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Research Article

Immunological and haemobiochemical changes induced by oxytetracycline in Black Bengal goats

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Abstract

Oxytetracycline (OTC) was administered intramuscularly in Black Bengal goats both therapeutically and sub-therapeutically. Today, OTC is commonly used at therapeutic dose in treatment of various animal diseases as well as extensively used at sub-therapeutic dose in animal feed as growth promoter. Extensive use of these drugs without labelled direction leads to various haemo-biochemical and immunological changes. There was drastic reduction in Hb level whereas significant reduction was noticed in TEC. TLC was reduced marginally. Lymphocytosis and eosinophilia with relative neutropenia was observed. However no alterations were found in monocyte and basophil counts. There was drastic fall in glucose level. There was significant decrease in serum protein too. However, the trend in ALT and AST level was found to be reduced and haphazard. The value of IgG was decreased significantly at both therapeutic as well as sub therapeutic doses indicating its immune-suppressive effects.

Keywords: Goat, Haemo-biochemical, Immunology, Oxytetracycline, Toxicity.

Oxytetracycline is commonly used for the prevention and/or treatment of diseases in livestock production. As a feed additive in sub-therapeutic doses, it contributes to the maintenance of optimal health and thus promotes growth in food-producing animals. It is known as a broad-spectrum antibiotic with a bacteriostatic effect on the wide range of gram negative and gram-positive bacteria. The mode of action lies in its binding to 30S ribosomal subunits of bacteria, thus inhibiting the protein synthesis. However, the use of this compound may result in residues in animal derived food products and can pose several health hazards. Excessive dosage of OTC can also alter haematological, biochemical and immunological parameters of livestock.

Material and methods

Clinically healthy black Bengal adult goats (1-1½ year age) weighing between 12-14 kg were used in this experiment. They were caged individually in custom made stainless steel metabolic cages. The animals were stall-fed and water was provided *ad libitum*. The composition of feed was 2 part wheat husk, 1 part crushed maize, 1 part crushed gram and 2 part green. The temperature of the animal room was maintained at 25 ± 3°C and provided with artificial lighting facilities. Before starting the experiment, the animals were dewormed once with a mixture of albendazole and rafoxanide (Vetalben-R, Indian Immunologicals) @ 7.5 mg/kg body weight.

Twenty four goats (24) were divided into 3 groups**.** Group-I consists of eight (8) goats as control, without administration of any OTC dose. Group–II consists of eight (8) goats which were administered sub-therapeutic dose of OTC @ 5 mg/ kg b.wt i/m on 0, 7, 14, 21, 28, 35, 42, 49, 56 days. Group-III consists of eight (8) goats which were administered therapeutic dose of OTC @ 10 mg/ kg b.wt i/m on 0, 7, 14, 21, 28, 35, 42, 49, 56 days. Blood samples (6 ml) were collected from jugular vein of each animal of three groups on (before application of oxytetracycline) 0, 7, 14, 21, 28, 35, 42, 49 and 56th days for estimation of haemo-biochemical and immunological parameters.

Hb was determined by Coffin (1953) and expressed as g/L. Total erythrocyte count, total leucocyte count and differential leucocyte count were estimated by Schalm *et al.* (1975). Blood glucose was estimated as per the method described by Jain (1986). The serum protein was estimated by Wooton (1974). Aspartate and alanine transferase activities were measured by the method of Yatazidis (1960) and expressed as pyruvic acid formed/ml of serum/hr.

A sandwich ELISA was performed for estimation of serum IgG levels in all the goats using the method of Heyman *et al.* (1984). The results were analyzed statistically for analysis of variance and least significant difference tests as per Snedecor and Cochran (1997).

Results and Discussion:

The hemoglobin (Hb) level decreased significantly from 28th day onwards in group II animals

as compared to control group whereas in group III the Hb level reduced drastically from 14th day onward in comparison to Group I (control). The Hb level was remained unaltered in Group I throughout study period. The total erythrocyte count (TEC) significantly (P<0.05) lowered from 56th day onwards in both group II and III. The TEC remain significantly (P>0.05) unchanged in Group I (control).The total leukocyte count (TLC) significantly (P<0.05) lowered from 49th day onwards in both group II and from 35th day onwards in group III. The TLC remain significantly (P>0.05) unchanged in Group I (control). Lymphocytosis (P<0.05) was observed in both groups II and III. The lymphocyte count has significantly (P<0.05) increased from 56th day onwards in group II and 35th day onwards in group III, whereas no significant (P>0.05) change was observed in group I. Neutropenia (P<0.05) was clearly evident in both groups II and III. The neutrophils count has significantly (P<0.05) decreased from 35th day onwards in group II and 28th day onwards in group III, whereas no significant (P>0.05) change was observed in group I. Eosinophilia (P<0.05) was clearly evident in both groups II and III. The eosinophil count has significantly (P<0.05) decreased from 42nd day onwards in group II and 35th day onwards in group III, although it was elevated sharply from 29th day onwards in therapeutic dose group, whereas no significant (P>0.05) change was observed in group I. Monocytopenia and Basopenia (P<0.05) was observed in both group II and III on long term administration of the drug @ therapeutic dose as compared to group I. Our present findings were in concordance with the findings of Fatema Elzhraa and Raheim (2004) that carried out hematological study on sixty cattle, Van Miert *et al.* (1983) who carried research on goats and Edosa and Sarafa (2005) who studied the effect of OTC on aquaculture, Nile Tilapia.

There was a drastic decrease in blood glucose level on long term administration @ therapeutic dose. However, there was not much alteration found @ sub-therapeutic dose. The significant (P<0.05) decrease in blood glucose level was noticed from 21 days onwards in group II and 7th day onwards in group III, whereas no significant (P>0.05) change was observed in group I. Glette *et al.* (1984) and Mody (1989) found out that at high concentrations of OTC administration, chemiluminescence and glucose oxidation were impaired. There was a severe decrease in blood glucose

Table I: Haemogram of Black Bengal goats following OTC administration (N=24, Mean ± SE).

Animal/ Day	Group I (n=8)	Group II (n=8)	Group III (n=8)
Hb (g/L)			
0	84.68 ± 4.15$^{a** x*}$	80.30± 5.32^{ayz}	77.43 ± 6.84^{az}
7	88.7 ± 8.54^{ax}	88.08 ± 1.25^{az}	63.68 ± 0.18^{byz}
14	87.93 ± 4.23^{ax}	79.08± 3.91^{ayz}	61.83± 4.84^{byz}
21	84.45± 6.37^{ax}	83.15± 3.66^{ayz}	58.60 ± 4.15^{bxy}
28	86.25 ± 4.49^{ax}	63.75± 6.17^{bxy}	56.50 ± 4.11^{bxy}
35	92.4± 3.16^{ax}	64.78± 5.43^{bxyz}	61.18± 4.17^{byz}
42	89.58± 3.84^{ax}	64.30±3.20^{bxyz}	56.53± 3.31^{bxy}
49	88.08 ± 3.59^{ax}	66.25 ± 5.83^{bxyz}	52.30± 2.90^{bxy}
56	92.43±4.01^{ax}	52.33±7.41^{bx}	42.30 ± 4.01^{bx}
TEC ($\times 10^{12}$/L)			
0	6.39 ± 0.68$^{a** x*}$	6.12 ± 0.39^{ay}	5.47 ± 0.25^{ay}
7	6.64 ± 0.69^{ax}	5.12 ± 0.13^{axy}	5.25 ± 0.28^{ay}
14	6.33 ± 0.43^{ax}	5.44 ± 0.28^{axy}	5.17 ± 0.08^{ay}
21	6.64 ± 0.32^{ax}	4.67 ± 0.16^{axy}	4.31 ± 0.41^{axy}
28	6.57 ± 0.80^{ax}	5.26 ± 0.38^{axy}	4.23 ± 0.24^{axy}
35	6.14 ± 0.28^{ax}	5.35 ± 0.20^{abxy}	4.47 ± 0.51^{axy}
42	6.60 ± 0.43^{ax}	5.14 ± 0.49^{abxy}	4.02 ±0.29^{axy}
49	5.95 ± 0.39^{ax}	5.00 ± 0.19^{abxy}	4.29 ± 0.27^{axy}
56	6.90 ± 0.42^{ax}	5.09 ± 0.30^{bxy}	3.52 ± 0.21^{bx}

Note: Group I → control; Group II → Sub-therapeutic administration; Group III → Therapeutic administration.

* Mean values with dissimilar superscript in a column vary significantly at (P<0.05) and denoted by superscript (x, y, z)

** Mean values with dissimilar superscript in a row vary significantly at (P<0.05) and denoted by superscript (a, b)

Table II: Haemogram of Black Bengal goats following OTC administration (N=24, Mean±SE).

Animal/ Day	Group I (n=8)	Group II (n=8)	Group III (n=8)
TLC (× 10^9/L)			
0	7.39± 0.44 $^{a** x*}$	7.47 ± 0.62^{ax}	6.04± 0.46^{ay}
7	7.68 ± 0.98^{ax}	7.80± 1.08^{ax}	6.00± 0.48^{ayz}
14	7.19 ± 0.56^{ax}	7.48±0.62^{bx}	6.05 ± 0.19 ^{ay}
21	7.08 ± 1.21^{ax}	6.41 ± 0.61^{ax}	5.19 ± 0.44^{axyz}
28	7.82 ± 0.89^{ax}	6.37 ± 0.31^{ax}	5.13 ± 0.35^{axyz}
35	7.67 ± 0.73^{ax}	6.47 ± 0.36^{abx}	5.05± 0.32^{bxyz}
42	7.46 ± 0.48^{ax}	6.18± 0.32^{abx}	4.96 ± 0.12^{bxyz}
49	7.60 ± 0.57^{ax}	6.76± 0.59^{bx}	4.28 ± 0.21^{bx}
56	7.65±0.54^{ax}	5.70 ± 0.70^{bx}	4.40±0.18^{bxy}
LYMPHOCYTES (× 10^9/L)			
0	2.57 ± 0.11 $^{a** x*}$	2.50 ± 0.02^{axy}	2.50 ± 0.12^{ax}
7	2.48 ± 0.97^{ax}	2.83 ± 0.01^{bxyz}	3.13 ± 0.40^{bxy}
14	2.59 ± 0.69^{ax}	2.20 ± 0.38^{ax}	3.08 ± 0.13^{axy}
21	2.69 ± 0.14^{ax}	2.68 ± 0.05^{axy}	3.10 ± 0.22^{axy}
28	2.70 ± 0.23^{ax}	2.90 ± 0.11^{axyz}	3.22 ± 0.07^{ay}
35	2.75 ± 0.12^{ax}	2.95 ± 0.16^{ayz}	3.70 ± 0.19^{byz}
42	2.64 ± 0.02^{ax}	2.95 ± 0.05^{byz}	3.65 ± 0.07^{cyz}
49	2.86 ± 0.04^{ax}	2.90 ± 0.07^{axyz}	3.65 ± 0.13^{byz}
56	2.73 ± 0.11^{ax}	3.43 ± 0.03^{bz}	4.13 ± 0.17^{cz}

Note: Group I → control; Group II → Sub-therapeutic administration; Group III → Therapeutic administration.
* Mean values with dissimilar superscript in a column vary significantly at (P<0.05) and denoted by superscript (x, y, z)
** Mean values with dissimilar superscript in a row vary significantly at (P<0.05) and denoted by superscript (a, b)

Table III: Biochemical parametersof Black Bengal goats following OTC administration (N=24, Mean±SE).

Animal/ Day	Group I (n=8)	Group II (n=8)	Group III (n=8)
Blood Glucose (m mol L^{-1})			
0	3.85 ± 0.31	3.50 ± 0.09	3.45 ± 0.06
7	3.15 ± 0.23^{ax}	3.15± 0.18^{ax}	2.85± 0.14^{azuv}
14	3.45 ±0.23^{ax}	2.65 ± 0.10^{auv}	2.85 ± 0.06^{av}
21	3.45 ± 0.14^{ax}	2.05 ± 0.23^{byz}	2.60 ± 0.21^{azu}
28	3.00 ± 0.25^{ax}	1.75 ± 0.65^{bz}	2.30 ± 0.09^{au}
35	3.18 ± 0.19^{ax}	1.25 ± 0.23^{bxy}	1.45 ± 0.06^{byz}
42	3.20 ± 0.17^{ax}	1.65 ± 0.18^{bxy}	1.15 ± 0.14 ^{by}
49	3.25 ± 0.21^{ax}	0.70 ± 0.09^{bx}	1.05 ± 0.10^{bx}
56	2.85 ± 0.12^{ax}	0.55 ± 0.06^{bx}	0.85 ± 0.14^{bx}
Serum Protein (g L^{-1})			
0	27.05 ± 1.24	24.75 ± 0.90	24.75 ± 1.25
7	27.45 ± 1.45^{ax}	19.6 ± 1.55^{bxy}	19.8 ± 1.19^{bu}
14	28.50 ± 1.30^{ax}	20.15 ± 0.51^{bxy}	15.80 ± 1.02^{cyzu}
21	27.85 ± 0.96^{ax}	17.90 ± 1.52^{bx}	17.15 ± 1.08^{bzu}
28	28.80 ± 0.44^{ax}	18.55 ± 1.42^{bx}	12.55 ± 0.79^{cxyz}
35	26.40 ± 1.10^{ax}	17.55 ± 0.54^{bx}	13.8 ± 0.50^{cxyz}
42	27.65 ± 1.21^{ax}	17.35 ± 0.76^{bxy}	11.35 ± 1.45^{cxy}
49	26.05 ± 1.87^{ax}	16.00 ± 1.85^{bx}	10.45 ± 0.76^{bx}
56	26.35 ± 1.09^{ax}	15.35 ± 0.48^{bx}	9.43 ± 0.66 ^{cx}

Note: Group I → control; Group II → Sub-therapeutic administration; Group III → Therapeutic administration.
* Mean values with dissimilar superscript in a column vary significantly at (P<0.05) and denoted by superscript (x, y, z)
** Mean values with dissimilar superscript in a row vary significantly at (P<0.05) and denoted by superscript (a, b)

level on long term administration @ therapeutic dose, whereas there was transient decrease found @ sub-therapeutic dose. The significant (P<0.05) decrease in blood protein level was evident from 7th day onwards in both group II and in group III(Table 2), whereas no significant (P>0.05) change was observed in group I.

Table IV. Immunological parameters [IgG(mg ml^{-1})] in Black Bengal goat following OTC administration. (N=24, Mean ± SE)

Animal/ Day	Group I (n=8)	Group II (n=8)	Group III (n=8)
IgG(mg ml^{-1})			
0	12.42 ± 0.20	12.42 ± 0.15	12.47 ± 0.18
7	12.19 ± 0.28^{ax}	11.72 ± 0.07 ^{ay}	11.57 ± 0.06 ^{ay}
14	12.39 ± 0.16^{ax}	11.55 ± 0.10^{byz}	11.45 ± 0.10 ^{by}
21	12.28 ± 0.09^{ax}	11.36 ± 0.09 ^{by}	11.32 ± 0.09^{byzu}
28	12.29 ± 0.20^{ax}	11.22 ± 0.04^{bzu}	11.11 ± 0.06^{byz}
35	12.16 ± 0.16^{ax}	11.07 ± 0.03^{buv}	10.63 ± 0.14^{bz}
42	12.34 ± 0.26^{ax}	10.76 ± 0.08^{cv}	9.96 ± 0.09^{bu}
49	12.32 ± 0.19^{ax}	10.13 ± 0.07^{cw}	9.28 ± 0.11^{bv}
56	12.33 ± 0.19^{ax}	9.78 ± 0.28^{cw}	8.81 ± 0.18^{bv}

Note: Group I → control; Group II → Sub-therapeutic administration; Group III → Therapeutic administration.
* Mean values with dissimilar superscript in a column vary significantly at (P<0.05) and denoted by superscript (x, y, z)
** Mean values with dissimilar superscript in a row vary significantly at (P<0.05) and denoted by superscript (a, b)

Similar reporting were done by NADA (2003), FatemaElzhraa and Raheim (2004).The significant (P<0.05) increase in ALT as well as AST level was evident on long term administration @ therapeutic dose. The significant (P<0.05) increase in ALT was observed from 42nd day onwards in therapeutic dose group while significant (P<0.05) increase in AST was noticed from 49th day onwards in therapeutic dose group although no significant changes were observed neither in control nor in sub-therapeutic dose group in both ALT and AST during study period. The present findings were found in accordance with the reporting of Van Miert and Van duin (1979) and Mody, (1989) in cattle and Van Miert *et al*. (1983) in goat, Kumar and Malik (2001) in buffalo. However, Nosaka and Sakamoto (1999) concluded that ALT and AST activities did not change significantly over time in humans after intramuscular injection of OTC.

The reduction in IgG level (mg ml^{-1}) was interpreted on long term administration of OTC @ therapeutic dose as well as @ sub-therapeutic dose. The significant (P<0.05) decrease in IgG level was evident from 14th day onwards in both group II and group III as compared to group I (control). The significant drastic (P<0.05) decrease in IgG level was evident from 42nd day onwards in both group II and from 35th day onwards in group III as compared to group I (control). The humoral immune responses were decreased as the immunosuppressive effects on long term administration of OTC. The present findings were similar with observations of Lien *et al*. (2007) in pigs, Diefy et al. (2003) and Rijkers *et al*. (1981) in sheep.

Conclusion

Although OTC is a wonder drug and drug of choice in many indications as well as extensively used as growth promoters, but, it has some adverse side effects on haematological, biochemical and immunological parameters of livestock on long term administration.

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Hepatoprotective effect of *Moringa oleifera* in experimentally induced hepatotoxicity in Cockerels

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Abstract

The ethanolic and aqueous extracts prepared from leaves of *Moringa oleifera* were evaluated against acetaminophen-induced hepatotoxicity in cockerels. Acetaminophen was given @ 500 mg/kg b wt orally to induce hepatocellular damage. Ethanolic extract of *Moringa oleifera* @ 200 mg/kg b wt helped in restoration of Hb, PCV, TEC, TLC and lymphocytes and heterophils as well as total protein, albumin and globulin, glucose, cholesterol, bilirubin and activities of AST, ALT, ALP and LDH. Liver sections of treated animals clearly showed normal hepatic cells and central vein thereby confirming hepatoprotective activity. Phytochemical analysis of ethanolic extract showed presence of alkaloids, coumarin, flavonoids, glycosides, resins, sterol, tannin and triterpenes.

Keywords: Cockerels, Hepatotoxicity, *Moringa oleifera*, phytochemial analysis

Hepatotoxicity is one of very common aliment resulting into serious debilities ranging from severe metabolic disorders to even mortality. To prevent hepatotoxicity some drugs or chemicals for antagonizing the toxins or to help the hepatocytes to regain its power of metabolism are administered. Uses of herbal liver stimulant/formulations become important in treating cases suffering from hepatic diseases. Herbs play a major role in the management of various liver disorders along with other system associated diseases. Various plants have been reported to possess hepatoprotective property. *Moringa oleifera* is a multipurpose tree with most of its parts being used (Anwar *et al.,* 2007). Its extracts have antihypertensive (Faizi *et al,* 1995), anti-inflammatory (Caceres *et al.,* 1992) and also antitumor (Guevara *et al.,* 1999) properties. It has also found to reduce the level of cholesterol (Mehta *et al,* 2003) and regulate the thyroid status (Costa *et al.,* 2005). Its hepatoprotective action against antitubercular drugs such as isoniazid and rifampicin induced liver injury has been documented (Kumar and Pari, 2003). The plant seed powder may be useful in chelation therapy (Kumari *et al.,* 2005; Sharma *et al.,* 2006). This paper reports the hepatoprotective activity of *Moringa oleifera* using liver function markers following experimentally induced hepatotoxicity in cockerels.

Material and Methods

The leaves of *Moringa oleifera* were procured from MRDC, Pantnagar and were shade dried and ground in a Willey Grinder at room temperature. For preparation of the ethanolic or aqueous extracts, 100 gm each powder of *Moringa oleifera* was soaked in 1000 ml of absolute ethanol or water for 48 hr at 37^0 C with continuous stirring. The contents were filtered and concentrated by evaporation at lower temperature (45-50^0 C) and reduced pressure using rotatory vacuum evaporator (Singh, 2001) and lyophilized to get the final extract residue. They were stored at 4^0C in refrigerator till further use.

Ethanolic and aqueous extracts of *Moringa oleifera* were analysed for major phytochemical groups, viz. alkaloids, anthraquinones, flavonoids, saponins, tannins, sterols, reducing sugars, glycosides, resins, triterpenes, proteins and coumarins by standard methods (Das *et al*., 1964, Harborne, 1973 Sofawara, 1982 and Arunadevi, 2003).

Total of 100, three-month-old cockerels belonging to same hatch, were procured from IPF, Pantnagar and randomly divided into five groups I, II, III, IV and V of 20 cockerels each. All the five groups had almost equal average body weight and maintained under standard deep litter managemental conditions. Gr I served as healthy control, Gr II received acetaminophen @ 500 mg/kg body weight orally for 7 days (Bhar *et al.*, 2009) served as infected control. Gr III received silymarin (as a standard reference) along with acetaminophen for 7 days and thereafter only silymarin was given upto 35^{th} day. In cockerels of Gr IV and V ethanolic and aqueous extracts of *Moringa oleifera* @ 200 mg/kg b wt (Patel *et al.,* 2008) along with

acetaminophen for 7 days and thereafter only extracts were given upto 35th day.

The blood samples were collected on day 0, 7, 15, 21, 28, 35 and 42 of treatment, for haematological and biochemical parameters. Hb, TEC, TLC, PCV and differential leukocyte count, glucose, total cholesterol, total protein, albumin, globulin, albumin: globulin ratio, blood urea nitrogen and serum bilirubin and activities of liver enzymes aspartate aminotransferase (AST), alanine aminotransferase (ALT), alkaline phosphatase (ALP) and lactate dehydrogenase (LDH) were studied by standard procedures.

Liver samples were collected in 10% buffered formalin for histopathological study and examined for any type of gross changes from each group on 7th, 21st and 35th day of treatment. The formalin fixed tissue pieces were serially dehydrated in alcohol and acetone, embedded in paraffin blocks and sections were cut and stained in hematoxyline and eosin stain for histopathological examination by standard procedures. The results were analysed as per the method described by Snedecor and Cochran (1994).

Results and Discussion

The ethanolic extract residue of *Moringa oleifera* was greenish brown in colour and oily in consistency, whereas its aqueous extract was light brown in colour and solid dry powder in consistency and maximum extract residue was obtained from aqueous extract (13.68%) and minimum from ethanolic extract (11.57%).

Significant decrease in Hb and PCV values were observed in Gr II as compared to Gr I, III, IV and V from 7th day onward up to the end of experiment. Gr V treated with aqueous extract also showed significant decrease in Hb value. A significant decrease in TEC and lymphocytic values and a significant increase in TLC and heterophils values were observed from 7th day onward upto the end of experiment in Gr II as compared to other treated and control groups. Ethanolic and aqueous extract of *Moringa oleifera* significantly restored these values to normalcy. The prominent reduction of Hb, PCV and TEC in acetaminophen treated groups in the present experiment could be attributed to toxic metabolite of acetaminophen, N – acetyl – p – benzoquinone, which was reported to cause oxidative stress and haemolysis of erythrocytes and hepatocytes leading to regenerative anaemia with methmoglobin and Heinz body formation (Vijayakumar *et al*., 2004). Disintegration of erythrocytes in the circulation might have resulted in reduction of haemoglobin content of blood, which in turn was associated with decrease in PCV and TEC (Chauhan *et al*., 2008). It shows that ethanolic extract of *Moringa oleifera* protects the disintegration of erythrocytes. This finding is also supported by Roa and Mishra (2004). Increased TLC values might be due to stress coupled with inflammatory changes in body tissue, which is responsible for phagocytosis of toxic substances and neutrophilia was induced by tissue demand for phagocytic function (Duncan and Prasse, 1977). An increase in heterophil count and decrease in lymphocyte count was also reported by Hedau *et al*. (2008) as was observed in this study. Roa and Mishra (2004) also found the restoration of TLC with the administration of *Moringa oleifera*.

