REMEDIATION CAPACITY AND GROWTH RESPONSES OF *Platanus orientalis* L., *Eucalyptus camaldulensis* Dehn., and *Populus nigra* L. TO DIFFERENT LEVELS OF LEAD AND CHROMIUM IN CONTAMINATED SOIL

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ABSTRACT

This study was conducted to evaluate the effect of contaminated soil with lead and chromium using three species of trees *Platanus orientalis, Eucalyptus camaldulensis,* and *Populus nigra* as a remediation tool which considers as a priority restoring method to keep healthy soil which support life system for all organisms. The experiment was conducted with three concentrations of Pb (200,400, and 600 mg.kg^{-1}) and three levels of Cr (100, 200, and 300 mg.kg^{-1}) along with control. The results reveals that root dry weight for orientale plane and poplar tree were reduced meanwhile, eucalyptus showed significant increase at 200 and 400 mg.kg^{-1} of lead. Shoot dry weight of poplar revealed a stimulatory effect (187.26, and 193.5 g) at 200 and 400 mg.kg^{-1} of lead respectively. Chlorophyl content decreased significantly linearly with the increasing the concentration of Pb and Cr. At the same time, proline content showed the highest values ((76.53, 35.325, and 45.020 µmoles. g^{-1}) at 300 mg.kg^{-1} of chromium in oriental plane, eucalyptus, and poplar respectively. Generally, the root system accumulates more Pb and Cr than leaves in which the bioaccumulation factor was < 1 for all species similarly the translocation factor except in eucalyptus (1.135, 1.013, and 1.018) at control, 200, and 400 mg.kg^{-1} of lead respectively as well as their leaves content of lead was morethan root. In conclusion the species showed differ responses to Pb and Cr.

Keywords: Soil contamination, Pb, Cr, phytoremediation, chlorophyll, biomass, proline, bioaccumulation.
***Part of Ph..D. Dissertation of 1st author.**

القدرة العلاجية و استجابة الدلب الشرقي و اليوكاليبتوس و القوغ الاسود للنمو في مستويات مختلفة منالرصاص و الكروم في التربة الملوثة

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المستخلص

اجريت هذه الدراسة لمعرفة تأثير التربة الملوثة بالرصاص والكروم على ثلاثة أنواع من أشجار الغابات (الدلب الشرقي اليوكاليبتوس, القوغ) وإمكانية استعمالها كوسيلة علاجية أساسية للحفاظ على تربة صالحة لها القابلية على إعطاء الدعم للنظام الحيوي لجميع الكائنات الحية. استخدمت في هذه التجربة ثلاث مستويات من الرصاص (200 , 400, و 600 ملغ. كلغم $^{-1}$) وثلاث مستويات من الكروم (100, 200, و 300 ملغ . كلغم $^{-1}$) حسب تصميم القطاعات العشوائية الكاملة. بينتالنتائج أن الوزن الجاف لجذور الدلب الشرقي والقوغ انخفض بإزدياد العناصر الثقيلة بينما اليوكاليبتوس سجل ارتفاعا معنويا عندتركيزي (200 و 400 ملغم. كلغم $^{-1}$)من الرصاص. الوزن الجاف للمجموعة الخضرية لنبات القوغ اظهر زيادة بلغت الى (187.26 و 193.5 غم) عند المعاملة (200 و 400 ملغم . كلغم $^{-1}$) من الرصاص على التوالي. كذلك اوضحت النتائج ان نسبة الكلوروفيل قل معنويا في جميع المعاملات وكلما زادت تراكيز المعاملات كلما كان التأثير أكبر. علاوة الى ذلك سجل محتوى البرولين أعلى نسبة (76.53 , 35.325 , و 45.020 مايكرومول. غم $^{-1}$) عند تركيز (300 ملغم . كغم $^{-1}$) من الكروم في الدلب الشرقي واليوكاليبتوس والقوغ الاسود على التوالي. بشكل عام كانت المجموعة الجذرية أكثر كفاءة بتجميع الرصاص والكروم من الاوراق على الرغم من أن نسبة تراكم العناصر كانت < 1 في جميع المعاملات والذي توافق ايضا باستجابة مشابهة في نسبة نقل العناصر من الجذور الى الاوراق والتي كانت < 1 في جميع المعاملات ماعدا اليوكاليبتوس حيث بلغت ,(1.135 ,1.013 , 1.018) لمعاملة المقارنة (200 , و 400 ملغم. كغم $^{-1}$) من الرصاص بالتتابع.

الكلمات المفتاحية: تلوث التربة، الرصاص، الكروم، العلاج بالنباتات، كلوروفيل، الوزن الجاف، البرولين، نسبة التراكم

