Effects of dietary chromium supplementation on blood biochemical constituents and enzyme activities in lactating crossbred cows

KUSUM RIKHARI, D. P. TIWARI and ANIL KUMAR

Department of Animal Nutrition, College of Veterinary and Animal Sciences, G. B. Pant University of Agriculture and Technology, Pantnagar-263145 (U. S. Nagar, Uttarakhand)

ABSTRACT: Crossbred lactating cows (16) were divided into 4 groups of 4 each and were supplemented with chromium at 0, 0.5, 1.0 and 1.5 mg per kg DM intake to discern the effect on blood biochemical constituents and enzyme activities. The feeding trial lasted for 90 days. Blood samples were collected from experimental cows at start and end of the feeding trial. The Cr-supplemented cows showed significantly higher blood glucose concentrations. There were no significant effects on the total protein, albumin, globulin, total cholesterol, total lipids and NEFA concentration in blood serum due to chromium supplementation. The Cr-supplemented cows showed significantly higher activities of serum glutamate pyruvate transaminase (SGPT) and serum glutamate oxaloacetate transaminase (SGOT) enzymes while the activity of alkaline phosphatase (ALP) remains unaffected.

Key words: Biochemical profile, blood, chromium, lactating crossbred cows.

The relevance of the trace element chromium (Cr) to human and animal nutrition has been known for more than 40 years. Chromium is present in feeds and fodder mostly in its trivalent stable form (Cr^{+3}). It is known for its in vivo antioxidative activity and favourable effects on the stability of proteins and nucleic acids (Anderson, 1994). Metabolically chromium enhances insulin activity by its presence in an organometallic molecule known as the glucose tolerance factor (GTF). A trivalent Cr^{+3} is important for inactive glucose tolerance factor. It is assumed that GTF consists of Cr^{+3}, nicotinic acid, glutamic acid, glycine and cysteine (Toepfer *et al*., 1977). Chromium enhances binding of insulin to cell membrane receptors and the optimization of insulin activity results in better regulation of glucose uptake by cells, improved control of blood glucose concentration and maximization of the energetic potential. Consequences of Cr deficiency which is probably associated with disturbed interaction between Cr and insulin includes lowering of glucose tolerance, increased insulin concentration, glycosuria, growth impairment, shortening of productive age, increased concentration of blood cholesterol and triacylglycerols, infertility and peripheral neuropathies (Anderson, 1994). Manifestations of Cr deficiency usually develop in animals affected by metabolic stress or exposed to physical strain. The aim of the present work was to assess the effects of supplementation of Cr on metabolism of lactating crossbred cows including possible ensuing effects on certain blood biochemical constituents.

MATERIALS AND METHODS

Crossbred lactating cows (16) were selected on the basis of their milk yield and stage of lactation and housed in a well ventilated shed having cemented floor with facilities for individual feeding. All the experimental cows were offered weighed quantity of chaffed green fodder consisting of green oats, mustard and berseem mix and wheat straw as dry fodder ad *lib*. at 8.00 A.M. daily. The concentrate mixture was given as per requirements (NRC, 2001). The total allowance of concentrate mixture was offered in 3 installments. The first installment was given at 10.00 A.M., whereas the other two installments were offered to cows at the time of milking at 2.30 A.M. and 2.30 P.M. daily. The feeding trail lasted for 90 days. Lactating cows in treatment groups were given as 100 g mustard cake enriched with chromium picolinate as top dressed immediately after offering the first installment of the concentrate mixture. The 4 dietary treatments were : Treatment 1, control (without chromium supplementation), Treatment 2, 0.5 mg chromium per kg diet DM as chromium picolinate, Treatment 3, 1.0 mg chromium per kg diet DM as chromium picolinate and Treatment 4, 1.5 mg chromium per kg diet DM as chromium picolinate along with concentrate mixture as per requirements. For preparing the treatments, the mustard cake was finely ground and chromium picolinate containing 12% chromium was mixed in it according to the levels of chromium in different

