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Tissue Trace Mineral Concentrations of Abattoir Sheep and Goats at the End of the Dry and Wet Seasons of Central Trinidad

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Abstract: *A study was conducted to evaluate serum and tissue micro-mineral concentrations of sheep and goats at the end of the dry and wet seasons in Trinidad. Tissue Samples for analysis were taken from the same liver lobe, the kidney cortex mainly (and outer medulla) and the cerebral hemispheres. Tissue sample trace concentrations were for Cu, Zn, Fe and Mn were determined by Atomic absorption spectroscopy.*

Sheep had lower mean liver Zn ($P<0.01$) and brain Zn ($P<0.05$) in the wet than in the dry season, while goats had lower liver Zn ($P<0.05$) and kidney Zn ($P<0.01$) in the wet season. Manganese levels were lower ($P<0.05$) in sheep livers in the dry than the wet season, and in brains of adults than in growing lambs. Tissue Zn levels in the wet season in some sheep and goats and particularly growing lambs probably indicated a state of marginal Zn deficiency. Also, in the dry and in the wet seasons, 40- 70% of sheep and goats had liver Cu below critical level (<25 mg/kg DM).

Keywords: *Liver, Kidney, Brain, Cu, Zn, Fe, Mn, season, age*

1. INTRODUCTION

In Trinidad, previous studies have demonstrated low concentrations of Cu and other minerals in tropical grasses (Youssef et al., 1999) and in the serum of sheep and goats (Youssef, 1985). Also, there have been sporadic occurrences, mainly in the dry and sometimes in the wet season, in Trinidad, of lambs and kids that have died within 6 months of birth of Cu deficiency swayback (Mohammed et al., 1995). Animals displayed an inability to stand at birth or hind limb ataxia progressing to paralysis in the delayed forms. Although the pathognomonic lesion of chromatolysis of the large motor neurons of the brainstem and spinal cord of swayback lambs and kids is unique to the condition, low serum and tissue Cu levels confirm the diagnosis of the condition. In particular, low serum Cu (<0.5 mg/L), liver Cu (< 25 mg/kgDM) and brain (< 6 mg/kg D (Whitelaw et al., 1982) levels support a diagnosis of the swayback condition in affected lambs and kids. However, marked seasonal related changes in blood plasma Cu have been reported by Gromadzka-Ostrowsk et al. (1986) over a three year investigation in primitive goats. In a study of Khan et al. (2007) seasonal variations in plasma Cu were detected in grazing goats in southern region of Punjab in Pakistan, where levels were higher in winter than in summer seasons. These variations could be attributed to the higher Cu levels in forage during winter than in summer, although no such difference was detected in feeds. Goats had their dietary Cu requirements fulfilled with the free supplement of salt lick all year. However, only lactating goats were marginally Cu deficient in summer. An increase in plasma Cu in June (hot season) compared with October/November (cool season) in goats in Oman (Osman et al., 2003) was suggested by authors to be related to generous supply of Cu rather than a genuine effect of season. However, there was a prevalence of swayback among kids born to grazing goats in summer in Oman rather than at other time (Ivan et al., 1990) suggesting the natural influence of season on Cu status. Variation of tissue Cu concentrations were also reported for location.

Comparisons of locations were reported by Tartour (1975) who did not find differences in serum Cu levels of sheep and goats, among regions in Western Sudan. In Saudi Arabia, Ali and Al-Noim (1992) found differences in serum Cu between grazing and intensively reared sheep at two locations. A high prevalence of plasma Cu deficiency in sheep and goats have been reported across districts in Kashmir valley (Yatoo et al., 2013). Liver Cu levels found in normal goats of Kenya and sheep of Ethiopia were between 9 - 18 and 16 - 51 mg/kg DM, respectively (Faye et al., 1991).

Although, little is known of these concentrations in goats and sheep in the wet and dry season, there are several reports on seasonal liver trace mineral concentrations in beef cattle from other tropical countries. For example, higher incidences of liver Cu deficiencies (<75 mg/kg DM) are reported in the wet than in the dry season in beef cattle from Malawi (Mtimuni et al., 1990), St. Croix (Wildeus et al., 1992) and Ethiopia (Khalili et al., 1993). Additionally Zn, Fe and Mn levels are lower in the wet than in the dry season in beef cattle from Malawi (Mtimuni et al., 1990). In contrast, liver Fe levels are higher in the wet than in the dry season in Ethiopia (Khalili et al., 1993b whereas liver Zn appears unaffected by season in beef cattle from Bolivia (McDowell et al., 1982b), Guatemala (Tejada et al., 1987) and St. Croix (Wildeus et al., 1992).

