

Studies on Changes in Some of Blood Constituents of Adult Cross-bred Cattle Fed Different Levels of Extracted Rice Bran

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Abstract: Feeding of two levels of extracted rice bran (ERB) along with wheat straw was studied in 6 adult crossbred cattle. After 121 days feeding of restricted - ERB the first phase of blood sample was collected from all the animals. Subsequently the animals were shifted to *ad lib.* feeding of ERB and the second phase of blood sample was collected after 42 days. The blood samples were analysed for biochemical constituents like haemoglobin, glucose, protein, albumin, globulin, inorganic phosphorus, AST (GOT), ALT(GPT), ACP, ALP, cholesterol, creatinine and urea nitrogen. The mean values of the blood biochemical constituents in the same order were 11.10 ± 0.17 and 10.41 ± 0.16 g/100ml., 56.99 ± 0.48 and 62.70 ± 0.97 mg/100ml., 7.20 ± 0.10 and 7.80 ± 0.20 g/100ml., 2.63 ± 0.07 and 2.07 ± 0.06 g/100ml., 4.52 ± 0.15 and 5.65 ± 0.23 g/100ml., 5.81 ± 0.08 and 7.02 ± 0.07 mg/100ml., 52.02 ± 1.93 and 59.38 ± 1.23 i.u., 20.63 ± 0.61 and 27.99 ± 1.34 i.u., 40.80 ± 4.61 and 56.53 ± 5.67 i.u., 8.03 ± 0.72 and 11.78 ± 0.77 i.u., 94.04 ± 2.36 and 79.24 ± 1.59 mg/100ml., 2.00 ± 0.17 and 1.00 ± 0.03 mg/100ml., and 15.02 ± 0.67 and 33.14 ± 0.67 mg/100ml. for restricted - and *ad lib.*-ERB fed animals respectively. All the mean values were differed significantly due to level of bran feeding.

Key words: Crossbred cattle, extracted rice bran, nutrient intake, enzymes, blood biochemical constituents

Introduction

Nutritional imbalances and excesses of farm animals are highly correlated with the turnover of fluids, salts and soluble organic materials in the system which reflected upon the secretions and excretions from the body and internal circulatory medium. Blood is a fluid connective tissue that is circulated throughout the vascular system carrying the vital factors to all cells in the body and serves as a principal transport medium for nutrients which also receives the waste products of nutrient metabolism from the tissue. Thus, blood biochemical screening in the nutritional research can act as an essential supportive clue for more accurate and reliable diagnosis of various physiological disorders and these pictures should be comprehensively interpreted by correlating with other nutritional parameters. Any disturbance on basic physiological process of the animal by dietary changes may lead to a chain reaction resulting in disruption of normal physiological activity and subsequently may also change the biochemical constituents of the body fluid (Sharma, 1973). In the present study, therefore, feeding of extracted rice bran at 2 levels (restricted and *ad libitum*) on the changes of blood biochemical profile was assessed.

Materials and Methods

Housing and Feeding: Healthy, 6 adult cross bred cattle of about 6½ years of age are freely given wheat straw (WS) as a basal diet with restricted amount of extracted rice bran to meet maintenance requirement of crude protein (CP) according to NRC (1989). After 121 days of experimental feeding trial the first phase of blood sample was collected from all the animals. Subsequently all the experimental animals were shifted to *ad libitum* feeding of ERB along with restricted amount of WS and the second phase of blood samples were collected after 42 days of feeding trial. In addition during the experimental feeding periods 1Kg of available green fodder was also offered as a carotene supplement to each animal throughout the experimental period. Clean drinking water was offered twice a day to all animals under uniform managerial conditions. The animals prior to experimentation were vaccinated for all common epidemic viral and bacterial diseases and twice dewormed against the helminths (albendazole @ 7.5 mg/kg B.W.).

