

EFFECTS OF CHROMIUM PICOLINATE ON GROWTH PERFORMANCE, CARCASS COMPOSITION AND SERUM TRAITS OF BROILERS FED DIETARY DIFFERENT LEVELS OF CRUDE PROTEIN

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Summary

An experiment was conducted to evaluate the effects of chromium picolinate on growth performance, nutrient utilizability, carcass composition, serum traits and *in vitro* protein synthesis of 3 day old Arbor Acres broiler chickens when dietary crude protein levels were varying in diets. Six replicates of eight chicks each (average initial weight = 59.4 g) were randomly assigned to three levels (low, medium, high) of dietary crude protein at two levels of chromium (0, 200 ppb Cr/kg diet) as chromium picolinate. Six chicks/treatment were randomly chosen for analyses of carcass composition, six additional chicks/treatment were randomly chosen for analyses of serum components, and a chick/treatment was chosen for *in vitro* culture of liver tissue.

Chromium picolinate did not affect feed intake, protein and fat utilizability, regardless of dietary crude protein level. But feed/gain ratio were more improved in groups fed the low protein diets added with chromium picolinate compared with groups fed the medium and high protein diets with chromium picolinate. Carcass fat tended to decrease whereas carcass protein tended to increase when added with chromium picolinate. Broilers fed diets with chromium picolinate exhibited lower serum triglyceride and nonesterified fatty acid concentrations than those fed without chromium picolinate ($p < 0.05$). Both secreted and retained proteins in cultured acinar cell were higher in groups fed diets with chromium picolinate than those fed diets without chromium picolinate ($p < 0.05$).

It could be suggested that chromium picolinate was effective in improving weight gain and nutrient utilizability when dietary crude protein was low ($p < 0.05$), and also effective in manipulating carcass fat when dietary crude protein level was high ($p < 0.05$).

(Key Words : Broiler, Chromium Picolinate, Crude Protein, Growth Performance, Nutrient Utilizability, Carcass Composition, Serum Traits)

Introduction

Chromium has been recognized as an essential element for mammals since Schwartz and Mertz (1959) first elucidated the significance of this metal in normal glucose utilization, while chromium is toxic like other micronutrients when given in excessive amounts.

It was known that chromium was widely distributed throughout the body, and was carried by the fractions of the serum proteins and was rapidly taken up by other tissues (Mertz, 1969; Mertz and Roginski, 1971). But little is known about the chemical form of chromium in the

tissues or serum proteins, although "glucose tolerance factor" was found as a natural chromium complex occurring in brewer's yeast and was assumed to activate the action of insulin and membrane transport (Mertz, 1969). Several investigations with both animals and humans provide evidence that the chromium occurs in many tissues and appears to be required for the action of insulin in controlling glucose metabolism. It also occurs in microsomes and nucleus, although its function in these organelles is unknown (Sargent et al., 1979).

Chromium deficiency in both humans and animals resulted in impaired glucose tolerance, elevated blood glucose levels, hyper-cholesterolemia and development of aortic plaques.

The exact mechanism whereby chromium participates in the function of insulin has not been elucidated but the theory ranged from a direct interaction of chromium with

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insulin (Mertz, 1969; Evans et al., 1973) to a role of chromium in the production of peripheral insulin receptors (Anderson, 1986). To date, the synergism between chromium and insulin has been studied primarily with regard to the hormone's regulation of carbohydrate and lipid metabolism. However, because insulin also has a key role in muscle metabolism (Felig, 1975), depletion of chromium in the body may impair the development of lean body mass. Glucose-independent effects of chromium on amino acid transport and utilization for protein synthesis also have been shown (Weser and Koolman, 1969; Okada et al., 1983, 1984).

