



The effect of Red Paprika Extract (*Capsicum Annuum*) in Feed on the Color Intensity Level of Comet Fish (*Carassius auratus*)

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Authors' contributions

This work was carried out in collaboration among all authors. All authors read and approved the final manuscript.

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ABSTRACT

This study aims to analyze the effect of red paprika flour in fish feed to increase the color intensity of the Comet fish juvenile. The research method used a Complete Randomized Design experiment consist of four treatments and three replicates. The treatments consisted of various levels of addition red pepper extract, namely treatment A (0%), B (3%), C (5%) and D (7%). The investigation includes color intensity of the fish. Survival rate and water quality parameters data were also taken. The color observation data were analyzed using the Kruskal-Wallis analysis, if there was a significant difference, the Z test was performed. The results of this study indicated that the addition of 5% red paprika extract to the feed was the best treatment with an increase in the color intensity level of comet fish at 4.71. While the comet fish Survival Rate (SR) data in all treatments were observed to be 100% and the water quality during the maintenance period was in the optimal range and met the standards. The average temperature value was 24.7-26.4°C, DO 5.6-5.8 mg/L and pH 6.67-7.53.

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1. INTRODUCTION

Indonesia is a country with a tropical climate and has enormous potential for fish resources. Indonesia has the potential annual ornamental fish income of 300 million per year, including 240 species of marine ornamental fish and 226 species of freshwater fish [1]. Several species of freshwater fish have been successfully cultivated, one of which is the comet fish (*Carassius auratus*). The comet fish (*Carassius auratus*) is one of the freshwater ornamental fish commodities that are much favored by ornamental fish hobbyists because of their color patterns and attractive body shapes, and can be kept in ponds or in aquariums [2]. As a fish that is included in the category of ornamental fish to get the best color, comet fish requires an environment and feed with good nutritional completeness. The addition of pigment source nutrients in fish feed can increase the concentration and distribution of color chromatophores in the skin tissue, thereby increasing the color intensity level [3]. One of the efforts to increase the color intensity level is the addition of carotenoids as components in the formation of red and yellow colors on fish [4].

Carotenoids are one pigment responsible for many types of color found in nature, including diversity and function. Various types of natural fat-soluble pigments are found in animals, plants, algae, fungi, photosynthetic bacteria and some non-photosynthetic bacteria. Carotenoids that are generally found in fish color are lutein (yellow-green), carotene (orange), doradexanthine (yellow), zeaxanthine (yellow-orange), cantaxanthine (orange-red), astaxanthine (red), echinenone (red) and taraxanthin (yellow) [5].

Paprika oleoresin is a product derived from red pepper (*Capsicum annuum* L.) [6]. It is an oil with high amounts of carotenoids, commonly used in the food industry as a colourant for sauces, soups, or meat-based meals. It is also used in cosmetic products such as liquid emulsions and creams [7]. The colouration capacity of paprika oleoresin is due to its high content of carotenoids, which represent the pigment pattern of red pepper. This includes seven carotenoids, of which capsanthin, epoxycapsanthin and capsorubin are exclusively synthesised in red pepper [8,9]. In addition, β -carotene and β -cryptoxanthin, both provitamin A carotenoids,

and the dihydroxylated xanthophylls zeaxanthin, cucurbitaxanthin A and violaxanthin are present. In this case, not only carotenoids with provitamin A activity are available, but also carotenoids with particular structural features that might exhibit biological properties, including antioxidant and non-antioxidant activities [10,11]. The oxocarotenoids capsanthin and capsorubin exhibit greater antioxidant capacity than other xanthophylls [12].

More than 25 different pigments have been identified in the fruits of paprika: green chlorophylls, yellow-orange lutein, zeaxanthin, violaxanthin, antheraxanthin, β -cryptoxanthin, β -carotene, etc. The red pigments capsanthin, capsorubin and cryptoxanthin are unique to the *Capsicum* species. The colour of paprika is determined by the proportion of red to yellow pigments [13,14], whereas β -, α -, γ -carotene and β -cryptoxanthin, as provitamins, contribute to its nutritive value. Capsanthin and capsorubin, which comprise 65–80%, contribute the red colour to paprika [15,16]. Capsanthin and capsorubin have a better radical scavenging ability than astaxanthin. Consequently, it is an important that carotenoids quench these free radical to produce harmless end products [17]. Peppers are a rich source of vitamin A and C, phenolic compounds and micro and macro elements [18].

Based on the description above, it is necessary to do research on the addition of red pepper extract (*Capsicum annuum*) in effective feed to improve the color quality of comet fish.

2. METHODOLOGY

2.1 Time and Place of Research

This research was conducted from March 2020 to May 2020 at the Aquaculture Laboratory of Building 4 Faculty of Fisheries and Marine Sciences Padjadjaran University, Indonesia.