Blood Collection: From all the experimental animals, the blood samples were collected by venipuncturing from jugular vein with a sterile, 16 gauge dry needle and clean, prepackaged plastic

Table 1: Chemical composition of experimental feeds on % DM basis

Attributes	ERB	WS
Organic matter (OM)	84.46	90.15
Crude protein (CP)	16.65	3.64
Ether extract (EE)	0.81	1.16
Total carbohydrates (TCHO)	67.00	85.35
Neutral detergent fibre (NDF)	52.54	74.66
Acid detergent fibre (ADF)	26.08	50.38
Hemi cellulose (HC)	26.48	24.28
Total ash (TA)	15.54	9.85
Acid insoluble ash (AIA)	5.74	5.81
Phosphorus (P)	1.21	0.12

ERB : Extracted rice bran,

WS : Wheat straw.

syringe in the morning prior to feeding and watering. The blood samples were collected in centrifuge tube containing an anticoagulant (disodium EDTA and sodium fluoride; Prasad 1992). It was then centrifuged at 3000 rpm of 15 minutes to separate the plasma. The separated plasma was then transferred into previously numbered plastic vials with the help of automatic microlit pipette capped and preserved at $\pm 20^{\circ}\text{C}$ for further analysis. About 2 ml of anticoagulated whole blood sample was also preserved for haemoglobin estimation. Though salt-EDTA is the anticoagulant of best the choice for most haematological procedures which caused shrinkage of RBC and highly interfered with determination of non protein nitrogen, urea-N, creatinine, alkaline phosphatase (Fraser, 1993). For serum separation, about 15 ml collected whole blood in the centrifuge tube was allowed to clot for about 30 minutes then rimmed with an applicator stick and centrifuged exactly at 2000 rpm for 10 minutes. The supernatant serum was carefully aspirated and placed in the clean & labeled plastic vial and preserved in deep freeze for further assay.

Analysis: The chemical composition of extracted rice bran and wheat straw samples was determined as per the methods detailed in AOAC (1990) and the fibre fractions viz. neutral detergent fibre (NDF), acid detergent fibre (ADF) and hemicellulose (HC) as outlined by Van Soest *et al.* (1991). The blood samples were estimated for blood biochemical constituents like AST and ALT, ALP, ACP glucose, haemoglobin, total protein, albumin, inorganic

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Table 2: Mean body weight and nutrient intake of experimental animals at different levels of ERB feeding

Attributes	ERB-restricted	ERB <i>ad-libitum</i>
Body weight (BW)		
Mean BW (kg)	370.12 ± 20.41	375.88 ± 21.03
Metabolic body size (KgW ^{0.75})	84.31 ± 3.52	85.29 ± 3.61
Dry matter intake (DMI)		
Wheat starw, (kg/d)**	3.91 ± 0.11 ^b	1.58 ± 0.05 ^a
ERB, (kg/d)**	2.07 ± 0.09 ^b	9.94 ± 0.30 ^a
Total DMI, (kg/d)**	5.98 ± 0.17 ^b	11.52 ± 0.35 ^a
% of BW**	1.62 ± 0.05 ^b	3.08 ± 0.10 ^a
g/KgW ^{0.75} /d**	70.99 ± 1.41 ^b	135.36 ± 2.49 ^a
Straw : ERB ratio**	65.30 : 34.70 ^a (± 0.75)	13.75 : 86.25 ^b (± 0.85)
Crude protein intake (CPI)		
Total CPI, (g/d)**	486.99 ± 16.98 ^b	1723.36 ± 45.02 ^a
g/100kg BW/d**	132.05 ± 2.91 ^b	461.16 ± 16.51 ^a
g/KgW ^{0.75} /d**	5.78 ± 0.08 ^b	20.26 ± 0.45 ^a
Organic matter intake (OMI)		
Total OMI, (g/d)**	5270.27 ± 147.82 ^b	9813.78 ± 293.73 ^a
Ether extract intake (EEI)		
Total EEI, (g/d)**	63.27 ± 1.66 ^b	91.23 ± 2.86 ^a
Carbohydrate intake (CHOI)		
Total CHOI, (g/d)**	4720.15 ± 130.56 ^b	7999.12 ± 248.18 ^a
Neutral detergent fibre intake (NDFI)		
Total NDFI, (g/d)**	3999.23 ± 110.85 ^b	6716.30 ± 219.44 ^a
Acid detergent fibre intake (ADFI)		
Total ADFI, (g/d)**	2486.48 ± 69.30 ^b	3413.81 ± 126.74 ^a
Hemi cellulose intake (HCI)		
Total HCI, (g/d)**	1503.64 ± 42.20 ^b	3289.20 ± 90.45 ^a

