

1 **ACCEPTED MANUSCRIPT**

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4 AND SKIN COLOR OF GOLDFISH (*Carrasius auratus*)

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18 **THE EFFECTS OF RED YAM FLOUR (*Ipomoea batatas* L.) ON THE GROWTH, SURVIVAL**  
19 **RATE AND SKIN COLOR OF GOLDFISH (*Carrasius auratus*)\*\***

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27  
28 Running title: Effects of red yam flour on goldfish  
29

30 **ABSTRACT**

31 This study evaluated the effects of commercial diets supplemented with 0%, 3%, 6%,  
32 9% and 12% red yam flour (*Ipomoea batatas* L.) on the growth, survival rate and skin colour of  
33 goldfish (*Carrasius auratus*). A completely randomized experimental design was developed with  
34 five treatments and three replicates. Seventy-five goldfish with the average initial length of 4.4 cm  
35 were assigned to fifteen experimental tanks. The experiment had lasted for twenty-eight days. The  
36 results showed that the growth and survival rate were not significantly affected by dietary  
37 treatments ( $P > 0.05$ ). However, red yam flour significantly affected the skin color of goldfish with  
38 the best concentration of 9%. It was concluded that the dietary administration of red yam flour of  
39 9% was found to be a suitable dietary supplementation to ensure skin pigmentation as well as there  
40 were not bad effects on fish growth and survival rate.

41  
42 **Keywords:** *Carrasius auratus*, Red yam flour, skin color.  
43

44 **INTRODUCTION**

45 Goldfish (*Carassius auratus*) is one of the most popular ornamental fish in Indonesia. The  
46 fish has a body color ranging from red, yellow, green, black and silvery. Since it was first  
47 discovered to be nurtured, there are approximately fifteen species of goldfish that have been  
48 recognized and favored by public. Goldfish is also one type of ornamental fish that attract many  
49 world fish markets. In order to produce the best quality goldfish, a good environment and high  
50 nutrients feed are needed. The nutritional content of feed will support color, health and quality of  
51 good fish.

52 For most ornamental fish, many studies focused to achieve high levels of their skin  
53 pigmentation together with body shape, fin shape and body size of the fish using synthetic pigments  
54 and natural sources (Kalinowski et al., 2005; Shahidi et al., 1998). These belong to the most  
55 important quality characteristic informing their market levels (Paripatananont et al. 1999). Fish,  
56 similar to other vertebrate animals, can not perform de novo synthesis of carotenoids (Goodwin,  
57 1984). Therefore, they depend on dietary supplies to obtain their own natural pigmentation. The  
58 color of fish is produced by the presence of chromatophoral cells existing in the dermis layer of skin

59 (Putra et al, 2012). Natural carotenoid substances can be found in plants and fruits in the form of  $\beta$ -  
60 carotene. Since the high cost of synthetic colourings, many studies on natural compounds such as  
61 *Chlorella zofin-giensis* (Bar et al. 1995) *Chlorococcum sp.* (Zhang et al. 1997); the green algae  
62 *Haematococcus pluvialis* (Harker et al. 1996; Yuan & Chen 2000 and *C. vulgaris* (Gouveia et al.  
63 1996) have been done. Other latest works have been done using paprika (Minh et al, 2014);  
64 *Spirulina platensis* (Mahdi et al 2013), *Medicago sativa* (Mahmut et al 2008); and carrot starch  
65 (Pardosi et al 2016) as sources of dietary carotenoids. One of alternative sources of a cheap and  
66 easily available carotenoid source in Indonesia is red yam. Red yam, among other vegetables, is  
67 known to contain the highest source of beta carotene. According to Fatimah (2013), red yam  
68 contains the highest beta-carotene among other sweet potatoes. It contains 9900 mg (32 967 SI) of  
69 beta-carotene per 100 grams. The thicker the colour of red yam is, the higher the content of beta-  
70 carotene will be. However, studies on the effects of dietary red yam flour in fish feed on the growth  
71 and skin colour of goldfish (*Carassius auratus*) have not been done yet.

