NUTRITIONAL STATUS AND HEMATO-BIOCHEMICAL PROFILE OF ANOESTRUS BUFFALOES OF MALWA REGION OF MADHYA PRADESH

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ABSTRACT

In Malwa region of Madhya Pradesh reproductive failure (anoestrus) is a major problem in buffaloes under field conditions due to under feeding and non availability of balanced ration. To find out the nutritional causes behind anoestrus, thirty anoestrus buffaloes {10 heifers (average body wt. 262.80±22.51 Kg)+20 buffaloes (average body wt. 461±10.83 Kg, milk yield 7.62±0.48 litre/h/d)} were selected randomly that have normal genitalia from ten villages of Indore district. Average daily feed intake of each animal was recorded and proximate principles, major elements like Calcium (Ca) and Phosphorus (P) and trace elements like iron (Fe), zinc (Zn), manganese (Mn), copper (Cu) and cobalt (Co) in available feedstuffs were determined to find out nutrient availability. Deficiency of various nutrients was calculated by comparing with the standard requirements of the animals. Blood samples were also collected from the animals and analyzed for different haemato-biochemical constituents. The average values for different blood parameters in heifers and lactating buffaloes were hemoglobin (Hb) 10.32±0.19 and 11.15±0.32 g/dl; blood glucose, 50.62±1.80 and 59.29±1.05 mg/dl; plasma protein 5.06±0.19 and 6.33±0.18 g/dl, respectively. Major mineral profile in heifers and lactating buffaloes were Ca, 8.13±0.38 and 9.59±0.25 mg/dl; Inorganic-P (iP) 5.06±0.14 and 5.07±0.17 mg/dl; Total-P 24±2.21 and 17.50±1.42 mg/dl, respectively. Trace mineral levels in heifers and lactating buffaloes were Fe 22.05±0.58 and 17.68±0.84ppm; Zn 1.04±0.05 and 0.87±0.05 ppm; Cu 0.90±0.05 and 0.77±0.04 ppm; Mn 0.84±0.05 and 1.14±0.10 ppm; Co 1.92±0.41 and 1.13±0.09 ppm, respectively.

Results indicated that values of Hb, blood glucose, total protein, Ca, iP, Zn were marginally low but Cu was deficient in heifers, while levels of iP, Cu and Zn were marginally low in lactating buffaloes. Fe levels were found high in both heifers and lactating buffaloes. It may be concluded that dietary deficiency reflected the hemato-biochemical profile of anoestrus buffaloes and a strategic supplementation is needed to these animals for exploitation of their genetic potential for optimum production and reproduction.

Keywords: nutritional status, blood profile, anoestrus, buffalo, Malwa, Madhya Pradesh

INTRODUCTION

Reproductive failure of dairy animals is the major area of concern now days in all over the country, which causes a huge economical loss to the

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dairy owners. Among the various factors affecting it, nutrition is one of the most important factors which receive less attention than what actually it should be. For normal development and activity of reproductive organs, feeding of balanced ration is of utmost importance, because most field cases of reduced fertility or sterility are of nutritional origin. The interaction between nutrition and reproduction needs particular attention in our country, to overcome nutritional inadequacies either in terms of quantitative or qualitative nutrient deficiencies/ imbalances.

The nutritional deficiency causes several infertility conditions in buffaloes and the highest (50.26%) prevalence was observed for anoestrus, while cases of repeat breeder, metritis, pyometra and prolapse were only about (25.69%) among the common field cases Malwa region of Madhya Pradesh (Shukla *et al*., 2007). Thus anoestrus remains a major condition which constitutes about half of the reproductive problems occurring in buffaloes.

In Malwa region of Madhya Pradesh most of the farmers rear crossbred cattle and buffaloes as the dairy animal and anoestrus is the major reproductive problem among dairy animals. Farmers are following traditional feeding practices, usually cotton seed cake is the only concentrate source fed to lactating cattle along with straw (wheat/ masoor/ gram/ soybean) and mineral supplementation is a rare inhabitant among the farmers (Mudgal *et al*., 2003). Other factors associated with anestrous are energy deficiency (Ling *et al*., 2007) and minerals also play important role in the regulation of hormones and enzymes for initiation of estrus (Dhoble and Gupta, 1986). This nutritional deficiency also reflects the blood profile. So that blood profile of anoestrus buffaloes of Malwa region was evaluated in this experiment.

