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SALIVA AND URINE AS INDICATORS OF MINERALS STATUS OF BUFFALOES

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ABSTRACT

The current work was carried out to investigate the effect of dietary minerals intake and minerals status in animal body on minerals concentration in saliva and urine of buffaloes. Twelve dairy buffaloes and 12 growing heifers almost similar in age and live body weight were fed traditional summer and winter rations. Also, two metabolism trails were conducted using barki rams to determine minerals balance of these rations. Samples of saliva, urine and feedstuffs were taken two times through the experimental period after three months of feeding traditional summer and winter rations for minerals determination.

The contents and intake of P, Mg, Na and Zn were higher, but the contents and intake of Ca, K, Cu, Mn and Fe were lower significantly ($P<0.05$) for summer ration compared with winter ration. Moreover, the concentrations of P, Na, K and Zn in saliva and the concentrations of Mg, Na and K in urine increased significantly ($P<0.05$) with increasing the intake of these elements. However, the concentrations of Ca, Mg, Cu, Mn and Fe in saliva and the concentrations of Ca, P, Cu, Zn, Mn and Fe in urine were not significantly ($P> 0.05$) affected by the intake of these elements.
The concentrations Ca, Mg and Mn were higher and the concentrations of P, Na, K and Cu were lower significantly ($P<0.05$) in saliva of dairy buffaloes compared with their growing heifers. While, the concentrations of Zn and Fe were not differing significantly ($P>0.05$). Moreover, the concentrations of Ca, P, Mg, Cu, Zn, Mn and Fe were higher significantly ($P<0.05$) in urine of growing heifers in comparison with dairy buffaloes. While, the concentrations of Na and K were nearly similar.
Keywords: Saliva, urine, minerals, buffaloes, summer and winter rations.

INTRODUCTION

The detection of mineral elements deficiencies or excesses involves clinical, pathological and analytical criteria, as well as response from specific element supplementation. Since minerals analyses are complicated and expensive, it is important to select and analyze the minimal number of plant and animal tissues (or fluids) that are indicative of the minerals status of animals. As in plants, mineral concentrations in animal tissues are influenced by many factors. Nevertheless, when appropriate interpretation is made, animal tissue concentrations are often better indicators of the minerals status of livestock than are either plant or soil concentrations (McDowell, 1985).

Forages are deficient in cobalt, copper, magnesium, sodium, phosphorus and zinc. Deficiencies can be predicted from a combination of analysis of tissues and fluids from the livestock fed forage. Analysis of saliva and urine provides additional diagnostic information of several minerals (McDowell, 1992). The best indices of sodium, potassium and phosphorus status are the concentration of these elements in saliva possibly more sensitive indices of the changing mineral status of the animal (Singh *et al.*, 1999). Salivary phosphorus secretion was directly related to phosphorus intake (Louvandini and Vitti, 1996). Moreover, Henkin *et. al.* (1975) found that salivary zinc concentration is more sensitive indicator of zinc status.

Sodium concentration in the urine is more accurate indicator of sodium deficiency (Underwood, 1980). Suspicion deficient of potassium is aroused by excess potassium excretion in the urine. An excess or lack of magnesium is immediately reflected in daily excretion of magnesium in urine, hence daily urinary excretion is a better criterion of magnesium supply (McDowell, 1992).

The objective of the present study was to investigate the relationship between dietary minerals intake and minerals concentration in saliva and urine of dairy buffaloes and their growing heifers fed traditional summer and winter rations.

MATERIALS AND METHODS

The current work was carried out at Mehallat Mousa Animal Production Research Station, Animal Production Research Institute, Agricultural Research Center. Twelve dairy buffaloes and 12 growing heifers of 4-6 and 1-2 years of age and weighed 600 and 250 kg on the average, respectively were fed 50 % concentrate mixture, 20 % berseem hay and 30 % rice straw (summer ration) and 40 % concentrate mixture, 40 % fresh berseem and 20 % rice straw (winter ration). The berseem hay and rice straw were brought from the local area and fresh berseem from Experimental Farm of Sakha Agricultural Research Station, while concentrate mixture was obtained from Tanta Oil and Soap Company, Tanta, Egypt. Daily feed intake covered TDN and CP requirements according to the Recommendation of Animal Production Research Institute (1997). Two metabolism trials were conducted to determine mineral absorption and retention using three Barki rams with an average body weight of 50 kg and 3 years of age fed the same rations used in feeding buffaloes. Rations were offered to cover the maintenance requirement of rams according to NRC (1985) for fifteen days preliminary period followed by seven days collection period. Metabolism crates permitted total collection and separation of feces and urine.