## 72 73 MATERIAL AND METHOD

### 74 Experimental design

75 The feeding trial was conducted in the Biology Laboratory of Faculty of Marine and  
76 Fisheries of Syiah Kuala University of Banda Aceh, Indonesia. It had been conducted for two  
77 months, from July to August 2017. Seventy five goldfish were purchased from a local market.  
78 Prior to the experiment, the fish had been acclimatized and fed with commercial ornamental fish  
79 for three days. Fish with the average length of 4.4 cm were randomly distributed into fifteen  
80 containers (volume 25 L each).

81 This research was done using an experimental method. The design used was Completely  
82 Randomized Design (RAL) with 5 treatments and 3 replications. The treatments used were:  
83 Treatment A: Control; without adding red yam flour; Treatment B: Red yam flour of 3%; Treatment  
84 C: Red yam flour of 6%; Treatment D: Red yam flour of 9%; Treatment E: Red yam flour of 12%.  
85 Fish maintenance had been lasted for 28 days and they had been fed three times a day at 09:00 am,  
86 12:00 pm and 17:00 pm at each treatment. The amount of feed given was 5% of the average weight  
87 of the fish's body.

88 The odd red yam were purchased from traditional market in Banda Aceh city. The red yam  
89 used in this study was dried first. Then, the red yam was ground using a laboratory grinder until it  
90 become flour, and then mixed the red yam flour with progol as binder in accordance with the  
91 dosage (2-3 g / kg of feed) in one container, stirred until evenly and adding water at a volume of  
92 150 ml / kg, poured the commercial ornamental fish feed into a different dosage of mixture of red  
93 yam flour and dried for 30-60 minutes. If the mixed feed color and smell changed, experimental

94 feed must be re-mixed During this period, the water temperature ranged from 25 ° C-32 ° C with  
95 pH from 6 to 9 and the dissolved oxygen was approximately 5 mg/l.

96

### 97 **Observation of fish color change**

98 The color measurements of the tested fish were adopted from Sitorus et al. (2015). Fish  
99 color were measured using Toca Color Finder (TCF) observed by 5 panelists who do not have  
100 visual impairments (color blindness and farsightedness). Fish color was transforming into a scores  
101 by scaling on color measuring paper. Color observation was done every 7 days for 28 days.

102

### 103 **Calculations and statistical analysis**

104 Absolute weight growth (AWG) = Fish weight at time-initial weight

105 Absolute length (AL) = Fish length at time-initial length

106 Survival rate (SR) (%) = 100 x (final fish number/initial fish number)

107 Specific growth rate (SGR) = (Initial weight-initial weight) x 100/days

108 Results were delivered as means± SE (Standard Error). All data were analyzed by one-way  
109 ANOVA using SPSS 14.0 for windows (Kalinowski et al., 2005). The level of significance was set  
110 at P > 0.05, and Duncan test was used to compare the mean values.

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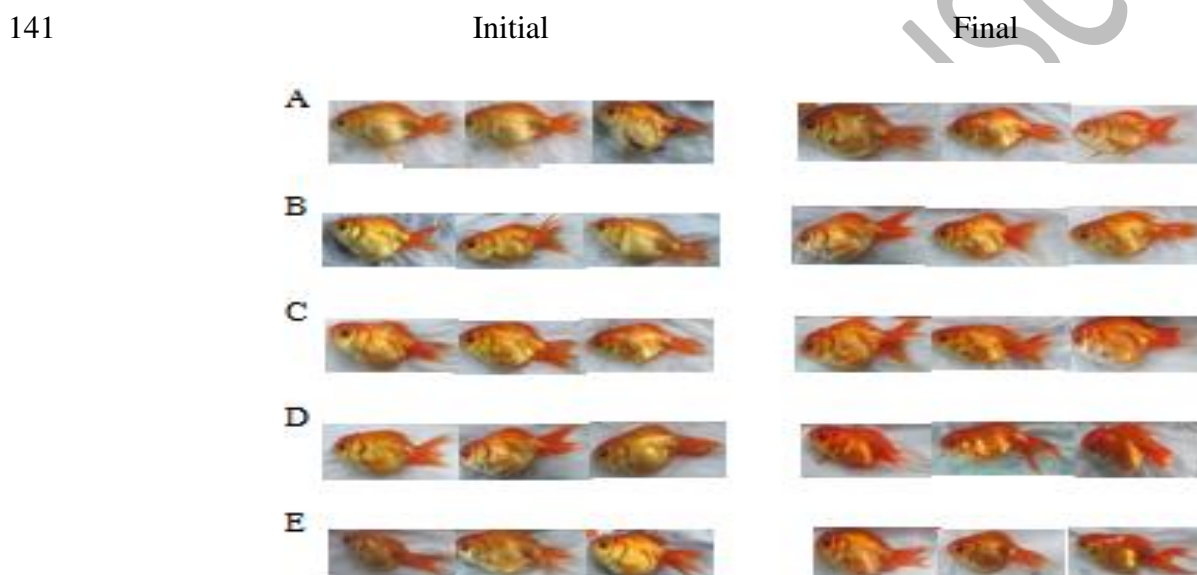
## 112 **RESULTS AND DISCUSSION**

