# EVALUATION OF VARIATION IN SOIL AND FORAGE MICRO-MINERAL CONCENTRATIONS IN A SEMIARID REGION OF PAKISTAN

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## Abstract

An investigation was conducted to evaluate the micro-mineral status of pasture having high population of small ruminants in Punjab, Pakistan. Soil and forage samples were collected fortnightly for two seasons. It was found that sampling period affected soil  $Cu^{2+}$ ,  $Zn^{2+}$  and  $Se^{2+}$  while all forage minerals except  $Se^{2+}$  were affected by sampling times. Seasonal effects were observed in soil  $Fe^{2+}$ ,  $Mn^{2+}$  and  $Se^{2+}$ , and forage  $Cu^{2+}$ ,  $Fe^{2+}$ ,  $Zn^{2+}$ ,  $Mn^{2+}$  and  $Se^{2+}$ . All soil mineral levels except  $Co^{2+}$  and  $Se^{2+}$  were found to be above the critical levels and likely to be adequate for normal growth of plants growing therein, whereas soil  $Co^{2+}$  and  $Se^{2+}$  were in severe deficient levels during both seasons for the normal plant growth. The levels of  $Fe^{2+}$ ,  $Zn^{2+}$ ,  $Co^{2+}$ , and  $Se^{2+}$  in soil were higher, whereas those of  $Cu^{2+}$  and  $Mn^{2+}$  were lower during winter than those during summer. Forages contained marginal deficient level of  $Co^{2+}$  during winter, those of  $Cu^{2+}$  and  $Se^{2+}$  during the summer. Moderate deficient levels of  $Fe^{2+}$ ,  $Zn^{2+}$ ,  $Mn^{2+}$  and  $Se^{2+}$  during the summer. Forage  $Co^{2+}$ ,  $Fe^{2+}$ ,  $Zn^{2+}$ ,  $Mn^{2+}$  and  $Se^{2+}$  during the summer. Forage the summer. Forage  $Co^{2+}$  and severe deficient level of  $Zn^{2+}$ ,  $Mn^{2+}$  and  $Co^{2+}$  were found during the summer. Forage  $Co^{2+}$ ,  $Fe^{2+}$ ,  $Zn^{2+}$ ,  $Mn^{2+}$  and  $Se^{2+}$  during winter were found to be adequate for the requirements of ruminants. Consequently, grazing animals at this location need continued mineral supplementation of these elements to prevent diseases caused by nutrient deficiency, and to support optimum animal productivity.

## Introduction

Under pasture systems, animals depend on forages to satisfy all of their nutritional requirements. Unfortunately, forages often do not provide all of the needed minerals, which animal require throughout the year. Many incidences of mineral inadequacies in forages and soils have been reported which are principal causes of reproduction failure and low production rate (McDowell, 1985; McDowell *et al.*, 1993; Vargas & McDowell, 1997). Mineral deficiencies likely to affect production of grazing livestock at pasture in most of the regions of the world include those of the major elements Ca, P, Mg, Na, S, and the trace elements Co, Cu, I, Mn, Se, and Zn (Little, 1982; Judson *et al.*, 1987; Judson and McFarlane, 1998).

Excessive intakes of minerals can also commonly have an adverse effect on animal health, the more commonly encountered problems have been associated with excessive intake of the minerals Cu, Mo, Fe, S, Na, K, and F. Signs of mineral disorders are often non-specific and in cases of marginal deficiencies may go unnoticed by the stock owners. The interpretation of such sign is also difficult if more than one mineral is deficient or the deficiency is associated with other disorders such as increased burdens of gastrointestinal parasites, especially since trace element deficiencies may increase the susceptibility of animals to disease (Suttle & Jones, 1989).

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The nutrition of grazing animals is a complicated interaction of soil, plant, and animal. Seasonal variability can markedly affect the dietary intake of minerals as a result of changes in composition, stage of growth and availability of pasture and to changes in the moisture content of the soil (Hannam *et al.*, 1980; Smith & Longeran, 1997). Pasture and soil tests are not perceived to be the initial tools for diagnosis of animal deficiencies. If pasture samples are taken in association with the animal samples an explanation of the predisposing pasture conditions may be assessed and subsequent routine plant analyses may be able to predict the variable incidence of the mineral problems. The advantage of this is the ease and comparative lower cost of plant tissue analysis when compared with the collection and testing of blood or tissue samples from animals.