There was significant decrease in the levels of glucose, bilirubin and cholesterol in Gr III, IV and V. Total proteins and albumin showed a marked reduction after induction of hepatopathy in untreated group from 7th day till end of experiment. Significant increase in the globulin values were recorded in all the treated groups. Increase in glucose and cholesterol might be due to the degenerative hepatic lesions and the altered values in the cholesterol might be due to the interference with the metabolism of fat in the liver. Nepolean *et al.* (2009), Sreelatha and Padma (2008) and Olugbemi *et al*. (2010) also observed the same findings. Kaneko (1989) and Mezey (1978) reported that proteins synthesized by the liver are frequently decreased in patients with liver diseases and this was manifested clinically by decrease in circulating proteins such as albumin. Globulins are intermediate proteins which are involved in antibody formation. Its higher level in *Moringa oleifera* fed group may be correlated with the immunomodulatory property of the herb (Chatterjee, 1994 and Sai Ram *et al.*, 2002). Due to the damage of hepatocytes, there was decreased elimination of bilirubin and thus an increase was observed. The increase in bilirubin was also observed by Selvakumar and Natarajan, (2008) from CCl_4 intoxication.

Increase in the activities of ALT, AST, ALP and LDH in Gr II reflected the damage of liver hepatocytes

and indirectly impairment of liver functions. Extracts of *Moringa oleifera* significantly reduced the elevation of these enzymes and it was also similar to the level of group treated with silymarin. There was significant increase in the ALP and LDH values in Gr V as compared to treated and healthy group on 7th day of experiment but it became normal from 14th day onwards. In this study, hepatoprotective effect of *Moringa oleifera* was evidenced by the improvement of ALT, AST, ALP and LDH levels. Treatment with *Moringa oleifera* extract suppressed acetaminophen induced AST, ALT, ALP and LDH elevations. The increase in serum enzymes suggest severe hepatocellular damage caused by leakage of these enzymes into circulation (Chung *et al.,* 2001).

A significant decrease in feed consumption and body weight was observed in Gr II as compared to Gr I, III, IV and V from 14th day onward till end of experiment. A significant increase in body weight was observed in the Gr IV at 42nd day of treatment as compared to treated and control groups which might be due to increase in function of hepatocyte and increased palatability of feed.

On phytochemical analysis of ethanolic extract of *Moringa oleifera* showed presence of alkaloids, coumarin, flavonoids, glycosides, resins, sterol, tannin and triterpenes. Similar results were shown by aqueous extract except absence of alkaloid and presence of reducing sugars and saponins.

The biochemical results obtained was comparable to the histopathological analysis of the liver sections obtained from each group. The healthy control group showed normal cellular architecture with sinusoidal spaces and central veins. Meanwhile, severe histopathological changes were clearly observed in intoxicated cockerels revealing centrilobular hepatic necrosis. The hepatic cords were irregularly distributed and distorted and the cells were rounded with opaque cytoplasm and showed mild vacuolated cells that suggested the fatty degeneration. When *Moringa oleifera* was given with acetaminophen, significant decrease in hepatocellular damage were observed. Normal liver architecture was seen in the liver sections obtained from cockerels. Similar observation was seen in silymarin treated group.

These results indicated that *Moringa oleifera* has hepatoprotective action. It increases the Hb, PCV, TEC, Lymphocytes, total protein, albumin, globulin and decreases glucose, total cholesterol, bilirubin, AST, ALT, ALP and LDH values to normalcy.

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Effect of herbal immunomodulators- *Azadirachta indica (Neem)* and Stresroak in vaccinated birds against Newcastle disease virus (NDV)

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Abstract

Ten different groups of birds consisting of 15 in each group were maintained. Groups I to III served as vaccine control, group I was inoculated with F strain of NDV, group II with inactivated oil adjuvanted R_2B vaccine and group III with both F and R_2B. Birds of the Stresroak treated groups (groups IV, V and VI) were vaccinated with F strain (group IV), R_2B strain (group V) and both F + R_2B (group VI). Group VII, VIII and IX were treated with *Azadirachta indica* (Neem), out of which group VII was inoculated with F strain, group VIII with R-$_2$B and group IX with both F + R_2B, whereas group X was kept as unvaccinated untreated control (UUC). Simultaneous vaccination was done on the 7th day of age with F strain of NDV and inactivated oil adjuvanted R_2B vaccine. Haemagglutination inhibition (HI) test was performed to determine the pre-vaccination and post-vaccination antibody titres against NDV groups of birds (I to IX) at different ages. Analysis of the sera samples at different time intervals revealed that the Stresroak and Neem treated cum vaccinated groups had higher HI antibody titres as compared to the untreated vaccinated groups at different ages. Statistical analysis showed that there was no significant variation between the antibody titres of the Stresroak and Neem treated groups, where the immunopotentiating response of *A. indica* was found significantly higher ($P < 0.01$) than that of Stresroak. Stresroak and Neem were found to augment humoral immune response thereby conferring protection on challenge with virulent ND virus.

Keywords: Birds, Immunomodulator, *Azadirachta indica*, Stresroak, Newcastle disease vaccine.

Newcastle disease is a dreaded disease that affects birds of all ages with heavy mortality (Hofstad *et al.*, 1984). Preventive measures of this disease include effective vaccination and proper scientific management of the birds (Sarmah, 2001). Simultaneous vaccination with F strain and inactivated mesogenic strain (R_2B) incorporated with oil adjuvant have been found to augment the immune response which protect the birds up to the market age (Pollard, 1982).

Stresroak has been found to augment humoral immunity in broiler birds vaccinated against Newcastle disease (Bora *et al.*, 1999). Another herbal drug *Azadirachta indica* (Neem) has been recognized and used, which can potentiate the immune responsiveness in the laboratory animals (Ray *et al.*, 1997) as well as in broiler birds (Sadekar *et al.*, 1998). The present study was conducted to evaluate the effect of herbal immunomodulators *Azadirachta indica* (Neem) and Stresroak on simultaneous vaccination with F strain and R_2B strain against Newcastle disease in broiler birds.

Materials and Methods

The freeze-dried of Newcastle disease virus (NDV) vaccine ampule was reconstituted in 2 ml phosphate buffered saline (PBS) for F strain of NDV and 2.5 ml sterile PBS (pH 7.4) for R_2B strain of NDV.

Propagation of viral antigen in a 9-days old embryonated egg through allantoic cavity route was done by the procedure described by Allan *et al.* (1978). To determine the EID_{50} of the virus strain, the method described by Reed and Muench (1938) was followed. The virus concentration was adjusted to 10^8 EID_{50} per ml. Inactivation of R_2B vaccine virus by mineral oil (Montanide Incomplete Seppic oil adjuvant) was done as per the method described by Allan *et al.* (1978).

Experimental designs (Table I) :- All the birds were vaccinated on the 7th day of age. The F strain vaccine was administered at a dose rate of 10^6 EID_{50} through oculonasal route and inactivated oil adjuvanted R_2B vaccine was administered at a dose rate of 10^8 EID_{50} by intramuscular route on the thigh muscle. Stresroak was given along with clean drinking water to groups IV, V and VI @ 5 ml/100 birds from first day to 10th day, @ 7.5 ml/100 birds from 11th day to 24th day and @ 10 ml/100 birds from 25th day to 56th day. *Azadirachta indica* (Neem) was administered along with feeds @ 2g/kg of feed from first day till completion of the experiment to groups VII, VIII and IX as recommended by Sadekar *et al.* (1998).

Immune response in NDV vaccinated birds was

evaluated by haemagglutination inhibition (HI) test at 0, 7th, 14th, 21st, 28th and 56th day of age from sera samples.

The birds challenged with virulent ND virus were observed for 10 days for any clinical symptom of the disease and protection per centage was evaluated.

Results were analysed as per the statistical procedures described by Snedecor and Cochran (1994).

Result and Discussion.

The maternal HI antibody titre with positive HI indices in prevaccinated chicks at day-old age was found and was in agreement with Gangopadhyay and Mallick (1970). This titre was found decline at 7 days of age with further decline at subsequent ages which colloborates the studies of Deka (2000).

The antibody titres in vaccinated groups of birds (groups I to IX) detected by HI test were determined and the findings are presented in Table II and fig. 1, 2 and 3.

The mean HI antibody titre (log 2 scale) of almost all the groups by the end of 2nd week started to rise gradually following vaccination cum treatment with Stresroak/neem ranging from 4.40 ± 0.24 to 5.00 ± 0.32 with 100 per cent HI positive indices while a slight decrease in R_2B alone vaccinated group II (3.80 ± 0.37) and control group X, where the HI antibody titre was 3.40 ± 0.40 with 80 per cent HI positive index.

Two weeks post vaccination, the HI titres of all the groups were significantly higher ($P < 0.01$) than the corresponding 14 days titre in almost all the vaccinated and treated groups except groups II (R_2B), IV (F + Stresroak), V (R_2B + Stresroak) and VIII (R_2B + Neem), where single vaccinated group I reached its peak at this age (21 days of age) with mean titre of 5.00 ± 0.63. All

Table I. Experimental designs-Vaccination and treatment schedules for different groups of birds

Experimental groups	Vaccine virus	Route of vaccination	Useof immunomodulators
I	F	Oculonasal	-
II	R_2B	Intramuscular	-
III	F + R_2B	Oculonasal+intramuscular	-
IV	F	Oculonasal	Stresroak
V	R_2B	Intramuscular	Stresroak
VI	F + R_2B	Oculonasal+intramuscular	Stresroak
VII	F	Oculonasal	*Azadirachta indica* (Neem)
VIII	R_2B	Intramuscular	*Azadirachta indica* (Neem)
IX	F + R_2B	Oculonasal+intramuscular	*Azadirachta indica* (Neem)
X	Unvaccinated untreated control (UUC)	Oculonasal+Intramuscular (0.1 ml PBS)	

Table II. Mean haemagglutination inhibition (HI) antibody titre (log 2 scale) ± standard error (SE) in chicks at different days of age.

Groups	Treatment combination	Mean ± SE HI titre (log 2 scale) at different days of age					
		0	7	14	21	28	56
I	F	6.60_{D}± 0.24	4.20_{B} ± 0.37	4.40_{B}^{b}± 0.24	5.00_{C}^{d}± 0.63	4.40_{B}^{b}± 0.51	3.60_{A}^{b} ± 0.40
II	R_2B	6.60_{D} ± 0.24	4.20_{AB} ± 0.37	3.80_{A} ± 0.37	3.80_{A}^{b} ± 0.20	5.20_{C}^{cd}±0.37	4.40_{B}^{c} ± 0.51
III	F + R_2B	6.60_{C}± 0.24	4.20_{A} ± 0.37	4.60_{A}^{b}± 0.24	5.60_{B}^{e} ± 0.40	6.20_{C}^{e} ± 0.37	6.60_{C}^{e} ± 0.81
IV	F + Stresroak	6.60_{C} ± 0.24	4.20_{A} ± 0.37	4.60_{A}^{b}± 0.24	4.40_{A}^{c} ± 0.40	5.60_{B}^{d} ± 0.24	4.40_{A}^{c} ± 0.40
V	R_2B+Stresroak	6.60_{C} ± 0.24	4.20_{A} ± 0.37	4.40_{A}^{b}± 0.24	4.40_{A}^{c} ± 0.24	5.00_{B}^{c} ± 0.45	5.20_{B}^{d} ± 0.49
VI	F+R_2B+Stresroak	6.60_{D} ± 0.24	4.20_{A} ± 0.37	4.40_{A}^{b}± 0.24	4.80_{B}^{cd} ± 0.20	5.80_{C}^{de} ±0.20	7.20_{E}^{f} ± 0.49
VII	F + Neem	6.60_{D} ± 0.24	4.20_{A} ± 0.37	4.80_{B}^{bc}±0.37	5.40_{C}^{de} ± 0.40	6.40_{B}^{e} ± 0.40	4.60_{A}^{c} ± 0.40
VIII	R_2B + Neem	6.60_{C} ± 0.24	4.20_{A} 0.37	4.40_{A}^{b}± 0.24	4.00_{A}^{bc} ± 0.37	5.40_{B}^{cd}± 0.24	7.80_{D}^{g} ± 0.37
IX	F + R_2B + Neem	6.60_{C} ± 0.24	4.20_{A} 0.37	5.00_{B}^{c} ± 0.32	5.20_{B}^{de} ± 0.49	7.00_{C}^{f} ± 0.55	7.60_{D}^{fg} ± 0.68
X	Control	6.60_{F} ± 0.24	4.20_{E} ± 0.37	3.40_{D}^{a}± 0.40	2.40_{C}^{a} ± 0.24	1.40_{B}^{a} ± 0.24	0.00_{A}^{a} ± 0.00

Means in a row bearing a common subscript do not differ significantly

Means in a column bearing a common superscript do not differ significantly

N.B. 1) Vaccination was done on 7th day of age with F strain and R_2B (inactivated oil adjuvanted) by oculonasal and intramuscular routes respectively.

2) F = F strain of NDV; R_2B = Inactivated oil adjuvant vaccine of NDV.

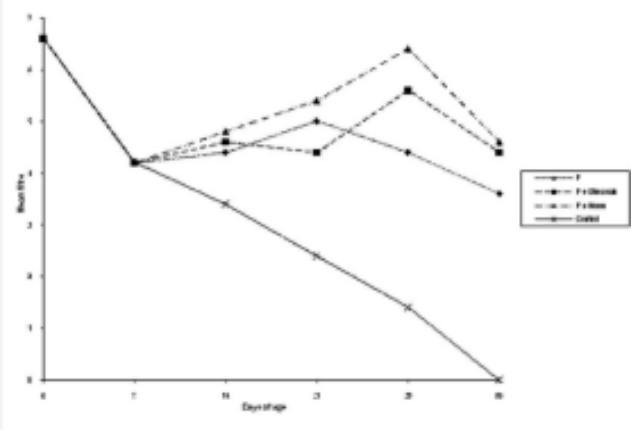

Fig 1. Variation in mean reciprocal HI antibody titre (log 2 scale) in birds vaccinated with F strain of Newcastle disease virus and its combinations with immunomodulators

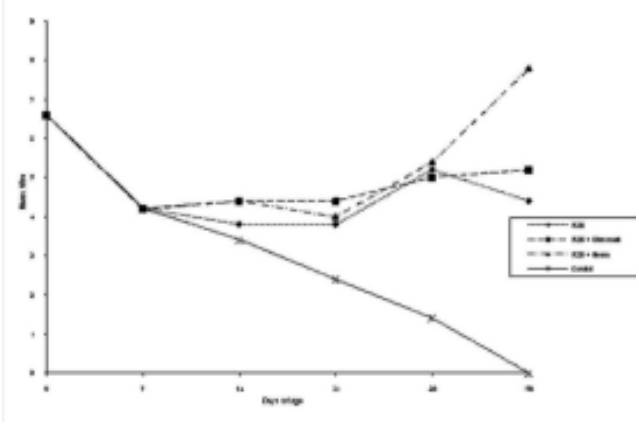

Fig 2. Variation in mean reciprocal HI antibody titre (log 2 scale) in birds vaccinated with R_2B strain of Newcastle disease virus and its combinations with immunomodulators.

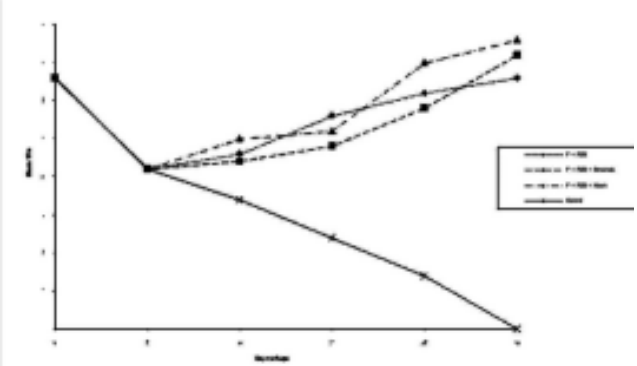

Fig 3. Variation in mean reciprocal HI antibody titre (log 2 scale) in birds vaccinated with F and R_2B strain of Newcastle disease virus and its combinations with immunomodulators.

the vaccinated groups (I to IX) exhibited 100 per cent HI indices, while the control (group X) showed 40 per cent HI index.

The reason for this variation in the rise of HI titres (at 14th day and 21st day) could be attributed to the triggering effect on the immune system of live F immunization in broiler's life (Samberg *et al.*, 1977).

At three weeks post vaccination, *i.e.* by the end of the 4th week (28th day) of age (Table II) the mean HI titre in all the vaccinated or treated birds of groups II to IX continued to rise and reached their peak levels in group II (R_2B), IV (F + Stresroak) and VII (F + Neem) with mean titres of 5.20 ± 0.37, 5.60 ± 0.24 and 6.40 ± 0.40 respectively, while group I (F) showed a slow declining phase from 3rd week onwards with 100% HI indices in all the groups (I to IX). On the contrary, the control group (group X) birds showed mean HI antibody titre of 1.40 ± 0.24 with negative HI index at this stage. The phenomenon of increase in HI titres at this stage might be from the fact that the inactivated oil adjuvanted R_2B virus antigen might have boosted the immune response slowly and gradually by releasing the virus particles from the trapping oil environment, thereby stimulating the immune responses. This study was in agreement with Samina *et al.* (1999).

The mean HI titres by the end of 8th week (56 days) of age (Table II), in chicks of group III (F & R_2B), groups V (R_2B + Stresroak), VI (F + R_2B + Stresroak), VIII (R_2B + Neem) and IX (F + R_2B + Neem) reached their peak . All the vaccinated groups (II to IX) showed 100% HI indices except group I that showed an 80% HI index, while the control birds (group X) exhibited a complete waning of maternal antibody. The significant increase in HI antibody titre in these groups might be attributed to the administration of oil adjuvanted inactivated R_2B vaccine alone or in combination with Stresroak (Kim *et al.*, 1989; Bora *et al.*, 1999 and Sarmah, 2001) as well as neem.

Amongst all the various vaccinated groups of birds, the group vaccinated with F strain of NDV vaccine (group I) had significantly lower ($P < 0.01$) HI antibody titre than all other vaccinated groups (II to IX). The vaccinated cum treated groups, groups IV, V and VI (Stresroak treated) and groups VII, VIII and IX (Neem treated) had significantly higher ($P < 0.01$) antibody titres. The birds of neem treated groups specially group VIII (R_2B + Neem) and IX (F + R_2B + Neem) had higher antibody titres, which was significantly high ($P < 0.01$)

amongst all the group combinations.

The present findings were also in conformity to a number of workers (El Sayed *et al.*, 1981; Kim *et al.*, 1989; Patel *et al.*, 1992; and Cajavec *et al.*, 1995), who obtained similar results on simultaneous and/or subsequent vaccination against ND in chickens by using live lentogenic strain vaccine as well as oil emulsion inactivated lentogenic/mesogenic strain vaccines.

The birds vaccinated with F and/or R_2B and receiving Stresroak reflected higher post vaccination HI titres at different ages as compared to the vaccinated birds without supplementation with immunomodulators. Thus, Stresroak might certainly be assumed to augment immune response in broiler chicks vaccinated with F strain/R_2B in ND. Similar findings were observed by Bora (1999) and Sarmah (2001) where they concluded that Stresroak possessed immunomodulatory properties when given in birds vaccinated against ND.

The birds vaccinated with F and/or R_2B receiving *Azadirachta indica* had the highest post vaccination HI titres at different ages as compared to that of vaccinated birds without treatment as well as vaccinated cum treated with Stresroak. From the present study, it was seen that *Azadirachta indica* certainly augmented the immune response in broiler chicks vaccinated with F and/or R_2B with the highest HI titres. As *Azadirachta indica* constituents azadirachtin, 3-deacetyl-3- cinnamoylazadirachtin, I-tigloyl-3-acetyl-II-methoxyazadirachtin which helps neem to acts as a strong immunomodulators. This observation was further substantiated by the findings of Sadekar *et al.* (1998).

Stresroak and Neem when given together was found to have a boosting effect on the antibody level in birds (Sarmah 2001). The constituents of stresroak are *Withania sominifera, Ocimum sanctum, Phyllanthus emblica, Magnifera indica and Shilajit*. Some of the ingredients like *Withania sominifera, Ocimum sanctum* and *Magnifera indica* have been reported to exhibit immunomodulatory effects (Ziauddin *et al.*, 1996)

Based on the present study, it might be concluded that simultaneous vaccination with NDV (live + inactivated) along with neem was the best and economic way of conferring protective immunity to the birds against NDV.

From the overall observations of the present study, it could be concluded that Stresroak and Neem had a beneficial effect on the birds, since these preparations not only augmented the humoral immune response but also conferred suitable immunity to challenge with virulent virus upto the market age of broilers, where the immunopotentiating response of *A. indica* was found significantly higher ($P < 0.01$) than that of Stresroak.

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Therapeutic potential of bovine colostrum in *E. coli* induced diarrhea in rabbits

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Abstract

Weaned rabbits, caged individually and maintained on standard ration, were utilized to evaluate the therapeutic potential of bovine colostrum. Three different doses were used to assess its antioxidant potential. The best antioxidant dose was subsequently used alone as well as in combination with standard therapy in *E. coli* induced diarrhea. The effect on clinical profile, serum IgG, hemato-biochemical parameters, erythrocytic lipid peroxidation and antioxidant enzyme activity were observed. Rabbits on the day of diarrhea were dull, depressed, showed loss of appetite and significant oxidative stress. The treatment regimens adopted revealed that bovine colostrum in conjunction with standard antimicrobial (ciprofloxacin) exhibited better therapeutic effect as compared to either alone. It was also observed that lyophilization had no detrimental effect on colostral IgG concentration, hence could be used as means of preservation for this nutraceutical.

Keywords: Antioxidant, Colostrum, Diarrhea, Lyophilization, Nutraceutical, Rabbits

Colostrum is the 'early' milk produced postpartum (Gopal and Gill, 2000). For a new born, bovine colostrum (BC) provides not only nutrition, but also protection against infection while immune system is still developing. It has a nutrient profile and immunological composition that differs substantially from 'mature' milk. It is rich in oligosaccharides, natural antimicrobials, immune regulation factors (Kelly, 2003) and antioxidant factors.

In commercial rabbit-fattening farms, enteritis caused by *Escherichia coli* is the main cause of morbidity and mortality in weaned rabbits. The gut of healthy weaned rabbits contains low levels of *E. coli* due to inhibitory influence of the cecal volatile fatty acids and the disease is associated with colonization and proliferation of *E. coli* in distal ileum and cecum (Blanco *et al.,* 1996).

Published reports have shown that colostrum and its components are effective against a wide range of common pathogens, including rotavirus, *Cryptosporidium spp.*, *Staphylococcus aureus*, *Candida spp.*, *Clostridium spp.*, Feline immunodeficiency virus, *Shigella spp.*, *Streptococcus spp.* and *E. coli* (Taillon and Andreason, 2000). The components of colostrum that convey its nutraceutical properties are nearly identical in structure and function among different species (Anderson *et al.,* 1989), thus can be used effectively to provide benefits across the species. BC is used in nutraceutical medicine due to the relative ease with which large amounts can be collected and processed, and its proven effectiveness when used to treat a variety of species.

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Materials and Methods

The first milking colostrum (3L) was collected from cows immediately after calving under proper sanitation to avoid contamination. It was frozen at -20° C and later on subjected to lyophilization to form freeze dried powder.

Lyophilized BC was analyzed for IgG content using standard method of single radial immunodiffusion (SRID) (VMRD Inc.) for identifying any loss of IgG on processing of BC.

New Zealand White (NZW) weaned rabbits of almost similar age group were acclimatized for seven days in the divisional laboratory animals shed before starting the experiment, were housed in individual cages and maintained on standard ration with *ad libitum* availability of drinking water.

The dose of BC was selected after evaluating the effect of three different doses of BC supplementation (Dose I= 200 mg/kg b wt *per os*, Dose II = 400 mg/kg b wt *per os* & Dose III = 600 mg/kg b wt *per os*) on lipid peroxidation (LPO), erythrocytic superoxide dismutase (SOD) and catalase (CAT) activity in healthy rabbits for 15 days. The dose which showed the best

anti oxidant effect was used for evaluating its therapeutic potential in *E. coli* induced diarrhea in rabbits.

The study was conducted using 30 healthy weaned NZW rabbits of similar age group divided in five groups of 6 animals each under standard feeding and managemental conditions.

Diarrhea was induced by oral administration of *E.coli* culture (Blake and Cantey, 1977) to rabbits of all the groups except healthy control group. The rabbits were fasted overnight and then challenged *per os* with 2 ml of the bacterial suspension (2 x 10^8 cfu/ml of *E. coli*) preceded by *per os* 5 ml of 10% sodium bicarbonate. They were then allowed food and water *adlib*. Diarrhea was recorded as present when either perineal soiling was seen or soft and unformed stool was recovered from rectal swabs. Fecal shedding was recorded as positive when lactose-fermenting Gram negative bacteria were recovered on MacConkey's plates after overnight incubation at 37^0 C or negative when no lactose fermenting bacterial growth was recorded on MacConkey's plates after overnight incubation.