There are few reports on mineral concentrations in serum, liver, and no reports on kidney and brain tissues of normal sheep and goats and those unaffected by Swayback and pertaining to season from Trinidad and other tropical countries.

The purpose of this research therefore, was to evaluate serum, liver, kidney, and brain tissue micro-mineral concentrations in abattoir sheep and goats in the dry and wet seasons in Trinidad. Additionally, because of a lack of information on sheep and goats, liver mineral concentrations would be compared with those of beef cattle from other tropical countries.

2. Materials and Methods

2.1 Animals Abattoir sheep were of Barbados Black Belly and West African hair type origin, whereas goats were of local Saanen and Anglo Nubian origin. Sheep and goats, comprising growing (mostly about 6 months) and dry or culled adult (2-6yr) animals, originated from several farms throughout southern and Central Trinidad. The animals foraged on a wide variety of local grasses, especially bamboo grass (*Paspalum fasciculatum*). Neither sheep nor goats were provided with adequate mineral supplements or oral anthelmintics.

Serum and tissue samples were collected from 39 sheep and 39 goats, at the end of the dry (April - May) and wet seasons (November - December) of Central Trinidad. Tissue samples represented those of 18 and 12 growing, including 21 and 27 adult sheep and goats, respectively (Table 1).

Table - 1 *Numbers of tissue samples collected from sheep and goats in the dry and wet seasons of Central Trinidad*

Animal	Growing			Adult			Total	
Season	Dry	Wet	Total	Dry	Wet	Total	Dry	Wet
Sheep[a]	6	12	18	13	8	21	19	20
Goats[a]	7	5	12	12	15	27	19	20

[a] Same numbers for serum, liver, kidney and brain tissues.

2.2 Sample Collections Blood samples were drawn by venepuncture (16 - 18 mm needle gauge) and collected in acid washed demineralized tubes. Clotted blood was centrifuged within four hours of collection to remove serum. Liver tissue, whole kidneys and brains were immediately collected from commercial abattoirs after fasting and captive bolt stunning. All serum and tissue samples were properly labeled and stored at -20°C.

Samples for analysis were taken from the same liver lobe, the kidney cortex mainly (and outer medulla) and the cerebral hemispheres. Oven dried tissue samples (20 - 25 g) were pre-ashed on a hotplate with 50% HNO_3, ashed overnight at 550°C, and finally solubilized with 10% HCl. Serum and tissue Zn, including tissue Fe and Mn mineral concentrations were determined according to Fick et al. (1979). All mineral determinations were carried out on a Pye Unicam SP2900 Atomic Absorption Spectrophotometer equipped with PU9090 Data Graphics System.

2.3 Statistics Tissue mineral means were tested separately in goats and sheep by Analysis of Variance using the General Linear Model procedure (Genstat Release 18.1 (PC/Windows 8) 23 December 2015 22:39:42 Copyright 2015, VSN International Ltd.). Means were tested between seasons and ages including interaction of season and age.

3. RESULTS

In sheep and goats, liver kidney and brain Cu did not vary (P>0.05) with season (Tables III and IV) or age (not shown). In sheep there was a higher incidence of serum Cu deficiency (< 0.5 mg/L) in the dry than in the wet season (58% cf., 35%), and in adult than in growing animals (64% cf. 42%). Additionally, there was a higher incidence of liver Cu deficiency (<25 mg/kg DM) in adult than in growing sheep (77% cf. 39%), but generally, a similar level of deficiency recorded in the dry and wet seasons (48% cf. 40%), respectively. In goats a somewhat similar incidence of serum Cu deficiency was observed in both seasons (79% cf., 85%) and in growing and adult goats (82% cf. 82%). A Similar incidence of liver Cu deficiency was also found in the dry and wet season (63% cf. 70%), and in growing and adult goats (73% cf. 82%).