Dairy buffaloes and their growing heifers were fed individually. Concentrate mixture was offered two times daily at 8 a.m. and 4 p.m., berseem hay or fresh berseem once daily at 11 a.m. and rice straw two times daily at 9 a.m. and 5 p.m. Fresh water was offered to animals three times daily. Minerals content of feedstuffs, traditional summer and winter rations is shown in Table (1). Minerals concentrations in drinking water were Ca 0.27, P 0.14, Mg 0.60, Na 0.41 and K 0.21 g/liter, Cu 0.03, Zn 0.05, Mn 0.63 and Fe 0.02 mg/liter as reported by Abou-Aiana (2003).

Samples of saliva and urine were taken two times through the experimental period after three month of feeding traditional summer and winter rations (in Augusts and February) from each animal at 2-3 hours after feeding. Saliva samples were obtained by forceps with dried collapsed cellulose sponges placed in the mouth by the maxilla and upper molar teeth in the area of the parotid duct orifice for about 2 minutes. Saliva was expelled from the sponges with disposable gloves and stored frozen in acid washed plastic bottles. Urine samples were taken by clitoral stimulation after the vaginal area was washed with warm water. Also, samples of concentrate mixture, fresh berseem, berseem hay and rice straw were taken at the same time of saliva and urine sampling.

Table (1): Minerals content of feedstuffs, summer and winter rations.

Items	Ca	P	Mg	Na	K	Cu	Zn	Mn	Fe
Feedstuffs	g / kg DM					mg / kg DM			
Concentrate mixture*	4.45	7.65	4.70	4.35	24.65	13.90	62.30	60.20	115.50
Berseem hay	10.30	3.20	3.60	2.85	38.50	12.20	42.60	25.40	135.40
Fresh berseem	12.40	2.10	2.50	2.70	45.90	14.50	35.75	42.50	180.70
Rice straw	2.40	0.80	1.15	2.20	9.50	4.90	15.70	12.50	35.80
Experimental rations									
Summer ration	5.01	4.71	3.42	3.41	22.88	10.86	44.38	38.93	95.57
Winter ration	7.22	4.06	3.11	3.26	30.12	12.34	42.36	43.58	125.64

*** Concentrate mixture consisted of 32% undecorticated cottonseed cake, 24%wheat bran, 22% yellow corn, 12% rice bran, 5% lineseed cake, 2% molasses, 2% limestone and 1% common salt.**

The samples were prepared for minerals determination according to the methods of AOAC (1990). Samples of feedstuffs and feces were dried in an oven at 60 ^{0}C four 48 hours, then weighed into crucibles and dry ashed at 500 ^{0}C for 5 hours. Ash was weighed and taken into 10 ml of HCL (10%) and diluted by 15 ml distilled water, then placed on hot plate until the solution completely evaporated. Thereafter, 10 ml HCL (10%) was added and the solution heated to observe ascending vapors (about 80-90 ^{o}c), following filtrated in volumetric flask (50 ml) and transferred into dry clean class bottles. While, saliva and urine samples were wet ashed using pure sulfuric and nitric acids (2:1 by volume) until most organic matter was destroyed. Then, filtrated in volumetric flask (50 ml) and transferred into dry clean class bottles. Minerals determined in feedstuffs, feces, saliva and urine samples as follows:

a- Calcium, magnesium, copper, zinc, manganese and iron were determined by Atomic Absorption Spectrophotometer (Perkin Elmer 2380).

b- Total phosphorus was determined according to the method described by Traung and Meyer (1939) using Spectrophotometer (Milton Roy Company Spectronic 20 D).

c- Sodium and potassium were determined by Flame Photometer (Jenway PFP 7).

The data obtained from chemical analysis were statistically analyzed using general liner models procedure adapted by SPSS (1997).

RESULTS AND DISCUSSION

Minerals intake from feedstuffs by dairy buffaloes and their growing heifers is presented in Table (2). Minerals intake revealed that the intake of P, Mg, Na and Zn were higher, but the intake of Ca, K, Cu and Fe was lower significantly (P<0.05) in summer ration compared with winter ration. These results might be due to the higher contents of P, Mg, Na and Zn and the lower contents of Ca, K, Cu and Fe in summer ration compared with winter ration as shown in Table (1). Moreover, average daily intake of macro and micro minerals by dairy animals was higher significantly (P<0.05) than that of growing heifers. These results could be attributed to the higher DM intake by dairy animals. All minerals except Mn in summer ration covered the recommended requirements being Ca 0.43-0.60, P 0.26-0.40, Mg 0.20-.035, Na 0.35, K 0.80%, Cu 10, Zn 40, Mn 40 and Fe 50 ppm on DM basis (NRC, 2001).

