



NICKEL, CO, CR, and MN METALS UPTAKE BY WHEAT AND GRASS PLANTS FROM SERPENTINITIC SOILS OF PENJWIN, KURDISTAN REGION- IRAQ

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ABSTRACT: Metal rich soils derived mainly from ultramafic (i.e. serpentinitic soils) are wide spread over Penjwin (Mlakawa) area. Three serpentinitic (S1, S2, S3) with one non-serpentinitic soils (S4) were used and these soils contain high concentrations of heavy metals such as Ni, Cr, Co and Mn. Results showed that total metal concentration ranged between 192.0 -1714.8 mg kg⁻¹, 289.0 -1264.2 mg kg⁻¹, 128.8-177.6 mg kg⁻¹ and 4125-4795 mg kg⁻¹ respectively, while after harvesting plants (wheat and grass) in the biological pots (CRD) experiment. The concentration of the total and potentially available metals Ni, Cr, Co and Mn decreased in the grass and wheat soil pots. But the CaCl₂-extractable and water-soluble metals increased in soils after harvesting the wheat and grass except for Co which decreased. In wheat and grass plants (wheat *Triticum aestivum* and canarygrass *Phalaris canariensis*), the uptake of Ni and Cr metals for soil S1 were above the normal range and the values were 14.47 mg Ni pot⁻¹ and 6.6 mg Cr pot⁻¹. The concentrations of both Ni and Cr metals were found to be at the toxic level. While Ni in other soil samples was observed within the normal range and tolerance limits, but Cr, in the other soil samples, was above the tolerance level and was slightly less than toxic level. Wheat could take up highest amounts of toxic metals when grown in serpentine soils. The study showed that none of plants (wheat and grass) were identified as hyperaccumulator.

Keywords: Nickel, metals, Wheat, Grass plants, Serpentinitic soils

INTRODUCTION

The Penjwin igneous complex represents an ophiolite sequence within the larger Zagros belt. It is a northwest-southeast trending elongated body within the Iraqi territories (Fig.1). The remnant parts were located within adjacent Iranian territories [20]. Serpentinitic soils in Penjwin area (Mlakawa village) have covered 35 km² and a wide range of these areas are used for cultivated plants such as wheat, barley, tomato, onion and other natural wild plants which are used for animal feeding. Soils developed from serpentinite rocks generally host distinctive vegetation, which is often stunted and interspersed with bare patches. These features have been attributed to the chemical and physical properties of serpentinitic soils. Plant growth is limited by many factors, including low nutrient levels, Ca deficiency, Mg toxicity and high concentrations of potentially toxic element, such as Cr, Ni and Co [2, 3, 29]. However, the factors controlling serpentine flora and vegetation differ from site to site (Proctor and Nagy, 1992). Plants grown on soils that developed from serpentine substrata can be divided into 'normal' and metal hyper accumulators [28]. Most plants grown on serpentinitic soils are of the former type and show only slightly elevated Ni concentrations in the shoot dry matter (about 5–100 μg g⁻¹) in comparison to those for other soil types (0.5–10 μg g⁻¹) [28]. While most serpentine plants are able to grow on these soils without excess uptake of elements, hyperaccumulator plants take up more than 1000 μg Ni g⁻¹ into their leaf dry matter [3]. The aim of investigations are to study the forms of heavy metals such as total, potentially available, CaCl₂-extractable, AB-DTPA-extractable and water-soluble fraction for Ni, Cr, Co and Mn in soils before and after harvesting (wheat and grass) grown in serpentinitic and non-serpentine soils. Also, to determine the total metals taken up by cultivated plants such as wheat and grass crops.

MATREIAL AND METHODS

Four soil samples were collected from Penjwin area at the depth 0 – 30 cm, which based on the difference in their Ni content. Soil samples (S1,S2 and S3) were taken from different positions of (high, medium and low) Ni contents, respectively (Fig.1). Soil samples, S1, S2 and S3 were developed from serpentinitic rock, while S4 was developed from non-serpentine rocks and it had been developed from clay stones as shown in Table (1). Composite soil samples were air dried and passed through a 2.0 mm sieve. Soil samples were stored in plastic bags prior to analyses. Some chemical properties such as (pH, CEC, ECe, OM, N, P, K, Ca and Mg) and soil texture were determined. The total metal determined by using 1:3 HCl: HNO₃ (Aqua Riga solution), bioavailable metal, Soluble metal and potentially available metal were determined by using CaCl₂, Distell water and dilute HNO₃, respectively.

