Abstract

The study was aimed to investigate magnesium level in soil samples, forages grown in that soil and plasma of grazers (goat and sheep) taken from District Bhakkar, Mianwali and Sargodha. The present study was carried out during 2016. The findings unveiled that the mean magnesium concentrations in soil of District Sargodha, Mianwali and Bhakkar varied from 40.49 to 50.14, 48.83 to 54.53 and 54 to 59.68 magnesium/kg sequentially. The highest Magnesium content was found in Bhakkar soil. The mean concentrations of Magnesium in forage samples were found between 32.75 - 39.13, 40.24 - 42.24 and 49.55 - 50.35 mg/kg for Sargodha, Mianwali and Bhakhar, independently. The average Magnesium contents in the blood plasma of goats fed on these forages were between 31.4 - 34.79 magnesium/L in Sargodha, 29.93 - 33.19 magnesium/L in Mianwali, and 20.76 - 30.85 mg/L from Bhakkar. In sheep blood samples, Magnesium levels in Sargodha, Mianwali and Bhakhar extended from 25.81 to 32.727, 25.14 to 31.43 and 15.93 to 17.76 mg/L, respectively, indicating that there is no need for magnesium supplementation for small grazers of this area.

Keywords: Blood; Forage; Goat; Magnesium; Metals; Sheep

Introduction

The health, growth and production rate of livestock rely on quality of forages [1, 2]. Forages are source of essential nutrients for grazing livestock. Plants also possess nutritional requirements for their growth that are meant to be delivered by soil [3]. The results of minerals deficiency in soils are retarded growth of plants and lower yields [4]. The level of minerals impacts on soil pH which ultimately affects plants [5, 6]. Mineral elements are required by the ruminants to fulfill the metabolic and growth need of the body. Forages contain adequate macro and microelements to meet these requirements [7, 8]. There is no information about the effect of season on the natural pastures and its mineral content, protein and fiber components. The adequacy and availability of mineral
element from pastures affects the health and performance of grazing livestock. The forage required by the goat and sheep must be replete with various nutrient so that it can help maintain better health of ruminants [9]. The extent to which the nutrients of the forages are available for livestock depends upon the quantity and bioavailability of nutrient found in the forage. It is often noticed that the nutrient found in the forage does not enter the animal body due to some reasons [10]. Magnesium, being involved in several metabolic pathways, is pivotal to the metabolism of plants [11, 12]. The chlorophyll molecules contain Magnesium in the center which plays a significant role in the formation of different product such as fats, carbohydrates and its products and play role in enzymes functioning in forages. The deficiency of Magnesium may cause the chlorotic patches and yellow color appears in the vein of older leaves and leaves become whirled. To overcome the deficiency of metal, its foliar spray prepared in water is applied to the plants [13, 14]. This research was aimed to find out the Magnesium residues in forages, soil samples and blood plasma of ruminants (Goat and Sheep) to evaluate the bioaccumulation and the relation of the well-being of animals to ingestion of forages having the mineral (Figure 1).

![Graphical Abstract](image)

**Figure 1. Graphical Abstract**

**Materials and methods**

Bhakkar, Mianwali and Sargodha districts were selected for the present investigation. Samples were taken from 10 sites in each district. These samples were thoroughly mixed to get composite samples in triplicate. Soil samples were taken with stainless-steel auger, 1.0 to 1.5 ft deep and stored in plastic bag. Forages such as Bajra (*Pennisetum glaucum*), Barsem (*Trifolium alexanderium*) and Oat (*Avena sativa*) were collected and sun-dried. From jugular vein, blood samples of five goats and sheep of each site were taken in heparinized tubes; plasma separated and was frozen at -20°C. Soil and forages samples were sun dried and then kept in the oven to dry for three days at 72°C. After drying, these samples (1g each) and blood plasma (1mL each) were digested by following standard wet digestion procedure [15] and then diluted to 50 mL and filtered. Samples were stored in tagged bottles for further process. To evaluate the magnesium concentration samples, Atomic Absorption Spectrophotometer (Model No. AA-6300,
Shimadzu, Japan) was deployed [16]. Data were statistically processed using Minitab 16 software. One-way ANOVA (Analysis of Variance) and LSD (Least Significant Difference) were used as advised by Steel and Torrie [17].