2.2 Materials and Methods

The equipment used in this study were 12 aquariums with sizes (30 x 18 x 20) cm³ with a density of 10 fish per aquarium, aeration equipment, bowl, digital scales with precision 0,1 g, Toca Colour Finder (TCF), thermometer, DO meter, dan pH meter. 120 juvenile comet fish (*Carassius auratus*) from Bandung, West

Java, Indonesia with a sized range of 3-5 cm, were used. The commercial red paprika oleoresin with the Pelangi brand as paprika extract as a feed ingredient for additional carotene sources, CMC (*Carboxymethyl Cellulose*), and the commercial feed as the main feed used for the mixture of red pepper extract. It was given twice a day as much as 3% of the fish weight. The method used in this research was an experimental method Complete Randomized Design (CRD) consisting of 4 treatments and 3 replications. The treatments given in the experiment were as follow: (A) Comercial feed without red paprika extract (control), (B) Comercial feed with the addition of 3% red paprika extract, (C) Comercial feed with the addition of 5% red paprika extract, and (D) Comercial feed with the addition of 7% red paprika extract.

This research was conducted for 35 days and observations of changes in color intensity were carried out every 7 days. Primary data measured in this study were changes in body color intensity of comet fish performed visually using the standard values obtained from the Toca Color Finder (TCF). The assessment starts from the smallest value of 1 to the largest score of 5 with a color gradation from faded yellow (TCF code 0405) to orange (TCF code 1015). Observation of survival rate and water quality as supporting data.

Observation of the effect of adding paprika oleoresin dosage on color intensity was carried out by focusing on two colors that were close to the body color of the fish. The measurement of fish color was carried out by five panelists who had an understanding of the color of ornamental fish and had no visual problems (color blindness and farsightedness) and had conducted training in advance. Observations were made visually by comparing the color of the fish with the color panel on the Toca Color Finder (TCF). The results of the data were then analyzed using the Kruskal-Wallis test. If there were significant differences, then Test Z was conducted. The data from the observation of water quality were analyzed in a descriptively comparative manner.

3. RESULTS AND DISCUSSION

3.1 Color Intensity Level

Observation of the increase in the color of the comet fish on the body of the fish every seven days for 35 days using the Toca Color Finder. The results showed that the addition of

carotenoids contained in the oleoresin of red peppers could affect the brightness of the comet fish. This was reinforced by the increase in color at the end of the study in the treatment with the addition of red paprika oleoresin during the observation period. The following is a graph of the color enhancement of various treatments in comet fish for 35 days with 4 treatments presented in Fig. 1.

Fig. 1 shows the increase in the value of the fish color score which indicates the effect of treatment. Based on the data in the graph (Fig. 1), on the 7th day it can be seen that there is an increase in the color score values of different treatments B, C and D from time to time. In treatment A (control) or without the addition of paprika extract showed the lowest increase in color value. This is because the feed given does not contain carotenoids. Ornamental fish feed was influenced by the amount and composition feed containing carotenoid sources [4]. The fish's body is unable to synthesize carotenoids without any addition from the outside, so the chromatophores cells not spread over the entire surface of the fish skin, causing the fish's skin to become pale [19].

Observations on the results of the study for 35 days showed that there was an increase in the color of the comet fish juvenile. The best result was treatment C with the addition of 5% red paprika extract. The addition of paprika extract in the feed began to take effect on the 7th day with a score of 2.31. On the 14th day it increased by a score of 2.87. On the 21st day it increased to a score of 3.44. On the 28th day it increased with a score of 4.16 and continued to increase until the 35th day with a score of 4.71.

The paprika extract in treatment B (3%) also experienced an increase in color on the 7th day with a score of 1.62. On the 14th day it increased to a score of 1.93. On the 21st day it increased with a score of 2.29. On day 28 it increased with a score of 2.73 and up to day 35 increased by a score of 3.07, but the color change was relatively not high due to the different doses of oleoresin peppers in treatment B.

In treatment D (7%) the addition of paprika extract was able to increase the intensity level of fish color on day 7 with a score of 2.49 to day 14 with a score of 3.24. However, the intensity decreased on the 21st day with a score of 2.93, then it could increase again until the 28th day

with a score of 3.40 and continued to increase until the 35th day with a score of 4.20.

