

EFFECT OF SUPPLEMENTING DIFFERENT SOURCES OF CHROMIUM TO DIET ON SOME PHYSIOLOGICAL TRAITS OF BROILER CHICKENS

By

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Abstract:*The experiment was conducted at the University of Ain Shams – Egypt, from January to March 2008, to study the effect and the comparison of chromium type that supplementation to diet, chromium chloride (CrCl_3) (inorganic chromium), chromium yeast (Cr yeast) and chromium picolinate (Cr pico) (organic chromium) , on some physiological traits of broiler chickens. A total of 480, one – day old chickens (unsexed) of Cobb strain were randomly allocated to four dietary treatments and each treatment consists of 120 birds (3 replicates each treatment). The treatments were control (A_1) without chromium supplementation, (A_2) 1 mg CrCl_3 /Kg diet, (A_3) 1 mg Cr yeast/Kg diet, and (A_4) 1 mg Cr Pico/Kg diet.*

The results indicated that Plasma glucose was lower significantly in A_3 compared with control group A_1 , while plasma total protein and globulin were significantly ($P \leq 0.05$) higher in the groups that were supplemented with Cr yeast (A_3) and Cr Pico (A_4) compared with other group . Plasma total lipid was significantly ($P \leq 0.05$) lowered in A_4 than the control group (A_1), meanwhile broilers of (A_2) and (A_3) were numerically lower in total lipid than the control group . Plasma cholesterol was significantly ($P \leq 0.05$) lowered in the broilers that were supplemented with all chromium sources. Low density lipoprotein (LDL) was significantly ($P \leq 0.05$) lower in (A_2) group, while other sources were numerically low in LDL as compared to the control (A_1). Spleen percentage increased significantly while plasma phosphorus decreased significantly in A_4 compared with control . Supplementing CrCl_3 (A_2) or Cr Pico (A_4) resulted in significant ($P \leq 0.05$) increase of chromium in liver, muscle while plasma chromium increased only in A_2 group .

It can be concluded that chromium supplementation to diet (either organic or inorganic forms) have been used in this study had a beneficial effect on some of physiological parameters that were measured.

INTRODUCTION

Poultry production has become one of the biggest agricultural industries and its improvement is one of the main objectives of the poultry industry.

For the past 4 decades, many nutritionists have been considered chromium as an essential element for humans and animals, (NRC , 1980). Because its involvement in carbohydrate, lipid, Protein and nucleic acid metabolic functions (Anderson and kozlovsky, 1985). Chromium compounds were also found to be a blood glucose tolerance factor (GTF) in rat experiments (walker, 1993), which promotes glucose metabolism, enhances glycogenesis from glucose and accelerates glucose transport and muscle build up, (Steele and Rosebrough, 1981), chromium is also a co – factor of insulin, promoting insulin activity (McCarty *et al.*, 1988), enhancing amino acid uptake, promoting lipogenesis from glucose and lipid storage in the liver and adipose tissues (Steele and Rosebrough, 1979). Chromium can also reduce blood levels of lipid, total cholesterol, low – density lipoproteins (LDL) and increases high density lipoproteins (HDL) (Press *et al.*, 1990). Chromium is also considered as antistress factor (Kegley and Spears, 1995) and increases immune response (Uyanik *et al.*, 2002). Chromium forms are varied in their bioavailability inside animal body (Mowat, 1997). Therefore, a need to reevaluate the efficiency of different supplementing forms of chromium (organic vs inorganic) have become a necessary issue.

The objective of the present work was to investigate the effects of dietary chromium supplementation of organic chromium chromium yeast (Cr yeast) vs chromium picolinate (Cr Pico) and inorganic chromium chromium chloride (CrCl₃) in broiler chickens diets on some physiological traits and tissue chromium concentration.

MATERIALS AND METHODS

The experiment were conducted at Broiler Nutrition Unit, Faculty of Agriculture Ain Shams University , Egypt during the period from January to March 2008.

The experimental birds were raised from day – old to 5 weeks of age in floor pens .Electrical heaters were used to maintain room temperature at 34°C during the first week of age, and then the temperature was decreased

gradually to 26°C during the third weeks of age. Artificial lighting was provided constantly during the experimental period. Water and feed were provided *ad lib* through the 5 weeks experimental period. The birds were vaccinated against avian influenza at one day old and Newcastle disease at 6, 18 days old.

Four hundred and eighty, one – day old Cobb broiler chickens were allocated randomly into four treatment groups of 120 birds (three replicated pen with 40 birds each).