The purpose of this investigation was to examine the potential for soil and plant analysis as indicators of likely mineral deficiencies or excesses of grazing livestock during different seasons.

#### **Materials and Methods**

The investigation was conducted at the Livestock Experimental Station Rakh Khaire Wala, district Leiah, in the province of Punjab, Pakistan. The ranch comprises 400 ha and receives annual precipitation of 250-750 mm restricted to July and August. The soils are sandy and vertisols. The ranch has about 7000 animals of which 2000 are breeding sheeps. The vegetation of the ranch consists of a variety of native and improved forages ranging from grasses, legumes, tree leaves and crop wastes available for grazing animals.

Soil and forage samples were collected eight times during the year (4 times during both the winter and summer seasons). Five composite, each of soil and forage, samples from the pasture assigned to the experiment were collected after each sampling period during each season. Each composite sample of soil or forage was derived from three sub-samples. Soil samples were obtained using a stainless steel sampling auger to a depth of 15 cm. The sub-samples of forages were collected from an area approximately 70 cm in diameter, and cut to a length of 3-6 cm to simulate grazing height. These samples were taken from the same area from which the soil samples were taken. Forage samples were cut using a stainless steel knife and placed in clean cloth bags on the site. Both the soil and forage samples were dried in an oven at 60  $^{\circ}$ C for 48 h and subsequently ground, using a Wiley mill, with a 1 mm stainless steel sieve (forage) or 2 mm sieve for soil. Ground samples were stored in plastic whirlpack sample bags until analysis. Soil minerals were extracted using Mehlich-1 method  $(0.05M \text{ HCl} + 0.0125M \text{ H}_2\text{SO}_4)$  following Rhue & Kidder (1983), while forage samples were prepared and digested according to the procedures of Fick et al., (1979).  $Se^{2+}$  analyses of soil and forages were achieved by fluorometry (Whetter & Ullrey, 1978), while Fe<sup>2+</sup>, Cu<sup>2+</sup>, Zn<sup>2+</sup> and Mn<sup>2+</sup> concentrations were determined by atomic absorption spectrophotometry on a Perkin-Elmer AAS-5000 (Anon., 1980). An atomic absorption spectrophotometer with graphite furnace and Zeeman background corrector was used to determine soils and forage Co concentrations.

The data were analyzed using a split-plot design (Steel & Torrie, 1980). Differences among means were ranked using Duncan's New Multiple Range Test (Duncan, 1955).

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### **Results and Discussion**

#### Soil

Soil  $Cu^{2+}$  was affected significantly by the sampling periods, but the effect of seasons on soil  $Cu^{2+}$  was non-significant (Table 1). The differences among the fortnights were non-consistent during both winter and summer seasons (Table 2). However, an increase in  $Cu^{2+}$  content at the 2<sup>nd</sup> and 3<sup>rd</sup> fortnights during both seasons was found.

In the present study, soil  $Cu^{2+}$  did not differ significantly during both seasons, but it was above the critical level (Rhue & Kidder, 1983) for the normal plant growth. The mean soil  $Cu^{2+}$  concentrations observed in the present study were similar to the values reported by Tejada *et al.*, (1985) and lower than those reported by Tiffany *et al.*, (2001). Copper availability to plants seems to be affected by soil pH. Aubert & Pinta (1977) and Sanders & Bloomfield (1980) suggested that available  $Cu^{2+}$  decreases with increase in soil pH. At higher pH,  $Cu^{2+}$  adhere to soil components and thus it may have led to decrease in  $Cu^{2+}$  in soil solution as cupric ions, which is the available form for plants. Soil  $Cu^{2+}$  availability is also related to soil organic matter. Kabata-Pendias & Pendias (1992) reported that  $Cu^{2+}$  binding capacity of any soil and  $Cu^{2+}$  solubility are highly dependent to the amount and kind of organic matter. In this study, the copper levels above critical value indicate the presence of higher organic matter in the soil.

Significant seasonal and non-significant sampling period effects were found on soil  $Fe^{2+}$  concentration (Table 1). The reduction in soil  $Fe^{2+}$  at all fortnights during winter progressed with time. In contrast, during summer the reduction in soil  $Fe^{2+}$  was observed only at the last fortnight (Table 2). Overall, soil  $Fe^{2+}$  was significantly higher in winter than that in summer.