Pollution Load Index (PLI) was measured by the formula proposed by Liu et al. [18].

\[ PLI = \frac{Metal\ contamination\ in\ soil\ under\ investigation}{Reference\ value\ of\ metal\ in\ soil} \]

Bio-concentration factor (BCF) was determined following Cui et al. [19].

\[ BCF = \frac{Concentration\ of\ metals\ in\ forages}{Concentration\ of\ metals\ in\ soil} \]

\[ BCF = \frac{Concentration\ of\ metals\ in\ blood\ plasma}{Concentration\ of\ metals\ in\ forages} \]

**Results and Discussion**

**Soil**

The non-significant effect of sites on Magnesium content in soil was observed (Table 1). The results revealed that Magnesium levels in soil were between 40.49-50.14 mg/kg in Sargodha District, 48.83-54.53 mg/kg in Mianwali, and 54-59.68 mg/kg in the soils of Bhakkar District (Figure 2). The critical limit of 9.10 mg/kg for Magnesium content was lower than the soil samples [20]. In the current work, soil Magnesium level was much lower than previously examined results [21]. This soil needs Magnesium containing fertilizers to enhance the plant growth and development [22].

**Forages**

The results showed that Magnesium content in forages was non-significantly (p<0.05) affected by sites (Table 1). Results revealed that Magnesium levels in forages were between 32.75-39.13 mg/kg in Sargodha District, 40.24-42.24 mg/kg in Mianwali, and 49.55-50.35 mg/kg in the forages of Bhakkar District (Figure 3). The lower Magnesium content in forage samples of Sargodha could be attributed to lower Magnesium assimilation capability of forages that might have affected the metal transfer in animals. Soil type, plant age, climate, plants species and soil pH can affect metal ions uptake [23].

**Blood of Goat and Sheep**

The non-significant effect of sites on Magnesium content in the blood of goats and sheep was revealed by analysis of variance of the collected data (Table 1). The results revealed that Magnesium levels in the blood of goats were between 31.4-34.79 mg/L in Sargodha District, 29.93-33.19 mg/L in Mianwali, and 20.76-30.85 mg/L in the blood of goats in Bhakkar District (Figure 4). Magnesium levels in blood of sheep were between 25.81-32.727 mg/L in Sargodha District, 25.14-31.43 mg/L in Mianwali, and 15.93-17.76 mg/L in the blood of sheep in Bhakkar District (Figure 5). Magnesium values obtained in the present investigation were similar to those reported by Prabowo et al. [24] and Meschy [25]. According to NRC [26], for small animals average blood Magnesium values ranged from 1.8 to 3.5 mg/L and the goat’s blood samples from all the three districts were above the safe limits. In the current research, the highest Magnesium levels were found in sheep blood samples taken from Sargodha and the lowest was found in those of Bhakkar district. Magnesium level in plasma samples of Sargodha and Mianwali was greater than the critical value (20 mg/L) suggested by Herd [27] but the values for Bhakkar district were within safe limits. Current research values for Magnesium were higher than those found by Stojković et al. [28].

**Correlation**

A significant correlation was noticed between soil-forage, between soil-blood plasma of selected grazers in Districts Sargodha and Mianwali. A significant correlation was found between forage-blood plasma and negative correlation between soil-forages and soil-blood plasma of goat and sheep in Bhakkar (Table 2).
Bio-concentration factor (BCF)

BCF of magnesium in forage was greater in Bhakkar and Mianwali as compared to Sargodha. The lowest BCF was observed in Sargodha while the maximum BCF was seen in Bhakkar. BCF of blood plasma of goats in Sargodha was greater as compared to that of Bhakkar. Similarly, BCF of blood plasma of sheep of Sargodha and Mianwali sampling was greater as compared to Bhakkar (Table 3). Bio-concentration factor serves as a pivotal element for determination of exposure to heavy metals moving along the food chain. The values of BCF > 1 indicate the capacity of plant for metal accumulation while BCF <1 conveys the idea of metal absorption without accumulation [29]. BCF for magnesium at Bhakkar, Sargodha and Mianwali sites was < 1.