A corn – soya diet was used. The chickens received starter ration from one day to 3 weeks of age (22% crude protein and 3000 kcal ME/kg diet) and grower ration was fed from 3 to 5 weeks of age, which contained 20% crude protein, and 3100 kcal ME/kg diet Table 1. The rations were formulated according to NRC (1994). All type of chromium that used in our experiment (mixed well with diet) was provided by Ahmed El-Batal Company – Egypt .

Table 1. Composition and calculated analysis of the experimental diets

Ingredient (%)	Starter (0-3 wks) %	Grower (3-5wks) %
Yellow corn	55.8	59.71
Soya bean meal (44%)	34.32	30.00
Corn gluten	3.33	2.80
Vegetable oil	2.79	4.00
Dicalcium phosphate	1.94	1.67
Limestone	1.14	1.14
Common salt	0.25	0.25
Vit & min. premix*	0.25	0.25
DL. methionine	0.18	0.18
Total	100	100
Calculated Composition %		
Crude protein	22.00	20.00
Kcal. ME/kg diet	3000	3100
Calcium	0.97	0.91
Available Phosphorus	0.50	0.45
Methionine + Cystein	0.91	0.78
Lysine	1.10	1.10

*Composition of vitamin and minerals premix. Each 3 kg of vitamin and minerals mixture contain: 12000000 IU vitamin A; 2000000 IU D3; 10 gI; 1gk; 1g B1; 5g B2 1500 mg B6; 10 mg B12; 10g pantothenic acid; 20g Nicotinic acid 1g Folic acid; 50mg Biotin. 500g

choline chloride: 4g copper: 300mg iodine: 30g iron: 60g manganese: 50g zinc: and 100mg selenium. Calculated composition according to (N.R.C.,1994)

Four different dietary treatments were used in this experiment as follows: Treatment 1 (A₁): without chromium supplementation to diet .Treatment 2 (A₂): 1 mg CrCl₃ /kg diet. Treatment 3 (A₃): 1 mg Cr yeast /kg diet. Treatment 4 (A₄): 1 mg Cr Pico /kg diet .

At the end of experimental period four birds were chosen randomly from each replicate and blood sample were collected for the measurement of physiological traits then the birds were slaughtered for the measurements of internal organ . liver ,gizzard ,heart , bursa , thymus ,and spleen ,were removed and weight as a percentage of body weight. The blood samples were collected in centrifuge tubes with EDTA. The tubes were Stoppard and centrifuged immediately (4000 rpm) for 15 minutes to separate plasma which was decanted into sterilized glass vials which were Stoppard tightly and stored in a deep freezer until test.

Plasma glucose was determined according to Trinder (1969) using commercial kits of (spectrum Co.).Plasma total proteins and albumin were determined by using colorimetric method according to Henry (1964) and Doumas et al., (1971) respectively using commercial kits of (Biodiagnostic Co.) .The concentration of plasma globulins was obtained by subtracting the albumin value from the value of total protein for each plasma sample. Plasma total lipids, triglycerides, total cholesterol, HDL were determined according to Knight *et al.* (1972) : Sidney and Barnard (1973); Richmond (1973) respectively using commercial kits of (Biocon CO.).LDL was determined according to Bergmenyer (1985) by calculation $LDL = \text{total cholesterol} - HDL - TG/5$. Thyroid hormones T3 and T4 were analyzed by using radioimmunoassay kits as described by Sharp *et al.* (1987).Plasma chromium levels were analyzed by using atomic absorption spectrophotometer as described by Perkin. (1982).Plasma calcium and phosphorus were determined by using commercial kits of (Giesse Diagnostics company) according to Gindler and King (1972).Uric acid was determined by enzymatic colorimetric test according to Arliss and Entwistle (1981), using commercial kits (Biodiagnostic CO.). Plasma creatinine was determined by using spectrophotometer according to Husdan and Rapaport (1968), using commercial kits (Biodiagnostic CO.).Aspartate aminotransferase (AST) and Alanine amino transferase (ALT) were determined according to Reitman and Frankel. (1957) using commercial kits (Biodiagnostic CO.) . Chromium determination of liver and muscles (mixed thigh and breast) by procedure used as follows:5 g add justly weighed of

muscle which was ashes in a muffle at 600 ° C for 3 hours .The ash was solved in concentrated hydrochloric acid at first, the washed in a limit quantity of hydrochloric acid 1 molar concentration and filtrate using ash less filtering papers .The filtrate was diluted properly and flame photometer was used for chromium determining (AOAC, 1980). The Cr was measured using Atomic Absorption (Spectrophotometer).