In this study, mean soil Fe<sup>2+</sup> concentration was different during both seasons. In both seasons Fe<sup>2+</sup> values were generally high compared to the critical level (Vieks & Lindsay, 1977). Adequate soil Fe<sup>2+</sup> values were also reported by Mooso (1982) and Merkel *et al.*, (1990) from Florida. These results may support the report of McDowell *et al.*, (1984) in which it was indicated that Fe<sup>2+</sup> deficiency is rare in grazing animals due to generally adequate content in soils and forages. However, Becker *et al.*, (1965) reported Fe<sup>2+</sup> deficiency in animals grazing on sandy soils in Florida. Similar seasonal trends in soil Fe<sup>2+</sup> fluctuation were also reported in some other studies (Tejada *et al.*, 1987; Prabowo *et al.*, 1991).

There was no seasonal effect on soil  $Zn^{2+}$  concentration, whereas the effect of sampling period was found to be significant (Table 1). Soil  $Zn^{2+}$  was found to be maximum at the 2<sup>nd</sup> fortnight during winter and at the 3<sup>rd</sup> fortnight during summer, whereas at the remaining fortnights the soil  $Zn^{2+}$  did not differ significantly (Table 2).

In the present study, soil  $Zn^{2+}$  contents across all samples during both seasons were almost similar, and these values of soil  $Zn^{2+}$  were above the critical level for normal plant growth (Rhue & Kidder, 1983). Similar values above critical levels have already been reported by Prabowo *et al.*, (1991) in Indonesia and Tiffany *et al.*, (2001) in North Florida. Slightly higher soil  $Zn^{2+}$  in winter than in summer as found in this study is in agreement with the findings of Velasquez-Periera *et al.*, (1997). In contrast, Pastrana *et al.*, (1991) found higher soil  $Zn^{2+}$  concentration in summer than that in winter. Extractable  $Zn^{2+}$  has been found to be affected by low pH and cultivation (Aubert & Pinta, 1977).  $Zn^{2+}$  may be more soluble and susceptible to leaching in low pH soils and high rainfall areas.

There was a significant effect of seasons but non-significant of fortnights on soil  $Mn^{2+}$  level (Table 1). The soil contained higher level of  $Mn^{2+}$  during summer than that during winter. A sharp decrease from fortnight 1 to fortnight 2 followed by an increase at both fortnights 3 and 4 was found during winter. In contrast, during summer, an increase up to fortnight 2 was observed followed by a consistent lag phase up to the last fortnight (Table 2).

The minimum dietary  $Mn^{2+}$  requirements of ruminants are not precisely known, but likely they range between 15-25 mg/kg for animals (Anon., 1980, 1990).  $Mn^{2+}$  requirements are substantially lower for growth than for optimal reproductive performance, and these are increased by high intakes of Ca and P. Although rarely reported for tropical regions, clinical signs suggesting  $Mn^{2+}$  deficiency have been observed in certain regions including Mato Grosso, Brazil (Mendes, 1977).

In the present investigation, soil  $Mn^{2+}$  levels across all samples were found to be high in summer and were above the critical level as suggested by Rhue & Kidder (1983). Similar  $Mn^{2+}$  levels and seasonal variation have earlier been reported by Tejada *et al.*, (1985) in Guatemala. Manganese is known for its rapid oxidation and reduction under variable soil environments. Oxidizing conditions may reduce  $Mn^{2+}$  availability, and reducing conditions may increase its availability (Kabata-Pendias & Pendias, 1992). When  $Mn^{2+}$  is reduced, its susceptibility to leaching increases. A significant difference was observed in forage  $Mn^{2+}$  level in different seasons. Forage  $Mn^{2+}$  was above the requirement of livestock during winter and below the required level during summer, although extractable soil  $Mn^{2+}$  was higher during summer. Similar trend in forage  $Mn^{2+}$  with respect to seasons had already been reported (Pastrana *et al.*, 1991; Velasquez-Pereira *et al.*, 1997).

Soil  $Co^{2+}$  concentration was not affected significantly by seasons or sampling periods (Table 1). A high elevation in soil  $Co^{2+}$  was observed at fortnight 1 during winter, which thereafter decreased and remained so up to the last fortnight, whereas during summer the soil  $Co^{2+}$  concentrations remained statistically unchanged throughout the season (Table 2).

