Vol 19 No 2, p 277-281

EFFECT OF COPPER SUPPLEMENTED SALT LICKS ON TOTAL AND TCA-SOLUBLE PLASMA COPPER CONCENTRATIONS IN OMANI CAMELS

Nur El Huda I.E.D. Osman
Department of Animal and Veterinary Sciences, College of Agriculture and Marine Science,
Sultan Qaboos University, Muscat, Oman

ABSTRACT

Fifty Omani camels raised in the Directorate General of Camels Affairs, Royal Affairs, Oman, half of which were given mineral salt licks containing copper (Cu-SL), and half were provided with salt licks without Cu (nCu-SL),were bled to study their plasma copper status. The camels were maintained on dates and fresh alfalfa; both contained adequate Cu levels at 7.57 and 9.4mg/kg DM, respectively. Means of total plasma Cu (TCu) in both groups were within deficient to marginal levels in other ruminants. Mean TCu was higher ($p=0.059$) in nTCu-SL than TCu-SL while TCA-soluble plasma Cu (TCA-Sol) was higher ($p=0.055$) in the TCu-SL than in the nTCu-SL. The proportion of TCA-sol/TCu was significantly higher ($p<0.001$) in Cu-SL. Total plasma Cu was significantly higher than TCA-soluble Cu ($p<0.001$) suggesting possible presence of thiomolybdates in the camels' plasma. High dietary Mo and S, and increased Fe were suggested to be the most possible cause of low Cu levels found in the camels.

Key words: Camel, Oman, plasma copper, TCA-soluble copper

Copper (Cu) is an important trace element that is involved in many biological activities including growth and development, (Harris, 1983; Underwood and Suttle, 2001), fertility and reproduction (Hidiroglou, 1979). The deficiency of Cu is manifested by many clinical symptoms including ataxia (Underwood and Suttle, 2001). Although Cu deficiency was reported in Omani livestock (Ivan *et al,* 1990; El Hag, 1990), these reports did not include camels. Personal communications with veterinarians in different Omani regions revealed that clinical symptoms of Cu deficiency were also seen in camels. The causes of Cu deficiency in Oman are not yet fully identified. However, the limited reports available suggest that the increased dietary levels of iron (Fe) (Ivan *et al,* 1990) and possible increased Mo and S (Osman, *et al,* 2000) may be within these causes. Salt licks with Cu are widely used in many Omani farms. No studies have been made to find the normal plasma Cu or the effects of these Cu-SL on plasma Cu status of Omani camels. This study was aimed to determine plasma Cu levels in camels kept under intensive system in Oman.

Materials and Methods

Animals

Present study was done on fifty female camels, (n=50) all above 8 years of age, were picked at from a large herd raised in the farms of the Directorate General of Camels Affairs, Ministry of Royal Affairs in Oman, north of Muscat. The herd, which was kept for racing and festival shows, was housed in partially shaded,well ventilated 30 X 100 m large pens at the rate of 42-50 camels/pen. The group-fed camels were offered fresh alfalfa (20 kg/head/d) and dates (5 kg/head/d), both were grown in farms belonged to the Diwan of Royal Court. The camels were also given mineralised salt licking blocks containing copper (Cu-SL) (300 mg Cu/kg, Red Mineral Blockies, Redency House, UK) *ad libitum*. Twenty five camels had the salt-licks without Cu (nCu-SL). Cu, Fe and Zn contents of the dates, alfalfa and the salt licks are presented in Table 1. Fresh water was freely provided. The animals were drenched with anthelmentic twice a year.

Table 1. Some mineral contents (mg/kg DM) in alfalfa, dates and salt blocks provided to camels.

	Copper	Iron	Zinc
Alfalfa	9.4	566.3	13.8
Dates	7.57	317.1	8.7
Salt block	300	1500	300

Chemical Analysis

Blood samples were collected in heparinised vaccutainer tubes from the jugular veins of the

SEND REPRINT REQUEST TO NUR EL HUDA I.E.D. OSMAN email: hudaisam@gmail.com

camels. Blood samples were centrifuged within one hour after collection, plasma separated and frozen until analysed. Total and TCA-sol Cu concentrations were measured in plasma samples using Atomic Absorption Spectrophotometry (AAS) after dilution of plasma in 6% butanol (1:2) for TCu and precipitation of proteins in 5% (v/v) trichloroacetic acid (TCA) (1:2) for TCA-sol.

