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Dynamics of Tissues Macro-Minerals Changes of Growing Camels (*Camelus dromedarius*) in Spring Season of Saudi Arabia

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Key words
Camels production, *Camulus dormiderous,* Rumen fluid Macro-minerals

ABSTRACT

The aim of our study was to evaluate the calcium (Ca), phosphorus (P), and magnesium (Mg) status of slaughtered camel tissues in five regions in Saudi Arabia during the spring season. Selected tissues (whole blood, liver, meat, rumen tissue and rumen fluid samples) were collected from 33 growing camels (one year old) from local slaughterhouses in five regions. Ninety-three biological samples were collected and prepared for analysis for Ca, P and Mg by ICP-MS. A significant variation was reported for the Ca, P, Mg and inorganic content of the liver by region. In meat samples, camels from northern and central regions had significantly higher levels of Ca ($P < 0.05$), P ($P < 0.001$), Mg ($P < 0.001$), and inorganic matter content ($P < 0.001$) compared to others. A significant variation in Ca, P and Mg concentration was reported in rumen fluid, rumen tissue and whole blood for camels in the eastern region compared to the central region. A negative trend correlation between rumen fluid P and rumen tissue P ($r = -0.899$; $P < 0.10$) was reported. In summary, variation for Ca, P, and Mg status of camels affected by regions can result from the differences in their concentration in soil, water, and forage, which are mainly related to environmental factors. Phosphorus is the only macro mineral that has shown a correlation between rumen fluid and rumen tissue. Further research is needed to examine mineral metabolism and the possibility of developing a supplementation program in some regions.

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INTRODUCTION

Camel production is considered a very important enterprise in Africa, Asia, and the Middle East in arid and semi-arid conditions (Faye and Bengoumi, 2018; Dowidar *et al*., 2022; Ola *et al*., 2022). Traditional farming is the most important management system for camels because, under very harsh environmental conditions, camels can convert low-quality feed into high-quality meat and milk (Eltaly *et al*., 2 023). Camel meat and dairy products are important for human consumption in Saudi Arabia (Abdelrahman *et al*., 2022a; Almundarij, 2023; Albarrak, 2023). Camel population in Saudi Arabia fluctuates from year to year, because of variations in environmental conditions such as drought, rainfall, variability, and global climate change (Abdelrahman *et al*., 2022b). It is known that mineral deficiencies and toxicity can affect camel production, reproductive performance, low feed efficiency, and many aspects of metabolism growth disorders in domestic animals (Faye and Bengoumi, 2018; Jafari, 2023). Therefore, minerals are very crucial for the productivity and reproduction performance and welfare of camels. Due to the different feeding conditions in different regions and seasons in arid and semi-arid areas, there is usually a variation in dromedary mineral concentrations in blood and tissues. Camel requirements for Ca, P and Mg are not well documented to cover their requirements and optimize their productivity. Ca, P and Mg are the most important macrominerals for animal growth, health, and productivity. Ca is a major component of bone, regulates nerve and muscle contraction, and activate many enzymes, blood clotting, and hormone release (Wilkens *et al*., 2020). P is considered to be very crucial in many metabolic processes in animal body since presenting in

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all body cells. It is one of the main constituents of bones and teeth, buffers, formation of high energy compound and synthesis of phospholipids and phosphoproteins (Hays and Swenson, 1985). Mg plays an important role in the formation of bones and teeth, activates the enzyme system, cofactor for oxidation phosphorylation process in cells (Murray *et al*., 2000). The maximum and minimum levels of minerals for camels in different physiological states are not as well established as for ruminants. Supplementation in dromedary camels is usually based on the estimated needs of other ruminants such as dairy cattle. Our expectations of camels and mineral requirements differ from other ruminants because camels differ in many aspects related to rumen and reticulum anatomy and many other factors related to the adaptation of camel biological systems to harsh conditions. Therefore, it is very important to understand the metabolism of each mineral in terms of bioavailability, absorption and excretion through intensive research and consequently to make solid recommendations for requirements like other ruminants. Therefore, the aim of our study was to determine the Ca, P and Mg concentration in whole blood, liver, meat, rumen tissue and fluid of growing male camels slaughtered in different regions in South Africa. In addition, a correlation study between all tissues was conducted to examine the relationship and role of each tissue in Ca, P, and Mg metabolism.

