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Profile of some marker enzymes and other biochemical constituents in the blood plasma of buffaloes during reticuloruminal impaction

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Reticuloruminal impaction (RRI), one of the most common digestive disorders of ruminants, is characterized by failure of digestion, slow passage or accumulation of feed in one or more stomach compartments leading to constipation, progressive emaciation and dehydration. Toxic substances/ metabolites that are produced in rumen get absorbed in circulation and subsequently their detoxification occurs in liver. Excessive production of toxic substances in rumen and other parts of digestive tract may cause overtaxation of liver. It is therefore imperative to elucidate the impact of reticuloruminal impaction on functional, metabolic and enzymatic activities of liver. Hence the present study was undertaken to monitor the activities of some liver specific enzymes and other biochemical constituents in blood plasma of buffaloes suffering from RRI.

Normal buffaloes (12) and buffaloes (12) with RRI were selected for this study from the dairy farm of the university and from the cases presented at the university clinical services complex respectively. The animals were divided into 2 groups: group 1: control, and group 2: buffaloes with RRI. Blood samples, collected in heparinized vials, were centrifuged in a cold centrifuge at 3 000 rpm for 15 min to separate plasma that was stored in a deep freezer at –20°C.

The activities of plasma arginase, sorbitol dehydrogenase (SDH), glutamate dehydrogenase (GDH) and aspartate amino transferase (AST) were estimated by the methods of Mia and Koger (1978), Gerlich (1983), Bulen (1956) and Reitman and Frankel (1957) respectively. Plasma levels of bilirubin and gamma glutamyl transferase (GGT) were determined by chemistry analyzer using autopak kits. Blood urea nitrogen (BUN) and plasma creatinine were estimated as per Wooton (1964) and Hawk *et al.* (1954) respectively. The data were subjected to student's t-test analysis (Snedecor and Cochran 1994).

The mean values of body temperature for groups 1 and 2 were 100.60±0.36°F and 102.30±0.20°F respectively. There was a nonsignificant increase in body temperature in the animals with RRI. These results were in agreement with Rao (1987).

The means of rumen motility were 3.00±0.15 and 0.55±0.12 per min for groups 1 and 2 respectively. This significant decline in rumen motility was in accordance with the findings of Singh *et al.* (1990).

Rumen pH in buffaloes with RRI varied within the normal limits i.e. 6.8 to 7.5 with the mean value of 7.2±0.08. This is in agreement with Singh *et al.* (1997).

AST: The mean plasma AST activity (Table 1) in control group was 116.27±6.88 IU/L which was in harmony with the mean plasma AST activity of 116.0±5.2 IU/L in buffaloes (Aujla 1992). Buffaloes suffering from RRI had a significant higher AST activity (272.33±41.46 IU/L) than control group. Similar findings were observed by Belluzi *et al.* (1979) in ruminal dysfunction. This increase could be due to affection of hepatic parenchyma and rumen musculature or as result of necrosis of liver because of toxemia (Dirksen 1970) or due to degenerative lesions in liver (Kaneko *et al.* 1997).

SDH and GDH: The mean plasma SDH activity was 2.43±0.30 U/L that was comparable to that reported in cattle and sheep (Shaw 1974). The enzyme level was 9.29±0.74 U/L in buffaloes with RRI, which was significantly higher ($P<0.01$) as compared to control animals. The mean plasma GDH activity of 3.71±0.45 U/L in control group was in agreement with the findings of Boyd (1962) but lower than that reported by Mullen (1976). The GDH level in group 2 was 15.05±1.22 U/L that was significantly higher ($P<0.01$) as compared to control group.

There is a paucity of literature to reveal relationship of

Table 1. Plasma enzyme activities (IU/L) in normal buffaloes and buffaloes with reticuloruminal impaction

Enzyme	Group 1	Group 2
AST	116.27±6.88	272.33±41.46**
SDH	2.43±0.30	9.29±0.74**
GDH	3.71±0.45	15.05±1.22**
GGT	16.67±2.17	19.33±3.06
Arginase	9.70±1.16	24.50±2.94**

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SDH and GDH activities with RRI or other ruminal disorders. However, SDH is a liver specific enzyme and normally it is low to negligible in plasma (Shaw 1974). Increase in plasma SDH activity is correlated to hepatic dysfunctions/damage (Shaw 1974, Alemn *et al.* 1977). Hoe and Wilkinson (1973) emphasized on estimation of GDH as a liver function test in large animals. This was supported by Mullen (1976) who stated that GDH was associated with microsomal fraction of hepatocytes and was released following acute liver damage.

