

Impact of Lead Toxicity on Germination, Plant Growth, Yield and Metal Uptake in Radish Plants

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ABSTRACT

The effects of lead on radish plants (*Raphanus sativus* L.) were investigated to understand the impact of lead accumulation on seed germination, plant growth and yield. Two germination and pot experiments were carried out during the 2006 and 2007 seasons, at the Faculty of Agriculture, Damanhour Branch, Alexandria University. Seeds and plants were treated with different concentrations of Pb: 0, 100, 200 and 300 ppm. Pb contents in both root and shoot tissues gradually increased with increasing the applied Pb concentration. More lead was accumulated in root tissues than in shoot tissues. The higher concentration of lead (300 ppm) caused a significant decrease in seed germination, plant height, leaves fresh and dry weights, leaves area and yield of (*Raphanus sativus* L.) Two polynomial quadratic equations were established to express the response of radish yield to Pb contamination. The experimental yield values and the corresponding calculated values were not significantly different as tested by high values of correlation coefficient.

INTRODUCTION

Lead (Pb) is one of the major heavy metals of the antiquity and has gained considerable importance as a potent environmental pollutant (Talanova *et al.* 1999). The background concentration of Pb in uncontaminated soil lies within the range of 10-50 ppm. However, in the soil with low-level contamination, Pb concentration can be expected to range from 30 to 100 mg/kg (Davies 1995). With rapid development in industry all around the world since the 20th century, the inputs of Pb to agricultural soils have been occurring through the combustion of gasoline containing Pb additives, human activities such as mining, smelting, electroplating, energy and fuel production, power transmission, intensive agriculture, sludge dumping and military operations (Nedelkoska and Doran, 2000). Excess Lead in environment can cause serious problems to all organisms (Herrero *et al.* 2003). Prenatal and early postnatal exposure to Pb resulted in damage to the central nervous systems. The damage is characterized by diminished intelligence, shortened attention span, and slowed reaction time. These effects are irreversible, untreatable, and lifelong (Wasserman *et al.* 1997). The absorption and transport of Pb by crops are of great concern, especially its accumulation in the edible part. Pb belongs among nonessential metals for plants and

has no known biological function (Wierzbicka 1995). The levels of Pb in soils that are toxic to plant are not easy to evaluate. However, it is generally agreed that soil Pb concentration ranging from 100 to 500 ppm are considered to be excessive (Kabata-Pendias, and Pendias, 1992). Pb inhibitory effects may be due to interference with enzymes essential for normal metabolic and development (Van Assche and Clijsters 1990), photosynthetic processes (Fodor *et al.* 1996), water and mineral nutrients absorption (Burzynski 1987), and changes in cell ultrastructure (Stefanov *et al.* 1995). The Pb content of plant grown on Pb-polluted areas is generally, highly correlated with the Pb concentration in soil, although the relationship differs among organs of the plants (Rooney *et al.* 1999). The uptake, transport, and accumulation of Pb by plants are strongly governed by soil and plant factors, and they differ significantly with plant species (Yang *et al.* 1993). The inhibitory and toxic effects of lead on seed germination and seedling growth have been reported in *Brassica pekinensis* (Xiong 1998), and *Pisum sativum* (Wierzbicka and Obidzinska 1998). Pb absorbed and accumulated in different plant tissues with the highest amount in the root tissues (Kumar *et al.*, 1995). Pb accumulation in roots takes place by binding with polysaccharides (Seregin and Ivanove, 1998) or binding to the cell walls in the roots and xylem vessels (Fathi, 1983) and thus might become immobile. Results of Jaja and Odoemena (2004) on tomato showed that increasing lead levels, significantly, decreased root length, shoot length, leaf area, fresh and dry weight. The response of *Brassica juncea* to lead was also studied by Begonia *et al.* (1998) who reported that root dry matter, total leaf area and shoot dry weight were reduced by lead treatment.

The leaves and roots of radish (*Raphanus sativus* L.) are edible; extracts of the leaves, flowers. and pods are useful against diseases caused by plant pathogenic bacteria, Also radish can be grown in a wide range of climates and their roots have useful medicinal property in the treatment of urinary complaints, piles, and gastro-diarrhea. (Chattopadhyay *et al.* 2006).