MATERIALS AND METHODS

Regions and temperature

The current study was conducted in five selected regions in Saudi Arabia with 33 growing camels, about 1 year old and 93 biological samples as shown in Table I.

All samples of this study were prepared and wet digested using acids (Cunniff and Washington 1997), then analyzed for Ca, P and Mg concentration using ICP-MS system. Tissues and fluids samples (whole blood, rumen, liver, meat and rumen fluid) have been prepared as follow: 0.5±0.001g of tissues samples, liver, meat, and rumen were weighed in acid-washed Teflon tube. Then, 3 ml of HNO_3, 1 ml of HCl, 1 ml H_2O_2 and 1 ml deionized H_2O was added to the sample tissues before placed in the digestion units. The samples were digested according to a preset temperature program as recommended by AOAC (1997). The same process was conducted for whole blood and rumen fluid using 1 ml sample sizs. Soil, water and forage samples were also analyzed prepared according to the above procedure. The digested samples were diluted in 0.1 normal HCl in a 25 mL volumetric flask, and 5 mL subsamples were taken in sterilized tubes for mineral analysis. The Ca, P and Mg concentrations were performed using an ICP-MS system with a Meinhard Nebulizer type A2. Argon (purity higher than 99.999% provided by AH Group, (Dammam, Saudi Arabia) was used to maintain plasma and as carrier gas. The operating conditions used for the ICP-MS determination were 1300 W RF power, 15 L min^{-1} plasma flow, 0.2 L min^{-1} auxiliary flow, 0.8 L min^{-1} nebulizer flow, 1.5 mL min^{-1} sample uptake rate. The axial and radial views were used for metal determination, while a 2-point background correction and three replicates were used to measure the analytical signal, with the processing mode being peak area. Emission intensities were recorded for the most sensitive lines free from spectral interference. Calibration standards were prepared by diluting the multi-element stock standard solution (1000 mg L^{-1}) in 0.5% (v/v) nitric acid. The calibration curves for all elements ranged from 1.0 ng mL^{-1} to 1.0 g mL^{-1} (1-1000 ppb).

Table I. Biological samples taken from camels from the five different regions, Saudi Arabia.

Regions*	Camel s' selected tissue					Total
	Meat	Liver	Whole blood	Rumen fluid	Rumen tissue	
Central (Al-Riyadh)	5	5	5	5	5	25
Dammam (Dammam)	4	4	4	4	4	20
Western (Mecca)	8	8	-	-	-	16
Southren (Najran)	7	7	-	-	-	14
Northren (Al-Jouf)	9	9	-	-	-	18
Total	33	33	9	9	9	93

Statistical analysis

Our results were analyzed using a fully randomized design using SAS version 9.4 (SAS Institute Inc. Cary, NC). Dependent variables of this study were the macromineral in sample tissues, whole blood and rumen fluid variables and independent variables were regions). The correlation coefficient between camel tissue minerals from the local slaughterhouse was performed using the Pearson correlation test at the $P < 0.05$ significance level.

RESULTS AND DISCUSSION

The mineral status in inedible and edible tissues has been extensively studied in most ruminants. Most of the articles focused on research related to nutrition and ruminant mineral deficiencies and welfare. In general, minerals are of great importance in animal and human

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diets typical of arid countries due to feed restrictions and low mineral composition. These in turn affect animal health and production performance in indoor or outdoor ruminant production systems. In addition to the factors, there are several other factors besides diet, such as geographical location, air temperature, type (monogastric and ruminant) and sex. In the current study, we aim to infer the role of ambient temperature, regions, and macromineral concentrations in examined tissues and fluids from local camels raised under arid and semi-arid conditions, Saudi Arabia

Temperature and macrominerals levels in soil, water, and forage

The mean ambient temperatures during the spring seasons for all regions are shown in Figure 1.

