# ACCUMULATION OF HEAVY METALS BY DIFFERENT PLANT SPECIES IN CONDITIONS OF COPPER EXCESS

## ACUMULAREA METALELOR GRELE DE CĂTRE DIFERITE SPECII DE PLANTE, ÎN CONDIȚII DE EXCES DE CUPRU

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***Abstract.*** *Trace element content has been determined in the plants of Vitis vinifera, clover, alfalfa, mixture of cereal grass crops, growing under Cu excess. It has been shown that Cu excess leads to its accumulation in the roots, which inhibits plant growth and some metabolic processes, retards the transport of trace elements to the above-ground organs of plants, and reduces Fe content in plants. A higher Cu content was accumulated in the plants of clover and mixture of cereal grasses. These crops can be used for phytoextraction of Cu excess. The study was conducted by the financial support of FEN of Moldova, grant #316 and RFFR 08-04-90111.*

**Key words:** trace elements, Cu excess, plants

***Rezumat.*** *A fost determinat conținutul de microelemente în organele plantelor de vița de vie, trifoi, lucernă şi amestec de ierburi cerealiere, în condiții de exces de Cu. S-a demonstrat că excesul de Cu în sol contribuie la acumularea lui în cantități superoptimale în organele plantelor, ceea ce influențează negativ asupra creşterii şi altor procese metabolice, la micşorarea conținutului formelor accesibile ale unor microelemente în plante. Cu reduce transportul Fe către organele aeriene, contribuie la apariția simptomelor de cloroză .Cultivarea plantelor acumulatoare de Cu (trifoi, graminee furajere) după defrişarea plantațiilor multianuale, poate constitui unul din elementele de perspectivă ale tehnologiei de fitoextragere a acestui element din sol. Cercetările au fost efectuate cu ajutorul financiar din Fondul Ecologic Național, grant 316 şi FCFFR 08-04-90111.*

**Cuvinte cheie:** microelemente, exces de Cu, organele plantelor

## INTRODUCTION

Anthropogenic environmental pollution is one of the main current problems. Heavy metal pollution of soil presents a great hazard. Accumulation of heavy metals in the superficial soil strata leads to a dynamic disbalance in the soil-plant-atmosphere system, to multiple deviations in the nutritive system, aggravation of population's health. The literature data report that the toxicity of heavy metals due their action on root growth and mitotic activity is represented in the following descendent order: Cu>Cd>Ni>Pb>Al>Zn. Soil pollution with heavy metals in Moldova is caused mostly by the orchards and viticulture plantations in the agriculture structure. The plantations are repeatedly treated with different pesticides along with copper- and zinc-containing products during the whole agricultural year. A growing tendency towards copper (Cu) pollution has been observed in the Tohatin silvan ecosystem (1). The earlier studies have revealed soil accumulation of copper, fluorine, and lead in super optimal amounts (4, 5 etc.). Cu is especially hazardous since its accumulation in some soils of Moldova by 10-15 times exceeds its concentration in comparison with the normal soils (background). Cu is an indispensable for plants element, it is a component part of a series of enzymes. However, the interval of the positive action of the microelement is very short. In comparison with other microelements, Cu accumulated in relatively small but super optimal amounts may have a toxic effect. The total copper toxicity effect is manifested through the following: tissue injury, decrease of membrane permeability, and loss of soluble substances by roots, reoxidation of lipids and deviations in photosynthesis, production of Cu surplus containing complexes. The aim of the research was to study the influence of Cu surplus on the growth and mineral status in different plants that present an interest for monitoring soil conditions and development of efficient techniques and procedures of their improvement, prevention and avoidance of pollutant penetration in the human's alimentary chain.

## MATERIAL AND METHOD

The experimental work was conducted in laboratory and in the vegetative complex of the Institute of Genetics and Plant Physiology, Mold. Acad. Sci. Growing Cu doses (150, 300, 450, and 600 mg/kg soil, concomitant treatments of Cu-1, Cu-2, Cu-3, and Cu-4) were administered in soil in the experiment carried out in the vegetal complex, Grape (cv. Codrinschi), alfalfa (cvs. Polis and Martum), mixture of cereal grass crops, clover (cv. Polis and Avanta) were used. The content of copper and other microelements were measured in soil and plants according to an atom absorption spectrophotometry method.

## RESULTS AND DISCUSSIONS

Biomass accumulation by plants is an important index of their state in different growing conditions. The data obtained in monitored conditions report that Cu has a stimulating effect on plant growth up to a certain concentration threshold. This tendency has also been observed by L. Y. Jiang et al. (2008). Further increase of Cu concentration in soil results in reduction of biomass quantity in comparison with the control. It is noteworthy that the growth diminution is more significant at the last dose – Cu-4 for annual plants. Grape is more sensitive to this element: biomass grew by 31,5 % in comparison with the control at the first Cu dose, while at Cu-3, it decreased by 19%.