In the present study, mean extractable soil  $\text{Co}^{2+}$  concentration was deficient in view of the critical level of 0.1 mg/kg (Kubota, 1968). These soil  $\text{Co}^{2+}$  values were not adequate compared to the requirement of plant growth. Similar low level of soil  $\text{Co}^{2+}$  has earlier been reported (McDowell *et al.*, 1989).

Analysis of variance of the data showed that both seasons and fortnights had a significant effect on soil  $Se^{2+}$  concentration (Table 1). Higher  $Se^{2+}$  was present in the soil during winter than that during summer. There was a consistent decrease in soil  $Se^{2+}$  with time in winter, whereas in summer, a change in soil  $Se^{2+}$  was not consistent (Table 2).

The present study showed that soil  $Se^{2+}$  content was very low and below the requirement for plant growth during both seasons. Similar levels of soil  $Se^{2+}$  had already been reported (Merkel *et al.*, 1990). In contrast, higher soil  $Se^{2+}$  levels were reported by Rojas *et al.*, (1993) and lower by Pastrana *et al.*, (1991). McDowell *et al.*, (1989) reported seasonal fluctuations in soil  $Se^{2+}$  levels, higher in dry season and lower in wet season quite parallel to what was observed in the present study. The mobility of  $Se^{2+}$  in soil depends upon soil pH, oxidation potential, organic carbon, calcium carbonate and cation exchange capacity (Banueles & Schrale, 1989).

Source of	Degree of	Mean squares						
variation	freedom	Cu <sup>2+</sup>	Fe <sup>2+</sup>	Zn <sup>2+</sup>	Mn <sup>2+</sup>	Co <sup>2+</sup>	Se <sup>2+</sup>	
Season (S)	1	0.0003ns	877.60**	3.06ns	285.23*	0.000004ns	0.007***	
Error	28	0.497	25.24	1.75	37.10	0.0003	0.00004	
Fortnight (FN)	3	1.58*	55.90ns	5.59*	10.70ns	0.00005ns	0.0007***	
S x FN	3	0.29ns	37.40ns	1.70**	88.69*	0.00008***	0.0003***	
Error	84	0.33	46.67	0.49	15.75	0.00004	0.00003	

 
 Table 1. Analysis of variance of data for mineral concentrations in soil at different (sampling periods) fortnights during the winter and summer seasons at sheep ranch.

\*, \*\*, \*\*\* = significant at 0.05, 0.01, and 0.001 levels respectively, ns = non-significant

Table 2. Micro-mineral concentrations of soil by season and sampling period at sheep ranch
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Element		Sampling Periods						
mg/kg	Season	т	п	тт	IV	Seasonal		
ing/kg		1	п		1,	means		
Cu <sup>2+</sup>	Winter	3.90+0.152	5.38+0.343	2.17+0.412	3.88+035	3.069		
	Summer	3.64+0.447	3.68+0.471	$2.47 \pm 0.23$	5.27+0.165	4.094		
Fe <sup>2+</sup>	Winter	72.30+2.09	52.80+4.89	58.00 + 2.40	42.00+5.44	58.45		
	Summer	51.60+3.93	50.60+2.18	30.20+2.52	42.00 + 2.44	48.85		
$\mathbf{Zn}^{2+}$	Winter	b	а	b	b	5.59		
		$6.29 \pm 0.202$	6.18+0592	$5.18 \pm 0.258$	6.06 + 0.340			
	Summer	b	b	b	b	4.28		
		$4.18 \pm 0.140$	5.70 + 0.581	$6.62 \pm 0.186$	$5.98 \pm 0.185$			
Mn <sup>2+</sup>	Winter	ab	С	bc	bc	61.65		
		72.80+3.48	57.60+2.14	57.80+2.27	59.40+2.09			
	Summer	ab	а	ab	ab	65.50		
		65.60 + 2.14	69.20+1.86	64.20+1.86	67.00+1.67			
Co <sup>2+</sup>	Winter	А	b	b	b	0.029		
		0.334 + 0.001	0.0280 + 0.001	0.0256 + 0.001	0.0282 + 0.001			
			3	6				
	Summer	b	b	b	b	0.028		
		0.0254 + 0.001	0.0246 + 0.002	0.0260 + 0.001	0.0298 + 0.002			
		6		6				
Se <sup>2+</sup>	Winter	a	b	b	с	0.079		
		0.0894 + 0.001	0.0788 + 0.002	0.0786 + 0.001	0.0698 + 0.002			
		9	7	4				
	Summer	de	d	de	e	0.058		
		0.0582 + 0.001	0.0562 + 0.001	0.0582 + 0.002	0.0540 + 0.002			
		7	7	4	3			

