RESEARCH ARTICLE

Effect of feeding betaine hydrochloride and bypass fat supplement on feed intake, milk yield and physiological parameters in lactating buffaloes during heat stress

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Abstract: Present study examined the effect of feeding betaine hydrochloride (BH) and bypass fat (BF) in 24 early lactating buffaloes during May to July, 2018 in Nadiad district of Gujarat state. Buffaloes (30-35 days in milk) yielding 10-12 kg milk/d were divided into three groups of eight each based on milk yield, fat% and stage of lactation. Buffaloes in T_1 group (control) were fed basal ration. In addition to basal ration, buffaloes in T, group were fed daily 30 g BH + 100 g buffer; while buffaloes in T₂ group were fed daily 30 g BH + 100 g buffer + 100 g BF for 60 days. Average thermal humidity index was 80.10 at 14:00 hr during the trial period. Study revealed that DMI improved (P=0.011) in T_a group as compared to control. Non-significant difference in milk yield was observed among the groups; however, reduction in milk yield was lower in T₂ & T₃ groups, as compared to control. Milk fat improved by 3.2% and 6.2% (P=0.002) in T₂ and T₃ groups, respectively. Rectal temperature (102.8, 102.7 vs 103.6 °F), respiration rate (46.0, 45.1 vs 60.1 breath/min.) and pulse rate (61.2, 58.5 vs 72.9 pulse/min.) at 14:00 hr were significantly (P<0.01) lower in buffaloes under T₂ and T₃ group as compared to T group. Average net daily income was increased by Rs. 36.02 and 47.43 in T₂ and T₃ group, respectively as compared to control.

Keywords: Betaine, Buffalo, Bypass fat, Heat stress, Milk yield

Introduction

Due to ongoing climate change, the climate of India has become increasingly volatile over the past several decades. There is accumulating empirical evidence that the changing environmental conditions are already adversely impacting the livestock productivity. Higher temperatures along with altered precipitation are likely to result in the problem of heat stress in dairy animals leading to decreased production. West et al. (2013) and Babinszky et al. (2011) reported lower feed intake by 5-20% and reduction in milk yield by 10-40% in dairy cows due to heat stress. Loss of productivity in dairy animals in India during heat stress is a wellknown menace.

India possesses the world's largest livestock population having 57% of the world's buffalo (*Bubalus bubalis*) population. India is the largest milk producing country in the world and buffalo is the main milk producing animal contributing >50% of the total milk production in India. Buffaloes are more vulnerable to heat stress at high ambient temperature due to less number of sweat glands and black coat colour leading to loss in their productive efficiency (Upadhyay et al. 2008). Exposure of buffaloes to the hot conditions causes a series of changes in the biological functions that include decrease in feed intake, efficiency and utilization, disturbances in metabolism of water, protein, energy and mineral balances, resulting in impairment of productive and reproductive efficiencies (Das et al. 2014).

Productivity loss during heat stress period can be minimized by providing strategic feed supplement. The provision of nutritional supplements to ameliorate the effects of heat stress in dairy animals is an attractive method and is potentially the fastest and easiest option for milk producers. One such supplement that has been recently studied is the organic amino acid derivative; dietary betaine (DiGiacomo et al. 2014). Physiologically, mammals utilise betaine as a methyl donor able to participate in protein and lipid metabolism, or when not catabolised, betaine can be used as an organic cellular osmoprotectant (Fernández et al. 1998; Huang et al. 2007). In addition, use of bypass fat supplement in early lactating animals increases the energy density of the diet enabling the animals to meet their energy and essential fatty acid requirements expressing their milk production potential (Sirohi

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et al. 2010). Keeping this in mind, present work has been undertaken to evaluate the effect of supplementing betaine hydrochloride and bypass fat on feed intake, milk yield and physiological parameters of lactating buffaloes during summer season.

