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Response of Lupin and Wheat to Soil Contamination with Heavy Metals

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THIS INVESTIGATION clearly indicates that characteristics of the test metals, their concentration and/or type of organ , or the plant itself , play a significant role in the physiological responses towards application of these elements. Lupin and wheat plants were cultivated in contaminated soils with either Cu, Zn, Cd or Ni for 4 weeks. None of the test elements affected any appreciable change in the dry weight gain by the roots of these plants.

Zn was highly toxic to dry weight gain by lupin , whereas Ni was least, if at all, effective. Wheat differed in its response, being more sensitive to the smallest volume of highly reactive Ni , as well as to the large-sized Cd with the largest atomic weight, more than copper or zinc. Lupin leaves can tolerate the lowest moderate levels of either test elements , without affecting either chlorophyll a or b content. Cu seemed to be without effect on chlorophyll-a content of wheat, but the other elements were suppressive, Zn being most and Ni being least effective. Cu and Ni were without effect on chlorophyll b, while Zn and Cd were inhibitory.Cd and Zn were effective in reducing the total carbohydrate content of lupin plants. Both elements are equally stable, whereas Cu and Ni are more reactive. Cd was most effective in lowering the total carbohydrate content of wheat, followed by Cu and Ni and least Zn.

Cd and Cu affected the nitrogen assimilation of both lupin and wheat. The four test elements mostly attenuated N, Na, K and Ca levels with minor effects on P or Mg.

Contamination of soils with heavy metals becomes more and more a problem in many countries. In areas where metal contaminated soils are used for food crop production, relatively mobile ions, such as Zn or Cd, may reach the human body through such crops.

Zenk (1996) proved that phytochelating peptides are induced in all autotrophic plants so far analyzed. Inspite of the fact that nucleic acid sequences and proteins are found in higher plants that have distant homology to animal metallothionines, there is no experimental evidence that these plant metallothionines are involved in the detoxification of heavy metals. De Filippis and Ziegler (1993) found that heavy metals caused a significant lowering of two key metabolites related to the early products of carbon dioxide fixation, while fructose-1,6-diphosphate was not affected. However, later products, as well as glycolate, increased with heavy metals. The immediate products of the enzyme, more strongly inhibited by metals, were present at their lowest levels. As cells aged, there was also recovery of metabolite concentrations. These observations are interpreted in terms of the direct action of heavy metals on enzymes, as well as on levels of high energy phosphates.

Ayaz and Kadioglu (1996), working with Lens found that germination percentage decreased with increasing heavy metal concentration, except for Zn where it was ineffective. Mocquot *et al.* (1996) found a significant decrease in root and leaf biomass at a concentration of 10μM copper. In contrast, changes in several enzyme activities occurred at lower copper concentrations in the nutrient solution. Peroxidase activity significantly increased in all investigated maize plant organs.

Nagoor and Ayas (1997) concluded that concentrations above 50 μg/ml of Cu or Cd inhibited germination, seedling growth, fresh and dry weights. Root growth suffered more than the shoot. Carbohydrate metabolism (α- ,β- amylases and α- invertase activities), reducing and non-reducing sugars content were altered by heavy metals. α-amylase activity in the embryo and endosperm was greatly lowered by high concentrations of heavy metals. After 48 hr of growth, copper stimulated β-amylase activity, while Cd lowered it in the embryo. Higher content of reducing and non-reducing sugars were noticed in both embryo and endosperm of metal-treated wheat seedlings.

Kalita (1997) and Rai *et al.* (1991) discussed the effects of 1–1000 μg $CuSO_4$/ ml on the growth of *Azolla–Anabaena*, collected from a rice field, and found that 1μg Cu / ml stimulated the growth . Biomass yield was higher after 21 days of treatment. All other concentrations inhibited growth and chlorophyll metabolism. Nitrogen accumulation was slightly stimulated by 1μg Cu / ml. Moustakas *et al.*(1997) found a positive relationship between leaf photosynthesis and grain production by wheat plants under copper stress. Changes in the rate of carbon dioxide assimilation were due to changes in stomatal conductance. Lower carbon dioxide assimilation, under copper stress, was associated with a lower chlorophyll content. Prasad and Ram (1991) proved that copper, at 5 ppm, increased potassium content, but decreased potassium uptake. Zinc, at 5 ppm, decreased potassium content of *Vigna radiata* plants, but seemed without effect on potassium uptake at harvest. Zinc is an essential metal entering in many cell physiological processes (Sigel, 1983) while high concentrations are toxic (Woolhouse, 1983 and Verklij and Schat, 1990). In addition, zinc has an essential role in protein, carbohydrate, nucleic acid and fat metabolisms.