MATERIALS AND METHODS

Thirty buffaloes {10 heifers (average body wt. 262.80±22.51 Kg)+20 buffaloes (average body wt. 461±10.83 Kg, milk yield 7.62±0.48 litre/h/d)} with normal genitalia (by per rectal examination) and without any clinical infection, showing anoestrus were selected randomly from 10 villages (Borkhedi, Harsola, Kevti, Piplihamalhar, Umaria, Panda, Rau, Rangwasa, Sonvay and Bhaslai) around Veterinary College, Mhow of Indore District (from Malwa region of Madhya Pradesh). Body weights (Kg) of the animals were determined by recording the length (inch) and girth (inch) of each animal and then putting them in Shaeffer's formula (Sastry *et al*., 1982). Feed offered and residues left of each animal were weighed with the help of spring balance at both times (morning and evening) for three consecutive days. Then average feed intake of each animal was calculated. The representative samples of each feed were subjected to proximate analysis (AOAC, 1995), Ca and P (Talpatra *et al.*, 1940) content and trace mineral estimations by Atomic Absorption Spectrophotometer.

Milk yield (litre) of each lactating animal was measured during milking (morning and evening) for three consecutive days. After that average milk yield was calculated. Availability of DM, DCP, TDN, major (Ca and P) and trace elements (Fe, Cu, Mn, Zn and Co) for each animal was calculated on the basis of chemical composition of feedstuffs and their intake. Selenium, carotene and vitamin E intake were worked out using values given in the literature. Finally, the nutrient availability of individual animal was compared with the standard nutrient requirements calculated out for specific body weight and productivity of individual animal with the help of feeding standards (Kearl, 1982) and thus the deficiencies/ excess to specific nutrient

was worked out.

Blood samples of around 15 to 20 ml were collected from all 30 anoestrus buffaloes (heifer and lactating) from jugular vein in a sterilized plastic tubes containing anti-coagulant heparin solution (0.2 mg/ml of blood).

The tubes containing blood samples were kept in ice and brought to the laboratory. In laboratory blood was analyzed for hemoglobin (Oser, 1979), glucose (Folin and Wu, 1920) and total phosphorus (AOAC, 1995) and remaining blood was centrifuged for separating the plasma. The plasma sample then stored in glass vials for analysis of protein (Teitz, 1986), Ca (Ichaylova and Iikova, 1971), inorganic P (Fiske and Subbarow, 1925) and trace minerals (Fe, Zn, Cu, Mn and Co) using atomic absorption spectrophotometer [Perkin Elmer Aanalyst 100, USA] after wet digestion.

RESULTS AND DISCUSSION

The chemical composition of specific feed ingredients being consumed by the animals has been presented in Table 1. The feeds being offered to the heifers were mainly the agricultural by products including wheat straw, gram straw, masoor straw and soybean straw, while wheat bran, cotton seed cake or at some places concentrate mixtures were also being used additionally in the ration of lactating buffaloes.

In Table 2, the availability of different nutrients to the animals was worked out and compared with the standard requirement and hence the excess or deficiency of specific nutrient has been presented. As compared to the standard requirements (Kearl, 1982), availability of DM was about 4-6% less, which may be associated with the deficiency of TDN too. Similar findings was also been reported by Mudgal *et al.* (2003) and Tiwary *et al*., (2007). Between the groups deficiency of major nutrients was observed in heifers for digestible crude protein which was only about 48% to that of the requirements, while limited deficiency (11%) was seen in lactating buffaloes and which may be associated with the supply of concentrate to the lactating buffaloes. Similar observations were also recorded by earlier workers (Sinha, 1982; Sohal *et al*., 1982; Mudgal *et al*., 2003 and Tiwary *et al*., 2007). The lower levels of energy and /or protein may be associated with the ovarian inactivity and anoestrus (Wiltbank *et al*., 1965) as negative energy balance depresses the ovarian activity by inhibiting pulsatile LH release (Butler and Smith, 1989).

When the major minerals were compared it showed that Calcium was the element supplied in excess (33 to 283%) to the requirement and which may be due to supply of higher amounts of leguminous straws in their ration. This reduces the availability of phosphorus on one hand and over supply the calcium on other hand as leguminous straws having a wider Ca: P ratio and having a fair deficiency of element phosphorus. Phosphorus is necessary for normal energy and phospholipids metabolism as well as normal skeletal development and its severe deficiency may delay the onset of puberty and postpartum anoestrus and increased incidence of cystic follicles because of inactive ovaries, whereas moderate and low conception rates (Pugh *et al*., 1985). Many other workers also have found lower levels of inorganic phosphorus in serum of anoestrus heifers /buffaloes than cyclic animals (Naidu and Rao, 1982; Kumar *et al*., 1992 and Dutta *et al*., 2001).