Means with the same letters do not differ significantly at  $P \le 0.05$  and Means are based on following number of samples of: soil (60) during each season

## Forage

Data for the different forage species were pooled within each season to assess the influence of fortnights and seasons, which was found to be significant on forage  $Cu^{2+}$  concentration (Table 3). During winter the lowest forage  $Cu^{2+}$  level was found at the last fortnight, whereas at the earlier three fortnights the difference in  $Cu^{2+}$  was not significant. In contrast, during summer, a consistent and statistically non-significant decrease in forage  $Cu^{2+}$  with time was observed. Overall, forage  $Cu^{2+}$  concentration was markedly higher in winter than that in summer (Table 4).

In the present study, forage  $Cu^{2+}$  level differed significantly during winter and summer. In winter, it was considerably high in the forage plants as compared to that in summer. The forage  $Cu^{2+}$  level in forage during winter was within the range of animal's requirement, but was on the borderline requirement during summer (Reuter & Robinson, 1997). Similar low forage  $Cu^{2+}$  levels have already been reported in Nigeria (Ogebe & McDowell, 1998; Ogebe *et al.*, 1995) and North Florida (Tiffany *et al.*, 2001), in Guatemala (Tejada *et al.*, 1987), Florida (Espinoza *et al.*, 1991) and Venezuela (Rojas *et al.*, 1993).  $Cu^{2+}$  deficiency can arise when high intakes of Mo and S occur, coupled with normal copper intakes (Underwood, 1981).

Seasonal and fortnights effects were found to be significant on forage  $Fe^{2+}$  levels (Table 3). Forage  $Fe^{2+}$  concentration was markedly higher in winter than that in summer. A slight reduction in forage  $Fe^{2+}$  level was observed with time during both seasons (Table 4).

Forage  $Fe^{2^+}$  concentration varied significantly by seasonal influence, and it was greater than the requirement of ruminants in winter and deficient in summer. Forage iron was higher during summer and lower during winter. Espinoza *et al.*, (1991) found variation in forage  $Fe^{2^+}$  concentration and higher percentage of  $Fe^{2^+}$  deficient samples in a study conducted in Florida. Vargas *et al.*, (1984) and Tejada *et al.*, (1987) did not find  $Fe^{2^+}$  deficient forage samples in Colombia and Guatemala, respectively. The absorption of  $Fe^{2^+}$  by plants is not always consistent and is affected by the physiological state of the plant, as well as changing condition of soil and climate (Kabata-Pendias & Pendias, 1992). The generally low forage  $Fe^{2^+}$  found in summer is in disagreement with the normal soil value found. It may be due to the type of forages deficient in iron below the requirements of animals. Feed  $Fe^{2^+}$  concentrations showed seasonal effect, being higher in winter than that in summer as reported in the results. Water  $Fe^{2^+}$  level was almost the same during both seasons and both feed and water  $Fe^{2^+}$  concentration seemed to complement the forage  $Fe^{2^+}$  level required by the ruminants.

Both seasons and sampling time had significant effects in changing the forage  $Zn^{2+}$  concentration (Table 3). Forage  $Zn^{2+}$  level was markedly higher during winter than that during summer. A decreasing trend in forage  $Zn^{2+}$  level was observed with time during both seasons (Table 4).

Forage  $Zn^{2+}$  concentration showed seasonal variation, with high concentration during the winter season and above the requirement of ruminants, but in summer all samples were deficient or below the critical level (McDowell *et al.*, 1993). Similar seasonal differences in forage  $Zn^{2+}$  levels were reported by Velasquez-Pereira *et al.*, (1997) in Nicaragua. Forage  $Zn^{2+}$  varied considerably depending on various ecosystem, characteristics, plant species, and stage of maturity. However, Kabata-Pendias & Pendias (1992) reported that  $Zn^{2+}$  concentration of certain forages from different countries do not differ significantly.