Mean values bearing different superscripts in a row differ significantly. ** P<0.01 (highly significant effect); Non significant effect (P<0.05).

phosphorus, cholesterol, creatinine, urea-N as per the method described by Reitman and Frankel (1957), Kind and king (1954), Bergmeyer (1984), Cooper and Mc Daniel (1970), Wong (1928), Hiller *et al.* (1927), Gustafsson (1976), Fiske and Subba Rao (1925), Zlatkis *et al.* (1953), Wootton (1974), Rahmatullah and Boyde (1980) respectively. The data pertaining to dry matter intake, nutrient intake and blood biochemical profile was analysed for variance and tested for statistical significance employing Duncan Multiple Range Test (Snedecor and Cochran, 1994).

Results and Discussion

The chemical composition of feeds offered to the experimental animals under two different feeding systems is given in Table 1. The proximate compositions and the fibre fraction of extracted rice bran (ERB) and wheat straw (WS) were within the normal range of common values for these feeds (Ranjhan, 1991; Prakash and Ramanathan, 1995). Average metabolic body size of the animals was 84.31 ± 3.52kg and 85.29 ± 3.61kg in restricted- and *ad lib.*-ERB fed groups respectively which showed a little fluctuation but statistically non significant effect between the groups. Such fluctuation in the mean body weight was due to shifting of animals from restricted to *ad lib.* - ERB feeding. The data on voluntary dry matter intake indicated that DMI was affected significantly by the dietary treatments. The ratio of extracted rice bran in the ration was about 35% and that of wheat straw 65% in restricted ERB fed animals but the corresponding value of the ratio in *ad lib.* ERB fed animals was 86% and 14% respectively, being significantly (P<0.01) different between the treatments. This indicated high palatability extracted rice bran being higher intake during *ad lib.*-ERB feeding. The intake of DM was similar to that recorded by Kishan *et al.* (1991) on feeding of high level of brans in the diet of adult buffaloes. The mean body weight and, DMI and nutrient intake per unit of live weight are presented in Table 2. The average nutrient intake of crude protein was 132.05 ± 2.91 g and 461.16 ± 16.51 g per 100 kg B.W. equivalent to 5.78 ± 0.08 g and 20.26 ± 0.45 g per kg metabolic body size of restricted and *ad lib.* -ERB fed animals respectively, which revealed statistically significant (P<0.01) between the dietary treatments. In the present study, the nutrients intake of

various proximate principles and fibre fractions like CHO, OM, EE, NDF, ADF and HC were significantly higher in *ad libitum* -ERB fed animals than that of restricted -ERB fed animals and this was in agreement to the previous reported values (Moran, 1983; Weston, 1985).