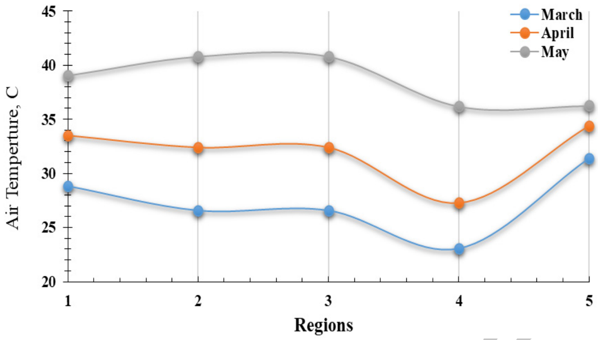

Fig. 1. The temperature during the spring season for the five regions (1: Central, 2: Eastern, 3: Western, 4: Southern and 5: Northern) during spring season.

Figure 1 shows that the ambient temperatures (Ta) were higher in May and lower in March for five regions during the spring season, but they were higher in western regions than other regions. In March and April, Ta was higher in the northern regions than other regions. This means that during this study Ta was different in the different regions. Figure 2 shows the average of Ca, P and Mg in the soil, water and feed from five different regions of Saudi Arabia during the spring season. Calcium was higher in the central region and lowest elsewhere, P was lower in the central region and tended to be higher in the southern, while Mg has a higher value in the western and lowest value in the northern region (Fig. 2A). In the water of each region, Ca was also higher in the central region and lowest in other places, Mg has a higher value in the western region and the lowest in the western region, while P decreased in the southern region, and tended to be higher in the center (Fig. 2B). Figure 2C shows the levels of macrominerals in the diet of camels. Whereas Ca was higher in the west at

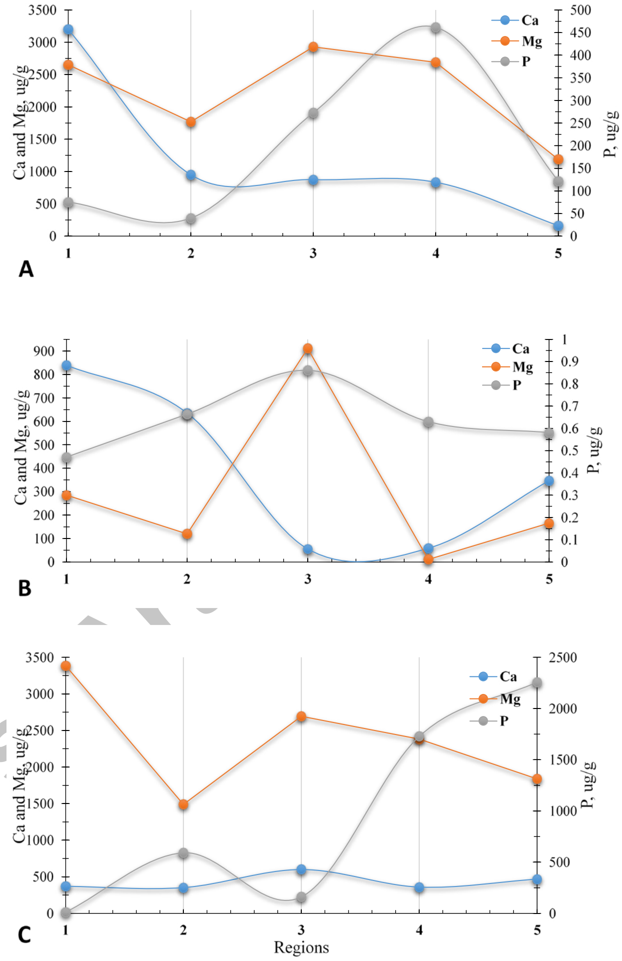

Fig. 2. The average of Calcium, Phosphorus and Magnesium concentrations in soil (A), water (B) and feeds (C) samples from the five regions during spring season. See Figure 1 for regions.

