



Effect of Molasses Feeding on Biochemical and Hormonal Parameters in Sahiwal and Karan Fries Heifers

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ABSTRACT

The present study was designed to investigate the effect of molasses feeding on biochemical and hormonal parameters in Sahiwal and Karan Fries heifers under two feeding regimes, i.e. feeding regime-1, as per the NRC (2001) and feeding regime-2 having 15 % higher energy (supplementation of molasses) than NRC (2001). Twelve healthy heifers (18-24 month) of each Sahiwal and Karan Fries were selected from Livestock Research Centre (LRC) of ICAR-National Dairy Research Institute (ICAR-NDRI). An initial 15 days were maintained as adaptation period for both feeding regimes. Actual experiment was conducted from 16th day onward for next 15 days. Blood samples were taken on 1st, 7th, 14th, 15th, and 16th day of each feeding regimes and analysed for various biochemical and hormonal parameters. The plasma glucose, Triiodothyronine (T₃) and Thyroxin (T₄) were significantly (p<0.05) higher during feeding regimes-2 as compared to feeding regimes-1 in both the breeds, while the plasma BUN levels were significantly lower during feeding regimes-2. The plasma NEFA level was found to be higher during feeding regimes-1 as compared to feeding regimes-2 in Sahiwal heifers. It is concluded that molasses supplementation in feed enhances the growth performance and reduces the non esterified fatty acid in both Sahiwal and Karan Fries heifers.

Keywords: Molasses, feeding, Karan Fries, Sahiwal

Molasses is the main byproduct of the sugarcane and is suitable source of energy. These are suitable way of supplying readily fermentable energy (glucose) to ruminants. Molasses and sugar supplementation to dairy ration can increase fiber digestibility, feed intake and lower blood urea nitrogen. After molasses feeding glucose level was increased due to production of propionate in the rumen (Sano *et al.*, 1999). Nutrient intake levels were causes variation in the metabolism of fat especially the mobilization of depot fat during low energy intake in the body. Due to this process there are changes in levels of non-esterified fatty acids (NEFA), β -hydroxybutyrate (BHBA), and glucose in the blood. NEFA and BHBA are released in the blood plasma when adipose tissue is mobilized to meet metabolic needs to the animals, primarily the needs of energy.

It is expected that blood urea nitrogen (BUN) would decrease upon constant protein intake and increasing dietary

energy (Chase *et al.*, 1993). The role of Triiodothyronine (T₃) on substrate metabolism was dependent on the amount of hormone available. The amount of available hormone was regulated by diet composition and calorie intake (Danforth and Burger, 1989). Dietary soluble carbohydrate is an important determinant of peripheral T₃ concentration. Thyroxin (T₄) and T₃ concentration have been shown to change following a meal in steers (Sticker *et al.*, 1996). Vaidya (2012) reported that the values of T₃ differed significantly (P<0.05) between breeds, seasons production levels in periparturient Sahiwal and Karan Fries cows.

Very scanty information are available on the effects of energy levels on the biochemical and hormonal parameters in Sahiwal and Karan Fries heifers, therefore, the investigations have been made to assess the effects of energy supplementation in the form of molasses.



MATERIALS AND METHODS

Climate and location

The experiment was conducted in the cattle yard of ICAR-National Dairy research Institute (NDRI), Karnal, Haryana, altitude of 250 meters above mean sea level and at 29°42' N latitude and 79°59' E longitude. The highest temperature goes up to 45 °C in summer and minimum temperature 3.5 °C to 4 °C in winter. The average rainfall is about 700 mm.

Animals and experimental design

Twelve healthy heifers (18-24 month) of each Sahiwal and Karan Fries were selected from the Livestock Research Centre (LRC) of ICAR- National Dairy Research Institute (NDRI), Karnal. The experimental animals were maintained as per the standard practices followed at the institute farm. These experimental animals were kept in separate shed throughout the entire experiment. Experiment was conducted after approval of the institutional animal ethics committee (IAEC). The heifers of both the breed were randomly divided into two groups based on their body weights so that the initial average body weight of each group was almost equal. The weight of the both the feeding regimes groups of the animals were taken on weekly intervals. The metabolic body weight and basal metabolic rate (BMR) was calculated with the help of formula:

$$M = W^{0.75} \text{ and } BMR = 70.5W^{0.734} \text{ respectively}$$

Where,

M = Metabolic body weight

BMR = The basal metabolic rate (kcal/day)

W = The body weight of the animal in kg.

Animals were let loose every week for exercise. The experiment on both groups of animals was conducted for 60 days under feeding regime-1 as per NRC, 2001 and feeding regime-2 (15% higher energy level over and above the NRC, 2001 by supplementation of molasses). The experimental animals were given adaptation period of 15 days prior to start of experiment. These animals were maintained on feeding regime-I for 15 days continuously and later on shifted to feeding regime-2 (15% higher

energy level) for next 15 days continuously. In between the both feeding regimes there were also given adaptation period of 15 days, so that animals become customized to the taste of molasses. The concentrate mixture was offered in the morning whereas, the chaffed green sorghum fodder was offered at 11:00 am. Concentrate mixture (CP 19.81% and TDN 70%) containing maize 33%, groundnut cake (oiled) 21%, mustard oil cake (oiled) 12%, wheat bran 20%, deoiled rice bran 11%, mineral mixture 2% and common salt 1%. The feeds offered to the animals and residue left were recorded fortnightly interval to find out the total dry matter intake (DMI) and ad-libitum water was given to the animals to find out the total water intake.

