

## Addition of Chromium ( $\text{Cr}^{+3}$ ) in the diets containing fermented yellow corn meal on jelawat, *Leptobarbus hoevenii*

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**Abstract.** Yanto H, Junianto, Rostika R, Andriani Y, Iskandar. 2017. Addition of Chromium ( $\text{Cr}^{+3}$ ) in the diets containing fermented yellow corn meal on jelawat, *Leptobarbus hoevenii*. *Nusantara Bioscience* 9: 214-219. This experiment aimed to find the optimal level of  $\text{Cr}^{+3}$  in the diets containing fermented yellow corn meal to increase the growth of jelawat (*Leptobarbus hoevenii* Bleeker, 1851). The completely randomized design had five levels of  $\text{Cr}^{+3}$ , they were A0 (0.52), A1 (1.55), A2 (3.03), A3 (4.52) and A4 (6.04) mg  $\text{kg}^{-1}$  in the diet. The source of  $\text{Cr}^{+3}$  was  $\text{CrCl}_3 \cdot 6\text{H}_2\text{O}$  which was fermented by *Saccharomyces cerevisiae*. The results showed that  $\text{Cr}^{+3}$  in diets could activate the insulin and regulate the blood glucose. The fastest increase in blood glucose occurs 5 hours after feeding on Chromium 1.5 mg  $\text{kg}^{-1}$ . The liver and muscle glycogen, protein and lipid of bodies, protein and lipid retentions, daily growth rate, and feed efficiency were significant ( $P < 0.05$ ). The diet containing  $\text{Cr}^{+3}$  1.55 mg  $\text{kg}^{-1}$  resulted in the highest for liver glycogen, body protein, body lipid, protein and lipid retentions, daily growth rate and feed efficiency. The  $\text{Cr}^{+3}$  level of 1.55 mg  $\text{kg}^{-1}$  in the diet contained 30% fermented yellow corn meal and 42.79% total carbohydrate was the best for the growth and feed efficiency of jelawat.

**Keywords:** Blood glucose, feed efficiency, growth rate, respiratory quotient

### INTRODUCTION

Jelawat (*Leptobarbus hoevenii* Bleeker, 1851), a freshwater fish, has a good prospect for aquaculture development. It is one of the high economic value fish and Indonesia's export commodities (Warta Pasarikan 2010). Then jelawat had been spawned successfully by the induced breeding technology, so the fingerlings of jelawat have been available to support the its culture. As an omnivorous fish, jelawat may consume various sources of food easily. The food habit has potentials to utilize various diet made of plant materials efficiently.

The fish has the different ability in utilizing the carbohydrate. The omnivorous fish can utilize the carbohydrate only about 30 to 40%, and the carnivores are about 10% to 20% of the total diet formulation due to the ability of the fish in producing amylase and insulin activity are low (Anderson and De Silva 2003). The increasing of carbohydrate utilization and sensitivity insulin are important for jelawat.

Selection of carbohydrate sources and fermentation increase the use of carbohydrate in fish. One of the resources is yellow corn meal containing the high carbohydrate level as an energy source, so it is usually used an ingredient in formulating the fish diet. In another way, the nutrition number in yellow corn meal can be increased by using fermented technology. The use of fermented ingredients increased the amount of nutrient and feed digestibility, growth and feeding efficiency in common carp, *Cyprinus carpio* (Suprayudi et al. 2012) and nile

tilapia, *Oreochromis* sp. (Mulyasari et al. 2013). The fermented yellow corn meal can be tried as the energy source in the diet for jelawat.

According to Gatlin III (2010), the addition of chromium ( $\text{Cr}^{+3}$ ) in diet could increase the sensitivity of insulin, so it could transfer the blood glucose into cells. Feeding the optimal level of  $\text{Cr}^{+3}$  in the diet increased the activity of Glucose-6-Phosphate Dehydrogenase (G6PDH) and 6-Phospho-gluconate Dehydrogenase (6PGDP) enzyme in the liver of hybrid tilapia, *O. niloticus* x *O. aureus* (Pan et al. 2003); the protein retention in common carp, *C. carpio* (Mokoginta et al. 2004); regulation of blood glucose in tilapia, *O. niloticus* (Setyo 2006); daily growth rate and feed efficiency to the African catfish, *Clarias gariepinus* (Aryansyah et al. 2007) and the RNA/DNA ratio, the growth rate and feed efficiency for baung, *Hemibagrus nemurus* (Sari et al. 2009). Giving the chromium in the diet to jelawat according to the need is important to be done in order to increase the carbohydrate metabolism and growth of jelawat.

