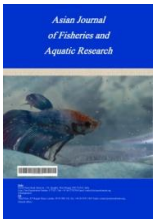

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Promoting Color Brightness of Clown Loach (*Chromobotia macracanthus* Bleeker) by Mixing Red Spinach (*Amaranthus tricolor* L.*)* Powder with Feed Stuff

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Authors' contributions

This work was carried out in collaboration among all authors. Author AY designed the study, performed the statistical analysis, wrote the protocol and wrote the first draft of the manuscript. Authors SIZ and AR managed the analysis of the study. Author YA managed the literature searches. All authors read and approved the final manuscript.

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ABSTRACT

The purpose of this research is to determine the optimal dose of the addition of red spinach powder to artificial feed on the brightness of clown loach. This research was conducted at Aquaculture Laboratory Building 4 Faculty of Fisheries and Marine Sciences of Universitas Padjadjaran from April to May 2020. This research method is experimental with a Completely Randomized Design consisting of four treatments and three replications. The red spinach powder addition treatment used 0%, 2%, 4%, and 6% of the feed amount. The parameters observed are color value as primary data by using Toca Color Finder, while the growth, survival rate, and water quality as support data. Color assessment results were analyzed using the Kruskal-Wallis test, if there were significant differences, Z test would be performed. Growth data and survival rates were analyzed using Analysis of Variance (ANOVA). Because the F test results were not significantly different, the Duncan test was not carried out. The results showed that the addition of 6% red spinach powder is the best treatment with an increased color brightness value of 5,63.

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Keywords: Amaranthus tricolor L.; Chromobotia macracanthus Bleeker; clown loach; red spinach powder; color brightness.

1. INTRODUCTION

Indonesia's ornamental fish industry is among the world's top five suppliers. In 2013 Indonesia's ornamental fish exports reached around 70 million USD or around 764 billion IDR, 20 percent more than the previous year (58 million USD) [1]. The prospect of ornamental aquaculture business is promising to be developed because it has advantages such as the technology is easily absorbed and applied. One of the icons of freshwater ornamental fish from Indonesia is clown loach (*Chromobotia macracanthus* Bleeker) which is often dubbed as fresh Nemo fish. In the international market of ornamental freshwater fish, clown loach is the major species [2]. World demand for clown loach continues from time to time because these fish have unique shapes and colors, but these fish haven't been able to be cultured effectively so that to fulfill world market demand still relies on catches from nature.

Besides physiology and ecology factors, aesthetics in ornamental fish also can increase the commercial value of exports [3]. Color is one of the decisive components in the assessment of ornamental fish. The mechanism of color enhancement towards a brighter direction is influenced by the chromatophore cells located in the epidermal layer. Color changes in fish can occur due to changes in the amount of pigment in the body of the fish. The causes of one color change include environmental stress, such as water quality, sunlight, and pigment content in the feed. Feeds affect the growth and health of fish, which can also affect the color of fish. Feed containing dyes or pigments certain things like carotene will increase the amount of pigment in the body of the fish so that the color will be brighter. Color brightness in fish can be increased by adding carotenoids to the feed which are the main components forming red and orange colors. Much pigmentation has been carried out on commercial fish using carotenoids and obtained significant results [4]. The addition of natural or synthetic carotenoids (artificial) in feed can increase the color quality of ornamental fish [5]. Red spinach is a plant that can be used as a natural coloring in food. The red color in red spinach is a betacyanin pigment that can be used as a natural coloring as well as being an antioxidant. Based on this, research about the addition of red spinach powder into artificial feed with different doses for the color brightness of clown loach is needed.

2. MATERIALS AND METHODS

The equipment used consists of 12 aquariums with sizes 40 cm length x 25 cm width x 27 cm height, water pumps, pump hoses, filters, plastic hoses, fiber tubs, pH meter, DO meter, thermometer, digital scales, Toca color finder (TCF), tray, small bowl, scoop, millimeter block, spoon, and zip-lock plastic. The material used consists of 60 clown loach sized 5-5,6 cm from Bekasi, West Java, self-made red spinach powder, carboxymethyl cellulose (CMC), and sinking feed with Mutiara brand containing 48% protein.