Collection of blood sample

The blood sample for biochemical parameters (glucose, BHBA, NEFA and BUN) and hormonal parameters (T_3 and T_4) was collected via jugular vein puncture into heparinized vacuum tubes on 1st, 7th, 14th, 15th and 16th day of each feeding regimes. Blood samples remained on ice until plasma was harvested following centrifugation at $1,500 \times g$ for 15 min and subsequently frozen at -20°C until analysis.

Estimation of Biochemical and Hormonal parameters

Glucose and β -Hydroxy butyrate were determined in plasma by using glucose and β -hydroxy butyrate, colorimetric assay kits, supplied by Cayman chemical company. Plasma NEFA and BUN were determined by using respective Bovine ELISA kits, supplied by Genx bio health sciences. Triiodothyronine (T_3) and Thyroxin (T_4) were determined in plasma using respective Bovine RIA Kits supplied by Beckman Coulter chemical company.

Statistical analysis

Data were analyzed using one way analysis of variance (ANOVA) by Statistical Analysis System (SAS, 2011) Software Programme, version 9.1 and results were expressed as mean \pm SE and considered statistically significant at 5% level.

RESULTS AND DISCUSSION

The result of biochemical and hormonal parameters of

Table 1: Biochemical and hormonal parameters in Sahiwal heifers under two different feeding regimes

Feed	BW(kg)	MBW(kg)	BMR(kg)	Glucose (mg/dl)	BHBA (mmol/L)	NEFA (μ mol/litre)	BUN (mmol/litre)	T ₃ (nmol/litre)	T ₄ (nmol/litre)
1	167.50 ^b ±4.08	46.54 ^b ±0.85	3023.36 ^b ±54.20	59.06 ^b ±1.28	0.242 ^a ±0.003	145.13 ^a ±5.12	9.65 ^a ±0.31	1.87 ^b ±0.09	50.76 ^b ±0.51
2	221.50 ^a ±7.91	57.38 ^a ±1.53	3710.45 ^a ±96.89	65.97 ^a ±1.15	0.237 ^a ±0.003	139.23 ^a ±8.73	7.52 ^b ±0.14	2.22 ^a ±0.09	58.60 ^a ±1.15

BW=Body weight; MBW=Metabolic body weight; BMR=Basal metabolic rate; BHBA= β -hydroxybutyrate; NEFA= Non-esterified fatty acids; BUN=Blood urea nitrogen; T₃= Triiodothyronine; T₄= Thyroxine

Means showing different superscripts in column differ significantly at 5% ($P<0.05$) whereas means of BW; MBW and BMR differs at 1% ($P<0.01$)

Table 2: Biochemical and hormonal parameters in Karan fries heifers under two different feeding regimes

Feed	BW(kg)	MBW(kg)	BMR(kg)	Glucose (mg/dl)	BHBA (mmol/L)	NEFA (μ mol/ litre)	BUN (mmol/ litre)	T ₃ (nmol/ litre)	T ₄ (nmol/ litre)
1	221.83 ^b ±15.63	57.34 ^b ±3.05	3707.61 ^b ±193.51	58.43 ^b ±1.08	0.261 ^b ±0.01	170.18 ^b ±4.83	11.66 ^a ±0.21	2.23 ^b ±0.09	57.02 ^b ±1.83
2	297.00 ^a ±13.47	71.47 ^a ±2.43	4599.94 ^a ±153.66	63.88 ^a ±0.87	0.259 ^b ±0.01	167.12 ^b ±4.14	9.66 ^b ±0.17	2.58 ^a ±0.09	64.74 ^a ±2.14

Means showing different superscripts in column differ significantly at 5% ($P<0.05$) whereas means of BW; MBW and BMR differs at 1% ($P<0.01$)

Sahiwal and Karan Fries heifers during feeding regime-1 and 2 are presented in Table 1 and 2. The mean body weight and metabolic body weight ($W^{0.75}$) of Sahiwal and Karan Fries heifers was significantly ($P<0.01$) higher in feeding regimes-2 as compared to feeding regime-1. Blood glucose and thyroid hormones (T₃ and T₄) were significantly ($P<0.05$) higher in feeding regimes-2 in both the breeds, whereas plasma NEFA and BHBA level was higher in feeding regime-1 than feeding regime-2, however, the values were found statistically non-significant ($p>0.05$). BUN was significantly ($P<0.05$) lower during feeding regime-2 compared to feeding regime-1 in both Sahiwal and Karan Fries heifers. Further, the mean values of the biochemical parameters (BHBA, BUN, and NEFA), hormonal parameters (T₃ and T₄) were significantly ($p<0.05$) higher in Karan Fries than Sahiwal heifers in both feeding regimes.

