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Short Communication

Efficiency of Canola (*Brassica Napus* L.) as an Accumulator of Heavy Metals in Wastewater Applications

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Abstract

This study was carried out to determine the elimination levels of heavy metal rise resulting from wastewater in agricultural areas irrigated with wastewater, by means of the extraction of the canola (*Brassica napus* L.) plant. Therefore, the summery Licolly variety of canola (*Brassica napus* L.) plant was grown by applying wastewater at different moisture levels (Control: 20 kPa, S1: 20 kPa, S2: 35 kPa, S3: 50 kPa, S4: 65 kPa, S5: 80 kPa). Total Cu, Zn, Pb, Cd, Cr, Ni, and Hg concentrations were determined in the harvested plants. At the end of this study, accumulation of Cd, Ni, and Pb could not be determined in the plants, and while the accumulation of Cu, Zn, and Cr was statistically significant, the accumulation of Hg was found to be insignificant.

Keywords: wastewater, heavy metal, canola (*Brassica napus* L.), irrigation

Introduction

The increase in the world's population and industrialization increases the pressure on water resources. When the need of the increasing population and of the developing industry is also taken into consideration, there occurs the requirement of reusing wastewater – which is defined as water that has been contaminated or whose properties have been partially or wholly altered as a result of domestic, industrial, agricultural, and other uses in agriculture, along with the efficient use of existing water resources.

Globally, about 20 million hectares of land are irrigated with municipal wastewater [1]. Besides the direct advantages of the use of wastewater in agriculture such as conservation of clean water resources, the use of less chemical fertilizer by farmers, and the provision of arid and semi-arid areas with a reliable water resource, it has indirect advantages such as prevention of the contamination of freshwater resources and the disposal of wastewater relatively hygienically at a low cost by local governments [2]. Apart from the above-mentioned advantages, there are also risk factors that might occur upon the reuse of wastewater in agriculture. Some risk factors are effective in a short period, and the intensity of the resulting effect varies depending on the degree of contact of people, animals, or the environment with these risk factors [3]. The other risk factors, however, are effective in a longer period and have an effect that increases upon the continuous use of treated water (e.g. soil salinity and effects of toxic chemicals) [4]. In order to reduce the pressure of these possible risk factors on the environment, they should be eliminated from the environment in which they are present. In spite of the use of methods such as isolation and immobilization, mechanical separation, the pyrometallurgical method, the electrokinetic

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method, the biochemical method, phytoremediation, and leaching in the depollution of soils contaminated by heavy metals [5], the method of phytoremediation out of these methods is biologically-based, simple, and low-cost [6, 7]. In addition, it is the most preferred method since it preserves the biological and physical structures of soil [8]. Many studies have been conducted on phytoremediation [9-12]. Plants of particular ability for the uptake of elements from the air, water, and soil and their accumulation are considered to be indicative and therefore they happen to be called bioaccumulators [13]. Canola (*Brassica napus* L.) is one of the most important plants widely used as a bioaccumulator of heavy metal. Canola is a plant that can be widely used in the food industry due to its rich oil content, in feed industry owing to its rich protein content, in apiculture thanks to its being among the first flowering plants in spring, and in biodiesel production [14].

This research aimed to eliminate in a short period the heavy metal rise in soil when wastewater was used as irrigation water via a plant that might be used in animal husbandry or technology and to provide favorable conditions for heavy metal limit values for later crop production. For this purpose, research was made on the efficiency of the canola plant as an accumulator of heavy metals in soils where heavy metal contamination was formed with the irrigation water applied at different levels.

Material and Method

The experiment was conducted as pot experiments in the glass greenhouse at the Agricultural Research and Application Center at Süleyman Demirel University in 2010. Some properties of the experimental soil are as follows: sand – 30.09%, silt – 41.20%, clay – 28.71%, $CaCO_3$ – 26.44%, pH – 7.77, electrical conductivity – 0.405 dS/m, and cation exchange capacity – 28.18 me 100/g. Its heavy metal contents are as follows: Cu – 39.27 mg/kg, Zn – 32.93 mg/kg, Pb – 16.51 mg/kg, Cd – 0.01 mg/kg, Cr – 60.92 mg/kg, Ni – 110.83 mg/kg, and Hg – 0.04 mg/kg.