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Source	Degree	Mean squares						
of variation	of freedom	Cu <sup>2+</sup>	Fe <sup>2+</sup>	Zn <sup>2+</sup>	Mn <sup>2+</sup>	Co <sup>2+</sup>	Se <sup>2+</sup>	
Season (S)	1	3573.74 ***	394490.00 ***	18250.16 ***	38160.32 ***	0.027***	0.027***	
Error	28	14.67	49.69	27.16	26.07	0.0003	0.001	
Fortnigt (FN)	3	71.11***	424.37***	272.62**	173.99***	0.008***	0.001ns	
S x FN	3	19.23**	7.27ns	45.36**	49.13**	0.004**	0.0007ns	
Error	84	3.73	6.92	5.78	8.22	0.001	0.0004	

 

 Table 3. Analysis of variance of data for mineral concentrations in forages at different (sampling period) fortnights during winter and summer seasons at sheep ranch.

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\*, \*\*, \*\*\* = significant at 0.05, 0.01, and 0.001 levels respectively.

ns = non-significant.

Table 4. Micro-mineral concentrations of forages by seasons and sampling period at sheep ranch.

Floment		Sampling Periods						
mg/kg	Season	Ι	п	III	IV	Seasonal means		
	¥¥724	а	b	b	с			
C 2+	winter	29.44+1.35	33.72+1.46	28.72+1.61	17.30+1.39	25.63		
Cu	<b>G</b>	d	e	e	e			
	Summer	12.00+0.71	8.20+0.36	6.56+1.60	4.48+0.12	7.81		
Fe <sup>2+</sup>	Winter	16.60 + 2.32	195.20+1.85	155.00 + 1.84	134.20+2.58	161.25		
	Summer	35.20+1.16	48.20+1.28	32.00+1.41	16.60+1.66	28.25		
Zn <sup>2+</sup>	Winter	а	b	b	с			
		68.64+1.50	65.24+1.63	59.16+0.91	46.06+2.21	59.53		
	Summer	d	e	e	e			
		18.80 + 1.66	14.80 + 0.86	14.80 + 0.66	12.20 + 1.28	15.65		
Mn <sup>2+</sup>	Winton	а	b	b	с			
	winter	77.38+1.82	71.78+1.65	69.90+1.19	64.28+2.28	67.59		
	Summer	d	d	d	d			
		14.00+1.36	12.80 + 0.86	15.00+0.71	10.40 + 0.75	17.00		
$C a^{2+}$	Winton	а	d	a	a			
	winter	0.1440 + 0.0136	0.1306 + 0.0174	0.1290 + 0.0116	0.1260 + 0.0093	0.107		
Co	Summer	b	cd	bc	cd			
		0.0760 + 0.0051	0.0380 + 0.0051	0.0570 + 0.0037	0.0380 + 0.0020	0.068		
a 2+	Winter	0.1120 + 0.0086	0.1000 + 0.0071	0.1120 + 0.0086	0.0774 + 0.0173	0.098		
Se <sup>2+</sup>	Summer	0.0500+0.0071	0.0540+0.0093	0.0540+0.0068	0.2460+0.0051	0.051		

Means with the same letters do not differ significantly at  $P \le 0.05$  and Means are based on following number of samples: of forage (60) during each season

There are some controversial reports on  $Zn^{2+}$  concentration in plants at the adult stage. For example; Underwood (1981) reported that as plants mature, their  $Zn^{2+}$  concentration decreases. In contrast, high concentration of  $Zn^{2+}$  has been found in old leaves of plants (Kabata-Pendias & Pendias, 1992).

A significant effect of seasons and fortnights was found on forage  $Mn^{2+}$  concentration (Table 3). High amount of  $Mn^{2+}$  was found in forage species sampled during winter than those during summer. A consistent decrease in forage  $Mn^{2+}$  was observed with time of sampling during winter, whereas during summer no change in  $Mn^{2+}$  was observed with time (Table 4).

Several studies indicated a high tolerance of  $Mn^{2+}$  in ruminants (Hansard, 1983). Mineral imbalance typified by excess of Fe<sup>2+</sup> and Mn<sup>2+</sup> may interfere with metabolism of other minerals (Lebdosoekojo *et al.*, 1980). Feed contained higher concentration of  $Mn^{2+}$  in summer and lower in winter, but was within the range of requirement of sheeps. While, water  $Mn^{2+}$  level was found to have no seasonal effect. The  $Mn^{2+}$ content in forage was not sufficient except during winter, although in feed it is within the required range of ruminants during both seasons. Non-consistent relationships between soil and forage  $Mn^{2+}$  were found as was already observed by Tejada *et al.* (1985, 1987) in Guatemala.

