IJAS Iranian Journal of Applied Animal Science

Growth Performance, Feeding Behavior and Physiological Responses of Young Growing Holstein Male Calves to Dietary Chromium-Methionine (Cr-Met) Supplementation Related to Body Weight and Age

Research Article

M. Yari[1*], M. Baharifar[2], A. Alizadeh Masuleh[3] and A. Mousaie[4]

[1] Department of Animal Science, Malayer University, Malayer, Iran
[2] Department of Animal Science, Islamic Azad University, Saveh Branch, Saveh, Iran
[3] Department of Embryology, Reproductive Biomedicine Research Center, Royan Institute for Reproductive Biomedicine, Academic Center for Education, Culture and Research (ACECR), Tehran, Iran
[4] Department of Animal Science, University of Jiroft, Jiroft, Iran

Received on: 27 Sep 2017
Revised on: 9 Nov 2017
Accepted on: 30 Nov 2017
Online Published on: Sep 2018

*Correspondence E-mail: m.yari@malayeru.ac.ir

Online version is available on: www.ijas.ir

ABSTRACT

This study was conducted to investigate the effect of dietary chromium-methionine (Cr-Met) supplementation on growth performance, feeding behavior and physiological responses of Holstein male calves during two different phases of body weight (BW). Twenty six (153.5±9.83 kg of BW) male calves were divided in two groups (n=13) and randomly assigned to treatments: 1) control and 2) Cr-Met (1.1 mg Cr/kg of diet on a dry matter (DM) basis). Each phase consisted of 1 week pen acclimatization, 3 weeks diet adaptation and 1 week sample collection. After the first phase, all calves received the basal diet (without Cr-Met) for 6 weeks. After this period, the second phase started (240.1±12.15 kg BW). During the second phase treatments changed between 2 groups. Calves fed with Cr-Met diet had lower DM intake and average daily gain ($P<0.05$) and rectal temperature ($P<0.01$) than the control group, whereas blood metabolites and feeding behaviors were similar between 2 groups during the first phase of experiment. During the second phase, calves fed with Cr-Met had greater DM intake, weight gain, average daily gain and standing time behavior ($P<0.05$) and lower blood glucose concentration, rectal temperature and resting time behavior ($P<0.05$) compared with calves fed with control diet. In conclusion, effect of feeding 1.1 mg Cr/kg DM as Cr-Met supplement to Holstein male calves on growth performance, blood glucose and resting and standing behavior might be related to body weight.

KEY WORDS blood glucose, chromium methionine, feeding behavior, Holstein male calves.

INTRODUCTION

Trivalent chromium (Cr) is an essential micro mineral for human and animals. Chromium potentiates insulin function via enhancing its binding to target cell receptors and also through improving its post-receptor signaling (Kegley *et al.* 2000; Vincent, 2001). Chromium in feed is weakly absorbed in animals, therefore dietary Cr supplementation in various chemical forms has been investigated over the past two decades. Supplementing Cr from highly bioavailable resources may provide the Cr requirement (Spears *et al.* 2012). Addition of Cr to diet has been demonstrated to increase blood glucose clearance rate and tissue insulin sensitivity (Kegley *et al.* 2000; Spears *et al.* 2012), enhance performance (Moonsie-Shageer and Mowate, 1993), and improve the immune response (Chang and Mowate, 1992) of growing calves mostly under stressed condition regardless of its supplemented source.

In human, greater blood free fatty acid concentration in obese people as a result of fat accumulation in non-adipose tissues (in particular liver and muscle) is the key factor to lower insulin sensitivity of peripheral tissues. Moreover, insulin resistance may attenuate the weight gain of growing animals. Insulin increases cellular glucose uptake, stimulates glycolysis, and promotes the hepatic and muscle carbohydrate, fat and protein synthesis (Baumgard *et al.* 2016). However, insulin sensitivity decreases throughout the first year of life in human (Ryan, 2000; Chen and Hess, 2008) and ruminant animals (Clarke *et al.* 2000). Eisemann *et al.* (1997) reported the decreased glucose metabolism and insulin sensitivity at heavier body weights in Holstein beef steers. Furthermore, as percentage of fat in the body tissues increased by age and weight, insulin sensitivity of tissues decreased in heifers (McCann and Reimers, 1985b).