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INTRODUCTION

Pollutants seems to contaminate large areas of soil and ground water thereby affecting plants, animals, and humans (2). Heavy metals toxicity is among the most important abiotic stresses to plants, their toxicity depends on their physicochemical properties (41), they enters the environmental through natural and anthropogenic activities, natural elements concentration depends on the geology of the area (8), while anthropogenic cause like wastewater irrigation, soiled waste disposal, automobile exhaust, mining and smelting processes, industrial activities and agricultural activities are the main sources of contamination with heavy metals when occurs in unwanted place, form, and levels that cause negative impact on the environment(48,20,31).The critical concern of heavy metals contamination arises from being common source of contamination, low solubility in biota, and their classification for having the ability to be carcinogenic and mutagenic (13). Lead is one of the toxic heavy metals which cause an oxidative stress, photosynthesis inhibition and DNA damage (24). In spite being with no known biological function in living organisms [27] it causes a broad range of toxic effects to living organism including morphological, physiological, and biochemical changes which impairs seed germination, root elongation, seedling development, plant growth, transpiration, chlorophyll content, and cell division (23,16). Chromium is one of the top 20 contaminants on the super fund priority list of toxic substances [10]. It exists in many oxidation states however, the most stable and common forms in soil are trivalent (CrIII) and hexavalent (CrVI) species (18). An inhibition of seed germination, nutrient balance, degrade pigment status, inhibition of antioxidant enzymes, and induce oxidative stress in plants was manifest as a result of chromium toxicity (32,33). Mechanical and physiochemical based remediation have been used to reduce or treat polluted soils but these techniques are costly and deteriorate the soil (28) whereas, phytoremediation is a good alternative by using green plants to remediate contaminated soil, sediments and water hence, it is low cost and environmentally friendly technique, and it reduce the ions in contaminated soil to low levels (38). Phytoremediation had been addressed as the most economic option for remediation costing 50-80 % lower than alternative strategies (15). Plants are grouped into two categories according to their surviving mechanisms in polluted environments, pseudometallophytes use exclusion strategy which comprise the avoidance of metal uptake and restriction of its translocation to shoot (12), and mettalophytes accumulate metals and store it in vacuoles to prevent metal toxicity (6). The phytoremediation strategies that are responsible for remediation of inorganic pollutants (heavy metals) are rhizofilteration (14), phytoextraction [37], and phytostabilization (43). Trees have been shown to be effective in remediating polluted site especially when it is inconvenient to use other treatments or when there is no time pressure on the reusing of the land (40). Trees reduces the overall flow of water down the soil by continuous transpiration which help to prevent the movement of heavy metals to ground and surface water there by reducing the chances of pollution (44). Mleczek *et al* (29) mentioned that phytoremediation by using of trees has gained an increasing attention due to some properties like growing well in poor-nutrient soil, deep root system, high biomass, fast growing, low cost, and high resistance to soil in purities. The limitation for some heavy metal's accumulation by plants is the low metals bioavailability in soil (35) whereas, chelates were used to enhance metal uptake by plants such as EDTA (ethylenediaminetetraacetic acid), EGTA [ethyleneglycol -bis (ß - aminoethyl ether), N, N, N', N-tetraacetic acid], CDTA (trans -1, 2 -diaminocyclohexane -N, N, N', N'-tetraacetic acid), and EDDHA [etylenediamine-di (o-hydroxyphenylacetic acid)]) (42). EDTA has been the most widely used chelating agent in studies of phytoremediation because of its high efficiency in extracting many metals (11). The objective of this study was to seeks the best candidate trees species for remediation of contaminated soil with lead and chromium by examining three common woody trees *Platanus orientalis* L., *Eucalyptus* *camaldulensis* Dehn., and *Populus nigra* L.

MATERIALS AND METHODS

This study was carried out from April till the end of October 2019 at the field of Forestry Department / Collage of Agricultural Engineering Sciences / University of Duhok located at the district of Sumail, Duhok at 473 m above sea level; Latitude: 36°51 N; and Longitude: 42°52 E. Maximum and minimum temperature, rainfall, and humidity during study period were obtained from Sumail metrological station as shows in Table 1.. The soil was collected from the top (20 cm) of soil profile from field of Agricultural Engineering Sciences, University of Duhok. Soil and loam were air dried, sieved through a 4 mm sieve, mixed together at 3 soils: 1 loam ratio. Random samples were prepared by drying, grounding, and sieving through 2 mm sieve for soil physical and chemical analysis according to A.O.A.C, (1) methods at soil department laboratory, Collage of Agricultural Engineering Sciences, Duhok university (Table 2). Stem cuttings of one year old shoots from *Platanus Orientalis* L., *Eucalyptus camaldulensis* Dehn, and *Populus nigra* L., were collected from Duhok city with uniform length and diameter, grown in sandy soil until fine roots induction (Two weeks), all cuttings were cultured using plastic pots with 10 Kg of soil in April 2018**.**

Table 1. Weather condition for studying period (2019

Months	Rainfull (mm)	Temperature Max	Temperature Min	Humidity
January	100.1	12.25	2.845	79.09
February	52.5	14.26	3.529	78.29
March	241.9	16.1	5.755	80.45
April	149.5	14.79	8.677	75.88
May	37.7	32.94	15.11	49.76
June	0	40.6	20.58	31.15
July	0	40.75	20.68	28.71
August	0	42.45	22.64	28.34
September	0	37.06	17.45	31.95
October	30.5	31.41	14.74	46.68
November	122	22.68	6.32	49.55
December	82.4	14.99	6.013	77.86

To study the effects of heavy metals contamination on the growth of the three species, an experiment was conducted in April 2019 when seedling became two years old which include adding two heavy metals (lead and chromium) as follows:

Control treatment (which contain the naturally occurring level of heavy metal), three concentrations of Pb (200,400, and 600 $mg.kg^{-1}$) were prepared by dissolving analytical grade Pb $(NO3)^2$ in (1250 ml) deionized distill water (19). Potassium dichromate ($K_2Cr_2O_7$) was used as a source of chromium to produce an artificially pollute soil with three concentrations (100, 200, and 300 $mg.kg^{-1}$ of soil (47). To enhance the uptake and the bioavailability of both Pb and Cr ethylene diaminetetraacetic acid disodium salt EDTA (Na2EDTA) was added to all pots in amount (0.5 mg. kg^{-1} of soil) 39. The treatments were arranged in a factorial experiment within Randomized Complete Block Design (RCBD) with five replicates. Data were statistically analyzed using SAS statistical program, and the differencesbetween various treatment means were tested with Duncan Multiple Range test at 5% level. At the end of the October 2019, the seedlings were uprooted carefully, separated to root and shoot using plant pruning scissors. Plants parts were washed to remove any soil residues, air dried for two days, then placed in oven for 72 hours at 70℃ (5) to record dry weight.

Table 2. Physical and chemical characteristic of the soil

Characteristics	Value	Measurement units
PH	7.4	
EC	0.242	$ds.m^{-1}$
Organic matter	1.71	%
Moisture content	2.07	%
Sand	34.5	%
Silt	27.1	%
Clay	38.4	%
Texture	Clay loam	
Total Nitrogen	0.15	%
Sulfate ($SO4^{-2}$).	4.26	$mg.kg^{-1}$
Nitrate ($NO^{-}3$)	6.89	$mg.kg^{-1}$
Phosphate($PO4^{-3}$)	14.34	$mg.kg^{-1}$
Chromium	23.4	$mg.kg^{-1}$
Lead	40.45	$mg.kg^{-1}$

samples were grinded into a fine powder for further measurements. Chlorophylls (a, b, and total) were estimated according to Wintermans and Demots (50) methods by using of absolute ethanol (99%) (49). Spectrophotometer (Pharmacia) was used at two wave length (665nm and 649nm) for chlorophyll a and b respectively as shows in the following equations:

Chl. a = (13.7) (A 665 nm) – (5.76) (A 649 nm)

Chl. b = (25.8) (A 649 nm) – (7.6) (A 665 nm)

