

Asian-Aust. J. Anim. Sci. Vol. 20, No. 12 : 1858 - 1864 December 2007

www.ajas.info

Influence of Varying Level of Sodium Bicarbonate on Milk Yield and Its Composition in Early Lactating *Nili Ravi* Buffaloes

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ABSTRACT: Influence of varying level of sodium bicarbonate (SB) supplementation on milk yield and its composition was examined in a randomized complete block design in early lactating *Nili Ravi* buffaloes during summer. Four iso-nitrogenous and iso-caloric diets were formulated. The diet 0B contained 0 while LB, MB and HB diets contained 0.50, 1.0 and 1.50% SB levels, respectively. The diets were randomly allotted to twenty buffaloes, five in each group. A linear increase in nutrient and water intake was recorded with increasing SB level. Buffaloes fed MB and HB diets showed higher nitrogen balance than those fed 0B and LB diets. A significant increase in blood pH and serum bicarbonate was noticed with increasing SB level. Urine pH increased significantly with increased SB level. A linear increase in milk yield was also noticed with increasing SB level. Milk fat% increased significantly in buffaloes fed MB and HB diets compared with those fed 0B and LB diets. Buffaloes fed HB diet had higher conception rate and less services per conception than those fed 0B diet. This study indicated that a high SB diet not only increased dry matter and water intake, milk yield, milk fat% but also increased conception rate in early lactating buffaloes during summer. (**Key Words:** Early Lactating Buffaloes, Sodium Bicarbonate, Nitrogen Balance, Milk Yield and Composition)

INTRODUCTION

High nutrients demand of dairy animals in early lactation can usually be fulfilled by high concentrate diet (Khan et al., 2006a), which generally results in low acetate to propionate ratio (Nisa et al., 2006). This leads to decreased feed consumption because of ruminal acidosis and thus low milk yield and milk fat content. Decreased feed consumption not only reduces milk yield (Khan et al., 2006b; Touqir et al., 2007) but it also impairs reproductive performance through delayed onset of ovarian cycle (Butler and Smith, 1989; Staples et al., 1990; Butler, 2000; Reist et al., 2000). Delayed resumption of ovarian activity has been attributed to low insulin like growth factor (IGF-1; Moore et al., 2000) due to decreased dry matter intake (DMI; Staples et al., 1990). Moreover, low IGF-1 also reduces the ability of follicles to produce sufficient estradiol by stimulating granulosa cells for estradiol production and thus reduces the chances of successful ovulation.

There may be many ways and means to enhance the feed intake in early lactation. One of the most promising is supplementing of sodium bicarbonate (SB). Low cost and

Received November 29, 2006; Accepted April 8, 2007

abundant availability of SB for the farmers of tropical region offer a potential nutritional economical tool to increase dairy animal productivity as SB increases DMI by counteracting ruminal and systemic acidosis. Sufficient scientific evidence is available regarding favourable effects of SB supplementation in dairy cows. However, limited scientific literature is available about its effects in buffalo (Bubalus bubalis). Moreover, physiological environmental condition and feeding strategies of buffaloes vary from that of exotic dairy cows in temperate region and SB responses of exotic dairy cow may not be of worth for direct application on buffaloes. Therefore, the present study was planned to determine the influence of varying levels of SB on nutrients intake, their digestibilities, nitrogen balance, milk yield and its composition and conception rate in early lactating Nili Ravi buffaloes during summer.

MATERIALS AND METHODS

The experiment was planned to determine the effect of varying levels of SB on nutrients (dry matter (DM), crude protein (CP), neutral detergent fiber (NDF) and acid detergent fiber (ADF)) intake, their digestibilities, nitrogen (N) balance, milk yield and its composition by early lactating *Nili Ravi* buffaloes. The experiment was

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Table 1. Ingredients and chemical composition of experimental diets varying in sodium bicarbonate for early lactating buffaloes