The mean values of haemoglobin level in blood observed some variation during the experimentation of dietary treatments. The difference between the ERB-restricted and *ad libitum* feeding of animals was statistically significant (P<0.05) but the values in both groups were within the normal range (Greatorex, 1957). The mean concentration of glucose content in the blood plasma was 56.99 ± 0.48 and 62.70 ± 0.97 mg/100ml for ERB -restricted and *ad lib.* feeding animal groups respectively, and comparable between the treatment groups which evinced significant increase in *ad lib.* feeding of ERB group, though the values were within the upper normal limit reported by Boyd (1984). This was due to higher niacin intake in ERB *ad lib.* group which results in increased level of glucose and protein (Horner *et al.*, 1989). The activity of enzyme transaminase and phosphatase gradually increased from restricted to *ad lib.* feeding of ERB group of animals. Similar finding was also reported by Prasad *et al.* (1990). The pattern of changes in the value of aspartate amino transferase (AST), alanine amino transferase (ALT), alkaline phosphatase (ALP) and acid phosphatase (ACP) may be considered normal and little relationship with the health of the animal particularly liver and skeleton which was similar to the reported by Kaneko (1997). The average value of total protein concentration in the blood plasma revealed a little increase during the *ad lib.* -ERB feeding experiment and the increase was statistically significantly higher (P<0.05) in *ad lib.* - ERB fed group. The values of the total protein in blood plasma of the adult crossbred cattle during the first phase as well as second phase of experiment were found to be within the range of reported values (Jain, 1996). There was a sharp decrease (P<0.01) in the plasma globulin concentration (4.52 ± 0.15g/100ml) in ERB -restricted fed group with an increase in albumin/globulin ratio (0.59 ± 0.04) than that the value of ERB -*ad lib.* fed group i.e. 5.65 ± 0.23g/100ml and 0.37 ± 0.03. The mean value of plasma albumin was dropped in *ad lib.* -ERB fed animals which showed a little lower from the normal range recommended by Boyd (1984).

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Table 3 : Mean values of blood biochemical constituents of experimental animals at different levels of ERB feeding

Attributes	ERB-restricted	ERB- <i>ad libitum</i>
Blood haemoglobin, (g/100ml)*	11.10±0.17 ^a	10.41±0.16 ^b
Plasma glucose, (mg/100ml)**	56.99±0.48 ^b	62.70±0.97 ^a
Serum - AST (GOT), (i.u)**	52.02±1.93 ^b	59.38±1.23 ^a
Serum - ALT (GPT), (i.u)**	20.63±0.61 ^b	27.99±1.34 ^a
Serum - ACP, (i.u)**	40.80±4.61 ^b	56.53±5.67 ^a
Serum - ALP, (i.u)**	8.03±0.72 ^b	11.78±0.77 ^a
Plasma total protein, (g/100ml)*	7.20±1.10 ^b	7.80±0.20 ^a
Plasma albumin, (g/100ml)**	2.63±0.07 ^a	2.07±0.06 ^b
Plasma globulin, (g/100ml)**	4.52±0.15 ^b	5.65±0.23 ^a
Albumin-globulin ratio**	0.59±0.04 ^a	0.37±0.03 ^b
Plasma inorganic phosphorus, (mg/100ml)**	5.81±0.08 ^b	7.02±0.07 ^a
Serum cholesterol, (mg/100ml)**	94.04±2.36 ^a	79.24±1.59 ^b
Serum creatinine, (mg/100ml)**	2.00±0.17 ^a	1.00±0.03 ^b
Serum urea nitrogen, (mg/100ml)**	15.02±0.67 ^b	33.14±0.67 ^a

AST : Aspartate amino transferase or, GOT : Glutamic oxalo acetic transaminase

ALT : Alanine amino transferase or, GPT : Glutamic pyruvic transaminase

ALP : Alkaline phosphatase, ACP : Acid phosphatase.

Mean values bearing different superscripts in a row differ significantly. * P<0.05 (significant effect), ** P<0.01 (highly significant effect).