The need of  $\text{Cr}^{+3}$  is different for every fish. For the examples, gurami, *Osphronemus gouramy* needs 1.50 ppm  $\text{Cr}^{+3}$  (Subandiyono 2004), common carp, *C. carpio* needs 1.59-2.16 ppm  $\text{Cr}^{+3}$  (Mokoginta et al. 2004), and nile, *O. niloticus* 4.50 ppm  $\text{Cr}^{+3}$  (Setyo 2006), and African catfish, *C. gariepinus* needs  $\text{Cr}^{+3}$  of 2.60 mg  $\text{kg}^{-1}$  (Aryansyah et al. 2007), and baung fish, *H. nemurus* is 3.2 mg  $\text{kg}^{-1}$   $\text{Cr}^{+3}$  in the diet (Sari et al. 2009). Conversely, feeding in the diet contained  $\text{Cr}^{+3}$  as much as 2 mg  $\text{kg}^{-1}$  did not affect the growth of hybrid tilapia (Pan et al. 2003), and the addition

of chromium picolinate 1.6 mg kg<sup>-1</sup> in the diet did not affect growth performance and the ratio of conversion of feed on salmon, *Onchorhynchus mykiss*, (Selcuk et al. 2010). Based on the above, the optimal levels of Cr<sup>+3</sup> in the diet containing fermented yellow corn meal is necessary to be determined to increase the growth and feed efficiency of jelawat.

## MATERIALS AND METHODS

### Experimental design

This research used completely randomized design with different levels of Cr<sup>3+</sup> in diets consisting of A0 (0.52 mg kg<sup>-1</sup>, as control), A1 (1.55 mg kg<sup>-1</sup>), A2 (3.03 mg kg<sup>-1</sup>), A3 (4.52 mg kg<sup>-1</sup>) and A4 (6.04 mg kg<sup>-1</sup>). Each treatment had 4 replications and 1 additional replication to complete the need of fish in blood glucose test.

### Experimental diets

There were five experimental diets in this research. The experimental diets were designed containing different levels of Cr<sup>+3</sup> according to the treatment. Based on the analysis, the fermented yellow corn meal had contained chromium 1.66 mg kg<sup>-1</sup>, so that the chromium was added in diets according to its deficiency and need according to the treatment. The organic chromium was produced using CrCl<sub>3</sub> 6H<sub>2</sub>O and *Saccharomyces cerevisiae* (Suryadi et al.

2011). The experimental diets used *Saccharomyces cerevisiae* with 0.9% kg<sup>-1</sup> to ferment the yellow corn meal (Suprayudi et al. 2012). All diets were also added by *Saccharomyces cerevisiae* so the protein contents of them were same. Then experimental diets were designed to contain the same protein and energy of 30% and 2,700 kcal kg<sup>-1</sup> respectively. The diet formulation and proximate analysis and Cr<sup>+3</sup> level of experimental diets could be seen in Table 1.

### Fish cultivation

Jelawat which were about 34.58±1.98 g kept in aquariums size 40 x 60 x 40 cm, and containing water volume of 72 L. The density of jelawat was 10 fish per aquarium for each replication, and 50 fish for each treatment. Jelawat has been raised for 60 days, and they fed 3 times a day at 8.00 a.m, 12.00 p.m, and 4.00 p.m with the experimental diets satisfyingly. The aquariums were cleaned in the morning before feeding it, and the water was replaced about 70% of the total volume. Aquariums had been equipped with an aeration system. According to the water quality measurement, the dissolved oxygen was 5.75-6.40 ppm, the temperature was 28-31 °C, the acid degree (pH) was 6.29-6.45 and ammonia total was 0.20-0.30 ppm. The water quality was sufficient to support the growth and life of jelawat. This resulted in 100% survival rate of jelawat in all experimental units.