Materials and methods

Location, animals and dietary treatments

Present study was undertaken during summer months (18^{th} May to 28^{th} July 2018) at an organized dairy farm of Dumral village in Nadiad district of Gujarat. Twenty-four early lactating (30-35 days in milk) buffaloes were divided into three groups of eight each, based on milk yield (10-12 kg/d), fat (6.8-6.9%) and stage of lactation (2-4 lactations). Buffaloes in control group (T_1) were fed basal diet comprising of 6-7 kg concentrate, 20 kg green fodder and 4.0 kg dry fodder (NRC, 2001). In addition to basal diet, animals in T_2 group were supplemented daily 30 g BH and 100 g buffer; and buffaloes in T_3 group were supplemented daily 30 g BH, 100 g buffer and 100 g BF for 60 days. Buffer contained 49 g sodium bicarbonate, 21 g magnesium oxide and 30 g potassium chloride. Bypass fat was procured from M/s. Berg Schmidt Indian Pvt Itd, Pune.

Temperature and humidity

Ambient temperature and relative humidity were recorded twice daily at 09:00 and 14:00 hrs in buffalo shed during the trial period. Daily maximum and minimum ambient temperatures were recorded and calculated thermal humidity index (THI) as per the equation THI = $0.72 (T_{db} + T_{wb}) + 40.6$; where T_{db} is the temperature of the dry bulb and T_{wb} is the temperature of the wet bulb (NRC 1971).

Feed, fodder and milk analyses

The chemical composition of feeds and fodder offered to buffaloes during the trial period was carried out as per AOAC (2005). All buffaloes were hand milked twice daily (06:00 and 18:00 hrs) and individual milk yield was recorded. The milk samples were collected at weekly intervals from individual buffaloes during both milking time. After proper mixing, pooled milk samples (100-150 ml) from each buffalo were collected and analysed for fat, SNF, protein and lactose contents by an automatic milk analyzer. Average feeding cost and realization receipt from the sale of milk was calculated based on the current market prices.

Physiological parameters

Rectal temperature, respiration rate and pulse rate were recorded twice daily at 09:00 and 14:00 hrs for 2 consecutive days in every week. The rectal temperature (°F) was recorded using a digital thermometer by inserting 3 inches in the rectum for about 2 min. The respiration rate per minute was recorded by counting the movement of flank, i.e. one outward and inward movement

considered as one respiration. The pulse rate per minute was counted by palpating coccygeal artery.

Statistical analysis

Data were analysed using one-way ANOVA with SPSS package programme (SPSS 9.00 software for Windows, SPSS Inc., Chicago, IL) as per Snedecor and Cochran (1994). When F-test was significant (P<0.05), Turkey's test was utilized to compare significant difference (P<0.05) among the groups.

Results and discussion

Chemical composition

Analysis of feeds and fodder offered to buffaloes in all three groups is given in Table 1. Bypass fat (Bergafat) contained 99% total fat (min.). Fatty acid composition contained about 77.22% palmitic acid ($C_{16:0}$), 1.21% myristic acid ($C_{14:0}$), 5.14% stearic acid ($C_{18:0}$), 12.49% olic acid ($C_{18:1n9c}$), 2.56% linoleic acid ($C_{18:2n6c}$) and 0.35 arachidic acid ($C_{20:0}$).

Weather conditions

The weather conditions during the experimental period (18^{th} May to 28^{th} July, 2018) were typically hot. The average ambient air temperature at 9.00 and 14.00 hrs during the experimental period was 32.03 ± 0.40 °C and 36.13 ± 0.96 °C, respectively. Whereas, average humidity at 9.00 and 14.00 hrs was $70.90 \pm 1.40\%$ and $60.71 \pm 2.44\%$, re-spectively. The average calculated THI was 70.22 ± 0.65 and 80.10 ± 1.07 units at 9.00 and 14.00 hrs, respectively during the trial period. The maximum THI was recorded at initial period (0 week) of experiment (85.24).