Although seed germination represents an initial and crucial phase in the life cycle of plants, virtually little information is available on the impact of lead on

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the action of gradually increasing concentrations of Pb on germinating radish seeds, plant growth and yield. Therefore this study was undertaken with a view to find out the effects of lead stress on seed germination, growth, biochemical constituents and mineral status of radish.

MATERIAL AND METHODS

Seed germination experiments

Two germination experiments were carried out in Petri dishes during April of 2006 and 2007 to determine the effect of lead on seeds germination, length of primary roots. Seeds of radish (*Raphanus sativus* L.) Balady cultivars were surface sterilized with 0.1% mercuric chloride solution and rinsed several times with distilled water. Sterilized glass petri dishes of approximately 15 cm in diameter, each containing 2 Whatman No. 1 filter papers were used as sowing container and media. Ten seeds of radish were spread uniformly in each of the Petri dishes. Pb Pollution was established by adding 20 ml of lead acetate solution at the various concentrations of 0, 100, 200, 300 ppm into the Petri dishes before sowing. Distilled water was used as the control treatment. Germination study was observed for 5 days. Percentage(%) germination was recorded when the radicle reached one mm in length. Each treatment was replicated five times and the solutions were replenished every other day. The germination percentage of the seeds, root length and lateral roots number were finally determined for each treatment. Root length was measured by placing the root on 1-mm squared graph paper. Tolerance indices (T.I.) were determined through use of the following formula given by Iqbal and Siddiqui (1992):

$$T.I. = \frac{\text{Mean root length in metal solution}}{\text{Mean root length in distilled water}} \times 100$$

Pot experiments

Two pot experiments were conducted on April during the two successive growing seasons of 2006 and 2007 at the Faculty of agriculture Damanhour Branch, Alexandria University. Plants were grown in pots in untreated soil (control) and in soil to which various concentrations of Pb had been added (100, 200, and 300 mg kg⁻¹ soil). Each pot contained 4 kg of air-dried soil (Clay loam, ECe=1.4 and 1.7 mmhos/cm; pH 7.5 and 7.8; Pb 1.12 and 1.26 ppm in the seasons of 2006 and 2007 respectively) to determine the effect of lead on the vegetative growth characters, lead content, yield of radish plants (cv. Balady) .

The physical and chemical characteristics of the Soil were determined according to the methods reported

by Page et al. (1982). Available lead was determined according to the method mentioned by Cottenie *et al.* (1982). and measured by atomic absorption spectrophotometer (AAS).

Experimental Layout

The experiments were set in completely randomized design with three replicates. Leads as lead acetate at the concentration of 0.0, 100, 200 and 300 ppm was used after dissolving in tap water and thoroughly mixed with the soil. In each pot 8 seeds were sown. All pots were irrigated with tap water whenever it was needed to keep the moisture in soil at about 70% of the total water holding capacity of the soil during the experimental period. Plants were thinned to three plants per pot one week after germination. Plants were harvested 50 days after sowing at edible stage. Roots and shoots were dried at 60C and weighed for the measurement of various growth parameters, chemical and mineral constituents.

Morphological Parameters

Morphological parameters, including shoot and root length, fresh and dry weight of leaves, and leaves area per plant. Leaves area were measured and calculated using the weight method as described by Fayed (1997).

Chemical Analysis

The collected plant samples were washed with tap water to remove the adhered soil particles and then washed several time with distilled water. The plant samples were oven dried at 70 C^o for 48 hours and ground in a mill with stainless steel blades .

Wet digestion procedure was performed according to Chapman and Pratt (1978). Lead contents were analyzed by atomic absorption spectrophotometer according to Cottenie *et al.*, (1982). Phosphorus percentage was determined calorimetrically as reported by Jackson, (1967). Potassium percentage was determined by flame photometer as described by Brown and Lilliand (1946).

