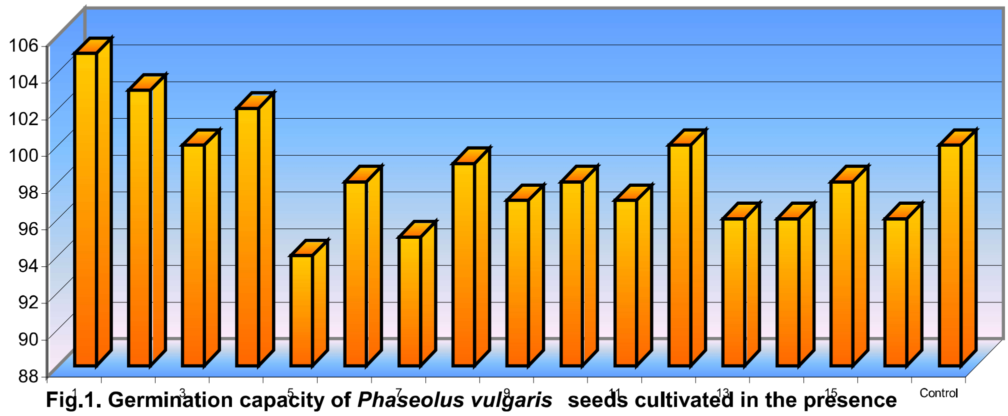

Fig.1. Germination capacity of *Phaseolus vulgaris* seeds cultivated in the presence of coordinative compounds of cobalt with monoethanolamine

The biomass accumulation is a complex process when it is determined by tissues growth (accomplished by cell division) and development processes (represented by qualitative modification and differentiation of new tissue from meristematice cells). If germination process is carried out by storage substances of endosperm, the following growing and development processes are realized based on exogenous mater and energy. Mineral elements necessary for plant nutrition after metabolisation remain in the organism for the rest oh its life. Therefore, dry mater determination of plant tissue offers the possibility to estimate the quantitative implication of mineral element in different metabolic processes.

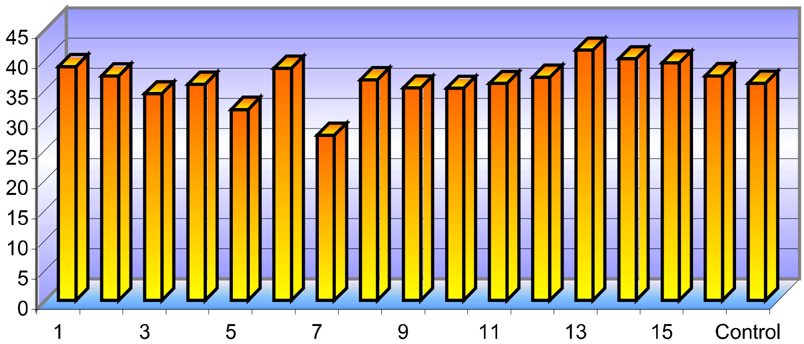

Fig. 2 The biomass variation of *Phaseolus vulgaris* plants growing in the presence of coordinative compounds of cobalt with monoethanolamine

In the case of our experiments, it was measured for ten 21-days old bean seedlings, the biomass and the dry substance content (expressed in %). Regarding the results presented in the table, it can be noted that biomass accumulation took place differently due to outer added substances. Comparing with the Control variant, in the case of variants **6**, **13**, **14**, **15**, **18**, an increased biomass accumulation could be noticed, but a decrease of dry substance. The highest biomass increase (41.53 g comparing with c 36,02 g for Control) was noticed for $[Co(Hetm)(etm)_2]NO_3$ (**13**), but, at the same time for this compound, the lowest content of dry substance- 3,2116% was noticed comparing to 7.0127% for Control. For experimental variants **1**, **2**, **8** and **12** the biomass accumulation was accompanied by the increase of dry substance content, and for variants **3**, **4**, **5**, **7**, **11**, even if biomass accumulation was lower than for Control, at the same time an increase of dry substance content in tissues was noticed. These differences can be explained by the complex structure of the used substances, by its having different functions in cellular processes. After all, it can be generally noted that a stimulating action at the level of growth and development of vegetal tissues exists. The only variants for which stimulation of any parameters were observed were **9** and **10**.

NO_3^- is the principal form of plant nitrogenous absorption. The nitrogenous need is increased during growing processes, based on cell division and which are not associated with important dry matter accumulation. In variants **6**, **13**, **14** and **15** we observed a higher value of biomass comparing with control variant which means a bigger availability of this anion in plants growing processes.