2.1 Research Methodology

This research was conducted at Aquaculture Laboratory Building 4 Faculty of Fisheries and Marine Sciences of Universitas Padjadjaran from April to May 2020. This research method is experimental with a Completely Randomized Design (CRD) consisting of four treatments and three replications. The percentage of the addition of red spinach powder used is as follows:

1. Treatment A: without the addition of red spinach powder (control)
2. Treatment B: addition of 2% red spinach powder per kg of feed (20 g/kg)
3. Treatment C: addition of 4% red spinach powder per kg of feed (40 g/kg)
4. Treatment D: addition of 6% red spinach powder per kg of feed (60 g/kg)

2.2 Experimental Set-Up and Fish Acclimatization

2.2.1 Preparation of the container and acclimatization

The container used in this research is an aquarium with a size of 40 x 25 x 27 cm as many as 12 units. Fiber tub and aquarium soaked using chlorine solvent for one day. Then the aquarium is rinsed and dried. Then the aquarium is filled with clean water with a water level of 10 cm (10 liters) in each aquarium. After the aquarium is filled with water, fixing a water pump, pump hose, water filter, and heater in each aquarium. Acclimatization is carried out for seven

days, this process is intended so that the test fish can adapt to the research conditions. In the acclimatization process, all the test fish were placed in a fiber tub and given commercial feed. Feeding is given twice at 08.00 a.m. and 04.00 p.m.

2.2.2 Feed preparation

2.2.2.1 Making red spinach powder

Making red spinach powder is as follows:

a) First of all, the red spinach is washed, then the leaves are separated.
b) The red spinach leaves that have been sorted are drained, then put in a baking sheet and put in the oven for 20 minutes.
c) The dry red spinach is mashed using a blender until it becomes smooth granular, then sifted using a flour sifter.

From 350 g of fresh red spinach leaves, 42.24 g of red spinach powder (8.3%) are produced.

2.2.2.2 Preparation of experimental feed

Making the test feed is done every three days. The procedure for making the test feed is by mixing the red spinach powder according to the treatment and CMC as much as 5% of the feed in the container. Then add 1 ml of water and stir until homogeneous. Then the commercial feed is put into the mixture and stirred again until evenly distributed. After that, it is continued with the drying process for 20 minutes by aerating.

2.2.3 Fish feeding

The test fish were stocked into the aquarium with a density of 1 fish / 2 liters. Then the fish were fasted for one day to equalize the physiological status of the fish. This research was conducted for 40 days with color assessments, weight measurements, and water quality measurements every ten days in the morning [6]. The frequency of feeding was carried out twice a day at 08.00 a.m. and 04.00 p.m., as much as 3% of the fish biomass per day [7].

2.2.4 Observations

2.2.4.1 Color grading

The parameters observed were the increase in the color value of the clown loach tail. The measuring tool used is the Toca color finder (TCF), which is a tool to identify color specifications. The color specification used is faded orange (TCF code 0614) to dark orange (TCF code 0916). The assessment starts from the smallest score of 1 to the largest score of 7 with a gradient of faded orange to dark orange.

The color measurement of the test fish was observed by three panelists who had no visual disturbances (myopic and color blind) and had been given prior training. Observations were made visually by comparing the original color of the fish in the Toca Color Finder (Table 1).

2.2.4.2 Survival rate

The survival rate observations were observed every day during the research period. The survival rate is calculated using a formula [8]:

$$SR = \frac{\text{Number of Fish at the End of the Experiment}}{\text{Number of Fish at the Beginning of the Experiment}} \text{ x } 100\%$$

2.2.4.3 Absolute weight rate

Absolute weight growth is calculated using the calculation formula [9]:

$$Wm = W_t - W_0$$

Information:
Wm : Growth in absolute weight (g)
Wt : Weight of biomass at the end of research (g)
Wo : Weight of biomass at the beginning of research (g)

