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Studying the effects of heavy metal on chlorophyll *a*nd sugar in one year-old seedlings organs of *Acer velutinum* specie

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ABSTRACT

Pollution is one of the most important factors inhibiting growth in the environment; therefore the effects of zinc pollution were studied in *Acer velutinum* specie. One year old seedlings of *Acer velutinum* specie were prepared from nursery, the concentrations of zero and one hundred thirty mg per liter of zinc chloride solution were added to the soil of seedlings pots after calculating and after passing a three-month period of seedling growth, the plant organs were removed, then the amount of zinc concentration in the samples was determined and data were analyzed. The results of the analysis showed that the highest amount of accumulation on the stem, root and soil in treatment concentrations is 87.75, 65.68 and 24.78 mg per kg and accumulation of zinc in total chlorophyll and sugar in treatment concentrations is 4.61 and 0.6028 mg per g, respectively, and accordingly *Acer velutinum* specie is suitable for refinement of soils contaminated with zinc.

1. Introduction

Accumulation of contaminants in the food chain is a basic threatening factor for human health. In most countries with parental industries contaminants are identified to a high rate. Contaminated soils and water are also dangerous for agricultural production and human consumption. Although, the first option is definitely preventing contamination but in general, these principles are not followed in industrial areas. Zinc (Zn), which at high pH (7.2 to 7.8), the amount of absorbed Zinc by the plant depends on the available forms of Zinc in soil [1]. Roots, especially in soils which are rich of Zinc often store more amount of Zinc compared to aerial parts. The presence of more than 500 mg of Zinc per kg of plants dry matter can cause toxicity to organisms which feed from them [2]. Also, much higher than 300 mg per kg cause toxicity in plants especially sensitive species [3]. Phytoremediation is one of the methods of soils bioremediation which in recent decades has received a lot of attentions. In this method, the resistant plants are used for remediation of soils contaminated with organic and inorganic compounds. The advantages which this method has over other methods are simplicity, low cost and the possibility of utilizing in large-scale. In this method, plant selection is of great importance. Plant selection depends on climatic conditions and also the amount of pollution [4].

In most industrial areas which are in need of treatment, there are mixtures of different contaminants at different concentrations in soil, underground water or wastewater. These hazardous wastes include a variety of salts, organic matter, heavy metals, trace elements and radioactive compounds [5]. Simultaneous refinement of multiple contaminants using chemical and traditional methods is both technically difficult and costly, while these methods damage soil biotic compounds [6]. Heavy metals such as zinc and copper are necessary for normal growth of plants, however, high concentrations of essential metals also leads to prevent the plant growth and toxicity. The plants tolerance range is to a certain amount and plants can perform detoxification with particular cellular mechanisms of heavy metal [7]. Due to the systemic characteristics of photosynthetic power with a high performance, plants are able to refinement the elements during their numerous metabolic functions. Therefore, the emerging phyto-technology of phytoremediation is highly regarded in the world for refinement of soil, underground water and wastewater contaminants due to being low cost and requiring low technology [8,9]. Baycu et al., in their study in 2006 revealed that concentrations of lead, zinc, cadmium and nickel were measured in the leaf of seven species of deciduous trees (*Acer negundo L.*, *Aesculus hippocastaneum L.*, *Ailanthus altissima*, *Fraxinus angustifolia*, *Acer velutinum L*, *Populus nigra L*, *Robinia pseudoacacia L*) in urban areas of Istanbul which the highest accumulation amount of cadmium and zinc, lead, and nickel was in *Populus nigra*, *Aesculus hippocastaneum L* and *Robinia pseudoacacia L*, respectively. Lead toxicity in plant none –mind ecotype (*NME*) *Elsholtzia argyi* causes the growth inhibition, significant reduction of plant height and root length, reduction of plant wet and dry weight, discoloration of leaves and their folding [10]. The aim of this study is to evaluate absorption of zinc in seedlings organs of *Acer velutinum* spices.

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2. Materials and methods

To perform this research, *Acer velutinum* was selected among the forest species and its one years old seedlings were prepared and transferred to the greenhouse and were held there for thirty days to adapt to the new conditions. Non-contaminated soil (natural soil used in this experiment) was collected from a depth of 0–30 cm of one of the nurseries then dried and passed through a 2 mm sieve. To prepare the soil contaminated with Zinc, after preparing the soil, the suitable salt of the desired element should have been prepared. Thus, to prepare zinc, zinc chloride salts manufactured by Merk, Germany were used. In this study, the method of spraying on soils with concentrations of zero and 130 mg of zinc per liter was used and the needed amount of solution was sprayed on the soil gradually and contaminated quite uniform with the soil and then the pots with one liter volume were filled with it. The Fifty sapling in same age and same size were selected and planted in pots, the pots were kept in the greenhouse and the soil moisture was maintained at capacity limit by weighted method, and irrigation, if necessary, was done with distilled water and after a three-month period of seedlings growth, shoots and roots were harvested and washed with water distilled, then dried in an oven at 70 °C in 48 h for two times and passed through a 2-mm sieve for analysis. Zinc content in plant samples was prepared after digestion of samples by dry digestion method and atomic absorption device [11].