All obtained data were statistically analyzed using SAS software program (1996). Comparisons among the means of different treatments were achieved using the revised least significant difference procedure at *P*= 0.05 level as illustrated by Al-Rawy and Khalf-Allah (1980).

RESULTS AND DISCUSSION

Effect of lead on seed germination

The effects of various concentrations of lead on seed germination of *Raphanus sativus* cultivar Balady are shown in Table(1). The results demonstrated that the percentage germination of radish seeds was not

significantly affected by lead treatments at lower concentration (100ppm Pb) compared to the control treatment in both seasons. However, the effects appeared significant at 200 and 300 ppm Pb treatments compared to control in both seasons. Lead treatment at 300 ppm reduced the germination percentage by 34.01 and 33.29%; 32.54 and 34.42 compared to the control after 3 and 5 days in the first and second seasons, respectively. Similar results have also been observed in *Brassica Pekinensis* by Xiong (1998). Hameed *et al.* (2001) also found that lead caused great reduction in germination percentage of *Spinacea oleracea* and *Lycopersicum esculentum*. The inhibitory effect of heavy metals on seed germination could be due to ionic toxicity or due to an osmotic effect (Redmann, 1974) or to the decreased level of auxin resulting from enhanced destruction of auxin by metal ion (Mukherji and DasGupta, 1972).

Effect of lead on root growth

Lead treatments, significantly decreased the root length (Table 2) at all concentrations than the control at 72 and 120 hours, in both seasons. After 72 hours, the reductions were 15.37, 33.03 and 47.71%; and 9.78, 34.13 and 49.27% of the control in the first and second seasons at lead concentration of 100, 200 and 300 ppm, respectively. Meanwhile, after 120 hours, the reductions were 21.79%, 36.19% and 51.95%; and 14.91%, 38.55% and 50.76% of the control in the first and second seasons at lead concentration of 100, 200 and

300 ppm, respectively. These findings are in line with those reported by Liu *et al.*, (1994) on onion. They found that Pb treatments inhibited the root growth of *Allium cepa* at concentration of 10^{-4} M. Obroucheva *et al.* (1998) reported that lead treatments affected both cell growth processes, i.e. cell division and cell elongation.

The lead inhibitory effect on cell division was noticed through the reduction of meristem size and decreased number of mature cells formed in a day. The inhibition of root growth after exposure to Pb may be due to a decrease in Ca in the root tips, leading to a decrease in cell division or cell elongation (Hausling *et al.*, 1988).

Early seedling growth was also inhibited in tomato and eggplant (Khan and Khan, 1983). Radish seedlings showed also low percentage of tolerance to lead treatment as compared to control. Tolerance to lead treatment at 300 ppm exhibited the lowest level as compared to lower concentrations in both seasons (Table 3). Iqbal and Siddiqui (1992) found also gradual decreases in plants growth with the increase in concentration of Pb. Tolerance to lead in some plant species was very low at higher concentrations. They concluded that the reason of low tolerance to lead might be due to occurrence of changes in physiological functions that took place during germination of those seedlings.

Table 1. Effect of lead acetate on seed germination of radish, during the seasons of 2006 and 2007

Pb (ppm)	Season 2006		Season 2007	
	Germination percentage after 72 hours	Germination percentage after 120 hours	Germination percentage after 72 hours	Germination percentage after 120 hours
	0	82.26 A*	98.24 A	83.36 A
100	76.74A	92.75 AB	78.54 A	93.33 AB
200	66.45C	84.36 B	64.39 B	86.76 B
300	54.28D	65.54C	56.23 C	63.92 C

*Values having a common alphabetical letter (s), do not significantly differ, using the revised L.S.D. test at P=0.05.

Table 2. Root growth responses of radish seedling to various lead levels during the seasons of 2006 and 2007

Pb (ppm)	Length of roots (cm) after 72 hours		Length of roots (cm) after 120 hours	
	2006	2007	2006	2007
0	4.36 A*	5.42 A	5.14 A	6.64 A
100	3.69 B	4.89 A	4.02 B	5.65B
200	2.92 C	3.57 B	3.28 BC	4.08 C
300	2.28 D	2.75 C	2.47 C	3.27 D

*Values having a common alphabetical letter (s), do not significantly differ, using the revised L.S.D. test at P=0.05.

