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Performance of lactating buffaloes as affected by varying concentrations of essential amino acids

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ABSTRACT: The study was designed to examine the influence of varying levels of limiting amino acids (lysine, methionine and threonine) supply to lactating Nili Ravi buffaloes on their nutrients intake and utilization, milk yield and composition and nitrogen balance. Fifteen lactating *Nili-Ravi* buffaloes in mid lactation with almost similar milk production were divided into three groups, five animals in each according to completely randomized design. Three iso-caloric and iso-nitrogenous diets with high, medium and low levels of lysine, methionine and threonine concentrations (%of crude protein) were formulated and represented as high essential amino acids (HEAA), medium essential amino acids (MEAA) and low essential amino acids (LEAA) concentrations, respectively. The study lasted for 100 days; first ten days were given for the adaptation to new diets while every six days after every month of the remaining period served as collection periods. The intake (% body weight) of dry matter (DM), crude protein (CP), metabolizable energy (ME), neutral detergent fiber (NDF) and acid detergent fiber (ADF) did not show any treatment effect. However, the intake of rumen un-degradable protein, lysine, methionine, and threonine reduced linearly ($P<0.01$) with decreasing concentrations of these nutrients in the experimental diets. A linear increase ($P<0.01$) in DM, NDF and ADF degradabilites was recorded with gradually decreased dietary RUP and EAA concentrations while CP digestibility remained unchanged. Milk yield, daily weight gain decreased linearly ($P<0.01$) with reducing dietary levels of EAA concentrations. A positive nitrogen balance was also observed in buffaloes fed all the experimental diets but it was similar statistically. Hematological analysis indicated decreased ($P<0.01$) mean corpuscular volume and hemoglobin concentrations in buffaloes fed gradual decreased dietary EAA concentrations.

Key words: essential amino acids, nutrient utilization, nitrogen balance, hematological aspects, buffaloes

INTRODUCTION

Sustainable milk production requires high intakes of protein especially essential amino acids (EAA; lysine methionine and threonine) for lactating animal to meet milk protein synthesis. Methionine and lysine have been suggested to be the most limiting AA for milk production when corn-based diets are fed to animals (NRC, 2001). Being cheaper high energy source, corn is extensively used as major feed ingredient in dairy rations world wide. Since dietary proteins are largely degraded by microbes in the rumen, so formulating a ration to provide high EAA concentrations can be difficult using locally available feed resources. The difficulty is further confounded when rumen degradation of feed ingredients is considered during feed formulation. Supplying rumen un-degradable protein (RUP) to meet the requirements of the host may balance the required nitrogen and energy ratio in the rumen but still may result in an inadequate supply of post ruminal EAA. In rapid growing and high producing dairy animals rumen micro-organisms also cannot synthesize enough protein to support high level of milk production rather the microbial protein is deficient in EAA. Therefore, high producing animal diet demands escape of some dietary protein from rumen fermentation and ample concentration of EAA for milk production. Additional RUP and EAA supply to dairy animals has been reported to optimize productivity (Ørskov ER.1982; Robinson

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et al., 1999). The NRC, (2001) have suggested that in a typical diet, approximately 40% of the protein eaten must be true protein that escapes rumen degradation, whereas 60% of the protein value can be a mixture of protein and non-protein nitrogen that is degraded and incorporated into the rumen microbes. The present study has designed to examine the influence of varying levels of EAA supply to lactating buffaloes on their nutrients intake and utilization, milk yield and composition and nitrogen balance.

MATERIALS AND METHODS

Fifteen lactating *Nili-Ravi* buffaloes having similar milk production were divided in to three groups (five buffaloes in each group) according to CRD for this study. Three iso-caloric and iso-nitrogenous diets with 3 levels of EAA (lysine, methionine and threonine) were formulated using NRC (2001) standards for energy, protein and EAA (Table 1). The buffaloes fed individually at ad lib intakes and were housed on concrete floor in separate pens. The study lasted for 120 days, first ten days were adaptation period while every first week of each month of the remaining period served as collection periods. The buffaloes were weighed at start of the study and fortnightly thereafter.