Dried samples of dates and alfalfa were analysed for Cu, Fe and Zn using AAS after dry digestion of material following a method based on that described by AOAC (1990).

Statistical analysis

The SPSS statistics 17 (2008) personal computer package was used for the statistical analyses. Analysis of variance was used to study the effects of Cu-SL on the measured TCu and TCA-sol (mg/l) and the calculated proportion of TCA-sol/TCu. T-test was used to study the difference between TCu and TCA-sol levels, and the correlation of these two levels within each camel group was done.

Results

In both groups of camels the mean TCu concentrations were within deficient to marginal levels (Table 2). Mean TCu concentration was higher (P<0.059) in nCu-SL than in Cu-SL camels. Mean TCA-soluble Cu was higher (P = 0.055) in the Cu-SL than the nCu-SL camels which resulted in a significantly higher (p<0.001) mean proportion of TCA-sol/TCu in Cu-SL than nCu-SL camels.

Table 2. Means (± SE) of measured TCu, TCA-soluble (mg/l) and the calculated proportion of TCu/TCA-soluble Cu in camels provided with salt licks not containing Cu (nCu-SL) or containing Cu (Cu-SL).

	nTCu-SL±SE	TCu-Sol±SE	Significance
TCu (mg/l)	0.65±0.030	0.55±0.045	P=0.059
TCA-sol (mg/l)	0.27±0.042	0.39±0.042	P=0.055
TCA-sol/TCu	0.41±0.061	0.74±0.063	***

*** :P<0.001.

There were significant differences between means of TCu and TCA-sol within groups of bled camels (Table 4), as indicated from the t-values. There was a significant correlation between TCu and TCA-sol in TCu-SL(P<0.001) but not in the nCu-S (P>0.05) groups.

Discussion

The major finding of this study was that a large proportion of camels, including those had Cu supplement in salt lick, had TCu levels within marginal to deficient levels, as suggested for cattle (Perry, 1980; Vijchulata *et al,* 1991). The TCu means of pooled total groups, as well as for that for the camels within normal range, was below levels reported in camels elsewhere (e.g. Naway, 1983; Moty *et al,* 1968). For instance, TCu mean levels in the current study were only 0.55 ± 0.05 – 0.65 ± 0.03 mg/l in the Cu-SL and nCu-SL, respectively, means of 0.9 – 1.0 mg/l were found in the dromedary camels in Sudan (Naway, 1983), 0.83 ± 0.067 mg/l in Egypt (Moty *et al,* 1968), 0.86 ± 0.24 mg/l in bactrian camels in China (Zong-Ping *et al,* 1994) and 94.3 ± 3.2 µg/100 ml in India (Ghosal and Shekhawat, 1992). Higgins and Kock (1986) reported values of 0.9 – 1.0 mg/l for the dromedary and 0.8 - 0.9 mg/l for bactrian camels. On the other hand a level of 57.6-72.4 µg/100 ml was reported in Sudanese adult camels (Mohamed, 2004), A 60.1 µg/100 ml was also recorded in Arabian camel in a neighbourig country, the UAE (Faye, *et al,* 2008), while a value of 20.5 µg/100 ml was reported in Iranian 5 year old female camels (Badiei *et al,* 2006). Furthermore, serum levels of 55.8-110.0 µg/100 ml were recorded in racing Arabian camels in the eastern region of this country in a recent report by Eltahir *et al* (2010), which confirms the findings of the present study. The dietary levels of Cu found in diet components given to the camels under investigation were adequate, which suggests secondary causes to influence plasma TCu levels in Omani camels.