Table 3. Root tolerance indices of radish seedling to various lead levels, during the seasons of 2006 and 2007

Pb (ppm)	Roots tolerance indices (cm) after 72 hours		Roots tolerance indices (cm) after 120 hours	
	2006	2007	2006	2007
	100	84.63 A*	90.22 A	78.21 A
200	66.97 B	65.87 B	63.81 B	61.45B
300	52.29 C	50.73 C	48.05 C	49.24C

*Values having a common alphabetical letter (s), do not significantly differ, using the revised L.S.D. test at P=0.05.

Effect of lead on vegetative growth characters:

The decrease in growth of radish was gradual and more depressed with increase in Pb concentration from 100 to 300 ppm. Root and shoots length of radish plants under lead treatment at 50 days are presented in Table (4). The root and shoot length decreased as the concentration of Pb in the soil increased. The lowest shoot and root length values were observed at 300 ppm Pb. The reductions were 44.09 and 51.9; 35.15% and 37.83%, compared to control treatment for shoot and root length in the first and second seasons respectively. Both leaves fresh and dry weights were significantly affected by Pb soil levels (Table 4). The effects of lead levels were severe at higher Pb concentrations. Reductions of leaves fresh and dry weight at 300 ppm relative to control plants were 61.76, 69.97%; and 57.74, 65.18% in the first and second seasons respectively. The reduction in roots development under excess Pb conditions was reported earlier for several other plants (Tomar et al., 2000). Pb at high levels may inhibit root and shoot growth directly by inhibiting cell division or cell elongation, or by a combination of both, resulting in limited exploration of the soil volume for uptake and translocation of nutrients and water (Khan and Frakland, 1983).

Leaf area of radish plants decreased with increased Pb content of the soil (Table 4). The reductions of leaf area of radish plants at the highest levels (300 ppm Pb) were 38.44 and 48.15% compared to control in first and second seasons respectively. Similar reductions in total leaf area due to excess lead have been observed by Khan and Frakland, (1983). The decrease in leaf area at high concentrations of Pb could be attributed to either a reduction in the number of cells or to a reduction in cell size as reported by Tomar et al. 2002 on mung bean.

Data presented in Table (4) illustrated that lead at all levels significantly inhibited root fresh and dry weights of radish plants and the inhibition was more severe at the highest Pb level. The reduction of root fresh and dry weight of radish plants grown in soil polluted with 300ppm Pb reached about 58.9 and 58.4% (1st season) and 65.58 and 60.06% (2nd season) compared to the untreated plants, respectively, The negative effects of lead on root fresh and dry weights of

radish plants may be due to its inhibitory effects on the uptake and translocation of some major and micro elements within plant roots un(Kabata Pendias and Pendias 1992); on the activity of some enzymes as well as on biosynthesis of photosynthetic pigments which were reflected on radish yield.

Biochemical Constituents

Photosynthetic pigments, such as chlorophyll a, chlorophyll b, and total chlorophyll contents of radish leaves decreased significantly with increasing Pb levels in the soil (Table 5). Excessive Pb treatment at the concentration of 300 ppm decreased significantly chlorophyll a, chlorophyll b and the total chlorophyll by 38.24%, 52.5% and 43.52% in first season and by 45.45, 50.52 and 47.31%, in the second season respectively. Excessive Pb might interfere with the synthesis of chlorophyll because of Pb treatments presumably block the synthesis and activity of enzyme proteins responsible for chlorophyll biosynthesis (Ahmed and Tajmir- Riahi 1993) or by causing impaired uptakes of essential elements such as Mg and Fe by plants (Burzynski, 1987).