The standard therapy (Antimicrobial) to be given to diarrheic rabbits was selected on the basis of antibiotic sensitivity testing.

The fecal consistency, depression and dehydration scores (0-3) were noted on the day of onset of diarrhea and after completion of treatment as per the method of Walker *et al.* (1998).

Freshly collected blood using EDTA as anti-coagulant was utilized to estimate PCV (microhaematocrit).

Total serum proteins were estimated using standard kit (Span diagnostics).

Serum IgG values of rabbits were estimated using SRID, VMRD inc.

The levels of LPO, SOD and CAT were estimated using 10% RBC hemolysate.

Statistical analysis

The data were analyzed statistically using ANOVA and't' test to find out the significance of difference in mean values (Snedecor and Cochran, 1994).

Results and Discussion

In this study no growth was observed after streaking the lyophilized BC on different media like MacConkey's agar, EMB agar and Heller's Enteric agar. Hence, it was considered safe for use in the study. Similar observations were made by Arguello *et al.* (2003) and Bilbao *et al.* (2001).

The IgG content of frozen BC and its lyophilized powder was 7600±178.92 mg/dl and 6933±169.51 mg/dl respectively indicating that lyophilization decreased the content of IgG non significantly ($p<0.05$) by 8.77% (Fig.1).This is in agreement with Elfstrand *et al*. (2002) and Nagaraja (2010).

Dosages of BC used in clinical studies generally ranged from 500 mg to 10 g/day for adult humans (Solomans, 2002). Dohler and Nebermann (2002) demonstrated 80% reduction in plasma endotoxin values in rats receiving 400 mg/kg b wt. Giffard *et al.* (2004) found significant improvement in fecal score of puppies aged 3-4 months using 500 mg BC powder/day. All the 3 doses (Dose I, Dose II & Dose III) of BC tested showed a significant ($P<0.05$) antioxidant effect as evident from significant reduction in LPO and increase in SOD and CAT activity in a dose dependent manner (Fig.2). This could be due to presence of various enzymatic antioxidants which include lactoperoxidase, catalase, superoxide dismutase, glutathione peroxidase and non-enzymatic antioxidants such as vitamins E, A, C, lactoferrin, selenium, copper, zinc and cysteine (Przybylska *et al*., 2007). The findings are in confirmation with Alberti-Fidanza *et al.* (2002), Choi *et al.* (2007) and Zhao *et al.* (2004). Based on the results, Dose III was selected for evaluating therapeutic potential of BC in *E. coli* induced diarrhea in rabbits.

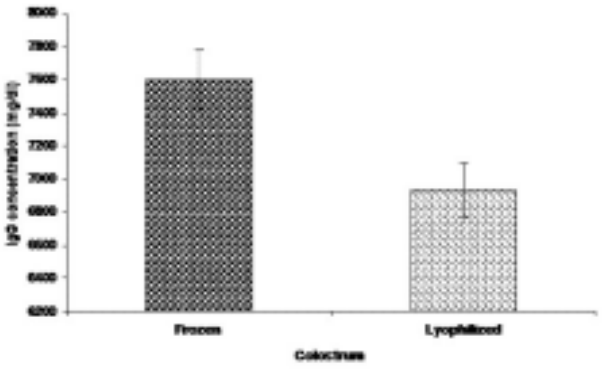
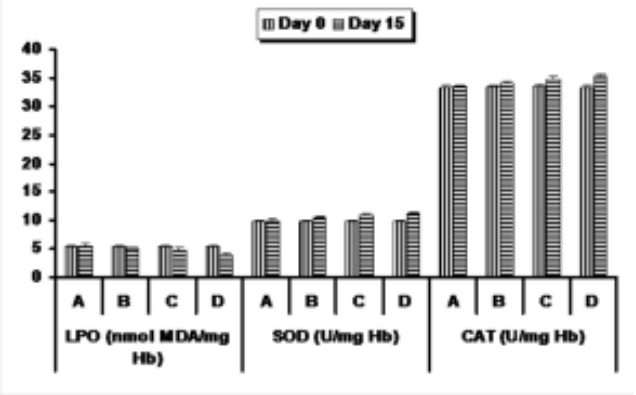


Fig.1: Effect of lyophilization on colostral IgG concentration



Gr. A : Control; Gr. B : BC Dose I; Gr. C : BC Dose II; Gr. D : BC Dose III

Fig.2: Effect of BC supplementation on oxidative stress indices

Diarrhea was successfully induced in rabbits by 64±8 hours after oral administration of *E. coli* culture containing $2x10^8$ cfu/ml. The administration of 10% $NaHCO_3$ before *E. coli* challenge was done to overcome the gastric acidity which could have adversely affected the survival of *E. coli* interfering with the induction of diarrhea. Similar findings were observed by Blake and Cantey (1977) and Cantey and Inman (1981). Day 3 was considered as day of onset of diarrhea as well as administration of therapies.

Antibiotic sensitivity testing revealed that *E. coli* was most sensitive to fluoroquinolones and the order of sensitivity observed was ciprofloxacin > ofloxacin > enrofloxacin > amikacin > ceftrioxone > cefotaxime. Ciprofloxacin was used @ 10 mg/kg b wt *per os* daily for three days. Milon *et al.* (1999) suggested that wide spectrum antibiotics such as B lactamases, lincomycin and clindamycin are highly toxic for the rabbit, essentially because they induce tremendous dysequillibrium of the digestive flora. Narrow spectrum antibiotics, such as polypeptides or quinolones may help to reduce mortality during epizootics.

Diarrheic rabbits, irrespective of groups, were found to be lethargic, dull, depressed and dehydrated with reduced appetite as evidenced by increased fecal, depression and dehydration scores. Similar clinical findings were observed in *E.coli* diarrhea of various species of animals (Changkija, 2002). In response to treatment regimen adopted there was a significant reduction in clinical scores (fecal, depression and dehydration scores) in groups receiving either

Table 1: Haemato-biochemical alterations in response to different treatments.

Parameters	Group	Days	
		3	7
PCV (%)	I	38.12±2.86	38.29±2.07[a]
	II	48.66±0.60	40.16±0.60[*a]
	III	47.26±0.77	45.08±0.28[b]
	IV	45.09±0.94	39.12±0.01[*a]
TSP (mg/dl)	I	6.81±0.18	6.88±0.10[a]
	II	8.93±0.40	7.13±0.10[*a]
	III	8.94±0.15	8.38±0.16[b]
	IV	8.83±0.11	7.01±0.81[*a]
LPO (nmol MDA/mg Hb)	I	5.85±0.19	5.85±0.32[a]
	II	7.67±0.11	5.91±0.12[*a]
	III	7.82±0.10	7.54±0.37[b]
	IV	7.72±0.16	5.88±0.21[*a]
SOD (U/mg Hb)	I	9.43±0.06	9.47±0.17[a]
	II	8.32±0.31	9.10±0.13[*a]
	III	8.31±0.09	8.71±0.11[b]
	IV	7.61±0.10	9.48±0.21[*a]
CAT (U/mg Hb)	I	31.29±0.09	31.32±0.27[a]
	II	29.10±0.28	30.61±0.32[a]
	III	29.14±0.21	30.11±0.37[a]
	IV	29.41±0.30	31.21±0.12[*a]
IgG (mg/dl)	I	1243.33±117.15	1238.33±146.21
	II	1235.00±164.81	1246.67±137.76
	III	1246.67±148.81	1246.67±144.26
	IV	1243.33±133.82	1250.83±123.83

Gr. I = Healthy control; Gr. II = Standard therapy (ST) only; Gr. III = BC only; Gr. IV = ST + BC

ciprofloxacin alone or a combination of ciprofloxacin and BC. Similar observations were made by Thapa (2005) and Harish (2008).

The haemato-biochemical alterations observed are given in Table 1. Significant increase in PCV on day 3 of all the groups was a direct reflection of the accompanying dehydration due to loss of water through diarrhea. This was further supported by an increase in total serum proteins as seen in all the groups. It was observed that hemato-biochemical parameters returned towards physiological values in response to standard treatment along with BC. Changkija (2002) reported significantly higher PCV and TSP values attributable to dehydration. Harish (2008) observed significant increase in total serum proteins in dogs suffering from enteritis.

As regards no significant change in serum IgG values in rabbits treated with either ciprofloxacin or BC or both, the finding is on the expected lines of general thinking that beyond the absorptive phase (24-48 hours after birth), IgG is not absorbed being a macromolecule. Further, very short period of treatment (3 days only) with BC in this study could be an additional reason for no increase in serum IgG. This is in agreement with Mero *et al.* (1997).

A significant oxidative stress was observed in the diarrheic rabbits as evidenced by increase in LPO and concomitant decrease in erythrocytic SOD and CAT activity. BC supplementation reduced the oxidative stress by enhancing the antioxidant enzyme activity. This might be supported by the fact that lactoferrin along with vitamin A, D, C and E contained in BC might act as an antioxidant in the GI tract and might provide a nutritional ingredient for the treatment of intestinal disorders associated with hyperpermeability, oxidative stress and inflammation (Shoji *et al.,* 2007). Vitamin C in colostrum might have contributed in maintaining the redox integrity of cells and thereby protecting against ROS generated during the respiratory burst and the inflammatory reactions (Wintergerst *et al.,* 2006). There are similar reports of increased lipid peroxidation and impaired antioxidant defense system in diarrheic patients (Nieto *et al.,* 2000, Murmu, 2006 and Moorthy and Murali, 2007).

The critical analysis of different parameters indicated that supplementation of BC @ 600 mg/kg b wt with a standard antibiotic therapy (ciprofloxacin) has given the best therapeutic effect as compared to either antibiotic or BC alone. This could be attributed to factors other than systemically absorbed IgG which include lactoferrin, oligosaccharides, proline rich polypeptides, cytokines, lactalbumin, EGF, IGF-I, IGF-II, TGF-α and TGF-β etc (Playford *et al.,* 2000). As regards the field applicability of the proven antioxidant potential of BC, its supplementation in newborns irrespective of the species during the critical stress period of first two months will be of great help in maintaining their proper health. It is known that during the first two months of their life, newborns are subjected to varied stress either as a result of changes in the diet and environment or diseases particularly of gastro-intestinal and respiratory tract. It may be concluded that therapeutic advantages might be gained by developing colostrum formulations especially tailored for individual conditions of gut, thereby reducing the incidence of gut infection while stimulating the gut repair.

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Association of Brucellosis with abortion, retention of placenta and repeat breeding in Bovine of organized and unorganized dairy farms of Jammu

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Abstract

The present study was conducted to investigate the seroprevalence of brucellosis in relation to abortion, retention of placenta (ROP) and repeat breeding in cattle and buffaloes from organized and unorganized dairy farms. A total of 81 serum samples (50 from buffaloes and 31 from cattle) were subjected to rose bengal plate agglutination test (RBPT) and serum tube agglutination test (STAT). The overall prevalence of brucellosis was found to be 14.81%. The prevalence was non-significantly higher (chi-square=0.069, P=0.793) in cattle (16.12%) as compared to buffaloes (14.0%). The risk of brucellosis was 2.059 times more in organized farms than in unorganized farms (95% C.I 0.644-5.557). The disease was found to be non-significantly associated with abortion (chi-square=0.0649, P=0.420) and retention of placenta (chi-square=0.069, P=0.793), however there was no association with repeat breeding (chi-square= 1.572, P=0.210).

Keywords: Seroprevelence, Brucellosis, Cattle, Buffalo, Abortion, Retention of placenta, Repeat breeding

Brucellosis has been a major long standing threat to livestock and mankind worldwide. The disease is caused by various species of the genus *Brucella*, which are facultative, intracellular bacteria capable of surviving and multiplying inside the cells of mononuclear phagocytic system (Jarvis *et al.,* 2002). The disease in bovines is characterized by abortion, retention of placenta, repeat breading and infertility (Singh *et al.,* 2002) and causes huge economic loss to dairy industry which includes, loss in milk production, low fertility rates, high cost of treatment and replacement of animals (McDermott and Arimi, 2002). Although the disease has been eradicated from bovines in some countries, but still the disease has worldwide distribution in livestock and human beings, including India (Seleem *et al.*, 2010) where it is endemic in nature (Renukaradhya *et al.*, 2002) and its prevalence greatly varying from one region to other. Thus the present study was planned with a primary aim to assess the epidemiological status of bovine brucellosis in Jammu region of Jammu and Kashmir State with special reference to its association with abortion, retention of placenta (ROP) and repeat breeding.

Materials and Methods

Animals with a history of frequent abortions, retention of placenta and repeat breeding from organized and un-organized dairy farms located in and around Jammu region formed the subject of study. A total of 81 (50 of buffaloes and 31 of cattle) blood samples were collected. Serum was separated and stored at -20º C till analysis. The samples were tested through RBPT method as described by Morgan *et al.* (1969) and by STAT method as described by Alton *et al*. (1975). A titre of 80 I.U per ml and above was considered positive, 40 I.U as doubtful and below 40 I.U as negative for brucellosis in case of STAT. The RBPT antigen and *Brucella abortus* agglutinating antigen for STAT was procured from the Division of Biological Products, IVRI, Izatnagar, Uttar Pradesh. The association of disease in relation to species (cattle and buffalo), management practices (organized and unorganized) and with the history of abortion, retention of placenta and repeat breeding was analyzed through Chi-square test by using an on line programme (http://statpages.org/ctab2x2.html). The sensitivity, specificity and over all agreement between two tests were analyzed by statistical methods as used by Samad *et al.* (1994).

Results and Discussion

Brucellosis testing revealed a prevalence of 16.12% in cattle and 14.0% in buffaloes, with an overall prevalence of 14.81%. Our finding is in agreement with Sharma (1995) who reported 14.61% prevalence in buffaloes of Punjab but is lower as compared to 22.5%

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reported by Gumber *et al*. (2004) and 18.26% reported by Aulakh *et al.* (2008).

The prevalence of brucellosis was non-significantly higher (Chi-square=0.069, P=0.793) in cattle (16.21%) as compared to buffaloes (14.0%). Singh *et al.* (2002) also reported similar finding. Extensive artificial insemination practiced in cattle, as compared to buffaloes may be the reason for higher prevalence in cattle. Moreover, the sale of problem breeders (probably infected) in market may also act as a source for introduction of brucellosis in new cattle herd, while buffaloes are usually culled by slaughter. There was no association of brucellosis with regards to the species because the odd ratio and the relative risk was less than one (Table 1).

In organized farms, the prevalence of brucellosis was non-significantly higher (19.04%) (Chi-square=1.238, P=0.266) as compared to unorganized farms (10.25%). Animals of organized farms were at 2.059 time's greater risk than animals of unorganized farms (95% C.I 0.596-7.035). Similar finding was also reported by Mehra *et al.* (2002). The odd ratio and the relative risk revealed the association of brucellosis with farm management (Table 2). The reason for higher sero-positivity in organized farms may be greater use of artificial insemination as compared to natural service, which provides a chance for magnified perpetuation of disease.

Brucellosis is reported to cause abortion, retention of placenta, repeat breeding, still birth and prolonged inter calving period due to early embryonic death (Roberts, 1999). Out of 81 animals screened, 32 animals had the history of abortion and of these, 6 animals (18.75%) were positive for brucellosis. The disease was non-significantly associated with abortion (Chi-square=0.649, p=0.420) and the risk of abortion was 1.654 times (95% CI 0.505-5.422) higher in animals with brucellosis than without brucellosis as revealed by odd ratio and relative risk (Table 3). Similar association was sort out by Aulakh *et al*. (2008) reporting risk of abortion 4.19 times more in animals having brucellosis than without brucellosis. The results are also in alignment with reports of other workers (Srinivasa *et al*.1999 and Sandhu *et al.* 2001).

During the study, 31 cases had a history of ROP, out of which 5 animals were positive for brucellosis. There was non-significant association between disease and ROP (chi-square=0.069, P=0.793) and the risk of placenta retention was 1.181 times (95% C.I 0.357-3.3931) higher in animals with brucellosis than without. No significant association between repeat breeding and brucellosis (Chi-square= 1.572, P=0.210) was found in the present study (Table 3). Our findings are in agreement with Bachh *et al.* (1988) and Aulakh *et al.* (2008) who reported higher prevalence of abortion and retention of placenta in bovines infected with brucellosis.

Out of the two serological tests used for diagnosis of brucellosis in 81 serum samples, RBPT revealed more positive samples (17) as compared to STAT [12 positives (80 I.U), 3 doubtful (40 I.U) and 1

Table 1: Association of brucellosis with Species.

Species	ST	SP	PP
Buffalo	50	7	14
Cattle	31	5	16.12
Total	81	12	14.81

(Chi Square= 0.069, P= 0.793), Odd Ratio 0.847 (95% CI 0.254-2.798), Relative Risk 0.868 (95% CI 0.315-2.454), ST – Samples Tested, SP- Samples Positive, PP- Per cent Positive

Table 2: Association of brucellosis with farm Management.

Type of Farm	ST	SP	PP
Organized	42	8	19.04
Unorganized	39	4	10.25

(Chi Square=1.238, P= 0.266), Odd Ratio 2.059 (95% CI 0.596-7.035), Relative Risk 1.857 (95% CI 0.644-5.557) ST – Samples Tested, SP- Samples Positive, PP- Per cent Positive

Table 3: Association of brucellosis with Abortion, Retention of placenta (ROP) and Repeat breeding.

History	Species	ST	SP	PP
Abortion	Buffalo	22	4	18.18
	Cattle	10	2	20
Total cases		**32**	**6**	**18.75**
(Chi Square= 0.649, P= 0.420), Odd Ratio (OR) 1.654 (95% CI 0.505-5.422), Relative Risk (RR) 1.531 (95% CI 0.554-4.206)				
ROP	Buffalo	17	2	11.76
	Cattle	14	3	21
Total cases		**31**	**5**	**16.12**
(Chi Square= 0.069, P= 0.793), Odd Ratio 1.181 (95% CI 0.357-3.931), Relative Risk 1.152 (95% CI 0.408-3.178)				
Repeat breeding	Buffalo	11	1	9.09
	Cattle	7	-	-
Total cases		**18**	**1**	**5.56**
(Chi Square= 1.572, P= 0.210), Odd Ratio 0.218 (95% CI 0.044-1.836), Relative Risk 0.318 (95% CI 0.053-1.657) ST – Samples Tested, SP- Samples Positive, PP- Per cent Positive				

negative (<40 I.U) titre]. The sensitivity and specificity of RBPT when compared with STAT was 100% and 92.75% respectively, with 93.82% overall agreement between the two tests. The results were in alignment with that of Sarumathi *et al*. (2003). Thus it was concluded that RBPT is more sensitive but less specific as compared to STAT for the diagnosis of brucellosis.

In sum, brucellosis is fairly prevalent in bovines of Jammu region and is associated with economic losses to the farmers, in the form of abortion and retention of placenta.

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Short Communication

Therapeutic efficacy of Anticoccidial drugs in Goats

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Abstract

The goal of present study was to evaluate the effectiveness of different drugs as a therapeutic agent against coccidiosis in goat. A total of 24 goat naturally infected with *Eimeria* sp. were divided in 4 groups and subjected to different therapeutic regimens. The therapeutic efficacy of the different drugs was evaluated on the 0 day, 3rd day and 1 week post-therapy and was based on the disappearance of clinical signs, reduction in OPG count. Amprolium hydrochloride, Sulphamethoxizole, Sulphaquinoxiline, and Turmeric revealed overall of 91.45, 70.49, 69.78 and 19.11% efficacy against coccidiosis in goats, respectively. The present study suggest that Amprolium hydrochloride is more effective as compared to sulpha drugs to check coccidiosis infection in goat. Turmeric powder did not show any significant reduction in fecal cyst counts of coccidiosis infection at the concentrations tested.

Keywords: Goats, Coccidia, Amprolium hydrochloride, Sulphaquinoxiline, Sulphamethoxizole

Goats have numerous internal parasites; one of the most important is the protozoan Coccidia. Kids are more susceptible to this infection and can be responsible for reduced animal performance and death. Nine different species of *Eimeria* can cause coccidiosis in goats, but some cause much more damage than others. Although a high per centage of adult animals may be infected, only a small per centage of them become sick (Dai *et al*., 2006). Coccidia only cause disease when their numbers become so great that pathological damage is done to the host. Age related resistance to clinical coccidiosis is reported in all ruminants. There is a steady decrease in oocyst count from 6 month to 6 years of age, followed by an increase in goats of 7 years and older, as immunity begins to wane in older goats (Kanyari, 1988). Hot, humid weather is particularly conducive to sporocyst development and out break of clinical coccidiosis are common in temperate region during summer. Clinical coccidiosis develops within 1-2 weeks of ingestion of a large dose of infective sporocysts leading to severe blood loss, abdominal discomfort, decrease appetite, crying and frequent rising up and lying down. Feces may first be unpelleted, then pasty and then, a watery yellow green to brown diarrhea develops. If dehydration becomes severe, animal will become recumbent with cold extremity and sub normal temperature (Smith and Sherman, 1994).

Materials and Methods

The present study was conducted on the experimental goats (24 in No.) of medicine department and were housed in the experimental shed of College of Veterinary and Animal Sciences. The goats were placed in pens in a barn with concrete floor. Wood shavings were applied over the ground as bedding material. Each pen had its own feeder and watering system. Clean water was available ad libitum to the animals at all times. The animals were sheltered in a barn during the evening hours and let out to pasture during the day. Twentyfour goats which were naturally infested with coccidiosis were randomly divided into four homogenous treatment groups of six animals. Groups were given treatment by Amprolium hydrochloride 20mg/kg (Yvore, 1984), Sulphamethoxizole + Trimethoprim 30mg/kg, Sulphoquinoxiline 1.3g/kg (Guss, 1977), and Turmeric 1g/kg orally. Clinical diagnosis was done by history, clinical examination and laboratory diagnosis was done on the basis of laboratory finding of fecal examination.

Table 1: Average OPG Count of Different Drugs at 0, 3, 7 days with efficacy in%.

Groups	Treatment	Average OPG Count			Efficacy (%)
		0 day	3rd day	1 week	
1	Amprolium	6141.667±497.41	950.000±427.785	525.000±196.850	91.4518
2	Sulphamethoxizole + Trimethoprime	6016.667±568.037	3375 ±853.082	1775±500.7494	70.498
3	Sulphaquinoxiline	6066.667±529.779	3133.333±688.234	1833.333±302.765	69.7802
4	Turmeric powder	6058.333±524.801	5583.333±800.416	4900±404.969	19.1197

Fecal samples were collected from the rectum of the animals. Aliquots (Feces) of two grams of the fecal sample from each goat were used for laboratory analysis. Fecal samples were analyzed on the day of feces collection by using a modified McMaster tech-nique (Paracount-EPG™, 1984). The number of oocysts counted on the McMaster slide was multiplied by 50 to get the parasite oocysts per gram of feces for each animal. Common supportive treatment camprising Hematinics (Feritas, inj.), Ringer Lactate, antispasmodic (Spasmovet), Liver extract and B complex was given.

Results and Discussion

Oocyst per gram of feces of group 1, 2, 3 decreased in the day 3, and week in all treated groups except group 4; the greatest reduction was observed in day 3. Amprolium hydrochloride, Sulphamethoxizole, Sulphaquinoxiline, and Turmeric revealed overall of 91.45%, 70.49%, 69.78% and 19.11% efficacy against coccidiosis in goats, respectively. Turmeric powder did not show any significant reduction in fecal oocysts counts of coccidiosis infection at the concentrations tested. The present study suggesting the Amprolium hydrochloride is more effective as compared to sulpha drugs to check coccidiosis infection in goat. Amprolium hydrochloride has shown maximum efficacy but it has got limitation as long term therapy of it can cause polioencephalomalacia. Sulpha drug have less efficacy than Amprolium hydrochloride but it has a added advantage of controlling secondry bacterial infection (Smith and Sherman1994). In addition to the use of coccidiostats, implementation of management changes is important in controlling outbreak of coccidiosis. Attempt should be made to reduce the exposure to infective sporocysts by removal of contaminated bedding and feed, reducing the stocking rate, and moving animal to new uncontaminated environment.