Table 1: Latitude and Longitude of the soil sampling positions.

Location	Latitude (North)	Longitude (East)	Elevation(m)	Geological information
S1	3536'22.7"	4554'7.17"	1398	Peridotite parent rock.
S2	3536'20.9"	4555'01.2"	1338	Banded Gabbro&Diorite parent rock
S3	3535'9.94"	4555'13.0"	1337	Course Gabbro parent rock
S4	3535'8.66"	4554'8.20"	1299	Qulqula Group parent rock

Soil samples from the Penjwin area were collected from the surface horizons 0-25 cm and sieved to pass a 4 mm sieve. A 250 g of clean gravel (8mm diameter) placed in the bottom of each 4000 cm³ pot and filled with 3000 g of the sieved soil. Fifty (50) seeds from two different plants; wheat (*Triticum aestivum*) and canarygrass (*Phalaris canariensis*) were placed on the surface of the soil in circular pattern, covered with a thin layer of soil and irrigated with tap water to 75% of soil water holding capacity. The pot experiment included 4 soil samples (S1, S2, S3 and S4), using factorial complete randomized design (CRD) with 3 replicates. 10 days after germination, the plants were fertilized (1.49 g N pot⁻¹, 1.5g P pot⁻¹ and 1.5 g K pot⁻¹) and then they were thinned to 20 plants per pot after 16 days.

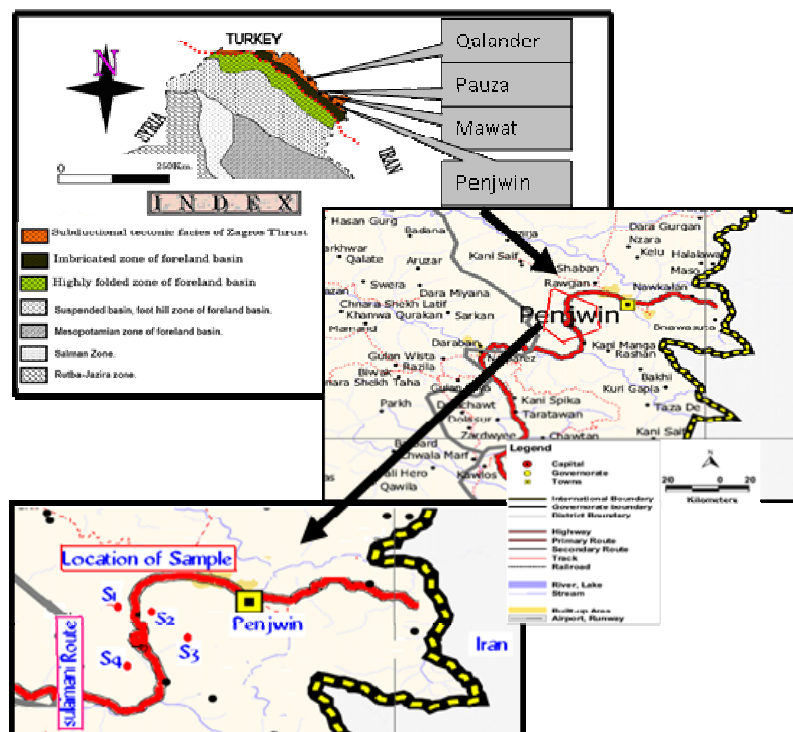


Figure 1: Sample location map.

At a maximum growth stage, the plants were harvested at 11 weeks after germination of wheat and grass plants were harvested at 13 weeks after germination. Plant shoots and roots were separated and dried at 65°C for 24-48 h, after they had been washed with tap water to clean the soils from the samples. The plant samples were weighed and milled by electrical miller and stored in the plastic containers for further analyses. Dried plant materials were digested using 1:1(H₂SO₄, H₂O₂). Statistical analyses were conducted using XL-STAT (version 7.5). Analysis of variance (ANOVA) was used to test the main The Least Significant Difference (LSD) test was done to find the significant differences among treatment at level (0.05).