Among trace elements the supply of iron and cobalt remained on plus side, while of zinc and copper remained deficient, while Mn was deficient only in buffaloes but not in heifers. The presence

Table 1. Macro and micro nutrient contents of feedstuffs (DM basis).

Feedstuffs	Wheat straw	Gram straw	Masoor straw	Soybean Straw	Wheat bran	Cotton Seed cake	Concentrate mixture
CP (%)	3.95±0.22	6.24±0.24	6.52±0.32	6.14±0.34	13.99±0.52	22.60±0.42	16.16±2.70
EE (%)	0.99±0.06	0.63±0.50	1.50±0.11	0.80±0.06	3.45±0.11	10.22±0.64	3.50±0.18
CF (%)	33.08±0.69	39.16±0.74	36.91±0.82	41.77±1.48	9.99±0.92	27.15±1.72	15.19±1.36
NFE (%)	50.31±0.66	45.95±1.20	46.21±0.87	45.52±1.74	68.71±1.63	35.75±1.15	49.29±2.51
TA (%)	11.64±0.64	7.98±0.34	8.84±0.30	6.59±0.38	4.51±0.74	4.25±0.22	13.98±1.79
AIA (%)	5.84±0.16	2.44±0.15	4.17±0.23	0.69±0.06	0.48±0.10	0.18±0.02	6.08±1.48
Ca (%)	0.23±0.02	1.54±0.08	1.46±0.07	0.94±0.04	0.21±0.01	0.22±0.01	0.32±0.05
P (%)	0.06±0.01	0.04±0.00	0.05±0.00	0.24±0.015	0.61±0.05	0.51±0.02	0.17±0.02
Fe (ppm)	414.76±6.47	364.14±12.8	605.28±8.05	461.99±62.70	298.51±15.23	275.24±16.96	258.45±5.94
Zn(ppm)	13.48±1.12	8.41±0.28	23.24±1.92	26.43±1.37	63.40±8.19	41.64±3.03	28.83±2.49
Mn (ppm)	39.81±1.18	15.68±0.85	87.07±7.35	67.63±2.38	71.37±3.22	15.80±0.56	18.82±1.37
Cu (ppm)	7.91±0.36	4.36±0.29	4.86±0.41	10.01±0.70	11.68±1.16	10.12±0.80	2.66±0.32
Co (ppm)	0.16±0.03	0.72±0.06	0.71±0.06	0.18±0.02	0.69±0.17	0.55±0.05	0.86±0.05

Table 2. Daily requirements and availability of nutrients in anoestrus buffaloes.

Parameters	Requirements for 300 kg Body wt.	Availability for 262.80± 22.51 Body wt.	Deficiency / Excess (%)	Requirements for 500 kg Body Wt and 8 lits /day production (7% fat)	Availability For 461±10.83 kg Body weight	Deficiency /Excess (%)
	Heifers			Buffaloes		
DMI (Kg)	5.99	5.75±0.43	4 (-)	12	11.33±0.47	6 (-)
DCP (g)	374	179.32±21.80	52 (-)	772	684.69±39.97	11 (-)
TDN Kg)	3.55	2.83±0.21	20(-)	7.28	6.06±0.26	17 (-)
Ca (g)	15	57.38±8.92	283 (+)	46.4	61.93±6.39	33 (+)
P (g)	12	8.16±1.28	32 (-)	35.8	24.41±2.15	31 (-)
Fe (mg)	299.5	2779.56±252.64	828 (+)	600	3801.73±239.87	534 (+)
Zn (mg)	179	118.24±13.74	34 (-)	480	294.24±21.01	39 (-)
Mn (mg)	239.6	353.96±38.74	48 (+)	480	396.24±24.83	17 (-)
Cu (mg)	59.9	44.61±3.22	26 (-)	120	91.40±4.09	24 (-)
Co (mg)	0.59	2.45±0.51	315 (+)	1.20	4.68±0.38	290 (+)
Se (mg)	0.6-1.8	0.78±0.06	Adequate	1.2-3.6	2.4	Adequate
Vit A (IU)	12000	5717.81±45	52 (-)	21000	11418.95±0.20	46 (-)
Vit E (IU)	15	9.49±1.54	38 (-)	15	57.96±4.98	286 (+)

of zinc is highly essential for certain enzymatic activities related to reproduction and indirectly it may act through the pituitary to influence the release of gonadotropic hormones or directly through complexing with specific legend in gonads (Miller, 1979). Deficiency of copper may also be reflected on reproductive behavior as well as performance of animals. Inactive ovaries, delayed oestrus and early embryonic death have been reported to occur due to deficiency of copper (Hidiroglou, 1979 and Singh and Vadnere, 1987).