Dang *et al.* (1990) proved that 50 mg Zn/kg soil slightly increased onion and Trigonella yield, whereas the yield decreased at higher zinc levels. Patil *et al.* (1998) applied 0, 10, 20, and 30 kg $ZnSO_4$ per hectare to 20 wheat cultivars. The mean grain yield was 2.26, 2.78, 2.83 and 2.91 ton/ha, respectively. Total chlorophyll content increased with increasing zinc rates. Tahlil *et al.* (2000) reported that seedlings of marrow cultivar, treated with 5µg Cd / g, 200 µg Cu / g, 500 µg Zn/g , had some ameliorative changes characterized by the appearance of a new isoform of peroxidase.

Addition of cadmium at 0–400 ppm / kg soil, cultivated with *Vigay composit*, lowered dry weight biomass of roots and shoots (Narwal *et al.*, 1990). Kuo and Huang (1992) noticed that cadmium in polluted soils increased Ca, Mg and K uptake by lettuce and soybean plants, fertilized by organic manure. Yadav *et al.* (1996) concluded that seed germination and wheat seedling growth were inhibited by exposure to cadmium, whereas peroxidase and total soluble sugars increased. Abdel-Aal and Abdel-Nasser (1996) reported that leaf area, fresh and dry weights and protein content of maize seedlings decreased by increased Cd concentration. The effect was greater on roots than shoots. Activity of δ-aminolevulinic acid dehydratase and phosphoenol pyruvate carboxylase were greatly decreased by Cd.

Bhattacharjee *et al.* (1996) observed a decreased activity of peroxidase and catalase in growing rice seedlings. Hydrogen peroxide content increased in leaves and roots with heavy metal treatment. Cadmium mediated greater membrane damage in rice. Fodor *et al.* (1996) suggested a change, caused by cadmium, in the fatty acid composition of the phospholipid fraction in the plasma membrane, which differed in wheat and sunflower roots.

Abdel-Sabour (1991) proved that 50 mg Ni / kg soil reduced dry matter in sandy loam and loam soils. Nickel concentration in stems and leaves increased linearly by increased nickel in the soil. It had a synergistic effect on Zn, Cu and N uptake and an antagonistic effect on Co uptake. Piccini and Malavolta (1992) showed that 1-4 mg / litre nickel in the irrigated waters induced toxic effects on roots and leaves of *Phaseolus* plants. Chlorophyll content decreased up to 50%, whereas Ca, Mg, Mn and Zn contents of the tissues were not affected. In the meantime, N, P, K, Cu and Ni increased.

Xylander *et al.* (1993) found that 10–100 µM affected growth and germination of *Spirodela polyrhiza*, while carbohydrates content increased due to inhibition of translocation of photosynthetic products. Shalygo *et al.* (1999) reported that 10^{-6} and 10^{-1} M nickel stimulated chlorophyll accumulation and synthesis of 5-aminolevulinic acid suggesting that the cation modified enzyme activity, but did not affect the enzyme amount.

The four metals, under consideration, are all bivalent, of the d-element of the periodic table. Copper belongs to group Ib, while zinc and cadmium belong to group IIb, whereas nickel belongs to group VIII. Cadmium has the highest molecular weight and ionic radius, while copper and zinc have almost the same molecular weight, whereas nickel has the least molecular weight. According to the electromotive force (bioreactivity), copper is less reactive and cadmium is more reactive than nickel, whereas zinc is more reactive than cadmium and nickel (Cotton and Wilkinson, 2000). The last electron level of either zinc or cadmium is completely filled, thus more stable, whereas that of copper or nickel is less filled (less stable).

The aim of this investigation is to detect the difference in response of a monocot and a dicot plant towards heavy metal contamination by elements of variable characteristics.

Material and Methods

120 pots were prepared, each containing 2 kg of mixed soil (2:1 ratio of peatmoss and sandy soil obtained from Ismailia Governorate). Soil characteristics are provided. The heavy metals were added as sulphates at 0, 50, 100, 200 and 400 mg/kg soil, then implanted with either lupin seeds (*Lupinus termis*) or wheat grains (*Triticum aestivum*) , kindly obtained from Agricultural Research Center, Giza, Cairo, Egypt. Hoagland solution (1/10 concentration) was added at field capacity every other day for 2 weeks, followed by ½ concentration at 3-day intervals.

Characteristics of the used soil

Type	PH value	Organic matter %	Soluble salt %	Field capacity	Humidity %	Chemical avaiable elements (ppm)							
						Fe	Mn	K	Mg	Zn	Cd	Cu	Ni
Sandy	7.9	0.484	1.17	653 cm^3/kg soil	3.2	7	10	90	134	0.7	0.83	3.11	0.7

Four weeks after sowing, the plants were collected to determine their fresh and dry weight. Fresh leaf samples were used for the determination of chlorophyll content, applying the methods of Metzner *et al.* (1965). Dry samples were used to estimate total carbohydrates and total nitrogen contents applying the methods of Fales (1951) and Delory (1949), respectively. Sodium and potassium were estimated, after wet digestion, by flame photometry, whereas calcium and magnesium were determined by atomic absorption, using a Pirken-Elmer apparatus No. 603. Phosphorus was assayed spectrophotometrically.