113 The results showed that red yam flour has significant effect on color change of the goldfish  
114 (*Carracius auratus*) (P<0.05) (table 1). Duncan's advanced test showed that the best color change  
115 occurred in treatment D (the dose of 9%) and the lowest shown in treatment A (control) (Figure 1).  
116 According to upstream et al. (2015), the color enhancement in fish varies according to the fish  
117 absorption capacity of the pigment type and the dose administered in each treatment. Based on  
118 these changes, the color of goldfish in each treatment was different and the dose used was different  
119 while the caratenoid material used was similar. In this study it is showed that the addition of 9% red  
120 yam flour was the best and the most effective treatment to improve the color of goldfish. According  
121 to Barus et al. (2014), fish takes a longer period to break the carotene material into color pigments if  
122 the pigment is present in large quantities. Panjaitan et al. (2016) mentioned that carotene added to  
123 the fish feed with excessive doses that to some extent affect the color of fish does not improve the  
124 color of the fish. Moreover, it can reduce the color value of fish.

125 An interesting finding seen on day 14, the color of the goldfish began to be brighter color at  
126 day-21 (Figure 1). It is in accordance with a previous research done by Sitorus et al. (2015), which  
127 stated that the administration of caratenoid in feed for two weeks showed the color improvement of  
128 goldfish. The provision of caratenoid for three weeks resulted in maximum color enhancement.

129 More than 3 weeks, the color will be stable due to the increase of carotenoid in the pigment cells  
 130 (chromatophore) of goldfish.

131 The fluctuation of color intensity is caused by the changes of chromatophore cells. The  
 132 changes are divided into two parts i.e. physiologically and morphologically. Physiological changes  
 133 are the changes caused by chromatophore cell activities that are spread and concentrated in  
 134 epidermis cells. The spread of chromatophore pigment stimulates the pigment to absorb the sun  
 135 perfectly, which then results in an increase of intensity of the fish body. Whereas, the pigment of  
 136 chromatophore that concentrates or gathers near the nucleus can decrease the intensity of fish body  
 137 color so that it will become darker or paler. The movement of pigments in the epidermal layer is  
 138 caused by external stimulation such as temperature, pH and light intensity. The morphological  
 139 changes are due to the amount and composition in the feed containing the carotenoid sources (Sari  
 140 et al., 2012).



142  
 143 Figure 1 Color changes in the goldfish (*Carassius auratus*) (A: Control; Red yam flour of 0%; B: Red  
 144 yam flour of 3%; C: Red yam flour of 6%; D: Red yam flour of 9%; E: Red yam flour of 12%)  
 145

146 The result of ANOVA test showed that feeding of red yam flour has no significant effects  
 147 on absolute weight growth, absolute length, specific growth rate and survival rate ( $P > 0.05$ ) (Table  
 148 1).