Table (2): Minerals intake from feedstuffs by dairy buffaloes and their growing heifers fed summer and winter rations.

Elements	Ration	Dairy animals	Growing heifers
DM intake (kg/ h/ day)	Summer	15.00 ± 0.09^{A}	7.50 ± 0.06^{B}
	Winter	14.50 ± 0.07^{A}	7.00 ± 0.05^{B}
Macro-minerals (g / h / day)			
Calcium (Ca)	Summer	75.08 ± 1.60^{bA}	37.54 ± 2.09^{bB}
	Winter	104.69 ± 1.81^{aA}	50.54 ± 1.49^{aB}
Phosphorus (P)	Summer	70.57 ± 0.44^{aA}	35.29 ± 0.57^{aB}
	Winter	58.87 ± 0.50^{bA}	28.42 ± 0.41^{bB}
Magnesium (Mg)	Summer	51.22 ± 0.70^{aA}	25.61 ± 0.92^{aB}
	Winter	45.10 ± 0.80^{bA}	21.77 ± 0.65^{bB}
Sodium (Na)	Summer	51.08 ± 1.15^{aA}	25.54 ± 2.00^{aB}
	Winter	47.27 ± 1.30^{bA}	22.82 ± 1.07^{bB}
Potassium (K)	Summer	343.13 ± 7.91^{bA}	171.56 ± 6.42^{bB}
	Winter	436.74 ± 8.57^{aA}	210.84 ± 4.58^{aB}
Micro-minerals (mg / h / day)			
Copper (Cu)	Summer	162.90 ± 1.48^{bA}	81.45 ± 1.93^{bB}
	Winter	178.93 ± 1.67^{aA}	86.38 ± 1.37^{aB}
Zinc (Zn)	Summer	667.20 ± 9.68^{aA}	333.60 ± 11.15^{aB}
	Winter	615.38 ± 8.55^{bA}	297.08 ± 7.95^{bB}
Manganese (Mn)	Summer	583.95 ± 5.65^{A}	291.98 ± 7.35^{B}
	Winter	631.91 ± 6.39^{A}	305.06 ± 5.24^{B}
Iron (Fe)	Summer	1433.55 ± 38.84^{bA}	716.78 ± 50.52^{bB}
	Winter	1821.78 ± 43.97^{aA}	879.48 ± 36.05^{aB}

^{a} and ^{b}: values for each element in the same column with different superscripts differ significantly (P<0.05).
^{A} and ^{B}: values in the same row with different superscripts differ significantly (P<0.05).

The effects of dietary minerals intake on minerals concentration in saliva of dairy buffaloes and their growing heifers fed traditional summer and winter rations are presented in Table (3). The concentrations of P, Na, K and Zn in saliva increased significantly (P<0.05) with increasing dietary intake of these elements. These results may be attributed to a significant increase in the absorption and retention (g or mg / h / day) of P, Na, K and Zn by rams with increasing the intake of these elements (P<0.05) as shown in Table (5). Moreover, the positive correlation between salivary P, Na, K and Zn concentration and dietary intake of these elements was 0.65, 0.82, 0.78 and

0.60, respectively. However, the concentration of Ca, Mg, Cu, Mn and Fe in saliva did not significantly (P>0.05) influence by dietary intake of these elements. These results agreed with those obtained by Ward (1966) Henkin *et al.* (1975), Tomas and Potter (1975), and Scott *et al.* (1985) They found that increasing dietary intake of P, Na, K and Zn resulted in increases of their concentrations in saliva.

Table (3): Minerals concentration in saliva of dairy buffaloes and their growing heifers fed summer and winter rations.