Completely randomized design (CRD) was used to study the effect of difference treatment in all traits. Duncan (1955) multiple range test was used to compare the significant differences between means. Data were analyzed using statistical analysis system (SAS, 2001) by assuming the following model.

$$Y_{ij} = \mu + T_i + e_{ij}$$

Were:

Y_{ij} : is the value of observation of traits.

μ : is The overall mean of traits.

T_i : The effect of treatments , control (A_1) , (A_2) , (A_3) and (A_4)

e_{ij} : Random error assumed to be mean equal to zero and variance is σ^2e ($N \sim 0, \sigma^2e$).

RESULTS AND DISCUSSION

Internal organs percentages as a relative weights to body weight of the chickens fed different experimental diets are shown in Table 2. Statistical analysis showed that liver weight, gizzard and heart percentage were not affected by dietary treatments, although, there was a slightly difference between experiment groups.

Table 2. Effect of chromium chloride ($CrCl_3$) chromium yeast (Cr yeast) chromium picolinate (Cr Pico) supplementation to diet on internal organs percentage in broiler chickens at 5 weeks of age.

Character (%)	Treatments (1 mg / kg diet)			
	Control	$CrCl_3$	Cr yeast	Cr Pico
	A_1	A_2	A_3	A_4
Liver	2.19	2.25	2.57	2.24
Gizzard	2.15	2.19	2.02	2.01
Heart	0.63	0.57	0.58	0.50

Lymphoid organs percentages are shown in table3. Neither bursa nor thymus percentage was affected by supplemented chromium to diet, whereas spleen weight percentage was significantly ($P \leq 0.05$) increased due to feeding Cr Pico – diet (A_4) 0.23%.

Table 3. Effect of chromium chloride (CrCl₃) chromium yeast (Cr yeast), chromium picolinate (Cr Pico) supplementation to diet on lymphoid organs percentage in broiler chickens at 5 weeks of age.

Characters %	Treatments (1 mg / kg diet)			
	Control	CrCl ₃	Cr yeast	Cr Pico
	A ₁	A ₂	A ₃	A ₄
Bursa	0.254 ^a	0.196 ^a	0.188 ^a	0.164 ^a
Thymus	0.554 ^a	0.633 ^a	0.490 ^a	0.627 ^a
Spleen	1.118 ^b	1.121 ^b	0.125 ^b	0.230 ^a

Means having different letters in the same row are significantly different ($P \leq 0.05$).

The effect of dietary chromium supplementation on blood glucose for the different groups is illustrated in Fig. 1

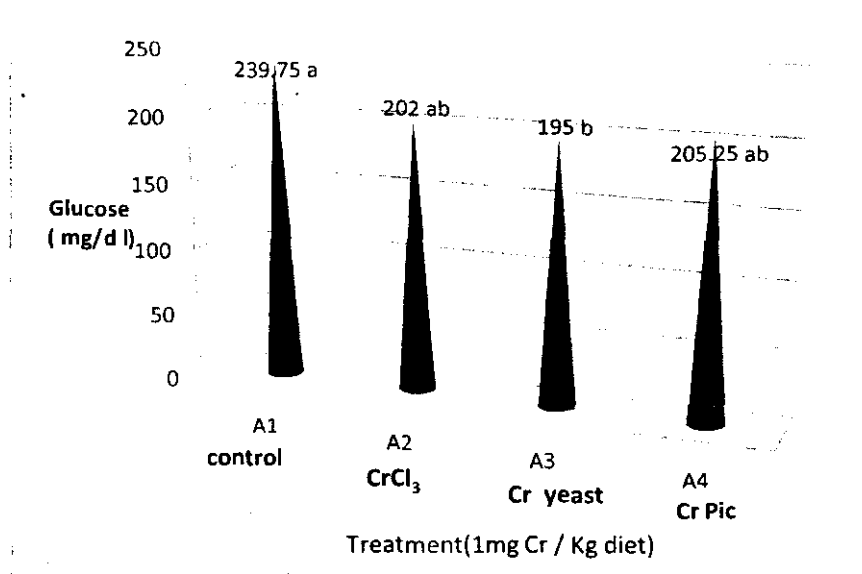


Fig. 1 : Effect of chromium chloride (CrCl₃), chromium yeast (Cr yeast), chromium picolinate (Cr Pico) supplementation to diet on plasma glucose concentration in broiler chickens at 5 weeks of age.