Due to practice of least greens supply in farmers vitamin A remained the most deficient among the animals and vitamin A is very important for maintaining the health status of epithelial tissue of the reproductive tract. The deficiency of vitamin E was only observed in heifers and which indicates the lacking of concentrate in their ration, but not in lactating buffaloes. The negative impact of insufficient vitamin E was also been observed on ovulation rates (Harrison *et al*., 1984) and postpartum activities (Arechiga *et al*., 1994) of the animals.

The average values of blood Hemoglobin, glucose and total phosphorus and plasma levels of Protein and different major and micro elements of the anoestrus buffalo heifers and lactating buffaloes are shown in Table 3. It was observed that Hb contents in anoestrus heifers (10.32±0.19 g/dl) and buffaloes (11.15±0.32 g/dl) were lower than the normal value (13.14±0.06 g/dl) as reported by Das *et al.* (2003). It may be due to deficient levels of copper. Similar lower values (8.79-10.24 g/dl) were also observed by other workers (Sharma *et al*., 1983 and Perumal *et al*., 2007). The average values of blood glucose and total protein were also found lower in anoestrus lactating buffaloes and in heifers compared to the values of healthy animals (Mandal *et al*., 2002 and Nayyar *et al*., 2003). Similar findings were reported by different workers (Sharma *et al*., 1983; Tandle *et al*., 1998; Jani *et al*., 2001; Sharma *et al*., 2004; Singh and Singh, 2005; Singh and Singh, 2006, Perumal *et al*., 2007). In contrast to above findings Giri and

Table 3. Hemato-biochemical profile* of anoestrus buffaloes.

Parameters	Heifers	Buffaloes
Hemoglobin (g/dl)	10.32±0.19	11.15±0.32
Glucose (mg/dl)	50.62±1.80	59.29±1.05
Total Phosphorus (mg/dl)	24±2.21	17.50±1.42
Total Protein (g/dl)	5.06±0.19	6.33±0.18
Calcium (mg/dl)	8.13±0.38	9.59±0.25
Inorganic Phosphorus (mg/dl)	5.06±0.14	5.07±0.17
Iron (ppm)	22.05±0.58	17.68±0.84
Zinc (ppm)	1.04±0.05	0.87±0.05
Copper (ppm)	0.90±0.05	0.77±0.04
Manganese (ppm)	0.84±0.05	1.14±0.10
Cobalt (ppm)	1.92±0.41	1.13±0.09

* The values of hemoglobin, Glucose and Total Phosphorus were reported in blood, while others in plasma.

Yadav (2001) and Jagathesan *et al.* (2006) reported that Hemoglobin, glucose and total protein values were in normal range in anoestrus animals and which may be associated with change in feeding practices of those animals.

The plasma concentration of total P, Fe, Mn and co were within range in heifers (McDowell *et al*., 1984) but Ca (8.13±0.38 mg/dl), inorganic phosphorus (5.06±0.14 mg/dl) and Zn (1.04±0.05 ppm) and Cu (0.90±0.05 ppm) were below the normal values (Underwood, 1977; Prasad and Rao, 1997; Das *et al*., 2002; Mandal *et al*., 2002; Das *et al*., 2003 and Sharma *et al*., 2004). In lactating buffaloes the values of Ca (9.59±0.25 mg/dl), total P (17.5±1.42 mg/dl), Fe, Mn and Co were in range but iP, Zn, and Cu were marginally low (Underwood, 1977; McDowell *et al*., 1984; Mandal *et al*., 1996; Paul *et al*., 2000 and Yadav *et al*., 2002). The concentration of Fe in plasma of heifers (22.05±0.58 ppm) and lactating buffaloes (17.68±0.84 ppm) observed many times higher than the reported normal values (Underwood 1977; Prasad and Rao, 1997; Das *et al*., 2002; Das *et al*., 2003 and Sharma *et al*., 2004) indicates the higher levels of it in the feed ingredients used.

It may be concluded that dietary deficiency reflected the hemato-biochemical profile of anoestrus animals and a strategic supplementation is needed to these animals for exploitation of their genetic potential for optimum production and reproduction.

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