**Table 1.** Formulation and content of nutrients in the experimental diets

Ingredients	Level of Cr <sup>+3</sup> (mg kg <sup>-1</sup> ) in the diets according to treatments				
	A0 (0.52)	A1 (1.55)	A2 (3.03)	A3 (4.52)	A4 (6.04)
Soybean meal (%)	8.50	8.50	8.50	8.50	8.50
Squid meal (%)	5.00	5.00	5.00	5.00	5.00
Fermented yellow corn meal (%)	30.00	30.00	30.00	30.00	30.00
Pollard (%)	12.00	12.00	12.00	12.00	12.00
Tapioca flour (%)	5.00	5.00	5.00	5.00	5.00
Fish oil (%)	2.00	2.00	2.00	2.00	2.00
Corn oil (%)	1.50	1.50	1.50	1.50	1.50
Palm oil (%)	0.80	0.25	0.25	0.25	0.25
Vitamin mix (%) <sup>1</sup>	1.00	1.00	1.00	1.00	1.00
Mineral mix (%) <sup>2</sup>	1.00	1.00	1.00	1.00	1.00
α Cellulose (%)	2.78	2.78	2.78	2.78	2.78
Cr <sup>+3</sup> yeast	0.00	0.08	0.19	0.31	0.42
Yeast	0.42	0.34	0.23	0.11	0.00
Proximate composition (% dry weight) and content of Cr <sup>+3</sup> :					
Crude protein (%)	31.84	31.57	32.05	32.10	31.93
Crude lipid (%)	9.90	10.37	11.05	10.07	10.61
Ash (%)	11.86	11.12	11.36	11.71	11.76
Crude (%)	4.55	4.14	3.64	3.58	3.10
Nitrogen free extract (%)	41.85	42.79	41.91	41.54	42.60
Total energy (kcal) <sup>3</sup>	2,735.17	2,777.36	2,836.18	2,834.46	2,807.75
Ratio energy/protein	8.59	8.80	8.85	8.83	8.79
Cr <sup>+3</sup> (mg kg <sup>-1</sup> )	0.52	1.55	3.03	4.52	6.04

Note: 1) every kg of vitamin mix contains: vit. A 3,000,000 IU; vit. D3 1,000,000 IU; vit. K3 1,200 mg; vit. E 7,500 mg; vit. B1 3,000 mg; vit. B2 4,500 mg; vit B6 3,000 mg; vit. B12 3,000 mg; vit. C 8,000 mg; Ca panthotenate 4,500 mg; folic acid 1,500 mg; biotin 1,000 mg; inositol 12,500 mg; nicotinamide 20,000 mg; choline chloride 15,000 mg; L-lysine 20,000 mg; DL-methionine 5,000 mg and 2) every 100 g of mineral containing: NaCl 1.00 g macro minerals; MgSO<sub>4</sub> 7H<sub>2</sub>O 15.00 g; NaH<sub>2</sub>PO<sub>4</sub> H<sub>2</sub>O 25.00 g; KH<sub>2</sub>PO<sub>4</sub> 32.00 g; Ca (H<sub>2</sub>PO<sub>4</sub>) 2H<sub>2</sub>O 20.00 g; Fe-citrate 2.50 g; Trace element mix 1.00 g; Ca-lactate 3.50 g; micro minerals ZnSO<sub>4</sub> 7H<sub>2</sub>O 35.30 g; MnSO<sub>4</sub> H<sub>2</sub>O 16.20 g; CuSO<sub>4</sub> 5H<sub>2</sub>O 3.10 g; CoCl<sub>2</sub> 6H<sub>2</sub>O 0.10 g KIO<sub>3</sub> 0.30 g and 45.00 g cellulose. 3) calculated according to an energy value of protein 4 kcal g<sup>-1</sup>, lipid 8 kcal g<sup>-1</sup> and carbohydrate 1.6 kcal g<sup>-1</sup>.

### The glucose tolerance test

In the final research, the glucose tolerance test was done by collecting jelawat from all replications from the same treatments in the 0.5 m<sup>3</sup> fiberglass tanks, and they were randomly divided into nine groups of times in taking the blood sample. Three jelawat were placed in the aquarium to take the blood samples as the repetition of treatments, and the jelawat was taken each hour. Then, jelawat has not been fed for 48 hours.

The blood sample was taken before feeding and then continuing after 1, 2, 3, 5, 7, 9, 11, and 18 hours feeding the fish. Jelawat was sedated with 12.55 ppm MS-222 (Tricaine Methane Sulfonate) to reduce the stress. The blood sample was taken from the caudal peduncle of jelawat using 2.5 ml syringe that has been rinsed with 3.8% anticoagulant solution (sodium citrate). The blood sample was inserted into 1.5 ml Eppendorf tube and centrifuged at 3,000 rpm for 10 minutes to analyze the blood glucose with a spectrophotometer.

### Determining the respiratory quotient and ammonia excretion

Determining the respiratory quotient (RQ) and ammonia excretion was aimed to determine the dominant nutrient in the diet which was catabolized by the fish. In the final of the research, the fish that had been combined from replications of every treatment was divided into three replications. Thus, there were 15 aquariums used and each unit was contained three fish. Then jelawat had not been feed for 24 hours. The water as the sample was taken after feeding the fish satisfyingly one time. The water was taken every hour for five hours, and it was started after the fish stopped eating, like 0 hours. During respiratory quotient and ammonia excretion measurement, the top of the aquarium was covered by styrofoam to block the air exchange, and the aeration was turned off.