Table – 2 *Tissue Micromineral Levels (mg/kgDM) in Sheep in the dry and wet Seasons*

Mineral	Critical	Tissue	Dry Season		Wet Season		Signif.[1]	Age x
	Levels		Mean[a]	SE	Mean[a]	SE	Season	Season
Cu	0.50[d]	Serum	0.46	0.070	0.61	0.065	NS	NS
		%BCL	58		35			
	25[b]	Liver	97.0	26.87	61.7	24.85	NS	NS
		%BCL	48		40			
	10[c]	Kidney	9.7	0.96	9.2	0.89	NS	NS
		%BCL	53		50			
	6[b]	Brain	11.1	1.16	11.3	1.07	NS	NS
		%BCL	26		26			
Zn	0.6[d]	Serum	0.75	0.057	0.77	0.052	NS	NS
		%BCL	42		5			
	84[b]	Liver	113	4.5	93.4	4.2	***	***
		%BCL	5		40			
		Kidney	99.7	3.90	92.6	3.61	NS	NS
		Brain	48	1.7	43	1.6	*	NS
Fe	180[b]	Liver	642	151.4	522	140.0	NS	NS
		%BCL	5		15			
		Kidney	401	77.5	315	71.7	NS	NS
		Brain	101	5.5	107	5.1	NS	NS
Mn	6[b]	Liver	7.3	0.52	8.8	0.48	*	NS
		%BCL	21		5			
		Kidney	5.4	0.23	5.5	0.21	NS	NS
		Brain	3.5	0.24	3.0	0.22	NS	NS

a Represents the means of 19 and 20 goats, respectively

[b,c,] McDowell (1985); Woolliams *et al*., (1986b); Whitelaw *et al*., (1982)

[d] mg/L

[1] *, ** Means differ at P<0.05 and P<0.01 levels of significance.

Table - 3 *Tissue Micromineral Concentrations (mg/kgDM) in Goats in the dry and wet Seasons*

Mineral	Critical	Tissue	Dry Season		Wet Season		Signif.[1]	Age x
	Levels		Mean[a]	SE	Mean[a]	SE	Season	Season
	0.50[b]	Serum	0.33	0.044	0.44	0.048	NS	*
		%BCL	79		85			
Cu	25	Liver	53.5	20.40	28.9	22.15	NS	NS
		%BCL	63		70			
	10	Kidney	10.4	0.67	9.9	0.73	NS	NS
		%BCL	42		55			
	6	Brain	10.1	1.8	8.2	2.0	NS	NS
		%BCL	0		20			
Zn	0.6[b]	Serum	0.79	0.074	0.70	0.080	NS	NS
		%BCL	42		0			
	84	Liver	165	12.5	122	13.6	*	NS
		%BCL	0		0			
		Kidney	96	1.49	87	1.62	***	NS
		Brain	40	6.1	38	6.7	NS	NS
Fe	180	Liver	783	319.5	343	346.9	NS	NS
		%BCL	40		40			
		Kidney	273	20.9	224	22.7	NS	NS
		Brain	103	8.5	111	9.3	NS	NS
Mn	6	Liver	10.9	0.73	10.3	0.79	NS	NS
		%BCL	11		5			
		Kidney	6.9	0.37	6.6	0.41	NS	NS
		Brain	3.8	0.21	3.5	0.23	NS	NS

[a] Represents the means of 19 and 20 goats, respectively

[b] mg/L

[1] *, ** Means differ at P<0.05 and P<0.01 levels of significance

In sheep, lower mean Zn was found in the wet than in the dry season in liver (*P*<0.001) and brain (*P*<0.05) tissues with an age **x** season interaction for liver Zn (*P*<0.001). Lower Zn was found in the wet than in the dry season in liver (*P*<0.05) and kidney (*P*<0.001) tissues of goats. Also, higher (*P*<0.05) serum Zn (mg/L) was found in growing (0.85) than in adult sheep (0.66). Percent of Zn deficient sheep livers (<84 mg/kg DM) was higher in the wet than in the dry season (40% cf. 5%), and contrastingly, in growing than in adult sheep (57% cf. 0%). Wet season liver, kidney and brain Zn in sheep expressed on a wet weight basis were 28, 18 and 10 mg/kg w.wt, respectively. Percentages of sheep and goats marginal deficient in serum Zn (< o.6 mg/L) in was higher (42% & 42%) in the dry than in the wet season (5% & 0%).