The hypothesis of present research was that in Holstein male calves undergoing high weight gain rate, the insulin function might decline because of insulin resistant with advancing body weight and age. Therefore, the Cr requirement might increase. The objectives of current study were to investigate the effects of feeding chromium-methionine (Cr-Met) on performance, feeding behavior and physiological responses of young male calves during two phases of body weight and age.

MATERIALS AND METHODS

Experimental design, diets and animals' management

Twenty-six growing Holstein male calves with average initial body weight of 153.7 ± 9.8 kg were used in this study. The calves were randomly assigned to two experimental groups (13 calves per each) including control (no Cr-Met supplementation) and Cr-Met group (receiving 1.1mg/kg of DM of Cr^{3+} from Availa Cr provided by Zinpro Company, USA). Basal diet (Table 1) formulated according to NRC (2001) for weaned calves. Each pen (23×9 m^2) consisted of two sections of shaded (8×9 m^2) and unshaded parts. Calves were cared for according to the guidelines of the Iranian Council of Animal Care (1995). The Current study conducted in 2 different phases which the first phase lasted from August 19 to September 24 and the second from November 5 to December 10 in 2013. During an adaptation period of 10 days, calves in each pen fed with basal diet (Table 1) without any Cr-Met supplementation. Body weight of each calf at the end of adaptation period recorded and used as covariate in final statistical model (Robinson *et al.* 2006).

After the adaptation period, calves received experimental diets for 21 days followed by 7 days of sample collection including group daily DM intake recording, blood sample collection, feeding behavior and body weight measurements (Robinson *et al.* 2006). After the first phase of experiment, all calves received the basal diet without Cr-Met addition for 6 weeks and then second phase of experiment started. Duration of second phase of the experiment and data collection procedures were similar to the first phase exception for Cr-Met addition, which switched between groups. Calves had free access to water throughout the study. During the current study, mean of weather temperature and maximum temperature humidity index (THI) were respectively 30 ˚C and 70 at the first phase and 15 ˚C and 57 for the second phase (Yari *et al.* 2010).

Table 1 Ingredients and chemical composition of basal diet

Ingredients	% of dry matter
Alfalfa hay (early flower)	50.0
Corn grain	13.0
Barley grain	13.0
Wheat bran	13.0
Canola meal	9.5
Sodium bicarbonate	0.3
Calcium carbonate	0.2
Salt	0.2
Mineral-vitamin mixture	0.8
Chemical composition[1]	
Crude protein (CP)	15.2
Neutral detergent fiber (NDF)	37.0
Non-fiber carbohydrates (NFC)[2]	39.0
Ether extract (EE)	3.2
Metabolizable energy (Mcal/kg DM)	2.34
Net energy for gain (Mcal/kg DM)	0.90

[1] Calculated with NRC (2001) software.
[2] NFC calculated as: NFC= 100 - (CP+NDF+EE+ash).

Dry matter intake, body weight gain and feed conversion ratio

Body weight gain was calculated as final individual body weight minus initial body weight. Average daily group DM intake during 7 days of sampling period within each phase was measured and was used as replication to compare statistically effects of treatments. To calculate individually DM intake daily group DM intake was divided by the number of calves within each pen. The feed conversion ratio (FCR) was determined for each period using individual body weight gain divided by average of group DM intake measured during 7 days (Robinson *et al.* 2006).

Rectal temperature, blood metabolites and feeding behavior measurements

Five calves from each pen were randomly selected and numbered for further measurements of rectal temperature, blood sampling and feeding behaviors.

Rectal temperature measurements performed before collecting blood samples with digital thermometer as described by Yari *et al.* (2010). Blood samples were drawn from the jugular vein of each calf between 11:30 and 12:30 h on day 27 in each period. Blood samples were immediately placed on ice to coagulate and moved to the laboratory and centrifuged at 3500 g for 15 min. Serum concentrations of total protein, glucose and blood urea nitrogen (BUN) were measured using an auto analyzer system (Biotecnica, Targa 3000, Rome, Italy) with commercial kits and associated procedures (Pars Azmon Co., Tehran, Iran). Serum insulin concentration was analyzed using ELISA method according to the insulin analysis kit (Insulin ELISA kit, monobindinc. Lake Forest CA USA) instruction.

