Comparative Evaluation of Therapeutic Trials in Experimentally Induced Lead Toxicosis in Crossbred Calves

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Introduction

Lead is an essential trace element but is considered to be one of the most potent metal toxicants to cattle. Several enzootics of lead poisoning in domestic animals had been recorded throughout the world where the source of the metal was the contamination of pasture or crops by nearby industrial lead operation.[1] The sources of environmental lead contaminating the food supplies include consumption of tetra ethyl lead enriched with gasoline, fodder crops, pastures alongside the highways, effluents from mines, smelters, foundry fumes, batteries, felt, linoleum, crankcase oil and newsprints. Continuous feeding of lead contaminated fodder may lead to serious health hazards particularly involving brain, liver and kidneys. Scanty information is available in the scientific literature on therapeutic responses in lead toxicosis vis-a-vis status of liver and kidney functions.[2] Therefore, the present study was undertaken to evaluate two different treatments in experimentally induced lead toxicosis by monitoring clinico-biochemical changes.

Materials and Methods

Animals and sampling procedures

Ten crossbred, 6-9 months old male calves were procured from a local market on the basis of good body condition. All the calves were kept under observation for about a month and were dewormed with broad spectrum anthelmintic. To establish base values, blood and urine samples were collected on three occasions from each animal. Subsequently lead toxicosis was experimentally induced in all the calves by oral administration of lead acetate @ 10 mg/kg body weight daily for 40 days. The calves were divided into two equal groups for therapeutic evaluation undertaken after 40 days of experimental induction. The samples were collected periodically throughout the period of experimental study.

Analysis procedures

The haematological indices, blood biochemicals and plasma enzymatic activities were determined as described elswhere.[2] Blood, plasma and urine concentrations of minerals were monitored by atomic absorption spectrophotometer (Model AA6, Varian Techtron, Melbourne, Australia).[3] The data were analyzed statistically by applying student's t-test at 5 per cent level of significance.

Treatment regimens

Group A: The calves were parenterally administered disodium ethylene diamine tetra acetate (Na_2EDTA; 8.5 % solution) - Calcium borogluconate (25% solution) combination (2:1)[2]. Na_2EDTA was given @ 100 mg/kg body weight.

Group B: The calves were parenterally administered Na_2EDTA - Calcium borogluconate combination at the same dose rate as calves of group A along with thiamine hydrochloride @ 5 mg/kg body weight. Supportive treatment in both the groups comprised of parenteral administration of 5 per cent dextrose solution and liver extract once daily for five days.

Results and Discussion

The clinical signs of inappetance, gastro-intestinal disturbances, loss of body condition, apparent blindness, grinding of teeth and nervous signs were observed in all the calves following induction of lead toxicosis. Following adoption of therapeutic measures, there was 80.00 and 100.00 per cent recovery in group A and B, respectively. Decrease in severity of nervous symptoms observed 24 hours after institution of therapy

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might be due to increased urinary lead excretion. Improvement in clinical signs in group B was observed in 3 days, whereas, animals of group A took five days for clinical recovery. The delayed clinical response in group B was ascribed to slow urinary excretion of lead (8.17±0.82 µg/ml) as compared to that in calves of group A (10.44±0.58 µg/ml) recorded on 3rd day post-treatment. The respective mean blood lead concentrations in group A and B during that period were 62.49±7.33 and 51.13±3.27 µg/dl.

Table 1. Haematological and biochemical changes before and after adoption of therapeutic measures in lead toxicosis in crossbred calves (Mean ± SE).