Although the effect of the Cu-SL on the available type of plasma Cu, the TCA-sol, was significant, it was not enough to raise the TCu levels to the normal range. Findings of this study suggest the presence of high dietary molybdenum (Mo) and sulphur (S) levels in the camel feeds but the diets were not analysed for either of these elements. The evidence of this suggestion is substantiated by the reduction of the proportion of TCA-sol/TCu and the significant differences between TCA-sol and TCu levels, which indicates a presence of thiomolybdates (TM) in the camels' plasma. Thiomolybdates are compounds that form in the rumen at the presence of high dietary Mo and S. A series of these compounds, known as di-, tri- or tetrathiomolybdates, result from a reaction of H_2S and molybdate ions in dilute neutral aqueous solutions to form a series of salts of type R 12Mo04_nSn where n=1 to 4 (Aymonino *et al,* 1969). These compounds were detected in plasma of sheep and deer maintained on high Mo and S diets (Osman, 1988). It is important to point out, however,

Table 3. Means (± SE) of measured TCu, TCA-soluble (mg/l) and the calculated proportion of TCu/TCA-soluble Cu in two groups of camels and that had their TCu within ranges of normal (≥ 0.70 mg/l) or low (< 0.70 mg/l).

Salt lick provided	nTCu-SL		TCu-SL	
TCu level (mg/l)	Normal (≥ (0.70)	Low (<0.7)	Normal (>0.699)	Low (<0.7)
N	9	16	6	19
TCu (mg/l)	0.82±0.042	0.56±0.034	0.76±0.055	0.47±.031
TCA-sol (mg/l)	0.39±0.059	0.20±0.044	0.60±0.072	0.30±0.041
TCA-sol/TCu	0.49±0.104	0.36±0.078	0.80±0.128	0.72±0.072

that the dietary levels of both Mo and S are important to define the TCu level in the affected animals. For instance, dietary Mo at levels less than 11.4 to 2.4 mg/kg and S at 2.3% DM reduced the TCu in both sheep and deer to variable degrees while higher levels of dietary Mo increased the TCu in both animal species (Osman and Sykes, 1989). The reduction in blood Cu to the level of 0.28 ± 0.17 mg/l was also observed in Chinese Bactrian camels fed on pastures containing Mo at 4.8 ± 0.25 mg/kg DM and Cu at 6.5 mg/kg DM (Zong-Ping, *et al,* 1994). Ivan *et al* (1990) reported a Mo level of 2.8 Mg/kg DM and 0.37 % of S in alfalfa samples in Oman. More recently Abu Damir *et al* (2008) reported a Mo and S content in Alfalfa as 2.2 mg/kg and 4.7 g/kg DM, respectively. These levels were high enough to affect Cu metabolism in sheep (Suttle, 1974). Furthermore, the fodder given to the animals under investigation was fresh alfalfa, a form suggested to further reduce the availability of dietary Cu in the presence of low levels of Mo compared to corresponding increases in hay (Suttle, 1978).

The high dietary iron levels in the diets and in the salt licks could further complicate reduce plasma levels in the camels in this study. The dietary iron levels in both alfalfa and dates given to these camels were within ranges that affect Cu metabolism in cattle (Bremner *et al,* 1983).

Cu salt licks given to some of the camels produced a significant improvement on the type of Cu form in the camels' plasma, the TCA-soluble form. The latter type comprise the form of plasma Cu that dissociate from protein upon application of trichloreacetic acid (TCA) and it is readily available for use by the animal body functions. This dissociation (solubility) reduces in the presence of thiomolybdates (Dick *et al*, 1975). This suggests that the higher level of Cu taken by these animals reduced the amount of free thiomolybdates in the rumen that was absorbed in the blood. Another evidence is the higher TCu recorded in the nCu-SL than in the Cu-SL camels, with lower TCA-sol/TCu proportion. The low correlation between TCA-sol and TCu in the nCu-SL camels suggests a high individual variation between camels in their response to the possible extra dietary Mo while the Cu supplementation reduced the individual variations.The amount of Cu in the salt licks, however, was not high enough to alleviate the effect of the secondary causes of Cu deficiency in these camels, especially that the salt licks themselves contained the unnecessary amount of iron. The positive response caused by Cu supplementation, however, with the high correlation between plasma Cu forms measured in this work, may suggest that dietary Cu supplementation to camels can be used to correct Cu deficiency in camels. This suggestion was also confirmed in Chinese camels, with Mo induced Cu deficiency, using Cu-salt blocks (Zonping, 2004), or using other means including oral administration with copper sulphate ((Zon-ping *et al,* 1994) or cupric oxide needle capsules (Abu Damir *et al,* 2008). The use of Cu-SL, however, was not successful in this lab when used in goats (Osman *et al,* 2000) and it was not recommended (Abu Damir *et al,* 2008).