Feeding behaviors were continuously monitored for 24 h at day 27 of each period as described by Yari *et al.* (2014). Feeding behavior was defined every 5 min as resting, eating, ruminating and standing.

Total time (min) spent on each activity was quantified by multiplying the total number of observations for that activity by 5.

Statistical analysis

Data from each phase was separately analyzed using mixed procedure of SAS 9.2 (SAS, 2003) by following statistical models:

$$Y_{ij}= \mu + T_i + B_j + e_{ij}$$
$$Y_{ijk}= \mu + T_i + B_j + D_k + FBW_l + e_{ijk}$$

Where:
Y (Y_{ij} and Y_{ijk}): observation of the dependent variable.
μ: fixed effect of population mean for the variable.
T_i: fixed effect of treatment (i=2; control and Cr-Met).
B_j: random effect of calf within treatment (j=13).
D_k: fixed effect of day (k=7; 1-7).
FBW_l: fixed effect of first body weight (l=13).
e (e_{ij} and e_{ijk}): random error associated with the related observation.

Model 1 was used for analysis of feeding behaviors, rectal temperature and blood metabolites. Experimental replicates were numbered calves within each pen during each period (n=5). Model 2 was used for analysis of DM intake which day of sampling (n=7) used as replication. Final body weight, body weight gain and FCR were analyzed by model 2 without the effect of day. The first body weight used as covariate in this model to improve the precision of analysis. The adjust Tukey test was used for multiple treatment comparisons using the LSMEAN statement of SAS 9.2 (SAS, 2003). The statistical differences were considered significance at $P \leq 0.05$ and trends at $P \leq 0.10$.

RESULTS AND DISCUSSION

Growth performance responses

The results of growth performance are shown in Table 2. Initial body weight of calves was not statistically different. During first phase, calves fed with diet supplemented with Cr-Met tended to have lower final body weight (P=0.06) and decreased DM intake, total weight gain and average daily gain (P<0.05), whereas FCR was similar between 2 group calves (P=0.16). At the beginning of second phase, body weight of calves was statistically similar. Calves fed diet with Cr-Met-supplemented had greater DM intake, body weight gain, average daily gain (P<0.05) and tended to have greater final body weight (P=0.06) while had similar FCR compared with calves fed diet without Cr-Met (P=0.16) during the second phase of experiment.

Feeding behaviors

During first phase, feeding behavior measurements were similar between 2 experimental groups (Table 3). Calves fed with Cr-Met diet had lower resting time and greater standing time behavior during second phase of the experiment compared with calves fed with control diet (P<0.05; Table 3).

Blood metabolites and rectal temperature

During the first phase, blood metabolites concentrations were similar between 2 group calves (Table 4). Calves fed with dietary supplemental Cr-Met had lower rectal temperature compared with calves fed control diet (P<0.01; Table 4). In addition, rectal temperature of calves decreased by feeding Cr-Met diet in the second phase (P<0.01). Calves fed with Cr-Met supplemented diet had lower blood glucose concentration (P<0.05) whereas other blood metabolites were similar between 2 groups in the second phase of experiment (Table 4).

The magnitude of Cr used in the present study as Cr-Met supplement (1.1 mg/kg DM) was below the maximum tolerable level for cattle which has been estimated at 100 mg of Cr/kg of DM (Spears *et al.* 2012).

During the first phase of study, there was a difference in the rate of body weight gain between 2 groups of calves while feeding behaviors and blood metabolites were similar. The relationship between Cr supplementation and body weight gain is still controversial. It has been resulted that using Cr supplementation in diet increased average daily gain in stressed, receiving feedlot calves (Chang and Mowate, 1992; Moonsie-Shageer and Mowate, 1993; Kegleyet *al.* 1997). Calves received supplemental dietary Cr yeast had 30% (Chang and Mowate, 1992) and 27% (Moonsie-Shageer and Mowat, 1993) increases in average daily gain compared with those not fed with Cr.