2.2.4.4 Absolute length rate

The absolute length growth is the difference between the length of the fish between the head and tail at the end of the study and the length of the body at the start of the study. Absolute length growth is calculated using a formula [9]:

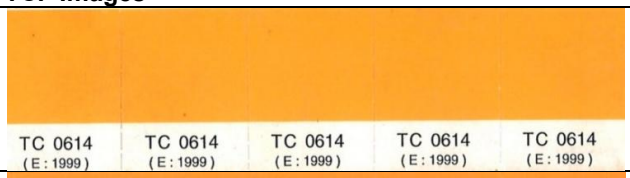
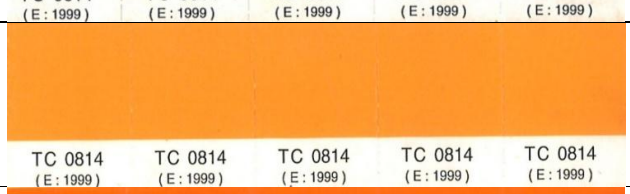
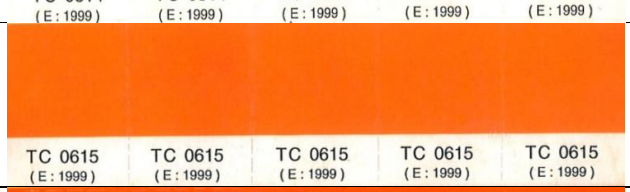
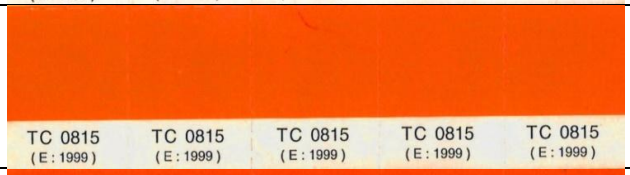
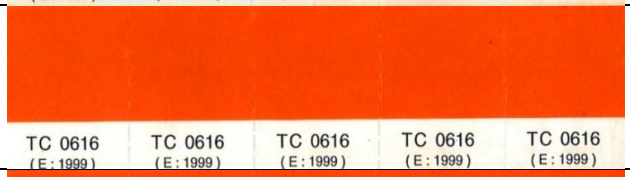
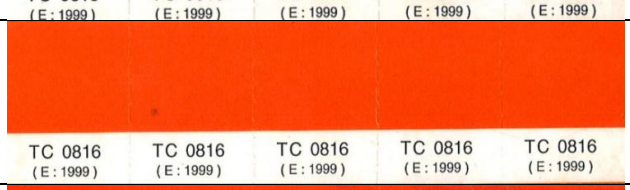
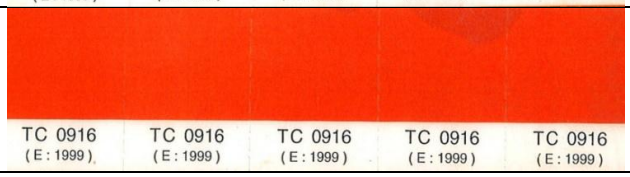
$$Lm = L_t - L_0$$

Information:
Lm : Growth in absolute length (cm)
Lt : Average length at the end (cm)
Lo : Average length at the beginning (cm)

2.2.4.5 Water quality parameters

Water quality measurements are carried out every ten days which include temperature, pH, and DO (Dissolved Oxygen).

Table 1. The TCF color code to be used

Number	TCF Images	Information
1		Score 1 TC code 0614
2		Score 2 TC code 0814
3		Score 3 TC code 0615
4		Score 4 TC code 0815
5		Score 5 TC code 0616
6		Score 6 TC code 0816
7		Score 7 TC code 0916

2.2.5 Data analysis

Data from the color observation of fishtails were analyzed using the Kruskal - Wallis test. Data from growth observations (weight and length) were analyzed using the F test with a 95% confidence level to determine the effect of treatment on parameters. If the treatment has a significant effect (F count> F table) then proceed with Duncan's multiple range test. The data from the observation of survival rate and water quality were analyzed descriptively and comparatively.