### Sample collection and chemical analysis

In observing the growth of body weight, jelawat in each unit was weighed every 15 days. At final, three fish were randomly taken from each unit experiment (replication), then they were combined in a place to do proximate analysis, glycogen analysis, and Cr<sup>+3</sup> on its muscles and whole body. Besides, three liver of jelawat were combined as one composite in each replication to do liver glycogen analysis. This was to satisfy the needs of liver glycogen analysis. Next, the rest of the fish (7 fish) in each experimental unit (replication) was collected according to the same treatments, and it was taken three fish for the blood glucose test and three fish in every treatment group for respiratory quotient and ammonia excretion test.

The proximate analysis was done to the ingredients of diet, experimental diets, jelawat body, and fish feces. The water content was measured by heating the sample material for 24 hours in an oven at temperature 65 °C, the levels of crude protein using Folch method (Takeuchi 1988). Blood glucose was analyzed based on Wedemeyer and Yasutake using spectrophotometer at a wavelength 635 nm (Handayani 2006). Liver and muscle glycogen levels of

jelawat were also analyzed using Wedemeyer and Yasutake method with sample glucose level measurement, then it plotted in the glucose standard curve, where 1 mg glycogen = 1.11 g glucose (Handayani 2006). Then, the chromium of jelawat's liver and muscle were analyzed with a spectrophotometer at a wavelength 350 nm (Takeuchi 1988).

### Data analysis

The observed variables were blood glucose level, liver and body glycogen, chromium in the whole body and muscle, protein and lipid body content, protein and lipid retention, daily growth rate, feeding efficiency, respiratory quotient and ammonia excretion. The data was analyzed statistically with one-way analysis of variance (ANOVA), except blood glucose descriptively. Differences were considered significant at P < 0.05. Analysis of variance and Duncan's multiple range test was assisted by PASW Statistic 18 software.

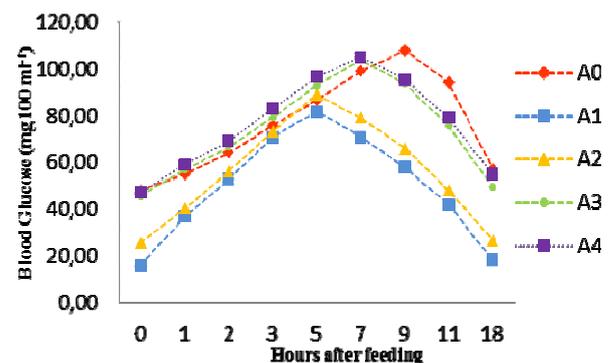
## RESULTS AND DISCUSSION

### Blood glucose pattern

The peak of blood glucose was different among the treatments. The highest level in treatment A0 (control) occurred 9 hours after feeding (postprandial), treatment A1 and A2 occurred after 5 hours and A3 and A4 occurred after 7 hours. Blood glucose levels returned to normal at 18 hours after feeding. The blood glucose pattern of jelawat after feeding the food fish can be seen in Figure 1.

### The chemical composition of body, liver, and muscle

The content of Cr<sup>+3</sup> in jelawat's muscle and body after doing the experiment were higher than before the experiment done. The amount of Cr<sup>+3</sup> in jelawat's muscle and body raised along with the increasing of Cr<sup>+3</sup> in the diets after the experiment. Levels of Cr<sup>+3</sup> in the muscle and body were significantly different (P<0.05). The glycogen level of the liver in jelawat fed the diets containing Cr<sup>+3</sup> of 1.55, 3.03, 4.52 and 6.04 mg kg<sup>-1</sup> (treatments A1-A4) were higher than the diet containing Cr<sup>+3</sup> of 0.52 mg kg<sup>-1</sup> (treatment A0). The



**Figure 1.** The blood glucose pattern of jelawat based on 18 hours treatment after feeding experimental diets in the different supplemented chromium

glycogen of liver in treatment A1 ( $113.33 \pm 1.96 \mu\text{g g}^{-1}$ ) was the highest and treatment A0 (control) was the lowest ( $56.84 \pm 1.98 \mu\text{g g}^{-1}$ ). The glycogen of liver was significant ( $P < 0.05$ ). The glycogen of muscle in treatment A1 ( $29.58 \pm 0.40 \mu\text{g g}^{-1}$ ) was the highest and treatment A0 (control) was the lowest ( $18.80 \pm 0.46 \mu\text{g g}^{-1}$ ). The glycogen of muscle was significant ( $P < 0.05$ ) (Table 2).