Results

Results in Table 1 indicate that 50 mg/kg soil of either Cu, Cd or Ni, insignificantly affected fresh weight of lupin shoots or roots. Higher elements level suppressed the fresh weight of lupin shoot, which was furthered by larger doses. On the contrary, doses of zinc were significantly suppressive to fresh weight gain by lupin shoots without affecting the gain by roots (except 400 mg/kg soil).

TABLE 1. Effect of different concentrations of various heavy metals on the fresh weight (in g) of 4-weeks old lupin or wheat seedlings (mean of 5 replicates).

Element	Plant	Organ	Concentration (mg / kg soil)					L. S. D. at 5%
			0	50	100	200	400	
Copper	Lupin	Shoot	5.75	5.16	3.85	4.89	3.59	0.71
		Root	0.35	0.33	0.29	0.25	0.21	0.08
		Total	6.10	5.49	4.12	4.14	3.80	0.77
	Wheat	Shoot	3.59	2.27	2.18	2.10	1.59	0.39
		Root	0.83	0.75	0.66	0.54	0.28	0.18
		Total	3.42	3.02	2.84	2.64	1.87	0.57
Zinc	Lupin	Shoot	6.00	4.71	4.41	4.27	3.10	0.65
		Root	0.37	0.35	0.34	0.31	0.22	0.10
		Total	6.37	5.06	4.75	4.58	3.32	0.77
	Wheat	Shoot	2.37	2.37	2.17	1.61	1.33	0.35
		Root	0.72	0.66	0.36	0.30	0.27	0.06
		Total	3.09	3.03	2.55	1.91	1.60	0.42
Cadmium	Lupin	Shoot	5.18	4.79	4.44	2.79	2.27	0.62
		Root	0.33	0.29	0.25	0.24	0.21	0.06
		Total	5.51	4.78	4.69	3.03	2.48	0.67
	Wheat	Shoot	2.27	1.18	1.19	0.64	0.40	0.23
		Root	0.82	0.20	0.14	0.12	0.08	0.04
		Total	3.19	1.38	1.33	0.76	0.48	0.29
Nickel	Lupin	Shoot	7.48	7.47	7.54	7.10	6.88	0.65
		Root	0.29	0.13	0.12	0.08	0.07	0.03
		Total	7.77	7.60	7.66	7.18	6.95	0.70
	Wheat	Shoot	2.52	1.52	1.19	1.10	1.06	0.34
		Root	0.61	0.66	0.66	0.35	0.26	0.01
		Total	3.13	2.18	1.85	1.45	1.32	0.36

Wheat was less sensitive to Cu than lupin, as the drop in fresh weight gain by the shoot or root started at 200 mg/kg soil, but was more sensitive to cadmium, as the fresh weight gain dropped by application of 50 mg or above. The same applies to Ni, though the root system was more tolerant to this element than lupin, since the drop in root fresh weight was noticed at 200 mg/kg soil or above, whereas the shoot seemed more tolerant. Wheat roots seemed more sensitive to zinc than those of lupin, as the drop in fresh weight gain by the shoot started at 200 mg, whereas of the root started at 200 mg / kg soil.

In the meantime, lupin shoots were more sensitive to copper than to cadmium, as the drop in dry weight gain started at 100 mg/kg soil for copper and at 200mg/kg soil for Cd. Nickel invariably affected shoot dry weight gain by lupin, whereas zinc seemed to be very toxic to lupin shoot dry weight gain, as it dropped even at 50 mg/kg soil. On the other hand, lupin root dry weight was hardly affected by either elements, except zinc that progressively lowered dry weight gain by increasing its concentration in the soil. Wheat shoot was most sensitive to cadmium and nickel, as the drop in dry weight started at the least applied dose of either elements and less sensitive to copper or zinc, since the drop in dry weight gain started at 100 mg / kg soil. The fluctuations in dry weight gain by the roots of either plants seemed irrelevant (Table 2).

TABLE 2. Effect of different concentrations of various heavy metals on the dry weight (in g) of 4-weeks old lupin or wheat seedlings (mean of 5 replicates).