3. RESULTS AND DISCUSSION

3.1 Color Brightness Level

The addition of red spinach powder to the feed with different concentrations gave an increase in the color value of clown loach. Assessment of color enhancement in clown loach was observed in the tail. Clown loach color changes were identified using the Toca Color Finder (TCF) tool. Based on the results of research that has been carried out for 40 days, it is found that the addition of red spinach powder increases the

color score in clown loach as in the following graph.

Observations on the 10th day showed that clown loach began to experience an increase in color values in treatment B (addition of 2% red spinach powder), C (addition of 4% red spinach powder), and D (addition of 6% red spinach powder). In the control treatment (without the addition of red spinach powder) there was no visible increase in the color value, this was because the fish were not given feed containing carotenoids so that the fish chromatophore cells would not spread to all over the skin and causing the fish to turn pale. This can be seen in the following Table 2.

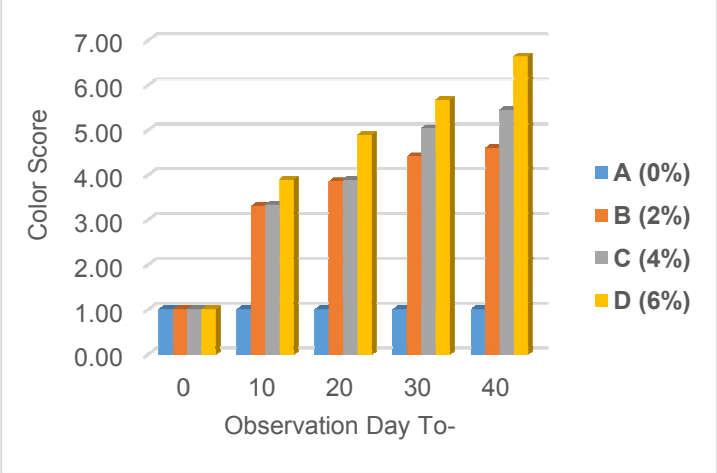


Fig. 1. Graph of clown loach color enhancement

Table 2. Image comparison of the color of clown loach tails per 10 days during the research

Treatment	Days to-				
	0	10	20	30	40
A (0%)					
B (2%)					
C (4%)					
D (6%)					

On the 40th day, there was an increase in orange color in each treatment which was added with red spinach powder (Table 2). The highest color score was in treatment D (addition of 6% red spinach powder) with an average score of 6,63 and the lowest score in treatment A (Control) with an average score of 1. The color value of clown loach in treatment B (addition of 2% red spinach powder), C (addition of 4% red spinach powder), and D (addition of 6% red spinach powder) continued to increase until the 40th day. This increase in color value is thought to be because clown loach still needs carotenoid content in their feed to be synthesized from faded orange to dark orange, and fish metabolism and absorption work optimally because the concentration given is corresponding with the fish's ability to synthesize carotenoids in red spinach powder. The dietary supply of carotenoids not only improves skin tone but also improves the economic value of ornamental fish [10].

Based on the Kruskal-Wallis test results, it shows that there is a significant difference between treatment A (control) and treatment B (2%), C (4%), and D (6%). During the research period, there was an increase in the color value of treatment B (2%) by 3,59, treatment C (4%) by 4,44, treatment D (6%) by 5,63, and treatment A (control) did not increase. The results presented in Table 3 show that the highest increase in the color value of clown loach was 5,63 in treatment D with the addition of 6% red spinach powder and the lowest color value of clown loach in treatment A without the addition of red spinach powder. The results showed that the addition of 6% red spinach powder resulted in the highest increase in the color value of clown loach. This is different from the research of adding 4% addition of red spinach flour treatment resulted in an increase in the color value of the best goldfish and at 5% treatment the color value of goldfish decreased [11]. The difference is presumably because the species used are different so that the results obtained are different.