599.2 µg/g and lowest in the south at 359.21 µg/g. Magnesium has a higher value in the central (3386.63 µg/g) and the lowest in the eastern region (1488.39 µg/g). Phosphorus was lower in the central region (11.61 µg/g) and tended to be higher in the northern region (2258.41 µg/g). From our results, we have found that fluctuations in Ta, fodder, water and soil minerals can cause the situation of mineral imbalances during spring. For example, water abundance or scarcity can result in higher/lower soil mineral content. Consequently, this means that there is a relationship between them in mineral content as suggested by Khan *et al.* (2007a). In a suptropical region, it was found that sheep fed an appropriate diet were tested for blood mineral levels, blood Ca was dependent on seasonal variation, and Na was more prevalent during the winter season (Khan *et al.* 2007b). Different Ta and geographic locations are determinants of blood and tissue mineral concentrations. For example, in addition to forage availability and quality and soil mineral properties (Ribeiro *et al*., 2019), Ca levels in bones of sheep during the wet and dry seasons are 22.1% and 21.2% of the DM, respectively (Khan *et al*. 2007b).

Macrominerals concentration in camels liver

Significant variation was reported for Ca (P<0.05), Mg (P<0.001), P (P<0.001) and percentage of inorganics (P<0.05) between all regions in camel meat. The Ca concentration in camel liver was significantly increased in the northern region and lower in the other regions. The liver P, north, and central were significantly highest, followed by other regions. In addition, Mg levels were highest in camels from the south and west and lowest in camel liver in the northern region, followed by the central and eastern regions. The highest percentages of inorganic liver material reported for camels reared in the Northern and Southern Regions followed in the Eastern, Central and Regional Regions, and the lowest for camels in the Western Region (Table II). The liver is the main storehouse for Mg, and a deficiency is associated with liver disorders and diseases (Gibbons *et al*., 1976; Liu *et al*., 2019), The high levels in our study can be attributed to Mg levels that reported in t grazing plants in the five studied regions or type of diet. Furthermore, soil, type of drinking water (Rosborg and Kozisek, 2016), and forage are the main sources of dietary minerals for camels reared in tropical and subtropical conditions (Saini *et al*., 2014; Pinotti *et al*., 2021). In spring, nutritional status includes supplemental grazing, and use of alfalfa, rhodes, and concentrate affected macromineral bioavailability of camels (Qureshi, 2020).

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Table II. The Macro-minerals concentrations (µg/g) of camels in liver, meat, whole blood, rumen fluid (µg/ml) in growing camels liver raised under tropical and sup-tropical regions during spring season.

Variables[1]	Regions					SEM2	P value
	Central	Eastern	Western	Southern	Northern		
Liver							
Ca	339.01[b]	351.01[b]	372.11[c]	343.10[b]	626.01[a]	21.00	*
P	6270.23[c]	5512.85[c]	3137.89[b]	3905.36[b]	8274.24[a]	129.28	***
Mg	749.24[b]	643.67[b]	3137.89[a]	3905.36[a]	1038.77[c]	245.15	***
IOR, %	1.41[a]	1.46[a]	1.16[b]	1.56[a]	1.59[a]	0.05	*
Meat							
Ca	358.03[a]	134.01[b]	282.10a[c]	193.12[c]	334.11[a]	32.91	*
P	4065.04[a]	3052.06[b]	2307.54[c]	1856.81[d]	3928.81[a]	171.82	***
Mg	978.54[a]	707.11[b]	524.17[c]	457.71[c]	814.49[a]	42.36	***
IOR, %	1.49[a]	0.94b[c]	0.88[c]	1.07[b]	1.52[a]	0.06	***
Whole blood							
Ca	0.89[b]	1.24[a]	-	-	-	0.07	**
P	89.91[b]	103.99[a]	-	-	-	7.18	*
Mg	6.33[b]	9.47[a]	-	-	-	0.63	***
Rumen fluid							
Ca	11.932[b]	19.085[a]				1.33	*
P	85.97[b]	163.86[a]				11.49	**
Mg	78.52[b]	180.61[a]				19.87	***
Rumen tissues							
Ca	16.36[b]	32.95[a]				4.38	**
P	235.85[b]	944.04[a]				35.95	**
Mg	42.76[b]	196.34[a]				6.23	***