Hypo-albuminemia is a common phenomenon in the farm animals which may be a result of malnutrition, maldigestion or malabsorption (Johnson *et al.*, 1982). However, no such problem was observed in the present study. This aspect requires further study for finding out the reasons for successful variation. From the perusal of the values of plasma inorganic phosphorus concentration in Table 3, it may be seen that it was statistically significantly higher (P<0.01) in ERB -*ad lib.* fed animals than that of restricted -ERB fed animals. This was on account of higher intake of available phosphorus content in the bran (Moran, 1983). However, both the mean values were within the range of values reported by Singh *et al.* (1990). Mean concentration of cholesterol in blood serum was dropped abruptly in ERB -*ad lib.* fed animals. The difference was found significant (P<0.01). This was due to the higher intake of fibrous diets which are responsible for lowering the cholesterol content in serum (Husseini *et al.*, 1976). In the *ad lib.* -ERB fed animals, the level of serum creatinine sharply decreased and differed significantly (P<0.01) from the value of restricted-ERB fed animals. The reason for decreasing in creatinine level (2.00 v/s 1.00 mg/100ml) on *ad lib.* feeding of ERB fed adult crossbred cattle might have inhibited to the body creatinine production (Walker, 1960). Mean serum urea nitrogen concentration was, in different levels of ERB fed animals, within the lower and upper limit recorded by Boyd (1984). Higher blood urea or NH₃ concentration was considered in incidences of urinary obstruction or hepatic dysfunction (Kaneko, 1997), which was not evident from urination pattern of animals. Unfortunately its related attributes like volume of urine output, specific gravity, pH etc. could not be noted to arrive at a possible conclusion. Urea-N normally increase with the increasing intake of protein and non protein nitrogen (Huber *et al.*, 1976). During the second phase (*ad lib.*) of ERB feeding the level of mean blood urea-N increased dramatically which showed significantly (P<0.01) different. Almost doubling of blood urea-N level (15.02 v/s 32.14mg/100ml) is an indicator of excess protein intake associated with less energy intake due to which ammonia (NH₃) released in rumen fermentation could not be efficiently utilized by the rumen microorganisms. It may be inferred from this study that feeding of different levels of extracted rice bran in adult cross-bred cattle has changed in the blood biochemical constituents.

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References

AOAC., 1990. Official Methods of Analysis, 15th revised edition, Association of Official Analytical Chemists. Washington, D.C., 20044, USA.

- Bergmeyer, H.U.,1984. Estimation of serum acid phosphatase activity by colorimetric method. Method of Enzymatic Analysis, 3rd revised edition., Weinheim, Academic Press. Inc., New York.
- Boyd. J.W., 1984. The interpretation of serum biochemistry test results in domestic animals. Veterinary Clinical Pathology, Veterinary Practice. Publishing Co. Volume XIII, No. II.
- Cooper, G.R. and V. Mc Daniel, 1970. Estimation of glucose by colorimetric method. Standard Methods of Clinical Chemistry, p: 159.
- Fiske, C.H. and Y. Subba Rao, 1925. The colorimetric determination of phosphorus. J. Biol. Chem., 66 : 375-380.
- Fraser, E.J., 1993. Cited from the Merck Veterinary Manual, 7th revised edition., Published by Merck and Co., Inc., Rahway. New Jersey, U.S.A.
- Greatorex, J.C., 1957. Observation on the haematology of calves and various breeds of adult dairy cattle. Br. Vet. J., 113 : 29-35.
- Gustafsson, Jan, E.C., 1976. Improved specificity of serum albumin determination and estimation of acute phase reactants by use of bromocresol green reaction. Clin. Chem., 22 : 616-622.
- Hiller. Mc Intosh and Van Slyke, 1927. J. Clin. Invest., 4 : 325. Cited in Practical Physiological Chemistry. 13th edition. By Philip, B., Hawk, Bernard, L. Oser and William, H. Summerson, p: 925.
- Horner, J.L, C.E. Coppock, G.T. Schelling, J.M. La Bore and O.H. Nave, 1989. Influence of niacin and whole cotton seed on intake, milk yield and composition, and systemic responses of dairy cows. J. Dairy Sci., 69 : 3087-3093.
- Huber, J.T., R.L. Boman and H.E. Henderson, 1976. Fermented ammoniated condensed whey as a nitrogen supplement for lactating cows, J. Dairy Sci., 59 : 1936-1940.
- Husseini, M.D., M.F. Krueger, R.C. Fanguy and J.W. Bradley, 1976. Blood serum and egg yolk cholesterol in hens as influenced by wheat middlings and oats in the diet. Poult. Sci., 55 : 1595-1598.
- Jain, N.C., 1996. In : Schalm's Veterinary Hematology. 5th revised edition, Lea and Febiger, Philadelphia, USA.
- Johnson, G.F., D.A. Zawie, S.R. Gilbertson and I. Sternlieb, 1982. Chronic active hepatitis in Doberman Pinschers. J. Am. Vet. Med. Assoc., 180 : 1438-1442.
- Kaneko, Jiro. J., 1997. Clinical biochemistry of domestic animals, 5th revised edition., Academic Press, Inc., New York, USA.
- Kind, P.R.M. and E.J. King, 1954. Estimation of serum alkaline phosphatase activity by colorimetric method. J. Clin. Path., 7 : 322-327.