Table 4. The relationships (correlation and t-values) between TCA-soluble and total plasma Cu levels measured in Omani camels given salt licks with or without Cu.

	Correlation	Significance	t-value	Significance
All camels	0.378	**	- 8.235	***
With Cu-SL	0.605	***	- 4.062	***
Without Cu-SL	0.337	P=0.99	- 8.913	***

**: P<0.01; *** :P<0.001.

It is worthwhile reporting that the apparently healthy Cu deficient animals can have some performance parameters affected including growth (Osman *et al,* 2008) or haematological parameters (Osman *et al,* 2009). Camels were found very tolerant to high dietary levels of Cu (Kinne *et al,* 2005) that indicates further increases in dietary Cu supplementation may be implemented with no hesitation. However, further studies are encouraged to specify the Cu status, normal plasma Cu concentrations, causes of Cu deficiency, implications

of Cu deficiency and best methods of alleviation of Cu deficiency in camels of Oman.

Acknowledgement

The author would like to thank Professors B.M. Musa and M.T. Abusamra (Royal Camel Affairs) for providing samples used in this study and toProfessor E. Johnson and Mr Andrew Ritchie (AVS) for advice and technical assistance.

References

Abu Damir H, Abbas TA and Alhaj Ali M (2008). Copper status in breeding and racing camels (*Camelus dromedarius*) and response to cupric oxide needle capsules. Tropical Animal Health and Production. 40: 643-648.

Aymonino PJ, Ranade AC, Dieman E and Müller A (1969). Study of the formation and relative reaction rates of different thioanions of molybdenum and tungsten. ZeitschriftfürAnorganische und Allgemeine Chemie 371:300-305.

Badiei K, Salah Mostaghani K, Pourjafar M and Parchami A (2006). Serum and tissue trace elements in Iranian camels (*Camelus dromedarius*). Comparative Clinical Pathology 15:58-61.

Bremner I, Phillipo M, Humphries WR, Young BW and Mills CF (1983). Effects of iron and molybdenum metabolism in calves. In: Trace Elements in Animal Production and Veterinary Practice: Occasional Publication No. 7 – British Society of Animal Producton. Edited by N.F. Suttle, R. G. Gunn, W. M. Allen, K.A. Linklater and G. Wiener.

Dick AT, Dewey DW and Gawthorne JM (1975). Thiomolybdates and the copper–molybdenum–sulphur interaction in ruminant nutrition. The Journal of Agricultural Science 85:567-568.

El Hag MG (1990). Some observations on an incident of copper deficiency in local dairy calves in the Sultanate of Oman. Sudan Journal of Animal Production 3:53-56.

Eltahir YE, Ali HM, Mansour MH and Mahgoub O (2010). Serum mineral contents of Omani racing Arabian camels (*Camelus dromedarius*). Journal of Animal and Veterinary Advances 9:764-770.

Faye B, Seboussi R and Askar M (2008). Trace elements and heavy metals status in Arabin camel. In: Impact of pollution on animal products, NATO Sciences for Peace and Security Series C: Environmental Security. Springer, Netherlands. pp 97-106.

Ghosal AK, Shekhawat VS (1992). Observations on serum trace elements levels (zinc, copper and iron) in camel (*Camelus dromedarius*) in the arid tracts of Thar Desert in India. Rev Elev Med Pays Trop, cababstractplus.org.

Harris ED (1983). Copper in human and animal health. In: Trace Elements in Health – A Review of Current Issues. Edited by J. Ross. Butterworths. pp 44-73.

Hidiroglou M (1979) Trace element deficiencies and fertility in ruminants: a review. Journal of Dairy Science 62(8):1195-1206.