Most diets are primarily composed of ingredients from plant origin, which are usually low in chromium (Schroeder, 1971; Giri et al., 1990). The absorption and utilization of chromium may be dependent on its association with an organic molecule (Mertz, 1969; Votava et al., 1973) such as picolinate (Evans and Johnson, 1980a, b). Although several investigations were conducted to evaluate the effects of chromium picolinate supplementation, it was rare about the interaction of chromium picolinate with other nutrients. Therefore, the purpose of this investigation was to assess the effect of chromium, in the form of chromium picolinate, on growth, serum and carcass traits and *in vitro* liver tissue culture of broilers when fed diets containing different levels of crude protein.

Materials and Methods

1) Experimental Design and Animal

Experimental diets were formulated to contain three levels of crude protein [80%, 100% and 120% of the requirement suggested by NRC (1984)] within two levels of chromium picolinate (0 and 200 ppb). All treatments had 6 replicates of 8 birds for each replicate.

Animals used in this study were broiler chicks of Arbor Acres produced by Han II Breeding Farm. At 3 days of age, experimental animals with similar initial body weight were fed the experimental diets for 6 weeks. A total number of 288 birds were used in this study. All feeding trials, metabolic trials and chemical analysis of experimental feed and excreta were conducted in the Animal Nutrition Laboratory, Department of Animal Science and Technology, College of Agriculture and Life Sciences, Seoul National University located in Suwon, Korea. Feeding trials were initiated in May 26, 1993 and terminated in July 7, 1993.

2) Experimental Diets

In this experiment, birds were fed a commercial diet (CP: 23%, 3,200 kcal ME/kg) for a preliminary period of three days. The three basal isocaloric diets (3,200 kcal/kg) were formulated to contain three different levels of dietary crude protein (80, 100, 120% of the NRC requirement, 1984). The formula and chemical composition of basal diets for starting and finishing period are presented in table 1. Each basal diet used in each period was supplemented with two levels (0, 200 ppb) of chromium picolinate. All the nutrients except protein were included to meet the National Research Council requirement (NRC, 1984).

3) Methods of Experiment

Feeding trial was conducted according to the procedures described by Kim et al. (1995). Nutrient utilizability was estimated using total excreta collection once a week over entire experimental period (6 weeks). Nutrient contents and amino acid concentrations in excreta and experimental diets, carcass composition, and blood components were also analyzed according to the methods described by Kim et al. (1995).

Liver acinar cell for culture was prepared according to the method by Choi et al. (1987). Culture medium was Eagle's MEM (Eagle, 1959) modified by Smith et al. (1982). To measure protein synthesis and amino acid uptake activity, shortly after the 18-h incubation, cells were collected, pooled (four dishes per treatment), and centrifuged at $1,000 \times g$ at $4^{\circ}C$ for 10 minutes. The synthesized protein was calculated based on the equation by Kim et al. (1995).

Statistical analyses for the present data were carried out by comparing means according to Duncan's multiple range test (Duncan, 1955), using General Linear Model (GLM) Procedure of SAS (1985) package program.

Results and Discussion

1) Growth Performance

Table 2 summarized the effects of crude protein and chromium picolinate on body weight gain, feed intake, feed conversion and mortality during the entire experimental period (1-6 weeks). The highest body weight gain was found when chicks were fed a diet containing 100% and 120% CP with no chromium picolinate and diets containing 100% CP with 200 ppb chromium picolinate ($p < 0.05$). At the 80% CP levels, 200 ppb chromium picolinate supplemented group showed higher body weight gain than no chromium picolinate supplemented group ($p < 0.05$). The chicks received a diet containing 100% and 120% crude protein with no