Ca is calcium; P is phosphorus; Mg is Magnesium and IOR is Inorganic matter%. [2]SEM is Standard error. [3]Means value within rows followed with different superscripts are significantly differ *is P < 0.05; *** is P < 0.001.

In addition to regional and seasonal variation as report by Abdelrahman *et al*. (2022b), the dry and rainy seasons. Also, affect some soil minerals in plant, which in turn can affect the minerals content in the animal's diet, including the various tissues of the body (Al-Ahmadi, 2014). Therefore, this variation may impact mineral status of the livers of camels slaughtered from five regions in the spring region of Saudi Arabia. The concentration in small ruminants (like in sheep liver; 6.66 mg/kg and 15.24 mg/kg, respectively), is very low compare to our results (Wilkens *et al*., 2012). On the other hand, our results were in the range of one-humped camel and cattle (El-Faer *et al*., 1991) and horses (Grace *et al*., 1999).

Microminerals concentration in camels meat

Meat is considered one of the main sources of protein for humans in South Africa and many developing countries in the Middle East and Africa. Our results showed significant differences between all regions for Ca, P of camel meat. Ca, P, Mg in camel meat were significantly higher in the central ($P < 0.05$ and $P < 0.001$) and inorganic fraction ($P < 0.05$) in the northern region. On the other hand, the significantly lowest levels in the camels were raised in the eastern region for Ca, in the southern for P and Mg, and in the western for the fraction of inorganic matter. In general, the proportion of Ca, P, Mg and inorganics in camel meat was significantly higher in the middle ($P<0.05$ and $P<0.001$); and the lowest camels come from the west (Table II). Phosphorus is the vital macro-mineral in meat (Pinotti *et al*., 2021). Our results for macro minerals were within those found in many published scientific papers (Elgasim and Alkanhal, 1992; Ali *et al*., 2011; Ibrahim *et al*., 2017; Siham and Daoud, 2015) and contradicted with those reported by Kadim *et al*. (2018). Calcium and P were higher in cattle than in camels. In general, the main variation in camel meat of macromineral profiles during the spring season could be related to the source and comtent of feed, water that provided by owner or the grazing plant which may also affected by soil.

Macrominerals concentration in camels tissues

There were significant fluctuations for Ca, P and Mg in whole blood and rumen tissue and fluid samples collected from slaughtered camels from central and eastern regions. Calcium, P, and Mg were in higher concentrations in the east for whole blood and rumen tissue and fluid samples compared to the central region. Furthermore, our results showed that the bioavailability of Ca and Mg in whole blood was low compared to other tissues within the same region (Table II). The changes in the concentration of macrominerals in different tissues can be taken as an indicator of the mineral status in camels and originate from different regions as mentioned before (Faye and Bengoumi, 2018).

Table III. Correlation between Ca, P and Mg minerals in meat, liver, whole blood and rumen tissue and fluid collected from Saudi slaughterhouses in five regions during spring season.