Pollution Load Index (PLI)

The PLI for magnesium was higher in Bhakkar soil samples as compared to Sargodha and Mianwali. The pollution level or contamination factor of magnesium was >1 and the soil is designated as contaminated (Table 4). The degree of pollution can be assessed using PLI. Soil is considered to be polluted if PLI>1, while the soil is considered to be non-contaminated if PLI<1 [30]. The pollution level of magnesium greater than 1 indicated contamination of soil.

Table 1. ANOVA for Magnesium concentrations in soil, forage and blood Plasma

| Magnesium | Sargodha | Mianwali | Bhakkar |
|-----------|----------|----------|---------|
| Soil      | 88.284 ns| 2.231 ns | 14.225 ns|
| Forage    | 38.492 ns| 2.306 ns | .481 ns  |
| B.G.      | 8.977 ns | 9.228 ns | 78.889 ns|
| B.S.      | 37.262 ns| 32.427 ns| 2.788 ns |
| Degree of freedom | 2 | Error | 8 |

B.G= Blood of Goats, B.S= Blood of Sheep

Table 2. Correlation of Magnesium between soil-forage and Forage-Blood of goat and sheep

| Soil  | Soil-Forages | Soil-B.G | Soil-B.S | Forage-B.G | Forage-B.S |
|-------|--------------|----------|----------|------------|------------|
| Sargodha | 1.000** | .961 | 720 | .962 | 718 |
| Mianwali | .709 | .872 | .990 | .274 | .606 |
| Bhakkar | -.860 | -.708 | -.989 | .248 | .926 |

B.G= Blood of Goats, B.S= Blood of Sheep

Table 3. Bio-concentration of Magnesium in districts Sargodha, Bhakkar and Mianwali

| BFC of Magnesium | Sites | Sargodha | Mianwali | Bhakkar |
|------------------|-------|----------|----------|---------|
| Soil-Forages     | 1     | 0.781    | 0.814    | 0.917   |
|                  | 2     | 0.809    | 0.836    | 0.837   |
|                  | 3     | 0.7804   | 0.835    | 0.894   |
| Forages-Blood of Goat | 1     | 0.868    | 0.816    | 0.622   |
|                  | 2     | 0.958    | 0.774    | 0.548   |
|                  | 3     | 0.889    | 0.733    | 0.412   |
| Forages-Blood of Sheep | 1     | 0.844    | 0.736    | 0.350   |
|                   | 2     | 0.788    | 0.744    | 0.318   |
|                   | 3     | 0.717    | 0.616    | 0.353   |
Table 4. PLI of Magnesium in districts Sargodha, Bhakkar and Mianwali

| PLI | Magnesium | Sargodha | Mianwali | Bhakkar |
|-----|------------|----------|----------|---------|
| 1   | 1.654      | 1.663    | 1.8      |
| 2   | 1.349      | 1.684    | 1.98     |
| 3   | 1.671      | 1.627    | 1.88     |

Figure 2. Magnesium content in soil of districts Sargodha, Bhakkar and Mianwali

Figure 3. Magnesium content in Forage of districts Sargodha, Bhakkar and Mianwali

Figure 4. Magnesium content in Blood of Goat of districts Sargodha, Bhakkar and Mianwali
Figure 5. Magnesium content in Blood of Sheep of districts Sargodha, Bhakkar and Mianwali

Conclusion
The values of Magnesium in soil and forage were above the critical limit indicating that there is no need for Magnesium fertilizers in these sites. Higher BCF in Sargodha and Mianwali as compared to Bhakkar district is evident of movement of this essential macro-mineral in the food chain hence denying the requirement of Magnesium supplementation for goats and sheep of the area under present investigation.

Author’s contributions
The experiment was conceived and designed: ZI Khan, K Ahmad, AB Gulshan & M Munir, Collected the samples and analysis were executed: S Siddique, IS Malik, Contributed reagents and analysis tools: M Nadeem, Collected data was analyzed statistically: H Bashir, Wrote initial draft of the study: S Nazar, K Wajid & A Ashfaq.

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