The treatment without the addition of red spinach powder did not increase the color value. This is because the fish cannot synthesize their coloring pigments without external additions, so they must be added to the feed [10]. In treatment B (2%) was not significantly different from treatment C (4%), this is presumably because the absorption rate of the orange pigment is influenced by the amount of carotenoid material present in the feed and the surface area of the fish body against the orange color. The color of clown loach for all treatments experienced fluctuating changes, this was due to the provision of carotene sources with different doses in each treatment, the response to different feeds, and differences in the absorption of clown loach to the carotene in the feed [12]. The greater the dose of the addition of red spinach powder, the change in color value in the clown loach will increase. The main source of skin pigmentation in fish is carotenoids. Fish color quality is brilliant because pigments play a role in the broad spectrum of colors in the fish body [7]. The appearance of color in fish is influenced by the content and ability or absorption of the fish to a given pigment source. It is suspected that fish need a longer time to break down carotenoids into color pigments if the number of carotenes in the feed is greater or the higher the carotenoid content in the feed, the more carotene is absorbed.

The increase in fish color values in treatment B (2%), C (4%), and D (6%) occurred because the fish absorbed the carotene contained in the feed well. Carotene, which is a source of pigment, will be absorbed by fish and then partially stored in the liver as a precursor for vitamin A and the rest will be channeled into fat tissue for color needs. Then the carotene is deposited in color cells (chromatophores) in the dermis [13]. Based on the results of this research, it can be said that adding red spinach powder to the feed as a source of carotene can increase the color of the clown loach, with the addition of 6% red spinach powder as a treatment to increase the color value of the best clown loach.

Table 3. Average clown loach tail color value

Treatment	Increased color brightness value
A (Control)	0^{a}±0
B (Addition of 2% red spinach powder)	$3,59^{b}$±0,93
C (Addition of 4% red spinach powder)	$4,44^{b}$±0,70
D (Addition of 6% red spinach powder)	$5,63^{c}$±0,56

Note: The number followed by the same letter notation means there is no significant difference with a 95% confidence level

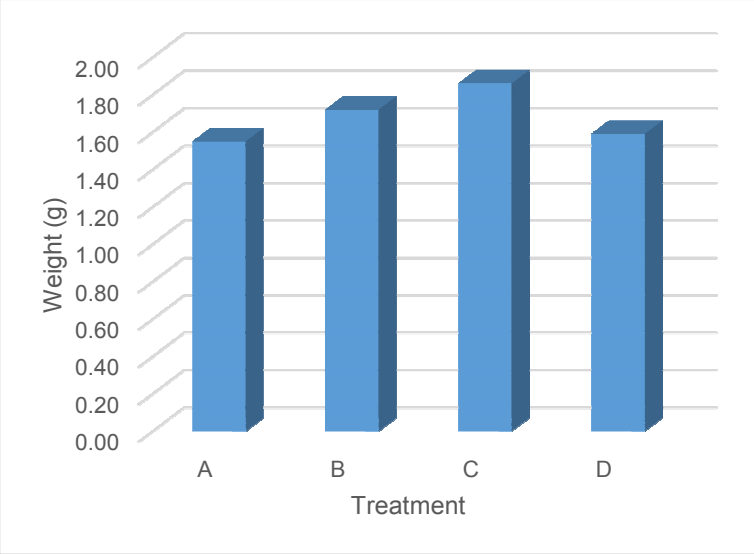


Fig. 2. Clown loach absolute weight growth graph

3.2 Growth

The parameters observed in this research are the increase in weight and absolute length. The growth of absolute weight and length were the supporting parameters observed to determine the effect of adding red spinach powder to artificial feed on the growth of clown loach.

3.2.1 Absolute weight growth

Clown loach weight measurements were carried out once every 10 days during the research period. The absolute weight gain of each treatment can be seen in the following graph.

During the research, clown loach experienced weight gain in each treatment. Based on the graph, it can be seen that the highest weight gain is in treatment C (addition of 4% red spinach powder) with a weight gain value of 1,86 g and the lowest weight gain is treatment A (control) with a weight gain value of 1,55 g. Clown loach growth is relatively slow [14]. This is presumably because clown loach is ornamental fish so that the increase in body weight does not increase significantly. The addition of red spinach powder to artificial feed did not affect the growth of clown loach. The results of the analysis of variance (Test F) showed that the absolute weight gain in the treatment with or without the addition of red spinach powder was not significantly different. Carotenoids given to goldfish do not affect growth [15]. The dietary inclusion of carotenoid pigments did not affect growth [16].