Dry matter intake

It is well known that dairy animal reduces its feed intake during the heat stress as a thermoregulation effort to prevent an excessive increase in body temperature via decreasing its heat production. West et al. (2013) reported that feed intake reduced by 5-20% in dairy cows due to heat stress.

In our study, initial dry matter intake (DMI) in all three groups were similar (14.05, 14.19 and 14.16; P=0.231). However, during the trial period, DMI reduced by 1.86, 1.71 and 1.42 kg in T_1 , T_2 and T_3 groups, respectively (Table 2). Despite reduction in all three groups, DMI in T_3 group was significantly higher (P=0.011) by 4.51% as compared to control group, which indicates that the heat stress has less effect on DMI in T_3 group as compared to control (Figure. 1). Non-significant improvement in DMI was observed in T_2 group as compared to T_1 group. Marai and Habeeb (2010) reported that the reduction in DMI is the most important effect of heat stress in tropical and sub-tropical conditions. Increased heat load decreases nutrient uptake in almost all species and in case of cattle, the nutrient uptake decreases upto

Fig. 1 Inter-relationship between THI and DMI



Table 1 Chemical composition of feeds and fodder (% DM basis)

Feed ingredients	СР	ΕE	CF	AIA	Ca	Р
Concentrate mixture	19.87	4.28	9.07	1.20	0.86	0.76
Wheat straw	3.12	1.64	39.10	6.26	0.74	0.10
Maize green fodder	7.42	2.29	28.92	6.58	0.66	0.27

about 30% of dry matter intake (Rhoads et al. 2013; Wheelock et al. 2010). A decrease in the dry matter intake in dairy cattle was found on exposure to heat stress (Rhoads et al. 2009). High environmental temperature reported to reduce feed intake in Friesian heifers (Smith et al. 2013).

Milk yield

As the lactation progress, milk production decline in all animals, but reduction in milk yield was less in treatment groups as compared to control group; may be due to supplementation of BH and BF in buffaloes. Milk yield was higher (P=0.334) by 5.69 and 6.73% in T_2 and T_3 group, respectively as compared to T_1 group. The average milk fat in T_1 , T_2 and T_3 was 6.93, 7.15 and 7.36%, respectively, which showed that the BH and BF supplementation in concentrate mixture of lactating buffaloes (T_2 and T_3) increased (P=0.002) fat content of milk as compared to control group (T_1).

Marai et al. (2009) recorded lower milk yield in buffaloes during summer than spring and winter seasons and decline in milk yield as a direct result of high environmental temperature. Wang et al. (2010) reported that by adding 100 or 150 g of anhydrous betaine significantly improved milk yield and fat during the early lactation of Holstein dairy cows. Zhang et al. (2014) studied that the addition of up to 15 and 20 g/day betaine caused milk yield to increase by 29.16 and 28.87 vs. 27.70 kg/d (control), respectively. Bouraoui et al. (2002) found that daily THI is negatively correlated with milk yield and observed that increasing THI value from 68 to 78 decrease milk production by 21% and DMI by 9.6%. Aggarwal and Singh, (2006) also reported that milk yield decreases by 0.41 kg/cow/d for each point increase in the THI values above 69.0. In our study also, we observed weekly inter-relationship between THI and daily milk yield of buffaloes during the whole trial period (Figure 2).

Economics

Daily feeding cost of ration was Rs. 209.59, 223.30 and 234.71 in T_1, T_2 and T_3 groups, respectively which was significantly higher (P=0.001) in T_2 and T_3 groups as compared to T_1 group. Higher feeding cost in experimental group was mainly due to the higher DMI and addition of supplemental cost. However, daily realizable receipt (Rs./head) from the sale of milk was higher by 9.12 and 13.30% (P=0.037) in T_2 and T_3 groups, respectively as compared to control group. Cost benefit ratio (net income per unit feeding cost) was found to be higher in $T_2(1.67)$ and $T_3(1.63)$ groups, as compared to T_1 (1.60) group. Thus, net daily income of farmer increased by Rs. 36.02 and 47.43 per animal after supplementation