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Efficacy of pimobendan in the treatment of congestive heart failure associated with Dilated Cardiomyopathy in dogs

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Abstract

Total 20 clinical cases of congestive heart failure (CHF) associated with dilated cardiomyopathy(DCM) were divided into two equal groups. Dogs of gr-I were treated orally with pimobendan 0.25mg/kg b.wt bid and gr-II with digoxin 0.22 mg/m^2 bid. In addition all the dogs orally received enalapril 0.5mg / kg b.wt once daily and furosemide 2mg/kg b.wt. b.wt bid and treatment was continued for 45 days. Therapeutic efficacy was determined by comparative assessment of M-mode left ventricular indices between dogs of two treatment groups. Combination of pimobendan, enlapril and furosemide was found to be effective in the treatment of CHF associated with DCM.

Keywords: Pimobendan, Dog, Congestive Heart Failure, Dilated Cardiomyopathy

Dilated cardiomyopathy (DCM) in dogs represents a complex clinical syndrome involving many pathophysiological mechanisms and characterized by chamber dilatation and myocardial systolic and diastolic dysfunction (Tidholm *et al*., 2001). Impairment of left ventricular function in dogs with DCM results in congestive heart failure (CHF), therefore strategy is directed towards relieving the signs of CHF and improved the quality of life. Owing to cumulative toxic effect of cardiac glycosides, their prolonged use is not without danger. Recent studies suggest that pimobendan appear to be safe and effective positive inotropic and inodilator in the clinical management of CHF secondary to DCM in dogs (Lombard *et al*., 2006). Therefore, an attempt was made to study the efficacy of pimobendan in the treatment of CHF associated with DCM in dogs.

Materials and Methods

The study was conducted on 20 clinical cases of CHF associated with DCM, brought to the Teaching Veterinary Clinical Service Complex, College of Veterinary Science, Tirupati. Dogs with clinical signs of cardiac insufficiency were subjected to detailed clinical examination, (Boddie, 2000) thoracic radiography (Fagin, 1988) and electrocardiography (Tilley, 1992). Dogs that were suspected for DCM were subjected to M-mode echocardiographic evaluation to confirm the diagnosis and quantifying the left ventricular dysfunction (Kienle and Thomas, 1995). Criteria for inclusion were (a.) M-mode echocardiographic evidence of fractional shortening below 20 per cent and visualization of dilated ventricles (b) absence of other significant imaging lesions by two - dimensional echocardiography. The dogs affected with CHF were divided into two equal groups. Gr-I dogs were treated orally with pimobendan @ 0.25mg/kg b.wt bid and Gr-II received orally digoxin @ 0.22mg/m^2 bid. In addition all the dogs received enalapril 0.5 mg/kg b.wt once daily and furosemide 2 mg/kg b.wt bid. Treatment was given for 45 days. The pet owners were advised to restrict physical activity and to give low salt diet to the animals. Therapeutic efficacy was known on the basis of comparative assessment of M-mode left ventricular contractility indices, (Fractional shortening and Ejection fraction) between dogs of Gr-I and II as on 0 and 45th day post-treatment. The data obtained were analyzed by paired-"t" test as per Snedecor and Cochran, (1994).

Results and Discussion

After 45 days of treatment, clinical signs were minimized and risk of clinical deterioration of heart failure was reduced in dogs of both treatment groups. However, some cases started showing signs of clinical improvement between 15 and 30 days of treatment in both groups. An appreciable reduction in pulmonary edema but not in heart size was observed in all the treated dogs. These finding are in agreement with report of Fuentes *et al*. (2002) and Atkins *et al*. (2002). The gradual reduction in pulmonary edema in CHF dogs can be attributed to the diuretic effect of furosemide and blocking of aldosterone induced renal retention of sodium and water by enalapril (Martin, 2003). The qualitative changes in electrocardiogram were significant between groups when compared to pre-treatment values.

Table 1: Left ventricular M-mode dimension in treatment groups (Mean±SE in cms)

Parameter	Treatment group	Pre-treatment	Post-Treatment
$LVID_d$	I	5.57±0.05	4.81±0.03**
	II	5.39±0.03	4.52±0.04**
$LVID_S$	I	4.32±0.04	3.52±0.02**
	II	4.25±0.05	3.41±0.03**
$LVPW_d$	I	074±0.02	0.87±.0.01**
	II	0.76±0.03	0.86±0.01**
$LVPW_S$	I	1.07±0.02	1.39±0.02**
	II	1.09±0.04	1.41±0.04**
IVS_d	I	0.85±0.02	$0.77±0.02^{NS}$
	II	0.86±0.01	$0.88±0.01^{NS}$
IVS_S	I	1.27±0.03	$1.26±0.02^{NS}$
	II	1.26±0.01	$1.28±0.03^{NS}$

Table 2. Mean + SE of left ventricular M-mode contractility indices in treatment groups (in per cent)

Parameter	Treatment group	Pre-treatment	Post-Treatment
Fractional	I	18.14± 4.38	29.04±0.72**
shortening	II	18.29± 0.19	24.58 ± 0.39**
	I	36.42± 0.68	58±61**
Ejection fraction	II	36.93±0.45	48.21±0.21**

**P<0.01.

The left ventricular dimensions of pre-treatment and post-treatment in two groups are shown in table1. Highly significant reduction in values $LVID_d$ and $LVID_S$ was observed in both treatment groups when compared to pre-treatment values. Highly significant elevation in values of FS and EF was observed in both groups when compared to pre-treatment values (Table 2).The pre treatment values of M –mode left ventricular contractility indices in all two groups were subjected to complete randomized design analysis. There was no significant difference in pre treatment values of dogs among the group I and group II. Hence pretreatment values out of total 20, ten values from each parameter were taken randomly and used as a pre treatment group. Therapeutic efficacy of the drugs were made on the basis of comparative assessment of left ventricular contractility indices among the group I and group II on pre and 45 day post treatment. There was highly significant difference in post-treatment values of contractility indices in Gr-I when compared to Gr-II. The positive inotropic effect of digoxin, angiotensin converting enzyme inhibitor action of enalapril and diuretic effect of furosemide are well documented (Martin, 2003). The elevation of ventricular contractility indices could be attributed to the combining calcium sensitizing properties with cyclic AMP phosphodiesterase – III inhibition action of pimobendan in group-I. The present findings concur with report of Fuentes *et al.* (2002). The study suggests that combination of pimobendan, enalapril and furesemide are useful in the management of CHF associated with DCM in dogs.

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Therapeutic efficacy of epipleural blockade for the treatment of aspiratory pneumonia in buffalo Calves

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Abstract

Therapeutic efficacy of epipleural blockade for the treatment of aspiratory pneumonia in buffalo calves studied & study revealed early and complete recovery in animals treated with epipleural blockade, steptopenicilline and prednisolone after induction of aspiratory pneumonia which may be due to control of inflamation.

Keywords: Epipleural Blockade, Treatment, Pneumonia, Buffalo.

Aspiratory pneumonia is a common problem in ruminants. It may occur due to regurgitation of ruminal contents in respiratory tract or faulty drenching (Fenwic, 1969). In the present study*, therapeutic effect of local anaesthesia using 0.5% procaine hydrochloride for epipleural blockade in aspiratory pneumonia has been evaluated on the basis of clinical, physiological and biochemical profiles in male buffalo calves.

Procaine has been used for therapeutic purposes in a number of conditions viz. nephritis (Sharda *et. al.,* 2001), cystitis (Sharda *et. al.,* 2007), spasmodic colic, gastro-enteritis, peritonitis and post-castration complications (Mols, 1957).

Twelve healthy, male buffalo calves, 1- 1½ years of age, weighing 60-75 Kg were randomly divided into two equal groups, A and B. Aspiratory pneumonia was created by intra-tracheal administration of 100 ml whole milk (Jadon and Kumar, 1993) and therapy was initiated after 48 hours. The animals of group A were treated daily with intramuscular administration of 1 gm streptopenicillin and 30 mg prednisolone IM daily for 7 days. Animals of group B were treated with epipleural blockade in addition to streptopenicillin and prednisolone as in group A. Bilateral epipleural blockade was performed by depositing 10 ml of 0.5% warm solution of procaine hydrochloride between 12th and 13th thoracic ribs on both sides of the body wall by inserting a 10 cm long, 18 G needle at the anterior edge of the 13th thoracic rib. The needle was directed towards the vertebral body at an angle of 20-30^{0} and advanced parallel to the edge of the 13th thoracic rib until it struck the body of vertebrae. The correct position of the needle in epipleural space was ascertained by absence of air on suction from the thoracic cavity. The entire procedure was repeated 4 – 5 times at 2 – 3 days interval.

The efficacy of the treatment was assessed by evaluating various clinico-physiological (respiration rate and rectal temperature), haematological (TLC and DLC), and biochemical parameters (plasma concentration of glucose, urea nitrogen, potassium and chloride).

All the pneumonic animals exhibited tachypnoea in early stages and inspiratory as well as expiratory dyspnoea later on. Serous nasal discharge, typical bubbling rales, occasional splashing sounds over the chest on auscultation and dry coughing in later stages were observed in the pneumonic animals. Respiratory symptoms were due to the lesions or dysfunctions of the respiratory tract reducing the air supply (Kumar and Singh, 1980). The elevation of rectal temperature also occurred in all the animals after induction of pneumonia. However early return to normal respiratory pattern and body temperature was noted in animals of group B as compared to group A. Similar findings have been reported by Sustronck *et al*. (1997).

A significant increase in TLC was followed by a gradual decrease in all animals. However the decrease was more in animals of group B (Table 1). A marked neutrophilia with corresponding lymphocytopenia observed in all animals after induction of pneumonia. Similar findings were also observed by Jadon and Kumar (1992).

Significant increase in plasma glucose level up

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to 2nd day post-pneumonia was noted in both groups of animals. However, gradual decline to near-normal levels by day 4 and 6 after appropriate treatment was observed in the animals of group B and A, respectively.

A slight non-significant increase in urea nitrogen was observed post-pneumonia, which gradually declined after therapy. The decline was more in the animals group B as compared to the animals of group A at respective time interval. This increase may have been due to dehydration or increased protein catabolism as a result of fever, stress or anorexia (Kaneko *et. al.,* 1997)**.**

Plasma potassium levels increased significantly in both groups post-pneumonia up to day 4. It gradually decreased after treatment and touched normal level on day 12. The increase in the potassium level initially could be due to maintenance of homeostasis (Jadon and Kumar, 1993). The initial decrease in chloride concentration may be attributed to the phenomena of 'chloride shift' caused due to the carbon dioxide imbalance. Plasma hypochloremia may therefore be associated with hypoventilation in the state of pneumonia (Coles, 1967). Moreover, osmotic effect of sodium ions causes passive movement of chloride ions alongwith

Table 1: Clinico-physiological, haematological and biochemical parameters (Mean ± SE) at different intervals.

Parameters	Groups	Post-treatment Days							
		0	1	2	3	4	5	9	12
Respiration rate/min	A	15 ±0.91	19.25 ±0.63**	25.75** ±0.48	22.75** ±1.12	20.25** ±0.85	16.25* ±0.48	14.25 ±0.48	13.00** ±0.41
	B	14.75 ±0.48	19.50** ±0.29	25.50** ±0.29	23.00** ±0.41	21.00** ±0.41	17.75** ±0.48	16.25* ±0.63	13.00** ±0.48
Rectal temperature (ºF)	A	100.95 ±0.10	100.65 ±0.57	102.15** ±0.12	102.1 ±0.44	103.65** ±0.22	103.35** ±0.21	102.40** ±0.58	101.2 ±0.58
	B	101.45@ ±0.33	101.50@@ ±0.50	101.50** ±0.37	101.6 ±0.54	102.25* ±0.30	103.00** ±0.28	102.75** ±0.25	101.55 ±0.25
Total leucocyte count (1000/mm^3)	A	7.225 ±0.42	9.550** ±0.50	13.175** ±0.30	12.850** ±0.33	12.175** ±0.24	11.500** ±0.15	10.155** ±0.19	9.078** ±0.23
	B	7.525@ ±0.17	9.687** ±0.24	13.013** ±0.49	12.813** ±0.23	12.288** ±0.11	11.280** ±0.13	9.665** ±0.27	8.960** ±0.34
Neutrophils (%) DLC	A	38 ±1.83	47.50** ±3.97	64.75** ±2.40	67.00** ±1.47	62.50**@@ ±1.66	54.50** ±1.76	49.50** ±1.70	45.25 ±1.80
	B	37.5 ±1.32	49.75**@ ±1.55	64.50** ±1.32	65.00** ±1.22	60.00** ±1.08	56.00** ±1.58	51.25**@ ±0.85	48.25**@@ ±1.11
Lymphocytes% (DLC)	A	57.5 ±2.02	47.25** ±3.73	31.25** ±2.72	28.75** ±1.80	33.50** ±1.55	41.75** ±1.65	47.00**@ ±1.29	50.25**@@ ±2.06
	B	57.5 ±1.04	45.75** ±1.31	31.25** ±1.55	30.50** ±0.96	36.25**@@ ±1.18	40.00** ±1.78	44.75** ±0.85	47.75** ±1.03
Plasma glucose (mg/dl)	A	70.25 ±2.50	72 ±2.42	78.00* ±2.74	74.75 ±3.75	72.75 ±3.20	69 ±3.76	68.25 ±3.35	69 ±4.08
	B	67.75 ±3.75	70.75 ±1.93	76.50** ±3.95	73.25 ±4.46	74.50@ ±2.02	68 ±0.95	67 ±0.82	67.25@ ±0.95
Plasma urea nitrogen (mg/dl)	A	17 ±2.50	18.25 ±5.12	17.75 ±2.29	19.25 ±2.21	21.25 ±2.94	18.75 ±1.80	19.5 ±0.75	18 ±4.39
	B	18.5 ±3.39	17.75 ±4.13	18.75 ±4.44	17.75 ±4.32	18 ±3.32	19.75 ±3.09	17.75 ±6.29	18.75 ±5.49
Plasma potassium (meq/L)	A	4.55 ±0.19	4.75 ±0.22	5.13** ±0.13	5.60** ±0.12	6.08** ±0.09	5.58** ±0.19	5.25** ±0.14	4.95* ±0.18
	B	4.7 ±0.19	4.88 ±0.22	5.38**@@ ±0.11	5.75** ±0.06	6.10** ±0.12	5.65** ±0.14	5.35** ±0.17	5.13** ±0.14
Plasma chloride (meq/L)	A	107 ±28.29	101 ±8.98	98.5 ±23.13	100.25 ±16.40	102 ±9.23	104 ±13.72	102.75 ±13.26	104.75 ±16.94
	B	99 ±9.21	95 ±15.62	93 ±12.63	95.5 ±11.73	96.5 ±9.85	97.25 ±12.65	97.25 ±22.00	98 ±19.00

* Significant at 5% level as compared to 0 day values, ** Significant at 1% level as compared to 0 day values

@ Significant at 5% level when compared between the groups, @@ Significant at 1% level when compared between the groups

them, thus causing a further decline in the concentration of the latter (Jadon and Kumar, 1993).

The findings of the above study revealed early complete recovery in animals of group B treated with the combination of epipleural blockade, streptopenicillin and prednisolone, after induction of aspiratory pneumonia. This early recovery might be due to controlling inflammation related-irritation associated with pneumonia by epipleural blockade. It was concluded that the management of aspiratory pneumonia with epipleural blockade using 0.5% procaine hydrochloride, streptopenicillin and prednisolone is quite effective and is superior to treatment with streptopenicillin and prednisolone alone.

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Haematobiochemical and Echocardiographic Changes in Downer Cow Syndrome

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Abstract

The study comprised of ten apparently healthy cross bred cows and 58 clinical cases of downer cow syndrome. Thirty eight of these cases were diagnosed within 30 days postpartum, eleven cases of downer cows were diagnosed after 30 days postpartum and nine were in advanced pregnancy. A significant decrease in haemoglobin, packed cell volume and total erythrocyte count was observed in downer cows when compared to apparently healthy cows. The elevation in total leucocyte count, and differential leukocyte counts were not statistically significant in downer cows compared to control cows. The values of serum calcium and phosphorous were significantly lower than that of control values. Hyperglycemia was noticed in all downer cows. Significantly increased levels of AST, CPK, serum urea nitrogen and creatinine were observed in downer cows when compared to control group. But no significant change was observed in serum magnesium and potassium levels in downer cows when compared to control animals. Echocardiographic examination showed normal cardiac function in downer cows.

Keywords: Downer Cow Syndrome, Haematology, Biochemical changes, Echocardiography.

Downer cow syndrome is characterized by inability of a cow to stand voluntarily from recumbency and is generally associated with hypocalcaemia, hypophosphataemia, hypomagnesaemia, hypokalemia, toxemia, septicemia and muscle and nerve injuries (Andrews et al., 1992). The disease occurs most commonly within the first 2 or 3 days after calving in high producing dairy cows. The etiological diagnosis and institution of prompt therapy of this clinical condition is always in favour of farm economy. So far, no systematic work has been done on this clinical condition in Andhra Pradesh. Keeping in view the above facts the present study was designed with the following objectives. a) To study the haematobiochemical changes in downer cows b) To evaluate the cardiac function of downer cows with echocardiography.

The study was carried out at Teaching Veterinary Clinical Service Complex, College of Veterinary Science, Tirupati. Ten apparently healthy cross bred cows between 4 to 6 years of age were selected as control group for obtaining normal data for comparison of parameters under study. Total 58 cross bred cows of 4 to 8 years of age with history of inability to rise voluntarily for the varied period of time were taken for this study. These clinical cases were either brought to teaching veterinary clinical service complex or attended at private farms in four districts of Rayalaseema region. Detailed history revealed that 5 cows were recumbent for one day, 18 cows were recumbent for two days, 11 for three days and rest of animals for four or more days. Thirty eight of these cases were diagnosed within 30 days postpartum, eleven cases of downer cows were diagnosed after 30 days postpartum and nine were in advanced pregnancy. The downer cows selected for present study were subjected to a detailed clinical examination and the data was recorded in a proforma specially designed for the data collection.

About two milliliters of whole blood was collected from each animal aseptically in vial containing heparin as anticoagulant for the estimation of total erythrocyte count, total leukocyte count, haemoglobin, packed cell volume and peripheral smears were made for differential leucocyte count as per the methods described by Coles (1986). Eight milliliters whole blood was also collected from each animal into centrifuge tube without anticoagulant for separation of serum. Serum concentration of calcium, phosphorus, magnesium, potassium, glucose, urea, nitrogen, creatinine, aspirate aminotranseferase (AST) and creatine phosphokinase (CPK) were estimated by standard methods in semi auto analyzer with span diagnostics commercial kits. Echocardiogram was performed in selected animals using IXOS vet ultrasound machine supplied by Esoate Pie Medical, Netherlands. A micro convex 2.5 mhz probe was used to perform echocardiogram by the method described by Hallowell *et al.* (2007). The statistical analysis of the data was carried out as per Snedecor and Cochran (1989).

In the present study, the age group of cows

affected include 4 years (n=19) and above 6 years (n=39). Clinically the alert downer cows appeared bright, alert, had normal defecation, urination and appetite was slightly decreased. The body temperature, pulse rate, respiratory rate were within normal range in alert downers. Total of 49 cows were in sternal recumbence while 9 cows were in lateral recumbence, non alert and anorectic with markedly accelerated cardiac and respiration rates. All the alert downers tried to stand up repeatedly but could not rise to their feet. They could raise their fore limbs but failed to put up weight on hind limbs. The mean, standard error and significance of haematalogical findings for normal and downer cows are given in table1. A significant decrease in haemoglobin, packed cell volume and total erythrocyte count was observed in downer cows when compared to apparently healthy cows, which might be associated with reduced feed intake. The present findings concur with earlier works (Wadhwa and Prasad, 2002). The elevation in total leucocyte count, and differential leucocyte counts were not statistically significant in downer cows compared to control cows. The mean ± SE values of serum biochemical of profile of control and downer cows are given in table-2. The values of serum calcium and phosphorous were significantly lower than that of control values. Hyperglycemia was noticed in all downer cows, which might be due to high cortisol level. Significantly increased levels of AST, CPK, serum urea nitrogen and creatinine were observed in downer cows when compared to control group. The rise in serum enzymes was the outcome of muscular damage. But no significant change was observed in serum magnesium and potassium levels in downer cows when compared to control animals. These findins are in agreement with report of Wadhwa and Prasad, (2007). Echocardiographic examination showed normal cardiac function in downer cows (Table 3and fig 1).

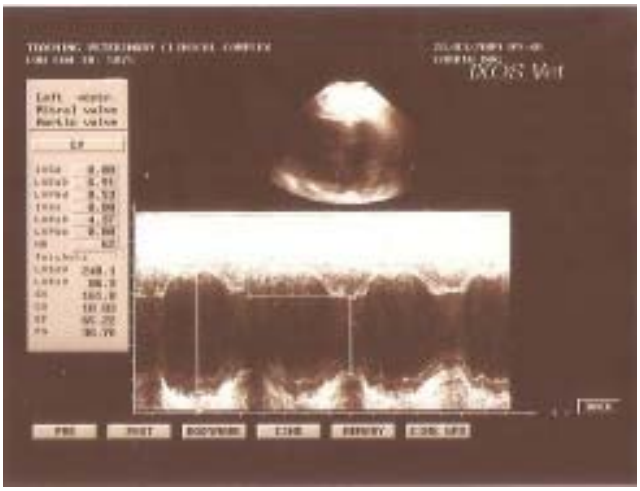

Fig. 1: M-Mode Echocardiogram in a downer cow

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Table 1. Haemotology in control and downer cows (Mean ± SE).

Parameter	Control cows	Downer cows
Haemoglobin (g/dl)	12.80 ± 2.12	8.21 ± 1.13 *
Packed cell volume (%)	39.51 ± 3.21	31.42 ± 2.12 *
Total erythrocyte count (10^6 µl/cmm)	6.32 ± 1.19	4.92 ± 1.21 *
Total leucocyte count(10^3 µl/cmm)	6.72±2.17	6.91±2.17 NS

*Significant (P<0.01) NS- Non significant

Table 2. Serum biochemical profile in control & downer cows (Mean ±SE).

Parameter	Control cows	Downer cows
Calcium (mg/dl)	10.71 ± 0.41	7.44 ± 0.51 *
Phosphorus (mg/dl)	5.47 ± 0.09	3.24 ± 0.29 *
Magnesium (mg/dl)	3.23 ± 0.09	3.32 ± 0.07 NS
Potassium (mg/dl)	4.71 ± 0.33	4.52 ± 0.33 NS
Glucose (mg/dl)	52.23 ± 2.37	72.12 ± 2.14*
Urea nitrogen (mg/dl)	23.12 ± 1.42	34.14 ±0.23 *
Creatinine (mg/dl)	0.7 ±0.12	1.39 ±0.01 *
AST (IU/L)	61.62 ±2.12	207.31 ± 6.21 *
CPK (IU/L)	60.12 ± 3.12	1577.21 ± 7.21 *

NS: non significant * Significant (P<0.01)

Table 3. Echocardiographic measurements in control & downer cows (Mean ± SE)

Parameter	Control cows	Downer cows
LVD_d (cm)	7.40 ± 0.33	7.10 ± 0.21 ^{NS}
LVD_s (cm)	4.30 ± 0.33	4.20 ± 0.34 ^{NS}
IVS_d (cm)	2.10 ± 0.04	2.12 ± 0.05 ^{NS}
IVS_s (cm)	3.40 ± 0.31	3.30 ± 0.2 ^{NS}
FW_d (cm)	1.20 ± 0.31	1.31 ± 0.2 ^{NS}
FW_s (cm)	1.50 ± 0.51	1.48 ± 0.6 ^{NS}
FS (%)	45.70 ± 8.21	44.21 ± 7.9 ^{NS}
EPSS (cm)	0.55 ± 0.42	0.61 ± 0.3 ^{NS}
AO (cm)	6.12 ± 0.07	6.21 ± 0.17 ^{NS}
LAD cm	10.21 ± 0.08	9.91 ± 0.29 ^{NS}

NS: non significant * Significant (P<0.01)

LVD_d (cm)	Left ventricular diameter in diastole
LVD_s (cm)	Left ventricular diameter in systole
IVS_d (cm)	Interventricular septal diameter in diastole
IVS_s (cm)	Interventricular septal diameter in systole
FW_d (cm)	Left ventricular free wall diameter in diastole
FW_s (cm)	Left ventricular free wall diameter in systole
FS (%)	Fractional shortening
EPSS (cm)	E- point to septal separation
AO (cm)	Aortic diameter in diastole
LAD cm	Left atrial diameter in systole

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Serotyping and isolation of Foot and Mouth Disease virus form tongue epithelium using Sandwich ELISA

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Abstract

For serotyping of FMD virus using sandwich ELISA, tongue epithelium were collected from different districts of Uttar Pradesh from 19 cattle and 6 buffaloes. Over all typeability was found 68.00% of which serotype O (58.82%) was the predominant, followed by A (41.17%). Serotype C and Asia-1 could not be recovered from any sample. Virus could be recovered from 8 samples of tongue epithelium using BHK-21 cell line (2 serotype O and 6 serotype A) and overall recovery of virus was 32.00%.