3.2.2 Absolute length growth

Clown loach length measurements were carried out twice, namely at the beginning and at the end of the research. The average increase in the absolute length of each treatment can be seen in the following graph.

Based on the graph above, it can be seen that the highest length addition is in treatment B (addition of 2% red spinach powder) and treatment D (addition of 6% red spinach powder) of 0,30 cm, while the lowest is in treatment A (control) of 0,21 cm. The clown loach growth rate is relatively slow, an increase in the length of the juvenile clown loach by 1 inch (2,5 cm) requires a maintenance time of 105 days [17]. Also with the breeding that has been carried out by the Ornamental Fish Research and Development Center, Depok, raising clown loach from larvae until they reach a marketable size of 2-2,5 inches requires a maintenance time of 6-8 months [18]. Based on the ANOVA (Analysis of Variance) test, it was found that the calculated f value was smaller than the f table, which means that there was no significant difference between treatments, this indicated that the addition of red spinach powder had no significant effect on the length increase of clown loach. Fish growth is influenced by internal factors and external factors. Internal factors include genetics, age, resistance to disease, and the ability to digest food. Meanwhile, external factors include the physical and chemical properties of the environment and the nutritional value of food

available in the feed. The main factors that can affect fish growth are food factors and environmental factors. Food factors such as nutrition, including the quality and quantity of food. Environmental factors such as temperature, dissolved oxygen concentration, salinity, and photoperiod [19]. The addition of carotenoids to the feed has no effect on growth and ornamental fish fed with the addition of carotene sources are thought to make more use of this dye to increase their body color.

3.3 Survival Rate

The survival rate of clown loach for 40 days of research shows the same results in each treatment, which can be seen in the following table .

Based on the Table 4, it can be seen that all treatments are not significantly different. The survival rate obtained is 100% in each treatment, the results of this survival rate are very good when compared with research on the addition of pumpkin flour to the feed for clown loach which results in a survival rate between 88.89-96,3% [20]. In this research, it appears that adding red spinach powder to fish feed does not affect mortality. This is presumably because the carotene content in red spinach powder, apart from being a source of color pigment, also does not endanger fish health. Apart from functioning as a source of color pigments, carotene has a role in protecting fish against light and can assist in the metabolism of the oxygen cycle. Carotene also plays other important functions as a basic ingredient of vitamin A, antioxidants, and immunoregulators [7].

3.4 Water Quality

The results of water quality measurements for each parameter are presented in Table 5 below.