Total chl. = (chl. a) + (chl. b)

Determination of proline was done following the procedure mentioned by Bates *et al* (1973) [7] via spectrophotometer (Pharmacia LKB) at the wave length of 520 nm using and Proline content determined by the bellow equation;

µmoles of proline / gram tissue = [(µg proline/ml) **x** ml toluene **x** ml salicylic acid)] **/** [(115.5 µg µmole **x** sample (g)

The samples were digested according to Patel *et al* (1997)[34] using of concentrated nitric acid (HNO3) with hydrogen peroxide (H_2O_2) (3:1) (43). Digested samples of roots and leaves were used to estimate the heavy metals content using atomic absorption spectrophotometer (GBC 932 AA, AUSTRALIA) as shown in following equation;

(p H = A r × / V W

After measurements of heavy metals concentration, the following data obtained; Biological accumulation factor (BAF) BAF=HM root/HM soil Translocation factor (TF) TF=HM leaf/ HM root

RESULTS AND DISUSSION

1-Effect of Pb and Cr on plant dry weight

Root dry weight were measured to detect the changes that could be happened as a result of heavy metals effects. The result in Figure 1a illustrate that the three species varied in their response. *Platanus orientalis* L. and *populus nigra* L. were decreased in all treatments and the influence of chromium was more than that of lead with oriental plane tree and poplar through recording the highest inhibition (25.2 and 83.67 gm) respectively at 300 $mg.kg^{-1}$ of chromium. On the other hand, *Eucalyptus camaldulensis* Dehn. increased with the increment of lead concentration to 200 and 400 $mg.kg^{-1}$ but 600 $mg.kg^{-1}$ showed insignificant reduction when compared to control. Meanwhile, there was a clear inhibition at 300 mg/kg^{-1} chromium that reached (35.70 gm) (picture 1). Results in Figure 1b showed significant reduction in shoot dry weight in *Platanus orientalis* L. in all treatments compared to control and the lowest value was recorded at 200$mg.kg^{-1}$ of chromium, while eucalyptus and poplar trees showed a stimulatory effect of lead treatments unlike the chromium treatment that significantly inhibited (69.773 and 132.89 gm) respectively when treated with 300 $mg.kg^{-1}$ of chromium.

Picture 1a . Root in control treatment b- Root at 300 mg.kg^{-1} of chromium

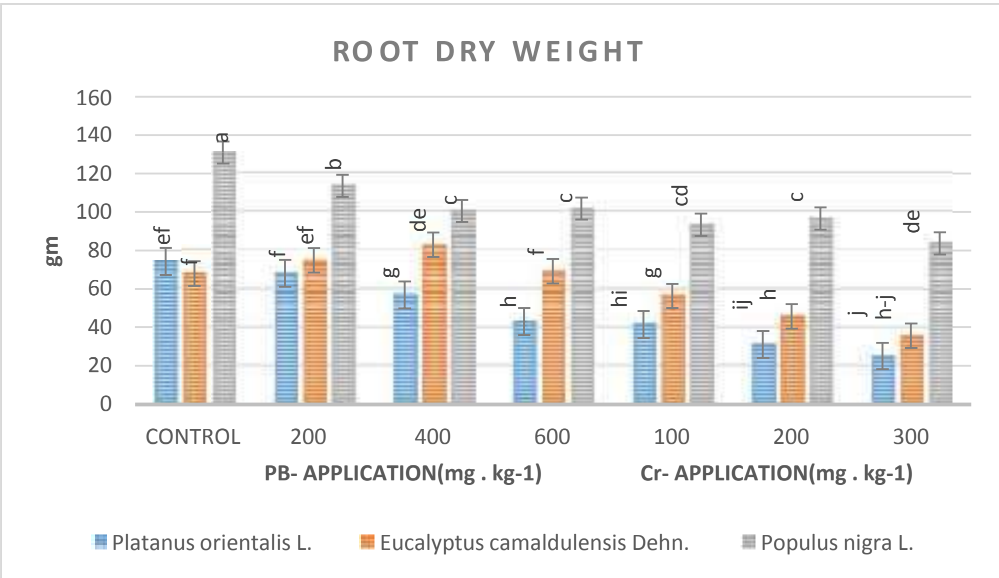

Figure 1a. Root dry weight under different Pb and Cr treatments

(Columns with the same letters are not significantly different according to Duncan's multiple range tests at 5% level)

1-Effect of Pb and Cr on chlorophyll content

Significant inhibition in the content of chlorophyll (a, b, and total) were observed in all treatments when compared with the control figure 2 (a, b, and c) and this inhibition were increased by rising the concentration of heavy metals. Chromium exhibits a higher effect on chlorophyll content than lead in all chlorophyll types that the 300 mg.kg^{-1} of chromium record the lowest values in all species (Picture 2).

Picture 2. Effect of treatments on chlorophyl content

(a) Control treatment **(b) 300 mg.kg^{-1} of chromium**

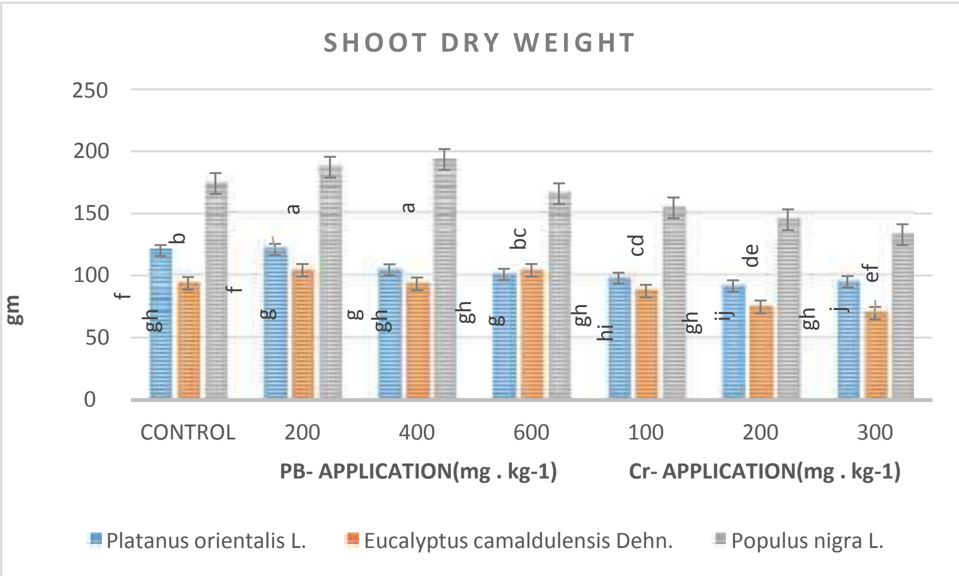

Figure (b. Shoot dry weight under different Pb and Cr treatments

(Columns with the same letters are not significantly different according to Duncan's multiple range tests at 5% level)