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- Kishan, J.M., Putan Singh and M.Y. Khan, 1991. Nutritive value of some economic rations for buffaloes. *Indian J. Anim. Nutr.*, 8 : 19-22.
- Moran, J.B., 1983. Rice bran as a supplement to elephant grass for cattle and buffalo in Indonesia. *J. Agri. Sci., (Cambridge)*. 100 : 709-716.
- NRC., 1989. National Research Council, Nutrient requirements of domestic animals. Nutrient requirement of dairy cattle. 6th revised edition. National Academy of Science, Washington, DC.
- Prakash, J. and G. Ramanathan, 1995. Proximate composition and protein quality of stabilized rice bran. *J. Food Sci. and Tech.*, 32 : 416-419.
- Prasad, B., 1992. *Veterinary Clinical Diagnostic Technology*, First edition, CBS, Publishers and Distributors, New Delhi, India.
- Prasad, C.S., K.T. Sampath and S.R. Sampath, 1990. Effect of feeding acid stabilized deoiled rice bran on the liver function and histopathological changes of various organs in cross bred calves. *Indian J. Anim. Sci.*, 60 : 1244-1245.
- Rahmatullah, M. and T.R.C. Boyde, 1980. An improvement in determination of urea using diacetyl-monoxime method with and without deproteinization. *Clinica Chemica Acta*. 107 : 3-9.
- Ranjhan, S.K., 1991. *Chemical Composition and Nutritive Value of Indian Feeds and Feeding of Farm Animals*, ICAR, Publication, New Delhi.
- Reitman, S. and S. Frankel, 1957. A colorimetric determination of serum glutamic oxalo-acetic and glutamic pyruvic transaminase. *Am. J. Clin. Path.*, 28 : 56-63.
- Sharma, G.N., 1973. Studies on changes in some of the important blood constituents of Tharparkar calves on two levels of protein feeding. *The Indian Vet. J.*, 50 : 569-577.
- Singh, H.P., A. Kumar and P.C. Chaudhuri, 1990. *Veterinary Clinical Guide*, 4th edn., Kalyani Publishers., New Delhi.
- Snedecor, G.W. and W.G. Cochran, 1994. *Statistical Methods*, 8th edn., Iowa State University Press, Ames, Iowa, U.S.A.
- Van Soest, P.J., J.B. Robertson and B.S. Lewis, 1991. Methods for dietary fibre, neutral detergent fibre and non starch polysaccharides in relation to animal nutrition. *J. Dairy Sci.*, 74 : 3583-3597.
- Walker, J.B., 1960. Metabolic control of creatine biosynthesis. Effect of dietary creatine. *J. Biol. Chem.*, 235 : 2357-2361.
- Weston, R.H., 1985. The regulation of feed intake in herbage-fed ruminants. *Proceeding of the Nutrition Society of Australia*, 10: 55-62.
- Wong, S.Y., 1928. Colorimetric determination of iron and haemoglobin in blood. *J. Biol. Chem.*, 77: 409-412.
- Wootton, I.D.P. 1974. Estimation of serum creatinine by colorimetric method. *Micro analysis in Medical Biochemistry*, 5th revised edn., Churchill Limited, Edinburgh, London.
- Zlatkis, A, Zak, B and Boyle, A.J. 1953. A new method for the direct determination of serum cholesterol. *J. Lab. and Clin. Med.*, 41:486-490.