



Haematological profile of subchronic oral toxicity of molybdenum in buffalo calves*

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In Punjab, areas around Sutlej belt (Ludhiana) are rich in molybdenum (Mo), so forages especially legumes grown on such soils serve as major source of molybdenosis in ruminants (Nayyar *et al.* 1977, Radostits *et al.* 2007). However, despite the association of chronic molybdenosis with haemopoietic disorders and alterations in other dairy animals (Sharma 2002, Radostits *et al.* 2007), little information is available on influence of long-term administration of excess dietary Mo on the haematological parameters in buffalos. Therefore, the present study was undertaken to investigate the impact of subchronic oral Mo toxicosis on various haematological parameters in buffalo calves.

An experimental trial was conducted on 8 healthy buffalo calves (80–150 kg) aged between 6 and 12 months, randomly divided into 2 equal groups. No salt was administered to the animals of control group whereas based on the results of the preliminary study animals of treatment group were orally administered sodium molybdate @ 3 mg/kg/day for 90 consecutive days. Concurrently, the same sets of animals were also maintained to investigate physical and biochemical alterations with excess dietary Mo (Varun *et al.* 2011).

To study the haematological indices, blood was collected in EDTA containing glass tubes on 0, 10, 20, 30, 40, 50, 60, 70, 80, 90 days of treatment period by jugular venepuncture. Various haematological parameters, viz. haemoglobin concentration (Hb), packed cell volume (PCV), erythrocyte sedimentation rate (ESR), total erythrocyte count (TEC), total leukocyte count (TLC), mean corpuscular haemoglobin

(MCH), mean corpuscular haemoglobin concentration (MCHC), and mean corpuscular volume (MCV) were analyzed in the blood as per standardized methods (Benjamin 2001). The collected data were subjected to the statistical analysis using t-test and results were expressed as mean±SE. Statistical significance was considered at $P \leq 0.01$ and $P \leq 0.05$ levels as per Snedecor and Cochran (1994).

The data pertaining to the long term effect of sodium molybdate (3 mg/kg/day for 90 consecutive days) on haematological parameters are presented in Tables 1 and 2. Daily oral administration of sodium molybdate resulted in a significant decline in the values of haemoglobin in buffalo calves. In the treatment group, maximum decline was observed to the extent of 47.3%, from pre-exposure level of 14.38 ± 0.09 g/dl to 7.57 ± 0.48 g/dl on 60th day of treatment period (Table 1). Our results on haemoglobin concentration were in agreement with Randhawa (1993) who reported a decrease in haemoglobin content in ruminants exposed to high Mo for long periods. The decreased haemoglobin content may be attributed to the defective iron metabolism and conditioned copper deficiency, which in turn is responsible for functional integrity of hematopoietic system (Radostits *et al.* 2007). Dietary sodium molybdate did not produce any significant change in the values of PCV. Non observance of any differences in PCV values in molybdenotic and control buffalo calves adds validity to the findings of Randhwa (1993).

Long term oral administration of sodium molybdate produced significant increase in ESR (Table 1). This finding may be attributed to enhanced rouleaux formation that is seen in several pathological conditions and toxicities (Brar *et al.* 2000). It was evident in our experiment that subchronic molybdenosis produced significant decline in TEC from the pre-exposure level of $7.10 \pm 0.95 \times 10^6/\text{mm}^3$ to $4.20 \pm 0.22 \times 10^6/\text{mm}^3$, a decline by 40.8% on the 40th day of treatment while no significant alteration was found in control animals (Table 1). Similar observations were recorded by Sharma (2002) consequent to Mo intoxication in cattle calves. The most probable reason behind this decrease was the failure of erythropoiesis that originated by induced copper deficiency and defective iron metabolism (Radostits *et al.* 2007).