The levels of protein and lipid of bodies in jelawat after doing the experiment were higher than before the experiment done. The protein of body in jelawat feeding the diet containing  $\text{Cr}^{+3}$  of  $1.55 \text{ mg kg}^{-1}$  (treatment A1) was the highest ( $59.86 \pm 0.60\%$ ) and treatment containing  $\text{Cr}^{+3}$  of  $0.52 \text{ mg kg}^{-1}$  (treatment A0) was the lowest ( $54.75 \pm 0.82\%$ ). The protein of body was significant ( $P < 0.05$ ). The lipid of bodies in jelawat feeding the diet containing  $\text{Cr}^{+3}$  of  $1.55 \text{ mg kg}^{-1}$  (treatment A1) was the highest ( $30.21 \pm 0.81\%$ ) and treatment containing  $\text{Cr}^{+3}$  of  $0.52 \text{ mg kg}^{-1}$  (treatment A0) was the lowest ( $24.74 \pm 0.60\%$ ). The lipid of body was significant ( $P < 0.05$ ).

The pattern of protein and lipid retentions were almost similar. The protein and lipid retention in diets containing the addition of  $\text{Cr}^{+3}$  in treatments A1, A2, A3, and A4 were higher than the control treatment (A0). The highest protein retention was resulted by treatment A1 ( $47.21 \pm 0.71\%$ ), and the lowest protein retention was resulted by treatment A0 ( $38.16 \pm 0.49\%$ ). The protein retention was significant

( $P < 0.05$ ). The highest lipid retention was resulted by treatment A1 ( $73.43 \pm 1.67\%$ ), and the lowest lipid retention was resulted by treatment A0 ( $57.15 \pm 1.91\%$ ). The lipid retention was significant ( $P < 0.05$ ).

#### Parameter utilization of diet

The lowest daily rapid growth was in treatment A0 ( $2.61 \pm 0.04\%$ ), and the highest was in treatment A1 ( $2.87 \pm 0.05\%$ ), but it was not significantly different to treatment A2 ( $2.82 \pm 0.09\%$ ) ( $P > 0.05$ ). The daily growth rate of jelawat was significant ( $P < 0.05$ ). The lowest feed efficiency was in treatment A0 ( $77.05 \pm 1.95\%$ ), and the highest feed efficiency was in treatment A1 ( $89.78 \pm 1.16\%$ ). The feed efficiency was significant ( $P < 0.05$ ) (Table 3).

#### Respiratory quotient and ammonia excretion

The entire treatments had a respiratory quotient (RQ) close to 1. The lowest RQ was in treatment A0 ( $0.81 \pm 0.02$ ) and the highest was in treatment A1 ( $0.97 \pm 0.02$ ). Respiratory quotient was significant ( $P < 0.05$ ). Otherwise, the highest ammonia excretion was in treatment A0 ( $0.0073 \pm 0.00044 \text{ mg g}^{-1}$  of body hour $^{-1}$ ) and the lowest was in treatment A1 ( $0.0039 \pm 0.00012 \text{ mg g}^{-1}$  of body hour $^{-1}$ ). Ammonia excretion was significant ( $P < 0.05$ ) (Table 4).

**Table 2.** The chromium levels of muscle and body ( $\text{mg kg}^{-1}$ ), liver and muscle glycogens ( $\mu\text{g g}^{-1}$ ), protein and lipid of the bodies (% dry weight), protein and lipid retentions (% dry weight) on jelawat before and after cultivation for 60 days.

