



SHORT COMMUNICATION

Trace Minerals and Biochemical Profile in Buffalo Calves Manifesting Coat Colour Depigmentation in the Fluoride Endemic South-West Punjab

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ABSTRACT

The present work aimed to study trace minerals and haemato-biochemical profile of buffalo calves manifesting depigmentation of coat colour (depigmented calves: DC) in comparison to calves having normal coat colour (normal calves: NC) in the fluoride endemic zone of the South-West Punjab. Plasma fluoride concentrations in NC and DC were higher than the normal but it did not vary between the groups. Plasma copper concentration in DC was lower than the critical limit of $0.70 \mu\text{gml}^{-1}$ and it was significantly ($p < 0.05$) lower as compared to NC. Hair copper and plasma ceruloplasmin activity did not vary between the groups. Plasma molybdenum, and plasma and hair zinc and manganese concentrations were normal and did not vary between the groups. Plasma iron concentrations in both the groups were higher than the physiological limit of $2.50 \mu\text{gml}^{-1}$. The hair iron concentrations were significantly ($p < 0.05$) higher in DC. The Hb, PCV, TEC, serum total proteins, albumin, globulin, plasma urea nitrogen, creatinine, glucose, cholesterol, triglycerides, ALP, AST, CK and GGT were normal in both the groups. It is concluded that depigmentation of coat colour in buffalo calves from the fluoride endemic South-west Punjab is due to deficiency of copper.

Keywords: Depigmentation, buffalo calves, trace minerals, fluoride, copper

The South-West region in the Punjab State of India is a fluoride (F) endemic zone. There had been reports of depigmentation of coat colour in buffalo calves from this region (Ozukum, 2012). Depigmentation of coat colour in ruminants is indicative of underlying nutritional deficiency particularly that of trace minerals viz. copper (Cu) and zinc (Zn) (Suttle, 2010) as these minerals play crucial part in melanin synthesis. In F endemic areas, ruminants may suffer from Cu and Zn deficiency due to antagonistic effects of F on Cu and Zn metabolism. The present work aimed to study status of trace minerals in buffalo calves manifesting depigmentation of coat colour in the F endemic zone of the South-West Punjab.

The study was conducted in the Mansa and Fazilka districts of the South-West Punjab, India. Dairy farms from the area were visited and buffalo calves (6-12 months age) manifesting depigmentation of hair coat were selected (depigmented calves: DC, $n=12$). Few calves

with normal hair and identical age and weight from the same herds were taken as control (normal calves: NC; $n=63$). Blood samples of the normal and depigmented calves were collected from jugular vein in heparin for plasma separation, without anticoagulant for serum harvesting and in EDTA for haemogram analysis. Hair samples were collected from the tail switch by using a stainless steel scissor. Plasma samples were wet digested as per Kolmer *et al.* (1951) and concentrations of Cu, Zn, iron (Fe), manganese (Mn) and molybdenum (Mo) were estimated by atomic absorption spectrophotometry (AAS, PerkinElmer A Analyst 700, USA). Plasma F was estimated by digital ion analyser (Orion 4 star bench top pH ISE meter, Thermo Scientific, Singapore) and F electrodes (Thermo Scientific, USA). Hair samples were washed as per IAEA (1978), wet digested as per Hershey *et al.* (1988) and concentrations of Cu, Zn, Mn and Fe were estimated by AAS. Activity of ceruloplasmin (Cp) in

plasma was estimated by the method of Houchin (1958). Red cell parameters (Hb, PCV, TEC) were estimated by Advia 2120 Haematology System (Siemens Medical Solutions Diagnostics, USA). Serum total proteins were analysed by Biuret method (Reinhold, 1953). Serum albumin and plasma urea nitrogen, creatinine, glucose, cholesterol, triglycerides, aspartate aminotransferase (AST) and gamma-glutamyl transpeptidase (GGT) were estimated on Microlab 300 (Merck, Netherlands) using diagnostic kits (Merck Specialties Pvt. Ltd., Goa). Serum globulin concentrations were calculated by subtracting values of serum albumin from the values of serum total proteins. Plasma alkaline phosphatase (ALP) and creatine kinase (CK) were estimated on System Vitros DT6011 Chemistry (Ortho-Clinical diagnostics, Johnson and Johnson, USA). Mean values for different parameters were calculated and compared between the study groups by using student's t-test using SPSS (Statistical Package for Social Sciences) for Window version 11.0.1[©] SPSS Inc. USA computer software program.

Trace minerals and haemato-biochemical profile of normal (NC) and depigmented calves (DC) are presented in Table 1 and 2. Plasma F concentrations in depigmented and normal calves were higher than the physiological limit of 0.10 μgml^{-1} , however the F concentrations did not vary between the groups. Higher intake of F primarily through drinking water was probably responsible for the higher plasma F contents in the calves. Plasma Cu concentrations in DC were significantly ($p<0.05$) lower as compared to NC. Moreover, the plasma Cu concentrations in Group II were lower than the critical limit of 0.70 μgml^{-1} given by Kincaid (1999) thus indicated Cu deficiency in the DC. However, hair Cu concentrations and plasma Cp activity (an indicator of functional Cu status) did not differ significantly between the study groups. Copper is an essential micronutrient required for melanization of hair coat. In Cu deficiency, reduced amino oxidase activity results in failure of conversion of tyrosine to melanin that leads to coat colour depigmentation in animals (Suttle, 2010). Several researchers had attributed coat colour depigmentation to hypocupraemia in dairy animals (Randhawa *et al.* 2006). On the contrary, Mee (1991) concluded that hair depigmentation was not a clinical sign of Cu deficiency in dairy animals. The plasma Mo levels were normal (i.e. $<300.0 \text{ ngml}^{-1}$) in both the groups, thus the Mo induced secondary Cu deficiency in these animals