Mean squares from the analysis of variance of the data for forage  $\text{Co}^{2+}$  concentration revealed that both seasons and sampling periods had a significant effect on  $\text{Co}^{2+}$ concentration in forage (Table 3). Higher values of forage  $\text{Co}^{2+}$  were found in winter than that in summer. During winter, the forage  $\text{Co}^{2+}$  level remained almost uniform except at the fortnight 2 where a very sharp depression in  $\text{Co}^{2+}$  concentration was recorded (Table 4). During summer, no consistent pattern of increase or decrease in forage  $\text{Co}^{2+}$  with time was found.

Forage  $Co^{2+}$  levels were deficient for ruminants during both seasons, because these were lower than the critical level (Anon., 1980). Similar  $Co^{2+}$  deficient forages were found in Nicaragua by Velasquez-Pereira *et al.*, (1997), in Florida, USA Espinoza *et al.*, (1991). Rojas *et al.*, (1993) found marginal to deficient  $Co^{2+}$  level. Tejada *et al.*, (1987) did not find differences in forage  $Co^{2+}$  concentrations among different regions in Guatemala, but the forage  $Co^{2+}$  level was higher than the critical values and also than the values reported in this work. It was observed in this study that forage  $Co^{2+}$  was deficient during both seasons, but was slightly higher than that in soil. Mtimuni (1982) suggested that there is readily available  $Co^{2+}$  in soil for plant growth even on  $Co^{2+}$  deficient soil. Similarly, Reid & Horvath (1980) illustrated that the level of  $Co^{2+}$  in the soil does not necessarily indicate its availability to plants.

 $\text{Co}^{2+}$  is often the most severe mineral deficiency of grazing livestock with the possible exception of P and Cu (McDowell *et al.*, 1984).  $\text{Co}^{2+}$  uptake by plants is dependent on  $\text{Co}^{2+}$  and  $\text{Mn}^{+2}$  concentration in soils. High soil  $\text{Mn}^{2+}$  depresses uptake of  $\text{Co}^{2+}$  in forages. In the present study, high levels of Mn were found in soil, which could have led to reduce  $\text{Co}^{2+}$  absorption by plants and subsequently, low levels in plant tissues. According to McKenzie (1967, 1975) the soils with high level of manganese oxide strongly bind free soil  $\text{Co}^{2+}$  to their surfaces leading to low availability of  $\text{Co}^{2+}$  to plants.

There was a significant influence of seasons on accumulation of  $Se^{2+}$  in forage species, while the sampling periods remained ineffective in affecting the forage  $Se^{2+}$  level (Table 3). Generally, forage  $Se^{2+}$  level was higher in winter than that in summer (Table 4).

Forage  $Se^{2+}$  was on borderline requirement of ruminants in summer, but it was higher in winter. Under central Florida conditions, similar values of forage  $Se^{2+}$  have earlier been reported by Espinoza *et al.*, (1991). In view of Pope *et al.*, (1979) increasing the concentration of S in the forage has a detrimental effect on  $Se^{2+}$  availability. A similar trend in the forage  $Se^{2+}$  levels was reported by Tejada *et al.*, (1987) in Guatemala. Gerloff (1992) reported that  $Se^{2+}$  concentration in plants is positively correlated with soil pH. Other factors affecting the  $Se^{2+}$  uptake are soil P, S and N concentrations. Among crops brassicas and legumes contain higher  $Se^{2+}$  than the other crops. It has been suggested that forage crops containing more than 0.1 mg/kg will protect livestock from  $Se^{2+}$  deficiency disorders (Gupta & Subhas, 2000). The low forage  $Se^{2+}$  found in this study was related to low  $Se^{2+}$  contents in the respective soil. According to Ammerman & Millar (1975) the  $Se^{2+}$  concentration of herbage generally reflects the status of the soil. The soluble  $Se^{2+}$  content in the soil showed a similar tendency to that of forage in this study.

Based on soil and forage analyses it is concluded that copper, selenium, zinc, manganese and cobalt deficiencies may be limiting sheep productions in this semi-arid region of Punjab, Pakistan. Therefore, supplementation studies are needed to determine the need and economic benefit of trace minerals supplementation.

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(Received for publication 13 April 2005)