RESULTS AND DISCUSSION

The soils S1, S2, and S3 developed from serpentinite rocks were low in clay content, while soil developed from non-serpentinite rocks (S4) were high in clay content Table (2). The CEC values for serpentinitic soils ranged from 11.04 to 18.71 cmol_c kg⁻¹ soil and the values were lower than the non-serpentinitic soil which was 41.72 cmol_c kg⁻¹ soil. This may be due to the low clay and organic matter contents, while the clay content in soil (S4) was higher. The serpentinitic soils had very low organic matter content which could be due to lack of vegetation. Also, the type of clay minerals may differ between the soils due to variation in the parent material which were rich in antigorite and chlorite [18]. On the other hand, the pH values which was more than 7.0 and ranged between 7.41 and 7.71. The sodium content and Ece were very low and ranged from 18 to 24 mg kg⁻¹, and from 0.45 to 0.68 dS m⁻¹, respectively. This was in agreement with the low Na content. Calcium and Mg content was higher than the Na and K content, in serpentinitic soils, and Mg content were higher than Ca. The Ca ion ranged between 220 to 350 mg kg⁻¹, while Mg ranged between 436 to 715 mg kg⁻¹. This was expected that the soils derived from serpentinitic rocks with high in Mg low in Ca and poor in available N and P. The S1 soil had been derived from prediotite while the S2 had been derived from gabbros [25].

The concentrations of water-soluble, CaCl₂-extractable, potentially available and total Ni, Cr, Co and Mn in the soil samples are shown in Table (3). Generally, these soils contain large amounts of total Ni, Cr, Co and Mn. The total concentration of Ni, Cr, Co and Mn in the soils derived from serpentinite rocks showed different results and the highest concentration of Ni, Cr and Co were in the soil sample S1. The serpentinitic soil samples (S1, S2 and S3) had different content of Ni, Cr, Co and Mn concentrations which reflect the variation of these metals in parent materials [28]. The serpentinitic soil weathered from prediotite and other ultramafic rocks contain more than 70% ferromagnesian silicate (mafic) minerals (Kurekeberg, 1984). Since the soil S1 was developed from the prediotite rocks, they were high in heavy metals such as Ni, Cr, Co and Mn and low in Ca (Brady *et al.*, 2005). And that was clearly shown in the results in Table (3). On the other hand, serpentine in location S2 is weathered from gabbro and diorite while serpentine in location S3 is weathered from cores gabbro [5].

These results were in agreement with the results reported by Wilson, [35]; Hunter and Horenstein, [10, 14] indicating that the content of Ni, Cr, Co and Mn were lower in the gabbro in comparison with prediotite.

Table 2: Chemical and physical properties of studied soils before planting.

Soil samples	pH	Ece dSm ⁻¹	Ca	Mg	Na	K	P	N	OM*	Sand	Silt	Clay	CEC
			mg kg ⁻¹										cmol _c kg ⁻¹
S1	7.71	0.45	320	715	18.2	10.6	5.11	300	0.1	751	223	26	18.71
S2	7.49	0.56	350	696	20.4	14.4	4.97	200	0.7	591	321	86	16.79
S3	7.41	0.54	235	540	22.6	0.87	0.79	600	0.1	531	397	72	11.04
S4	7.45	0.65	220	437	24.9	17.4	3.58	100	6.2	311	251	33	41.72

The results in Table (3) indicate that the amount of these metals in S2 and S3 which derived from gabbro had a lower metal content than the soil S1 which is derived from prediotite. Soil S4 which is developed from non-serpentinite contained higher Ni and Co content than soil S3 and it was 281.6 mg Ni kg⁻¹ soil and 139.0 mg Co kg⁻¹ soil. This might be due to the contamination from the serpentine.