Elements	Ration	Dairy animals	Growing heifers
Macro- minerals (mg/100 ml)			
Calcium (Ca)	Summer	2.60 ± 0.18^{A}	2.25 ± 0.14^{B}
	Winter	2.75 ± 0.23^{A}	2.40 ± 0.18^{B}
Phosphorus (P)	Summer	25.20 ± 0.45^{aB}	28.85 ± 0.90^{aA}
	Winter	20.70 ± 0.60^{bB}	23.70 ± 0.75^{bA}
Magnesium (Mg)	Summer	1.60 ± 0.05^{A}	1.40 ± 0.04^{B}
	Winter	1.45 ± 0.06^{A}	1.25 ± 0.04^{B}
Sodium (Na)	Summer	358.40 ± 9.50^{aB}	380.60 ± 6.21^{aA}
	Winter	335.20 ± 7.30^{bB}	348.20 ± 6.85^{bA}
Potassium (K)	Summer	30.40 ± 2.10^{bB}	32.40 ± 2.30^{bA}
	Winter	34.90 ± 1.65^{aB}	36.55 ± 2.35^{aA}
Micro- minerals (mg/100 ml)			
Copper (Cu)	Summer	0.35 ± 0.03^{B}	0.46 ± 0.04^{A}
	Winter	0.41 ± 0.04^{B}	0.50 ± 0.04^{A}
Zinc (Zn)	Summer	3.10 ± 0.17^{a}	3.20 ± 0.18^{a}
	Winter	2.38 ± 0.15^{b}	2.40 ± 0.15^{b}
Manganese (Mn)	Summer	1.30 ± 0.15^{A}	1.15 ± 0.12^{B}
	Winter	1.45 ± 0.17^{A}	1.30 ± 0.16^{B}
Iron (Fe)	Summer	6.70 ± 0.55	6.50 ± 0.52
	Winter	6.85 ± 0.65	6.65 ± 0.45

[a] and [b]: Values for each element in the same column with different superscripts differ significantly (P<0.05).

[A] and [B]: Values in the same row with different superscripts differ significantly (P<0.05).

Although dietary P and Zn intake covered the recommended requirements of these elements according to NRC (2001) as shown in Table (2). Yet, it was obvious that salivary P and Zn concentration was below the normal level being 31.83 ± 3.36 mg / 100 ml (Dua *et al.*, 1986) and 3.80 ± 0.50 mg / 100 ml (Swanson and King, 1982), respectively. The previous results may be attributed to the negative absorption and retention of P and Zn as shown in Table (5). These results are in harmony with those obtained by Dua and Care (1998).

The concentration of Na and K in saliva ranged from 335.20 and 30.40 to 380.60 and 36.55 mg / 100 ml, respectively. These values were above the normal levels of Na and K being 322.30 ± 14.12 and 28.67 ± 7.20 mg / 100 ml, respectively as obtained by Dua *et al.* (1986). These results may be attributed to the high positive absorption and retention by rams as shown in Table (5). Moreover, the concentrations of Ca, Mg and Mn were higher, but the concentrations of P, Na, K and Cu were lower significantly ($P<0.05$) in saliva of dairy animals compared with growing heifers. These results were in accordance with those obtained by Allam.(1989). It was obvious from the previous results that the concentration of P, Na, K and Zn in saliva of dairy buffaloes and growing heifers was a great indicator of the intake, availability and status of these elements.

The concentrations of minerals in urine of dairy buffaloes and their growing heifers fed traditional summer and winter rations are presented in Table (4). The concentration of Mg, Na and K in urine increased significantly ($P<0.05$) with increasing dietary intake of these elements, for the positive correlation between them being 0.57, 0.75 and 0.70, respectively.

Table (4): Minerals concentration in urine of dairy buffaloes and their growing heifers fed summer and winter rations.

Elements	Ration	Dairy animals	Growing heifers
Macro- minerals (mg/ 100 ml)			
Calcium (Ca)	Summer	22.45 ± 2.30^{B}	25.85 ± 3.50^{A}
	Winter	23.30 ± 3.30^{B}	27.20 ± 1.90^{A}
Phosphorus (P)	Summer	4.80 ± 0.50^{B}	5.10 ± 0.58^{A}
	Winter	4.65 ± 0.42^{B}	4.95 ± 0.50^{A}
Magnesium (Mg)	Summer	30.75 ± 2.35^{aB}	32.60 ± 3.05^{aA}
	Winter	25.65 ± 3.40^{bB}	27.80 ± 1.70^{bA}
Sodium (Na)	Summer	202.20 ± 8.75^{a}	208.70 ± 10.50^{a}
	Winter	170.75 ± 7.80^{b}	171.60 ± 8.50^{b}
Potassium (K)	Summer	730.85 ± 80.10^{b}	744.45 ± 67.95^{b}
	Winter	865.40 ± 52.20^{a}	880.80 ± 65.40^{a}
Micro- minerals (mg/ 100 ml)			
Copper (Cu)	Summer	0.035 ± 0.005^{B}	0.048 ± 0.004^{A}
	Winter	0.040 ± 0.006^{B}	0.060 ± 0.005^{A}
Zinc (Zn)	Summer	0.026 ± 0.002^{B}	0.032 ± 0.002^{A}
	Winter	0.022 ± 0.002^{B}	0.028 ± 0.002^{A}
Manganese (Mn)	Summer	0.007 ± 0.001^{B}	0.009 ± 0.001^{A}
	Winter	0.009 ± 0.001^{B}	0.013 ± 0.002^{A}
Iron (Fe)	Summer	0.030 ± 0.003^{B}	0.042 ± 0.004^{A}
	Winter	0.038 ± 0.003^{B}	0.048 ± 0.005^{A}