TABLE 1. FORMULA AND CHEMICAL COMPOSITION OF THE BASAL DIETS FOR EXPERIMENT

Levels of dietary protein ¹ (%)	Starter			Finisher		
	80	100	120	80	100	120
Ingredient (%):						
Corn, yellow	71.28	56.95	42.95	73.50	60.00	48.94
Soybean meal	8.90	24.28	30.44	6.80	18.00	26.59
Fish meal	2.40	2.41	2.30	1.00	0.60	0.40
Corn gluten meal	9.50	9.20	12.99	7.60	9.00	11.50
Wheat bran	2.40	0.00	2.40	5.00	4.40	3.94
Tallow	2.00	4.01	5.92	2.50	4.70	5.62
Limestone	2.15	2.20	2.20	2.30	2.30	2.21
Salt	0.30	0.30	0.30	0.30	0.30	0.30
Vit.-min. mix. ²	0.45	0.45	0.45	0.45	0.45	0.45
Lysine	0.57	0.15	0.00	0.50	0.20	0.00
Antibiotics	0.05	0.05	0.05	0.05	0.05	0.05
Total	100.00	100.00	100.00	100.00	100.00	100.00
Chemical composition :						
ME (kcal/kg)	3,209	3,200	3,200	3,202	3,220	3,201
Crude protein (%)	18.45	23.00	27.00	16.04	20.02	24.00
Lysine (%)	1.20	1.20	1.25	1.00	1.01	1.05
Methionine (%)	0.43	0.49	0.58	0.36	0.43	0.50
Calcium (%)	0.90	0.95	0.97	0.87	0.88	0.86
Avail. phosphorus (%)	0.40	0.43	0.48	0.37	0.40	0.42

¹ 80, 100 and 120% of protein levels suggested by NRC (1984).

² Vit.-min. mixture contains followings in 1 kg; vitamin A, 10,000 IU; vitamin D₃, 1,500 IU; vitamin K₃, 5 mg; vitamin E, 15 mg; vitamin B₂, 8 mg; vitamin B₁₂, 0.008 mg; Ca-d-pantothenate, 8 mg; niacin, 25 mg; folic acid, 0.4 mg; biotin, 0.2 mg; choline, 500 mg; pyridoxine, 1 mg; B.H.T. 125 mg; Co, 0.85 mg; I, 1.29 mg; Zn, 100 mg; Mn, 110 mg; Cu, 8.75 mg; Se, 0.15 mg; Fe, 35 mg.

chromium picolinate showed the highest feed intake ($p < 0.05$).

Feed conversion was significantly different among treatments ($p < 0.05$). Chicks fed a diet containing 120% crude protein with 200 ppb chromium picolinate presented the best feed conversion ($p < 0.05$), whereas the worst feed conversion was obtained at the 80% crude protein with no chromium picolinate ($p < 0.05$). Among crude protein levels, the 120% crude protein group showed the best feed conversion ($p < 0.05$), and between the chromium picolinate levels, the better feed conversion was obtained at the 200 ppb chromium picolinate supplement ($p < 0.05$). The mortality was lower in groups fed 100% CP diets with 200 ppb chromium picolinate.

From these results, it can be concluded that chromium picolinate reduced feed intake of chicks ($p < 0.05$), yet did not decrease the body weight gain. Consequently, feed conversions of chicks fed diets containing 200 ppb chromium picolinate were superior to those of non-chromium picolinate supplemented groups ($p < 0.05$).

Interaction between crude protein and chromium picolinate was not found in body weight gain, feed intake and feed conversion.

2) Nutrient Utilizability

As shown in table 2, the utilizability of dry matter and crude ash was not affected by dietary crude protein and chromium picolinate levels, and utilizability of crude protein was significantly different among treatments ($p < 0.05$). It was found that the utilizability of crude protein was increased ($p < 0.05$), as the dietary crude protein levels increased from 80% to 120%. But there was no significant difference between chromium picolinate levels.

Among the levels of crude protein, crude fat utilizability was the highest ($p < 0.05$) at 100% CP and the lowest at 80% CP ($p < 0.05$), but the levels of chromium picolinate did not affect crude fat utilizability. At 80% CP, chromium picolinate supplemented group showed significantly higher utilizability of crude fat than non-supplemented group ($p < 0.05$).