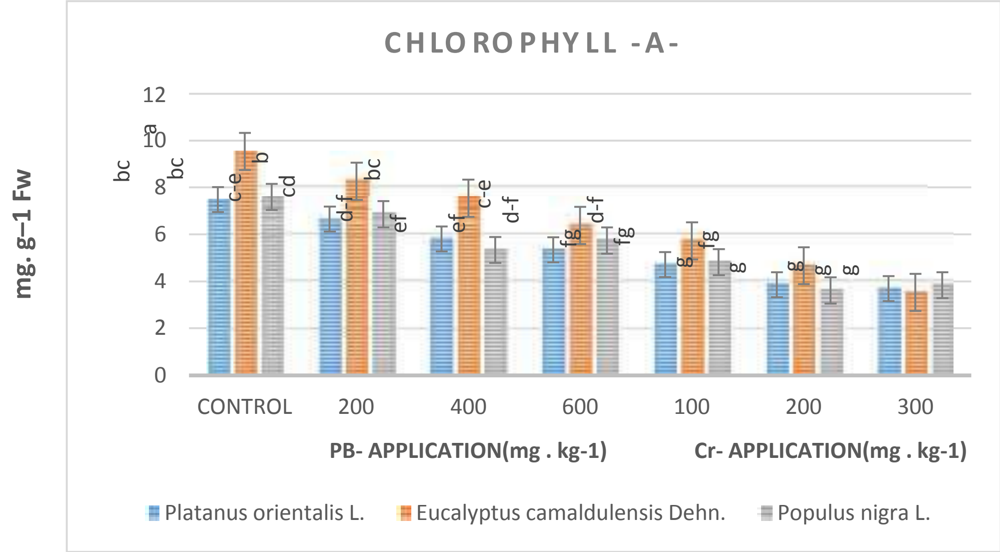

Figure 2a. Chlorophyll (a) content under different Pb and Cr treatments

(Columns with the same letters are not significantly different according to Duncan's multiple range tests at 5% level)

2-Effect of Pb and Cr on proline content

The proline content was used to assess the tolerance of tested species to both Pb and Cr stress. The proline content *Platanus orientalis*, increased significantly in comparison with control but it was not linearly correlated with the increasing concentration of heavy metals (figure 3). *Eucalyptus camaldulensis* recorded a significant increase when treated with 600, and 200 mg.kg^{-1} of lead, 300 mg.kg^{-1} of chromium respectively. Whereas, a fluctuation was shown in *Populus nigra* in proline content by recording a highest value when was treated with 300 mg.kg^{-1} of chromium.

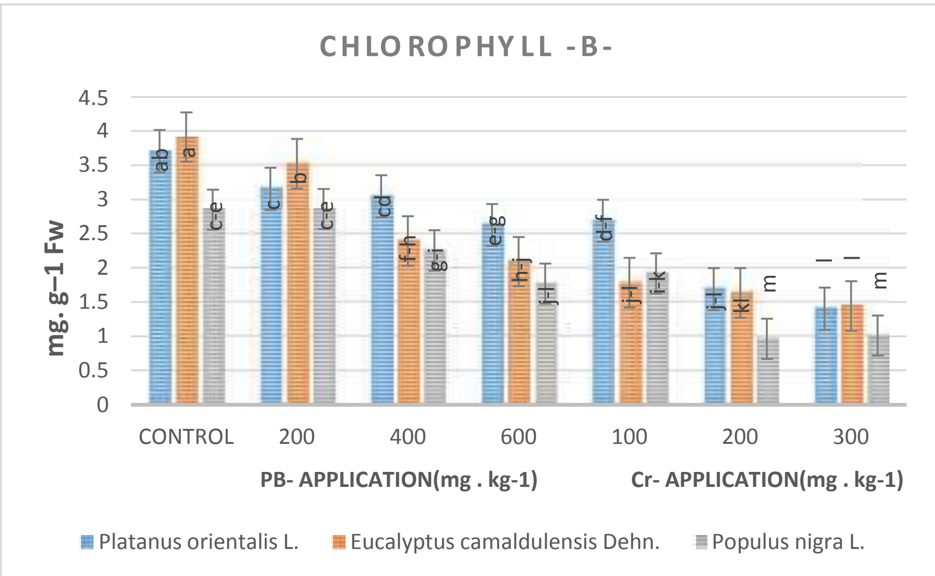

Figure 2b. Chlorophyll (b) content under different Pb and Cr treatments

(Columns with the same letters are not significantly different according to Duncan's multiple range tests at 5% level)

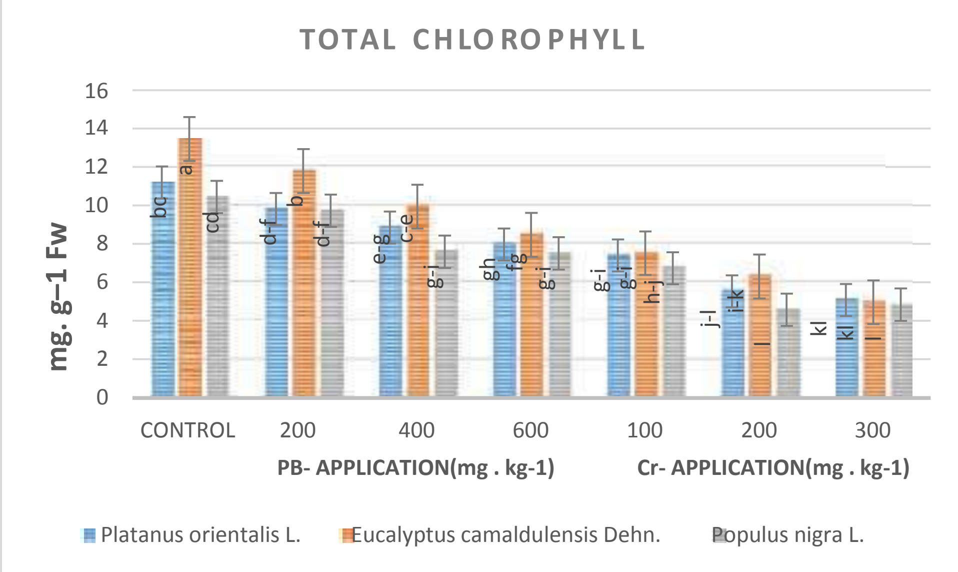

Figure 2 c. Total chlorophyll content under different Pb and Cr treatments

(Columns with the same letters are not significantly different according to Duncan's multiple range tests at 5% level)