[a] and [b]: Values for each element in the same column with different superscripts differ significantly ($P<0.05$).
[A] and [B]: Values in the same row with different superscripts differ significantly ($P<0.05$).

Also, minerals balance by rams as shown in Table (5) revealed that urinary excretion of Mg, Na and K expressed as g / h / day increased significantly ($P<0.05$) with increasing the intake and absorption of these elements. The present results agreed with those obtained by Poe *et al.* (1985) and Davenport *et al.* (1990) They found that increasing the intake of Mg, Na and K resulted in increases in their concentrations in urine.

However, the concentrations of Ca, P, Cu, Zn, Mn and Fe in urine were not significantly ($P> 0.05$) affected by the intake of these elements. Similar results were obtained by Ivan and Grieve (1975), Oluokum and Bell

(1985) and and Drew *et al.* (1990). Moreover, the concentrations of minerals except Na and K were higher significantly (P<0.05) in urine of growing heifers in comparison to dairy animals. These results were in agreement with those obtained by Kilmer *et al.* (1981), Hansard (1983) and Tucker *et al.* (1992). The previous results obviously showed that urinary excretion of Mg, Na and K was a good indicator of the intake and availability of these elements.

Moreover, it is necessary to carry out minerals metabolism trails for helping us to interpret some of the data obtained from the field experiments. Minerals metabolism by rams fed traditional summer and winter rations is illustrated in Table (5). Minerals intake by rams fed summer ration was lower in Ca, K, Cu, Mn and Fe, but higher in P, Mg, Na and Zn compared with feeding winter ration (P<0.05). Minerals excretion in feces except Na and the excretion of Mg, Na and K in urine increased significantly (P<0.05) with increasing the intake of these elements.

Table (5): Minerals metabolism by rams fed traditional summer and winter rations.

Elements	Ration	Intake	Excretion		Absorption	Retention
			Feces	Urine		
Macro-minerals (g / h / day)						
Calcium	Summer	6.26^b	2.85^b	1.35	3.41	2.06
	Winter	8.66^a	4.85^a	1.50	3.81	2.31
Phosphorus	Summer	5.88^a	6.40^a	0.40	-0.52^a	-0.92^a
	Winter	4.87^b	6.00^b	0.35	-1.13^b	-1.48^b
Magnesium	Summer	4.27^a	2.78^a	0.30^a	1.49	1.19
	Winter	3.73^b	2.40^b	0.20^b	1.33	1.13
Sodium	Summer	4.26^a	0.30	2.65^a	3.94^a	1.31^a
	Winter	3.91^b	0.28	2.45^b	3.63^b	1.18^b
Potassium	Summer	28.59^b	1.75^b	15.50^b	26.84^b	11.34^b
	Winter	36.14^a	2.15^a	19.55^a	33.99^a	14.44^a
Micro-minerals (mg / h / day)						
Copper	Summer	13.58^b	8.20^b	0.90	5.38	4.48
	Winter	14.81^a	9.20^a	1.00	5.61	4.61
Zinc	Summer	55.60^a	61.40^a	4.20	-5.80^a	-10.00^a
	Winter	50.93^b	59.00^b	4.05	-8.07^b	-12.12^b
Manganese	Summer	48.66^b	58.90^b	3.05^b	-10.24	-13.29
	Winter	52.30^a	61.90^a	3.25^a	-9.60	-13.05
Iron	Summer	119.46^b	60.70^b	8.70^b	58.76	50.06
	Winter	150.77^a	90.80^a	9.40^a	59.97	50.57

[a] and [b]: Values for each element in the same column with different superscripts differ significantly (P<0.05).