The potentially available fractions of different heavy metals are shown in Table (3) indicating that Mn was the highest concentration than others metals in serpentine soils. The highest value of metal concentration were found in soil S1, and lower value of Ni and Cr were found in Soil S2 and lower value of Co and Mn were found in soil S3 (Table.3). The results of potentially available metal nearly reflect with total metal content in soil. The concentration of extractable Ni, Cr, Co and Mn extracted with CaCl_2 are shown in Table (3) the results indicated that the amount of metals extracted by CaCl_2 were much lower compared to amount extracted by both total and acid extraction. A dilute CaCl_2 -extract is considered to be comparable to soil solutions. The majority of the mean metals were recovered in CaCl_2 were similar to water soluble fractions. The results in (Fig.4) indicate that all the released metal mean values in CaCl_2 or water soluble were less than 0.5% of total extracted amount except for Co which was slightly more than 0.5%. But Mn metal had the lowest recovery percentages which was less than 0.1%. This implies that low amounts of exchangeable metals maybe present in the forms of oxide and/or oxyhydroxide which were extracted by acids.

Table (3): The Total, potentially available, CaCl_2 and Water -soluble extractable fraction of selected metals in the initial soil

Soil Sample	Different form of metals	Initial			
		Ni	Cr	Co	Mn
S1	Total-aqua riga	1714.8	1264.2	177.6	4675.0
	Potentially available HNO_3	276.0	73.5	124.0	553.0
	CaCl_2 -extractable	1.1	1.3	2.0	1.2
	Water -extractable	1.5	0.6	1.6	0.4
S2	Total-aqua riga	382.0	880.4	133.4	4295.0
	Potentially available HNO_3	37.5	42.5	91.5	110.0
	CaCl_2 -extractable	1.9	0.5	2.0	1.0
	Water -extractable	1.1	1.3	1.2	0.6
S3	Total-aqua riga	192.6	326.8	128.8	4125.0
	Potentially available HNO_3	55.0	66.0	87.0	57.0
	CaCl_2 -extractable	1.7	1.2	1.6	0.9
	Water -extractable	1.5	0.8	1.5	0.4
S4	Total-aqua riga	281.6	289.6	139.0	4795.0
	Potentially available HNO_3	57.0	64.0	90.5	456.5
	CaCl_2 -extractable	1.4	1.6	0.8	0.9
	Water -extractable	1.3	1.1	1.6	0.7

The results in Table (4) showed that the mean uptake values varied between the metals in soils and plants. Generally, a comparison between the metals indicates that the highest amount of uptake was for Mn in both wheat and grass plants, while the lowest uptake was Co metal. The results showed that the mean Mn uptake was higher in wheat than in grass, and roots had higher Mn uptake than shoots in both wheat and grass for all soils. Also, the plant uptake Mn was slightly higher in plants grown in soils developed from serpentinite than non-serpentine soils. The results Mn analysis in Table (4) showed that the highest significant ($p < 0.05$) of Mn content was ($28.26 \text{ mg pot}^{-1}$) in the wheat roots.

This value was below the critical level according to Kabata-Pendias and Pendias [16] the normal concentration level of Mn ranged from 30 to 300 mg kg^{-1} plant. So, it neither affects the plant growth nor would cause metal plant toxicity. In spite of that the soils were rather high in Mn concentration than the other metals (Table.3). The results in Table (4) show that the uptake values of Ni metal ranged from 2.00 to 14.47 mg pot^{-1} and from 0.62 to 2.47 mg pot^{-1} for wheat shoots and roots respectively, while in grass shoots and roots, it ranged from 1.51 to 2.06 mg pot^{-1} and from 0.54 to 1.34 mg pot^{-1} , respectively.

The highest mean uptake value for Ni in wheat and grass grown in the soils was found in soil S1 and it was much higher than the other soils, but there was similarity in the mean Ni uptake in the other soils S2, S3 and S4. This is also reflected in the variation in the initial metal content in the soils (Table.3).The highest significant accumulation values of Ni (14.47 mg pot⁻¹) in wheat shoots were observed in serpentine soil S1. According to WHO [34] the accumulation of Ni in wheat crop should not exceed (0.05-5 mg kg⁻¹). The results indicated that the concentration of Ni in the wheat grown in soil S1 was higher than the other soils.On the other hand, the same trend was found for Cr and the result in Table (4) showed that the highest significant value of Cr was (6.60 mg pot⁻¹) which found in wheat shoots in soil S1. According to Kabata-Pendias and Pendias (2001) the normal values of Cr in plants were ranged from 0.1 to 0.5 mg kg⁻¹, and the excessive or toxic values ranged from 5 to 30 mg kg⁻¹. The results of present study (Table 4) showed that the lower concentration of Cr which taken up by wheat and grass were more than the normal range in plant. Since Cr was a non essential element to plants and is known to be toxic metal.The Co metal taken up by plant in wheat and grass was nearly similar and the results showed that the wheat and grass taken up the Co from soil solution in the similar ranges, the higher accumulation of Co in shoots rather than roots (Fig.5). The data analyses (Table.4) showed that the highest significant (p<0.05) uptake of Co value (1.91 mg pot⁻¹) in the grass shoots from soil sample S4, which is developed from non-serpentine and had a higher available fraction of Co, may be contaminated from serpentine soils.