The data obtained showed that blood glucose of all chickens groups which received diets supplemented by inorganic CrCl₃ (A₂) and organic Cr yeast and Cr Pico(A₃ and A₄) chromium were 202.00, 195 and 205.25 mg/dl respectively lower when compared with those control group (A₁) 239.75 mg/dl. The data showed that Cr yeast (A₃) dietary decrease plasma glucose significantly ($P \leq 0.05$) than A₁ group

Plasma proteins Table 4 increased significantly ($P \leq 0.05$) due to inclusion of organic chromium Cr yeast and Cr Pico (A_3 and A_4) (3.15, 3.59 g/dl) into broiler chickens diet simultaneously plasma albumin values were not affected by treatments so that plasma globulin of chickens which received diets supplemented with Cr yeast (A_3) and Cr Pico (A_4) was higher ($P \leq 0.05$) 1.72 , 2.12 g/dl respectively, than those of the control group (A_1) 1.04 g/dl.

The effects of supplemental chromium on lipid derivatives in broiler chickens are presented in Table 4. There was a reduction in plasma total lipids. In groups which received diets supplemented by inorganic and organic chromium, plasma total lipids concentration for different treatments were 4.77, 3.8, 4.22 and 3.37 mg/dl for A_1 , A_2 , A_3 and A_4 group respectively. The lowest ($P \leq 0.05$) total lipid was obtained in the Cr Pico groups (A_4) than the control. There was no significance difference among control group and other chromium diet groups. While there were no significant effects of Cr supplementation on triglycerides. The triglycerides level, was the highest in the control group among the treated groups. The concentration were 174.00 , 140.33 , 137.5 and 146.00, in A_1 , A_2 , A_3 , A_4 respectively . Analysis of variance of the recorded data indicated a non significant differences between Cr supplemented groups and the control in plasma HDL. levels were 88.75, 99.25 88.5 and 88.75 mg/dl for A_1 , A_2 , A_3 and A_4 respectively.

Table 4. Effect of chromium chloride ($CrCl_3$) chromium yeast (Cr yeast), chromium picolinate (Cr Pico) on plasma total protein, Albumin, globulin , total lipid , triglycerides and High Density Lipoprotein(HDL) in broiler chickens at 5 weeks of age.

Characters	Treatments (1 mg / kg diet)			
	Control	$CrCl_3$	Cr yeast	Cr Pico
	A_1	A_2	A_3	A_4
Total protein (g/dl)	2.43 ^c	2.83 ^{bc}	3.15 ^{ab}	3.59 ^a
Albumin (g/dl)	1.39 ^a	1.67 ^a	1.42 ^a	1.47 ^a
Globulin (g/dl)	1.04 ^c	1.16 ^{bc}	1.72 ^{ab}	2.12 ^a
Total lipid (mg/dl)	4.77 ^a	3.90 ^{ab}	4.22 ^{ab}	3.37 ^b
Triglycerides (mg/dl)	174.00 ^a	140.33 ^a	137.50 ^a	146.00 ^a
HDL (mg/dl)	88.75 ^a	99.25 ^a	88.50 ^a	88.75 ^a

Means having different letters in the same row are significantly different ($P \leq 0.05$).

Plasma cholesterol levels of broiler chickens significantly ($P \leq 0.05$) reduced by feeding diets with inorganic and organic chromium Fig.2. The values of plasma cholesterol concentration for different treatments were 149.5, 115.5, 119 and 120.7 mg/dl for the A₁, A₂, A₃ and A₄ respectively.

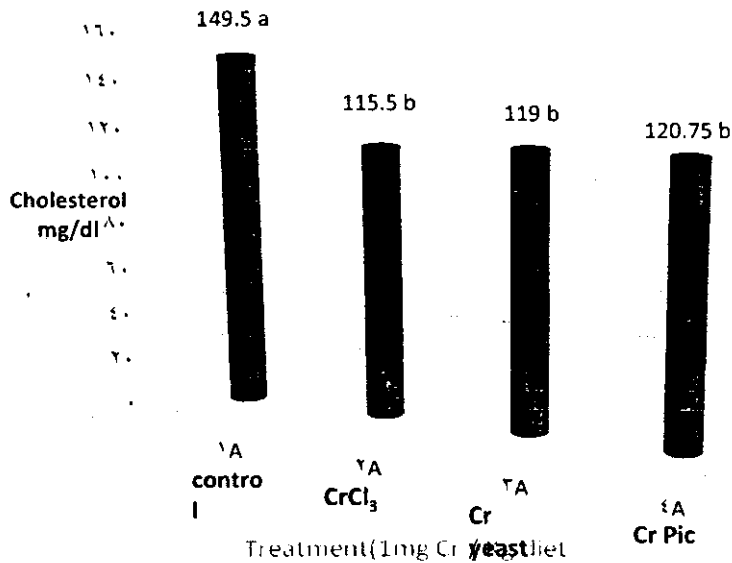


Fig. 2 : Effect of chromium chloride (CrCl₃), chromium yeast (Cr yeast), chromium picolinate (Cr Pico) supplementation to diet on plasma cholesterol in broiler chickens at 5 weeks of age.