GGT: The mean plasma GGT level was 16.67±2.17 IU/L in group 1 which was in harmony with the values reported by Aujla (1992). The mean activity of GGT in group 2 was 19.33±3.06 IU/L that was nonsignificantly higher as compared to control group level. The GGT assay seems to be a straight forward way to distinguish between hepatocellular and obstructive disease (Righetti and Kaplam 1972). It offers high specificity in suspected extrahepatic obstruction. If majority of GGT is not associated with liver parenchymal cell, but rather with the bile duct epithelium then there may not be a large increase in GGT activity as was with RRI induced hepatic dysfunction in the present study.

Arginase: The mean plasma arginase activity for normal buffaloes was 9.70±1.16 U/L, which was comparable to 8.54±0.77 U/L in goats (Jain *et al.* 1995). Buffaloes with RRI had a significant higher ($P<0.01$) enzyme activity as compared to control group (Table 1). Plasma arginase is an indicator of active liver cell damage in ruminants (Harvey and Obeid 1974). Randhawa *et al.* (1989) also revealed higher arginase activity in the blood of acidotic buffalo calves due to hepatic damage, which they confirmed by histopathological examination of liver in their study.

BUN: The mean value of BUN in normal buffaloes was 28.58±3.03 mg/dl, which was non significantly lower than that of control group (Table 2). Similar findings were reported by Panciera *et al.* (1990) and Singh *et al.* (1990) in their studies on ruminal derangement.

During ruminal dysfunctions, tissue extract and putrefied ingesta liberated toxic amines such as histamine in rumen, which on absorption in system increase BUN level (Dain *et al.* 1995). There is also failure of urea recycling process and it is not properly utilized by rumen microbes. These factors might be responsible for increased level of BUN in the present study.

Creatinine: The mean concentration of creatinine in group 1 was 1.33±0.12 mg/dl. For group 2, the mean value was 2.08±0.11 mg/dl that was significantly higher than that of control group. Similar findings were reported by Panciera *et al.* (1990) and Singh *et al.* (1990) during ruminal disorders in bovines. Elevated plasma level of creatinine is indicative of renal damage. Excessive feeding of wheat straw, which is a common cause of reticuloruminal impaction, can lead to renal insufficiency due to formation of oxalate crystals. This might be the cause of increased plasma creatinine level.

Table 2. Concentrations (mg/dl) of BUN, creatinine and bilirubin in normal buffaloes and buffaloes with reticuloruminal impaction

Parameter	Group 1	Group 2
BUN	28.58±3.03	35.37±4.73
Creatinine	1.33±0.12	2.08±0.11**
Bilirubin	0.24±0.03	1.05±0.07**

Mean values significantly different (*$P<0.01$) from control.

Bilirubin: The mean level of bilirubin for control group was 1.24±0.03 mg/dl, which was within the normal range as reported by Kaneko *et al.* (1997). Buffaloes suffering from RRI had significantly higher ($P<0.01$) value of bilirubin. This was in accordance with the findings of Pienkowski (1969) which stated that during ruminal dysfunctions there was absorption of toxic products from rumen or elementary tract leading to cellular disturbance of liver parenchyma (Pienkowski 1969). Gopinath and Ford (1970) also considered bilirubin to be a valuable index of acute hepatic damage.

Therefore, a significant increase in the activities of liver specific enzymes, viz. AST, SDH, GGT and arginase in plasma and creatinine and bilirubin concentrations points towards hepatic and renal dysfunctions during RRI in buffaloes.

SUMMARY

A study was conducted to monitor the activities of some liver specific enzymes and other biochemical constituents in blood plasma during reticuloruminal impaction (RRI) in buffaloes. Twenty-four buffaloes were divided into 2 groups of 12 each. Group 1: control; group 2: buffaloes with RRI. There was no significant difference in the gamma glutamyl transferase (GGT) activity and blood urea nitrogen (BUN) level of both groups. The plasma activities (IU/L) of aspartate amino transferase (AST), sorbitol dehydrogenase (SDH), glutamate dehydrogenase (GDH) and arginase were 272.33±41.46, 9.29±0.74, 15.05±1.22 and 24.50±2.94, respectively, in buffaloes with RRI; which were significantly higher than those of control group. A significant rise in plasma creatinine and bilirubin levels (mg/dl) was also observed in the diseased buffaloes. These changes were indicative of severe hepatic and renal dysfunctions during reticuloruminal impaction in buffaloes.

REFERENCES

Alemn P, Forsyth G W and Searcy G P. 1977. A comparison of parameters used to assess liver damage in sheep treated with carbon tetrachloride. *Canadian Journal of Comparative Medicine* **41**: 420–27.

Aujla T K. 1992. 'Clinico-pathological studies in some buffalo diseases'. M.V.Sc. Thesis, P. A. U., Ludhiana, Punjab (India).