Plasma NEFA and BHBA levels are recognized as indicators for body fat mobilization and negative energy balance in dairy animals (Butler and Smith, 1989; Butler *et al.*, 2003). Decreased plasma glucose levels have been associated with low energy intake (Veenhuizen *et al.*, 1991) and increase as heifers were supplied more readily soluble carbohydrate (molasses) and progress towards a

more positive energy balance, (Jorritsma *et al.*, 2003). Effects of feeding extra glycogenic nutrients on plasma NEFA, BHBA, and glucose levels are presented in Table 1 and 2. Kanjanapruthipong *et al.* (2010) observed that low caloric intake during pre- partum period resulted in greater concentration of BHBA during postpartum in dairy cows. Feeding concentrate containing 68% of sorghum especially 6 weeks prior to parturition while increasing ration energy and protein levels during the last week before calving lead to decrease in NEFA and BHBA concentration and increase in serum glucose concentration (Strzetelski *et al.*, 2010).

Studies carried out by Knegsel *et al.* (2010) also indicated the lower NEFA levels due to feeding of glycogenic nutrients to dairy cattle. Vanderhaar *et al.* (1999) showed a negative correlation between blood NEFA level and the energy balance in cattle. Drackley *et al.* (2003) found a decrease in plasma NEFA and BHBA levels after feeding extra glycogenic nutrients. Plasma glucose concentrations were increased after feeding extra glycogenic nutrients (Miyoshi *et al.*, 2001). Ruppert *et al.* (2003) found no increase in BHBA, as an indicator of NEB, when fat was added to a corn silage diet, in contrast to an alfalfa silage diet. Since corn silage is mainly glycogenic (a high



proportion of C3 nutrients) and alfalfa silage is mainly lipogenic (a high proportion of C2 nutrients), it seems logical that BHBA levels on a corn silage diet are less increased than BHBA levels in cows on an alfalfa silage diet upon fat addition. In the corn silage diet the extra C2 nutrients of the fat addition are easier to metabolize because of the higher availability of C3 nutrients, compared to the alfalfa silage diet. It seems that the effect of dietary fat addition depends on the nature of other nutrients in the diet, suggesting the C2/C3 balance to be a factor in the concentration of plasma BHBA. Lower NEFA and higher glucose levels were reported in cows fed with steam-flaked sorghum compared to steam-rolled corn (Santos *et al.*, 2000). This can be explained by a higher ruminal starch digestibility and resulting higher ruminal propionate secretion of steam-flaked sorghum compared to steam-rolled corn. This was in line with Simas *et al.* (1995) who found elevated blood glucose levels in cows fed steam-flaked sorghum, compared to dry rolled sorghum, which has lower ruminal starch degradability.

To monitor the body weight changes and body condition score, BUN can be a useful tool for monitoring the protein energy status of cattle. Increasing dietary energy intake while holding protein intake constant would be expected to decrease BUN. This was demonstrated in an experiment with bulls, where diets were provided with 75 and 150% of maintenance energy requirement but equal CP intake. It was found that at the high and low level of energy intake BUN averaged 5.6 mg/dL and 19.7 mg/dL, respectively (Chase *et al.*, 1993). Increased energy intake in feed are associated with reduced BUN (Thomas and Kelly, 1976; Tyagi *et al.*, 2009 and Lien *et al.*, 2010). Quantitatively, biological activity of T_3 is higher than T_4 (Oppenheimer *et al.*, 1987). The positive correlation between circulating thyroid hormone concentration and energy balance is well known in many species including cattle (Cassar-Malek *et al.*, 2001). Energy intake in lab rodents appears to suppress TRH expression in the PVN. Fall in TSH production and alteration in pattern of glycosylation on newly synthesized TSH leads to reduced TSH bioactivity. Thus, as a consequence of low energy intake, fall in T_3 and T_4 levels, leads to central hypothyroidism (Flier *et al.*, 2000). The dominant signal for the suppression of TRH expression in the PVN of the brain is an energy-induced drop in the level of leptin hormone (Flier *et al.*, 2000). Leptin mediate neuroendocrine responses by energy supply or

deprivation via acting through hypothalamic nerve centers (Zhang *et al.*, 1994). The results of the present study are in the line with the values reported by (Capuco *et al.*, 2001; Cassar-Malek *et al.*, 2001) i.e. positive correlation between circulating thyroid hormone concentrations and energy balance in many species of livestock. Singh and Upadhyay (1997) also reported breed differences in blood glucose levels and indicated higher energy demand for the crossbred cows.

CONCLUSION

From the study it is concluded that molasses supplementation in feed, increased body weight, metabolic body weight, basal metabolic rate, glucose, thyroid hormones and reduced blood urea nitrogen in both Sahiwal and Karan Fries heifers and non-esterfied fatty acids were reduced in feeding regimes-2.

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