Table 2 Growth Performance of Holstein young calves fed supplemental chromium methionine (Cr-Met) at different body weights

Item	**Treatment**		**SEM**	**P-value**
	Control	Cr-Met		
First period (body weight range from 150 to 190 kg)				
Initial BW (kg)	159.9	147.6	9.83	0.39
DMI (kg)	7.1^a	6.1^b	0.158	< 0.01
Final BW (kg)	188.6	181.5	2.47	0.06
BW gain (kg)	35.3^a	27.4^b	2.49	0.03
ADG (kg)	1.3^a	1.0^b	0.09	0.03
FCR	6.81	7.64	0.933	0.53
Second period (body weight range from 220 to 260 kg)				
Initial BW (kg)	223.7	242.0	12.15	0.29
DMI (kg)	8.5^b	8.8^a	0.06	< 0.01
Final BW (kg)	255.8	260.1	1.530	0.06
BW gain (kg)	22.8^b	27.5^a	1.50	0.04
ADG (kg)	1.08	1.31	0.071	0.04
FCR	9.05	7.96	0.539	0.16

BW: body weight; DMI: dry matter intake; ADG: average daily gain and FCR: feed conversion ratio.
The means within the same row with at least one common letter, do not have significant difference (P>0.05).
SEM: standard error of the means.

Table 3 Feeding behaviors of Holstein male calves fed diet supplemented with chromium methionine (Cr-Met) at different body weights

Items (minute/day)	**Treatments**		**SEM**	**P-value**
	Control	Cr-Met		
First period (body weight range from 150 to 180 kg)				
Eating	276.0	256.0	19.99	0.50
Rumination	397.0	426.0	16.05	0.24
Resting	376.8	381.2	21.51	0.89
Standing	390.0	377.0	17.20	0.60
Second period (body weight range from 220 to 260 kg)				
Eating	254.0	279.0	19.93	0.40
Rumination	413.0	395.0	16.45	0.46
Resting	435.0^a	345.0^b	26.97	0.05
Standing	338.0^b	421.0^a	14.14	< 0.01

The means within the same row with at least one common letter, do not have significant difference (P>0.05).
SEM: standard error of the means.

Table 4 Blood metabolites and rectal temperature of Holstein young calves fed with chromium methionine (Cr-Met) supplemented diet at different body weights

Item	**Treatments**		**SEM**	**P-value**
	Control	Cr-Met		
First period (body weight range from 150 to 180 kg)				
Glucose (g/dL)	75.6	78.6	3.55	0.85
Insulin (µIU/mL)	12.4	11.8	3.12	0.75
Ins:glu	0.17	0.15	0.040	0.40
Urea (g/dL)	22.2	23.9	1.380	0.27
Total protein (g/L)	8.7	8.7	0.17	0.80
Rectal T (˚C)	39.9^a	38.7^b	0.18	< 0.001
Second period (body weight range from 220 to 260 kg)				
Glucose (g/dL)	83.8^a	70.2^b	3.79	0.04
Insulin (µIU/mL)	13.7	6.8	3.13	0.22
Ins:glu	0.17	0.10	0.035	0.38
Urea (g/dL)	24.0	25.4	1.38	0.55
Total protein (g/L)	7.9	7.6	0.21	0.35
Rectal T (˚C)	39.4^a	38.8^b	0.12	< 0.001

Ins:glu: insulin to glucose ratio and T: temperature.
The means within the same row with at least one common letter, do not have significant difference (P>0.05).
SEM: standard error of the means.

Similarly, calves fed with Cr-nicotinic acid complex also displayed increased average daily gain by 11% (Kegley *et al.* 1997) comparing with those on the control diet.

Some studies have reported no differences in average daily gain, DM intake, FCR and final body weight of steers fed either 0.0 or 0.2 mg Cr/kg DM as Cr yeast (Chang and

Mowate, 1992) or 3 mg Cr/kg DM as a Cr amino acid chelate (Mathison and Engstrom, 1995). In addition, steers fed 0.4 mg Cr/kg DM had decreased average daily gain, DM intake, FCR, final body weight when compared to those fed 0.0 and 0.2 mg Cr/kg DM (Pollard *et al.* 2002). Feeding 3 mg Cr/kg DM as a Cr amino acid chelate (Mathison and Engstrom, 1995) decreased hot carcass weight when compared to the control diet. Our results from the first phase are in consistent with findings of Pollard *et al.* (2002) who reported that organic Cr supplementation in the level of 0.4 ppm decreased average daily gain, DM intake, FCR and final body weight.