Ingredients 0B LB MB HB						
Wheat straw	40.0	40.0	40.0	40.0		
Corn grain cracked	15.0	15.0	15.0	15.0		
Molasses	12.0	12.0	12.0	12.0		
Wheat bran	10.2	9.70	9.20	8.70		
Sunflower meal	10.0	10.0	10.0	10.0		
Canola meal	6.25	6.25	6.25	6.15		
Vegetable oil	3.0	3.0	3.0	3.0		
Urea	1.80	1.80	1.80	1.90		
DCP ¹	1.50	1.50	1.50	1.50		
Salt	0.25	0.25	0.50	0.50		
NaHCO ₃	0	0.5	1.00	1.50		
Chemical composition						
NE _L (Mcal/kg)	1.50	1.51	1.51	1.51		
CP^2	16.0	16.0	15.9	16.0		
RDP^3	10.50	10.50	10.50	10.70		
RUP^4	5.5	5.5	5.4	6.3		
NDF^5	40.90	40.70	40.70	40.2		
ADF^6	26.20	26.10	26.0	25.91		
NFC^7	30.12	30.02	30.01	30.25		
Ca	0.69	0.69	0.70	0.69		
P	0.66	0.66	0.67	0.66		
Na	0.19	0.32	0.46	0.59		
K	1.54	1.53	1.52	1.52		
Mg	0.28	0.28	0.29	0.28		
Cl	0.43	0.43	0.43	0.43		
S	0.22	0.23	0.23	0.22		

Means within the same row having different subscripts differ significantly (p<0.05).

Diet 0B contained no supplementation of SB while LB, MB and HB diets contained 0.5%, 1.0% and 1.5% SB, respectively.

conducted at Animal Nutrition Research Center, Institute of Animal Nutrition and Feed Technology, University of Agriculture, Faisalabad, Pakistan.

Four diets 0B, LB, MB and HB were formulated to have 0, 0.5, 1.0 and 1.5% SB supplementation, respectively. The diets were formulated to be iso-nitrogenous and iso-caloric using NRC (2001) values for energy and protein (Table 1). Twenty early lactating *Nili Ravi* buffaloes were randomly allocated, to four dietary treatments in a randomized complete block design. Average weight of the animal was 492±24.5 kg. The experiment lasted for five months (June to October, 2005).

Buffaloes were housed on concrete floor in separate pens and no mechanical means were used to control the house temperature. The first month was adaptation period while last 10 days of each month was collection period. The diets were mixed daily and fed twice (0300 and 1400 h) a day at *ad libitum* but at 10% weigh back during collection period. The buffaloes were milked twice (0330 and 1430 h) daily.

Feed intake and milk yield were recorded daily and their representative samples were taken for analysis. Nutrient digestibility was determined by using total collection method. During four collection periods, each comprising of ten days, complete collections of urine and faeces were made according to the procedure described by Williams et al. (1984). The faeces of each animal were collected daily, weighed, mixed thoroughly and 20% of it was sampled and dried at 55°C. For urine collection, small special metal buckets fitted with plastic pipe were made to surround the vulva and plastic pipe. This plastic pipe ended in a large container (30 lit.). The urine excreted by each animal was acidified with 50% H₂SO₄ and 20% of it was sampled and preserved at -20°C (Nisa et al., 2006). In the end of each collection period, the preserved urine samples were composited by animal after thawing and 10% of the composited sample was used for analysis. During collection period, faeces were collected daily, dried at 55°C, bulked and mixed at the end of each collection period. Feed and faecal samples were analyzed for NDF by the method described by Von Soest et al. (1991). The AFD was determined by the method of Goering and Van Soest (1970). Feed samples were also analyzed for CP, Na, K, Cl, Ca, P, Mg, and S using methods described by AOAC (1990). Blood samples were collected in heprinized syringes from Juggler vein to determine the pH (AOAC, 1990). Blood serum was harvested to determine bicarbonate (HCO₃) by the method devised by Harold (1976). Milk samples were also analyzed for protein, fat, solid not fat, total solids and lactose using the methods devised by AOAC (1990). The N balance was determined by using equations as described by NRC (2001).

Statistical analysis

The data were analyzed using Randomized complete block design. In case of any significance means were separated by Duncan's Multiple Range Test (Steel and Torrie, 1984).

RESULTS

Nutrients intake and digestibilities

A linear increase in dry matter and water intake was recorded with increasing the level of SB (Table 2). The maximum (16.30 kg/d) and minimum (12.60 kg/d) DMI was recorded in buffaloes fed HB and OB diets, respectively. Buffaloes fed HB diet consumed 29.37% more feed than those fed 0B diet. The DMI by buffaloes fed LB and MB diets was 13.4 and 14.60 kg/d, respectively. However, DMI in buffaloes fed MB and HB diets remained unaltered.