TABLE 2. GROWTH PERFORMANCES AND NUTRIENT UTILIZABILITY OF BROILER AS AFFECTED BY THE LEVELS OF DIETARY PROTEIN AND CHROMIUM PICOLINATE (1-6 WEEKS)

Chromium picolinate Protein ¹	0 ppb			200 ppb			SE ²
	80	100	120	80	100	120	
Weight gain (g)	1,809 ^c	2,064 ^a	2,073 ^a	1,855 ^{bc}	2,031 ^a	1,993 ^{ab}	25.85
Feed intake (g)	3,761 ^{ab}	4,009 ^a	3,884 ^{ab}	3,827 ^{ab}	3,788 ^{ab}	3,674 ^b	36.61
Feed/gain	2.08 ^a	1.94 ^a	1.87 ^b	2.06 ^a	1.87 ^b	1.84 ^b	0.02
Mortality (%)	4.17 ^{ab}	9.38 ^a	0.00 ^b	2.08 ^{ab}	0.00 ^b	2.08 ^{ab}	1.04
Nutrient utilizability (%)							
Crude protein	64.7 ^b	74.3 ^{ab}	78.1 ^a	74.6 ^{ab}	72.5 ^{ab}	72.7 ^{ab}	1.52
Crude fat	93.0 ^b	96.5 ^a	96.4 ^a	95.4 ^a	96.1 ^a	95.0 ^{ab}	0.35
Between chromium	Weight gain	Feed intake	Feed/gain	Mortality	Crude protein	Crude fat	
0	1,982	3,884	1.96 ^a	4.17	71.0	95.3	
200	1,960	3,763	1.91 ^b	1.39	73.3	95.5	
Among protein	80	3,794	2,07 ^a	3.13	69.6	94.2 ^b	
100	2,047 ^a	3,898	1.90 ^b	3.75	73.3	96.3 ^a	
120	2,033 ^a	3,779	1.86 ^c	1.14	74.5	95.7 ^{ab}	
Interaction (P value)							
Chromium × protein	0.4651	0.1772	0.0837	0.0960	0.1087	0.0464	

¹ 80, 100 and 120% of protein levels suggested by NRC (1984).

² Pooled standard error.

^{ab,c} Means with different superscripts within the same column or row are significantly different ($p < 0.05$).

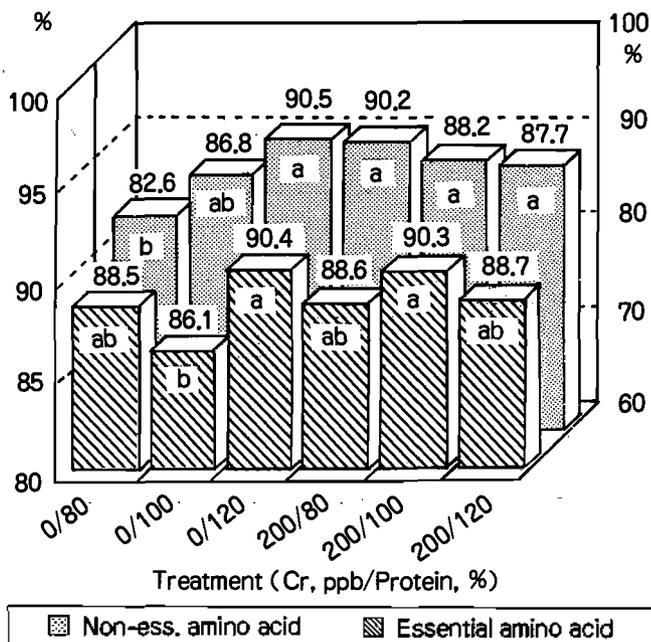


Figure 1. Effects of dietary protein and chromium picolinate on amino acid utilization of broilers at 6 weeks old

^{ab} Mean values with different superscripts within the same row are significantly different ($p < 0.05$).

Interaction between crude protein and chromium picolinate was found in crude fat utilization ($p < 0.05$).