At the second phase of this study, calves fed with diet supplemented with Cr-Met, with heavier body weight, had lower blood glucose concentration, numerically lower insulin and insulin/glucose ratio compared with calves in control group. Environmental condition such as heat stress, basal diet composition and physiological stage can affect the insulin function and glucose metabolism in cattle (McCann and Reimers, 1985b; Yari *et al.* 2010; Spears *et al.* 2012). The recorded THI during this study might be considered as mild (during first phase) to weak (during second phase) heat stress for young calves. Chromium-methionine feeding during heat stress has been reported to improve performance in dairy cows (Nikkhah *et al.* 2011) and dairy calves (Yari *et al.* 2010) as a result of improved insulin function. Forbes (1980) explained that with increased fatness and days on feed, adipocytes have been reduced ability to remove glucose from the blood because of their decreased sensitivity to insulin. Radunz *et al.* (2012) also stated that steers had increased insulin resistance as they were longer on a corn-based finishing diet. Spears *et al.* (2012) reported that heifers supplemented with Cr at the level of 0.47 mg/kg DM from Cr propionate for 43 d were more insulin sensitive because of decreased insulin, glucose concentration and insulin to glucose ratio. Feeding Cr-Met at the level of 0.02 and 0.04 mg/kg $BW^{0.75}$ (0.25 and 0.50 mg/kg DM, respectively) improved tissue insulin responses through glucose tolerance test (Yari *et al.* 2010).

In a study on growing calves with functional rumen, feeding Cr as Cr-Met (BW range from 288 to 312 kg) at two levels of 0.4 and 0.8 ppm, did not change final body weight and DM intake while glucose clearance rate after an insulin infusion and insulin response to an intravenous glucose challenge increased (Kegley *et al.* 1997). These variable results may reflect differences in Cr status of the calves, the amount of stress to which the calves had been exposed, physiological stage, body weight and age, the amount and bioavailability of Cr in the basal diet, or the bioavailability of the supplemental Cr sources (Kegleyet *al.* 1997; Spears *et al.* 2012; Yari *et al.* 2010).

In the study of Chang and Mowat (1992) and Moonsie-Shageer and Mowate (1993), feeding Cr at the level of 0.2 and 0.4 ppm improved DM intake and average daily gain in under-shipment stressed young calves but in pre-weaning calves, feeding 1 ppm Cr did not influence DM intake and average daily gain (Depew *et al.* 1998). In some studies, feeding Cr did not affect DM intake and average daily gain (Kegley *et al.* 1997; Kegley *et al.* 2000; Depew *et al.* 1998) mainly because of Cr levels and sources in diet, severity of stress and physiological stage of animal. Feeding growing finishing pigs with 0.025, 0.05, 0.10 and 0.2 ppm of Cr supplements improved linearly dry matter intake (DMI) and average daily gain (ADG) while feeding at the level of 0.4 and 0.8 ppm decreased DM intake and average daily gain (Page *et al.* 1993). Close to the Cr dosage used in current study, Mousaie *et al.* (2014) reported that feeding 0.8 mg Cr-Met/kg DM in growing lambs under transportation stress resulted in increased DM intake and average daily gain and improved FCR. Current data provide evidence that 1.1 mg Cr/kg DM can be effectively used for improve DM intake, body weight gain and average daily gain in calves with > 200 kg of body weight.

Blood metabolites

Calves fed with diet supplemented with Cr-Met during the second phase of this experiment had lower blood glucose concentration. Blood glucose level can be a direct index of energy metabolism in the body of animal. The level of circulating insulin in cattle is strongly correlated with the body weight gain rate, and feed restriction would result in a decline in its concentration (Hayden *et al.* 1993; Kegley *et al.* 2000; Spears *et al.* 2012). Decreased glucose concentration in calves fed with Cr-Met during the second phase may be due to their increased insulin sensitivity. Hayden *et al.* (1993) reported decreased plasma concentration of glucose corresponded with a decreased concentration of insulin in steers. In the second phase, decreased numerically insulin concentration and significantly glucose concentration in calves fed with Cr-Met may indicate higher disposal rate of glucose from peripheral blood by tissues through increased tissue insulin sensitivity. This higher glucose uptake by tissues may be a possible explanation for better growth performance of calves fed supplemental Cr-Met during the second phase of experiment.