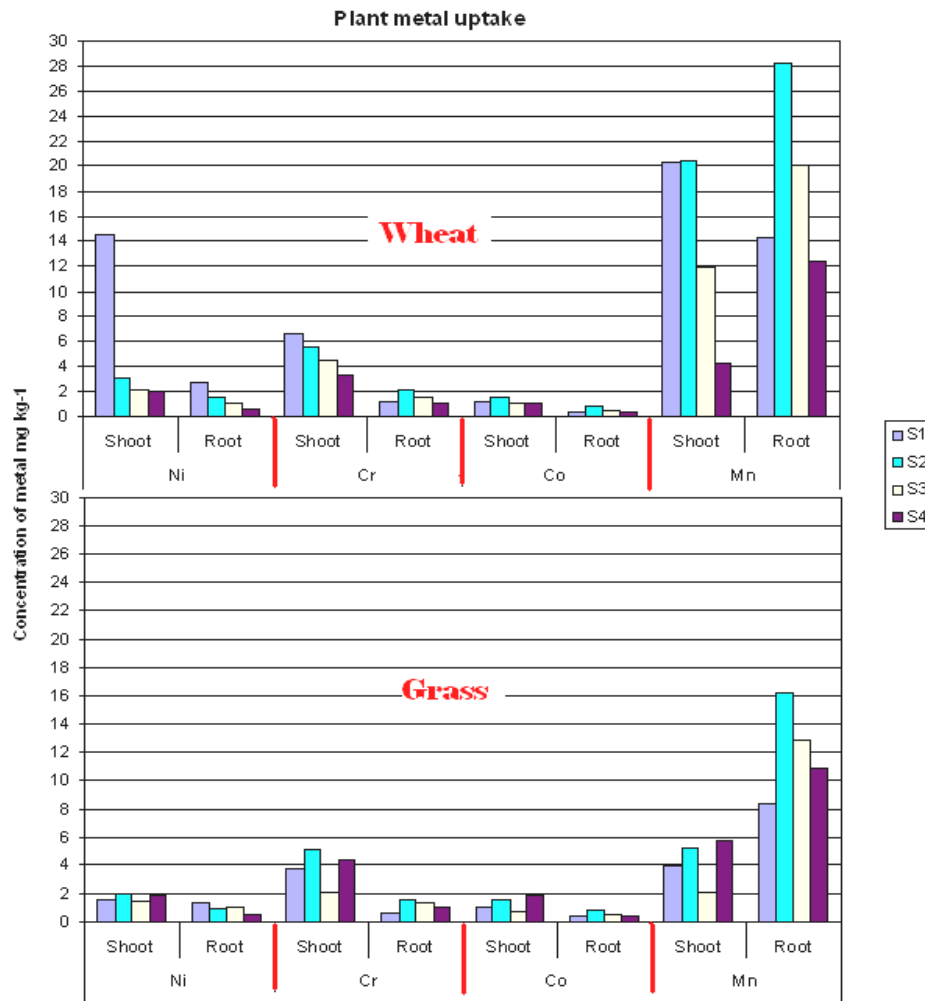


Figure (2): Plants (wheat and grass) metal uptake and accumulation in shoots and roots.

Table 4: Plant metals concentration and uptake in (Shoot and Root) from different soil samples.