Table 1. Ingredients and chemical composition of experimental diets, g/ kg

Ingredients, g/kg	HEAA[1]	MEAA[1]	LEAA[1]
Maize broken	30.00	200.00	150.00
Wheat straw	450.00	250.00	120.00
Wheat bran	150.00	200.00	300.00
Rice polishing	10.00	0.00	50.00
Cotton seed cake	60.00	15.00	20.00
Maize oil cake	0.00	0.00	95.00
Maize gluten Meal 30%	83.50	50.00	10.00
Canola meal	10.00	10.00	10.00
Soy bean meal	48.00	55.00	20.00
Corn steep liquor	60.00	60.00	60.00
Cane molasses	80.00	142.00	148.00
Mineral mix	15.00	15.00	15.00
Urea	3.50	3.00	2.00
Chemical composition, %			
Dry matter	87.62	85.83	85.37
Crude protein	14.02	14.18	14.12
Rumen un-degradable protein	50.31	41.31	32.17
Lysine %CP	3.10	2.43	1.85
Methyionine %CP	1.86	1.20	0.95
Treonine %CP	3.20	2.84	1.86
Lysine%of RUP	1.56	1.01	0.60
Methionine %RUP	0.94	0.50	0.31
Threonine %RUP	1.61	1.17	0.60
Neutral detergent fiber	46.87	32.35	30.88
Acid detergent fiber	26.98	17.56	13.86
Acid detergent lignin	5.44	3.35	2.85
Total ash %	8.89	9.30	9.19
Metabolizable energy, Mcal/ kg	2.09	2.22	2.25

[1]HEAA, MEAA and LEAA diets contained high, medium and low essential amino acids concentrations, respectively

RESULTS AND DISCUSSION

Nutrients Intake, Digestibilities, Nitrogen Balance

Dry matter (DM), crude protein (CP), neutral detergent fiber (NDF), acid detergent fiber (ADF) and ME intake and nitrogen balance in buffaloes fed different EAA concentrations remained unaltered, however, there was a linear decrease in EAA and RUP intakes with reducing levels of these nutrients in the diets (Table 2). The linear decrease in intake of EAA and RUP in buffaloes fed HEAA, MEAA and LEAA diets was because of gradual decreased dietary concentration of these nutrients in the experimental diets.

Dry matter and NDF digestibility increased linearly with gradually decreased dietary concentration of EAA and RUP in the diets but CP digestibility was similar statistically. The diets were iso-nitrogenous which did not influence CP digestibility, but improved DM and NDF digestibility in buffaloes fed decreased EAA and RUP concentrations might be attributed to gradual dietary increased RDP concentration which might have increased supply and availability of N in diverse forms (ammonia N, peptides and amino acids). This might have accelerated rumen microbial fermentation activities leading to improved feed consumption (DelCurto et al., 1990). A positive correlation between increased rumen fermentation and dry matter digestibility has been reported previously by Nisa et al. (2004). Availability of N in diverse forms has also been documented to facilitate the synchronization of carbon and nitrogen units in rumen which has direct positive influence on rumen microbial proliferation and enzyme production which might have favored increased DM digestibility in buffaloes fed decreased EAA and RUP diets (Javaid et al., 2008; Nisa et al., 2008). Increased nutrient digestibility due to increased ruminal dynamics by availability of nitrogen in diverse form has also been documented by Haddad Goussous (2005). Therefore improvement in nutrient digestion in the current investigation might be due to increased ruminal dynamics by availability of nitrogen in diverse form to the host.

Weight Gain, Milk Yield and Milk Fat

A linear ($P<0.05$) decrease in daily weight gain and milk yield was observed in buffaloes fed reducing EAA and RUP concentrations, however, milk fat content didn’t differ (Table 4). This increased milk production and weight gain with increasing EAA concentrations might be attributed to better availability of dietary nitrogen in diverse forms which might have enhanced rumen microbial multiplication, per unit enzyme production and overall fermentation activities in the rumen leading to more volatile fatty acid production (Javaid et al., 2008). This might have enhanced nutrients supply for more milk synthesis. Furthermore, increasing dietary RDP concentration has been reported to increase rumen ammonia content (Nisa et al., 2008). In the current study all the buffaloes were in positive N balance which led to spared more nutrients for milk production and daily gain. Ammonia, being alkaline in nature might have increased rumen pH which might have favored shifting the fermentation pattern in favour of acetate and butyrate production and ultimately increased milk production.

Hematology

Observations of WBC, RBC, Hct, and MCHC remained unaltered across all diets, however, MCV and MCH concentrations decreased with gradual reduction of dietary RUP concentration (Table 3). Gradual reduction in MCV and MCH concentration with decreased dietary EAA and RUP concentration might have decreased the sustainable supply of EAA required for the normal biosynthetic activities aimed to synthesize these blood constituents. Gradual reduction in dietary RUP concentration might have limited the supply of these amino acids leading to their respective decreased contents in the blood, despite the fact that diets were iso-nitrogenous (Bergman and Heitmann, 1980). Another plausible explanation of decreased MCV and MCH concentration might be that gradual increase in RDP fraction of dietary CP couldn’t ensure similar amino acid supply at post ruminal level to avoid any variation in blood amino acid profile which might have altered availability and sequence of amino acid supply for the synthesis of these blood constituents.