Calves fed with both experimental diets had similar blood total protein and urea concentrations. Our findings are in consistent with results of Moonsie-Shageer and Mowate (1993) who reported that feeding Cr supplement at the level of 0.2, 0.4 and 1 ppm did not cause changes in blood urea and total protein concentration of stressed calves.

However, Kegley *et al.* (2000) showed that feeding 0.4 and 0.8 ppm of Cr-Met increased blood urea at 2 h after feeding time. In current study, a lack of difference between 2 groups in the case of blood urea and total protein may be related to the sufficient supply of protein in the body, balanced protein to energy ratio and appropriate ruminal nitrogen metabolism (Kegley *et al.* 2000).

Feeding behavior and rectal temperature

Feeding behaviors in the first phase were similar among calves but during the second phase, calves fed with Cr-Met supplement had lower resting time and higher standing time. There are little information about the reasons for different types of behavior in calves. However, the possible reason for decreased resting (lying) time may be due to bedded floor system and space allowance per each animal.

In a study conducted on heifers (Mogensen *et al.* 1997) a correlation between lying periods and daily gain was found. In that study, heifers housed on slatted floor with few lying periods also had a low daily gain. They demonstrated that heifers with changing position problems also had reduced gain. However, in the current study there were enough allowance space and deep-bedded system. Moreover, calves on Cr-Met diet had improved growth performance at the second phase of experiment. Therefore, decreased resting time and increased standing time might be related to other animal physiological responses, which are unknown for us, and remain to be elucidate in future studies.

There are a few evidences regarding the effects of dietary Cr supplements on feeding behavior and rectal temperature of calves. Szyszka *et al.* (2012) in a study on steers showed that the lying behavior of steers challenged with lipopolisacharides (LPS); as an acute health challenger) and parasite *Ostertagia ostertagi* (as a chronic health challenger) lasted while rectal temperature did not change. Induced acute and chronic stress in their study had time dependent effect on body weight gain. In LPS-treated animals, standing and lying time were not affected while, *O. ostertagi*-challenged animals, showed increased standing and lying periods and average meal duration (Szyszka *et al.* 2012). Over the last decade, there has been a growing scientific interest in feeding behavior of cattle especially dairy cows. Mechanisms controlling feed intake are dependent on the interaction between diet and physiological state of animals. Age, pregnancy, lactation, and adiposity are main factors affect physiological state. That is characterized by differences in insulin sensitivity of tissues and plasma levels of insulin and likely other hormones such as growth hormone, and leptin (Allen and Piantoni, 2014). Improved performance of calves fed with Cr-Met in the second phase of this study with reduced blood glucose might likely indicate that peripheral tissues have used nutrients with more efficiency that have been resulted to change in resting and standing behavior time.

Calves fed with Cr-Met had lower rectal temperature during 2 experimental phases. Physiological and hormonal changes in the body during stress condition may result in increased rectal temperature (Moonsie-Shageer and Mowate, 1993; Yari *et al.* 2010). Chromium metabolism in animal and human body is strongly influenced by stress condition, its intensity and duration. In the study of Arthington *et al.* (1997), feeding Cr did not reduce rectal temperature when calves challenged with type 1 hepatitis B virus (HBV-1) and rectal temperature increased (Arthington *et al.* 1997). However, our results are in consistent with findings of Moonsie-Shageer and Mowate (1993) who reported feeding Cr to calves challenged with pig red blood cell and calves under stress of shipment, rectal temperature decreased. The reduction in rectal temperature in calves fed with Cr may be because of Cr impact on lowering cortisol in stressed calves (Moonsie-Shageer and Mowate, 1993; Yari *et al.* 2010; Ghorbani *et al.* 2012). The mean of THI of 70 for the first phase and 57 for the second phase of current study could not be considered as heat stress condition for young calves (Yari *et al.* 2010). Therefore, it seems that calves in this study did not undergo stress condition and reduction in rectal temperature in response to feeding Cr-Met may be related to other physiological effects of Cr which are remained to be clarified. Altogether, current data and previous results (Yari *et al.* 2010; Ghorbani *et al.* 2012) support the hypothesis that Cr-Met effects on calves performance, physiological responses and feeding behaviors dependent mainly on age, body weight and rate of weight gain.