Parameters	Initial	Levels of $\text{Cr}^{+3}$ ( $\text{mg kg}^{-1}$ ) in the diets according to treatments				
		A0 (0.52)	A1 (1.55)	A2 (3.03)	A3 (4.52)	A4 (6.04)
$\text{Cr}^{+3}$ of muscle	$0.21 \pm 0.02$	$0.24 \pm 0.09^a$	$0.57 \pm 0.06^b$	$0.68 \pm 0.13^b$	$0.88 \pm 0.08^c$	$0.97 \pm 0.04^c$
$\text{Cr}^{+3}$ of body	$0.26 \pm 0.02$	$0.30 \pm 0.03^a$	$0.62 \pm 0.03^b$	$0.76 \pm 0.01^c$	$0.94 \pm 0.02^d$	$1.07 \pm 0.05^e$
Glycogen of liver	-	$56.84 \pm 1.98^a$	$113.33 \pm 1.96^c$	$105.71 \pm 1.75^d$	$70.86 \pm 0.99^c$	$60.00 \pm 1.26^b$
Glycogen of muscle	-	$18.80 \pm 0.46^a$	$29.58 \pm 0.40^c$	$28.62 \pm 0.33^d$	$26.26 \pm 0.43^c$	$23.76 \pm 0.23^b$
Protein of body	51.70	$54.75 \pm 0.82^a$	$59.86 \pm 0.60^c$	$57.55 \pm 0.30^b$	$55.29 \pm 0.59^a$	$54.88 \pm 0.19^a$
Lipid of body	20.31	$24.74 \pm 0.60^a$	$30.21 \pm 0.81^c$	$29.73 \pm 0.24^c$	$28.77 \pm 0.35^b$	$28.08 \pm 0.37^b$
Protein retention	-	$38.16 \pm 0.49^a$	$47.21 \pm 0.71^d$	$46.46 \pm 0.93^d$	$43.22 \pm 0.78^c$	$40.33 \pm 1.68^b$
Lipid retention	-	$57.15 \pm 1.91^a$	$73.43 \pm 1.67^d$	$72.30 \pm 1.667^{cd}$	$70.21 \pm 0.79^{bc}$	$66.99 \pm 3.19^b$

Note: Means with a common superscript letter in the same row were not significantly different ( $P > 0.05$ ) by Duncan's test.

**Table 3.** Initial and final weight, daily growth rate, feed consumption and feed efficiency of jelawat cultivated for 60 days.

Variables	Levels of $\text{Cr}^{+3}$ ( $\text{mg kg}^{-1}$ ) in the diets according to treatments				
	A0 (0.52)	A1 (1.55)	A2 (3.03)	A3 (4.52)	A4 (6.04)
Initial weight (g)	$35.10 \pm 1.51$	$34.23 \pm 1.13$	$33.83 \pm 1.03$	$33.88 \pm 1.03$	$35.30 \pm 1.19$
Final weight (g)	$164.83 \pm 3.96$	$186.78 \pm 1.71$	$179.78 \pm 2.23$	$170.73 \pm 2.42$	$169.05 \pm 1.24$
Daily growth rate (%)	$2.61 \pm 0.04^a$	$2.87 \pm 0.05^c$	$2.82 \pm 0.09^c$	$2.74 \pm 0.03^b$	$2.65 \pm 0.05^a$
Feed consumption (g)	$1,684.00 \pm 13.44^a$	$1,699.25 \pm 18.19^a$	$1,689.75 \pm 18.84^a$	$1,684.50 \pm 14.25^a$	$1,687.25 \pm 18.55^a$
Feed efficiency (%)	$77.05 \pm 1.95^a$	$89.78 \pm 1.16^d$	$86.38 \pm 1.49^c$	$80.06 \pm 0.87^b$	$78.54 \pm 1.43^{ab}$

Note: Means with a common superscript letter in the same row were not significantly different ( $P > 0.05$ ) by Duncan's test

**Table 4.** Respiratory quotient (RQ) and ammonia excretion.

Variables	Levels of $\text{Cr}^{+3}$ ( $\text{mg kg}^{-1}$ ) in the Diets According to Treatments				
	A0 (0.52)	A1 (1.55)	A2 (3,03)	A3 (4,52)	A4 (6,04)
Respiratory quotient (RQ)	$0.81 \pm 0.02^a$	$0.97 \pm 0.02^c$	$0.93 \pm 0.05^{bc}$	$0.91 \pm 0.01^b$	$0.86 \pm 0.02^a$
Ammonia excretion ( $\text{mg g}^{-1}$ of body hour $^{-1}$ )	$0.0073 \pm 0.00044^b$	$0.0039 \pm 0.00012^a$	$0.0049 \pm 0.00040^a$	$0.0070 \pm 0.00140^b$	$0.0071 \pm 0.00031^b$

Note: Means with a common superscript letter in the same row were not significantly different ( $P > 0.05$ ) by Duncan's test