Organ	Meat	Liver	Whole blood	Rumen tissue	Rumen fluid
Calcium concentration					
Meat	1				
Liver	0.25 0.75	1			
Whole blood	0.575 0.43	-0.62 0.38	1		
Rumen tissue	-0.77 0.23	-0.110 0.91	-0.64 0.357	1	
Rumen Fluid	0.35 0.65	0.53 0.47	-0.33 0.673	0.28 0.72	1
P concentration					
Meat	1				
Liver	-0.16 0.84	1			
Whole blood	-0.68 0.32	-0.15 0.85	1		
Rumen tissue	0.81 0.19	-0.43 0.57	-0.13 0.87	1	
Rumen fluid	-0.54 0.46	0.74 0.26	-0.17 0.84	-0.91 0.10	1
Mg concentration					
Meat	1				
Liver	-0.62 0.38	1			
Whole blood	-0.82 0.18	0.28 0.72	1		
Rumen tissue	-0.15 0.85	0.51 0.51	-0.43 0.57	1	
Rumen fluid	-0.89 0.11	0.84 0.05*	0.49 0.512	0.56 0.44	1

Correlation coefficient of each macrominerals between different tissues

The rumen plays an important role in the microbial digestion of various dietary components in ruminants. Thus, rumen fluid and rumen tissue showed the mechanism of dietary nutrients including minerals. Since camels are classified as pseudo-ruminants, different microbial digestion and absorption through the epithelial tissue barrier of the rumen wall can be expected compared to

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true ruminants. In addition, the correlation test of each mineral between different biological tissues including rumen fluid and tissue is considered crucial to understand the mechanism of digestion and absorption. Correlation analysis between Ca for tissues examined revealed no significant correlation between all tissues for Ca (Table III), but meat and rumen tissue Ca were numerically correlated ($P < 0.23$) (Table III).

Phosphorus data reported in this study shows a numerical negative correlations ($P<0.10$) between rumen fluid and rumen tissue. Furthermore, a positive correlation was reported between rumen fluids and liver ($P<0.26$), rumen tissue with meat ($P<0.19$), but not significant (Table III).

The current study found surprising results for the relationship between macrominerals of selected slaughtered camels compared to other ruminants. Calcium, P and Mg in the rumen fluid, whole blood and liver appeared to be negatively correlated with meat, expected rumen tissue numerically correlated ($p > 0.05$) with camel meat, indicating that macromineral content in tissues including rumen fluid, whole blood and liver negatively affected the meat values. Macromineral bioavailability is reduced, possibly due to changes occurring in the rumen environment of camels compared to other ruminants. The higher the macroelement content in these tissues, the lower the expected meat tissue ratio between rumen tissue and meat. The same trend, the relationship between macrominerals in rumen tissue and whole blood was negative for Ca and P and positive for Mg, meaning that the higher levels of Ca and P in rumen tissue and whole blood can lead to lower minerals in liver tissue and vice versa for Mg Our data were consistent with Abdelrahman *et al*. (2021) in growing lambs. Several complex relationships and interactions between Ca, Mg, and P occurred in camels tissues as found in ruminants (Littledike and Goff, 1987). The bioavailability of macrominerals is influenced by several factors, such as rumen pH and the presence of other minerals in high concentrations in the feed (Spears, 2003). In addition, methods of mineral supplementation such as the use of long-acting trace elements as a rumen bolus (Alhidary *et al*., 2016), or injection (Pogge *et al*., 2012) in camels.

Finally, the conclusion of this study showed that the differences in the Ca, P and Mg statuses of the affected camels in each region may be due to differences in their concentrations in the soil, water and forage during spring, which are mainly caused by many environmental factors. Phosphorus is considered essential for the microbial fermentation process in the rumen and is clearly reflected in the interrelationship between rumen fluid P and rumen tissuesl P. Therefore, further investigations are needed to investigate the metabolism of rumen macrominerals using in vivo or in vitro rumen culture techniques. The possibility of developing a supplementation program in some regions during specific region must be considered.

DECLARATIONS

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IRB approval

This research work was approved by the Departmental Committee on Ethics and Animal Welfare, King Saud University, Saudi Arabia (KSU/CFAS/201).

Ethical statement

This study was conducted under the approval number KSU-SE-22-21 by the Ethics Committee at King Saud University, Riyadh, Saudi Arabia.

Data availability statement

Data are available upon request.

Statement of conflict of interest

The authors have declared no conflict of interest

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