Belluzi G C, Sabatim P, Corbella E and Ubaldi A. 1979. Changes in alanine and aspartate amino transferase in cattle with metabolic acidosis and metabolic alkalosis. *Veterinary Bulletin: Abstract*

7485.

Boyd J W. 1962. The comparative activity of some enzymes in sheep, cattle and rats; normal serum level and changes during experimental liver necrosis. *Research in Veterinary Science* **3**: 256–68.

Bulen W A. 1956. The isolation and characterization of glutamate dehydrogenase from corn leaves. *Archives of Biochemistry and Biophysics* **62**: 173–83.

Dain T A, Neal A L and Dougherty R W. 1995. The occurrence of histamine and tyramine in ruminal ingesta of experimentally overfed sheep. *Journal of Animal Sciences* **14**: 930–55.

Dirksen G. 1970. Acidosis. *Proceedings of the 3rd International Symposium on the Physiology of Digestion and Metabolism in the Ruminant.* Phillipson A T (Ed.) pp. 612–25.

Gerlich U. 1983. *Methods of Enzymatic Analysis.* 3rd edn, Vol. 3, pp. 112–17. Verlay Chemic, Deerfield Beach, Florida, USA.

Gopinath C and Ford E J H. 1970. The effect of napthyl isothiocyanate on the liver of sheep and calves. *Journal of Pathology* **100**: 269–80.

Harvey D G and Obeid H M A. 1974. The application of certain liver function tests including serum alkaline phosphatase estimations to domesticated animals in Sudan. *British Veterinary Journal* **130**: 544–55.

Hawk P B, Oser B L and Summeson W H. 1954. *Practical Physiological Chemistry.* pp. 555. McGraw-Hill Book Co., London.

Hoe C M and Wilkinson J S. 1973. Liver function: A review. *Australian Veterinary Journal* **49**: 490–501.

Jain A, Dhanotia R S and Dixit N K. 1995. Activity of certain serum enzymes in normal goats. *Indian Veterinary Journal* **72**: 583–85.

Kaneko J J, Harvey J W and Bruss M L. 1977. *Clinical Biochemistry of Domestic Animals.* 5th edn, pp. 120–38. Academic Press, New York, U. S.

Mia A S and Koger H D. 1978. Direct colorimeteric determination of serum arginase in various domestic animals. *American Journal of Veterinary Research* **39**: 1381–83.

Mullen P A. 1976. The diagnosis of liver dysfunctions in farm animals and horses. *Veterinary Record* **99**: 330–34.

Panciera R J, Martin T, Burrows G E, Taylor D S and Ricel L E. 1990. Acute oxalate poisoning attributed to ingestion of curly dock (*Rumex crispus*) in sheep. *Journal of American Veterinary Medical Association* **196**: 1980–84.

Pienkowski M. 1969. Estimation of the functional state of liver in cows with acid indigestion. Annals Universitatis. *Marial Curie-Sklodowska, Lublin Polonia, Sectio dd* Vol. **24(18)**: 209–22.

Randhawa S S, Gupta P P, Roy K S, Ahuja A K and Rathor S S. 1989. Biochemical, pathological and histoenzymological studies on experimental acute lactic acidosis in buffalo calves. *Buffalo Journal* **5**: 13–24.

Rao T D S. 1987. Importance of impaction of rumen in bovines. *Livestock Adviser* **11**: 25–27.

Reitman S and Frankel S. 1957. *Gradwohl's Clinical Laboratory Methods and Diagnosis.* 7th edn, Vol. I. C. V. Morby Co., St. Louis, U.S.A.

Righetti A A B and Kaplan M M. 1972. Disparate responses of serum and hepatic alkaline phosphatase and 5'nucleotidase to bile duct observation in the rat. *Gastroenterology* **62**: 1034–39.

Snedecor G W and Cochran W G. 1994. *Statistical Methods.* 8th edn. Oxford and IBH publishing Co., New Delhi.

Shaw F D. 1974. Sorbitol dehydrogenase in diagnosis of liver diseases of ruminants. *Australian Veterinary Journal* **50**: 277–78.

Singh J, Gill B P and Kwatra M S. 1990. Napier grass (*Pennisetum purpureum*) poisoning in bovines and its treatment. *Indian Journal of Veterinary Medicine* **10**: 142–44.

Singh N, Kumari R and Akbar M A. 1997. Effects of tranquilizers on biogenic amines, volatile fatty acids and milk production in clinical cases of primary indigestion in buffaloes. *Indian Veterinary Journal* **74**: 590–93.

Wooton I D P. 1964. *Microanalysis in Medical Biochemistry.* 4th edn. J & A Churchill Ltd, 1047, Gloucester palace, London-W1.