was ruled out. Randhawa *et al.* (2009) had reported no variation in plasma Cu concentration between leucodermic and normal buffaloes. However, they observed significantly higher plasma Mo levels in the leucodermic buffaloes as compared to the normal buffaloes and suggested that the higher Mo levels in the leucodermic buffaloes were probably responsible for rendering the Cu physiologically unavailable as Mo forms complexes with Cu.

Table 1: Trace minerals profile of normal and depigmented buffalo calves (Mean \pm SE)

Parameter	Normal calves	Depigmented calves
	(n=63)	(n=12)
Plasma		
Cu (μgml^{-1})	0.71 \pm 0.01	0.65 \pm 0.07*
Mo (ngml^{-1})	79.88 \pm 15.78	94.55 \pm 30.70
Zn (μgml^{-1})	1.33 \pm 0.01	1.79 \pm 0.19
Fe (μgml^{-1})	3.67 \pm 0.28	2.53 \pm 0.52*
Mn (ngml^{-1})	69.55 \pm 7.90	61.47 \pm 18.32
F (μgml^{-1})	0.17 \pm 0.04	0.21 \pm 0.04
Hair		
Cu	6.02 \pm 0.18	6.11 \pm 0.49
Zn	60.92 \pm 2.68	67.18 \pm 9.93
Mn	27.86 \pm 4.10	42.89 \pm 13.08
Fe	204.16 \pm 38.04	369.67 \pm 167.89*
Cp activity (mgdl^{-1})	10.58 \pm 0.62	10.55 \pm 1.52

*Significant at ($p<0.05$).

Concentrations of Zn in plasma and hair were normal and did not vary between the study groups. Thus any role of Zn in causing the depigmentation in calves was ruled out. Zinc had been reported to play a part in the melanin synthesis and low levels of Zn had been recorded in skin of the human vitiligo patients (Kedar *et al.* 1978). Moreover, Randhawa *et al.* (2009) had observed that the supplementation of zinc oxide prevented relapse of leucoderma in cattle. Plasma Fe concentrations in both the study groups were considerably higher than the physiological limit of 2.50 μgml^{-1} , which could be due to high dietary intake of Fe through forages. Excess of Fe had been reported in the soil-plant system in the most parts of India (Bhandari *et al.* 2006). Significantly ($p<0.05$) higher hair Fe concentrations in the DC were in agreement with Randhawa *et al.* (2009). They related increased Fe contents of hair of leucodermic buffaloes with Cu deficiency that caused impairment of Fe metabolism and

thus its deposition in tissues including hair. Plasma and hair Mn concentrations did not vary between the normal and depigmented calves.

Table 2: Haemato-biochemical profile of normal and depigmented buffalo calves (Mean±SE)

Parameter	Normal calves	Depigmented calves
	(n=63)	(n=12)
Hb (gdl ⁻¹)	9.72±0.21	9.71±0.49
PCV (%)	31.03±0.75	31.09±1.52
TEC (x10 ⁶ µl ⁻¹)	7.79±0.23	7.30±0.32
Total proteins (gdl ⁻¹)	7.08±0.14	7.65±0.30
Albumin (gdl ⁻¹)	2.97±0.10	3.34±0.29
Globulin (gdl ⁻¹)	4.11±0.14	4.31±0.32
Urea nitrogen (mgdl ⁻¹)	11.06±0.51	10.32±1.62
Creatinine (mgdl ⁻¹)	1.34±0.04	1.18±0.07
Glucose (mgdl ⁻¹)	78.78±2.17	68.26±4.82*
Cholesterol (mgdl ⁻¹)	123.59±4.77	114.67±10.79
Triglycerides (mgdl ⁻¹)	13.97±1.47	13.62±3.49
ALP (uL ⁻¹)	228.94±17.25	201.17±49.28
AST (uL ⁻¹)	64.23±3.75	64.50±7.81
GGT (uL ⁻¹)	6.85±0.83	6.08±0.84
CK (uL ⁻¹)	175.51±10.23	171.17±20.46

*Significant at (p<0.05).

Haemogram (i.e. Hb, PCV, TEC), protein-energy status (serum total proteins, albumin, globulin, plasma urea nitrogen, creatinine, glucose, cholesterol and triglycerides concentrations), and activities of several enzymes suggestive of tissue injury (i.e. ALP, AST, CK, GGT) were normal and did not vary between the study groups, which ruled out protein-energy malnutrition or any active inflammatory process in the buffalo calves.

SUMMARY

Trace minerals and haemato-biochemical profile of buffalo calves manifesting depigmentation of coat colour was assessed in comparison to normal calves in the fluoride endemic zone of the South-west Punjab. Copper deficiency was detected in the depigmented calves; whereas excess of F and Fe, and normal Mo, Zn, Mn, haemogram and protein-energy status was observed in normal and depigmented calves. It is concluded that depigmentation of coat colour in buffalo calves from the fluoride endemic South-west Punjab may be due to deficiency of copper.

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