In sheep and goats, mean liver, kidney and brain Fe was not affected by season (Tables III and IV) or age. Forty percent of goats had low Fe (<180 mg/kg DM) in both seasons, with 57% evident in adult goats. However, percentages of sheep livers with Fe levels above 600 and 1000 mg/kg DM in the dry and wet seasons were 37% and 21%, and 35% and 15%, respectively. Percentages of sheep kidneys with Fe levels above 400 mg/kg DM were 32% and 30% in the dry and wet seasons, respectively.

Manganese in sheep was lower in liver tissues (*P*<0.05) in the dry than in the wet season.Twenty one percent of sheep livers were Mn deficient (<6 mg/kg DM) in the dry season. Brain Mn was higher (*P*<0.05) in growing lambs (3.6) than in adult (2.9 mg/kg DM) sheep. However, tissue Mn did not vary with season or age in goats.

4. Discussion

The absence of seasonal variation in liver Cu in sheep and goats contrasted with the higher wet season incidence of liver Cu deficiency reported for beef cattle of Malawi (Mtimuni et al., 1990), St. Croix (Wildeus et al., 1992) and Ethiopia (Khalili et al., 1993b). The higher incidence of liver Cu deficiency found in adult than in growing sheep was probably an indication that Cu storage reserves had declined with age. However, goats were deficient regardless of season or age.

Liver and kidney Cu levels in abattoir goats were similar to those found in normal goats from Oman (Ivan et al., 1990. However, liver Cu levels were lower than those reported for slaughter housed goats from Eastern Sudan (Abu Damir et al., 1983) and Western Sudan (Tartour, 1975), but higher than that of goats from Kenya (Hedger et al., 1964). Liver Cu levels in sheep were lower than those reported in Western and Eastern Sudan (Tartour, 1975; Abu Damir et al., 1983) but higher than those reported for Kenya (Hedger et al., 1964) and Ethiopia (Faye et al., 1991). Kidney Cu levels in sheep were lower than that of sheep from Eastern Sudan (Abu Damir et al., 1983), whereas brain Cu was similar to that reported in the U.K. by Lewis et al., (1974).

The higher incidence of marginal liver Zn deficiency in sheep in the wet than the dry season is analogous to the findings for beef cattle from Malawi (Mtimuni et al., 1990). Liver Zn levels in sheep in the dry season expressed on a wet weight basis were similar to findings for slaughter housed sheep from Eastern Sudan (Abu Damir et al., 1993) and for growing lambs kept on Zn sufficient diets (Ott et al., 1965). However liver Zn (w.wt) in the wet season was similar to that of growing lambs kept on Zn deficient diets (Ott et al., 1965), and of Australian sheep (Langlands et al, 1987). Kidney Zn (w.wt) in sheep in the wet season was also lower than that of Eastern Sudan (Abu Damir et al., 1983) and Australia (Langlands et al., 1987).

The higher serum Zn found in growing than adult sheep was probably associated with increased intestinal absorption rates in young animals (Miller et al., 1968). In contrast the higher incidence of marginal liver Zn deficiency in growing lambs probably represents a more long term effect of their liver Zn status (Binnerts, 1989).

In goats, liver and kidney Zn levels in the dry season were similar to those reported for normal goats (Groppel and Hennig, 1971). However, liver Zn levels in the wet season were probably marginally deficient, since levels were similar to those reported for kids born from dams kept on Zn deficient diets (Groppel and Hennig, 1971). Brain Zn in goats was similar to those reported by Hainlein (1980), while that of sheep was lower than the expected value of 13 mg/kg DM as suggested by Davis and Mertz (1986). The apparent anomaly of high serum marginal Zn deficiency observed in sheep and goats in the dry than in the wet season probably represents a short term effect of dietary Zn intakes.

The low Fe status in liver of goats suggested that Fe storage was low and implied that a higher incidence of anaemia probably existed in this species. The relatively high Fe found in some livers and kidneys of sheep at both seasons suggested that Fe-Cu imbalances may have existed at the tissue level in these animals (Woolliams et al., 1986b) probably associated with the occasional manifestation of swayback lambs from adult ewes. The level of Mn deficiency in sheep in the dry season resembled findings for beef cattle from the southwest region of Guatemala (Tejada et al., 1987). The higher brain Mn found in growing than in adult sheep was probably age related. In both seasons, liver Cu in sheep and goats and liver Fe in goats, including wet season liver Zn in sheep and goats were probably limiting animal production.

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