2-Lead content in plants

Results in Figure 4 a reveal that significant increases in lead contents in root linearly correlated with the increasing concentration of lead in soil except poplar trees at 600 mg.kg^{-1} which was non significantly differences from 400 mg.kg^{-1} treatment. Meanwhile, eucalyptus showed the higher accumulation of lead in roots (320.11 mg.kg^{-1}) at 600 mg.kg^{-1}. Significant increase shown in lead content of leaves when compare to control figure 4 (b). The highest value (254.433 mg.kg^{-1}) was recorded by eucalyptus at 400 mg.kg^{-1}. On the other hand, increasing the concentration of lead caused significant increment in lead content of leaves which reached its highest value (93.5 mg.kg^{-1}) at 600 mg.kg^{-1} of lead in *Platanus orientalis*. Poplar species recorded the highest content (112.533 mg.kg^{-1}) when treated with 400 mg.kg^{-1} of lead. Results in Figure 4 c reveals a reduction in the bioaccumulation factor in oriental plane tree as a result of increasing the concentration of lead in soil that record the highest value in control treatment (0.596) which was significantly different from other treatments. Eucalyptus shows less fluctuation in bioaccumulation factor with control that showed the highest value (0.668) followed by non insignificant decreases at (200 and 400 mg.kg^{-1}). Meanwhile, the highest concentration of lead shows the lowest bioaccumulation factor in eucalyptus 0.533. At the same time, significant inhibition was observed in poplar tree at 200 and 600 mg.kg^{-1} compared with both control and 400 mg.kg^{-1} Figure 4 c. Eucalyptus records the highest translocation factor when compare with plane and poplar trees which reaches 1.135, 1.013, and 1.018 in control, 200, and 400 mg.kg^{-1} respectively, but significant reduction and toxicity symptom obtained with increasing the level to 600 mg.kg^{-1} of lead (Picture 3) which was similar to poplar when treated by both 400 and 600 mg.kg^{-1} in comparison with control Figure 4d.. Moreover, the reduction in translocation factor was only significant (0.367) at 400 mg.kg^{-1} in oriental plane tree.

Picture 3 Symptoms of toxicity appeared on plant at high concentration of metal applied

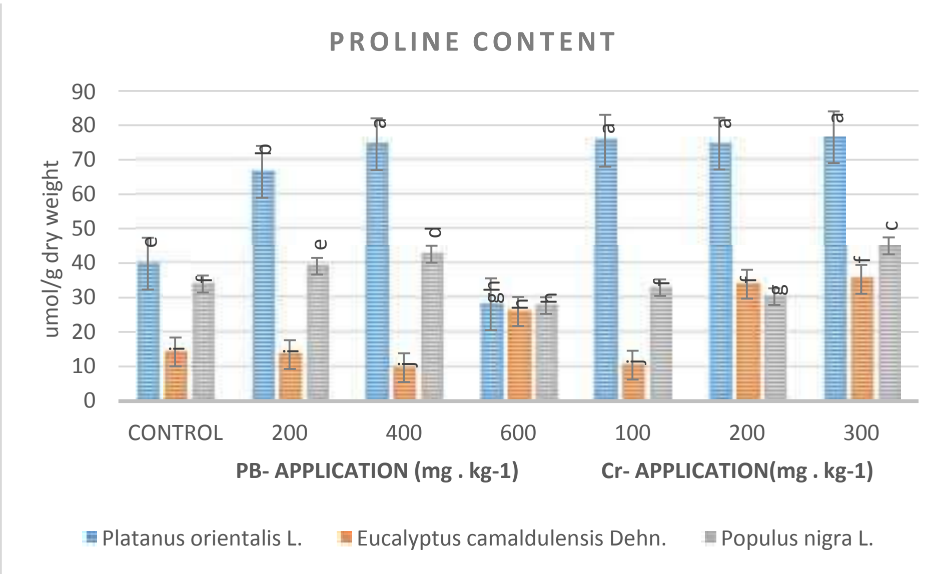

Figure 3. Proline under different Pb and Cr treatments

(Columns with the same letters are not significantly different according to Duncan's multiple range tests at 5% level)