## Discussion

Feeding the diets containing different levels of  $\text{Cr}^{+3}$  affected the levels of  $\text{Cr}^{+3}$  in the muscle and body of jelawat. The higher the  $\text{Cr}^{+3}$  in the diets, the higher the levels of  $\text{Cr}^{+3}$  in the muscle and body of jelawat (Table 2). Watanabe et al. (1997) and Gatlin III (2010) stated that as an essential micromineral,  $\text{Cr}^{+3}$  plays an important role in the metabolism of carbohydrates, especially to improved the regulation and utilization of glucose. The function of  $\text{Cr}^{+3}$  with the utilization of blood glucose can be seen on the speed and peak of blood glucose levels in a glucose tolerance test. The peak of blood glucose is the balanced point between blood glucose as the result of digestion and glucose entry into the cells. When the digestive blood glucose is higher than the transferred glucose into the cells of the body, then it needs the higher and longer time for the peak glucose to be accomplished. The experimental diet with concentration  $1.55 \text{ mg kg}^{-1} \text{ Cr}^{+3}$  had the lower peak and need a shorter time to be accomplished than the other diets, and it was the optimal level of  $\text{Cr}^{+3}$  in the diet to transfer the blood glucose into target cells. This is consistent with the research results of Mehrim (2014), in which blood glucose of tilapia (*O. niloticus*) at low ( $127 \text{ mg dL}^{-1}$ ) when fed containing chromium picolinate optimal of 400 ppb. Then levels of chromium carbochelate were 18 and  $36 \text{ mg kg}^{-1}$  in the diet decreased blood glucose significantly in pacu, *Piaractus mesopotamicus* after 24 and 48 hours of feeding (De Castro et al. 2014). The low peak of blood glucose was due to transfer into the target cells running optimally.

The transferring of blood glucose into the cells cannot be separated from the role of insulin affected by the chromium as the cofactor (Shiau 1997; Watanabe 1997; Gatlin III 2010), and the chromium has the optimum range score to do the optimal function (Aryansyah et al. 2007). The chromium biological function decreased such as in the diet not contain chromium in non-optimal range (Subandiyono 2004). Therefore, the chromium level must be optimum in the diet. It can improve the insulin activity, so the blood glucose is transferred into the target cells optimally as the metabolic energy. This causes the activity of various enzymes related to carbohydrate metabolism also increased in cells. For example, feeding chromium of  $2 \text{ mg kg}^{-1}$  in the diet increased the activity of various enzymes for carbohydrate metabolism, such as fructose 1,6-diphosphatase (FDPase), glucose-6-phosphatase dehydrogenase (G6PDH), and 6-phosphogluconate dehydrogenase (6PGDH) in tilapia liver cells (*O. niloticus* x *O. aureus*) (Pan et al. 2003). Then the excess of glucose is stored as the glycogen in the liver cells and the muscles through glycogenesis process.

The chromium also has the role in glycogenesis involving the various enzymes for the formation of the glycogen. In the liver cells and muscles, the insulin works to stimulate to form the glycogen as the energy reserves (Bedner and Mayes 2006). Feeding 2 ppm of chromium to the tilapia, *O. niloticus* x *O. aureus* (Shiau 1997), and feeding 1.5 ppm on gurami, *Osphronemus gouramy* (Subandiyono 2004) increased the glycogen in their livers and muscles. Feeding  $0.8 \text{ mg kg}^{-1}$  of chromium in the diet

to *Labeo rohita* increased the liver glycogen (Giri et al. 2014). Therefore,  $1.55 \text{ mg kg}^{-1}$  chromium in the diet was optimum to produce the highest glycogen of liver and muscle in jelawat.

The utilization of glucose as metabolic energy in target cells gives the opportunity for the body cells to utilize the protein and lipid efficiently as the growth support. Therefore, the increasing of protein and lipid of the body were also due to the metabolism of amino acid and lipid relating to the insulin activity in the body. The insulin hormone is also called "hormone of abundance" which means insulin hormone leads directly to the accumulation of excess carbohydrate, lipid, and protein (Suryadi et al. 2011). Therefore chromium as a cofactor of insulin should be optimized in the diet to maximize the insulin function physiologically. For example, feeding 3.20 ppm chromium produced the highest RNA and RNA/DNA ratio in baung, *M. nemurus* (Sari et al. 2011), addition of chromium picolinate of  $0.8 \text{ mg kg}^{-1}$  in the diet increased the muscles protein *L. rohita* be  $28.95 \pm 0.91 \text{ mg g}^{-1}$  (Giri et al. 2014), and the addition of 400 ppb chromium picolinate in the diet increased protein of muscles to be 63.7% in tilapia, *O. niloticus* (Mehrim 2012 and Mehrim 2014). The activity of insulin doesn't only encourage the protein synthesis but also improves the lipid accumulation. Therefore it causes the content of the protein and lipid of the body becomes high. This phenomenon could also be seen in the other research such as gurami, *O. gouramy* (Subandiyono 2004), common carp, *C. carpio* (Mokoginta et al. 2004), tilapia, *O. niloticus* (Mokoginta et al. 2005), African catfish, *C. gariepinus* (Aryansyah et al. 2007), baung, *M. nemurus* (Sari et al. 2009). Besides, supplementation of chromium picolinate (Cr Pic.) prevents the decrease of cortisol level as the main hormone relate to the nutrient catabolism, and it increases the content of insulin-like growth factor-I (IGF-I) producing protein deposition (Xi et al 2001). For example, pacu (*P. mesopotamicus*) produces the lowest cortisol when feeding chromium carbochelate as much as 18 and  $36 \text{ mg kg}^{-1}$  in the diet (De Castro et al. 2014).