Parameters		Sampling time (days)						
		0	20	30	40**	45	50	55
Hb (g/dl)	A	11.12 ±0.87	10.80 ±0.58	9.96 ±0.44	8.40 ±0.43	8.30 ±0.34	8.60 ±0.21	8.95 ±0.38
	B	10.96 ±0.51	10.44 ±0.28	9.16 ±0.25*	7.45 ±0.18*	8.00 ±0.40*	8.50 ±0.45*	8.90 ±0.48
PCV (%)	A	32.00 ±2.19	31.20 ±1.01	28.80 ±1.85	24.00 ±1.26*	25.00 ±1.73	25.00 ±1.29*	28.00 ±1.41
	B	30.80 ±1.74	30.40 ±0.74	29.20 ±2.15	21.00 ±0.57*	25.00 ±1.70	27.50 ±0.95	26.50 ±0.95
Plasma creatinine (mg/dl)	A	1.07 ±0.15	—	—	2.47 ±0.51*	—	—	1.25 ±0.14
	B	1.11 ±0.15	—	—	2.43 ±0.53*	—	—	1.34 ±0.06
BUN (mg/dl)	A	17.75 ±1.52	—	—	29.18 ±0.64*	—	—	22.19 ±0.75
	B	18.86 ±1.62	—	—	31.43 ±0.96*	—	—	22.54 ±2.33
AST (IU/L)	A	52.41 ±4.36	55.05 ±3.45	68.74 ±3.84*	73.39 ±3.09*	69.36 ±9.05	61.93 ±2.56	53.96 ±4.79
	B	41.09 ±4.40	59.09 ±4.71*	69.99 ±4.46*	69.94 ±6.34*	74.34 ±2.65*	47.23 ±3.45	53.11 ±4.43
ALP (IU/L)	A	56.63 ±3.97	67.91 ±4.29	76.29 ±2.40*	76.68 ±3.81*	74.26 ±3.29*	73.32 ±1.52*	65.88 ±2.46
	B	55.64 ±3.04	62.26 ±5.84	81.68 ±3.83*	75.31 ±2.68*	79.05 ±1.56	68.54 ±1.93	70.46 ±6.46
GGT (IU/L)	A	17.00 ±1.00	—	—	38.20 ±4.17*	—	—	21.00 ±1.29
	B	18.20 ±0.73	—	—	33.25 ±1.10*	—	—	21.00 ±2.27
LDH (IU/L)	A	263.80 ±3.97	—	—	404.40 ±25.25*	—	—	281.50 ±4.19
	B	263.60 ±5.91	—	—	355.25 ±17.28*	—	—	284.25 ±5.45
Blood lead (µg/dl)	A	18.17 ±7.70	70.45 7.53*	79.54 ±0.03*	100.99 ±6.62*	36.92 ±5.44	42.61 ±9.70	22.72 ±4.63
	B	22.72 ±8.03	74.99 ±5.79*	79.54 ±8.03*	102.26 ±4.63*	28.40 ±7.33	34.09 ±4.63	17.04 ±7.33
Urine lead (µg/ml)	A	0.02 ±0.02	0.22 ±0.03*	0.24 ±0.04*	0.49 ±0.05*	10.10 ±0.19*	1.92 ±0.14*	0.02 ±0.02
	B	0.02 ±0.02	0.19 ±0.05*	0.29 ±0.05*	0.47 ±0.05*	12.38 ±0.23*	1.72 ±0.14*	0.05 ±0.02

*Significant at 5 per cent level (P/0.05).
** Therapy instituted after 40 days.
(—) Not analyzed; A-Treatment Group A; B- Treatment Group B.

Haematological indices revealed normocytic and normochromic anemia following induction of toxicity. Both dyshaemopoiesis and haemolysis played a part in the production of anemia in chronic lead poisoning.[4] The improvement in mean Hb and PCV values after adoption of therapy might be due to improvement in erythropoietic activity of bone marrow following significant reduction in lead concentration in the system. The mean plasma AST, ALP, LDH and GGT enzymatic activities and BUN and plasma creatinine increased significantly by the 40th day of induction of toxicosis. However, significant improvement was recorded in plasma enzymatic and blood biochemical values 15 days after adoption of therapeutic measures and the values were comparable to zero day values in both the therapeutic groups. The significant increase in plasma activities was reflective of degenerative changes involving various organs[5] following exposure to toxic doses of lead. Improvement in these values observed following treatment in both the groups was suggestive of regenerative processes occurring in various vital organs following increased elimination of lead from the system. Significant increase in plasma GGT activity by the 40th day of induction reflected hepatitis.[5] Significant decline in plasma GGT after therapy was ascribed to significant reduction in blood lead concentration and positive effect of liver tonic on hepatic regeneration.

A significant increase in BUN and plasma creatinine levels in lead toxicotic calves was reflective of renal parenchymal damage and impaired kidney functions leading to decreased glomerular filtration rate.[6] Toxic doses of lead are capable of destroying a large quantity of renal parenchymal and may induce changes in BUN and plasma creatinine levels.[5] Histopathological changes in kidney sections in the present study, reflective of degenerative changes, supported the above findings.

Blood lead concentration showed over a five-fold increase in both groups by the 40th day of induction. However, following adoption of therapeutic measures, a significant decline in lead content was observed on the 43rd day as compared to 40th day values in both the groups. The decline in blood lead content after therapy might be ascribed to increased urinary excretion of lead in both the groups probably due to binding of lead with EDTA. A significant increase recorded in urinary lead levels after adoption of therapeutic measures might be due to the chelating effect of NA_2EDTA with lead. It had been suggested that when calcium salt of EDTA was administered to an animal, in the presence of lead ions or lead bound loosely to proteins, lead replaced calcium in the molecule to form PbEDTA because of the greater solubility constant of PbEDTA. The PbEDTA compound has been reported to be non-ionizable at physiological pH and hence non-toxic[7].

The enhanced efficacy of the combination of thiamine and EDTA, in reducing blood levels, increasing urinary lead excretion and restoring lead induced biochemical alterations, suggested a promising role of thiamine in combination with EDTA in the treatment

of lead intoxication. It was considered that thiamine might be facilitating cell penetration by $CaNa_2EDTA$ through reducing its ionic character for a more effective chelation of intracellular bound lead. Probably, the main advantage of the combination therapy was that the lower doses of fairly toxic chelating agents such as $CaNa_2EDTA$ could be used as suggested by Lilis and Fischbein.[8] Also, it was considered that if the metal dissociated from the chelating agent, it might be chelated by the other complimentary agent.[9]

It was concluded that lead toxicosis in calves resulted in anemia, hepatitis and impairment of kidney functions. Therapeutic measures adopted in group B were more effective than those of group A in restoring early and absolute clinical recovery in lead toxicosis in crossbred calves.