3) Amino Acid Utilizability

Figure 1 summarized the utilization of essential and non-essential amino acids of experimental diets fed to broilers.

The utilization of essential amino acids was not affected by chromium picolinate levels. However, as the dietary crude protein level was increased, the utilization of essential amino acids was improved ($p < 0.05$).

The utilization of non-essential amino acids by chicks fed 80% CP diet with no chromium picolinate was lower than that of other groups ($p < 0.05$). Between the chromium picolinate levels, there was no significant difference in non-essential amino acid utilization. When dietary crude protein level was high, the utilization of non-essential amino acid was increased ($p < 0.05$).

Chicks fed diets containing 80% CP with no chromium picolinate and 100% CP with no chromium picolinate showed lower utilization of amino acids than other chicks ($p < 0.05$).

120% CP and 100% CP diet showed better utilization

of amino acid than 80% CP diet ($p < 0.05$).

4) Carcass Composition

The effects of dietary crude protein and chromium picolinate levels on carcass composition are outlined in table 3.

Crude protein content of broiler chicks was significantly influenced ($p < 0.05$) by the levels of dietary crude protein and chromium picolinate.

Crude fat content was lowest in 120% CP diet with 200 ppb chromium picolinate and highest at 80% CP diet with no chromium picolinate ($p < 0.05$).

TABLE 3. EFFECTS OF DIFFERENT LEVELS OF CRUDE PROTEIN AND CHROMIUM PICOLINATE ON CARCASS COMPOSITION AND THE CONTENT OF SERUM INSULIN, GLUCOSE, TRIGLYCERIDE AND NONESTERIFIED FATTY ACID (NEFA)

Chromium picolinate Protein ¹	0 ppb			200 ppb			SE ²
	80	100	120	80	100	120	
Carcass composition (%)							
Crude protein	37.7 ^c	43.1 ^{ab}	44.3 ^a	38.7 ^{bc}	43.2 ^{ab}	46.7 ^a	0.81
Crude fat	52.9 ^a	49.3 ^{abc}	45.5 ^{cd}	51.8 ^{ab}	46.5 ^{bcd}	42.8 ^d	0.91
Crude ash	5.5 ^c	5.7 ^{bc}	6.8 ^a	5.6 ^c	6.4 ^{ab}	7.0 ^a	0.14
Serum trait							
Insulin (μ IU/ml)	5.8 ^c	7.8 ^{bc}	13.7 ^a	6.7 ^c	14.3 ^a	12.2 ^{ab}	0.80
Glucose (mg/dl)	215 ^b	231 ^{ab}	245 ^a	236 ^a	236 ^a	239 ^a	2.60
Triglyceride (mg/dl)	106.7 ^a	79.1 ^b	65.9 ^{bc}	80.7 ^b	60.8 ^c	53.0 ^c	2.94
NEFA ³ (μ Eq/l)	1,288 ^a	773 ^{bc}	692 ^{bc}	845 ^b	682 ^{bc}	626 ^c	35.56
Between chromium	Crude protein	Crude fat	Crude ash	Insulin	Glucose	Triglyceride	NEFA
0	41.7	49.2	6.0	9.4	231	83.7 ^a	931 ^a
200	42.9	47.0	6.4	11.1	237	64.9 ^b	714 ^b
Among protein	80	100	120	80	100	120	
	38.2 ^b	43.1 ^a	45.5 ^a	38.2 ^b	43.2 ^{ab}	46.7 ^a	1,076 ^a
	43.1 ^a	47.9 ^b	44.1 ^c	51.8 ^a	46.5 ^{ab}	42.8 ^b	727 ^b
	45.5 ^a	44.1 ^c	6.9 ^a	12.9 ^a	242 ^a	59.5 ^b	656 ^b
Interaction (P value)							
Chromium \times protein	0.7888	0.8635	0.4750	0.0756	0.0738	0.5090	0.0025

¹ 80, 100 and 120% of protein levels suggested by NRC (1984).