Plasma LDL was lowered (not significant) in chromium supplemented group as compared to the control group (Fig. 3). A₂ treatment showed a significantly ($P \leq 0.05$) reduction in LDL (13.66 mg/dl) than the control group (A₁).

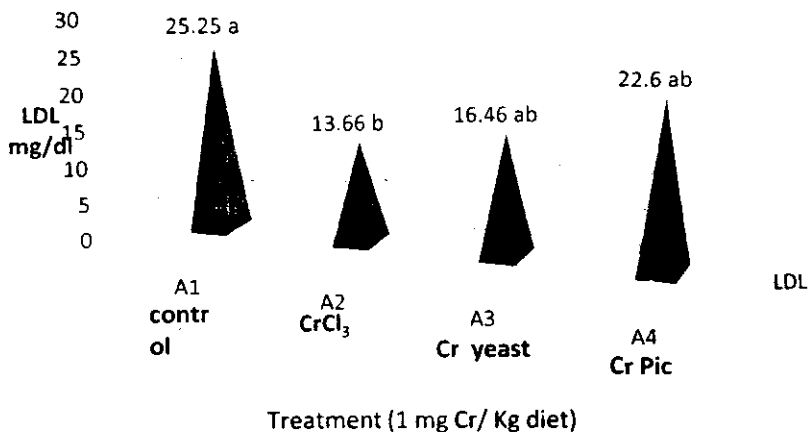


Fig. 3: Effect of chromium chloride (CrCl₃), chromium yeast (Cr yeast), chromium picolinate (Cr Pico) supplementation to diet on plasma Low Density Lipoprotein (LDL) in broiler chickens at 5 weeks of age.

Table 5 represents the effect of chromium supplementation on plasma calcium and phosphorus in broilers. Blood calcium concentration was not affected significantly by supplementing chromium to diets. The concentrations were 4.47, 4.23, 4.23 and 4.4 mg/dl in A₁, A₂, A₃ and A₄ respectively; while phosphorus concentration was significantly ($P \leq 0.05$) lower in broilers which received diet with Cr Pico (A₄) (4.00 mg/dl) as compared to control group (5.94 mg/dl).

The effect of supplemental chromium on the plasma T₃ and T₄ levels of broiler chickens are summarized in Table 5. The data showed a no significant effect of the inorganic CrCl₃ (A₂) and organic Cr yeast and Cr Pico (A₃ and A₄) chromium supplementation on levels of T₃ and T₄, but there was a slightly increase in the level of T₄ hormone in the chickens fed dietary chromium, A₂, A₃ and A₄ 12.52, 12.56 and 12.75 ng/dl respectively as compared with the control group (A₁) 12.20 ng/dl.

Plasma albumin values were not affected by treatments, a lack of Cr effect on plasma albumin was also reported by (Ibrahim *et al.*, 2005 ; Karam *et al.*, 2007).

In general there were a reductions in plasma total lipids, triglycerides (table 4), cholesterol and LDL (Fig. 2 and 3), due to supplementing Cr (significant or not significant), this result confirms the findings of Abraham *et al.*, (1982 a, b) who reported that Cr is essential for lipids metabolism. Also Chen *et al.*, (2001) found a reduction in lipid parameters resulted from increasing Cr level in broiler diets. While HDL was not affected, these results are similar to the findings of Chen *et al.*, (2001) who reported that dietary Cr have not a significant influence on HDL in turkeys.

Table 5 shows that, blood calcium was not affected significantly by adding Cr into the diets. There was a single ($P<0.05$) reduction in phosphorus concentration in chickens received Cr picolinate these results are in harmony with those of Uyanik *et al.*, (2002) who found an influence of Cr on phosphorus metabolism in laying hens.

The decreasing in phosphorus level (Cr Pico) may result from the relation of Cr and Phytic acid and conjugation between them produces Cr – phytat complex, consequently, the free phosphorus level in blood will be decreased.