CONCLUSION

Growing young Holstein male calves responses such as dry matter intake, body weight gain, blood glucose, rectal temperature as well as standing and resting feeding behavior to Cr-Met supplementation with the level of 1.1 mg Cr/kg DM in a diet with forage to concentrate ratio of ~ 50:50 under normal condition changed as their body weight and age increased. These data may indicate that glucose metabolism of young male Holstein calves differ as their body weight increase and therefore may influence on Cr requirements.

ACKNOWLEDGEMENT

The authors thank the Islamic Azad University, Saveh Branch for lab equipment and partial financial support of the current study. The authors also thank the Sana Dam Pars company (Dr Pejman Ajilchi) to provide Availa Cr and partial financial support of this research.

REFERENCES

Allen M.S. and Piantoni P. (2014). Physiological mechanisms controlling feeding behavior. *J. Anim. Sci.* **92,** 935-942.

Arthington J.D., Corah L.R., Minton J.E., Elsasser T.H. and Blecha F. (1997). Supplemental dietary chromium does not influence ACTH, cortisol, or immune response in young calves inoculated with bovine herpesvirus-1. *J. Anim. Sci.* **75,** 217-223.

Baumgard L.H., Hausman G.J. and Sanz Fernandez M.V. (2016). Insulin: Pancreatic secretion and adipocyte regulation. *Domest. Anim. Endocrinol.* **54,** 76-84.

Chang X. and Mowat D.N. (1992). Supplemental chromium for stressed and growing feeder calves. *J. Anim. Sci.* **70,** 559-565.

Chen X. and Hess S. (2008). Adipose proteome analysis: focus on mediators of insulin resistance. *Expert Rev Proteomic*. **5,** 827-839.

Clarke L., Firth K., Heasman L., Juniper D.T., Budge H., Stephenson T. and Symonds M.E. (2000). Influence of relative size at birth on growth and glucose homeostasis in twin lambs during juvenile life. *Reprod. Fertil. Dev.* **12,** 69-73.

Depew C.L., Bunting L.D., Fernandez J.M. and Tompson D.L. (1998). Performance and metabolic response of young dairy calves fed diets supplemented with chromium Tripicolinate. *J. Dairy Sci.* **81,** 2916-2923.

Eisemann J.H., Hantington G.B. and Catherman D.R. (1997). Insulin sensitivity and responsiveness of portal-drained viscera, liver, hindquarters and whole body of beef steers weighing 275 or 490 kilograms. *J. Anim. Sci*. **75,** 2084-2091.

Forbes J.M. (1980). Hormones and metabolites in the control of food intake. Pp. 145-160 in Digestive Physiology and Metabolism in Ruminants. Y. Ruckebusch and P. Thivend, Eds. Springer, Dordrecht, Netherlands.

Ghorbani A., Sadri H., Alizadeh A.R. and Bruckmaier R.M. (2012). Performance and metabolic responses of Holstein calves to supplemental chromium in colostrum and milk. *J. Dairy Sci.* **95,** 5760-5769.

Hayden I.M., Williams J.E. and Collier J.J. (1993). Plasma growth hormone, insulin-like growth factor, insulin, and thyroid hormone association with body protein and fat accretion in steers undergoing compensatory gain after dietary energy restriction. *J. Anim. Sci.* **71,** 3327-3338.

Iranian Council of Animal Care. (1995). Guide to the Care and Use of Experimental Animals, vol. 1. Isfahan University of Technology, Isfahan, Iran.

Kegley E.B., Galloway D.L. and Fakler T.M. (2000). Effect of dietary chromium-L-methionine on glucose metabolism of beef steers. *J. Anim. Sci.* **78,** 3177-3183.

Kegley E.B., Spears J.W. and Brown T.T. (1997). Effect of shipping and chromium supplementation on performance, immunity response, and disease resistance of steers. *J. Anim. Sci.* **75,** 1956-1964.