² Pooled standard error.

³ Nonesterified fatty acid.

^{a,b,c} Means with different superscripts within the same column or row are significantly different ($p < 0.05$).

When dietary crude protein level was increased from 80 to 120%, crude protein content of carcass was increased ($p < 0.05$), whereas crude fat content was decreased ($p < 0.05$). Crude fat content of broilers fed diet containing 200 ppb chromium picolinate was lower than the crude fat content of broilers on the non-chromium picolinate diet ($p < 0.05$).

Crude ash content was highest with 80% CP diet with 200 ppb chromium picolinate and lowest at 120% CP diet with no chromium picolinate ($p < 0.05$).

It was found that crude protein content and crude ash content increased as dietary crude protein level increased

from 80 to 120, whereas crude fat decreased. Chromium picolinate diet not affect carcass composition of broilers ($p < 0.05$).

Overall, broilers fed diets containing 120% CP with 200 ppb chromium picolinate showed the highest crude protein content and the lowest crude fat content in carcass composition ($p < 0.05$). Chicks fed a diet with 80% CP and no chromium picolinate showed the lowest crude protein content and the highest crude fat content ($p < 0.05$).

These results agreed with the previously reported data. Anderson et al. (1989) reported that supplemental

chromium from $\text{CrCl}_3 \cdot 6\text{H}_2\text{O}$ increased the percentage of turkey breast, McCarty (1993) showed that chromium picolinate reduced body fat in rat and Evans (1989) reported that chromium from chromium picolinate increased lean body mass and body fat loss of men in a weight training program. Britton et al (1968) reported that chromium supplementation increased nitrogen retention in lambs.

5) Total Cholesterol versus High Density Lipoprotein (HDL) in Serum

The effects of dietary crude protein and chromium picolinate on the ratio of total cholesterol versus HDL in serum of broiler chicks are presented in figure 2.

As shown in figure 2, the HDL/total cholesterol ratio was highest at 120% CP with 200 ppb chromium picolinate and lowest at 100% CP with no chromium picolinate ($p < 0.05$). It was also significantly improved with 80% and 100% CP diet added with chromium picolinate ($p < 0.05$).

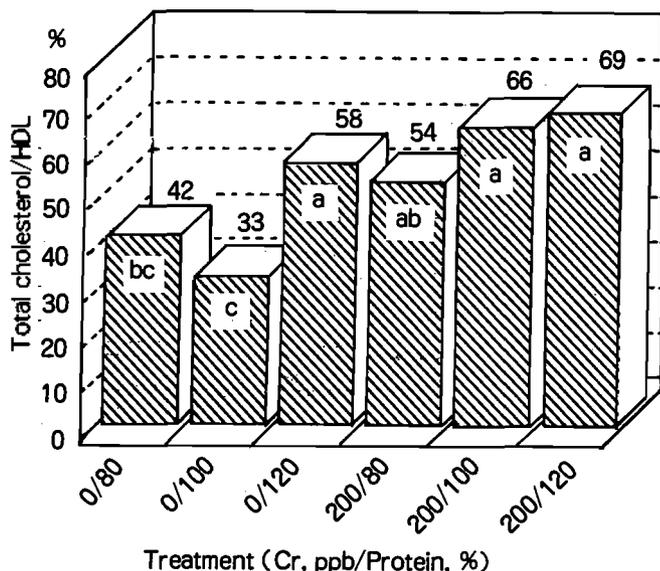


Figure 2. Effects of different levels of crude protein and chromium picolinate on the HDL/total cholesterol ratio

^{abc} Mean values with different superscripts within the same row are significantly different ($p < 0.05$).

Among crude protein levels, 120% CP group showed higher HDL/total cholesterol ratio than 100% and 80% CP groups ($p < 0.05$). And chromium picolinate supplemented groups showed higher HDL/total cholesterol ratio than other groups ($p < 0.05$).