A constant increase in CP and NDF intake was observed with increasing the SB level of diet. Buffaloes fed HB and

¹ Dicalcium phosphate. ² Crude protein.

³ Rumenly degradable protein. ⁴ Rumenly undegradable protein.

⁵ Neutral detergent fiber. ⁶ Acid detergent fiber.

⁷ Non-fermentable carbohydrate.

Table 2. Influence of varying levels of sodium bicarbonate on nutrients intake in early lactating buffaloes

Nutrients (kg/d)	0B	LB	MB	HB	SE
Intake					
Dry matter	12.60^{c}	13.40^{b}	14.60^{ab}	16.3 ^a	0.46
Dry matter	2.61 ^c	2.85^{b}	2.97^{ab}	3.19^{a}	0.074
(as % BW)					
Crude protein	2.02^{c}	2.14^{b}	2.34^{ab}	2.61 ^a	0.86
ADF^1	3.30^{c}	3.51^{b}	3.81 ^{ab}	4.22^{a}	1.78
NDF^2	5.15 ^c	5.45 ^b	5.91 ^{ab}	6.55^{a}	2.82
Water (L/d)	75.8 ^c	78.6 ^b	86.5 ab	98.5 ^a	4.12

Means within the same row having different subscripts differ significantly (p<0.05).

Diet 0B contained no supplementation of SB while LB, MB and HB diets contained 0.5%, 1.0% and 1.5% SB, respectively.

0B diets consumed maximum (2.61 kg/d) and minimum (2.02 kg/d) CP, respectively. Buffaloes fed LB and MB diets had 2.41 and 2.34 kg/d CP intake, respectively. Similar trend was observed for N intake (Table 4). Increase in N balance was also noticed with increasing the SB level of diets. The NDF intake was maximum (6.15 kg/d) and minimum (5.15 kg/d) in buffaloes fed HB and 0B diets, respectively. Similar trend was noticed for ADF intake (Table 2). However, a nonsignificant increase in nutrients digestibilities was noticed with decreasing the SB level of diets.

Blood pH and serum bicarbonate

The blood pH was maximum (7.516) in buffaloes fed HB and minimum (7.351) in those fed 0B diets (Table 5). Buffaloes fed LB and MB diets had 7.371 and 7.412 blood pH, respectively. A constant increase in serum HCO₃ was noticed with increasing the SB level of diets. The maximum

Table 3. Influence of varying levels of sodium bicarbonate on nutrients digestibilities in early lactating buffaloes

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Nutrient	0B	LB	MB	HB	SE
Digestibility (%)					
Dry matter	68.5	67.1	67.1	66.12	0.21
Crude protein	73.51	72.85	72.77	72.78	0.11
ADF^1	63.01	62.51	62.33	62.41	0.27
NDF^2	62.11	61.45	61.33	61.02	0.12

Means within the same row having different subscripts differ significantly (p<0.05).

Diet 0B contained no supplementation of SB while LB, MB and HB diets contained 0.5%, 1.0% and 1.5% SB, respectively.

(26.30 mmol/L) and minimum (21.55 mmol/L) HCO_3 was recorded in buffaloes fed HB and 0B diets, respectively. Buffaloes fed LB and MB diets had 23.52 and 26.31 mmol/L HCO_3 , respectively (Table 5).

Urine pH

A linear increase in urine pH was observed with increasing the SB level of diet (Table 5). The minimum (6.05) and maximum (8.01) urine pH was noticed in buffaloes fed 0B and HB diets, respectively. Buffaloes fed LB and MB diets had 6.52 and 7.47 urine pH, respectively.

Milk yield and composition

The maximum (15.4 kg/d) and minimum (13.52 kg/d) milk yield was recorded in buffaloes fed HB and OB diets, respectively (Table 8). Buffaloes fed LB and MB diets produced 14.02 and 14.87 kg/d milk, respectively. However, the difference in milk yield in buffaloes fed MB and HB diets was non-significant.