Keywords: Cattle, FMD, Sandwich ELISA, Serotyping.

FMD poses serious limitation on the development of the livestock industry and there are restrictions upon international trade (Lobo *et al.,* 1976). Sandwich ELISA test has been found specific, time saving and made use of freshly harvested FMDV infected BHK-21 fluid as antigen (Pattnaik and Venkataramanan, 1989). Virus isolation remains the ultimate proof of the presence of live FMDV. The virus adopted directly on to BHK-21 cells has a higher infectivity titre than that adopted through primary calf kidney cells. There are isolated reports on the occurrence of this virus from different parts of the country. The present study was, therefore, undertaken to record the serotype involved and isolation of FMD in some districts of Uttar Pradesh.

Total 25 tongue epithelium samples from 19 cattle and 6 buffaloes were collected from different districts of Uttar Pradesh for serotyping and virus isolation (Table 1). Sandwich ELISA test for serotyping of FMD virus serotype, i.e. O, A, C and Asia-1 was performed as per the method described by Bhattacharya *et al*. (1996). The tongue epithelium samples were processed for isolation of serotype O, A, C and Asia-1 on BHK-21 cell line after getting reagents and BHK-21 cell line from Central FMD virus typing Laboratory, I.V.R.I, Mukteshwar. Virus isolation was confirmed and cytopathic effects (CPE) were observed on BHK-21 cell lines (Fig. 1 and 2) which were characterized by rounding of cells and finally detachment of the cells.

Out of 25 tongue epithelium samples collected from cattle and buffaloes, 17 specimens were found positive for the presence of FMDV giving overall typeability per centage as 68.00%. Of these 17 serotyped, 10 were characterized as serotype O and 7 as serotype A (Table 1) while serotype C and Asia-1 were not found. Common occurrence of serotype O in

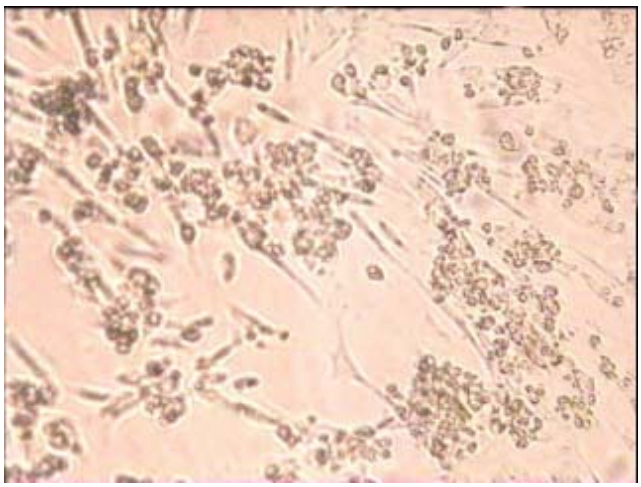

Fig. 1: Showing CPE on BHK-21 cell line on 3rd day's post-infection propagation 40 X

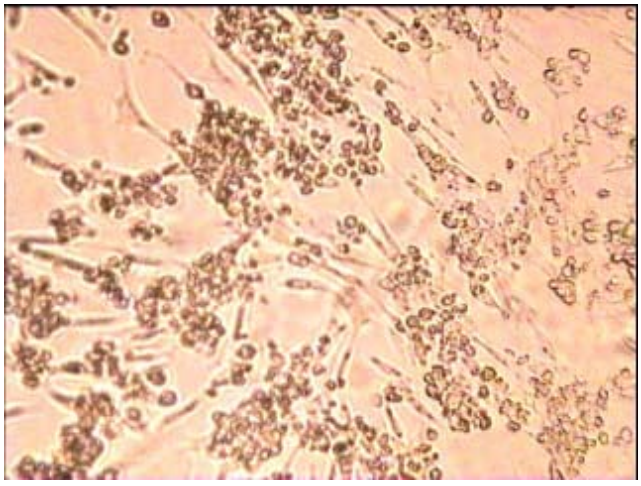

Fig. 2: Showing CPE on BHK-21 cell line on 6th day's post-infection propagation 40 X

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Table 1: Distribution of FMD Virus types.

S. No	Species	Total No. of samples	Virus Recovered	Types of Virus			
				O	A	C	Asia-1
1	Cattle	19	13	8	5	-	-
2	Buffalo	6	4	2	2	-	-
3	**Total**	**25**	**17 (68.00%)**	**10 (58.82%)**	**7 (41.17%)**	**-**	**-**

Table 2: Recovery of virus from field specimens tongue epithelium.

S. No.	Sample No.	Sample	Age of Animal	Districts	CPE on BHK-21	Type of Virus
1	UPl	Tongue Epithelium	5 yr	Meerut	+	A
2	05 PNT/09-10		6 yr	Moradabad	+	O
3	08 PNT/09-10		4 yr	Rampur	+	O
4	10 PNT/09-10		3 yr	Bareilly	+	A
5	14 PNT/09-10		9 yr	Bareilly	+	A
6	15 PNT/09-10		7 yr	Agra	+	A
7	Brl		2.5 yr	Bareilly	+	A
8	23 PNT/09-10		3.5 yr	Bareilly	+	A

cattle and buffaloes is in concurrence with the findings of Prasad *et al*. (1992), Sarma *et al*. (1992), Mannet *et al*. (1998) and Shah *et al.* (2011) in India. Several scientists from different countries have reported that sandwich ELISA is a good test for serotyping of FMDV (Oliver *et al*., 1988; Pattnaik and Venkataramanan, 1989 and Alonoso *et al*., 1992). All the 25 samples of tongue epithelium were processed for virus isolation on BHK-21 cell line and virus could be recovered from 8 samples (2 serotype O and 6 serotype A) (Table 2) with an overall recovery per centage of 32.00%. Possible reasons for poor recovery of virus from tongue epithelium may be either samples were not collected at the proper time of clinical manifestation of disease or were not suitably preserved. The combined use of ELISA with polyvalent antisera and cell culture inoculation was the most effective procedure for identifying FMDV in epithelial samples from field (Alonoso *et al*., 1992).

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Comparative evaluation of various tests for detection of brucellosis in swamp buffalo

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Abstract

Brucellosis was detected in organized and unorganized swamp buffalo herds of Assam with the prevalence rate of 13.89% in A-B ELISA followed by 12.96%, 8.33%, 6.48% and 9.62% respectively in DTHT, STAT, RBPT and MRT. The highest rate was recorded in she buffaloes (17.31%) followed by heifers (11.42%) and bulls (9.52%) in A-B ELISA. Comparative evaluation of all the tests revealed that A-B ELISA detected maximum (17.31%) positive cases of brucellosis, followed by DTHT (15.38%) , STAT (13.46%),RBPT (13.46%) and MRT (9.62%) in she buffaloes.Higher prevalence rate of brucellosis was detected in unorganized herds (15.60%) than organized herds(8.16%)

Keywords: Brucellosis, Delayed type of hypersensitivity test, Prevalence

Brucellosis is the most widespread and economically ravaging reproductive disease of sexually mature animals. In India, 8.7% cattle and 10.2% buffaloes have been found to be reactors (Renukaradhya *et al.,* 2001),Diagnosis of brucellosis based on antibody demonstration is not conclusive. Combination of serodiagnosis and hypersensitivity test could give a comprehensive diagnosis about prevalence of brucellosis in a herd. Present report highlights on the detection of *Brucella* reactive swamp buffalo using both serological and cell mediated immunodiagnostic test.

Milk and serum samples from a total of 108 swamp buffaloes comprising of 52 she buffaloes, 35 heifer and 21 bulls belonging to four different organized and unorganized herd of Assam were investigated for brucellosis. The animals were kept in semi intensive system and were allowed for natural breeding.

For demonstration of antibodies milk (5) and sera (25) samples were collected and same animals were subjected for delayed type of hyper sensitivity test. Detection of brucella specific antibody in serum was done by rapid plate test and serum agglutination test as described by Alton *et al* (1975b).

Colored antigens and brucella plain antigens were procured from IVRI, Izatnagar. Duplicate sera samples were subjected for Avidin-Biotin ELISA (A-B ELISA) using the kit provided by PD-ADMAS, Banglore, India. Further differentiation of infected antibody from that of vaccine antibody was done by 2-Mercaptoethanol Test (2-MET) as described by Alton *et al*. (1975b).

Milk ring test (MRT), Rose Bengal Plate test (RBPT) and Serum Tube Agglutination test (STAT) were performed as per the method described by Alton *et al* (1975b). Delayed type of hyper sensitivity test (DTHT) was done according to the method described by Bercovich *et al* (1989).

The overall prevalence of brucellosis was 13.89%. The highest prevalence rate (17.31%) was recorded in she buffalo (Table-1). In breeding bulls (9.52%) of age group 3.5–5 years and (11.42%) heifers of age group 1.5–3 years were identified as *Brucella* seropositive. Heifers exposed to brucella organism or acquired congenitally in utero from the infected dams may develop self-limiting infection or remain as latent carriers (Morgan *et al*.,1971) Serological examination alone therefore will not eliminate the infection in heifers and combination of tests was necessary to detect reactors.

It was observed that all the affected animals gave a STAT titre (ranging from 1: 40 – 1: 320) against plain antigen of Brucella *abortus*. The difference might be due to the extent of infection. However, age or sex of the affected animals did not have any effect on the titres (Pati *et al*. 2000*).*

In the present study, prevalence of brucllosis in unorganized herds was found to be higher (15.60%). than organized herds (8.16%) which indicate that management exerts a major influence in the prevalence

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Table 1: Comparative evaluation of MRT, RBPT, STAT, A-B ELISA and DTHT for detection of brucellosis.

Categories of animals	No. of animals tested	No. of samples/animals positive										
		MRT		RBPT		STAT		A-B ELISA		DTHT		Statistical significance
		No	%	No	%	No	%	No	%	No	%	
She buffalo	52	5	9.62	7	13.46	7	13.46	9	17.31	8	15.38	NS
Heifer	35	—	—	0	0	1	2.85	4	11.42	4	11.42	NS
Bull	21	—	—	0	0	1	4.76	2	9.52	2	9.52	NS
Total	**108**	**5**	**9.62**	**7**	**6.48**	**9**	**8.33**	**15**	**13.89**	**14**	**12.96**	

NS = Non significant

of the disease. As majority of the buffalo herds of Assam are unorganized. Poor management, ill housing system and sanitary condition or over crowded population of the unorganized herds may influence spread and maintenance of the disease. Chakraborty (1988) has also reported higher incidence of brucellosis in unorganized herds than organized herds.

In the present study it was observed that DTHT could detect higher per centage of brucellosis (12.96%) than the other three tests (MRT, RBPT and STAT) for which DTHT could effectively be used in field condition as rapid screening test. Comparative evaluation of all tests revealed that A-B ELISA detected maximum (17.31%) positive cases of brucellosis, followed by DTHT (15.38%), STAT (13.46%), RBPT (13.46%) and MRT (9.62%) in she buffaloes. Therefore, ELISA had been advocated as it was less complexes, more time saving, sensitive procedure and it overcomes many of the short comings often encountered in other assays like RBPT or STAT (Mahato *et al*., 2004).

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Serum protein level in primary hepatic disorders in dogs

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Abstract

Biochemical parameters such as liver enzymes and bile acids are often poor diagnostic tool in evaluating liver diseases in dogs. Thus severely diseased dogs with late stage cirrhosis and a poor prognosis may have almost normal serum activities of liver enzymes and only moderately increased serum concentration of serum bile acids where as dogs with acute hepatitis and chronic progressive hepatitis and with a good prognosis may have greatly increased levels. The present study was performed to find out the importance of protein estimation (qualitative/quantitative) as a diagnostic and prognostic tool in evaluating liver diseases in dogs. Significantly lower values of total protein, albumin & globulins have been observed in cases of intrahepatic portosystemic shunt than that of hepatitis and cirrhosis in dogs. In SDS – PAGE bands of albumin and prealbumin were less thick than that of healthy dogs while bands of α_1 antitrypsin and β-globulins and C reactive proteins were found to be increased in cases of hepatitis which may be due to proteins of acute inflammation.

Keywords: protein, liver, dog, electrophoresis, albumin, SDS- PAGE.

More than 200 proteins have been identified in the serum of man and animals. Of these albumin is most abundant and synthesized exclusively in the liver. The liver also is the site of synthesis of all blood coagulation proteins except factor VIII. It has long been recognized that changes in a selected group of constitutive plasma proteins occur during disease process. Liver being the site of synthesis of many serum/plasma proteins, a change is logically expected in their concentration in hepatic diseases. The present investigation was, therefore, undertaken to study the changes in protein profile of dogs with primary liver diseases.

Six healthy dogs and 82 dogs with primary hepatic diseases (hepatitis/hepatosis 47; cirrhosis/ fibrosis 21; and intrahepatic portosystemic shunt 14), diagnosed on clinical, enzymological, biochemical, urological and ultrasonographic investigations, were included in the present study. Dogs with ascites were subjected to electrocardiographic evaluation and only those not having cardiac involvement were included in this study. Serum total proteins and albumin (biurate method) were estimated colorimetrically using kits. Electrophoretic separation of serum proteins was done by SDS –PAGE as per method deseribed by Laemmli (1970) using vertical mimigel electrophoretic apparatus (Biorad USA). The proteins in the serum were subjected to electro phoretic separation in 10% separating and 5% stacking gel.

Among hepatic diseases, the mean serum total proteins level was significantly (P< 0.01) lower (3.04-0.64 g%) in intrahepatic portosystemic shunt (intrahepatic PSS) as compared to total serum proteins levels of dogs with hepatitis (5.03±0.27 g%) or cirrhosis (4.49±0.42 g%). Similarly albumin levels were also lower (1.06±0.21 g%) in intrahepatic portosystemic shunts (Table 1).The value of serum globulin was significantly higher in cases of hepatitis (3.24±0.23 g%) than that of intrahepatic portosystemic shunt. A/G ratio in hepatitis, cirrhosis and intraherpatic PSS were 0.79 ±0.11, 082±0.14 and 0.54±0.43, respectively. Marked decrease in A/ G ratio in cases of intrahepatic PSS appears to be due to hypoalbuminemia, an important feature of chronic liver diseases (Kaneko, 1999). Decrease in serum albumin values has also been observed by other workers in hepatopathies (Nalini Kumari *et al.,* 1998), intrahepatic PSS (Johnson *et al.*, 1987; Rutgers, 1993) and also in cirrhosis (Lucena *et al.,* 2001; Thornburg *et al.*, 1983). Decrease in serum albumin values in cases of hepatic cirrhosis, is also well established. Low level of albumin as observed in the present study was also observed by Barrett *et a*l., (1976) in three out of four cases of intrahepatic PSS. It appears that lower value of albumin in hepatic diseases, may reflect either increased volume of distribution than impaired hepatic synthesis as observed in humans with cirrhosis having ascites or dilutional hypoalbuminemia owing to retention of sodium and water in cirrhosis or leaking of albumin directly from hepatic lymph into

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Table 1. Serum protein fratcionization of dogs with hepatic diseases (Mean ±SE)

Groups	Total Protein ** (g%)	Albumin ** (g%)	Globulin** (g%)	A/G ratio
Hepatitis (n=47)	5.03±0.27[a] (1.99-7.5)	1.8±0.10[a] (0.46-3.04)	3.24±0.23[a] (0.56-6.04)	0.79±0.11 (0.15-3.75)
Cirrhosis (n=21)	4.49±0.42[a] (2.4-9.28)	1.70±0.11[a] (0.88-2.4)	2.83±0.34[a] (0.64-6.96)	0.82±0.14 (0.3-2.75)
Intrahepatic PSS(n=14)	3.04±0.64[b] (0.30-6.68)	1.06±0.21[b] (0.08-2.13)	1.97±0.42[b] (0.21-4.58)	0.53±0.03 (0.27-0.71)

Different superscripts differ significantly, ** ($p<0.01$) Values in parenthesis are range

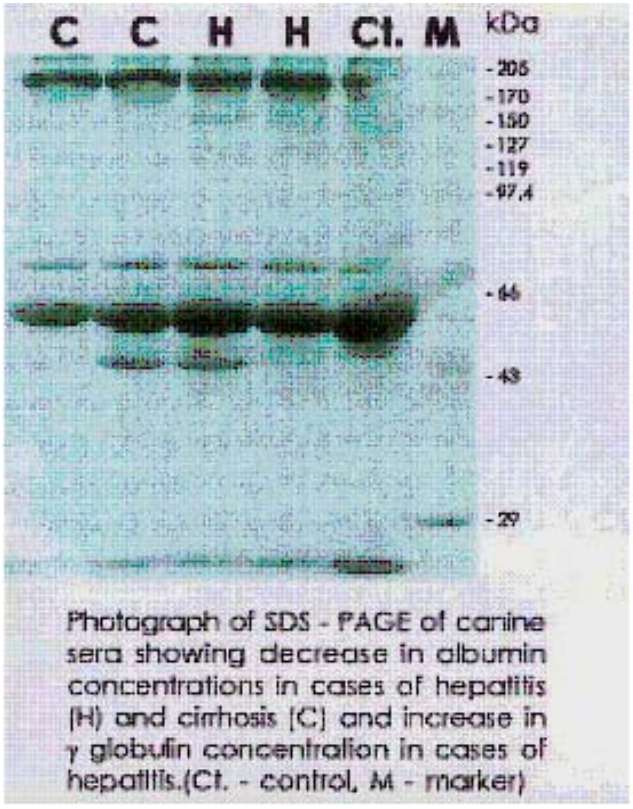

Fig 1.

ascites (Center, 1994) or decreased nutrient uptake (Kindmark and Laurell, 1972; Skrede et al. 1975; Hiramatsu, 1976; Chio and Oon, 1979). Comparatively higher values of globulins in hepatitis can be explained on the basis of increased systemic immune response to foreign antigens due to reduced kupffer cell number and function in hepatic damage (Center, 1994). Mostly changes in plasma proteins appear late in liver damage (Jain, 1986).

Comparative less thickness of bands of albumin (59,000 dalton) and pre-albumin (54,000 dalton) in SDS- PAGE in hepatitis and cirrhosis indicated a decrease in their concentration (Fig 1). Hypoalbminemia in these chronic liver diseases could be ascribed to switch in production of proteins by liver towards the increased synthesis of acute phase proteins and concomitant decrease in the synthesis of albumin (Eckersall and Conner, 1988; Koj *et al.*, 1988) or due to impaired liver function. In human also, hypoalbuminemia has been reported in acute toxic hepatitis, chronic active hepatitis, cryptogenic cirrhosis and hepatic tumors (Skrede et al., 1975). In the present investigation, decrease in albumin concentration in cirrhosis was more marked. Increase in α_1 antitrypsin (a protein of 45,000 dalton) in hepatitis and cirrhosis agrees with similar increase in most type of liver diseases in man (Skrade *et al.*, 1975). The erraticness of α_1 antitirypsin may be due to stage of hepatic diseases, as absence of this protein has been reported in late stage of cirrhosis (Sevelius and Anderson, 1995). A band of 150,000 dalton in cases of hepatitis in dogs, indicated increase in γ-globulins in hepatitis may be due to auto-antibody production in response to release of antigen from liver (Anderson and Sevelius, 1992)

Appearance of a band of 140,000 dalton in hepatitis was possibly due to development of C-reactive protein, an acute phase positive protein that increases in inflammatory diseases (Kaneko, 1999). An increased concentration of C3 (another acute phase protein) as compared to that of cirrhosis could be ascribed to acute inflammatory process. It appears that the serum protein profile revealed by electrophoresis can be valuable complement to other diagnostics aid in telling about the stage of disease.

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Effect of lactational status on trace element profile of Vrindavani Cattle

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Abstract

Study group comprised of thirty five Vrindavani cattle and divided into five equal groups (n=7) in different lactations for evaluating status of Copper (Cu), Iron (Fe), and Zinc (Zn). Cattle in first and second lactation were placed in group I, cattle in third and fourth lactation in group II, cattle in fifth and sixth lactation in group III, cattle in seventh and eighth lactation in group IV and dry cattle were assigned to group V. Significant difference was observed with respect to blood copper levels only in group IV and V cattle, for blood iron levels in group II and V, while no significant difference was observed among different groups for blood zinc concentrations.

Keywords: Copper, Iron, Lactational status, Vrindavani cattle, Zinc.

Minerals occupy an important place in animal nutrition for production as well as health (McDonald *et al*., 2009). Their normal concentrations mainly depend on dietary supplementation, absorption, presence of other minerals, homeostatic control mechanism of the body, state of production and the species of animal involved. Vrindavani is synthetic cross-bred cattle strain which carries 50–75% inheritance from exotic temperate cattle breeds (Holstein-Friesian, Jersey and Brown Swiss) and 25-50% inheritance from the native indigenous Hariana breed, has been developed by Indian Veterinary Research Institute, Izatnagar. The milk yield per lactation (305 days) is up to 3000 liters with high fat per centage (4-4.5%).

Trace elements play major role in host resistance to infections including intramammary infections as well as in productivity of dairy animals. They are of utmost significance in both production as well as health of cattle. Any deficiency of these micro-minerals in diet of animals is responsible for reduced milk yield, increased susceptibility to diseases as well as reproductive disorders like infertility, anestrous etc (Sharma *et al*., 2003a; Kumar *et al*., 2004a).Therefore the study was performed with objective to evaluate blood trace mineral concentration in Vrindavani cattle and to study the effect of lactational status on trace minerals viz., Cu, Fe, and Zn.

The study was carried out at Cattle and Buffalo (C&B) farm of LPM section, IVRI, Izatnagar during the month of April to May 2010. Selection of cattle was made randomly from the farm herd of Vrindavani cattle kept and maintained on similar feeding and management practices. These were further divided into five groups, according to their lactation numbers. Seven cattle were assigned in each group (n=7), with the following categorization: Group I (lactation no. 1-2) Group II (lactation no. 3-4), Group III (lactation no. 5-6), Group IV (lactation no. 7-8) and Group V (dry cattle).

Blood samples (10 ml) were collected using sterile disposable syringe during early morning hours by jugular venipuncture. The blood samples were transferred to sterilized acid washed test tubes containing EDTA as anticoagulant and immediately transported to laboratory on ice pack.

Blood concentrations of Cu, Fe, and Zn were determined after nitric acid/perchloric acid wet digestion as per Kolmer *et al*., (1951) and analyzed by atomic absorption spectrophotometry.

Differences in blood concentration of Cu, Fe, and Zn among different groups were compared using one way analysis of variance (ANOVA) to determine the level of significance between groups (Snedecor and Cochran, 1994).

Values of mean blood concentration of Cu, Fe, and Zn in Vrindavani cattle in different lactations are given in Table 1. Mean blood concentration of Cu varied from 0.62±0.05 to 1.08±0.06 µg/ml among different lactational groups. Highest blood Cu concentration was observed in group V cattle while lowest values were recorded in group III cattle, followed by group II. The present findings are attributed to the fact that Vrindavani cattle might reach their maximal performance level in terms of milk yield in their V-VI lactation. The lower

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Table 1: Concentration (mg/ml) of Copper (Cu), Iron (Fe) and Zinc (Zn) in blood samples of cattle (Mean±SE).

Group	Cu	Fe	Zn
I	0.89±0.08^{ab}	74.78±5.83^{abc}	1.98±0.12
II	0.80±0.07^{bc}	62.35±4.17^{c}	2.54±0.21
III	0.84±0.07^{bc}	77.85±3.04^{ab}	2.51±0.45
IV	0.62±0.05^{c}	65.69±7.78^{bc}	1.84±0.14
V	1.08±0.06^{a}	89.28±1.18^{a}	2.59±0.32

Values with different superscripts differ significantly (P<0.05)

blood Cu concentration in group III and IV is attributed to mobilization of Cu in milk, as only little placental transfer of Cu occurs during pregnancy and calf is dependent on colostrum as an initial source of copper (Ullrey *et al*., 1977). The present findings corroborates with the findings of other reports published (Kumar, 2006; Smart *et al*., 1981). Akhtar *et al*. (2009) also observed increased serum Cu levels in dry Nili-Ravi buffaloes during advanced pregnancy while significant decrease in Cu concentrations during lactation.