Phosphorous and potassium contents

At all levels of applied Pb the concentrations of P and K in leaves and roots of radish plants decreased significantly compared to corresponding control values. As well as, the accumulation of phosphorus and potassium were also found to be high in tops than in roots (Table 6). Radish leaves showed reductions of 35.71 and 32.61 in P content at 300ppm Pb compared to 25.31 and 30.88 in K content in 2006 and 2007 seasons, respectively. On the other hand at the same lead treatment, reductions of 35.94 and 36.83; and 33.22 and 36.17 were detected in roots content of P and K in the first and second seasons respectively. Similar trends were also reported by Walker et al. (1997), who found that Pb decreased the uptake of K, Ca, Mg, Fe and NO₃⁻ in seedlings of *Cucumis sativus* and decreased the uptake of Ca, Mg, K and P in *Zea mays*. Also, Paivoke (2002) reported that Pb influenced the overall distribution of nutritional elements within the different organs of the plant. Phosphorus content was found to be negatively correlated with soil Pb.

Table 4. The effect of lead on morphological parameters of radish plants during 2006 and 2007 seasons

Lead concentration ppm	Leaves (cm)	Roots length (cm)	Leaves fresh weight (g)	Leaves dry weight (g)	Leaves area (cm ²)	Root fresh weight (g)	Root dry weight (g)
2006							
0	29.12 A	14.28 A	49.69 A	4.52 A	902.13 A	40.41 A	2.69 A
100	24.13 A B	13.18 A	38.26 B	3.75 A B	822.08 A	33.10 AB	2.25 AB
200	20.59 BC	11.29 B	27.29 C	2.72 B C	660.24 B	25.73 C	1.62 B C
300	16.28 C	09.26 C	19.00 D	1.91 C	555.38 C	16.61 D	1.12 C
2007							
0	30.27 A	15.73 A	57.31 A	4.94 A	1011.5 A	44.63 A	3.13A
100	27.64 B	13.09 B	42.54 B	4.13 B	966.24 B	35.24 B	2.54 B
200	19.27 C	12.25 B	26.24 C	2.57 C	634.81 C	24.47 C	1.50 C
300	14.56 D	9.78 C	17.21 D	1.72 D	524.45 D	15.36 D	1.25 D

Table 5. Effect of Pb on chlorophyll concentration of leaves of radish plants during 2006 and 2007 seasons

Lead concentration ppm	chlorophyll a (mg/g dry wt) in leaves	chlorophyll b (mg/g dry wt) in leaves	Total chlorophyll
2006			
0	3.4 A	2.0 A	5.4 A
100	3.0 B	1.7 A	4.7 B
200	2.7 B	1.2 B	3.9 C
300	2.1 C	0.95 B	3.05D
2007			
0	3.3 A	1.9A	5.2A
100	3.2 A	1.6 AB	4.8 A
200	2.4 B	1.3 BC	3.7 C
300	1.8 C	0.94 C	2.74 D

*Values having a common alphabetical letter (s), do not significantly differ, using the revised L.S.D. test at P=0.05

Table 6. Effect of Pb on elemental content of leaves of radish plants during 2006 and 2007 seasons

Lead concentration ppm	P%	K%	Pb (ppm)
2006			
0	0.84 A	4.03 A	19.6 D
100	0.76 B	3.73 B	118.7 C
200	0.61 C	3.32 C	184.4 B
300	0.54 D	3.01 D	250.6 A
2007			
0	0.92 A	4.21 A	22.1 D
100	0.85 A	3.85 B	108.9 C
200	0.74 B	3.13 C	214.8 B
300	0.62 C	2.91 D	285.4 A

Lead contents

The results of this study showed that the concentration of Pb increased significantly to different extents in both tops and roots (Tables 6 and 7). When soil Pb concentration reached 300 ppm, the lead contents in roots and leaves were as high as 497.2 and 250.6, µg/g; and 468.6 and 285.4 µg/g, in the first and second seasons respectively. Thus the supply of Pb stimulated the accumulation of Pb not only in roots but also in tops. However, the accumulation of Pb was higher in roots than in tops at different Pb levels. This

result agreed with that of Malkowski *et al.* (2002) who showed that the Pb concentration in the roots of corn seedlings increased with increasing Pb concentration. About 90% of Pb accumulated in a number of species of the Brassicace family and other plants were in the roots (Kumar *et al.*, 1995). Roots can accumulate Pb up to 3-50 times more than leaves (Wozny *et al.*, 1995). Lead translocation from roots to shoots takes place by loading in the xylem sap and translocating to the aboveground parts through the transpiration stream (Briat and Lebrun, 1999). Pb accumulation in the root

takes place by binding with polysaccharides (Seregin and Ivanove, 1998), complexing with organic acids or binding to the cell walls in the roots and xylem vessels (Fathi, 1983) and thus might become immobile. The low allocation ratio of Pb between the roots and shoots may also be due to Pb movement in the root tissues which is usually prevented by the endodermis (Wierzbicka, 1987).