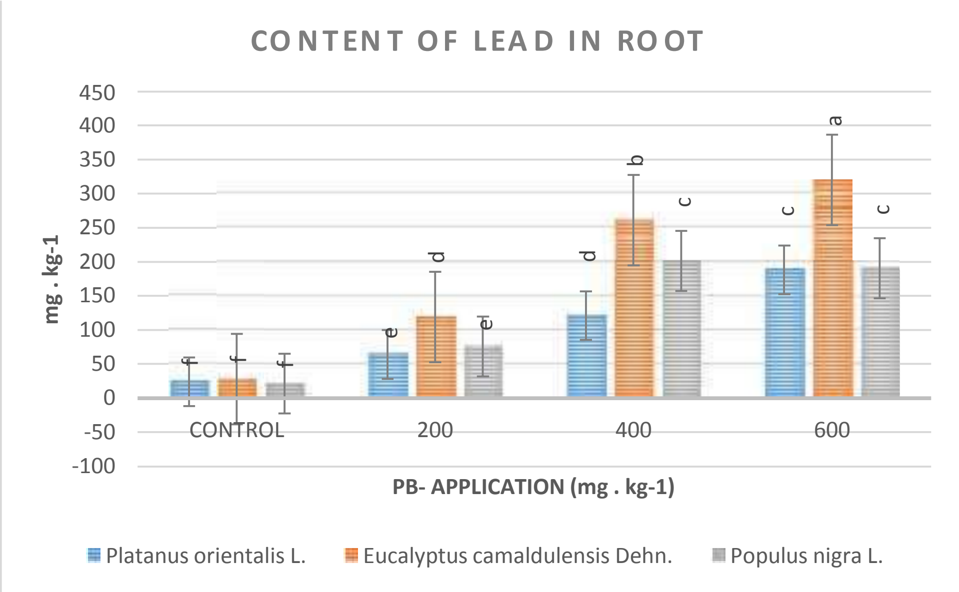

Figure 4 a. Effect of treatments on lead content in roots

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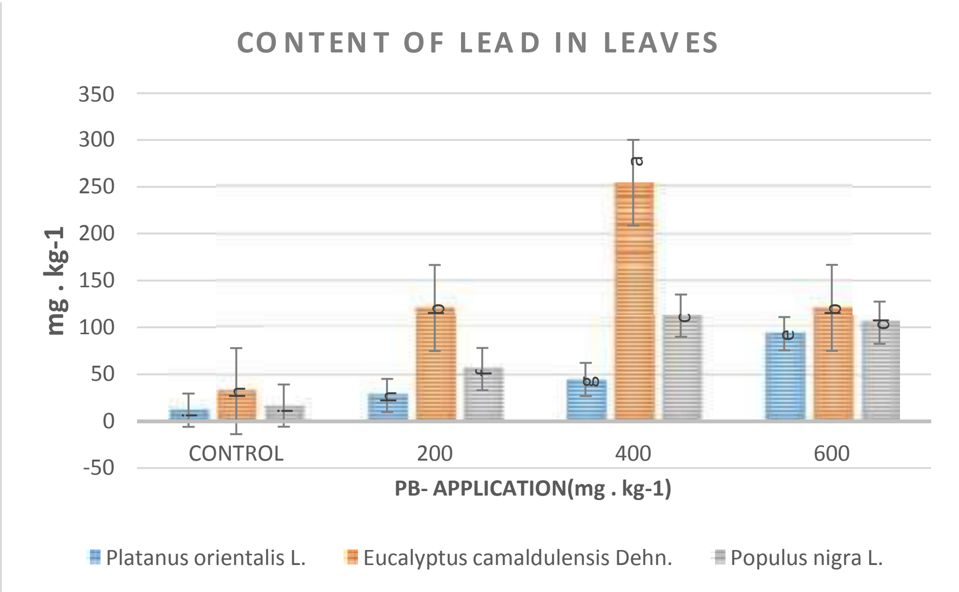

Figure 4 b. Effect of treatments on lead content in leaves

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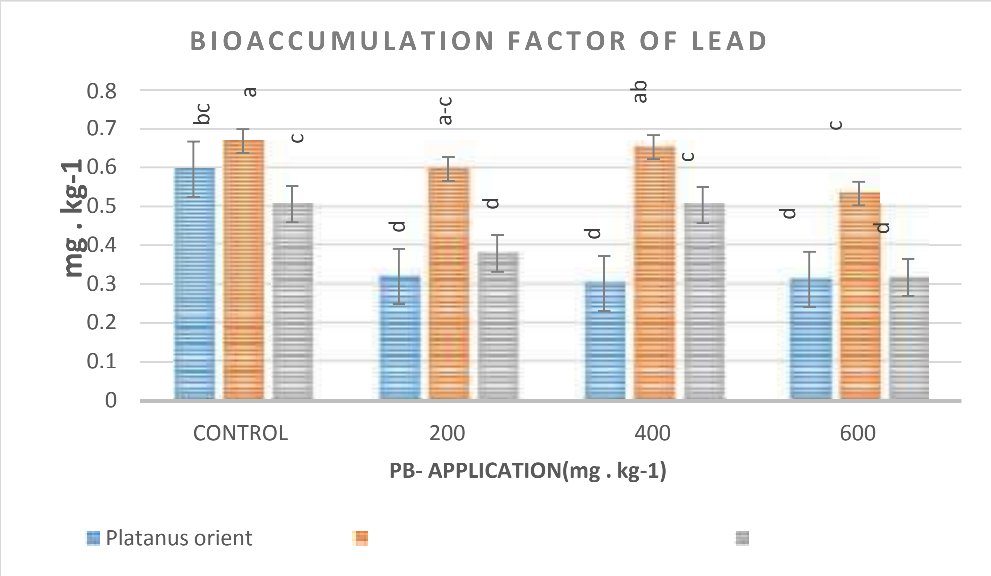

Figure 4 c. Bioaccumulation factor of lead (BAF)

(Columns with the same letters are not significantly different according to Duncan's multiple range tests at 5% level)

2-. Chromium content in plants

The chromium content in root increased during the increasing chromium levels in oriental plane and poplar trees figure 5 (a) which recorded their highest values (95.84 and 91.977 mg.kg^{-1}) respectively when treated with 300 mg.kg^{-1}. Whereas, eucalyptus reached its maximum chromium content (146.333 mg.kg^{-1}) in root when treated with (200 mg.kg^{-1}). Figure 5b illustrates the concentration of chromium in soil and their content in leaves. *Platanus orientalis* showed a linear correlation which reaches its highest value of (27.32 mg.kg^{-1}) at 300 mg.kg^{-1} of chromium. Meanwhile, eucalyptus and poplar record the highest values (69.82 and 39.818 mg.kg^{-1}) respectively at 200 mg.kg^{-1}.

The bioaccumulation factor of chromium reached its maximum value (0.476) in oriental plane when treated with 200 mg.kg^{-1} which was significantly differed from other treatments. Whereas, eucalyptus and poplar record the highest value (0.853 and 0.597 mg.kg^{-1}) respectively at 100 mg.kg^{-1} of chromium Figure 5 c. The three tested species showed fluctuation in the translocation factor of chromium with *Platanus orientalis* recorded the highest value (0.391) at 100 mg.kg^{-1}. While, control treatment has the highest translocation factor (0.838) in eucalyptus. Poplar reached the maximum translocation factor at (200 mg.kg^{-1}) figure 5 (d).