Element	Plant	Organ	Concentration (mg / kg soil)					L. S. D. at 5%
			0	50	100	200	400	
Copper	Lupin	Shoot	0.76	0.74	0.48	0.38	0.24	0.05
		Root	0.04	0.04	0.03	0.02	0.01	N.S.
		Total	0.80	0.78	0.52	0.40	0.25	0.05
	Wheat	Shoot	0.44	0.39	0.32	0.28	0.25	0.06
		Root	0.08	0.06	0.05	0.03	0.03	0.01
		Total	0.52	0.45	0.37	0.31	0.28	0.07
Zinc	Lupin	Shoot	0.89	0.69	0.63	0.54	0.40	0.09
		Root	0.08	0.07	0.06	0.05	0.05	0.01
		Total	0.97	0.76	0.69	0.59	0.45	0.10
	Wheat	Shoot	0.59	0.48	0.35	0.28	0.27	0.09
		Root	0.07	0.06	0.05	0.05	0.04	0.02
		Total	0.66	0.54	0.40	0.33	0.31	0.11
Cadmium	Lupin	Shoot	0.81	0.80	0.74	0.38	0.24	0.08
		Root	0.04	0.03	0.03	0.03	0.03	0.01
		Total	0.85	0.83	0.77	0.41	0.27	0.09
	Wheat	Shoot	0.38	0.21	0.20	0.14	0.09	0.03
		Root	0.07	0.04	0.03	0.02	0.02	0.01
		Total	0.45	0.25	0.23	0.21	0.11	0.04
Nickel	Lupin	Shoot	0.77	0.83	0.87	0.76	0.68	0.09
		Root	0.04	0.04	0.04	0.03	0.03	0.01
		Total	0.81	0.87	0.91	0.79	0.71	0.10
	Wheat	Shoot	0.40	0.34	0.29	0.23	0.13	0.05
		Root	0.05	0.05	0.03	0.02	0.01	0.01
		Total	0.45	0.39	0.32	0.25	0.14	0.06

The fluctuations in chlorophyll a-content of lupin seedlings were minor, following exposure to either of the test-elements, except Ni that sharply attenuated its accumulation, with its increasing level in the soil. The largest dose of Cu or Cd favoured the accumulation of chlorophyll a. Similarly Cu did not affect chlorophyll a-accumulation in wheat, but the other elements dramatically reduced chlorophyll a-accumulation, most prominently Zinc, then cadmium and least by Ni. Chlorophyll b of lupin increased by increased Zinc-concentration, but decreased with the other elements, more prominently in the presence of Cd, followed by Ni and least by Cu. Chlorophyll b of wheat was slightly affected by Cu or Ni, but sharply dropped by Cd or Zn, the latter being more effective than the former.

Accordingly, chlorophyll a / b ratio of lupin-seedlings increased to an almost constant level by copper treatment, irrespective of its concentration in the soil and progressively increased by increasing Cd-level, whereas Ni or Zn caused a drop in such ratio, that was furthered by increased concentration of either elements. Cu slightly increased chlorophyll a/b ratio of wheat, but Zn was more effective, especially at 200 mg/kg soil. Either Cd or Ni reduced chlorophyll a/b ratio that was furthered with a rise of concentration of either metals (Table 3).

TABLE 3. Effect of different concentrations of various heavy metals on pigmentation (mg/1g leaf dry weight) of 4-weeks old lupin or wheat seedlings (mean of 5 replicates).

Element	Plant	Pigment	Concentration (mg / 1 kg soil)					L. S. D. at 5%
			0	50	100	200	400	
Copper	Lupin	Chl a	5.821	5.073	5.107	5.527	6.732	0.380
		Chl b	0.857	0.459	0.418	0.512	0.600	0.090
		Chl a/b	6.792	11.052	12.396	10.795	11.220	
		Total	6.678	6.532	5.519	7.039	7.332	
	Wheat	Chl a	12.663	12.187	12.899	13.100	11.957	1.390
		Chl b	4.432	4.122	4.088	4.025	3.689	0.750
		Chl a/b	2.857	2.957	3.155	3.255	3.241	
		Total	17.095	16.309	16.987	17.125	15.646	
Zinc	Lupin	Chl a	4.416	4.328	4.480	4.044	4.340	0.390
		Chl b	0.808	0.848	0.840	1.004	1.192	0.100
		Chl a/b	5.465	5.204	5.333	4.028	3.641	
		Total	5.224	5.176	5.320	5.048	5.532	
	Wheat	Chl a	13.656	14.444	13.016	8.192	4.728	0.28
		Chl b	5.125	3.995	2.505	1.150	0.985	0.280
		Chl a/b	2.665	3.616	5.196	7.123	4.800	
		Total	18.881	18.439	15.521	9.342	5.713	
Cadmium	Lupin	Chl a	5.284	5.508	5.692	5.722	6.810	0.320
		Chl b	0.970	0.539	0.564	0.341	0.286	0.160
		Chl a/b	5.447	10.219	10.092	16.783	23.811	
		Total	6.254	6.046	6.256	6.067	7.096	
	Wheat	Chl a	12.804	10.002	10.472	7.588	5.770	0.420
		Chl b	4.883	4.371	4.511	3.513	2.911	0.240
		Chl a/b	2.630	2.288	2.321	2.159	1.985	
		Total	17.737	14.373	14.983	11.092	8.688	
Nickel	Lupin	Chl a	3.978	3.680	3.238	3.443	2.153	0.390
		Chl b	0.939	0.723	0.644	0.681	0.550	0.070
		Chl a/b	4.236	5.090	5.028	5.026	3.915	
		Total	4.918	4.403	3.882	4.124	3.703	
	Wheat	Chl a	17.577	17.423	12.514	10.299	8.738	1.040
		Chl b	5.670	5.469	5.361	5.155	0.570	0.570
		Chl a/b	3.100	3.182	2.267	1.998	1.583	
		Total	23.247	22.892	17.515	15.454	14.259	