Moreover, the absorption and retention of P, Na, K and Zn increased significantly (P<0.05) with increasing the intake of these elements. Also, metabolism trails by rams revealed negative absorption and retention for P, Zn and Mn and positive absorption and retention for the other minerals. The negative absorption and retention of P, Zn and Mn might be attributed to increasing dietary K and Fe contents (Table 1) and intake (Table 2), since they had antagonistic effect on the absorption and retention of P, Zn and Mn. The evaluation of feeds and feed supplements as sources of minerals depends not only on what the feed contains (the total content or concentration as determined physico-chemically) but alos on how much of the total minerals can be absorbed from the gut and used by animal's cells and tissues. These results agreed with those obtained by Durand *et al.* (1982). and Georgievskii (1982).

From these results it could be concluded that the analyses of P, Na, K and Zn in saliva and Mg, Na and K in urine of farm animals were found to be a great indicator concerning the intake and status of these elements.

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اللعاب و البول كدلائل على حالة العناصر المعدنية فى الجاموس

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تم إجراء هذه الدراسة على ١٢ جاموسة حلابة و ١٢ عجلة نامية متماثلة تقريبا فى العمر و الوزن، غذيت على العلائق التقليدية خلال فصلى الصيف و الشتاء و التى تتكون من ٥٠ % علف مركز + ٢٠ % دريس برسيم + ٣٠ % قش أرز خلال فصل الصيف، ٤٠ % علف مركز + ٤٠ % برسيم طازج + ٢٠ % قش أرز خلال فصل الشتاء لدراسة تركيز العناصر المعدنية فى اللعاب و البول و تأثرها بالمأكول و علاقتها بحالة هذه العناصر فى جسم الحيوان. أيضا أجريت تجربتين هضم على العلائق الصيفية و الشتوية باستخدام الكباش البرقى لتقدير التمثيل الغذائى للعناصر المعدنية. أخذت عينات مواد العلف و اللعاب و البول مرتين خلال فترة التجربة بعد ثلاثة شهور من التغذية على العلائق الصيفية و الشتوية لتقدير العناصر المعدنية و قد أوضحت النتائج الأتى:-

ارتفع محتوى كل من الفوسفور و الماغنسيوم و الصوديوم و الزنك و المأكول منهم بينما انخفض محتوى كل من الكالسيوم و البوتاسيوم و النحاس و المنجنيز و الحديد و المأكول منهم (على مستوى ٠,٠٥) فى العليقة الصيفية بالمقارنة بالعليقة الشتوية.

وجد ارتباط موجب معنوى (على مستوى ٠,٠٥) بين تركيز كل من الفوسفور و الصوديوم و البوتاسيوم و الزنك فى اللعاب و تركيز كل من الماغنسيوم و الصوديوم و البوتاسيوم فى البول مع المأكول من هذه العناصر. فى حين أن تركيز كل من الكالسيوم و الماغنسيوم و النحاس و المنجنيز و الحديد فى اللعاب و تركيز كل من الكالسيوم و الفوسفور و النحاس و الزنك و المنجنيز و الحديد فى البول لم يتأثر معنويا بالمأكول من هذه العناصر.

ارتفع تركيز كل من الكالسيوم والماغنسيوم و المنجنيز و انخفض تركيز كل من الفوسفور و الصوديوم و البوتاسيوم و النحاس (معنويا على مستوى ٠,٠٥) فى لعاب الجاموس الحلاب بالمقارنة بالعجلات النامية. بينما تركيز كل من الزنك و الحديد لم يختلف معنويا. بالإضافة إلى ذلك ارتفع تركيز كل من الكالسيوم و الفوسفور و الماغنسيوم و النحاس و الزنك و المنجنيز و الحديد (معنويا على مستوى ٠,٠٥) فى بول العجلات النامية بالمقارنة بالجاموس الحلاب. فى حين أن تركيز كل من الصوديوم و البوتاسيوم متماثل تقريبا.

يتضح من هذه الدراسة أن تركيز كل من الفوسفور و الصوديوم و البوتاسيوم و الزنك فى اللعاب و كذلك تركيز كل من الماغنسيوم و الصوديوم و البوتاسيوم فى البول يعطى دليلا جيدا للمأكول و حالة هذه العناصر فى الحيوانات المزرعية.