There were no significant differences in T3 and T4 hormone between dietary treatment (table 5). Adding the Cr (both organic and inorganic) in chick diets decreased glucose level in blood, this may be due to increase insulin secretion and consequently decrease thyroid hormones secretion, this may be due to the antagonistic relationship between thyroid hormone and insulin.

The results shows that the dietary Cr supplementation had no effect on plasma uric acid or serum creatinine (table 6). Cr (organic, inorganic) levels used in the present study were safe for birds and had no deleterious effects on kidney function. These results were in agreement with those obtained by Mohamed and Afifi (2001) who reported that there was no effect of $CrCl_3$ on uric acid and creatinine.

The results show that dietary Cr had no effect on plasma ALT, AST (table 6). The Cr supplementation had no deleterious effect on liver function. This result is in agreement with that obtained by Mustafa (2007).

Using inorganic (CrCl_3), organic (Cr yeast, Cr Pico) led to a significant increase the liver, muscle, plasma chromium levels (table 7). This result was in agreement with those obtained by Uyanik *et al.*, (2002) who recorded that chromium supplementation increased serum Cr concentration.

The overall result of this work indicated that supplementing chromium did have a beneficial effect of some of the parameters measured.

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الملخص العربي

تأثير اضافة مصادر مختلفة من الكروم للعليقة في بعض الصفات الفسلجية لفروج اللحم

لمى خالد البندر و ضياء خليل ابراهيم وعيسى حسين المشهداني

كلية الزراعة جامعة بغداد

اجريت التجربة في جامعة عين شمس بجمهورية مصر العربية، للمدة من شهر كانون الثاني الى شهر اذار عام ٢٠٠٨ لدراسة تأثير اضافة انواع مختلفة من عنصر الكروم ثلاثي Cr⁺³ في بعض الصفات الفسلجية.

هدفت التجربة الى المقارنة بين استخدام الكروم العضوي ويتضمن كروم البايكولنت (Cr Pico) chromium picolonte وكروم الخميرة (Cr yeast) chromium yeast، والكروم غير العضوي والمتمثل بكلوريد الكروم (CrCl₃) Chromium chloride.

استعمل في هذه التجربة ٤٨٠ فرخ لحم Cobb بعمر يوم واحد ووزعت عشوائيا على اربع معاملات في كل معاملة ١٢٠ طير وبواقع ٣ مكررات لكل معاملة في كل مكرر ٤٠ طير وكالاتي: المعاملة A₁: السيطرة (control) بدون اضافة الكروم الى العليقة. المعاملة A₂: ١ ملغم من CrCl₃ / كغم علف. المعاملة A₃: ١ ملغم من Cr yeast / كغم علف. المعاملة A₄: ١ ملغم من Cr Pico / كغم علف.

واشارت النتائج الى حصول انخفاض معنوي ($P \leq 0.05$) في الكلوكونز للمعاملة A₃، وارتفاع معنوي للبروتين الكلي والكلوبيولين ليلازما الدم في المجاميع التي اضيف الى عليقتها Cr yeast (A₃) و Cr Pico (A₄). انخفض الدهن الكلي معنويا ($P \leq 0.05$) في طيور المعاملة A₄ مقارنة بمعاملة السيطرة، بينما اظهرت طيور المعاملتين A₂ و A₃ انخفاضا حسابيا في مستوى الدهن مقارنة بمعاملة السيطرة. كما انخفض معنويا ($P \leq 0.05$) كوليستيرول بلازما الدم لدى الطيور التي غذيت على جميع انواع الكروم. كما انخفض معنويا ($P \leq 0.05$) LDL في الطيور التي اضيف الى علاقتها CrCl₃ (A₂)، بينما اضافة مصادر الكروم الاخرى أدت الى انخفاض حسابي في LDL مقارنة بمعاملة السيطرة. ان النسبة المئوية للطحال ارتفعت معنويا في المعاملة A₄ مقارنة مع السيطرة بينما نلاحظ انخفاض في تركيز فسفور البلازما في A₄ مقارنة مع السيطرة. اضافة CrCl₃ (A₂) او Cr Pico (A₄) أدت الى زيادة معنوية ($P \leq 0.05$) في مستوى الكروم في الكبد، العضلات و في البلازما للمعاملة A₂ فقط.

يمكن الاستنتاج بان مستويات الكروم سواء العضوي والغير عضوي له تأثير ايجابي على بعض الصفات الفسلجية التي تم قياسها في هذه الدراسة