Blood Fe levels varied from 62.35±4.17 to 89.28±1.18 µg/ml in cattle in different lactations. Significantly (P< 0.05) lower values of blood Fe were recorded in lactating cattle of groups II and III compared to dry cattle of group V. Blood Fe concentrations in remaining two lactating cattle groups (group I and IV) were comparable and non-significantly different, though lower than dry cattle of group V. Kumar, (2006) reported higher iron concentration in dry cattle and buffaloes compared to lactating cattle and buffaloes from different districts of Bihar. Deficiency of Fe is normally not common in ruminants as they frequently get access to the Fe source through water, soil or feeds high in Fe concentration except in young animals or milk fed calves (Kincaid, 1999).

Average blood Zn concentration non-significantly (P<0.05) varied from 1.84±0.14 to 2.59±0.32 µg/ml in various lactational groups of cattle. Randhawa *et al*. (2009) reported no significant difference in plasma Zn concentrations in dairy animals in different age groups of both organized and rural dairy units of Punjab. Our findings regarding blood Zn are in accordance with those reported by Randhawa *et al*. (2009).

Study concluded that significant difference was observed regarding blood concentration of Cu and Fe among some of the lactational groups with higher concentrations in dry cattle compared cattle in lactating groups. Blood Zn concentrations revealed no significant difference among different lactational groups of Vrindavani cattle.

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Clinicobiochemical and ultrasonographic investigation of acute prostatitis in dogs

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Abstract

Acute prostatitis was diagnosed in five cases. Transrectal digital palpation, ultrasonography, prostatic fluid cytology and clinico biochemical parameters were studied in detail. The mean prostatic length, depth, width and volume in the present study were 5.77 cm, 4.66 cm, 4.95 cm and 68.76 cm^3 respectively. The per centage of band forms was found to be elevated in all the cases. Treatment was given depending on the culture and sensitivity result of prostatic fluid.

Keywords: Prostate, Cytology, *E. coli*, Urethral discharge.

Krawiec and Heflin (1992) and Dorfman *et al.*(1995) opined that dogs become predisposed to prostatitis by increased number of bacterial organisms in the periprostatic urethra, compromise of local immunity, disease of urinary tract, altered prostatic tissue and fluid flow as in the cases of benign prostatic hyperplasia (BPH) and cysts. Infection was mostly of ascending nature from the urethral flora, along with hematogenous spread or extension from testes, epididymis or peritoneal cavity. *Escherichia coli* was the most common bacterial organism identified in dogs with bacterial prostatitis, followed by *Staphylococcus aureus*, *Klebsiella* spp., *Proteus mirabilis*, *Mycoplasma canis*, *Pseudomonas aeruginosa*, *Enterobacter* spp., *Streptococcus* spp., *Pasteurella* spp., and *Haemophilus* spp.(Ling *et al.,* 1983; Barsanti and Finco, 1986; Johnston *et al*., 2000).

Materials and Methods

Dogs presented to the Veterinary College Hospital, Mannuthy and Kokkalai with the clinical signs suggestive of prostatic diseases such as tenesmus, dyschezia, dysuria, arching of back and rear limb weakness formed the materials for the present study.

Clinical observations, breed, age and previous history were recorded. Based on ultrasonography, transrectal palpation, hemato – biochemical and prostatic fluid cytology studies, five dogs were found to be associated with acute prostatitis.

Whole blood, serum and urine samples were collected on day 0 for estimation of haematocrit, haemoglobin, erythrocyte sedimentation rate (ESR), total erythrocyte count (TEC), total leucocyte count (TLC), differential leucocyte count (DLC) platelet count, total serum protein, albumin, globulin, A: G ratio, serum creatinine, blood urea nitrogen (BUN), calcium, glucose, testosterone, total acid phosphatase, prostatic acid phosphatase, urinary pH, detection of blood pigment, specific gravity and presence of protein. The serum samples from all the dogs were subjected to Brucella antibody test.

During ultrasonography, changes in echogenecity and prostatic volume were recorded. Prostatic volume was calculated using the formula, Volume (cm^3) = $L \times W \times H \times 0.523$. The length (L) is defined as the maximum diameter of gland along the urethral axis in sagittal image, height (H) as the maximum diameter perpendicular to the axis of length. On transverse images, the height is defined as the diameter of the prostate on a line separating the two lobes of the gland and the width (W) as the maximum diameter perpendicular to the axis of the height.

Prostatic fluid was collected from all the cases by prostatic massage combined with catheterization and subjected to cytology, culture and sensitivity. Prostatic smears were prepared from the collected prostatic fluid and stained with leishman's stain and observed under 100 x magnification.

Response to the treatment adopted was evaluated based on clinical improvement.

Results and Discussion

Age of the affected dogs ranged from 3.6 – 9 years with a mean of 5.3 year. Dogs presented with prostatitis had symptoms such as anorexia, urethral discharge, dyschezia and hematuria that were also reported by Krawiec and Heflin (1992), Dorfman *et al.*

(1995), Johnston *et al*. (2000) and Smith (2008). Clinical signs such as straining to defecate and constipation may result secondary to displacement and narrowing of large intestine due to prostatomegaly as opined by Hoffer *et al*. (1977). Haematuria might be due to irritation of the bladder by ammonia released from retained urine (Kiren, 2008). Urine retention or incontinence or dysuria may occur from impingement upon the bladder and urethra as reported by Hoffer *et al*. (1977).

Fever (103.4^0 F) was observed in dogs with prostatitis in the present study. The same observation was also made by Krawiec and Heflin (1992), Dorfman *et al*.(1995). Three dogs with prostatitis had caudal abdominal pain, which agrees with the findings of Smith (2008). The warmness and pain on palpation of the prostate gland of all the dogs indicated prostatitis. The variation in position of the prostate gland might be due to variation in degree of prostatomegaly with regard to inflammation. These observations were similar to the report of Davidson (2003) and Smith (2008).

Ultrasonographic examination revealed diffused increase in echogenisity of prostatic parenchyma and 40% of dogs had multifocal hyperechoic areas in prostatic parenchyma. The mean prostatic length, depth, width and volume in the present study were 5.77 cm, 4.66 cm, 4.95 cm and 68.76 cm^3 respectively. Duque *et al.* (2009) also observed diffused increase in prostatic echogenisity and enlarged prostate with 5.28 cm length and 4.9 cm width in prostatitis. The occurrence of multi focal hyperechoic areas in 40% dogs in the present study might be due to more severe localized infection. Hyperechoic particles in urinary bladder of three dogs might be due to spread of inflammation from prostate. In the absence of micturition or ejaculation, urethral pressure moved the prostatic fluid cranially into the bladder by prostatic fluid reflux as opined by Romagnoli (2007). Small kidneys with indistinct corticomedullary junction might be due to ascending grade of infection from prostate (Jayathangaraj *et al*., 1993).

Table 1: Concentration (µg/ml) of Copper (Cu), Iron (Fe), and Zinc (Zn) in blood samples of cattle (Mean±SE).

Groups	Cu* (µg/ml) Cu	Fe (µg/ml) Fe	Zn* (µg/ml) Zn
I	0.89±0.08^{ab}	74.78±5.83^{abc}	1.98±0.12
II	0.80±0.07^{bc}	62.35±4.17^{c}	2.54±0.21
III	0.84±0.07^{bc}	77.85±3.04^{ab}	2.51±0.45
IV	0.62±0.05^{c}	65.69±7.78^{bc}	1.84±0.14
V	1.08±0.06^{a}	89.28±1.18^{a}	2.59±0.32

Values with different superscripts differ significantly (* P< 0.05)

Prostatic fluid cytology of all the cases revealed large number of neutrophils. Bacteria-laden neutrophils in prostatic fluid could be observed in 40% of the cases. Ling *et al.* (1983) opined that bacteria-laden white blood cells in canine prostatic fluid were indicative of active infection. Cultural examination of prostatic fluid from the five dogs revealed *E. coli.* Ling *et al.* (1983), Barsanti and Finco (1986), Johnston *et al*. (2000) and Dhanya (2004) reported that *E. coli* was the most common bacterial organism identified in dogs with bacterial prostatitis.

Mild anemia with hemoglobin of 11.08 g%, RBC of 4.95 million /cu.mm and PCV of 30.14%, leucocytosis (14880/cu.mm) and neutrophilia (83%) with left shift (2%) observed in the present study is in agreement with the findings of Johnston *et al.* (2000), Davidson (2003), Smith (2008) and Duque *et al*. (2009). The mean platelet count was within normal range. There is no report regarding the platelet count in dogs with prostatitis. Normal total protein (6.8 g/dl), mild hypoalbuminemia (2.16g/dl) and hyperglobulinemia (4.64g/dl) were observed in the present study. There is paucity of literature regarding this. The elevation of mean creatinine (4.42 mg/dl) and BUN (39.6 mg/dl) value was observed in the present study which has also been reported by Jayathangaraj *et al.* (1993) and Duque *et al.* (2009). Most of the dogs with prostatic infection may also have bladder infection and renal failure (Barsanti *et al*. 1983) and this was confirmed with the present case by ultrasonogram of bladder and increased serum creatinine. The mild hypoglycaemia and hypocalcaemia observed in the study may be due to anorexia associated with prostatitis.

Barsanti and Finco (1979) suggested that most of the prostatic infections were secondary to migration of bacteria from the urethra although spread through blood, semen and rectal flora was also possible.

The level of serum total acid phosphatase (6.32 U/L) and prostatic acid phosphatase (2.9 U/L) were elevated in dogs with prostatitis. Elevated human serum Prostatic acid phosphatase (PAP) concentrations were present in benign prostatic hypertrophy, prostatitis, following urethral catheterization and prostatic massage as suggested by Wadstrom *et al.* (1984) in prostatic adenocarcinoma (Babaian and Orlando, 1986) and

gastrointestinal tract tumors (Sorbin *et al.,* 1986). Prostatic acid phosphatase is androgen dependent and its increase in serum was probably due to degeneration of prostatic secretory cells induced by increased dihydrotestosterone concentration within the gland (Corazza *et al*.,1994). No correlation was found between serum testosterone level and prostatitis as the value was within normal range. Lowseth *et al.* (1990) estimated serum testosterone in healthy dog as 92-2550 pg/ml.

The serum samples from all the dogs were negative for Brucella antibody test (Fig. 4). *Brucella canis, Brucella suis* and *Brucella abortus* are all capable of causing prostatitis in dogs as opined by Barr *et al*. (1986). Haematuria and proteinuria were observed in two cases. Similar observations were also made by Davidson (2003), Smith (2008) and Duque *et al.* (2009). Prostate gland is in close proximity to the microflora of distal urethra. This may be the reason for prostatitis being mostly coexistent with nephritis and cystitis as observed in the present study.

Culture of urine sample of two dogs revealed *E. coli*. This may be due to possible contamination of the fluid from the urinary bladder or urethra, while in the rest of the two cases, urine was found sterile, and the prostatic fluid revealed a pathogen. This evidence emphasizes the fact that the isolated pathogen originates certainly from the prostate gland.

Three animals responded successfully to the treatment with enrofloxacin, which is in agreement with Duque *et al.* (2009). Enrofloxacin could easily diffuse through blood- prostate barrier and achieve therapeutic concentration in prostatic fluid. The case with renal failure, which developed secondary to prostatitis, was effectively treated with enrofloxacin, fluid and anti ulcer therapy. The serum creatinine value of 5.6 mg/dl was reduced to 2.4 mg/dl with the above treatment , whereas one case with highly elevated creatinine 12.4 mg/dl did not respond to the treatment and succumbed, this might be due to advanced renal failure.

The unsatisfactory response showed by the other dog might be due to mild benign prostatic hyperplasia which was masked by inflammation.

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Haematobiochemical profile of dogs with hepatic and splenic affections

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Abstract

The present study was conducted on 32 dogs suffering from various hepatic and splenic affections to evaluate various haemato-biochemical changes. The blood and serum samples were estimated for Hb, TLC, DLC, platelet count, liver function tests, kidney function tests, total protein and albumin. The diagnosis was confirmed by clinical, radiographic, ultrasonographic, Ultrasound guided biopsy (USGB)/ Ultrasound guided fine needle aspiration biopsy (USG-FNAB) findings. Hepatic affections were characterized by marked elevated SGPT and AKP values along with mild to moderate anemia and neutrophilic leucocytosis. Dogs with non neoplastic splenic affections had moderate elevation in the TLC, SGPT, SGOT and AKP values. Severe anemias along with marked elevated AKP values were recorded in neoplastic conditions of spleen.

Keywords: Dogs, Haematology, Radiography, Ultrasound guided biopsy, Ultrasound guided fine needle aspiration biopsy.

Radiography and ultrasonography are the major tools for confirming the liver and spleen disease. However, haemato-biochemistry is also considered as an important preliminary tool for proceeding towards the correct diagnosis and treatment protocol (Meyer, 2005). The present study was undertaken to evaluate the haemato-biochemical profile of dogs in confirmed cases of hepatic and splenic disease.

Materials and Methods

The present study was conducted on 32 cases of dogs of either sex, aged 4 months to 12 years and body weight ranging from 2 to 40 kg presented for the affections of liver and spleen. Various hematological and biochemical parameters were determined. Radiography and ultrasonography was performed in all the animals. Confirmatory diagnosis was made by ultrasound guided biopsy (USGB)/ultrasound guided fine needle aspiration biopsy (USG-FNAB). The haemato-biochemical parameters were correlated with the histopathological/cytological findings.

Results and Discussion

Out of the 32 animals, 21 were confirmed to have liver involvement and 11 had splenic involvement based on USGB/USG-FNAB findings. Radiography revealed hepatomegaly in most of the animals with liver involvement. Ultrasonographically, the liver was hypoechoic in general with hyperechoic areas in all the cases of hepatosis. In animals with hepatitis, hyperechoic hepatic parenchyma was observed in comparison to splenic echotexture.

In hepatosis (N=8) clinical signs ranged from vomiting, abdominal distension, reduced feed intake and black colored loose faeces. The hematological values revealed Hb-8.10±1.54 g/dL, TLC-48.19± 17.65 $X10^3$/µL, DLC (N-88.00±1.89%, L-9.75±1.79%, E-2.25± 0.96%). The anemia in these cases may due to increased degradation of RBC in most hepatobiliary disease (Bush, 2002). Hepatosis is considered to be non-inflammatory and is characterized by degenerative changes in hepatocytes without inflammatory cells. The blood SGPT (326.75±124.98 µ/L), SGOT (148.75 ± 24.70µ/L and AKP (615.50±164.45µ/L) values were elevated in all cases. The BUN, creatinine, total protein and albumin were within the normal range. Total platelet count was adequate in all the animals except in one case where it was found to be inadequate. The increased level of SGOT may also be associated with leakage following altered membrane permeability (Dial, 1995). However, the elevated values of SGOT along with SGPT in the present study indicated liver damage and SGOT is considered important if elevated along with SGPT (Meyer, 2005).

Hepatitis was diagnosed in 13 animals on the basis of histopathological/cytological findings of the biopsy samples collected under ultrasound guidance. All the animals were dull and depressed, anorectic for 15-20 days, had vomiting, diarrhoea, abdominal distension, black loose faeces, and yellowish urine. The hematological values revealed Hb- 7.32±0.63 g/dL, TLC-36.61± 8.82 $X10^3$/µL, DLC (N-90.77±1.32%, L-6.92±1.41%, M-0.46±0.27%, E-1.85± 0.79%) in case of hepatitis. Total platelet count was adequate in most

of the cases.

The animals were usually anemic had neutrophilic leucocytosis. The possible cause of degradation may be the increased transit time of erythrocytes through the spleen due to reduced portal blood flow and/or increased fragility of red blood cells due to high levels of bile acids (Rothuizen and Meyer, 2000). The neutrophilic leucocytosis is characteristic of acute inflammatory conditions (Johnson, 2000; Bush, 2002 and Verma, 2005) The blood SGPT (279.15±52.80 µ/L), SGOT (182.31±42.13 µ/L) and AKP (556.38±93.42 µ/L) values were elevated in all cases which could be attributed to hepatic cell damage or biliary obstruction associated with hepatic inflammation (Bush, 2002).The BUN, Creatinine, total protein and albumin were within the normal range in all except in one animal where BUN (218 mg/dL) and creatinine (16 mg/dL) levels were very high which could be the cause of increase in the mean values of BUN and creatinine in this group. The AKP level was markedly elevated (1210 µ/L) in one case. The increase in SGPT and AKP value can be attributed to hepatocellular injury caused by various stimuli or factors. The SGPT is a liver specific cytosolic enzyme in dogs (Kramer and Hoffman, 1997).

Among the splenic affections (n=11) neoplastic conditions were found in two cases and nine cases were found to be non-neoplastic based on USGB/ USG-FNAB findings. In the two animals with neoplastic splenic diseases the survey radiographs revealed a round radiodense mass at mid ventral abdomen which was indicative of change in size, shape and surface contour Ultrasonographic examination revealed extremely enlarged, hypoechoic and congested spleen; filling almost half of the abdomen. The cytological examination revealed lymphoma in first case and haemangiosarcoma in the second case. In animal which was diagnosed with lymphoma, marked lymphocytic leucocytosis (TLC-19.68 $X10^3$/µL, L-88%, N-12%) was observed while neutrophilic leucocytosis (TLC- 18.55 $X10^3$/µL, N-90% L-08%, E-02%) was recorded in the case diagnosed as haemangiosarcoma. Total platelet count was moderately increased in both animals. The blood SGOT (201.50±38.50 µ/L), SGPT (218.50 ± 8.50 µ/L) and AKP (1032.00±317.00 µ/L) values were markedly elevated in both cases. The value of AKP was 8-10 times more than the upper limit of the normal range and such increases are usually indicative of the neoplastic conditions in dogs (Bush, 2002). The BUN, Creatinine, total protein and albumin levels were within the normal range.

The diseased spleen was found to be non neoplastic in nine animals based on USGB/ USG-FNAB findings (Haematopoiesis in four cases and suppurative splenitis in rest of the cases). The ultrasonographic findings showed gross enlargement of spleen with uniform to hyperechoic echotexture in most of the cases. The haematological values revealed Hb-7.89± 1.19 g/dL, TLC-46.21±18.89$X10^3$/µL, DLC (N-87.11± 2.86%, L-10.00± 2.43%, M- 0.22 ± 0.22% and E-2.44± 1.09%). The blood SGPT (189.89± 73.59 µ/L), SGOT (136.11± 29.05 µ/L) and AKP (535.56±219.28 µ/L) values were moderately elevated. The BUN, Creatinine, total protein and albumin levels were within the normal range. Usually the splenic diseases were difficult to diagnose because the physical signs and haematobiochemical changes could often be variable and nonspecific (Bhadwal, 1997).

It was concluded that hepatic affections were characterized by marked elevation of SGPT and AKP values along with mild to moderate anemia and neutrophilic leucocytosis. Dogs with non neoplastic splenic affections had moderate elevation in the TLC, SGPT, SGOT and AKP values. Severe anemias along with marked elevated AKP values were recorded in neoplastic conditions of spleen.

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Clinical Article

Management of PPR outbreak in an organized goat farm

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Abstract

During an outbreak of PPR at a goat farm, total of 103 animals were affected and one died. Post mortem examination of dead goat revealed lesions suggestive of PPR. The disease was was confirmed by heparinized blood collected from live animals & lymphnodes from dead animals. The flock was treated with antiviral tablets Acyclovir and antiviral spray TH4.

Keywords: Goat, PPR, treatment.

Peste des petits ruminants (PPR) is a highly infectious and often fatal viral disease of sheep and goats (Ramachandran *et al.,* 1992). Its incidence is increasing at the organized goat farms. The mortality (90%) and morbidity (50%) are high in the recent past years (Rodostitis *et al*., 2007). This paper evaluates the efficiency of antiviral spray and tablets in the management of affected flock.

Total 103 goats of various age groups were transported from north India to the goat farm in Tamil Nadu. The flock consisted of breeds like Sirohi, Beetel, etc. After a week of arrival, the animals showed clinical signs like nasal discharge, reduction in feed intake, gastroenteritis and one animal aborted and died. Lymphnodes from the dead goat and heparinized blood samples from infected animals were sent to Madras Veterinary College, Chennai, for the confirmation of the disease. On postmortem, mucus membranes were found severely congested, intestinal and uterine mucosa highly inflamed with two aborted full grown foetuses. Lungs were congested with pneumonic changes.

The goats were confirmed to be suffering from PPR based on laboratory report from Madras Veterinary College. They were treated with ACIVIR[1] @10 mg/ kg.b.wt for 5 days, sulphamethoxazole trimethoprim combination[2] @ 10 mg /kg b.wt once daily and Meloxicam[3] @ 5 mg / kg b.wt daily for 5 days. TH_4 [4] sprayed on the animals and inside the animal sheds. As supportive therapy Dextrose normal saline and B complex were also given in standard doses.

Migratory flocks of nomads have been thought to be a source of infection of PPR in new areas (Dhand *et al.,* 2002). In this study Acyclovir was found to be effective as antiviral therapy. The TH_4 spray helped in reduction of viral load inside the sheds. Secondary bacterial infection was controlled by sulphamethaxazole-trimethoprim therapy.

The present study found that usage of antiviral tablets (ACIVIR) and antiviral spray (TH_4) were helping in control of PPR. The mortality was restricted to 2% and morbidity was also gone down to zero.

1.Marketed as Acivir by M/S Cipla pharmaceuticals. Mumbai.
2.Marketed as Robatran by M/S TTK helath Care Limited, Chennai
3.Marketed as Melonex by M/S Intas pharmaceuticals, Ahemadabad
4.Marketed as TH_4 contains glutaraldehyde and quarternaryammonium compound by M/S Laboratory Sogeval laboratories, France.

Summary

103 goats were affected with PPR and all but one was recovered by using ACIVIR and TH_4 spray on the animal. The mortality and morbidity were forbidden.

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Diagnosis and management of multiple liver cysts using USG in a dog

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Abstract

A non- descript male dog of 14 yr age had history of anorexia, reluctance in movement and occasional vomiting since last 15 days. Ultrasonographic, radiographic and cytological examinations of aspirated fluid suggested multiple cysts in liver and around the kidney. There was severe leukocytosis and neutrophilia along with gradually increasing blood urea nitrogen and serum creatinine level.The animal showed improvement on next day after ultrasound assisted aspiration (a total 3 sessions) of fluid, but dog died after 6 days.

Keywords: Dog, Liver cysts, Ultrasound

Large cysts may result in abdominal distension, lethargy, vomiting and polydipsia and these cysts can be identified with radiographs or ultrasonography. USG is useful in guiding aspirates and biopsies (Larson, 2007). Liver cysts are fairly common with advancing age and may be serious in cases of polycystic, liver cancer and *Echinococcus* infection. The present paper reports diagnosis and management of multiple cysts using USG assisted aspiration of fluid.

A male non-descript dog of 14 yr age weighing 30 kg was presented with the complaint of anorexia, increased temperature (103^{0}F), reluctance in movement and occasional vomiting since last 15 days. The animal had distension of upper abdomen and pain. The faeces was scanty but urination was normal. The animal was severely depressed but Hb, TEC, AST, ALT and bilirubin levels were within normal range on day 1, 4 and 8. Urea and creatinine values were increased progressively. Haematological examination revealed severe leukocytosis (70,200/µl) with neutrophilia (92%) which gradually came down towards normal levels after treatment. The value of amylase was drastically increased (1478 IU/l) on 8th day as compared to the 1st day level (480 IU/l). The animal was negative for *Ehrlichia canis* and microfilaria. Stool analysis did not revealed any abnormality except occult blood (++++) present in high amount. The ultrasonographic examination revealed multiple anechoic area during scanning of liver which is suggestive of liver cyst, whereas, a scanning of kidneys revealed no cystic lesions. Radiographic examination revealed ground glass appearance with loss of all detail at upper abdomen whereas, lower abdomen had visible gas filled intestine. The liver was enlarged and noticed up to the level of 4th lumbar vertebra which, resulted in shifting of diaphragm cranially and had reduced excursion. Confirmed diagnosis was made by collection of cystic fluid (100 ml in one time from different places) by ultrasound assisted aspiration technique. The cytological examination of aspirated fluid revealed that the cystic fluid was of non-parasitic origin.