Protein and lactose % remained unaltered due to SB alteration. However, protein and lactose yield increased

Table 4. Influence of varying levels of sodium bicarbonate on nitrogen balance in early lactating buffaloes

Items	0B	LB	MB	HB	SE
Nitrogen intake (g/d)	322.56 ^c	343.04 ^b	373.76 ^{ab}	417.28 ^a	18.22
Faecal nitrogen (g/d)	80.64	85.71	94.66	104.09	3.01
% of intake	25.01	25.48	24.68	23.66	0.28
Apparent absorption (g/d)	241.92 ^c	257.33 ^b	279.1 ^{ab}	313.19 ^a	9.66
% of intake	75.01	70.01	74.67	75.05	1.56
Urinary nitrogen (g/d)	65.04 ^c	69.23 ^b	78.87^{ab}	83.6 ^a	2.05
Apparent retention (g/d)	176.88 ^c	188.1 ^b	200.24 ^{ab}	229.59 ^a	10.13
% of intake	54.83	54.83	53.57	55.02	1.26
Nitrogen balance (g/d)	103.12 ^c	107.3 ^b	119.44 ^{ab}	134.3 ^a	11.23
% of intake	31.97	31.23	31.96	31.98	0.25

Means within the same row having different subscripts differ significantly (p<0.05).

Diet 0B contained no supplementation of SB while LB, MB and HB diets contained 0.5%, 1.0% and 1.5% SB, respectively.

Table 5. Influence of varying levels of sodium bicarbonate on blood pH, bicarbonate, urine pH of early lactating buffaloes

	0		<u> </u>		
Nutrients	0B	LB	MB	HB	SE
Blood pH	7.351 ^d	7.371°	7.412 ^b	7.516 ^a	0.03
$HCO_3 (mmol/L)$	21.55 ^d	22.92°	24.91 ^b	26.3 a	0.98
Urine pH	6.05^{d}	6.52°	7.47 ^b	8.01 ^a	0.32

Means within the same row having different subscripts differ significantly (p<0.05).

Diet 0B contained no supplementation of SB while LB, MB and HB diets contained 0.5%, 1.0% and 1.5% SB, respectively.

¹ Acid detergent fiber. ² Neutral detergent fiber.

¹ Acid detergent fiber. ² Neutral detergent fiber.

Table 6. Influence of varying levels of sodium bicarbonate on milk yield and its composition in early lactating buffaloes

	0B	LB	MB	HB	SE
Milk yield (kg/d)	13.52 ^c	14.02 ^b	14.87 ^{ab}	15.4 ^a	0.378
Protein (kg/d)	0.461^{c}	0.474^{b}	$0.5.05^{ab}$		0.11
Fat (kg/d)	0.852^{c}	0.904^{b}	0.981^{ab}	1.032^{a}	0.14
Total solids (kg/d)	$2.23^{\rm c}$	2.32^{b}	2.58^{ab}	2.68^{a}	0.25
Solid not fat (kg/d)	1.38 ^c	1.42^{b}	1.60^{ab}	1.65 ^a	0.17
Lactose (kg/d)	0.757^{c}	0.784^{b}	0.836^{ab}	0.867^{a}	0.11
Concentrate (%)					
Fat	6.3 ^b	6.45 ^b	6.6 a	6.7 ^a	0.10
Protein	3.41	3.38	3.4	3.9	0.10
Total solids	16.5 ^b	16.55 ^b	17.35 ^a	17.42^{a}	0.12
Solid not fat	10.2	10.1	10.75	10.72	0.12
Lactose	5.6	5.59	5.62	5.63	0.10

Means within the same row having different subscripts differ significantly (p<0.05).

Diet 0B contained no supplementation of SB while LB, MB and HB diets contained 0.5%, 1.0% and 1.5% SB, respectively.

with increasing the SB level (Table 6). Milk fat % increased with increasing the SB level of diets. The maximum milk fat was 6.7% in buffaloes fed HB diet while minimum was 6.3% in those fed 0B diet, respectively. Milk fat % in buffaloes fed MB and HB diets was non-significant, however, it was significantly higher than those fed 0B and LB diets. Total milk solids were higher in buffaloes fed MB and HB diets than those fed 0B and LB diets (Table 6).

Conception rate

Buffaloes fed HB and MB diets had 100% conception rate. Buffaloes fed OB and LB diets had 33.3 and 66.6% conception rate, respectively (Table 7). The services per conception was minimum (1.67) in buffaloes fed HB diet while buffaloes fed OB, LB and MB diets had 2.67, 2.33 and 2.33 services per conception, respectively (Table 7).