Crop	S1		S2		S3		S4		
	shoot	Root	shoot	Root	shoot	Root	shoot	Root	
	Concentration mg kg ⁻¹								
Wheat	Ni	295.13	317.93	72.67	95.47	63.67	86.47	80.20	84.67
	Cr	134.53	148.87	134.07	134.67	134.27	134.40	133.87	134.93
	Co	23.20	59.60	35.20	49.53	30.87	45.13	43.27	49.73
	Mn	414.67	1836.67	471.00	1713.33	361.33	1720.00	170.33	1686.67
	uptake								
	Ni	14.47	2.47	3.15	1.57	2.13	1.01	2.00	0.62
	Cr	6.60	1.16	5.81	2.22	4.50	1.57	3.33	0.99
	Co	1.14	0.46	1.53	0.82	1.03	0.53	1.08	0.36
	Mn	20.33	14.30	20.43	28.26	12.11	20.06	4.24	12.31
	Dry weight kg / pot								
	0.049	0.008	0.043	0.016	0.034	0.012	0.025	0.007	
Concentration mg kg ⁻¹									
Grass	Ni	61.73	276.33	62.27	91.00	74.13	83.50	71.73	84.07
	Cr	150.73	135.33	154.40	156.73	158.80	169.90	162.73	165.07
	Co	39.62	85.87	47.13	81.33	55.80	69.40	69.87	74.80
	Mn	156.00	1726.67	157.00	1656.67	163.00	1610.00	213.33	1646.67
	Uptake								
	Ni	1.55	1.34	2.06	0.89	1.51	1.00	1.96	0.56
	Cr	3.79	0.66	5.11	1.54	3.23	2.03	4.44	1.09
	Co	1.00	0.42	1.56	0.80	1.13	0.83	1.91a	0.49
	Mn	3.93	8.37	5.19	16.24	3.31	19.22	5.82	10.89
	Dry weight kg / pot								
	0.025	0.005	0.033	0.010	0.020	0.012	0.027	0.007	

Generally, this result indicated that Co uptake by grass plants was more than wheat. [12] found that the grass was able to exude a class of organic compounds termed as siderophore (mugineic and arnic) capable of enhancing the availability of trace elements through the decreases of pH in the soil, Which in turn leads to Co uptake by plant and then accumulation in shoots increases. The concentration of Co taken up by wheat and grass in present study was between the normal ranges according to [16] (0.02-1 mg kg⁻¹), only with the exception of the Co uptake by grass shoots (1.91 mg pot⁻¹) which was more than the normal range, but still it was far less than the excessive or toxic (15-50 mg kg⁻¹) level [16]. It has been found that Co concentration in wheat plants ranged from 0.03 to 0.27 mg kg⁻¹ [15, 23, 7]. Also, Sillanpää and Jansson [32] reported that the average concentration in wheat plants was 0.11 mg kg⁻¹. The results in Table (4) show that Co taken up by wheat and grass was more than that published by other investigators, simply because Co was taken up by plant passively and it is one of the essential elements for plant growth at normal level, whereas at high level of Co, plants are known to have phytotoxicity effects on plant [17].

The results of total, potentially available (HNO₃-extractable), CaCl₂-extractable and water soluble metals (Ni, Cr, Co and Mn) after harvesting wheat and grass plants are shown in Fig (2). After harvesting plants, the water-soluble and CaCl₂ -extractable Ni, Cr and Mn increased in all soil samples, however Co decreased in all soil samples. Also, the results indicated that total and potentially available metals decreased after harvesting wheat and grass.

The results in Table (5) show that the percent values of the reduction in total and HNO₃-extractable metals in soils after harvesting were very close, and it was higher in HNO₃-extractable than the total reduction. But the reduction percentage was very low for Mn HNO₃-extractable compared with other metals. Also, the reduction percentage was lower in soil after harvesting grass compared to wheat.

On the other hand, the lowest reduction percentage was in total Co for both wheat and grass. But the water-soluble and CaCl₂-extractable metals increased in soils after harvesting for both crops. This may be due to the effect of root acid exudates such as (malice, citric and oxalic acid...etc.), which may decrease the rhizospheres pH and increase the availability of the metals [23]. This is even clearer in Table (5), the highest reduction is in potential metal fractions.