Higgins AJ and Kock RA (1986). 1. A guide to the clinical examination, chemical restraint and medication of the camel. In: The Camel in Health and Disease. Edited by Andrew Higgins. Published by Bailliere Tindall, London. pp 21-40.

Ivan M, Hidiroglou M, Ismaily SI, Al-Sumry HS and Harper RB (1990). Copper deficiency and posterior paralysis (Shalal) in small ruminants in the Sultanate of Oman. Tropical Animal Health and Production 22:217-225

Kinne J, Nag P and Wernery U (2005). Serum copper levels in dromedaries after long exogenous copper supplementation. The Veterinary Journal 169:444-453.

Mohamed HE (2004). The Zinc and Copper content of the plasma of Sudanes camels (*Camelus dromedarius*). Veterinary Research Communications 28:359-363.

Moty IA, Muller Ael and Zaafer SA (1968). Copper, iron and zinc in the serum of Egyptian farm animals. Sudan Agriculture Journal 3:146-151.

Naway NG (1983). Studies on the normal clinical values, haematology and serum constituents of the one humped camel (*Camelus dromedarius*). MVSc thesis, May 1983; Dept of Medicine, Pharmacology and Toxicology, Faculty of Veterinary Science, University of Khartoum.

Osman NHI (1988). Comparative Studies on the Effects of Molybdenum and Sulphur on Copper Metabolism in Sheep (*Ovis aries*) and Red Deer (*Cervus elaphus*), PhD thesis, Lincoln College, University of Canterbury, New Zealand, September 1988.

Osman NHI and Sykes AR (1989). Comparative effects of dietary Molybdenum concentration on distribution of copper in plasma in sheep and red deer (*Cervus elaphus*). Proceedings of the New Zealand Society of Animal Production 49:15-19

Osman NHI, Johnson EH and Al-Busaidi RM (2000). Plasma copper status in kids. 2. Total and TCA-soluble levels in kids given copper-salt lick. Volume 1 in Proceedings of International Conference on Goats. 15-18 May 2000, Tours, France. pp 99.

Osman NHI, Al-Busaid RM and Johnson EH (2009). The effect of breed, sex and total plasma copper level on haematology of growing kids of three Omani goat breeds, 13th Seminar of the FAO-CIHEAM Sub-Network on Sheep and Goat Nutrition. León, Spain, October 14-16, 2009.

Perry TW (1980). Mineral requirements of beef cattle. In: Beef cattle feeding and nutrition. Academic Press, New York. pp 33-48.

Suttle NF (1974). Effects of molybdenum and sulphur at concentrations commonly found in ruminant diets on the availability of copper in sheep. In: Hoekstra, W.G., Ganther, H.E.; Mertz, W.; Editors. Trace element metabolism in animals-2 Baltimore: University Park Press. pp 612-614.

Suttle NF (1978). Effects of sulphur and molybdenum on the absorption of copper from forage crops by ruminants. In: J.C. Borgan; Editor. Proceedings, Symposium on Sulphur in Forages, Waxford, Ireland. Dublin: An Foras Taluntais. pp 179-211.

Underwood EJ and Suttle NF (2001). Copper. In: The mineral Nutrition of Livestock. CABI Publishing, UK. pp 238-342.

Vijchulata P, Yaowapaksopon J and Thanindratarn B (1991). Mineral status in Chieng Mai province. In: Proceedings of the Third International Symposium on the Nutrition of Herbivores. Rasa Sayang Hotel, Peneng, Malaysia, 25-30 August 1991. Compiled by: M. Wang Zahari, Z. Ahmad Tajuddin and H.K. Wong. Published by The Malasian Society of Animal Production (MSAP), c/o/ Department of Animal Science, UniversitiPertanian Malaysia, 434000 UPM, Serdang, Selengor, Malaysia. pp 54.

Zonping L (2005). Studies on rickets and osteomalacia in bactrian camels (*Camelus bactrianus*). The Veterinary Journal 169:444-453

Zong-Ping L, Zhuo M and You-Jia Z (1994). Studies on the relationship between sway disease of Bactrian camels and copper status in Gansu province. Veterinary Research Communications 18:251-260.