The relationship between applied Pb and changes in the Pb concentration of the leaves and roots was described mathematically (Figs1and2) by polynomial quadratic equations described in Snedecor and Cochran (1989)

$$Y_i = B_0 + B_1 X_i + B_2 X_i^2 \quad (1)$$

Where Y_i is the predicted Pb content corresponding to lead applied rates x_i .

Table 7. Effect of Pb on elemental concentration of roots of radish plants during 2006 and 2007 seasons

Lead concentration ppm	P%	K%	Pb ppm
2006			
0	0.64 A	2.86 A	24.1 D
100	0.59 A	2.59 B	226.9 C
200	0.48 B	2.23 C	318.77 B
300	0.41 C	1.91 D	497.2 A
2007			
0	0.76 A	2.93 A	27.3 D
100	0.67 B	2.62 AB	202.5 C
200	0.53 C	2.32 B	306.9 B
300	0.48 D	1.87 C	468.6 A

Table 8. The polynomial quadratic equations expressing lead contents in roots and leaves of radish plants as affected by lead application in 2006 and 2007 seasons

Polynomial Quadratic Equations	
Season 2006	
Y roots = - 0.0006x ² + 1.693 x + 33.985	R ² = 0.9832
Y leaves = - 0.0001x ² + 0.802 x + 21.445	R ² = 0.9976
Season 2007	
Y roots = - 0.0003x ² + 1.5296 x + 33.705	R ² = 0.992
Y leaves = - 0.0004x ² + 1.0173 x + 19.38	R ² = 0.9963

Table 9. Experimental and predicted lead contents of roots and leaves of radish plants as affected by applied lead in 2006 and 2007 seasons

Season 2006					
Lead content (ug/g dry wt.) Roots			Lead content (ug/g dry wt.) Leaves		
Treatments	Exp	Pre	Treatments	Exp	Pre
Pb0	24.1	34.0	Pb 0	19.6	21.45
Pb100	226.9	197.33	Pb100	118.7	100.65
Pb200	318.7	348.67	Pb200	184.4	177.85
Pb300	497.2	488.01	Pb300	250.6	253.05
R= 0.9916			R= 0.9954		
Season 2007					
Lead content (ug/g dry wt.) Roots			Lead content (ug/g dry wt.) Leaves		
Treatments	Exp	Pre	Treatments	Exp	Pre
Pb0	27.3	33.71	Pb0	22.1	19.38
Pb100	202.5	183.67	Pb100	108.9	117.11
Pb200	306.9	327.63	Pb200	214.8	206.84
Pb300	468.6	465.59	Pb300	285.4	288.57
R= 0.9960			R= 0.998		

B_0 is the intercept, represents Pb content without Pb application, B_1 and B_2 are the linear and quadratic coefficients respectively and X is the level of Pb applied.

Polynomial quadratic equations in Table(8) and Figs.(1and2) were used to predict lead contents in leaves and roots of radish plants in both seasons. The models appeared to be suitable for predicting the response of radish plants to Pb application. Since the estimated values of lead content by the models were found to be closely associated with the measured ones as evidenced by the high R values (Table 9). Such a mathematical approach may help the investigators to quantitatively predict and compare lead contents as affected by the levels of Pb pollution.