Chlorophyll measurements were performed using Arnon method [12]. To this end, 0.05 g of leaf tissue with 5 ml acetone was mixed at a concentration of 80%.The resulting mixture was centrifuged for 10 min at 13,000 rpm. Involves passage of the extract was separated and measured its volume to 8 ml of chlorophyll using spectrophotometer at a wavelength of 645 nm and 663 nm respectively. Method Omokolo was used for sugar extraction [13]. 0.1 g of tissue stem wet with 5 ml 80% ethanol was mixed in a porcelain mortar and then 10 min in water bath at 70 °C was placed. Then, to evaporate the alcohol extracts were obtained at 70 °C.To remove chlorophyll a ratio of 1 to 5 chloroform was added for 10 min at 10000 rpm was placed in a centrifuge. Glucose measurements were performed using Mc Creay [14]. Three milliliters Anthrone extract is added with 100 ml of boiling water for 20 min and we spectrophotometer at a wavelength of 620 nm using a sugar solution is determined.

Composite soil samples were transported to the laboratory and then dried, crushed and passed through a 2 mm sieve was. Sample digestion and release of elements method Sposito was performed [15]. Accordingly, taking into account soil moisture to 2 g soil samples 12.5 ml of nitric acid was added 4 M. Sample overnight at a temperature of 80 °C water bath in the bathroom were stored, and then the resulting solution is passed through filter paper. Concentration of the solution was determined by atomic absorption.

Data obtained from plant tests were organized in SPSS Statistics software. To analyze the data, first, in order to determine the metal accumulation in shoot and root the analysis of variance was done and to compare the effects of zinc accumulation on the shoot and root Duncan method was used.

3. Results

The following graph shows that the amount of zinc accumulation in shoots is 87.75 mg per kg in treatment concentration and 37.96 mg per kg in control concentration (Fig. 1).

The following graph shows that the amount of zinc accumulation in roots is 65.68 mg per kg in treatment concentration and 33.32 mg per kg in control concentration (Fig. 2).

The following graph shows that the amount of zinc accumulation in soil solution is 24.78 mg per kg in treatment concentration and 0.44 mg per kg in control concentration (Fig. 3). Sample digestion and release of element (Zinc) method Sposito was performed [15].

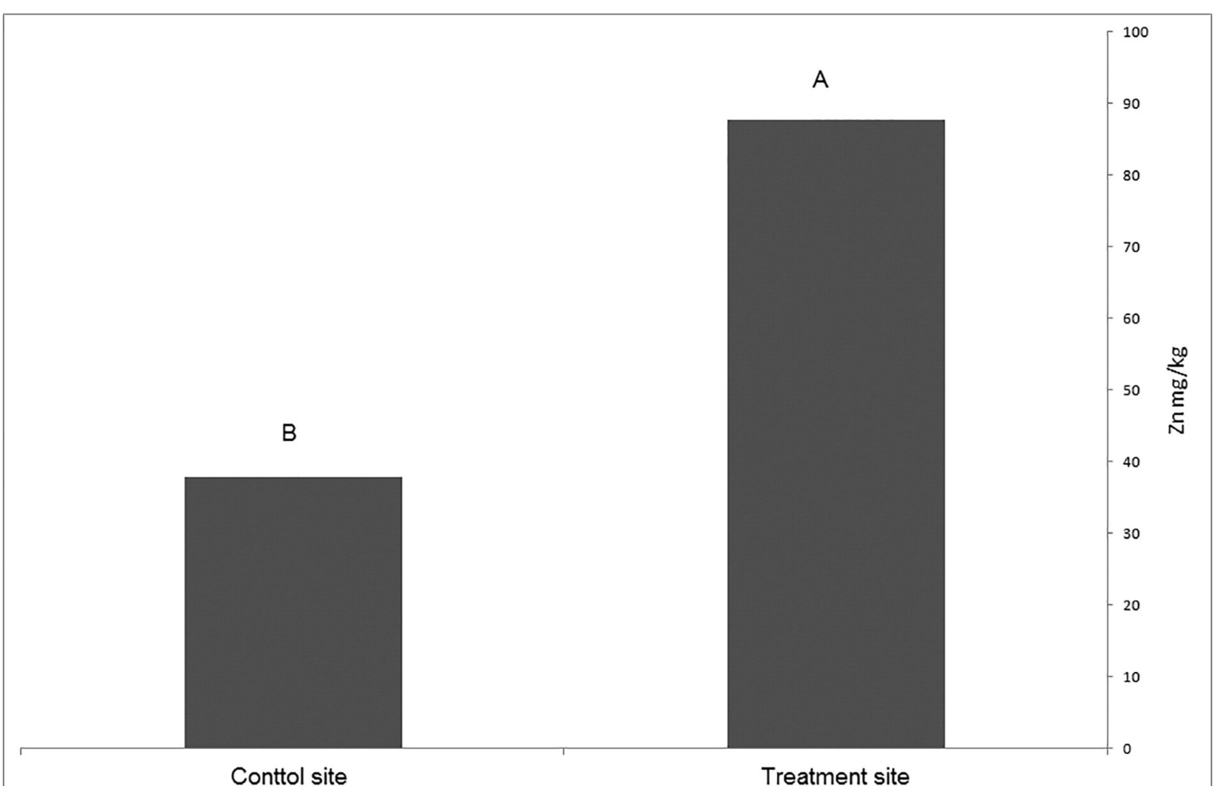

Fig. 1. Comparing the amount of zinc accumulation in the stem of *Acer velutinum* seedlings.

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