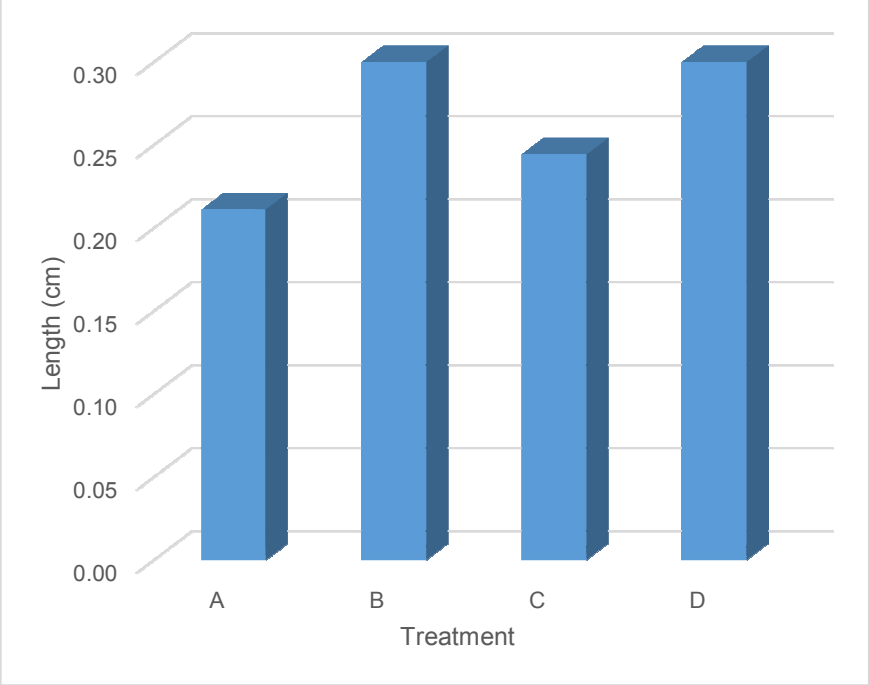


Fig. 3. Clown loach absolute length growth graph

Table 4. Result of observation of clown loach survival rate

Treatment	Survival rate (SR)
A (Control)	100%
B (Addition of 2% red spinach powder)	100%
C (Addition of 4% red spinach powder)	100%
D (Addition of 6% red spinach powder)	100%

Table 5. Results of observation of clown loach water quality

Treatment	Temperature (^{0}C)	pH	DO (ppm)
A	26,23	7,60	5,21
B	26,31	7,61	5,28
C	26	7,57	5,20
D	26,38	7,64	5,26
References	22 – 28 (Research Workshop on Freshwater Ornamental Fish Cultivation 2008)	6,5 – 7,0 (Research Workshop on Freshwater Ornamental Fish Cultivation 2008)	>5 (Maulidiyanti et al. 2015)

Based on the Research Workshop for Freshwater Ornamental Fish Cultivation, Depok, the temperature suitable for the life habits of clown loach is 22 - 28 ^{0}C. During the research, the water temperature was in the range 26 - 26.38 ^{0}C, meaning that the temperature in the research media was almost the same as this reference and supported the life of clown loach during the research. Based on the Research Workshop for Freshwater Ornamental Fish Cultivation, Depok, the appropriate pH for clown loach life is slightly acidic to neutral, namely the range of 6.5-7.0. During the research, the pH was still in the normal range, namely 7.57-7.64 so that clown loach could survive. Optimal DO values for fish can reduce the proportion of metabolic energy and energy loss, thereby increasing the proportion is allocated to growth [21]. For optimal fish growth, oxygen content must be maintained above 5 ppm. During the research, dissolved oxygen levels were in a good range of dissolved oxygen concentrations between 5.20-5.28 ppm, so that clown loach had a 100% survival.

4. CONCLUSION

Based on the results it can be concluded that the addition of 6% red spinach powder to the feed for 40 days was able to increase the best color value of clown loach with an increase in the color value of 5,63. The addition of red spinach powder to feed did not affect the growth of the clown loach.

CONSENT

All authors declare that 'written informed consent was obtained from other approved parties for publication of this case report and accompanying images.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

REFERENCES

1. Meyliana, Widjaja HAE. E-Commerce implementation to support ornamental fish breeders in Indonesia. International Conference on Information Technology Systems and Innovation (ICITSI); 2015.
2. Legendre M, Satyani D, Subandiyah S, Sudarto L, Pouyaud E, Baras J. Slembrouck. Biology and culture of the clown loach *Chromobotia macracanthus* (Cypriniformes, Cobitidae): 1-hormonal induced breeding, unusual latency response and egg Production in Two Populations from Sumatra and Borneo Islands. Aquat. Living Resour. 2012;25:95-108.
3. Arulvasu C, Ramya. Meena S, Chandirasekar D, Sivaganam S. Evaluation of Natural sources of carotenoid pigments from Rosa Rubiginosa on Growth, Survival and Coloration of *Xiphophorus Helleri* Fish Fry. European Journal of Biological Sciences. 2013;5(2):44-49.
4. Das AP, S. Prasad B. Carotenoids and Pigmentation in Ornamental Fish. Journal of Aquaculture and Marine Biology. 2016;4(4):1-3.
5. Sujath BJS, Shalin JJ, Palavesam A. Influence of four ornamental flowers on the growth and colouration of orange sword-tail Chiclidae Fish (*Xiphophorus helleri*, Heckel, 1940). International Journal of Biology Medicine Resource, 2011;2(3): 621–626.
6. Solichin I, Kiki H, dan Henhen S. The effect of addition of rebon flour to artificial feed on the chroma value of chef goldfish (*Carassius auratus*). Faculty of Fisheries and Marine Science. Universitas Padjadjaran. Journal of Fisheries and Marine. 2012;3(4):185-190.
7. Wagde MS, Subodh K. Sharma, Bhanu K. Sharma, Amrita P. Shivani, Naresh R.