Inhibition of plant growth under the influence of growing Cu doses is associated with the changes occurring in plant metabolic processes. Evaluation of chlorophyll content in leaves showed that it drops with the Cu dose increase in the nutrient medium.

The content of Cu accessible forms in soil varies significantly and is higher under grape plants (table 1). Intensive accumulation of acid-soluble forms of microelements is observed under the plants that are occasionally treated with different pesticides and fertilizers. Evaluation of Cu content in the soil of experimental pots demonstrated that it grew linearly with the increase of Cu dose introduced in soil. The Cu content was higher (327.5 mg/kg) in the Cu-4 pots where nothing was planted (plantlets' pots).

Absorption of heavy metal ions by plants depends on their concentration in soil. Cu penetration in plants is directed by plant biological particular features: there are plants sensible to copper major concentrations in the environment, tolerant plants and plants that are accumulators of this microelement. Copper quantity in the plant above-ground part, as a rule, varies within lower limits as compared with other heavy metals. However, Cu concentration can reach major values in roots. Detection of Cu-accumulating plants is one of the promising procedures of phytoextraction of toxic element amounts.

Examination of the plants pot grown in the vegetative house has demonstrated a difference between the plants studied and plant organs in view of accumulation of Cu and other microelements (table 2). The element concentration lifted in the high Cu dose treatments in comparison with the control, in the first place, in roots. The highest Cu concentration was revealed in the roots of annual plants, a lower one in grape roots. The Cu content grows simultaneously with the increase of exogenic Cu dose in the roots of cereal grass crops from the beginning, while in Cu-4 it decreases. Alfalfa accumulated quite a large amount of Cu in the above-ground part, as well. Alfalfa has been already mentioned in literature as a Cu-accumulating plant. Our data demonstrate that the Cu concentration in alfalfa aerial organs is 130.4 mg/kg d.w. The total Cu content in roots + aerial organs made 361,42 mg/kg d.w. The roots of this type of plants, in our opinion, do not have the so called threshold of sensibility to Cu.

Evaluation of the content of other microelements in grape organs showed disorders in the plant mineral status caused by Cu surplus. The correlation between the dose of Cu incorporated in soil and Cu and Fe content in the roots of grape grafts is well pronounced. The increase of Cu dose is accompanied by a linear accumulation of this element in roots and decrease of Fe content in this organ. Decrease of Zn content was observed in sprouts, which is likely to be caused by transport inhibition by the above- ground organs. Cu had no significant influence on Mn content in graft organs. Super optimal Cu concentration in soil does not only decrease the content of the Fe accessible for plants but impedes its transport by the above-ground organs.

*Table 1*

**Content of trace elements în soil under the different plants, mg/kg soil**

| Plant selection | Cu | | Zn | | Fe | | Mn | | Ni | |
|---|---|---|---|---|---|---|---|---|---|---|
| | Acidosol ubile | bufer of acetat | Acidosol ubile | bufer of acetat | acidoso lubile | bufer of acetat | acidoso lubile | bufer of acetat | acidoso lubile | bufer of acetat |
| Stauceni, ploughed field | 2.20 | 1.80 | 2,0 | 0.16 | 55,0 | 7,19 | 65,80 | 1,20 | 2,20 | 0,70 |
| Stauceni – 1, vineyard | 68.70 | 9,90 | 5,00 | 1,70 | 158,00 | 3,40 | 114,80 | 50,50 | 3,30 | 1,50 |
| Stauceni – 2, vineyard | 69,0 | 12.00 | 4,00 | 0,50 | 170,00 | 6,6 | 110,00 | 54,10 | 3,30 | 1,56 |
| Anenii Noi – 1, vegetable garden | 17,20 | 0,70 | 6,80 | 0,17 | 215,00 | 7,8 | 169,9 | 1,20 | 5,10 | 0,80 |
| Anenii Noi – 2, vegetable garden | 13,20 | 0,50 | 7,60 | 0,07 | 255,00 | 6,7 | 147,40 | 2,10 | 5,00 | 1,60 |