Table 2. Nutrients intake and their digestion

Parameters	HEAA[1]	MEAA[1]	LEAA[1]	SEM[1]	Linear	Quadratic
Nutrients Intake % body weight						
Dry matter	2.85	2.92	2.78	0.09	0.78	0.65
Crude protein	0.40	0.41	0.40	0.01	0.84	0.73
Rumen un-degradable protein	0.20[a]	0.17[b]	0.12[c]	0.01	0.001	0.70
Lysine	0.49[a]	0.37[b]	0.28[c]	0.03	0.00	0.16
Methyonine	0.29[a]	0.19[b]	0.15[c]	0.02	0.00	0.00
Treonine	0.51[a]	0.43[b]	0.29[c]	0.03	0.00	0.09
ME	0.25	0.25	0.25	0.02	0.94	0.93
NDF	6.14	6.40	6.33	0.56	0.90	0.91
ADF	3.51	3.65	3.61	0.32	0.91	0.91
Nutrient digestion						
DM	63.95[c]	65.83[b]	68.86[a]	0.63	0.001	0.16
CP	75.00	76.50	76.05	0.98	0.69	0.67
NDF	53.82[c]	55.66[b]	57.71[a]	0.56	0.001	0.88
Daily weight gain	0.81[a]	0.5675[b]	0.5175[b]	0.043	0.001	0.06

[1]HEAA, MEAA and LEAA diets contained high, medium and low essential amino acids concentrations, respectively; SEM standard error of the means
[a,b,c] Means within row bearing different superscripts differ significantly ($p<0.05$)

Table 3. Milk production and weight gain

Parameters	HEAA	MEAA	LEAA	SEM	Linear	Quadratic
Milk production, Kg/day	14.86[a]	12.25[b]	10.84[c]	0.52	0.00	0.09
Fat%	6.08	6.18	6.33	0.11	0.39	0.92
4%FCM, Kg/day	15.06	14.16	13.82	0.24	0.03	0.52
Daily weight gain	0.81[a]	0.5675[b]	0.5175[b]	0.043	0.001	0.06

[1]HEAA, MEAA and LEAA diets contained high, medium and low essential amino acids concentrations, respectively; SEM standard error of the means
[ab] Means within row bearing different superscripts differ significantly ($p<0.05$)

Table 4. Nitrogen Balance

Parameters	HEAA	MEAA	LEAA	SEM	Linear	Quadratic
Nitrogen Intake g/d)	353.37	339.39	340.30	4.59	0.28	0.47
Fecal Nitrogen	77.60[c]	106.40[b]	128.00[a]	6.91	0.00	0.62
Urine Nitrogen	152.06[a]	134.88[ab]	131.75[c]	3.76	0.02	0.28
Milk Nitrogen	22.735	22.19	22.6575	0.244246	0.91	0.38
Nitrogen Balance	89.9325	91.4675	89.0425	2.94825	0.91	0.78
Blood Urea Nitrogen	21.07[a]	18.00[b]	16.16[c]	0.67	0.00	0.25

[1]HEAA, MEAA and LEAA diets contained high, medium and low essential amino acids concentrations, respectively; SEM standard error of the means
[a,b,c] Means within row bearing different superscripts differ significantly ($p<0.05$)

Table 5. Hematology

Parameters	HEAA	MEAA	LEAA	SEM	Linear	Quadratic
WBC Nx10^3/uL	10.91	12.80	11.19	0.96	0.91	0.25
RBC Lx10^6/uL	5.45	4.88	5.58	0.28	0.98	0.25
HB L g /dL	10.54	8.91	9.00	0.47	0.10	0.16
MCV H fL	66.60[a]	59.39 [ab]	57.08[b]	1.91	0.04	0.45
Hct L %	32.84	30.13	30.95	1.22	0.35	0.22
Plt *L x10^3/uL	188.01	248.44	218.31	22.0	0.68	0.37
MCH H pg	23.90[a]	17.45[ab]	16.38[b]	1.55	0.04	0.38
MCHC L g/dL	33.30	29.10	28.70	0.99	0.05	0.41

[1]HEAA, MEAA and LEAA diets contained high, medium and low essential amino acids concentrations, respectively; SEM standard error of the means
[ab] Means within row bearing different superscripts differ significantly ($p<0.05$)

CONCLUSION

The findings of the present study reflect that varying levels of lysine, methionine and threonine have the potential to alter nutrient digestibility and milk production. However, further studies, keeping in view the level of amino acid degradability, should be executed with grater number of animals before recommending any concentration level of these amino acids in buffaloes.

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