treatments viz., 42 mg chromium picolinate was added to 100 g ground mustard cake in order to supplement 0.5 mg Cr/ kg DM (T2), 84 mg chromium picolinate was added to 100 g mustard cake to supplement 1.0 mg Cr/ kg DM (T3) and 126 mg chromium picolinate was added to 100 g mustard cake to supplement 1.5 mg Cr/ kg DM (T4) assuming 10 kg dry matter intake per animal daily. After mixing the required quantities of chromium for different treatments in mustard cake, the respective 100 g chromium enriched mustard cake was offered as top dress in the concentrate mixture, however, the animals in control group (T1) were given 100 g mustard cake without chromium daily. The weight of feed offered and residue left was recorded daily. The animals were offered fresh and clean drinking water twice daily. Blood samples were collected at the start and end of the feeding trial. The samples of blood for analyses were collected from jugular vein into disposable test tubes. The serum was harvested and analyzed for concentrations of glucose, total cholesterol, total protein, albumin, globulin, non-esterified fatty acids (NEFA) and catalytic activities of serum glutamate pyruvate transaminase (SGPT/ALT), serum glutamate oxaloacetate transaminase (SGOT/AST) and alkaline phosphatase (ALP) enzymes in blood serum. The blood glucose was estimated (GOD-POD enzymatic method) using span diagnostic kit. The total protein (UV-end point method), albumin (UV-end point method), cholesterol (CHOD-POD enzymatic method), SGOT, SGPT and ALP activities (photoelectric colorimeter with the help of standard graph) were measured using Merck diagnostic kits. The non-esterified fatty acids (NEFA) in total lipid extracted by using 'folch-wash' method (Folch *et al*., 1957) from serum was estimated by titrating it against KOH in the presence of phenolphthalein indicator (Cox and Pearson, 1962). The data were analyzed statistically for variance by applying completely randomized design (Snedecor and Cochran, 1980) using the analytical programme STPR 3.

RESULTS AND DISCUSSION

The glucose concentration in blood ranged from 63.59 mg/dl in control to 80.79 mg/dl in chromium supplemented group (1.0 mg Cr/ kg DM). There was linear increase in blood glucose concentration with the increasing level of chromium supplementation (Table 1). The values of blood glucose concentration in all the chromium supplemented groups were significantly ($P<0.05$) higher than the control group while these were statistically similar in all the chromium supplemented groups. The results are in agreement with the findings of Sano et al. (2000) and Subiyatno et al. (1996) who attributed due to the increased gluconeogenesis in Cr-supplemented cows. Yang et al. (1996) also reported elevated values for blood glucose concentration in chromium supplemented cows. However, no effects on the concentration of blood glucose due to chromium supplementation in cows have also been observed (Pechova et al., 2002; Williams et al., 2004 and Al-Saiady *et al*., 2004). Similarly, reports also exists that Cr supplementation significantly reduces serum glucose level compared with control group in goats (Haldar *et al.*, 2006) and in beef cows (Stahlhut et al., 2006). The total

Table 1: Average blood serum biochemical profile and enzyme activities in lactating crossbred cows fed ration supplemented with different levels of chromium

Particulars	Treatments/Groups			
	T1 (Control, no supplemental chromium)	T2 (0.5 mg Cr/ kg DM)	T3 (1.0 mg Cr/ kg DM)	T4 (1.5 mg Cr/ kg DM)
Glucose (mg/dl)*	63.59^{b}±3.19	77.69^{a}±4.73	80.79^{a}±4.68	78.09^{a}±3.74
Total protein (g/dl)	7.79±0.18	7.82±0.14	8.06±0.08	8.09±0.06
Albumin (g/dl)	3.95±0.12	4.18±0.06	4.29±0.09	4.25±0.04
Globulin (g/dl)	3.85±0.12	3.64±0.10	3.77±0.08	3.84±0.05
Total cholesterol (mg/dl)	203.87±23.84	160.74±15.73	213.74±25.85	201.4±7.79
Total lipids (mg/ml)	8.98±0.30	7.85±0.27	8.43±0.52	9.09±0.42
Non-esterified fatty acids (g/100ml)	1.13±0.04	0.95±0.04	1.16±0.10	1.04±0.06
Serum glutamate pyruvate transaminase (units/L)*	20.66^{b}±3.91	28.73^{ab}±0.44	34.48^{a}±3.99	35.36^{a}±3.15
Serum glutamate oxaloacetate transaminase (units/L)**	32.71^{b}±7.79	57.17^{a}±8.95	67.63^{a}±8.02	84.42^{a}±3.26
Serum alkaline phosphatase (units/L)	165.0±23.68	179.85±13.84	184.8±30.39	179.03±12.88

[ab]Values bearing the different superscripts in a row differ significantly from each other; *P<0.05, **P<0.01.