Keer. Effect of Natural β-carotene From Carrot (*Daucus carota*) and Spinach (*Spinacia oleracea*) on Colouration of an Ornamental Fish-Swordtail (*Xiphophorus helleri*). Journal of Entomology and Zoology Studies. 2018;6(6):699-705.
8. Opasola OA, Adewoye SO, Fawole OO. Growth performance and survival rate of *Clarias gariepinus* fed *Lactobacillus acidophilus* supplemented diets. IOSR Journal of Agriculture and Veterinary Science (IOSR-JAVS). 2013:3(6):45-50.
9. Effendie MI. Fisheries biology. Nusatama Library Foundation. Yogyakarta; 1997.
10. Gupta SK, Jha AK, Pal AK, Venkateshwarlu G. Use of natural carotenoids for pigmentation in fishes. Natural Product Radiance, 2007;6(1):46-49.
11. Saputra R, Mulyadi M. Phil, Rusliadi. The influence of additional red spinach flour (*Amaranthus hypocondriacus*) in made feed of color quality to goldfish. Journal. Faculty of Fisheries and Marine. Riau University; 2017.
12. Jannah RR, Raharjo EI, Rachimi. Effect of addition of marigold flower flour (*Tagetas erecta*) in feed on the color quality of clown loach (*Chromobotia macracanthus*) seeds. Journal. Muhammadiyah University Pontianak; 2016.
13. Goodwin T. The biochemistry of carotenoids. Animals. Science Edition. 1984;II.
14. Permana A, Alimuddin W. Hadie, Priyadi A. Growth response of clown loach (*Chromobotia macracanthus* Bleeker 1852) juveniles immersed in water containing recombinant growth hormone. Indonesian Aquaculture Journal. 2015;10(2):125-130.
15. Alma AC, Juan E. Pablo, G. Adrian, Maurilio. The effect of marigold (*Tagetas erecta*) as natural carotenoid source for the pigmentation of goldfish (*Carrasius auratus* L.). Research Journal of Fisheries and Hydrobiology, 2013;8(2):31-37.
16. Besen KP, Melim EWH, da Cunha L, Favaretto ED, Moreira M, Fabregat TEHP. Lutein as a natural carotenoid source: Effect on growth, survival and skin pigmentation of goldfish juveniles (*Carassius auratus*). Aquaculture Research, 2019;(50):2200-2206.
17. Satyani D, Meilisza N, Solichah L. Description of juvenile length growth of cultured clown Loach in hapa system with a stocking density of 5 Fish per Liter. Proceedings of the Aquaculture Technology Innovation Forum. 2010;395-402.
18. Aras KA, Nirmala K, Soelistyowati Sudarto DT. Light spectrum manipulation on growth and color quality of clown loach yuwana (*Chromobotia macracanthus* Bleeker 1852). Indonesian Journal of Iktiology. 2015;16(1):45-55.
19. Dutta H. Growth in fishes. Department of Zoology. N. C. College Jaipur, 2009:97-112.
20. Azuri R. The effect of adding pumpkin (*Cucurbita* sp) flour to the feed with different doses on the brightness of the color of clown loach (*Chromobotia macracanthus*). Journal. Faculty of Fisheries and Marine Science. Riau University; 2018.
21. Nariswari S, Yustiati A, Rostini I, Suryadi IBB. Effect of stocking density on water quality of tilapia (*Oreochromis niloticus* Linnaeus, 1758) in round container with current and venturi Aeration System. World Scientfic News. 2020;145:397-408.

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