DISCUSSION

Nutrients intake and digestibilities

Increased DM and water intake in buffaloes fed high SB might be attributed to higher rumen pH (Tucker et al., 1991; West et al., 1987) and blood HCO₃ (Shahzad et al., 2007a) acid base balance (Sanchez et al., 1994). Increased rumen pH in dairy cows fed SB has also been confirmed by mixed model analysis of Hu and Murphy (2005). The buffaloes fed HB diet might have increased ruminal buffering capacity in addition to increased water intake and ruminal fluid dilution (Russel and Chow, 1993). Similar findings were reported by Rogers et al. (1982a) who observed increased DMI in dairy cows when high SB was supplemented. They stated that in addition to buffering effect, SB also increased ruminal osmotic pressure and liquid dilution rate. In rumen sodium bicarbonate is converted into sodium (Na) and bicarbonate (HCO₃) and they impart non-buffering and buffering effects, respectively (Schneider et al., 1986). It is hypothesized that

Table 7. Influence of varying levels of sodium bicarbonate on conception rate and services /conception in early lactating buffaloes

Parameter	Diets ¹					
	0B	LB	MB	HB		
CR ² (%)	33.33	66.67	100.0	100.0		
S/C^3 (No)	2.67	2.33	2.33	1.67		

¹ Diet 0B contained no supplementation of SB while LB, MB and HB diets contained 0.5%, 1.0% and 1.5% SB, respectively.

rumen buffering reduces the extent of acidity produced by volatile fatty acids production in rumen and improves systemic acid base status (Erdman, 1988). It is also proposed that propionate decreases feed intake of ruminants by stimulating oxidative metabolism in the liver (Allen, 2000). Oxidative metabolism in the liver had been shown to affect satiety in rats (Langhans et al., 1985). They proposed that oxidative metabolism in the liver affected feed intake by hyperpolarizing cell membrane potentials. Moreover, non-buffering effect of SB due to solute action increases rumen osmotic pressure and liquid dilution rate (Rogers et al., 1979; 1982). The non-buffering effects, inturn, increase influx of water and acclerate flow of liquid digesta from the rumen (Rogers, 1979), which is associated with increased efficiency of fiber digestion and microbial protein synthesis (Rogers, 1988). A slight increase in nutrients digestibilities in buffaloes fed 0B diet might be due to increased retention time because of significantly reduced feed intake. It is documented that positive co-relation between nutrients retention time and digestibilities exist (Sarwar et al., 1996).

Blood pH and serum bicarbonate

A constant increase in blood pH with increasing the SB level of diets might be attributed to gradual increase in sodium intake. Sodium absorption takes place in posterior segment of the intestine, when excess of Cl, in exchanges of H^+ to maintain the electrical neutrality of the body. In the present study, absorption of high sodium content compared to Cl might have decreased serum H^+ concentration by giving birth to increased blood pH and HCO_3 (Table 3) Similar findings have been reported by Jackson et al. (2001) who reported increased serum HCO_3 in calves fed SB (1.75%). Waterman et al. (1991) also reported an increase in blood H^+ with reducing cation level like Na.

The slight acidic blood pH in buffaloes fed 0B and LB diets might be attributed to the fact that phosphate and ammonia buffer system function for hydrogen ion excretion. Hydrogen ions combine with phosphate or ammonia after entering the renal tubules and a HCO₃ ion is formed that enters the extracellular fluids to further buffer acid in the extracellular fluids (Guyton, 1991). The 0B diet had low Na and high Cl. The Cl is anionic in nature and high Cl in OB diet might have overcome the capacity of kidneys to excrete sufficient hydrogen ion to maintain a constant blood pH,

² Conception rate. ³ Services per conception.

resulting in slight systemic acidosis (Shahzad et al., 2007b). Moreover, buffaloes fed HB diet tended to have high blood pH because of high Na exchange in place of H⁺ from the posterior segment of intestine which might have resulted in high HCO₃⁻ production and H⁺ excretion (Tucker et al., 1992). However, the increased pH was within the normal range.