The increasing of chromium supplementation which is too high can suppress the biological function of another essential mineral. With the capacity of limited transferrin, the  $\text{Cr}^{+3}$  was one of the very potential minerals as the contender in utilizing the facility as the ion  $\text{Fe}^{+3}$  carrier (Subandiyono 2004). This condition caused the decreasing of hemoglobin (Hb) in sangkuriang, *C. gariepinus* when it was fed by the chromium compared to the non-chromium in the diet (Hastuti and Subandiyono 2011). Chromium level was too high also reduce the level of hemoglobin and the number of erythrocytes on tilapia, *O. niloticus* (Mehrim 2014). The decrease in supplying  $\text{Fe}^{+3}$  causes the interference in the process of energy oxidation. Besides, the chromium which is too high can suppress the function of another various mineral, such as  $\text{Na}^+$  on the sodium pump system (sodium pump) (Subandiyono 2004). Finally, the decrease of  $\text{Fe}^{+3}$  impairment supply and  $\text{Na}^+$  function causes the interference in the metabolism and the low growth rate in fish.

The highest muscle protein and lipid accumulation in jelawat feeding the diet containing  $\text{Cr}^{+3}$  of  $1.55 \text{ mg kg}^{-1}$

produced the highest protein and lipid retention. Finally, it also produced the highest daily growth rate and feed efficiency. Many researchers suggested that feeding the chromium optimally in diet to the common carp, *C. carpio* (Mokoginta et al. 2004); tilapia, *O. niloticus* (Mokoginta et al. 2005); gurami, *O. gouramy* (Subandiyono 2004); African catfish, *C. gariepinus* (Aryansyah et al. 2007); baung, *H. nemurus* (Sari et al. 2009); nila, *O. niloticus* (Mehrim 2012); *L. rohita* (Giri et al. 2014) resulted the highest growth rate and feed efficiency.

The respiratory quotient (RQ) is closely related to the kinds of nutrient catabolized in the fish body. The normal RQ is slightly above 0.8 indicating that the catabolized nutrient is the mixing between protein, lipid, or carbohydrate, and RQ between 0.8 – 1.0 indicating the catabolized nutrient is fat (Subandiyono 2004). Based on RQ, the nutrient catabolized by jelawat which consumed the diet containing  $\text{Cr}^{+3}$  of 0.52 mg  $\text{kg}^{-1}$  was the mixing between protein, lipid, and carbohydrate. The nutrients in great quantities catabolized by jelawat consuming the diets containing  $\text{Cr}^{+3}$  of 1.55, 3.03, 4.52 and 6.04 mg  $\text{kg}^{-1}$  were carbohydrate. Jelawat which was consumed the diet with  $\text{Cr}^{+3}$  of 1.55 mg  $\text{kg}^{-1}$  had the higher ability to utilize the carbohydrate efficiently than the others because it had the highest score. Unlike the RQ, the ammonia excretion of jelawat feeding the diet containing  $\text{Cr}^{+3}$  of 1.55 mg  $\text{kg}^{-1}$  was only a few in using the protein as the metabolic energy source. It is shown by the lowest ammonia excretion. It means that the jelawat feeding the diet with the level of  $\text{Cr}^{+3}$  1.55 mg  $\text{kg}^{-1}$  was able to reduce the use of protein and lipid as a metabolic energy. This phenomenon is similar to the research of Subandiyono (2004), that was the gurami, *O. gouramy* fed optimal  $\text{Cr}^{+3}$  (1.50 mg  $\text{kg}^{-1}$ ) resulted in the highest RQ and the lowest ammonia excretion among other treatments. Both of these phenomena proved that the content of  $\text{Cr}^{+3}$  as much as 1.55 mg  $\text{kg}^{-1}$  in the diet was able to utilize carbohydrates as a source of metabolic energy, and utilized protein and lipid efficiently to support the growth of jelawat.

To conclude, the diet containing  $\text{Cr}^{+3}$  of 1.55 mg  $\text{kg}^{-1}$  and 30% fermented yellow corn meal and 42.79% total carbohydrate was the best for the growth and feed efficiency of jelawat.

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