*Table 2*

**Content of trace elements in plants under the different roses of $CuSO_4$ in soil, mg/kg d.w.**

| Tip of plants | Doze of Cu, introduced în soil | Plant organs | % of ashes | Cu | Fe | Mn | Zn | Ni |
|---|---|---|---|---|---|---|---|---|
| Grape | Control | roots<br>above-ground parts of seedlings | 9,43<br>7,8 | 3,65<br>2,42 | 101,75<br>18,25 | 40,54<br>54,21 | 1,71<br>1,28 | 3,21<br>1,60 |
| | Cu - 4 | roots<br>above-ground parts of seedlings | 9,55<br>7,34 | 28,32<br>2,46 | 67,23<br>10,20 | 31,03<br>49,91 | 1,30<br>1,12 | 2,01<br>1,92 |
| mixture of cereal grass crops | control | above-ground parts<br>roots | 12,48<br>26,86 | 5,61<br>34,91 | 79,75<br>381,14 | 119,80<br>286,05 | 1,74<br>4,20 | 4,24<br>5,70 |
| | Cu - 4 | above-ground parts<br>roots | 6,34<br>30,79 | 74,17<br>437,21 | 39,24<br>162,87 | 51,98<br>130,85 | 2,21<br>3,68 | 1,77<br>6,92 |
| clover (cv. Polis) | control | above-ground parts<br>roots | 9,24<br>29,64 | 10,62<br>48,15 | 24,39<br>154,85 | 62,83<br>160,25 | 3,23<br>3,14 | 2,22<br>2,12 |
| | Cu - 4 | above-ground parts + roots | 9,27 | 361,42 | 66,65 | 144,14 | 2,97 | 2,08 |

It may be caused by the competition between the two elements by phytosiderophores and/or at xylem loading. Comparison of the data on Fe content in plant organs reliably evidences about the antagonism existing between the two elements. A similar tendency is also observed in annual plants. Reduction of microelement transport from roots to aerial organs in the stress conditions provoked by Cu toxicity is also underlined by other authors (3, 6).

The Cu content was recalculated in plants and the biomass accumulated was recorded. The data confirmed that Cu extraction from soil by annual plants was much higher than that by grape plants. Evaluation of the Cu content remained in the pot soil after experiment termination and calculation of the element extracted from soil by plants permitted us to assess what part of the element introduced in soil was immobilized, in other words, transferred in the forms inaccessible for plants. The calculation showed that about 50% of the Cu introduced in soil was transferred in inaccessible forms.

The current scientific works make it possible to take a series of measures to contribute to decontamination of soil from the accumulate Cu and to avoid penetration of the toxic element into food products. Different measures are utilized in modern agriculture to detoxicate the heavy metal polluted soil: lime treatment of acid soils, phosphoric fertilizer application, soil chiseling, screening for tolerant plants etc. One of the promising procedures is phytoextraction of heavy metals from soil. The plants that accumulate elevated element amounts and produce quite a volume of biomass may be used for extraction. Presently, biological bases and technical conditions are being prepared to develop a procedure of heavy metal excess phytoextraction from soil. The data presented confirm that the problem regarding development of efficient soil decontamination after perennial plantation uprooting is urgent.

Thus, the findings reported demonstrate that Cu surplus in soil contributes to its excessive accumulation in soil and the organs of annual plants and grape, which has a negative impact on plant growth and other metabolic processes, decrease in the content of the accessible forms of some elements in soil, in the first place, Fe and Zn. Cu impedes Fe transport by above-ground organs, decreases Fe content in grape sprouts and leaves and contributes to appearance of visual chlorosis symptoms. Utilization of Cu-accumulating plants (alfalfa, cereal grass crops) after perennial plantation uprooting may serve as one of the promising elements in the technology of Cu phytoextraction from soil.

## REFERENCES


1. **Calugareanu N., A. Begu**, **2004** - *Poluarea cu metale grele a unor ecosisteme silvice din regiunea de Centru a Republicii Moldova.* Mediul Ambiant, N 2 (13), 2004, p.11-12.
2. **Jiang L. Y., Yang X. E., Chen J. M., 2008** - *Copper Tolerance and Accumulation of Elsholtzia splendens Nakai in a Pot Environment.* Journal of Plant Nutrition, Volume 31, Issue 8, pages 1382-1392

**3. Prasad M., Freitas H., 2000 -** *Removal of toxic metals from solution by leaf, stem and root phytomass of Quercus ilex. L. (holly oak).* Environ. Pollut. 100, #2: 277-283.
4. **Veliksar Sofia, Toma S., 1996** - *Ecologichesci bezopasnie metodi primenenia udobrenii na vinogradnicah i v sadah*. Chishinau, 45 p.
5. **Veliksar Sofia, Toma S., 2001 -** *Vzaimosviazi mezshdu soderzshaniem microelementov v organah vinogradnih rastenii i v pochve.* Intern. Conference „Soil, climate, crop" Chishinau, p. 12-17.
**6. Zhao Shu-Lan, Duo Li-An, 2002** - *Initial growth effect and ecological threshold of Festuca arundinaceae L. under progressive stress of* $Cu^{2+}$ *and* $Zn^{2+}$// Shengtai xuebao=Acta Ecol. Sci, 7:1098-1105.