Urine pH

Alteration in urine pH reflected alteration in blood pH and kidneys played a vital role to minimize this change by making the urine pH alkaline due to more Na content in buffaloes fed MB and HB by means of excreting more HCO₃ and conserving H⁺ (Roche et al., 2003). Increase in urine pH with increasing the dietary Na content has also been reported by Waterman et al. (1991). Moreover, increased urine pH had been used as an indicator of metabolic alkali load (Sanchez, 2003). The findings of the present study are in line with other workers (Jackson et al., 1992; Mosel et al., 1993; Jackson and Hemken, 1994; Pehrson et al., 1999) who reported increased urine pH with increasing the dietary Na when sodium bicarbonate was added in diet. Maximum (9.0) and minimum (4.50) fluctuation in urine pH had been reported by Roche et al. (1999) with supplementing varying sodium bicarbonate to increase dietary sodium concentrations. Similar findings are also reported by Jackson et al. (2001) who reported increased urine pH (8.09) in dairy claves fed high dietary Na or K content compared to those (6.80) fed low or without sodium or bicarbonate supplementation (Jackson et al., 2001).

Milk yield and composition

Increased milk production in buffaloes fed HB diet was due to increased DMI. These findings are supported by Tucker et al. (1988) who reported increased milk production in lactating cows fed high SB compared to those fed low SB diet. Similar results were reported by Block (1994) who indicated that high Na or K contents from sodium or potassium bicarbonate increased milk production in lactating cows. He further stated that lactating cows had higher metabolic rate that tended to make the cellular environment acidic due to more CO₂ production. A high SB diet due to high Na content has alkalogenic nature and reduces the extent of that acidity and thereby increases cellular glucose uptake.

Unaltered protein and lactose % due to alteration in SB level are in concordance with findings of other workers (Tucker et al., 1988; Block, 1994). In the present study, increased milk fat % was recorded in buffaloes fed HB diet. High sodium bicarbonate diet had the tendency to increased ruminal pH, which might have shifted the fermentation pattern in favour of acetate and butyrate production (Kolver

and De Veth, 2002). It might have resulted in increased *de novo* fatty acid synthesis which accounts for 60% of bovine milk fat (Bauman and Davis, 1974) and hence increased milk fat content. Decreased propionate and increased acetate molar proportion by increasing dietary SB level has also been reported by Staples and Lough (1989). Reduced milk fat content in buffaloes fed low SB diet might be attributed to high propionate and low acetate rumen molar concentrations as positive relationship between milk fat and molar proportion of acetate has been documented by Hu and Murphy (2005). Deceased propionate concentration has also been reported to negatively co- related with milk fat content (Coppock et al., 1986).

In the present study, 40% wheat straw, this is rich in effective fiber. A high sodium bicarbonate when fed in blend with high effective fiber counteracted the acidic environment of rumen and this might have ensured high acetate to propionate ratio by optimizing cellulolytic microbial activity. The combined effect of these two might have increased milk fat synthesis. These findings are supported by Roche et al. (2005) who reported an increased milk fat when sodium bicarbonate was added in diet.

Conception rate

Increased conception rate in buffaloes fed HB diet might be attributed to increased DMI which might have increased IGF-1 (Pate, 1999). Moreover, ability of follicles to produce sufficient estradiol for successful ovulation depends upon IGF-I that has stimulatory effect on granulosa cells for estradiol production, thus it is hypothesized that high level of IGF-I might have improved ovarian activity through accelerating the follicle growth (Butler and Smith, 1989). These findings are in concordance with those reported by Reist et al. (2000) who indicated that cows with higher serum IGF-1 (65 ng/ml) ovulated while low serum (47.5 ng/ml) IGF-1 cows could not ovulate at 21 days postpartum. Moore et al. (2000) also observed increased plasma IGF-1 in cows fed high bicarbonate compared to those fed low carbonate diet. They also observed delayed resumption of ovarian activity in cows with low IGF-1, indicating the effect of low DMI.

In buffaloes fed 0B diet, decreased DMI might have resulted low IGF-1 level which delayed onset of ovarian cycle, low conception rate and high services per conception (Butler and Smith, 1989; Staples et al., 1990; Butler, 2000; Reist et al., 2000). However, high conception rate (100%) in buffaloes fed HB and MB diets might be due to small number of animals per treatment which is the main limitation of this study but this facilitated the authors for closer observance of peak heat periods for more timely breeding as compared with large groups in pasture or paddock holdings. However, detailed study involving greater number of animals is warranted.

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