149 Table 1 Changes in color, growth, survival and specific growth rate of goldfish for 28 days rearing.

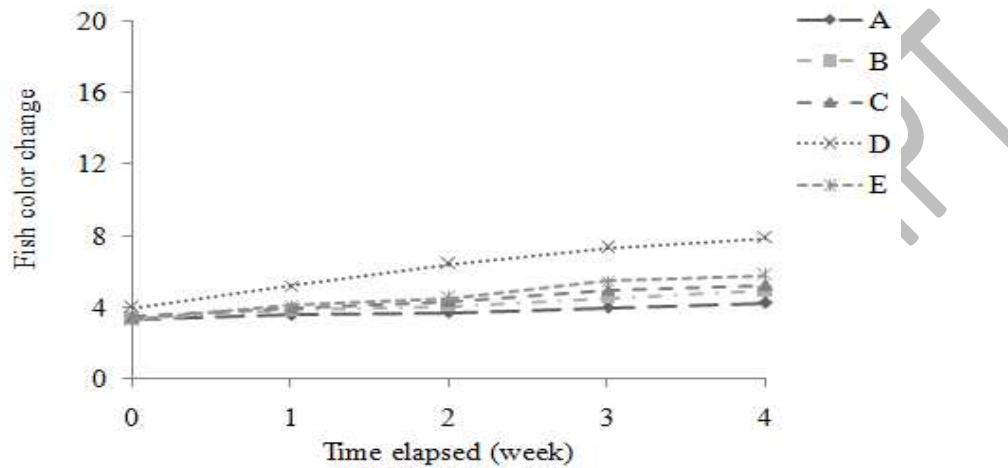
Treatment	Test parameters				
	Color change	Absolute weight growth (gram)	Absolute length (mm)	Specific growth rate (% / day)	Survival rate (%)
A (control)	$0.93 \pm 0.31^a$	$2.44 \pm 0.57^a$	$11.99 \pm 1.60^a$	$2.28 \pm 0.26^a$	$100 \pm 0.00^a$
B (3%)	$1.20 \pm 0.00^{ab}$	$2.90 \pm 0.33^a$	$13.75 \pm 0.75^a$	$2.71 \pm 0.46^a$	$100 \pm 0.00^a$
C (6%)	$1.80 \pm 0.53^b$	$2.35 \pm 0.42^a$	$13.08 \pm 2.76^a$	$1.93 \pm 0.50^a$	$93.33 \pm 11.55^a$

D (9%)	$3.87 \pm 0.31^d$	$2.17 \pm 0.65^a$	$12.46 \pm 3.22^a$	$2.21 \pm 0.53^a$	$100 \pm 0.00^a$
E (12%)	$2.47 \pm 0.31^c$	$2.18 \pm 0.43^a$	$10.48 \pm 2.93^a$	$2.24 \pm 0.22^a$	$100 \pm 0.00^a$

150 Description: Different superscripts in the column show significant differences (P <0.05) between  
 151 treatments based on Duncan Test.

152 The results of the study of color changes of goldfish during the study period of 28 days in  
 153 each treatment can be seen in Figure 2.