A recent study demonstrated that total cholesterol,

LDL (low density lipoprotein)-cholesterol and the related transport protein apolipoprotein B were significantly decreased while apolipoprotein A₁, the HDL-cholesterol related protein, was elevated when the supplement contained chromium picolinate (Press et al., 1990).

6) Insulin, Glucose, Non-esterified Fatty Acid (NEFA) and Triglyceride in Serum

Content of serum insulin, glucose, triglyceride and NEFA is presented in table 3.

Serum insulin in broilers fed 100% CP diet with 200 ppb chromium picolinate was greater than that of other groups ($p < 0.05$) while serum insulin in broilers fed 80% CP diet with no chromium picolinate was smallest ($p < 0.05$). Among dietary crude protein, serum insulin content was higher at 100% CP and 120% CP than at 80% CP ($p < 0.05$).

Content of serum glucose was not affected by chromium picolinate. As dietary crude protein levels increased, the content of serum glucose became higher ($p < 0.05$).

Broilers received diets containing 80% CP diet with no chromium picolinate showed the lowest content of serum triglyceride ($p < 0.05$). Chromium picolinate supplemented groups presented lower serum triglyceride content than other groups ($p < 0.05$). Serum triglyceride and NEFA were reduced by chromium picolinate supplementation ($p < 0.05$) at all crude protein levels.

Among dietary crude protein levels, 120% and 100% CP groups had higher serum triglyceride than the 80% CP group ($p < 0.05$).

An increase in NEFA would imply that the process of fat degradation or lipolysis in adipose tissue was increased (Mersmann and MacNeil, 1985).

Interaction between dietary crude protein and chromium picolinate was found in the content of serum NEFA.

7) *In vitro* Protein Synthesis

The effects of dietary CP and chromium picolinate (0 or 200 ppb) on retained protein and secreted protein in liver acinar cell culture are summarized in table 4. Secreted and retained protein from liver tissue of chicks fed diets supplemented with 200 ppb chromium picolinate was significantly ($p < 0.05$) higher than those of chicks fed non-chromium picolinate diet. Secreted and retained protein of liver tissue was not significantly affected by crude protein levels even though secreted and retained protein tended to increase as dietary crude protein level increased.

These results mean that supplementation of 200 ppb

chromium picolinate can increase secreted protein and retained protein in liver tissue up to 29.1% and 21.6%.

chromium picolinate was not found in secreted protein and retained protein.

Interaction between dietary crude protein levels and

TABLE 4. EFFECT OF DIETARY PROTEIN AND CHROMIUM PICOLINATE ON THE SECRETED AND RETAINED PROTEIN BY ACINAR CELL CULTURE (DPM/MG)

Chromium picolinate Protein ¹	0 ppb			200 ppb			SE ²
	80	100	120	80	100	120	
Secreted protein ³	2,639.6	2,599.2	2,671.3	3,130.6	3,669.9	3,417.4	230.3
Retained protein ⁴	3,406.1	3,628.1	3,716.3	3,906.7	4,584.5	4,590.4	276.7
Between chromium	Secreted protein			Retained protein			
0	2,636.7 ^b			3,583.5 ^b			
200	3,405.9 ^a			4,360.5 ^a			
Among protein	80	2,885.1		3,656.4			
	100	3,134.5		4,106.3			
	120	3,044.3		4,153.3			
Interaction (P value)	Chromium × protein			0.5840			0.7514

¹ 80, 100 and 120% of protein levels suggested by NRC (1984).

² Pooled standard error.

³ The amount of secreted protein was determined by the incorporation of [³H]-lysine (0.5 μCi/ml) into TCA-insoluble material.

⁴ The amount of retained protein was determined by the incorporation of [³H]-lysine (0.5 μCi/ml) into TCA-insoluble material.

^{a,b} Means with different superscripts within the same column or row are significantly different (p < 0.05).

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