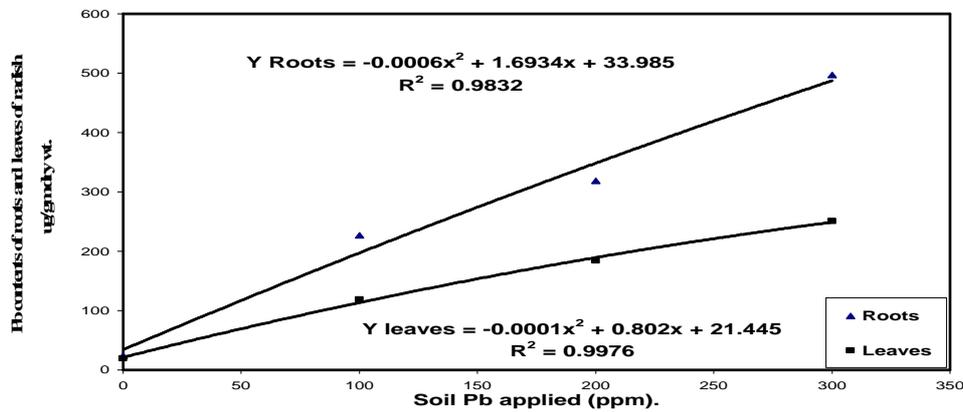


Fig. 1. The relationship between Pb applied and Pb contents of radish roots and leaves in 2006 season

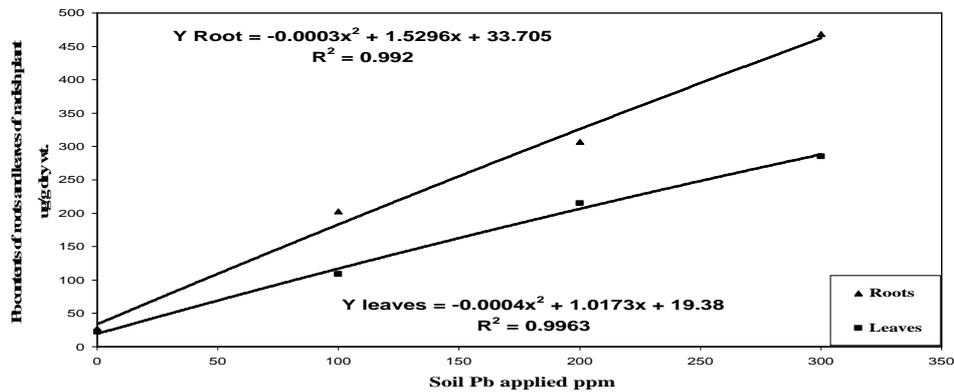


Fig. 2. The relationship between Pb applied and Pb contents of roots and leaves of radish plants in 2007 season

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الملخص العربي

دراسة تأثير تلوث الأراضي بالرمصاص على الإنبات والنمو والمحصول والتركيب الكيماوي للفجل

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النمو الخضري ومحصول الجذور والاوراق مقارنة بالتربة الغير ملوثة في كل من موسمي النمو. وقد ارتبطت اقل القيم المسجلة لهذة الصفات وذلك عند تلوثها بالرمصاص بأعلى مستوى (٣٠٠ جزء في المليون) في موسمي النمو.

أمكن الحصول على علاقة كمية لدراسة تأثير العلاقة بين تلوث الارض بالرمصاص ومحتوى الجذور والاوراق من عنصر الرصاص وتم التعبير عنها رياضياً بمعادلتين من الدرجة الثانية لكل من الجذور والاوراق في كل من موسمي النمو.

اجريت تجربتان في الاصح خلال موسمي ٢٠٠٦، ٢٠٠٧ بكلية الزراعة- فرع دمنهور- جامعة الاسكندرية بهدف دراسة تأثير معاملات تلوث الأراضي بالرمصاص على الانبات وصفات النمو الخضري ومحصول الجذور والاوراق والتركيب الكيماوي لنبات الفجل البلدى حيث استخدمت اربع مستويات مختلفة من الرصاص وهى ٠، ١٠٠، ٢٠٠، ٣٠٠ جزء في المليون تمت اضافتها على صورة محلول من محلات الرصاص. اظهرت النتائج ان تلوث التربة بالرمصاص ادى الى انخفاض معنوى في الانبات وصفات