The dog was given fluid therapy (150 ml dextrose + 150 ml ringer lactate) iv for 3 days and ceftriaxone sodium along with tazobactum @ 562.5 mg im for 5 days followed by ampicillin and cloxacillin @ 500 mg bid im for 7 days. The animal was further given antacids and multivitamins as a supportive treatment. The decrease in neutrophils and leukocyte may be due to the effect of antibiotic. Lukewarm soap water was applied as enema and the animal defaecated within 5 min after application of enema. A slight improvement was recorded at every morning after aspiration of cystic fluid followed by fluid therapy attributed by intake of food with cessation of vomiting that indicates pressure of cysts on stomach wall might be the cause of vomiting. Zatelli *et al.* (2007) managed 19 dogs by cyst drainage and alcoholization. The role of percutaneous aspiration of the hydatid liver cyst using sclerosant has not yet been unanimously proved (Losanoff *et al.,* 2002). In the present case after every aspiration (a total 3 session), there was improvement but again there was accumulation of cystic fluid and the condition was further deteriorated. The dog died after 6 days of treatments. Hepatic cyst associated with clinical signs should promptly resected (Fossum, 2007) but in the present case, animal was so depressed, the surgical intervention could not be undertaken.

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Tail chasing in a German Shepherd dog: a compulsive obsessive disorder

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Abstract

A case of tail chasing in a German Shepherd dog was reported. Clinical examination did not reveal any abnormality and skin scraping was negative for ectoparasites. Treatment with Clomipramine and Clonazepam combination brought successful recovery.

Keywords : Clomipramine , Clonazepam, German Shepherd , Tail chasing.

Tail chasing or 'spinning' is an obsessive compulsive disorder (OCD) seen most often in Bull Terriers (Dodman, 1997). It is a stereotypic behavior characterized by ritualistic, repetitive, constant sequence of aimless movements and most often noticed in confined animals.

A 5 years old German Shepherd male dog was presented to Referral Veterinary Polyclinic, IVRI with a history of tail biting, continuous circling and inappetance. All the physiological parameters were found within normal range. Clinical inspection and skin scraping examination revealed absence of ectoparasites in tail region. The wound, due to self-mutilation, was found in the tail region which could be contributory to further irritation.The case was tentatively diagnosed as a behavioral abnormality.

The treatment included Clomipramine @ 25 mg (Total dose) daily orally for 3 weeks in morning and Clonazepam@1 mg (Total dose) orally for 3 weeks at night. Along with this Silymarine @ 1 tsp orally for 1 month was given as a supportive therapy. Owner was advised to provide regular heavy exercise to the pet to lose extra calories and divert attention, avoid fatty diets, and environmental stress including long time social isolation.

The causes of tail chasing may include behavioral causes like attention seeking, boredom and anxiety (Seksel and Lindeman, 2001), medical causes like physical injuries, irritation and neurological (Dodman *et al.,* 1996), or environment stress. Female dogs are more likely to be obsessive tail chasers. The severity and age of onset vary and environmental factors and hormonal changes of puberty or heat cycles, certain types of anesthesia, and the stress of undergoing a surgical procedure such as neutering have all been implicated as triggers (Moon-Fanelli and Dodman, 1998).

Clomipramine has been found effective in controlling signs of OCD and /or separation anxiety, noise phobia in dogs in conjunction with behavioral and environmental management (Seksel and Lindman, 2001). In German–Shepherds, the addition of anti-convulsant (Clonazepam) to an anti –obsessional drug regimen (Fluoxetine, Paroxitine and Imipramine) was found effective (Dodman *et al*., 1993; Foa *et al*., 1992). Tail chasing in dogs may be associated with cholesterol elevation, thus HDLP and LDLP cholesterol level may be used as biochemical makers for compulsive tail chasing (Yelchin *et al.,* 2009). Hence avoidance of fat rich diet in this case may be helpful in management of OCD. The successful management of tail chasing depends on owner compliance and understanding that

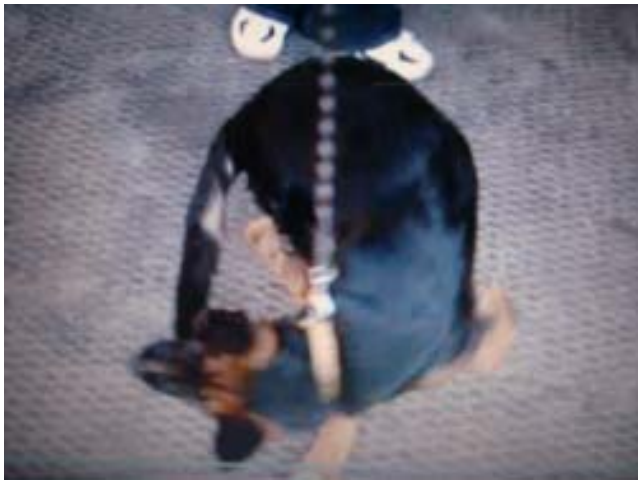

Fig. Tail Chasing by GSD.

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OCD cannot be cured, but can be well controlled (Overall and Dunham, 2002).

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Epidemilogical study of *E. canis* in Bangalore

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Abstract

Incidence of ehrlichiosis has been reported according to age, gender, breed and season. Out of 90 cases, 40 were found positive for canine ehrlichiosis. Occurrence in dogs aged >1 – 3 years were higher. Higher percentage of male dogs was affected with ehrlichia infection. Higher incidence was observed during summer compared to winter season.

Keyword: Epidemiology; E. Canis; Bangalore

Over the past several decades, tick-borne diseases have emerged as important threat to mammals worldwide, and have gained notoriety because of growing concern in changing climate conditions which favour vector-borne disease transmission.

Canine ehrlichiosis is a tick-borne disease caused by *Ehrlichia canis* an obligate intracellular rickettsia (Price and Sayer, 1983) and is mainly transmitted by the brown dog-tick *Rhipicephalus sanguineus* (Smith *et al.,* 1976). Canine ehrlichiosis has been reported to occur in Asia, Africa, Europe and America (Baneth *et al*., 1996). Mudaliar (1944) reported *E. canis* for the first time in India from Chennai. In the present paper incidence of ehrlichiosis has been reported according to age, gender, breed and season.

A total number of 90 clinically suspected cases of dogs presented to Veterinary College Hospital, Hebbal and Teaching Veterinary Clinical Service Complex, Yelahanka, Bangalore, during the period of September 2008 – April 2009 were selected for the present study. Dogs with clinical signs of pyrexia, tick infestation, depression, lymphadenopathy, melena, petechial haemorrahges, epistaxis were tentatively selected. Diagnosis was made by buffy coat smear examination using Giemsa's stain and confirmation was done by doing nested Polymerase Chain Reaction.

In the present study out of 90 cases, 40 (44.44%) were found positive for canine ehrlichiosis.

Out of the 40 positive cases dogs aged from 22 days to 13 years were affected with ehrlichiosis which is in agreement with the findings of Nims *et al.* (1971) and Harikrishnan *et al.* (2001)

Occurrence in dogs aged 1–3 years were higher (15%) which correlates with the findings of Moreira *et al.* (2003) and Bindu *et al.* (2006). However Tresamol *et al.* (1998) reported no significant difference between age groups. (Fig.-1)

In the present study higher per centage of male dogs (55%) were affected with ehrlichia infection compared to female dogs (45%) as also reported by Nims *et al.* (1971) and Costa *et al.* (2007). However, Harrus *et al.* (1997), Tresamol *et al.* (1998) and Bindu *et al.* (2006) analysed no sex correlation with *Ehrlichia* spp. infection.

Among 40 positive cases German shepherd had the higher (50%) occurrence compared to other breeds like Labrador retriever (22.6%), Non-descript (7.5%), Mastiff (5%), Spitz (5%), Doberman (2.5%), Dalmatian (2.5%), Dachshund (2.5%), and Golden Retriever (2.5%). GSD are more susceptible which correlate with the observations of Walker *et al.,* 1970; Bindu *et al.* (2006).

The finding supports the hypothesis that GSD are more susceptible to the disease due to weaker cellular immune response against *E. canis.* (Nyindo *et al*., 1980 and Harrus *et al*., 1997)

Higher incidence was observed during summer

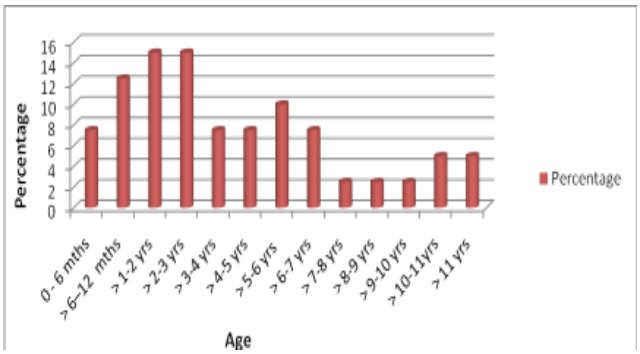

Fig. 1 Agewise occurrence of ehrlichiosis in dogs during Sept. 2008 – April 2009

season (March – April) (52.5%) compared to winter season (November – January) (30%) which coincides with the findings of Harrus *et al.* (1997) who also observed 79 per cent ehrlichiosis cases during summer.

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Therapeutic management of Ivermectin toxicosis in a dog- A case report

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Abstract

A 3 yrs old, sexually intact female spitz dog was presented after having consumed approximately 50 mg (6.25 mg /kg b wt) of an oral ivermectin preparation used for treatment of intestinal parasites in large animals. Results of haematological findings were unremarkable, except for mild leucopenia (4,543/µl), neutrophilia (92%), lymphopenia (7%) and eosinopenia (0%), compatible with stress. Therapeutic management with atropine sulphate, balanced electrolyte solution and methyl cobalamin was found effective in alleviating the acute symptoms.

Keywords: Atropine sulphate, dog, Ivermectin toxicosis.

Ivermectin is commonly used anti parasite drug that causes neurologic damage to the parasite, resulting in paralysis and death. Its toxicity is seen when an overdose of the drug is administered or consumed or in pets with increased sensitivity to the drug. The most common causes of ivermectin toxicity is due to administration of excessive doses (10- 20 X the recommended dose) and breed sensitivity. Toxicity results in any number of clinical signs ranging from mild to extremely severe, including death. Picrotoxin (Sivine *et al.,* 1985) or physostigmine (Lovell, 1990) or neostigmine (Pathania and Kant, 2010) has been used for ivermectin toxicosis. The present report describes ivermectin toxicosis and its management with atropine sulphate in a dog massively overdosed (50 mg) by its owner.

A 3 yrs old 8 kg b.wt. sexually intact female spitz dog was examined after having overdosed 16 hours before presentation with approximately 50 mg (6.25 mg /kg b wt) of an ivermectin preparation used for large animals treatment for intestinal parasites. Clinical examination showed hypothermia (99.8^{0}F), bradycardia (60 beats /min), respiration (13 beats/min) with weak shallow thoracic excursions, excessive drooling, bilateral blindness, head pressing, bilateral mydriasis, disorientation, weakness, inability to rise and severe depression. On neurological examination, the dog was stuporous, with head pressing, dilated pupils and reduced menace and pupilary light reflexes for both eyes. Withdrawal reflex were detected in all limbs, with normal to exaggerated patellar and gastrocnemius reflexes. The findings were suggestive of diffuse intracranial disease. Hence, ivermectin toxicosis was diagnosed by history of exposure to high dose of ivermectin and clinical signs. Haematobiochemical parameters *viz* Hb, TEC, PCV, TLC, DLC, ALT, AST, BUN, serum creatinine, total protein and albumin were estimated on day 0 of presentation. General supportive care consisting of a balanced electrolyte solution @ 40 ml/kg b. wt. iv, atropine sulphate @ 0.07 mg/ kg b wt iv and methylcobalamin @ 1ml im od for three days was administered. After 3 day of treatment, methylcobalamin 1 tablet orally for 7 days was advised.

On day 2, dog had little milk and began walking with assistance while complete recovery was observed on day 5. Vision, as assessed by menace response, returned on day 4 of treatment. Results of haematological findings were unremarkable, except for mild leucopenia (4,543/µl), neutrophilia (92%), lymphopenia (7%) and eosinopenia (0%) compatible with stress. Biochemical analysis was within normal reference range.

In simple stomach animals ivermectin is upto 95% absorbed after oral administration and presumably undergoes hepatic metabolism and excretion, with more than 95% of tritium leveled ivermectin found in faeces and only a small portion found in urine (Pullian and Preston, 1989). Ivermectin toxicosis results from increased CNS action of γ- aminobutyric acid (GABA). Once in CNS, ivermectin or its metabolites stimulate release of GABA from pre synaptic CNS neurons and increase post synaptic receptor site binding of GABA. This increased GABA activity increases post synaptic chloride conductance, resulting in neuronal hyperpolarisation and depressed neuronal function. This effect is global, causing brainstem, cerebellar, and

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cortical deficits resulting in clinical signs (Hadrick *et al.,* 1995).

The main stay of management is supportive and symptomatic care. Atropine sulphate, which is a muscarinic receptor antagonist and does not cross the blood brain barrier, was found effective to overcome ivermctin toxicosis (neurological signs, drooling and bradycardia) in present study. Picrotoxin has been proposed as specific antidote but has narrow margin of safety and is not considered the best treatment for ivermectin toxicosis. Physostigmine, reversible inhibitor of acetylcholine esterase that can penetrate the blood brain barrier has shown some effect in comatose animals due to increased concentration of acetylcholine in affected animal (Lovell, 1990). Muhammad *et al.* (2004) and Pathania and Kant (2010) used neostigmine for ivermectin toxicity in cats and pomerarian pup, respectively.

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Organochlorine poisoning in dog and its successful treatment

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Abstract

Organochlorine poisoning was diagnosed in five dogs (two Rottweiler and three Labrador) of two year age with the history of watery diarrhea and emesis after the accidental intake of Parrysulfan mixed water on the previous day. All the affected animals were treated symptomatically and had an uneventfully recovery.

Keywords: Organochlorine, poisoning, dog, treatment.

Endosulfan is an organochlorine insecticide, ubiquitous environmental pollutant, actually toxic and has poisoned numerous people, livestock and wildlife. Endosulfan has resulted in congenital birth defects, reproductive health problems, cancer, immunosuppression, neurological and neurobehavioral problems. Organochloride pesticides may be absorbed through the intact skin as well as by inhalation and the gastrointestinal tract. Its mode of action as a CNS stimulant leads to convulsions. It is rapidly metabolized and its metabolites are excreted in the urine and faeces (Choudhary *et al.,* 2003). However literature on endosulfan toxicity in dogs appears to be lacking. The present paper records endosulfan poisoning in dogs and its successful treatment.

Three Labrador dogs (two female and one male) of two year age and two Rottweiler male dogs of the same age were presented to the Teaching Hospital complex with the history of watery diarrhea and emesis after the accidental intake of endosulfan mixed water (half liter mixed with five liters of water) on the previous day intended for agricultural use.

Endosulfan contamination does not appear to be widespread in the aquatic environment, but endosulfan has been found in agricultural runoff and rivers in industrial areas where it is manufactured or formulated (IPCS, 1984). Organochlorine insecticide can be absorbed orally and topically, with absorption being rapid due to the lipid solubility of these compounds (Aslani, 1996).

Four other dogs of the same owner died before reporting to the hospital with symptoms of convulsion, salivation, emesis and diarrhoea. The clinical symptoms observed in the present case were in agreement with those of Choudhary *et al*. (2003) in rats. The endosulfan blocks the inhibitory receptors of the CNS, disrupts the ionic channels and destroys the integrity of the nerve cells. Acute toxic effects include dizziness, vomiting, hypersensitivity, tremors, lack of coordination and convulsions. Chronic exposure may result in permanent damage to the nervous system which may manifest in various kinds of neurological problem (Gandolfi and Cheney, 1984).

History revealed that all those animals which had an accidental exposure to the pesticide, had vomiting and diarrhoea. Physiological parameters of the affected animals such as rectal temperature 38.2°C-39.0°C, pink conjunctival mucus membranes, pulse rate 110-120/min, respiratory rate 22-26/min and other parameters were well within the normal range.

There is no specific antidote for endosulfan poisoning in animals (Osweiler, 1996). It was opined that in case of oral exposure, the animal should be given an absorbent or mineral oil (Raisbeck, 2006). Mineral oil may be more effective than saline cathartics or sorbital (Messonnier, 2001). All the affected animals reported to the Clinical Complex were treated with Inj. Normal Saline @ 10ml/kg b wt iv, Liquid Paraffin @ 1ml /kg orally and liver extract B.Complex 1-2 ml im. After twenty four hours of treatment, all the five animals recovered uneventfully.

Acknowledgement

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Canine Ehrlichiosis and its therapeutic management

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ABSTRACT

The present study was conducted on eight clinical cases of canine ehrlichiosis, irrespective of age, sex and breeds presented with a history of fever, anorexia and in some cases clotting abnormalities leading to bilateral nasal bleeding. Clinical examination revealed fever, increased respiratory rate, dehydration, dullness, pale mucus membrane with marked anaemia and enlargement of lymph nodes. Giemsa stained blood smears prepared from all 8 dogs revealed presence of small spherical cocco-bacillary ehrlichia inclusions in monocytes. Blood examination revealed significant decrease in Hb, TEC and PCV. Seven out of eight dogs treated with oxytetracycline followed by doxycycline, antipyretic and haemocoagulant along with dextrose saline, multivitamins and haematinics resulted into clinical recovery.

Keywords: Dog, Ehrlichiosis, Canine, Treatment

Ehrlichia canis infection has been recognized worldwide. It is transmitted by brown dog tick *Rhipicephalus sanguineus* (Kumar *et al*., 2010) and characterized by reduced blood cellular elements (Ashuma *et al.,* 2005). It affects lymphatic system, particularly the immune system and so best related as AIDS of the canine world. Natural ehrlichiosis infection often goes undetected during subclinical phase or because of non-specfic symptoms which mimic other diseases and thus misinterpreted (Pusterla *et al.,* 1997). The cases can be managed using oxytetracycline (Ettinger and Feldman, 2000; Harikrishnan *et al.,* 2002; Singh *et al*., 2009; Kumar *et al*., 2010).

The present study was undertaken on 8 clinical cases of canine ehrlichiosis, irrespective of age, sex and breed which were presented to Veterinary Teaching Hospital of the college with the history of fever, anorexia, oedema of limbs and abdominal region and serous ocular discharge in all cases and clotting abnormalities leading to bilateral nasal bleeding in three of them. Most of these cases were previously treated with broad spectrum antibiotics without any significant improvement. Clinical examination of the dogs revealed elevated rectal temperature (105-107 ^{0}F), increased respiratory rate, dehydration, dullness, pale mucus membrane with marked anaemia and enlargement of lymph nodes. In three cases, erythematous pustules were prominent in abdominal and medial sides of the hind limbs. Haematological examination revealed significant decrease in Hb, TEC and PCV. Examination of Giemsa stained blood smears revealed presence of small spherical cocco-bacillary ehrlichia inclusions in monocytes which were identified as *E. canis*.

All the dogs were treated with oxytetracycline @ 20 mg/kg b wt intravenously daily for 5 days followed by doxycycline @ 10 mg/ kg b wt orally twice a day upto 21 days. Phenyl butazone @ 30 mg/kg b wt intramuscularly during febrile phase and N- butanol and citric acid @ 1 ml intramuscularly in dogs with bilateral nasal bleeding for 5 days were also advocated. Normal saline during febrile phase and 5% dextrose saline during non-febrile phase were given intravenously for rehydration and maintenance of nutritional status. Liver extract and vitamin B-complex were administered @ 2ml intramuscularly on alternate days for eight occasions along with oral haematinic @ 1 TSF orally once daily for subsequent 30 days. The tick infestation was controlled by application of coat cleansing shampoo followed by dips of 12.5% deltamethrin mixed with water @ 2 ml per liter at weekly interval.

There was marked clinical improvement in majority of cases after 72 hours of treatment as evidenced by normal body temperature and regression of erythematous pustules. However one dog collapsed after 3 days of treatment, which could be attributed to hemorrhages and concurrent infection in chronically infected dog (Ettinger and Feldman, 2000 and Singh *et al*., 2009)

Bleeding disorders manifested by epistaxis, melena and petechiae, pallor due to anaemia, severe weight loss, debility and neurological signs due to

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dehydration were the typical signs in affected dogs as reported by Harrus *et al.* (1997) and Ettinger and Feldman, (2000). Intravenous administration of oxyteracycline followed by oral administration of doxycycline has been proved useful in early recovery. Singh *et al*. (2009) and Harikrishnan *et al.* (2002) also recommended the use of oxytetracycline along with doxycycline for treatment of chronic canine ehrlichiosis. Use of vitamin-B complex and liver extract along with tick control by deltamethrin dip has been advocated by Kumar *et al.* (2010) also.

Prognosis of the disease is reported to be good in acute phase but it becomes worse as the disease progresses (Ettinger and Feldman, 2000).

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Osteodystrophy with facial hyperostosis due to chronic renal failure in two dogs

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Abstract

Clinical cases of osteodystrophy secondary to chronic renal failure in adult dogs were diagnosed. Anemia in the present cases thought to be due to chronic renal disease. Vomiting in the dogs might be associated with stimulation of chemoreceptor trigger zone by uremic toxins, decreased excretion of gastrin resulting in increased gastric acid secretion and gastrointestinal irritation. Other clinical parameters noted.

Keywords: Osteodystrophy, renal failure, dogs.

Two male Labrador and Rottweiler dogs aged five to seven years weighing around 30 and 40 kg respectively were referred to the Teaching Veterinary Hospital of Institute with the history of swollen face, inappetance, weakness, occasional vomiting, polyuria for last 20-30 days.Physical examination revealed normal rectal temperature, tachycardia, tachypnea, pale mucus membrane, moderate degree of dehydration and hard swelling of nasal and frontal bones, which was not painful on palpation. Hematological findings were anemia,leucocytosis with neutrophilia. Serum biochemistry revealed increased urea, increased creatinine, hyperphosphatemia, increased alkaline phosphatase and normal calcium, alanine amino transferase and bilirubin (table 1). Urine analysis findings were reduced specific gravity (1.015), scattered distribution of epithelial cells, occasional distribution of RBC, WBC, bacteria, mild proteinuria and some granular casts. Rostroventral radiographic view of upper jaw and skull revealed generalized reduced bone density of jaw and head area. The maxilla and mandibles were enlarged and poorly mineralized. An ultrasonographic examination showed small and irregular shape kidneys and poor corticomedullary differentiation in kidneys was observed. Cytological examination of fine needle aspirate of the swollen jaw area ruled out possibility of bone tumor/ inflammatory swelling. The dogs were treated with Ringers lactate@ 250 ml iv (Albert Davis), dextrose normal saline@ 250ml iv (Wokhardt), ampicillin @ 10mg/kg b.wt. im (Roscillin, Ranbaxy), metoclopromide@ 0.5 mg/kg b. wt. im (Perinorm, IPCA), multivitamins @ 2ml im(Polybion, Merck), furosemide @ 2mg/kg b.wt. im(Lasix,Hoechst Marion Russel), phosphorus binders (Gelusil, Parke Davis) and advised phosphorus-restricted diet. The treatment was done twice a day. The Labrador dog died after four days of start of treatment.

Table1: Clinical and hematobiochemical parameters of dogs

Parameters	Case 1 (Labrador)	Case 2 (Rotweiller)
Temperature (^{0}F)	100	100.6
Heart rate/minute	138	130
Respiration/per minute	64	58
Hemoglobin (g/dl)	3.8	8.8
TLC (µl)	27000	13480
Neutrophils (µl)	23700	10260
Lymphocytes (µl)	3300	3220
BUN (mg/dl)	183	36
Creatinine (mg/dl)	19.6	4.5
Calcium (mg/dl)	8.6	10.1
Phosphorus (mg/dl)	27.9	4.8
AKP (U/L)	204	549
ALT (U/L)	15	26
Bilirubin (mg/dl)	0.4	0.1

Rottweiler dog showed marked clinical improvement after 7 days of start of treatment. Animal became alert and active, started taking food and stopped vomiting. Owner did not bring the dog after 7 days and treatment was discontinued. Animal remained stable for about one month and after that again animal's condition deteriorated. This time, dog was presented with more severe signs of vomiting, anorexia, polyuria, weakness and swollen face. Hematobiochemical findings revealed severe anemia (Hemoglobin-5.2g/dl), markedly elevated BUN (180mg/dl), Creatinine (6.0mg/dl), hyperphosphatemia (14.8mg/dl). Same treatment as above was advised for next 7 days. The dog died two days after start of treatment.