Mathison G.W. and Engstrom D.F. (1995). Chromium and protein supplements for growing-finishing beef steers fed barley-based diets. *Canadian J. Anim. Sci.* **75,** 549-558.

McCann J.P. and Reimers T.J. (1985b). Insulin response to glucose in estrous and di-estrous obese and lean heifers. *J. Anim. Sci.* **61,** 619-625.

Mogensen L., Christian C.K., Tind S.J., Hindhede J. and Harder Nielsen L. (1997). Association between resting behaviour and live weight gain in dairy heifers housed in pens with different space allowance and floor type. *Appl. Anim. Behav. Sci.* **55,** 11-19.

Moonsie-Shageer S. and Mowate D.N. (1993). Effect of level of supplemental chromium on performance, serum constituents, and immune status of stressed feeder calves. *J. Anim. Sci.* **71,** 232-238.

Mousaie A., Valizadeh R., Naserian A.A., Heidarpour M. and Kazemi Mehrjerdi H. (2014). Impacts of feeding selenium-methionine and chromium-methionine on performance, serum components, antioxidant satus, and physiological responses to transportation stress of baluchi ewe lambs. *Biol. Trace Elem. Res.* **162,** 113-123.

Nikkhah A., Mirzaei M., Khorvash M., Rahmani H.R. and Ghorbani G.R. (2011). Chromium improves production and alters metabolism of early lactation cows in summer. *J. Anim. Physiol. Anim. Nutr.* **95,** 81-89.

NRC. (2001). Nutrient Requirements of Dairy Cattle. 7th Ed. National Academy Press, Washington, DC, USA.

Page T.G., Southern L.L., Ward T.L. and Thompson D.L. (1993). Effect of chromium picolinate on growth and serum and carcass traits of growing-finishing pigs. *J. Anim. Sci.* **71,** 656-662.

Pollard G.V., Richardson C.R. and Karnezos T.P. (2002). Effects of supplemental organic chromium on growth, feed efficiency and carcass characteristics of feedlot steers. *Anim. Feed Sci. Technol*. **98,** 121-128.

Radunz A.E., Fluharty F.L., Relling A.E., Felix T.L., Shoup L.M., Zerby H.N. and Loerch S.C. (2012). Prepartum dietary energy source fed to beef cows: II. Effects on progeny postnatal growth, glucose tolerance, and carcass composition. *J. Anim. Sci.* **90,** 4962-4974.

Robinson P.H., Wiseman J., Udén P. and Mateos G. (2006). Some experimental design and statistical criteria for analysis of studies in manuscripts submitted for consideration for publication. *Anim. Feed Sci. Technol.* **129,** 1-11.

Ryan A.S. (2000). Insulin resistance with aging: Effects of diet and exercise. *Sports Med.* **30,** 327-346.

SAS Institute. (2003). SAS®/STAT Software, Release 9.2. SAS Institute, Inc., Cary, NC. USA.

Spears J.W., Whisnant C.S., Huntington G.B., Lloyd K.E., Fry R.S., Krafka K., Lamptey A. and Hyda J. (2012). Chromium propionate enhances insulin sensitivity in growing cattle. *J. Dairy Sci.* **95,** 2037-2045.

Szyszka O., Tolkamp B.J., Edwards S.A. and Kyriazakis I. (2012). The effects of acute versus chronic health challenges on the behavior of beef cattle. *J. Anim. Sci.* **90,** 4308-4318.

Vincent J.B. (2001). The bioinorganic chemistry of chromium (III). *Polyhedron*. **20,** 1-26.

Yari M., Nikkhah A., Alikhani M., Khorvash M., Rahmani H. and Ghorbani G.R. (2010). Physiological calf responses to increased chromium supply in summer. *J. Dairy Sci.* **93,** 4111-4120.

Yari M., Valizadeh R., Naserian A., Jonker A., Azarfar A. and Yu P. (2014). Effects of including alfalfa hay cut in the afternoon or morning at three stages of maturity in high concentrate rations on dairy cows performance, diet digestibility and feeding behavior. *Anim. Feed Sci. Technol.* **192,** 62-72.