Summary

An experimental study on lead toxicosis induced by oral administration of lead acetate @ 10 mg/kg body weight in ten crossbred, 6-9 months old male calves, was undertaken to study its effect on plasma enzymes and blood biochemicals and to evaluate the therapeutic response with two different sets of treatment. Blood biochemical analysis revealed anemia, significant increase in mean plasma activity of AST, ALP, LDH and GGT enzymes associated with significant increase in BUN and plasma creatinine contents by 40th day of induction of lead toxicosis. Blood biochemical alterations were suggestive of impairment of hepatic and kidney functions. The clinical response with two sets of treatments undertaken in two groups of toxicotic calves revealed that the treatment adopted in group B comprising parenteral administration of Na_2EDTA-calcium borogluconate combination along with thiamine hydrochloride was more effective than treatment comprising Na_2EDTA-calcium borogluconate combination alone used in group A in restoring blood biochemical and plasma enzymatic alterations with early and absolute clinical recovery probably through reduction in blood lead levels and increased urinary excretion of lead.

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Abstract

Bovine tuberculosis in Swedish deer farms; epidemiological investigations and tracing using restriction fragment analysis

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Bovine tuberculosis was eradicated from Sweden after a program lasting many years. By 1991, no tuberculosis in wildlife had been discovered for 50 years and the last case in cattle had occurred 13 years before. In 1991, the disease was identified in a herd of farmed follow deer (*Dama dama*) and over the next three years nine other infected herds were identified. Investigation revealed that all the infected deer were either deer that had been imported into Sweden from the United Kingdom in 1987 or had been in contact with them. Restriction fragment analysis of eight isolates of *Mycobacterium bovis* from five of the herds showed that the isolates had identical patterns of DNA fragments, which indicated a common source of infection. Among more than 800 isolates of *M. bovis* that have been analyzed, these patterns were identical to those of only two previous isolates, both of which came from British deer. These results indicate that the eight Swedish strains of *M. bovis* and the two British strains may have a common source of infection.

Reduced Cardiac Functional Capacity as a Consequence of the Double-muscled Conformation Selection in the Belgian White and Blue Breed

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Introduction

In the field of meat production in Belgium, the interest in double-muscled cattle is increasing because of the exceptional commercial value of the animals produced by such a selection. However, double-muscled cattle have a lower aerobic metabolic capacity than do conventional cattle.[1] Several steps in the oxygen-transport pathway might be responsible for this limited maximal oxygen consumption. Among these, the cardiovascular system has been suspected as a potential critical step.[1,2] The present study was performed in order to test this hypothesis by direct hemodynamic measurements.

Material and Methods

The study was conducted in two parts. For the first part of the study, 41 Friesian calves, considered as conventional, and 19 Belgian White and Blue double-muscled calves were studied repeatedly during their growth. A total of 123 and 70 recordings were collected in the conventional and double-muscled group, respectively. In these calves, central venous (CVP), right ventricular (RVP), pulmonary arterial (PAP), pulmonary capillary wedge (PW) and systemic arterial (SAP) pressures were obtained by means of fluid filled catheters positioned under pressure monitoring, heart rate (HR) was calculated from the ECG tracings and cardiac output (CO) was measured using the thermodilution technique. Stroke volume (SV), cardiac and stroke indices (CI and SI, respectively), and pulmonary and systemic vascular resistance (PVR and SVR) were calculated from the measured parameters.

For the second part of the study, 41 and 55 sets of hemodynamic records were collected from a group of 6 conventional and 6 double-muscled calves, respectively. In these calves, CVP, PAP, PW, SAP, SV and CO were measured as in the first part of the study. Right ventricular pressure was recorded by means of a microtip catheter and the maximal value of its first derivative with respect to time (Max dP/dt) was calculated from the tracings obtained by means of a differentiator amplifier. Right ventricular end-diastolic and end-systolic volume (EDV and ESV, respectively) and ejection fraction (EF) were measured by means of the thermodilution technique and using Swan Ganz catheters equipped with a rapid response thermistor. In each group of calves, the mean right ventricular pressure-volume loop was constructed from EDV, ESV, peak systolic RVP, proto- and end-diastolic RVP, and diastolic PAP values. The systolic elastance slopes (Ees) was calculated from the peak systolic RVP/ESV ratio. Diastolic elastance slope (Eed) was calculated from the [end-diastolic RVP - proto-diastolic RVP] / [EDV - ESV] ratio.

Statistical Analysis

The results obtained in the 2 groups were compared using a random linear nested model including the effect of calf, breed, and body weight.

Results and Discussion

The results of the first part of the study demonstrated that global cardiac performance, as expressed in terms of CO, SV, CI or SI, is significantly lower in double-muscled than in conventional calves, suggesting that the global cardiac performance is significantly poorer in double-muscled calves.

In a heart of given volume, performance is governed by 4 determinants: heart rate, preload, afterload and contractility.[3,4] Heart rate, CVP, diastolic RVP and

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