Table 4 shows that either Cd or Zn drastically reduced the total carbohydrate content of lupin, a response that continued and furthered by raising the level of these elements in the soil. Copper or nickel were less effective, since the drop in total or Zn, by an increased level of the metals. The total carbohydrate content of carbohydrates started at 100 mg /kg soil and continued, to a less extent than that of Cd, wheat was least sensitive to zinc and most sensitive to cadmium, whereas copper or nickel was moderately effective. Under all conditions, the drop in total carbohydrates progressivly increased by increasing level of either metals, except for Cu or Zn, where the drop in the wheat total carbohydrates remained unaffected by increased level of either elements.

TABLE 4. Effect of different concentrations of various heavy metals on the total carbohydrates of 4-weeks old lupin or wheat seedlings (mean of 5 replicates).

Element	Plant	Concentration (mg / 1 kg soil)					L. S. D. at 5%
		0	50	100	200	400	
Copper	Lupin	180.58	180.19	180.34	154.78	146.48	15.30
	Wheat	227.22	194.82	190.56	180.11	180.56	17.30
Zinc	Lupin	178.79	157.05	143.94	130.26	11.02	10.45
	Wheat	239.33	222.61	215.45	213.46	210.13	16.30
Cadmium	Lupin	187.01	166.67	143.56	119.26	118.71	12.80
	Wheat	217.22	195.37	215.00	178.33	168.52	15.90
Nickel	Lupin	187.13	186.66	179.63	153.16	146.85	13.80
	Wheat	241.11	216.85	211.85	207.60	185.74	11.90

Table 5 shows that either Cu or Zn slightly or hardly affected the total nitrogen of lupin shoots at 50 –100 mg / kg soil. Higher levels of either metals attenuated nitrogen-accumulation in this plant. Cadmium or nickel were almost equally suppressive to total nitrogen-accumulation in lupin; a response that was furthered with an increased level of this metal in the soil.

Table 6 shows that lupin nitrogen was least sensitive to zinc, since it dropped by application of 400 mg /kg soil, but most sensitive to Cd, since it dropped with increased Cd-level from 50 mg / kg soil on wards Ni or Cu were moderately suppressive to total nitrogen-accumulation, since the drop started at 200 mg/kg soil, but the former was more effective than the latter. Similarly, wheat nitrogen was most sensitive to Cd application and least sensitive to Ni, but moderately sensitive to Cu or Zn, the former being more effective than the latter.

TABLE 5. Effect of different concentrations of various heavy metals on the total nitrogen content (mg/1g dry shoot) of 4-weeks old lupin or wheat seedlings (mean of 5 replicates).

Element	Plant	Concentration (mg /kg soil)					L. S. D. at 5%
		0	50	100	200	400	
Copper	Lupin	13.42	14.21	12.64	11.33	11.01	1.11
	Wheat	9.43	8.25	6.23	6.15	5.43	0.68
Zinc	Lupin	13.88	13.85	13.31	12.15	10.43	1.06
	Wheat	9.83	9.05	8.12	7.51	6.13	0.79
Cadmium	Lupin	14.32	12.45	10.31	8.43	6.22	1.09
	Wheat	10.22	7.52	6.05	5.12	4.33	0.86
Nickel	Lupin	14.13	12.15	9.43	8.13	7.51	1.01
	Wheat	10.05	9.83	9.65	5.42	7.52	0.96

Lupin phosphorus was hardly affected by either levels of Cu or Cd, but most sensitive to Ni. In the meantime, Zn favored the accumulation of phosphorus up to 200 mg / kg soil above which phosphorus content dropped below the control level. Also, nickel slightly lowered phosphorus accumulation, whereas Cu or Cd slightly favored phosphorus accumulation that was hardly affected by an increased level of either metals. On the other hand, Zn remarkably attenuated phosphorus accumulation in wheat seedlings, a response that was partially alleviated by an increased level of the metal.

Lupin sodium was hardly affected by either levels of copper or zinc, but the drop in its content in the presence of nickel was hardly affected by concentration of either elements. In the meantime, the progressive increase in Lupin sodium with Cd level returned to control level at 400 mg / kg soil. Wheat sodium was slightly, if at all, affected by either levels of Cu or Zn, but it was very susceptible to nickel, where the drop in its content progressively increased by increasing level from 50 – 400 mg / kg soil. On the other hand, 50 or 100 mg / kg soil seemed without effect on wheat sodium content, but the higher level significantly increased this component. Lupin potassium was drastically lowered by an increased level of either Cd or Ni both of which seemed equally effective Copper seemed without effect, except for 400 mg / kg soil, where a slight drop in potassium content was observed. Zinc accelerated potassium accumulation to almost the same extent regardless the metal concentration.