The soil used in the experiment was pounded when the air was dry and sieved through a 4-mm plastic sieve, and 44 kg of soil was weighed and placed into 50-kg pots. The moisture in the pots was saturated up to the field capacity, and some 16 canola seeds were planted in each pot on 12.04.2010. After the completion of germination, they were reduced so that 10 plants could remain in all pots.

The wastewater used in the experiment was obtained from the Wastewater Treatment Plant of Isparta Municipality. Its pH was 7.57, and its electrical conductivity was 0.825 dS/m. Some of its heavy metal contents were found to be as follows: Cu – 0.0097 mg/l, Zn – 0.098 mg/l, Cr – 0.014 mg/l, Ni – 0.0183 mg/l, and Hg – 0.815 mg/l. However, the elements of Pb and Cd could not be detected.

Soil moisture tension was measured by means of the tensiometers (Soilspec brand, SST102G) placed into the soil (20 cm in depth) contained in the pots. Readings of the tensiometers were performed in kPa – the unit of pressure.

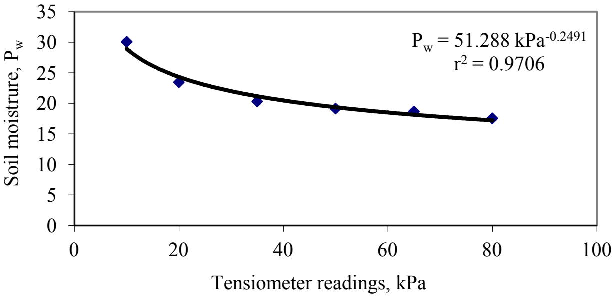

Fig. 1. The relationship between tensiometer readings and moisture content in the experimental soil.

The tensiometers were calibrated to determine the irrigation water amount applied according to the treatments. The calibration curve and equation were obtained from the relationship between the values of soil moisture (Pw) and soil moisture tension (kPa). The calibration curve and its equation are provided in Fig. 1 (P_w=51.288 $kPa^{-0.2491}$, r^2=0.9706).

Irrigation was performed at 5 different soil moisture tensions apart from the control application in order to determine the effect of wastewater application at different soil moisture levels on the heavy metal uptake by canola. When the soil moisture tension reached 20±3 kPa in the control treatment, irrigation was performed with potable water. The wastewater treatments were arranged to be irrigated when they reached 20±3 kPa (S1), 35±3 kPa (S2), 50±3 kPa (S3), 65±3 kPa (S4), and 80±3 kPa (S5). The soil moisture of each treatment was saturated up to the field capacity in every irrigation. Water collection containers were placed under the pots so as to prevent heavy metal loss through percolation, and the percolated water was reused in the next irrigation. The irrigation water amounts applied to the control, S1, S2, S3, S4, and S5 were 99.0, 145.8, 137.3, 134.9, 105.9, and 99.2 liters, respectively.

Some three plants were randomly selected from each pot on 27.07.2010, and they were harvested by cutting them about 1.5 cm above the soil surface.

Methods of Laboratory Analysis

Heavy metals (Cu, Zn, Pb, Cd, Cr, Ni, and Hg): 0.5 g of the plant specimens that had been filtered through distilled water, dried, ground in an agate mill and homogenized and 0.25 g of soil were obtained; 10 ml of HNO_3 was added to them; and the process of wet ashing was carried out in a MARS-5 (Microwave Accelerated Reaction System, CEM Inc., the U.S.A.) microwave apparatus. In these specimens, heavy metals were determined using the Perkin-Elmer 5300 DV ICP-OES apparatus.

Statistical Analysis

Minitab 16 software was used in the analyses of variance of the data obtained through relating the elemental concentration values determined for the plant specimens to yields and in the comparison of means.

Table 1. Accumulation of heavy metals in plants according to experimental treatments.

Treatments	Heavy metals (mg/pot)						
	Cu	Zn	Cd	Cr	Ni	Pb	Hg
Control	0.672 b	0.418 b[#]	Nd	0.035 c	Nd	Nd	0.005
S1	1.404 a	0.933 a	Nd	0.117 a	Nd	Nd	0.102
S2	1.127 a	0.977 a	Nd	0.061 b	Nd	Nd	0.012
S3	1.364 a	0.885 a	Nd	0.043 bc	Nd	Nd	0.020
S4	1.252 a	0.857 a	Nd	0.052 bc	Nd	Nd	0.015
S5	1.174 a	0.763 a	Nd	0.040 c	Nd	Nd	0.011
	**	*		**			Ns
LSD	0.2938	0.2788		0.02396			

**significant at the level of 0.01, *significant at the level of 0.05, Nd – not detected, Ns – insignificant, [#]The differences between the means denoted with the same letter are insignificant.