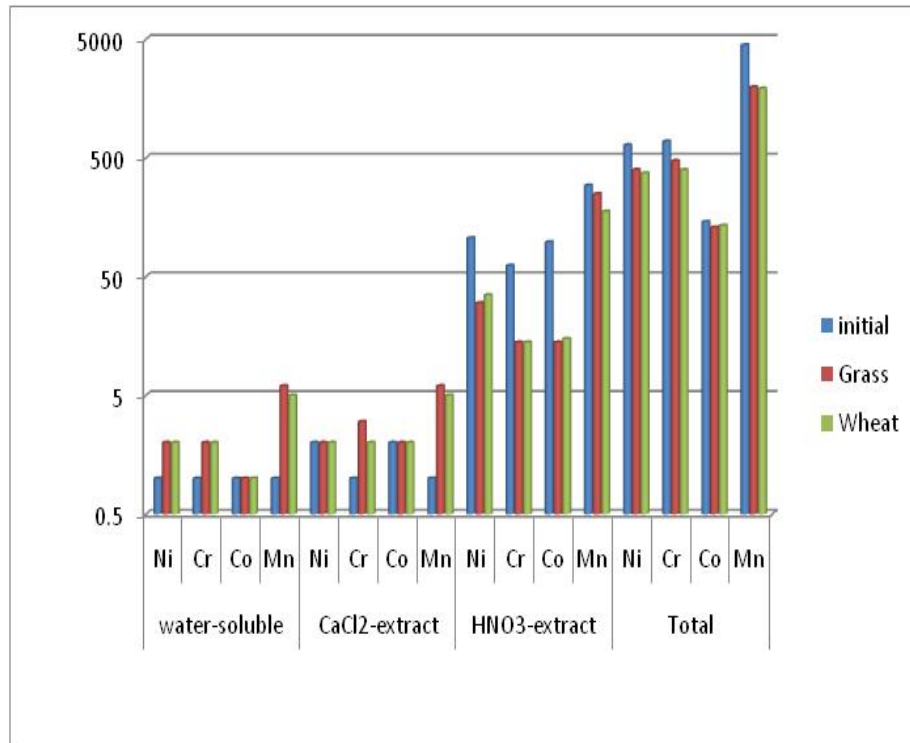


Fig 2: Soils extractable amounts of metal fractions between initial and after harvesting grass and wheat.

Table 5: Percent of the reduction in total and HNO₃-extractable metals in soils after harvesting.

Soils	HNO ₃ -extract				Total			
	Ni	Cr	Co	Mn	Ni	Cr	Co	Mn
Grass pots % reduction	72	78	85	15	38	31	10	56
Wheat pots % reduction	67	77	85	40	42	43	7	57

The correlations between initial metals in soils and after harvesting plants are shown in Table (6). The results indicated that there were not significant ($p > 0.05$) correlation between concentration of metals initial soils and after harvesting grass and wheat for potentially available Ni, Cr and Mn, except Ni which was significant at $p < 0.01$ in grass only. But the relationship between initial potentially available Co and after harvesting grass and wheat was positively significant ($p < 0.01$). For the total Ni, Cr, Co and Mn the results indicated that there were significant ($p < 0.05$) correlation between initial metal concentrations and after harvesting wheat and grass. Except for Cr which was not significantly ($p > 0.05$) correlated in wheat only.

The correlation between water-soluble or CaCl₂-extractable metals from initial soils and after harvesting wheat and grass were negatively not significant ($p > 0.05$). This variations may reflect the presence of these metals in different minerals and different solubilities. While results showed that the total and potentially available metals were identified as significant sources of metals in serpentine soils.

Table 6: Correlations coefficient between initial metal in soil and after harvesting plants

metals	Correlations coefficient							
	soluble		CaCl ₂ -extractable		HNO ₃ -extractable		Total-Aqua regia	
	Grass	Wheat	Grass	Wheat	Grass	Wheat	Grass	Wheat
Ni	-0.94	0.31	-0.28	0.72	0.99**	0.85	0.99**	0.99**
Cr	-0.62	-0.24	-0.51	-0.70	0.55	0.51	0.98*	0.83
Co	-0.02	0.41	0.73	0.42	0.99***	0.99***	0.99**	0.99*
Mn	-0.77	-0.39	0.14	-0.09	0.61	0.43	0.98*	0.99**

* Significant value of metal concentration at 0.05 levels

** Significant value of metal concentration at 0.01 levels

*** Significant value of metal concentration at 0.001 levels

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