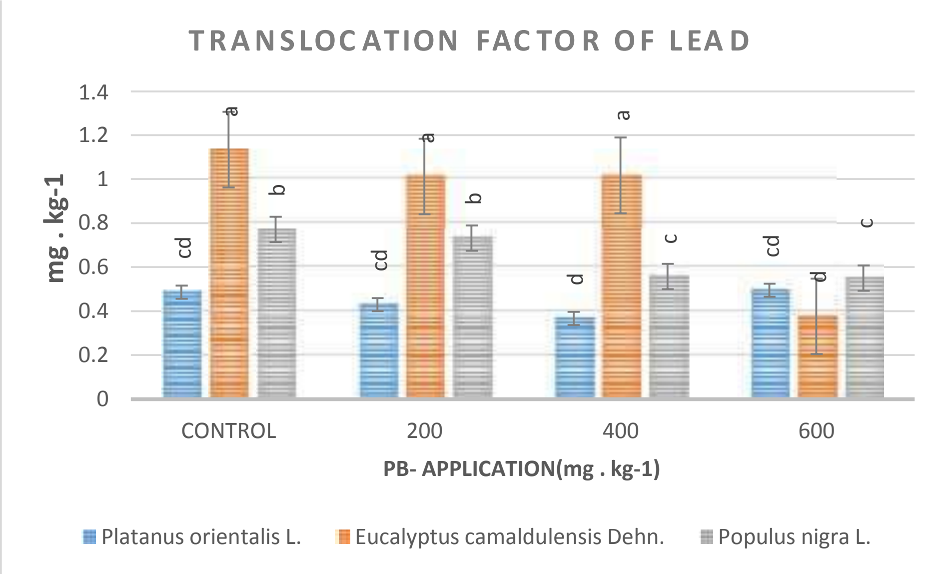

Figure 4 d. Translocation factor of lead (TF)

(Columns with the same letters are not significantly different according to Duncan's multiple range tests at 5% level)

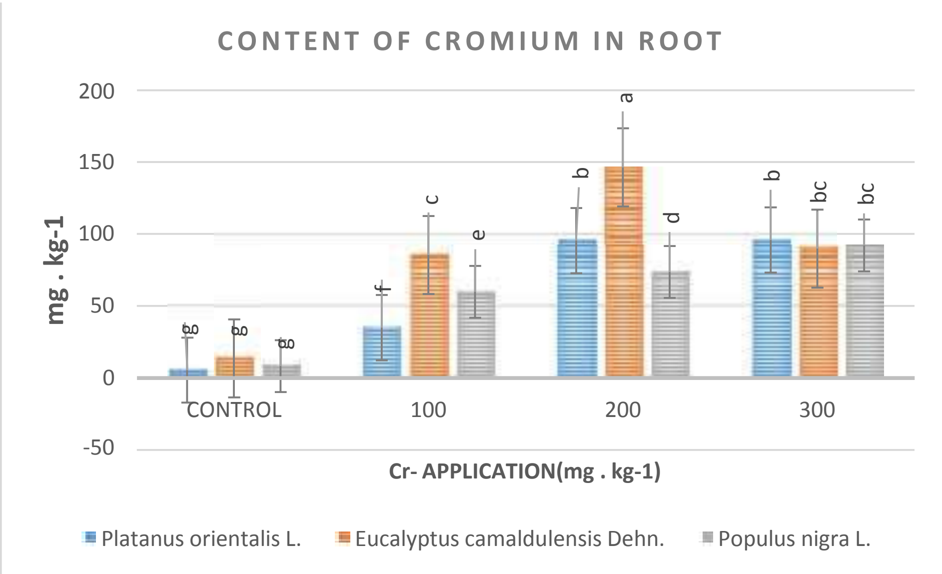

Figure 5 a. Effect of treatments on chromium content in roots

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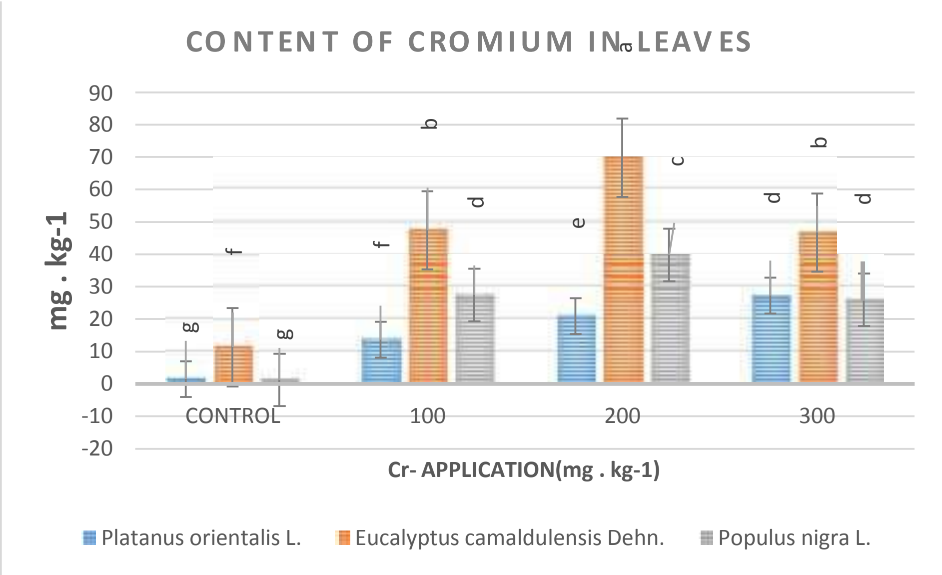

Figure 5 b. Effect of treatments on chromium content in leaves

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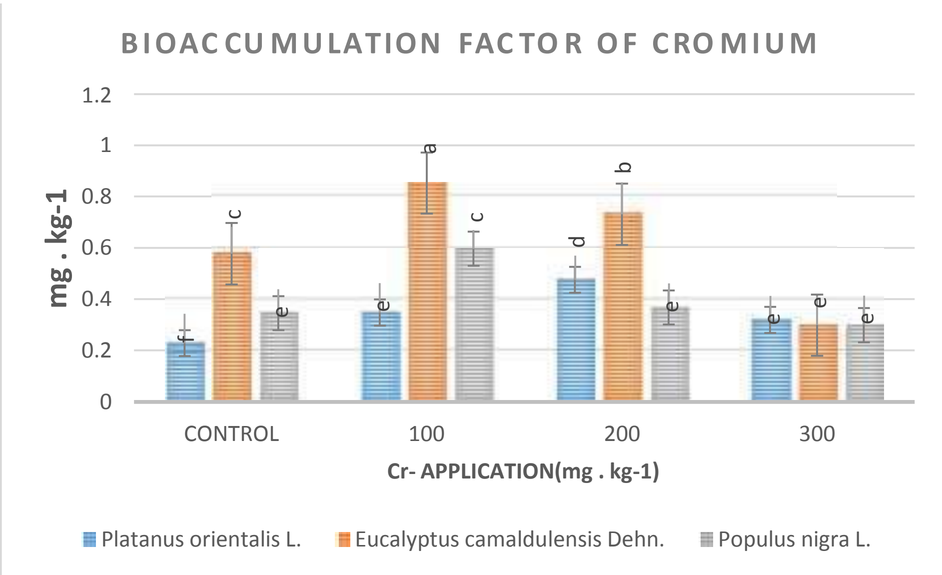