TABLE 6. Effect of different concentrations of various heavy metals on the elements content (mg/1g d. wt. shoot) of 4-weeks old lupin or wheat seedlings (mean of 5 replicates).

Heavy metal	Plant	Element	Concentration (mg / 1 kg soil)					L.S.D. at 5%
			0	50	100	200	400	
Copper	Lupin	N	14.42	15.21	12.64	11.33	11.01	2.21
		P	6.88	8.14	7.75	8.01	8.40	1.72
		Na	44.14	48.44	52.42	43.40	41.65	8.73
		K	40.62	38.06	43.12	38.25	36.40	2.69
		Ca	24.00	16.13	17.22	17.43	14.87	2.34
		Mg	8.61	7.53	9.88	7.62	7.97	1.76
	Wheat	N	9.43	8.25	6.23	6.15	5.43	1.35
		P	11.92	19.55	19.65	18.84	16.23	3.26
		Na	7.76	6.06	7.05	5.42	5.21	N.S.
		K	81.61	59.81	56.06	47.83	46.59	3.37
		Ca	3.82	3.66	2.72	3.53	2.57	0.71
		Mg	4.40	3.68	3.83	6.86	3.88	1.28
Zinc	Lupin	N	13.88	13.85	13.40	12.15	10.43	2.12
		P	5.86	7.31	8.09	7.69	4.23	1.11
		Na	51.37	52.69	60.65	47.29	41.53	10.64
		K	44.73	59.31	60.03	64.13	55.34	6.08
		Ca	21.37	16.73	16.47	16.67	16.91	3.20
		Mg	8.43	7.22	9.46	8.75	7.00	0.95
	wheat	N	9.83	9.05	8.32	7.51	6.13	1.58
		P	11.14	6.59	7.94	9.62	11.62	2.37
		Na	9.39	10.59	11.44	7.48	8.78	2.49
		K	72.13	60.25	68.15	78.75	84.19	13.35
		Ca	4.31	4.71	4.72	6.49	6.31	1.16
		Mg	4.71	3.06	3.66	5.94	6.25	1.99
Cadmium	lupin	N	14.32	12.45	10.31	8.43	6.12	1.59
		P	6.05	5.03	6.45	6.24	7.25	1.82
		Na	40.99	45.97	49.65	56.89	44.62	4.79
		K	44.84	38.84	38.62	28.57	25.49	4.77
		Ca	23.45	19.66	19.67	20.37	16.58	2.45
		Mg	8.50	7.99	8.93	9.12	9.31	1.01
	wheat	N	10.22	7.53	6.05	6.12	4.33	1.73
		P	9.13	10.96	10.19	10.69	10.19	0.69
		Na	7.88	6.72	7.31	9.97	10.88	1.27
		K	87.72	65.00	62.62	66.69	55.32	4.81
		Ca	3.06	3.19	6.06	3.75	3.88	0.43
		Mg	4.19	3.75	4.25	4.15	4.81	0.38
Nickel	lupin	N	14.13	12.15	9.43	8.13	7.51	2.01
		P	5.76	3.82	4.23	4.28	4.23	0.49
		Na	47.44	42.63	42.63	44.28	42.78	2.16
		K	42.63	33.09	34.19	34.44	29.99	0.95
		Ca	20.94	19.13	18.94	17.00	15.81	2.62
		Mg	7.25	8.88	6.31	6.50	6.60	0.75
	Wheat	N	10.05	9.83	9.65	8.42	7.52	1.92
		P	11.15	12.31	12.31	12.02	11.15	2.63
		Na	6.06	6.69	5.79	4.67	4.03	0.72
		K	81.61	59.81	56.66	57.83	46.59	14.57
		Ca	6.37	6.37	7.12	7.63	7.56	0.86
		Mg	4.47	5.75	4.50	3.69	4.00	0.94

Lupin calcium was attenuated by all the test elements, copper being more effective and more prominently by an increased level of the elements in the soil. Copper lowered the calcium content of wheat, whereas Zn, Cd or Ni increased it particularly at the high doses of these elements. Lupin or wheat magnesium was invariably affected by either elements, irrespective of their concentration in the soil.

Discussion

The results of this investigation clearly indicate that characteristics of the tested heavy metals, their concentration and/or type of organ or the plant itself, play a significant role in the physiological responses towards application of these elements. At their moderate or high level, the four test elements hindered water uptake by the plants to different extents. Copper was more effective on lupin than wheat, whereas the reverse was the case regarding the other elements. Cu belongs to Group Ib, like zinc, yet it is less stable (lagging one or two electrons on the outer shell) and larger in size than zinc (ionic radius 0.93 and 0.88 A° , respectively).