Table 2 Effect of feeding betaine and bypass fat on feed intake, milk yield and economics

Parameters	T ₁	T ₂	T ₃	SEM	P value
Feed intake					
Initial DMI (kg/d)	14.05	14.19	14.16	0.09	0.231
DMI (kg/d) after 60 days of trial	12.19 ^a	12.48 ^{ab}	12.74 ^b	0.09	0.011
Milk yield and composition					
Initial milk yield (kg/d)	12.85	12.78	12.81	0.22	0.326
Milk yield (kg/d) after 60 days	11.59	12.25	12.37	0.23	0.334
Fat (%)	6.93ª	7.15 ^{ab}	7.36 ^b	0.06	0.002
SNF (%)	9.15ª	9.36 ^b	9.34 ^b	0.04	0.038
Protein (%)	3.40	3.42	3.45	0.01	0.222
Lactose (%)	5.24	5.29	5.30	0.02	0.439
Economics					
Daily feeding cost (Rs./head)	209.59ª	223.30 ^b	234.71°	2.50	0.001
Realizable receipt from sale of milk (Rs./head/d)	545.39ª	595.13 ^b	617.95 ^b	11.77	0.037
Average gross income (Rs/head/d)	335.81	371.83	383.24	10.93	0.242
Cost benefit ratio	1:1.60	1:1.67	1:1.63		

^{abc}Means bearing different superscripts in a row differ significantly (P<0.05)

of BH alone and along with BF in T_2 and T_3 group over T_1 group, respectively.

Physiological parameters

The rectal temperature and respiration rate is recognized as an important measure of physiological status as well as ideal indicator for assessment of stress in dairy animals. Rectal temperature is generally considered to be a useful measure of body temperature and changes in rectal temperature indicates changes of a similar magnitude in deep body temperature (Ganaie et al. 2013). In our study, supplementation of BH, buffer and BF helped to reduce heat stress in buffaloes. Average rectal temperature (103.6, 102.8, 102.7 °F; P=0.01), respiration rate (60.1,

46.0, 45.1 per min; P=0.01) and pulse rate (72.9, 61.2, 58.5 per min; P=0.01) were significantly lower at 14:00 hr in T_2 and T_3 groups as compared to control group (Table 3). Physiological parameters indicate that the buffaloes in control group were more prone to heat stress as compared to supplemental groups. Gaughan et al. (2005) suggested that the addition of exogenous osmolytes significantly reduce the rectal temperature, and increases the feed intake and heat endurance of buffaloes during heat stress. The results of our study showed that the rectal temperature significantly declined with BH and BH with BF supplemented groups. This trend might be related to the function of BH, which helps animals to alleviate heat stress.

Attribute	Time (hrs) T ₁	T ₂	T,	SEM	P value
Rectal	9.00	101.5	101.3	101.6	0.08	0.398
temperature (°F)	14.00	103.6 ^b	102.8ª	102.7ª	0.13	0.01
Respiration rate	9.00	44.1	42.0	40.9	0.67	0.133
(breath/min.)	14.00	60.1 ^b	46.0ª	45.1ª	1.60	0.01
Pulse rate	9.00	56.7	55.0	55.0	0.63	0.346
(Pulse/min.)	14.00	72.9 ^b	61.2ª	58.5ª	1.47	0.01

Table 3 Physiological parameters of buffaloes

^{abc}Means bearing different superscripts in a row differ significantly (P<0.05)

Conclusions

Study indicates that supplementation of betaine hydrochloride, buffer and bypass fat in the ration of buffaloes improved dry matter intake, milk production as well as its component and also helped in maintaining crucial physiological parameters of buffaloes during hot condition. This supplementation during summer season increases the net daily income of farmer by 14% in comparison to control.

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