154

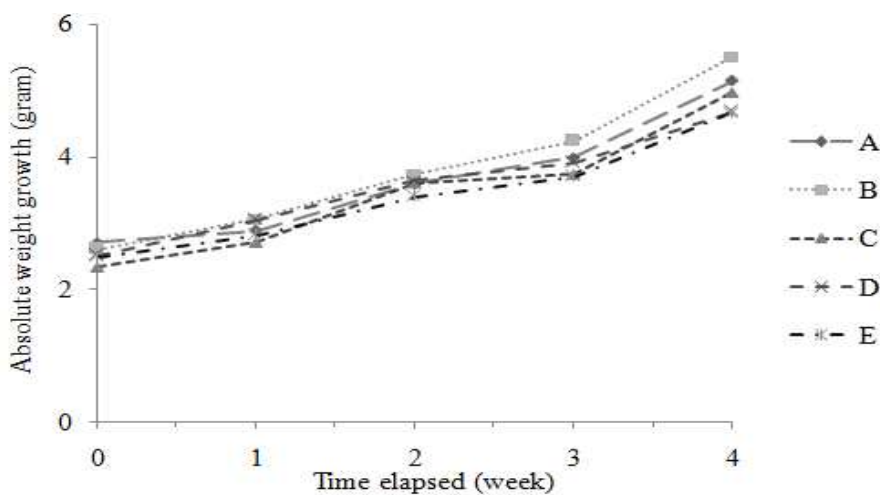


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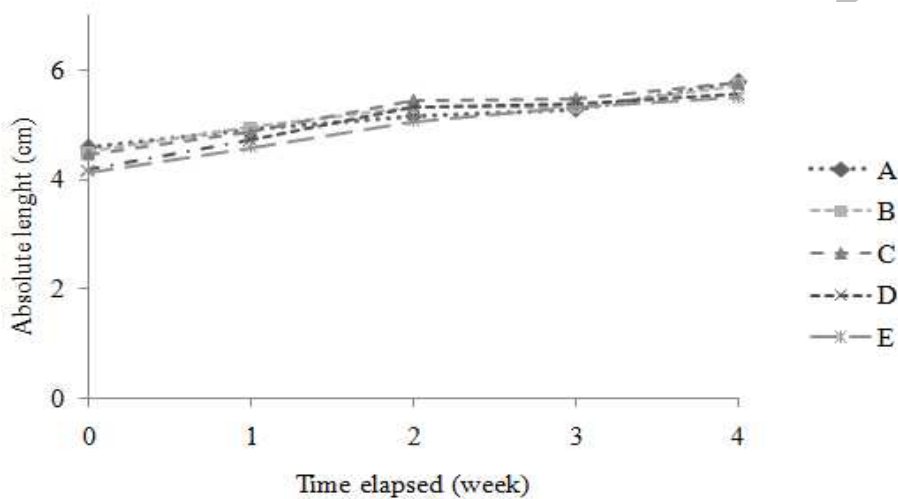
157 Figure 2 Fish color changes of goldfish (A: Control; Red yam flour of 0%; B: Red yam flour of 3%; C: Red  
 158 yam flour of 6%; D: Red yam flour of 9%; E: Red yam flour of 12%)  
 159

160 The growth parameters (absolute weight growth, absolute length growth and specific growth  
 161 rate) of goldfish increase in each treatment; both control feed and artificial feed with the addition of  
 162 red yam flour (Figure 3 and 4) but no significant different (P > 0.05). This indicates that fish are able  
 163 to utilize the feed given for their growth and body maintenance (Putra et al, 2016). Feeding  
 164 increasing the growth of goldfish is considered to meet the nutritional needs of goldfish. It can be  
 165 seen from the nutritional content in the feed and the resulted performance. According to Subamia et  
 166 al. (2013), growth occurs when there are excess energies that once; the available energy was used  
 167 for metabolism, digestion and activities. The result of ANOVA analyses showed that the treatment  
 168 of red yam flour with different doses did not give significant effect to the growth parameters of  
 169 goldfish, namely absolute weight growth, absolute length growth and specific growth rate (P>  
 170 0.05).



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Figure 3 Absolute weight growth of goldfish (A: Control; Red yam flour of 0%; B: Red yam flour of 3%; C: Red yam flour of 6%; D: Red yam flour of 9%; E: Red yam flour of 12%)



176  
177

Figure 4 Absolute length growth of goldfish (A: Control; Red yam flour of 0%; B: Red yam flour of 3%; C: Red yam flour of 6%; D: Red yam flour of 9%; E: Red yam flour of 12%)

181 The survival rates of goldfish during the 28-day study period gave no significant effects in  
182 each treatment. It is allegedly due to the lack of protein content in red yam flour. The survival of  
183 goldfish is also influenced by water quality. According to Noviyanti et al. (2015), the optimal water  
184 temperature for the goldfish is around 25 ° C-32 ° C. The temperatures obtained in maintaining  
185 media ranged between 27 ° C-29 ° C. The optimal degree of acidity (pH) of water for goldfish is  
186 around 6-9. The water quality parameters measured during the study were DO, temperature and pH  
187 within feasibility limits for the maintenance of goldfish. The over tolerance of water quality of fish  
188 may cause fish stress. It is in accordance to Barus et al. (2015), stated that an ornamental fish in a  
189 state of stress can cause its color to fade. It is due to the gathering of pigment granules in the middle  
190 of the cell, so that the fish loses its color.

## CONCLUSION

The administration of red yam flour (*Ipomoea batatas L.*) at different concentration in fish feed has a significant effect on color improvement with the optimal dose in D treatment (dose of 9%). However, the administration does not significantly affect the increases of absolute weight, absolute length, daily growth rate and the survival of the goldfish (*Carassius auratus*). Red yam flour can be considered as a potential feed supplement to enhance the economical value in ornamental fish markets.

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