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In the current investigation, subchronic molybdenosis produced significant decline in TLC when compared with control group (Table 2). In contrast, Randhawa (1993) reported a nonsignificant decline in TLC values. Additionally, significant differences were also recorded between treatment and control group for MCH, MCHC and

Table 1. Effect of repeated oral administration of sodium molybdate (3 mg/kg/day) for 90 consecutive days on haematological indices (mean $\pm$ SE) in buffalo calves

Haematological parameters (units)	Days									
	0	10	20	30	40	50	60	70	80	90
<i>Haemoglobin (g/dl)</i>										
Control	12.87 ± 0.79	11.43 ± 0.99	8.81 ± 0.92	8.34 ± 0.47	9.99 $\pm 0.17^A$	10.89 $\pm 0.65^A$	10.83 $\pm 0.83^A$	10.30 $\pm 0.57^A$	10.58 $\pm 0.42^a$	10.20 $\pm 0.73^A$
Treatment	14.38 ± 0.09	13.93 ± 0.37	11.16 ± 0.60	8.91 ± 0.89	8.65 $\pm 0.41^B$	8.52 $\pm 0.26^B$	7.57 $\pm 0.48^B$	7.82 $\pm 0.47^B$	7.87 $\pm 0.44^b$	7.68 $\pm 0.39^B$
<i>Packed cell volume (%)</i>										
Control	40.41 ± 3.90	32.28 ± 1.56	38.26 ± 2.03	29.90 ± 0.80	28.43 ± 0.32	33.27 ± 1.21	36.29 ± 2.99	35.23 ± 1.23	34.37 ± 1.20	35.93 ± 1.69
Treatment	43.61 ± 1.80	35.91 ± 3.02	36.39 ± 2.27	34.09 ± 2.16	33.49 ± 2.41	30.66 ± 1.12	36.75 ± 1.98	34.77 ± 1.27	35.44 ± 1.80	34.66 ± 1.34
<i>Erythrocyte sedimentation rate (mm/h)</i>										
Control	17.40 ± 2.00	46.80 ± 3.20	20.40 ± 2.20	34.80 ± 2.40	24.00 ± 2.30	27.60 ± 2.60	25.20 ± 2.40	19.20 $\pm 2.20^a$	20.40 $\pm 2.20^A$	18.00 $\pm 2.20^a$
Treatment	24.00 ± 2.40	56.40 ± 3.60	19.80 ± 1.80	28.80 ± 2.60	19.80 ± 1.80	31.80 ± 2.80	33.00 ± 3.00	31.80 $\pm 2.60^b$	31.20 $\pm 3.00^B$	30.60 $\pm 2.60^b$
<i>Total erythrocyte count ($10^6/\text{mm}^3$)</i>										
Control	6.95 ± 0.80	5.80 ± 0.85	4.92 ± 0.43	6.87 $\pm 0.38^a$	6.42 $\pm 0.31^a$	6.75 $\pm 0.42^a$	7.55 $\pm 0.51^a$	7.15 $\pm 0.32^a$	7.40 $\pm 1.00^A$	7.20 $\pm 0.26^a$
Treatment	7.10 ± 0.95	5.40 ± 0.25	6.82 ± 0.83	4.60 $\pm 0.35^b$	4.20 $\pm 0.20^b$	4.92 $\pm 0.08^b$	5.02 $\pm 0.21^b$	5.00 $\pm 0.15^b$	4.95 $\pm 0.17^B$	4.85 $\pm 0.17^b$

^{A,B}Within a column and a haematological variable, values with different superscript alphabets differ significantly ($P \leq 0.05$). ^{a,b}Within a column and a haematological variable, values with different superscript alphabets differ significantly ($P \leq 0.01$).

Table 2. Effect of repeated oral administration of sodium molybdate (3 mg/kg/day) for 90 consecutive days on total leukocyte count and erythrocyte indices (mean $\pm$ SE) in buffalo calves