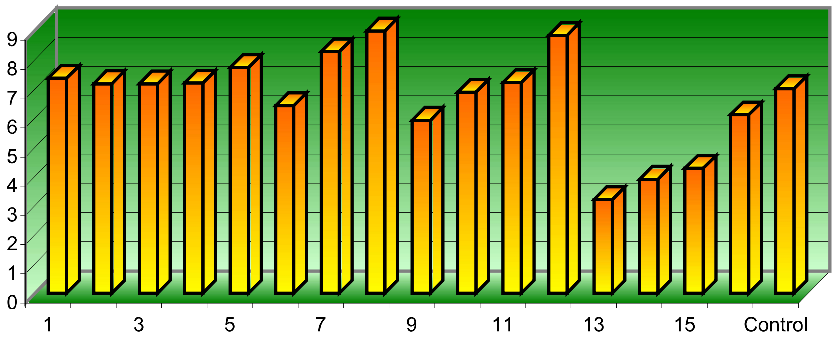

Fig. 3 The dry biomass content of *Phaseolus vulgaris* plants growing in the presence of coordinative compounds of cobalt with monoethanolamine

As much as nitrogenous, sulfur is a plastic element witch is a component part of some amino acids, proteins and vitamins. It plays an important role in cells differentiation processes and it is associated with dry matter accumulation.

In our experiments we observed a variation of assimilating pigments quantity in function of the experimental variant used.

The highest value of chlorophyll *a* was observed at compound no. **8** (2,32 mg/g s.u. compared with 2,18 mg/g d. s. in the control case), and for chlorophyll *b* – on compound no. **11** (0,84 mg/g s.u. compared with 0,72 mg/g s.u. . in the witness case). It is notable the fact that substances which have in their composition SO_4^{2-} have a stronger stimulating effect upon the assimilator pigments biosynthesis then the compounds with NO_3^-, even if in scientific literature there are many references of the role of nitrogen in the biosynthesis role of chlorophylls. The stimulating effect of treatment variants 5, 8, 12 on carotenoid biosynthesis is very important, because they have a protector role against UV radiations therefore their utilization like anti-mutagenic compounds form could be of perspective.

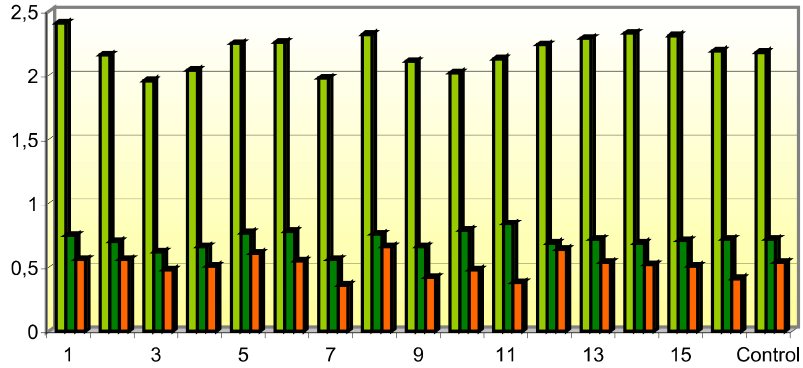

Fig. 4 The assimilatory pigments content of *Phaseolus vulgaris* plants growing in the presence of coordinative compounds of cobalt with monoethanolamine

Chl a (mg/g d.s.) Chl b (mg/g d. s. u.) Carotenoids (mg/g d. s.)

CONCLUSIONS

1. We can observe a weak diminution of germination seed capacity at almost all experimental variants. An exception were variants 1, 2 and 4. The obtained data showed a slowing of germination process, by their action on the unleashing factors of germination.
2. The majority of the studied substances contributed to the growing of biomass and accumulated dry substance.
3. The biggest increase of biomass (41.53 g according to 36,02 g for the witness sample) was registered at $[Co(Hetm)(etm)_2]NO_3$ compound (**13**). At the same time, the same compound registered the lowest content of dry substance.
4. Substances with SO_4^{2-} anion, had a more powerful stimulating effect on assimilating pigments' synthesis that compounds with NO_3^- anion.
5. $[Co(Hetm)_3](HSO_4)(SO_4)\cdot H_2O$, $\{[Co(Hetm)_3][Co(etm)_3]\}_2(SO_4)_3\cdot 8H_2O$, $[Co(Hetm)_3]_2(SO_4)_3 \cdot 4H_2O$ contributed to an increase of the carotenoids content, thus a better protection effect against UV radiations.

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