Results and Discussion

Accumulation of Heavy Metals in the Plant

The effect of the different amounts of wastewater applied depending on different kPa values on Cu, Zn, and Cr that accumulated in plants was found to be statistically significant, whereas their effect on Hg was insignificant. Despite the presence of Ni, Cd, and Pb in the soil used as growing media and of Ni in the wastewater used as irrigation water, these three elements could not be detected in the harvested plants (Table 1). The fact that the forms of Ni that were uptakable by the plant were low (particularly despite the presence of Ni in both soil and wastewater) caused it to remain below the concentration limits that were detectable in the plant. The same condition applies to Pb and Cd that were found in soil, although not in the irrigation water, but that could not be detected in plants.

Out of the elements under examination, Cu is the one that was determined at the highest amount in plants, followed by Zn and Cr. In the studies carried out with treatment sludge, it was stated that zinc and copper accumulated more within canola. Mathe-Gaspar and Anton [15], established that the concentrations of Zn, Cu, Pb, and Cd determined in the above-ground part of the rape plant cultivated in soil with Pb, Zn, Cd, and Cu concentrations being at the levels of >1,000, >3,000, >18, and >280 mg/kg, respectively were 1,145±376, 18.60±7.53, 15.23±11.82, and 8.09±2.43 mg/kg, respectively. Likewise, Solhi M. et al. [16] reported that it was Zn that accumulated the most in the aboveground part of the canola plant cultivated by applying treatment sludge and Stingu A. et al. [17] reported that *Brassica napus* was especially suitable for Cu bioaccumulation. In the study, the highest accumulation of Cu was observed in S1 (1.404 mg/pot) in the wastewater applications, while the lowest value was determined in S2 (1.127 mg/pot). On the other hand, the accumulation of Cu in the control application was 0.672 mg/pot. Even though the accumulation of Cu in the control application was statistically significant in comparison with the accumulation of Cu in the wastewater applications, the difference among the increasing wastewater applications was insignificant. In other words, although the accumulation of Cu in the aboveground parts of plants decreased depending on the decreasing wastewater applications apart from treatment S2, these decreases were statistically insignificant.

When the obtained results are examined in terms of Zn, the highest accumulation of Zn among the wastewater applications was recorded in treatment S2 (0.977 mg/pot), while the lowest accumulation was found in S5 (0.763 mg/pot). On the other hand, the accumulation of Zn in the control application was 0.418 mg/pot. The difference between the control treatment and the wastewater applications was statistically significant in the accumulation of Zn, as in the accumulation of copper. However, the decreasing wastewater application also caused a decrease in Zn that accumulated in the aboveground part of the plants; nevertheless, this decrease was statistically insignificant.

The highest Cr that accumulated in the plant was obtained in S1 (0.117 mg/pot), followed by S2 (0.061 mg/pot). On the other hand, the lowest accumulation of Cr among the wastewater treatments was detected in S5 that was in the same statistical group with the control application (0.040 mg/pot). In terms of chromium, it might be stated that the Cr uptake also increased with the increasing irrigation water amount. In a study by Budak F. et al. [18], the amounts of Cr accumulating in the stem for Brassica juncea plant and in the stem and leaves for *Brassica oleracea*, irrigated with water containing 2, 6, 13, 26, and 52 ppm of Cr, displayed some ordering in parallel with the applications.

In terms of the accumulation of Hg in plants, the difference between the control treatment and the wastewater applications and the difference in the means among the wastewater applications are not very clear. When all treatments are taken into consideration, it is seen that the lowest accumulation of Hg was in the control (0.05 mg/l), whereas the highest accumulation was in S1 (0.102 mg/pot). The fact that the highest accumulation of Cr and Hg among the wastewater applications was in treatment S1, where moisture in the soil was kept around the field capacity in the wastewater applications can be interpreted as the facilitation of the uptake of these elements under moist conditions by plants.