Haematological parameters (units)	Days									
	0	10	20	30	40	50	60	70	80	90
<i>Total leukocyte count ($10^3/\text{mm}^3$)</i>										
Control	8.30 ± 0.57	6.75 ± 0.27	5.22 ± 0.26	4.60 ± 0.25	4.45 $\pm 0.17^A$	5.65 $\pm 0.22^a$	5.35 $\pm 0.41^a$	9.22 $\pm 0.43^a$	8.40 $\pm 0.30^a$	7.10 $\pm 0.45^B$
Treatment	8.85 ± 1.40	6.05 ± 0.99	5.00 ± 0.25	4.10 ± 0.25	3.50 $\pm 0.30^B$	4.75 $\pm 0.12^b$	3.65 $\pm 0.23^b$	5.90 $\pm 0.15^b$	5.85 $\pm 0.32^b$	5.27 $\pm 0.36^A$
<i>Mean corpuscular haemoglobin (pg/dl)</i>										
Control	17.34 ± 3.15	21.08 ± 1.89	18.59 ± 2.62	12.15 $\pm 0.16^A$	15.98 $\pm 0.72^A$	16.18 ± 1.13	14.56 ± 1.27	14.40 ± 0.41	14.39 $\pm 0.73^A$	14.28 ± 1.04
Treatment	23.38 ± 4.88	26.38 ± 2.18	23.88 ± 3.29	20.16 $\pm 3.17^B$	20.77 $\pm 1.37^B$	17.36 ± 0.75	15.39 ± 1.43	15.66 ± 0.83	23.57 $\pm 3.86^B$	15.92 ± 0.85
<i>Mean corpuscular haemoglobin concentration (g/dl)</i>										
Control	26.97 ± 1.46	36.56 ± 4.68	23.21 ± 2.36	28.25 ± 2.38	33.84 ± 2.98	32.63 ± 2.60	30.43 $\pm 2.40^a$	29.49 $\pm 0.89^a$	31.17 $\pm 2.37^A$	28.66 $\pm 1.30^A$
Treatment	33.17 ± 2.24	38.78 ± 0.39	22.52 ± 3.05	25.92 ± 1.91	26.33 ± 1.55	28.07 ± 1.70	20.46 $\pm 0.99^b$	24.69 $\pm 1.64^b$	22.38 $\pm 1.20^B$	22.56 $\pm 1.38^B$
<i>Mean corpuscular volume (fl)</i>										
Control	63.09 ± 8.89	65.03 ± 11.81	78.04 ± 7.18	44.54 $\pm 3.40^a$	44.73 $\pm 2.35^a$	50.54 $\pm 2.42^A$	449.08 $\pm 4.24^A$	50.06 $\pm 3.21^a$	46.87 $\pm 4.11^a$	50.79 $\pm 3.48^A$
Treatment	71.20 ± 13.45	67.87 ± 5.12	57.52 ± 8.43	76.03 $\pm 6.57^b$	81.31 $\pm 9.40^b$	62.50 $\pm 2.28^B$	74.45 $\pm 7.14^B$	69.54 $\pm 1.63^b$	71.75 $\pm 3.38^b$	72.47 $\pm 5.45^B$

^{A,B}Within a column and a haematological variable, values with different superscript alphabets differ significantly ($P \leq 0.05$). ^{a,b}Within a column and a haematological variable, values with different superscript alphabets differ significantly ($P \leq 0.01$).

MCV (Table 2) when buffalo calves were exposed to Mo rich diet. Furthermore, MCH values declined significantly from 23.38 ± 4.88 pg/dl, the pre-exposure level, to 20.16 ± 3.17 pg/dl (13.7%) on 30th day of treatment. There was a fall in MCHC value to the extent of 38.3% by the 60th day of treatment. However, when compared with control group MCV values showed variable results, the value declined non-significantly ($P > 0.05$) till the 20th day and thereafter there was a significant increase ($P \leq 0.01$) to 81.31 ± 9.40 fl on 40th day of treatment (Table 2).

The significant alterations in blood parameters in the present investigation merely reflected severe anaemia and leucopenia in molybdenotic buffalo calves. Further, as evidenced by major perturbation in analysed blood parameters with significant decrease in TLC, oral administration of excess Mo containing diet for a prolonged period alters immune functions in buffalo calves, validated earlier reports conducted on cattle (Ward and Spears 1999). Taken together, the results of present investigation indicated that it would be prudent to avoid excessive dietary Mo in buffalo calves.

SUMMARY

An experimental trial was conducted to assess the influence of excess dietary molybdenum for prolonged period of time on haematological profile of healthy buffalo calves. Repeated oral administration of sodium molybdate @ 3 mg/kg/day for 90 consecutive days produced profound decline in the mean values of Hb, TLC, TEC, and MCHC in buffalo calves. It is suggested that exposure to excess dietary molybdenum should be avoided in buffalo calves.

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