It is interesting to note that neither of the test elements regulated in any appreciable way or change in dry weight of the roots of either plants, indicating that translocation to the root was immune for the deleterious effects, even with high levels of these elements. Still, the rapid translocation of the absorbed heavy metals to the shoot may play a significant role in this respect.

Zn (a stable element of Group Ib, of moderate ionic radius) was highly toxic to dry weight gain by lupin, whereas Ni (belonging to group VII, an unstable, highly reactive element with the smallest ionic radius and atomic weight) was least, if at all effective. In the meantime, it is logical that lupin responds more to Cu than Cd, for it is a less stable element, with a lower ionic volume. However, the high toxicity of the large-sized, stable element of moderate atomic volume (Zn) indicates the role of the biological barriers (*e.g.*, chelating agents) in this respect. Wheat (a monocot plant) differed in its response, being more sensitive to the highly reactive Ni with the smallest volume, as well as to the large-sized, highly stable Cd with the largest atomic weight, than to copper or zinc. This indicates the difference in attitude and metabolism between the types of plants.

One may recall that Yang *et al.* (1996) classified crop plants according to their sensitivity to cadmium into tolerant (wheat and rice) and non-tolerant (*Cucumis sativus* , *lactuca sativa* and *Phaseolus vulgaris*). They emphasized that the percentage of cadmium, combined with proteins in the Cd-tolerant crops, was lower than in the non-tolerant crops. Organic and Cd-hydrophosphates increased in the Cd-tolerant crops. In the crops less tolerant to cadmium, large molecular proteins were rich in cadmium.

Poongothi *et al.* (1998) proved that sorghum, grown with 0 – 7.5 ppm Zn or 0-50 ppm Cd, had an increased dry matter yield with increasing zinc rate and yield decreased with increasing Cd-level. Ewais (1998) observed that Cd or Ni inhibited shoot growth, but were less suppressive to roots of weeds.

The results also indicate that lupin leaves can tolerate the low or moderate levels of all the test elements, without affecting either chlorophyll a or b content. Even a large dose of these elements seemed without effect (Zn), or it increased (Cu or Cd) or decreased (Ni) this component.

Zinc and copper are cofactors for the enzymes, responsible for the photosynthetic system. Nickel, being the most active of the test elements, seemed to replace Mg or K during the biosynthesis of the chlorophyll a molecule. It seems that Zn activity was reserved for the transformation of chlorophyll a to chlorophyll b, whereas the other test elements were just suppressive.

On the other hand, Cu seemed without effect on chlorophyll a content of wheat, but the other elements were suppressive, Zn being most and Ni being least effective. This indicates that the biological barriers against these elements,except for Cu, were far less persent in wheat than in lupin. Still, copper seemed without effect on chlorophyll b, but nickel shared it in this respect. Zn and Cd were still inhibitory. This indicates that these two elements attack one and the same target during chlorophyll biosynthesis (probably δ-aminolevulinic acid synthetase or dehydrase. Cu seemed to have a minor effect on the mechanism of transformation of chlorophyll a to chlorophyll b, but nickel seemed to stimulate such process in both plants, coupled with Zn (lupin) or Cd (wheat).

In this connection, Gil *et al.* (1995) found that chlorophyll a showed similar trends to total chlorophyll. Chlorophyll a / chlorophyll b ratio increased with Cd concentration. Prasad (1996) reported that the net carbon dioxide uptake was affected more significantly through a decrease in chlorophyll content and through damage to the electron transport system. Chugh and Sawhney (2000) found that the rate of photosynthesis, chlorophyll content, activation of photosystem I and II , in lupin seedlings, decreased with increasing concentrations and/or exposure to Cd. Cadmium had a more pronounced effect on Ps II than on Ps I. Activity of photosynthetic enzymes showed far greater inhibition 12 days after treatment.

Concomitant with the drop in chlorophyll a and b and inspite of the differences in configuration and activity of the test elements, Cd and Zn were almost equally effective in reducing total carbohydrate content of lupin plants. The same applies to Cu and Ni,though to a less extent. Cd and Zn are equally stable elements, regardless of the other differences, whereas Cu and Ni are more reactive. In the meantime, Cd was most effective in lowering the total carbohydrate content of wheat, followed equally by Cu and Ni and least by Zn. This might reflect the difference in the role of these elements in the photosynthetic pathway and carbohydrate metabolism between the two plants.

Samarakoon and Rauses (1997) found that 2.0 mM Zn inhibited carbohydrate movement in *Phaseolus vulgaris*, leading to decreased carbohydrate concentration, which coincided with sucrose accumulation, though starch was reduced. They attributed the drop in carbohydrate content by heavy metals, such as Ni, to depression in autophotosynthesis.

Cadmium seemed to attack one common target during nitrogen assimlation, by either lupin or wheat. It is active most probably during the early stages of this process, forming stable complexes, particularly with enzymes. The same applies to copper though it was less effective than cadmium, at least by being a more active element, with lower atomic volume and molecular weight than cadmium. Equally effective was Ni on lupin and Zn on wheat. These two elements exchange place as the least effective on total nitrogen content of the test plants.