Figure 5 c. Bioaccumulation factor of chromium (BAF)

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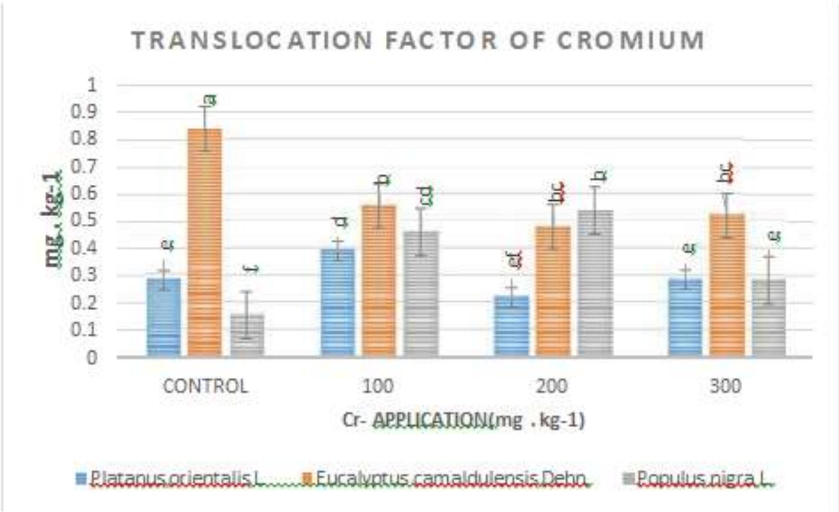

Figure 5 d. Translocation factor of chromium (TF)

Columns with the same letters are not significantly different according to Duncan's multiple range tests at 5% level)

Ecophysiological responses of plants to heavy metals toxicity has been reported by many researchers by manifesting their effecting on the growth parameters. Generally, plant dry weight was affected by both Pb and Cr, the inhibition of root dry weight exceeds that of shoot dry weight. Meanwhile, chromium treatments have a greater negative effect on root dry weight than lead and the reduction was more in *Platanus orientalis* than other species this may returns to the fact that heavy metals may have interaction with essential micro nutrient and in turn affect their uptake and transport by plants which influence plant growth and their physiological function (45). A depression in growth parameters mainly biomass production, root hair formation, and root elongation were observed in young tress [21]. Toxic ions like Al, Zn, Cu, Pb, Cd, and others may inhibit root growth and at higher concentration may lead to root necrosis and death. Reduction in root and shoot dry matter as well as in the total dry matter was observed with the increase in heavy metal contamination level (26). Reduction in all types of chlorophyll (a, b, and total) was observed and this inhibition was concentration dependent and was stiffer in chromium treatments. This reduction may return to the facts that these metals have the ability to replace magnesium (central atom of chlorophyll) which hampers the light-harvesting capability of chlorophyll thereby affecting photosynthetic process and this may interpret the reduction in the biomass. Chlorophyll -a- fluorescence measurement mentioned to be a good indicator to manifest the effect of stress in plants [22] according to that it seems that three tested species was more stressed under chromium treatments. Two key enzymes that control chlorophyll biosynthetic pathway has been reported to affect heavy metals: protochlorophyllide reductase and d-aminolaevulinic acid (ALA)- dehydratase theses enzymes are found to be sensitive to different metals [46]. The concentration of chlorophyll a, b, and total were decrease significantly when industrial wastewater with different concentration of heavy metals was used in irrigation 17. Proline is an amino acid that produced when plants exposed to a wide variety of environmental stresses 36. Its production aim to increase plant resistant under stress induce environment by regulation of osmotic potential which enable the plants to cope with osmotic stress 4, protect against plant denaturation 30, and control the production of reactive oxygen species ROS 3. The proline production in this experiment was more in chromium treated plant than of lead in most treatment which indicate that the species shows the effects of stressful environment in thepresence of chromium which may be resulted from; reduction in proline utilization, decrease in proline degradation, hydrolysis of protein, or increase in proline synthesis 9. Regarding lead and chromium accumulation in plant part root accumulation ability was more than shoot this may be due to the different patterns of behavior and mobility of heavy metal within a tree. Lead, Chromium and copper tend to be immobilized and held in root whereas, Cd, N1 and Zn are more easily translocated to aerial tissues 35. The accumulation patterns of metals seem to be independent to the increasing concentration of tested metals this may returns to the toxicity symptoms that appeared on plants at higher concentration of metals, this fact was reported by Zaki and Ridha (50). In all treatments the bioaccumulation factor was <1 which for a good phytoremediation candidate tree the BAF should be >1 however, a considerable amount of lead and chromium were absorbed by studied plants they just need to assist their remediation ability especially when the concentration of heavy metals is high. Translocation factor were <1 for both heavy metal except *Eucalyptus camaldulensis* when treatedwith (200 and 400 mg.kg-1) of lead this seems to be the result of the tendency of lead and chromium to be held in root.

Conclusion

The aim of this study was to test the tolerance and accumulation capacity of three tested speciesto lead and chromium contamination in order to seeks the best candidate for the remediation of contaminated soil with these heavy metals. Result obtained showed that species showed variation in their responses to lead and chromium. The order of tested trees capacity to show a tolerant character were *Eucalyptus camaldulensis* > *Populus nigra* > *Platanus orientalis.* It can be concluded from this study that the three species have the ability to tolerate moderately contaminated soil with lead and chromium since death were not occurred in any observation therefor, they can be used in these areas not only to decontaminate the soil but also to prevent the leaching of these metal to the ground water by continuous transpiration thereby preventing the contamination of new areas.

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