Efficiency of Heavy Metal Elimination by Plants

Fig. 2 shows the amount of heavy metals added to the medium with the increasing wastewater application (IWA), the mean amounts of heavy metals eliminated excessively by the plant (EP), and the IWA/EP ratios. The fact that the IWA/EP ratios remained the same for the increasing amount of wastewater applications means that the rates of increase in the amounts of heavy metals eliminated by the plants equaled the rate of increase in the amounts of heavy metals added to the medium. For Cu, the IWA/EP ratio was 1.9 in treatments S1, S2, S3, and S5, while it was 1.8 in treatment S4. The lowest ratio for Zn was determined in treatment S4 (23.6), followed by S2 (24), S1 (27.7), S5 (28.1), and S3 (38.6) in the ascending order. Regarding Cr, the IWA/EP ratios display greater variations among the applications. The lowest ratio was in S1 (24.7), followed by S2 (74.6), S4 (84.1), S3 (235.1), and S5 (290.4) in ascending order. The close values of the IWA/EP ratios for Cu and Zn according to the experimental treatments show that the efficiency of canola plant at the elimination of Cu and Zn might be independent of the amounts of Cu and Zn added by irrigation water. Nevertheless, it is seen that the amount of Cr uptaken by the plant increased with the amount of Cr that increased in soil with irrigation water.

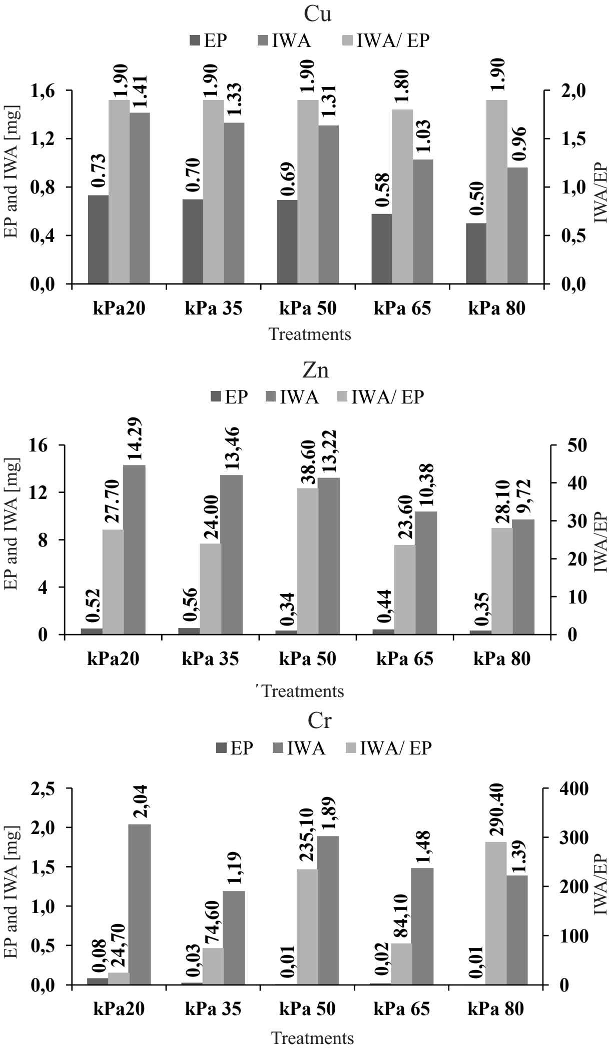

Fig. 2. Amounts of heavy metals added to the growing media with wastewater (IWA), mean amounts of heavy metals excessively eliminated by the plant (EP), and the IWA/EP ratios.

Conclusion

An increase in the amounts of Cu, Zn, Cr, and Hg that accumulated in the growing media was observed with increasing irrigation water. The heavy metal uptaken by the plant increased as compared to the control. In the plants, Cu and Zn accumulated the most. The increase in the irrigation water amounts applied to the medium had no effect on the uptake of Cu and Zn, while it affected Cr uptake.

The values of IWA/EP, an indicator of the efficiency of canola plant at the elimination of heavy metals, were the same in terms of Cu in all treatments irrigated with wastewater, whereas they varied in Zn and Cr. According to the values of IWA/EP, it was concluded that Cu was the heavy metal that canola could most efficiently eliminate from the soil in the wastewater applications.

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