In this respect, Dahiya *et al.* (1994a) observed that soil amendment with 10 mg Cd/kg soil decreased dry matter yield of wheat, both in absence and presence of nitrogen fertilizer. Dahiya *et al.* (1994b) also, concluded that cadmium decreased uptake of N, P, K and Na. Singh and Antil (1996) concluded that nitrogen uptake was decreased by Zn application to wheat plants. Ni or Cu were moderately suppressive. Siesko *et al.* (1998) observed a change in protein synthesis, accompanied by depressed growth after a 21 days incubation of *Potamogeton pectinatus* with 1 mM $CdCl_2$. Ewais (1998) reported a change in root and shoot protein content of *Chaenopodium ambrosiodies* and *Digitaria sanguinalis* by the addition of Cd or Ni to the soil, the effect being less apparent on shoots.

The four elements mostly attenuated N, Na, K, Ca levels with minor effect on P or Mg. Singhania and Bajpai (1995) showed that Zn caused a significant increase in phosphorus content of rice grains and straw. Kuo and_Huang (1992) proved that Cd impaired the uptake of Ca, Mg and K in lettuce and soybean plants.

Conclusion

Plant needs minerals in small amounts , any increase in these quantities in the soil either by nature or man causes pollution of the soil and injure the plant, human and the animal .The threshold concentration of heavy metals varied with the element , its molecular weight and bioreactivity , also plant species could affect their response to the applied metal.

The four tested metals caused inhibition in growth and photosynthetic pigments for both lupin and wheat plants which increased with increasing the level of heavy metal in soil . At lower levels of applied metal , pigments proved that Wheat is more resistant than lupin . There was a remarkable decrease in total carbohydrate content of both plants , although some resistance shown by wheat as compared with lupin. In general, Cd and Ni were more toxic than Cu and Zn.

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مدى استجابة نباتي الترمس والقمح لتلوث التربة بالعناصر المعدنية الثقيلة

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تهدف الدراسة إلى خصائص العناصر التي تحت البحث طبقا لتركيزاتها و/أو نوع النبات المستخدم (ذوات الفلقة الواحدة – ذوات الفلقتين) تلعب دورا واضحا في الاستجابة الفسيولوجية لإضافة تلك العناصر.

تم زراعة نباتي الترمس (ذوات الفلقتين) ونبات القمح (ذوات الفلقة الواحدة) في تربة ملوثة تحتوي على تركيزات ٥٠ ، ١٠٠، ٢٠٠ ، ٤٠٠ ملجرام/كم تربة لكل من عناصر النحاس – الزنك – الكادميوم أو النيكل لمدة أربعة أسابيع.

وجد أن العناصر المستخدمة لم تؤثر على زيادة الوزن الجاف لجذور كلا النباتين.أظهر الزنك أعلى زيادة في التأثير على الون الجاف لنبات الترمس بينما كان النيكل أقلهم تأثيرا. اختلفت استجابة نبات القمح للتلوث بالعناصر فكان أكثر حساسية للعناصر ذات الأحجام الصغيرة كالنيكل (ذو النشاط العالي والوزن الجزئي المخفض) وأيضا للعناصر ذات الأحجام الكبيرة كالكادميوم (ذو الثبات العالي والوزن الجزئي المرتفع) عن عنصري النحاس والزنك.

تحملت أوراق نبات الترمس التركيزات المخفضة والمتوسطة لكل العناصر التي تحت الدراسة دون تأثير محتوى كلورفيل أ أو ب.لم يؤثر عنصر النحاس على محتوى كلوروفيل أ –– بينما العناصر الأخرى كانت مثبطة حيث كان عنصر الزنك أعلاهم أما النيكل فكان أقلهم تأثيرا. بالنسبة لكلوروفيل ب لم يظهر عنصرا النحاس والنيكل أي تأثير بينما كان عنصرا الزنك والكادميوم مثبطان لكلوروفيل ب كما أثرا على تقليل المحتوى الكربوهيدراتي الكلي لنبات الترمس وكانا متساويان في ثباتهم العنصري بينما أظهر عنصرا النحاس والنيكل أعلى نشاطا. أما بالنسبة لنبات القمح فقد أظهر عنصر الكادميوم تاثير واضح في تقليل محتوى الكربوهيدرات الكلي ثم تلاه عنصري النحاس والنيكل ذوي التأثير المتساوي ثم عنصر الزنك ذو التأثير الأقل.

أثر عنصرا الكادميوم والنحاس على التمثيل النيتروجيني لكل من نباتي الترمس والقمح. حثت العناصر الأربعة التي في الدراسة غالبا على تراكم مستويات النيتروجين والصوديوم والبوتاسيوم والكالسيوم بينما كانت ذات تأثير ضئيل على تراكم كل من عنصري الفوسفور أو الماغنيسيوم.