The dogs were diagnosed to have chronic renal failure on the basis of history, haematobiochemical, urinalysis and ultrasonographic findings and the osteodystrophy observed in these dogs were due to chronic renal failure. Anemia in the present cases thought

to be due to chronic renal disease. Vomiting in the dogs might be associated with stimulation of chemoreceptor trigger zone by uremic toxins, decreased excretion of gastrin resulting in increased gastric acid secretion and gastrointestinal irritation (Nelson and Couto, 1998). The skeletal lesion like swelling and reduced density of upper jaw bones observed in these dogs could be due to renal secondary hyperparathyroidism, which resulted from impaired renal excretion of phosphate and consequent hyperphosphatemia (Rosol and Capen, 1996). Bones of the skull and mandible are most severely affected and marked proliferation of the connective tissues associated with the maxilla causes distortion of the face (Ettinger and Feldman, 2000). Neutrophilic leucocytosis might be due to bacterial infection in the urinary tract. The serum calcium concentration was within the normal range, which may be due to continued coordination of PTH and 1,25-dihydroxy vitamin D in calcium metabolism (Rosol and Capen, 1996).

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An outbreak of infectious keratoconjunctivitis due to *Moraxella bovis* in a nomadic Buffalo herd

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Abstract

The present report gives an account of an outbreak of infectious keratoconjuncitivitis in a herd of 65 nomadic buffaloes. A female buffalo of one year's age was presented in university hospital with the history of redness in eyes, mucopurulent discharge and epihora. Animals in the herd were examined for eye lesions. Isolates from eye swabs collected for microbiological culture were identified as *Moraxella bovis* and culture sensitivity test revealed high sensitivity for chloramphenicol, oxytetracycline, ceftriaxone, cefataxime and ciprofloxacin. Flies collected were identified as *Haematobia spp* and *Musca spp.* Morbidity rate of IKC, on the basis of clinical signs and demonstration of *Moraxella bovis* in the buffalo herd was 32.7 per cent. Incidence of IKC was higher in calves. Affected animals were treated with chloramphenicol eye drops instilled tid for five days and parental single injection of long acting oxytetracycline @ 20 mg/kg b.wt deep im. All affected buffaloes recovered without any sequel.

Keywords: Buffalo, Infectious keratoconjuncitivitis, *Moraxella bovis,* Treatment.

Pinkeye is non-fatal ocular disease and has great economic impact on the livestock industry in terms of losses resulting from decreased weight gain, milk production, cost of treatment and labor, decreased value of calves and adults due to disfigurement of the eyes (Smith, 2001). The present communication reports an outbreak of infectious keratoconjuncitivitis in a nomadic buffalo herd.

A female buffalo of one year's age was presented in university clinics with the history of redness in eyes, mucopurulent discharge and epihora. Nomadic buffalo herd of 65 animals comprised of 34 adult lactating and pregnant buffaloes, one bull, 18 heifers and 12 calves. Animals were examined for bulbar, palpebral conjunctival mucous membranes, sclera and cornea, any discharge and foreign bodies. Eye swabs were collected for microbiological culture and identification of causative agents (Quinn, 1999) and culture sensitivity test was done by method of Kirby *et al,* (1966). Flies were collected for identification. A total of 21 buffaloes out of 65 were affected, which included 4 adults, 7 heifers and 10 calves. Conjunctival mucous membranes were severely congested with mucopurulent discharge and epiphora attracting the flies around the eyes. Mishra *et al.* (1995) observed red and swollen conjunctiva with serous ocular discharge, blepharospasm and photophobia as clinical signs in an outbreak of IKC. Culture was identified and confirmed as *Moraxella bovis* on basis of colony characteristics (flat round, grayish white, friable colonies surrounded by narrow zone of complete hemolysis on sheep blood agar), biochemical tests of colonies (positive for oxidase, catalase, nitrate reduction and gleatinase and negative for urease and phenylalanine deaminase) and examination of Gram's stained smears of culture (short plump and paired rods). Culture sensitivity test revealed high sensitivity for chloramphenicol, oxytetracycline, ceftriaxone, cefataxime and ciprofloxacin and resistant for gentamicin.

Morbidity rate of IKC, on the basis of clinical signs and demonstration of *Moraxella bovis* in the buffalo herd was 32.7 per cent. Incidence of IKC in calves, heifers and adult buffaloes were 83.3, 38.88 and 11.76 per cent, respectively. Inverse relation was found between age and infection of IKC in the animals. Takele and Zerihun (2000) also reported higher prevalence of IKC in younger and crossbred animals of Ethiopia. *Moraxella bovis, M. urethralis and Actinobacillus sp.* have been earlier identified and reported as an etiology of pink eye from affected buffaloes (Achdijati *et al.,* 1983).

Flies were identified as *Haematobia spp* and *Musca spp* and were incriminated as mechanical transmitter of *M. bovis* in buffaloes. Lyutskanov (2001) isolated bacteria in the viscera of *Musca spp* and confirmed role of flies as a mechanical vector in the mechanism of transmission of IKC. All the affected animals were treated with boric acid solution eye wash, chloramphenicol eye drops tid for five days and parentral single injection of long acting oxytetracycline @ 20 mg/

kg b.wt deep im. Samea *et al*. (1994) reported complete recovery from IKC in buffaloes treated with chloramphenicol ophthalmic ointment. However, Chadli (1992) reported high recovery rate in cattle group treated with single im injection of long-acting oxytetracycline as compared to group treated with chlortetracycline ophthalmic ointment. All affected animals recovered without any sequel to IKC. Control of flies was found difficult in nomadic buffalo herd. An outbreak of infectious keratoconjuncitivitis in the nomadic buffalo herd is reported and its successful management is discussed. It was concluded that early treatment prevents sequel of IKC.

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ISVM Awards & Rules

The members of the ISVM are requested to send their applications in quadruplicate (4 copies) for the below mentioned awards with all relevant supportive documents including photocopy for proof of age, date of enrollment as life members etc., as required for specific award, through their controlling officer/Head of the department verifying their credentials/contributions.

General rules applicable to all the awards:-

1. Only members of ISVM are eligible to apply for any of the awards
2. Incumbent executive committee members of ISVM are not eligible for consideration of any of these awards, exception being FISVM.
3. Recipient of any ISVM award during last two years is not eligible to apply or to be considered for any of ISVM award
4. Persons applying for more than one award should indicate the preference order.
5. The minimum quorum for all the awards except FISVM is two.
6. The General Secretary, in consultation with the President, may reject application for any award for want of required quorum or incomplete application.
7. The General Secretary and President reserve all rights to accept/reject any application without assigning any reason for the same.
8. All the applicants will be required to submit half page write-up for the citation along with their original applications.
9. A sum of Rs. 250/- will be payable by the applicants as demand draft in favour of Indian Society for Veterinary Medicine, for each award applied as the processing fee to the ISVM.
10. The award may not be given for a year if an applicant does not get the qualifying minimum score.

1. SHRI RAM LAL AGRAWAL GOLD MEDAL

The Gold Medal has been instituted by M/s Indian Herbs Research & Supply Co. Saharanpur (Uttar Pradesh) in the memory of its Founder, Late Shri Ram Lal Agrawal. The award shall be conferred annually upon a scientist who is a life member of the Indian Society for Veterinary Medicine in recognition of his/her distinguished work/ contribution in the field of Veterinary Medicine. In every third year, the award shall be given to a scientist who has contributed significantly in the field of Indigenous Veterinary Medicine. The individual competing for this Gold Medal, should be of minimum 45 years of age as on 1st January of the year of evaluation.

2. INTAS YOUNG SCIENTIST AWARD

The ISVM award silver trophy to a young scientist (life member of ISVM) below 32 years of age on 1st of January of the year of presenting his/her own research findings at the time of convention of ISVM. Besides the award, citation will also be given. Entries in duplicate of full length research paper typed in double space on bond paper with zerox copy of the proof of age should be sent on or before the last date prescribed to the General Secretary, ISVM along with the certificate from the Head of the Department, verifying the place of research work and year during which work was done. The paper to be presented for this award shall not find a place as Abstract in the proceedings of the convention. Only the author's name will be given who will present the paper himself/ herself. The award in the form of trophy and a certificate to the selected young scientist shall be given at the Annual Convention in the same year. If the research work presented by the young scientist is the part of his/her post-graduate thesis, the same has to be mentioned clearly along with a certificate from the Advisor (Guide). The name of the guide will also figure in the award certificate. The executive committee may reject incomplete application/award for want of required quorum (minimum of 2 candidates). They also reserve all rights to accept/ reject the application without assigning any reasons for the same.

3. DR. D.C. BLOOD GOLD MEDAL

Dr. D.C. Blood Gold Medal will be awarded every year from the interest accruing on the corpus fund of Rs.

10,000/- deposited for the said purpose out of the savings of ISVM convention held at A.P.A.U., Hyderabad, 1990. The award will comprise a gold plated medal and a citation. The award is open for the life members of ISVM of the age above 32 and below 45 years as on 1st January of the year of evaluation.

4. SMT. P.Z. SHARMA GOLD MEDAL FOR CANINE MEDICINE

The Gold Medal will be awarded from the interest accrued on the corpus funds of Rs. 10,000/- deposited for the said purpose by Dr. S.Z. Sharma, Veterinary Clinic Sukhmani, TVPD Scheme, 10th Road, Mumbai-49 (Maharashtra).The medal will be awarded every alternate year from 1994 onwards (the date of commencement of award) to a life member of the ISVM in recognition to his/her outstanding contributions in the field of Canine Medicine during the last 5 years in the form of quality research and clinical publications, generating innovative ideas and development of newer techniques.

5. S.K. MYLSAMY GOUNDER GOLD MEDAL FOR POULTRY MEDICINE

The award is open for life members of ISVM. The applications indicating teaching, research and extension and other activities in the field of Poultry Medicine will be invited by the General Secretary every year and must be submitted by aspirants through respective Heads of Department/Office, verifying their credentials/contributions on or before the prescribed date.

6. DR. G.N. DUTTA MEMORIAL AWARD

This award will be conferred to an applicant who has completed 5 years as life member of ISVM and is of minimum 45 years of age as on 1st January of corresponding year. The award will be given in recognition of his/her meritorious research contributions during the last 10 years to the Veterinary Medicine especially in area of management of infectious diseases in animals.

7. P. K. DAS GOLD MEDAL

This award will be conferred to life member of ISVM in recognition of his/her outstanding contributions in the field of Clinical Toxicology during the last 5 years in the form of quality research and clinical publications, generating innovative ideas and development of newer techniques.

8. AWARD OF FELLOWSHIP OF ISVM (FISVM)

The life members of ISVM for the past 10 years are only eligible for the award of FISVM. ISVM Fellowship is awarded only to the members having master's degree in Veterinary Medicine/Preventive Medicine/Clinical Medicine and their designation should not be lesser than the rank of Professor. The fellowship is awarded to a person of high professional standing, who has rendered commendable service for the cause of Veterinary Medicine. The application for FISVM is required to be submitted along with comprehensive information about academic and professional achievements, distinguished published work/contributions in the field of Veterinary Medicine. A demand draft of Rs. 2000/- in favour of "Indian Society for Veterinary Medicine" must be enclosed with the application as the required fee for FISVM. The eligible ISVM members are requested to submit their application with detailed information under the following subheads: Academic and research achievements. Total particulars of employment; National and ISVM awards received;Total number of research and clinical publications (not popular articles) in peer reviewed Indian and Foreign Journals (enclose the list); List of published reviews in Journals and international proceedings/books; Books/Monographs published; Number of research and clinical publications relevant for specific award (enclose list) ;Contributions to the advancement of Veterinary Medicine; List of papers presented in international conferences/symposium held abroad and any other scientific achievements not covered in above cited information.

9. FIELD VETERINARIAN AWARD

i) The Field Veterinarian Award shall be given to a field veterinarian who is a life member of ISVM.

ii) The award shall be given in the form a plaque and certificate at ISVM convention based on the oral presentation by the applicant in National Symposium.

iii) The presentation shall be judged by a committee of three members nominated by the executive committee for

this purpose.

10. ISVM MERIT AWARD FOR POST GRADUATE RESEARCH:

There shall be two ISVM Merit Awards annually – one for a student pursuing PhD .degree in the discipline of Veterinary Medicine and one for a student pursuing MVSc. degree in the discipline of Veterinary Medicine (Veterinary Clinical / Preventive Medicine). The value of award for a Ph.D. and M.V.Sc student shall be Rs.5000/ -andRs.4000/-respectively to be given once during the degree programme of the student. The applications for the award must reach the office of ISVM by (stipulated date) duly recommended by the guide and forwarded by the Head of the Department concerned.

11. BEST CLINCAL ARTICLE AWARD

It will be awarded to the best clinical article published in the Indian Journal of Veterinary Medicine during the year immediately preceding the year of annual conference of ISVM. The articles will be sent to three Judges by the Editor and based on the score; the best article will be selected and placed before the executive committee members for approval. The award will be in the form of a plaque and certificate to the first author, and certificates to co-authors to be given every year at the ensuing ISVM convention. 4. The criteria for scoring marks shall be as follows:Title 5 marks; Materials & Methods 10 marks; Results & Discussion 10 marks; Contribution to Science 5 marks

12. BEST RESEARCH ARTICLE AWARD

It will be awarded to the best full length research article published in the Indian Journal of Veterinary Medicine during the year immediately proceeding the year of annual conference of ISVM. The research articles will be sent to three Judges by the Editor and based on the score; the best research article will be selected and placed before the executive committee members for approval. The best research article award will be in the form of a plaque and certificate to the first author, and certificates to co-authors to be given every year at the ensuing ISVM convention. The criteria for scoring marks shall be as follows: Title 5 marks; Materials & Methods 10 marks; Results & Discussion 10 marks; Contribution to Science 5 marks

13. ISVM APPRECIATION AWARD

The ISVM Appreciation Award will be given to a life member of the Society. It will carry a citation and plaque of honour. The proposal for the award with justification will be made by the President, General Secretary, Organizing Secretary and Editor/Associate Editor during the executive committee meeting at the time of annual convention on the basis of services rendered by a life member for the betterment of ISVM/IJVM affairs. The executive committee will approve a maximum of 4 persons each year for this award from the names proposed as above.

Award Application procedure

The eligible members are requested to submit application for the awards/fellowship (SerialNo.1 to 10) with detailed information under the following subheads with detail description on specific need of the individual award: Academic and Research Achievements; Total particulars of employment; National / international and ISVM awards received Total number of research and clinical publications(not popular articles); in peer reviewed Indian and Foreign Journals(Enclose the list); List of published reviews in journals and international proceedings/ books; Books/monographs published; Number of research and clinical publications relevant for the specific award(Enclose the list); Contribution to the advancement of Veterinary Medicine; List of papers presented in the international Conference & Symposia

Remark Note:

(i) Members of the incumbent executive committee of ISVM are not eligible for consideration of any of these awards except for Fellow ISVM.

(ii) Recipient of any of the ISVM awards during last 2 years is not eligible to apply/or be considered, for example, any person getting the award in 2005 shall be eligible to apply in 2008 and so on.

(iii) Person applying for more than one award should indicate preference order.

(iv) Person competing for any of these awards cannot become member of any of the Judging Committee

PROCEEDINGS

General Body Meeting of ISVM held on 2nd Feb 2012 at 5PM| at College of Veterinary Science and Animal Husbandry, Selesih, Aizawl, Mizoram

The meeting was presided by the President, ISVM, Dr Susovan Roy and following executive Body Members were present on the dais.

Dr S.S. Randhawa – Vice President

Dr R. K. Bhagerwal – Vice President

Dr S. Dey – Editor

Dr R. C. Patra – General Secretary

The General Body meeting was attended by 74 life members of the society. The President welcomed the esteemed members of the society. The following decisions were made.

1. Dr Mahesh Kumar was nominated as the Election Officer for the next election for selection of new Executive Body members.
2. Veterinary College, MHOW shall hold the 31st Convention of ISVM with Dr R. K. Bhagerwal as the Organizing Secretary.
3. Dr R. D. Sharma field Veterinarian Award was instituted and the call for application will be made from the year, 2012
4. It was decided to re-look into the score-card for different categories of award from the society. For that matter a committee was formed with Dr A. K. Galhot as the Chairman and Dr S. S. Randhawa and Dr S. Prathaban as the members.
5. A committee was also formed with Dr N. N. Pandey as Chairman and Dr R. K. Bhagerwal and Dr D. C. Nauriyal as members to prepare guidelines for submission of abstracts to be published in the proceeding of National Symposium of ISVM.
6. The life membership fee of the Society was increased to Rs 1500 and this will be effective from 1st April 2012.

The meeting ended with thanks

Scientific Recommendation of 30th ISVM Annual Convention and the National Symposium on "Health Strategies including Biotechnological Interventions and Animal Welfare to augment productivity of animals with special reference to North Eastern Region." Held at College of Veterinary Science and Animal Husbandry, Selesih, Aizawl, Mizoram w.e.f. 1st to 3rd February

The Executive Body Meeting was held on 31st Jan 2012 at 6PM. The following members were present.

Dr Susovan Roy	–	President
Dr S.S. Randhawa	–	Vice President
Dr S. Dey	–	Editor
Dr D. B. Mondal	–	Associate Editor
Dr R. C. Patra	–	General Secretary
Dr M. Chandrasekhar	–	Secretary Southern Region
Dr Rakesh Ranjan	–	Member, Editorial Board
Dr Gunjan Das	–	Secretary, North East Region
Dr Mahesh Kumar	–	Immediate Past President
Dr N. N. Pandey	–	Organizing Secretary
Dr D. S. Nauriyal	–	Member, Editorial Board

The president of ISVM welcomed all the members, and the General Secretary detailed the financial position of the society in absence of the treasurer. The organizing secretary described the three-day technical program for the 30th Annual convention and National Symposium.

ISVM AWARD – 2011

It was decided to honour the following ISVM members in the inaugural function of the Symposium.

1. Dr A. K. Galhot – Fellow of Indian Society for Veterinary Medicine
2. Dr K. Nalini Kumari – Smt Ava Roy Gold Medal
3. Dr P. K. Dash Gold Medal – Dr P. Selvaraj
4. Dr G. N. Dutta Gold Medal – Dr N. Sahoo
5. S. K. Mylsamy Gounder Gold Medal for Poultry Medicine – Dr Prakash Bhatt
6. ISVM Honour to immediate past Organizing Secretary – Dr D. V. Keskar
7. ISVM Appreciation Award
   Dr K. K. Sardar
   Dr Kranti Sharma
   Dr Kalyan Sharma
   Dr G. B. Rajesh
   Dr Rakesh Ranjan
8. Best Research Article (published in IJVM) Award - Dr D. Chandrasekhar and Co-workers
9. Best Clinical Article (published in IJVM) Award – Dr Satish Kumar and Co-workers

The agenda relating to holding of 31st Annual Convention of ISVM, was discussed as there was four proposal to hold the said conference. The matter was taken to General Body for discussion.

The Executive Body decided to nominate the Dr Mahesh Kumar, Ex-President of the Society and Professor and Head, Department of Medicine, College of Veterinary Science and Animal Husbandry to facilitate the society by discharging the function of Election Officer for the forthcoming election for formation of Executive Body.

Dr R. D. Sharma Field Veterinarian Award -The Executive body agreed for the proposal and the matter was taken to General Body.

The meeting ended with thanks to all.

GENERAL GUIDELINES FOR CONTRIBUTORS

The Indian J. Vet. Medicine is published twice in a year, June and December. It contains review articles (guest), original/applied research articles, clinical observations, preliminary reports of scientific studies and short communications on Veterinary Medicine and Animal Health. In addition, the journal also publishes Letters to the Editor, Tips to Vets and other relevant informations.

Manuscripts. The manuscripts are accepted on the basis of scientific importance and suitability for publication on the understanding that they have not been published, submitted or accepted for publication elsewhere wholly or partly in any language. The copyright of papers, accepted for publication, belongs to *The Indian Society for Vet. Medicine*.

The official language of journal is English. The articles should be sent to **The Assoc. Editor, Indian Journal of Veterinary Medicine, Division of Medicine, IVRI, Izatnagar-243 122, Bareilly, U.P., India**. The manuscript should be typewritten on one side of the paper with wide margins and double spacing throughout except in abstracts, footnotes and references which should be in single spacing. It should be sent in duplicate. Each page of the manuscript should be numbered on the top corner including title page, references, tables, etc. All the pages should contain running title of the paper and surname of author(s) at the top.

Small corrections, if necessary, in the manuscript may be inserted in between the lines but the space where they should go, must be clearly indicated. Large corrections should preferably be typed on separate sheets and attached at proper places.

The manuscript should be organized in the following order in general:

Title with author(s) name(s) and complete address for correspondence with PIN code

Abstract, Keywords, Introduction, Materials and Methods, Results, Discussion, Acknowledgement, if any, References, Tables, Figures

Title: Papers should be headed with full title, the initials and surname(s) of the author(s) and address of the Institution where the work was carried out. A shortened version of the title should also be supplied for running headlines. The serial titles are not acceptable, so each paper should have an individual title.

Abstract: This should not exceed 300 words and should outline briefly the purpose of the study, important findings and conclusions. Repetition and generally known information should be avoided.

Keywords: Important and relevant 4-6 keywords be mentioned.

Introduction: This part should state briefly the nature and purpose of the work together with the important findings of previous workers.

Materials and Methods: The author(s) should describe materials, methods, apparatus, experimental procedure and statistical methods in detail to allow other workers to reproduce the results. Sub-heading may be used in this part.

Results: The experimental data should be presented clearly and concisely. Information presented in tables and figures should not be repeated.

Discussion: This should focus the interpretation of experimental findings. Do not repeat data presented in the introduction or information given in the result. References in this part should be cited as follows…..as observed by Kumar *et al.* (1984) or in parentheses…….. were found (Dwivedi *et al.,* 1983; Singh and Singh, 1984).

Acknowledgement(s): This should be short. Grants and technical helps provided should be acknowledged.

References: All publications cited in the text should be presented in the form of a list of references arranged alphabetically according to authors' surnames. Don't give serial numbers. Use the following system for arranging the references.

For periodicals: name(s) and initials of author(s) year of publication, title of the paper, abbreviated title of the journal (in conformity with the World list of Periodicals), volume number (bold), colen, first and last page numbers.

a. For periodicals:

Bartley, E.E., Wheatcroft, K.L., Claydon, T.J. Fountaine, F.C. and Parrish, D.V. 1951. Effect of feeding aureomycin to dairy calves. *J. Anim. Sci.* **10**: 1036-1038.

b. For books:

Snedecor, G.W. and Cochran, W.G. 1994. *Statistical Methods.* VIII edn. Iowa State University Press, Iowa, USA, pp. 287-192.

c. For chapter in a book:

Thomas, J.R. and Charles, C.C. 1997. Calcium regulating hormones and diseases of abnormal mineral metabolism. In: *Clinical Biochemistry of Domestic Animals*. Kaneko, J.J., Harvey, J.W. and Bruss, M.L. (eds) V. edn. Academic Press, London, pp. 619-702.

d. For thesis:

Singh, S.K. 1998. Studies on clinico-biochemical changes in Downer cow syndrome. M.V.Sc. thesis, Punjab Agriculture University, Ludhiana, India.

e. For proceedings of symposia/conference:

Shah, R.L., Kataria, J.M., Arya, S.C. and Verma, K.C. 1996. Study on inclusion body hepatitis in broiler chicks. *Proc. XX World Poult. Congress* held on Sept. 2-5, 1996, New Delhi, Vol. IV, pp. 313-314.

Tables: These should be as few as possible and typed on separate sheets and numbered in roman numerical. Each table should have a brief and self-explanatory title.

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Body weight	b wt	Litre	l	Calory	cal
Meter	m	Centimeter	cm	Microlitre	ì l
Counts per minute	cpm	Millligram	mg	Cubic centimeter	cm^3
Millilitre	ml	Degree centigrade	^0C	Minute(s)	min
Degree Fahrenheit	^0F	Once a day	od	Decilitre	dl
Parts per million	ppm	Gram	g	Percent	%
Hour(s)	hr	Picogram	pg	Inch	in
Revolution per min	rpm	Intramuscular	im	Second(s)	sec
Intraperitoneal	ip	Square centimeter	cm^2	Intravenous	iv
Subcutaneous	sc	Kilo calories	kcal	Thrice a day	tid
Kilogram	Kg	Year(s)	yr	Twice a day	bid
Volts	V				

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