protein concentration in blood serum of lactating cows ranged from 7.79 g/dl in control to 8.09 g/dl in the group supplemented with 1.5 mg Cr/kg DM and these values did not differ significantly amongst the different treatments. These results are in agreement with the findings of Bailoni and Simonetto (2001) who reported increased serum total proteins with two levels (400 and 1600 ppb) of chromium supplementation in meat lambs. In contrary Al-Saiady et al. (2004) reported decreased concentration of total blood protein by adding chromium to the diet of lactating cows. However, chromium supplementation may play a role in reducing gluconeogenesis from amino acids. A possible protein-sparing effect of supplemental chromium was noted in studies with simple stomached animals (Lindemann *et al*., 1995). The serum albumin and globulin levels varied from 3.95 g/dl (T1, control) to 4.29 g/dl (T3, 1.0 mg Cr/ kg DM) and 3.64 g/dl (T3, 1.0 mg Cr/ kg DM) to 3.85 g/dl (T1, control), respectively. There was no significant difference among the treatments for the values of serum albumin and globulin concentrations while the higher concentration of albumin was observed in chromium supplemented animals than the control and lower concentration of globulin was recoded in the chromium supplemented groups than the control. The addition of chromium to the diet of lactating cows did not show any effect on serum albumin, while globulin was decreased as reported by Al-Saiady *et al.* (2004) which supports the results of present study. The finding of Gentry et al. (1999) also supports the results that lambs fed high protein diet had elevated plasma albumin ($P < .04$) with Cr vs control. Similar values have been reported earlier in crossbred cows fed different levels of UDP and plane of nutrition (Kumar *et al.*, 2006). Serum total cholesterol concentration in lactating cows ranged from 160.74 mg/dl in the group supplemented with 0.5 mg Cr/ kg DM to 213.74 mg/dl in the group supplemented with 1.0 mg Cr/ kg DM but the values did not differ significantly amongst the different groups. The values observed for total cholesterol concentration were also in the normal range (Chauhan, 1995; Kaneko *et al*., 1997). No significant differences in serum cholesterol concentration between chromium supplemented and control group of dairy cows have also been reported (Pechova *et al*., 2003) which corroborates with the present findings. On the contrary, Bailoni and Simonetto (2001) observed increased serum cholesterol levels in meat lambs due to chromium supplementation in the diet. The studies in humans showed a decline in serum total cholesterol accompanied by a concurrent increase in the proportion of HDL cholesterol (Anderson, 1994). In dairy cows the decrease in total cholesterol concentrations in serum was detected (Pechova *et al*., 2002) when the elevation exceeds the physiological values of 2.5 - 5.2 mmol/l. Similarly, Haldar *et al*. (2006) reported that Cr supplementation significantly reduced serum cholesterol ($P = 0.0001$) levels in adult goats compared with those in the control group.

The total lipid concentration in blood serum of lactating cows varied from 7.85 mg/ml (0.5 mg Cr/ kg DM) to 9.09 mg/ml (1.5 mg Cr/ kg DM) and did not differ significantly among the different treatments. These values were slightly lower than the values reported by Tandon *et al*. (2006) which ranged from 9.08 to 9.68 mg/ml serum in growing heifers and Mewara *et al*. (2008) which ranged from 10.15 to 12.66 mg/ml blood serum in lactating cows. Serum non-esterified fatty acids (NEFA) concentration varied from 0.95 g/100 ml to 1.16 g/100 ml in different groups of animals. These values were also not affected significantly due to chromium supplementation. These results are in agreement with the findings of Pechova *et al*. (2003), Besong *et al*. (1996) and Williams *et al*. (2004) who reported that there was no significant effect on concentration of serum NEFA in dairy cows due to chromium supplementation. Likewise Ireland (1999) found no decrease in NEFA concentration in the blood of dairy cows after the administration of chromium. However, Stahlhut *et al*. (2006) and Sumner *et al*. (2007) reported increased concentration of non-esterified fatty acids (NEFA) in serum due to chromium supplementation which is also a dose-dependent, quadratic manner in beef cows and in growing heifers, respectively. It is also reported that chromium supplementation has the greatest impact on serum NEFA concentration as chromium from chromium-Met decreased serum NEFA concentration in Holstein cows (Bryan *et al*. 2004). Likewise, Sano et al. (2000) reported that plasma concentrations of NEFA increased ($P < 0.05$) in both of diets (chromium supplemented vs control) in sheep. Gentry *et al*. (1999) reported that lambs fed high protein diet declined post-prandial plasma NEFA concentration with Cr vs control.

The activities of serum glutamate pyruvate transaminase (SGPT) and serum glutamate oxaloacetate transaminase (SGOT) enzymes were significantly ($P<0.05$) and ($P<0.01$) increased, respectively, in the chromium supplemented groups when compared with control. There was no significant difference in alkaline phosphatase (ALP) enzyme activity among the treatments, however, a linear increase in alkaline phosphatase enzyme activity was observed with the

increasing levels of chromium supplementation. The results of present study corroborated with the findings of Uyanik et al. (2002) who reported increased serum ALP activity with chromium supplementation in broilers. However, in an earlier report there was no effect on the activities of SGOT and ALP due to supplementation of 5 mg chromium to each animal daily but supplemental chromium 10 mg per animal daily in Holstien cows during peri-partal period decreased the activities of SGOT (Pechova et al., 2002). In the present study all the blood parameters were in normal range. It can be concluded that chromium supplementation up to 1.5 mg/kg diet DM maintained the normal blood biochemical parameters as well as enzyme activities without any adverse effects.

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