Feed treatment with the addition of carotenoids derived from paprika oleoresin changed better color intensity. In the results of this study, both at the addition level of 3%, 5% and 7% there was an increase in color intensity compared to 0%, and increased with the level of red pepper oleoresin giving. Carotenoids are hydrophobic compounds that are not easily solubilised in the aqueous environment of the gastrointestinal tract of fish; therefore, digestion, absorption and transport processes are associated to lipids [20]. The intestinal absorption of carotenoids involves several steps, including disruption of the food matrix, dispersion in lipid emulsions and solubilisation into mixed bile salt micelles, before being carried to the enterocyte brush border where the absorption takes place [21,22]. Therefore, the hydrophobic carotenoids can be digested and absorbed properly by fish.

The color values in treatments B, C and D experienced an increase in color until day 35, it was because the use of paprika oleoresin as a source of color additives in the form of artificial carotenoids mixed in commercial feed can improve the color quality of comet fish juvenile [23]. This is because the comet fish seeds require carotenoid materials found in the feed to be synthesized into orange color. Furthermore, when the proper dose is combined with the ability of the fish to synthesize carotenoids from additional feed, comet fish absorption and metabolism are optimized. Carotenoids in red pepper extract range from 800-1500 mg/kg [24].

In paprika, capsanthin exists in a form where it is acylated with fatty acids, and as a result, it contains oxygen and is more polar than β -carotene; the different structures of capsanthin may exert different functional effects in the human body [25]. Among the different varieties, red paprika as a ripe fruit is preferred by consumers, and capsanthin, one of the xanthophylls, is the major carotenoid present in red paprika [26]. Capsanthin, one of the xanthophylls, has eleven conjugated double bonds ending in one or two polar ketones and efficiently absorbs green light to give a red-orange hue [27,28]. Carotenoids such as xanthophylls are lipidsoluble molecules that follow the absorption pathway of dietary fats [29,30]. Capsanthin is a polar carotenoid and is likely to be localized at the polar surface of

lipoproteins consisting of phospholipids and apoprotein [29,26]. In the ripe fruits of red paprika, capsanthin is esterified with fatty acids. The amount of esterified capsanthin increases as the fruit ripens, and in the ripe fruit, esterified capsanthin accounts for 70–80% of the total capsanthin [31].

Based on the results of the Kruskal-Wallis test (Table 2), there was a significant difference between the treatment without the addition of the extract and the treatment with the added extract of paprika. The highest increase in color of comet fish occurred in treatment C (5%) and the lowest was in treatment without the addition of red paprika extract. The value of color change in treatment C (5%) can be seen starting to experience an increase in the highest color intensity until the 35th day. This indicated that the addition of 5% paprika extract in the feed was the best result and had a significant effect ($P < 0.05$) on the color change of comet fish, but it was not significantly different from treatment D with the addition of 7% paprika extract.

This red paprika has been extensively studied that act as the natural pigment sources for fish [32, 33, 34, 35, 36, 37]. Generally, the skin color of the fish affected by several factors which include carotenoid sources, dietary lipid combination, fish species, and environmental condition [38, 39, 40, 32, 41]. The study from [42] showed that the addition of red pepper (*Capsicum annum*) juice to the color intensity level of koi fish (*Cyprinus carpio* L.) found the best effect was found in the addition of 5% paprika. The study from [35] contradicts that showed when increasing the paprika to 8% showed could improve the skin color of the fish. Same result the study from [43] showed that the caudal and dorsal either muscle or fin showed the highest color at the fish which fed with 8% paprika.

3.2 Survival Rate

The survival rate of comet fish with the addition of paprika extract had no significant effect ($p < 0.05$) on the survival rate of comet fish. From the observations until the 35th day, the percentage of survival rate in all treatments was 100%. It is suspected that the test fish used had high durability and during the research, the quality of the water in the maintenance container was always maintained.

Carotenoids are among the most significant antioxidants of pepper besides phenolics and flavonoids which act synergistically as efficient free radical scavengers [44,45,46]. The most powerful antioxidant carotenoids of pepper are capsanthin and capsorubin, which are the characteristic xanthophylls of the red fruits [47]. Pepper fruit serves as one of the primary dietary sources of provitamin A carotenoids which helps to counter vitamin A deficiency (VAD) and consequently prevents abnormalities in growth, development, immune function and vision [48]. Capsanthin, capsorubin, β -carotene and lutein are the carotenoids of pepper that possess the ability to complement and support the dermal photoprotection system against UV radiation via their strong antioxidant defence, mainly as singlet oxygen quencher and peroxy radicals scavenger [49,50].

3.3 Water Quality

Observation of water quality was one of the parameters that must be observed, because

water quality is one of the factors that influence cultivation activities. Water quality parameters observed in this study were temperature, DO, and pH. Observation of water quality in the study was carried out every 7 days. The results are presented in Table 3.

The quality of water during the study was maintained in good order by means of regular water changes. Water quality measurements during the study showed conditions in the aquarium were in normal conditions, meaning that the water quality conditions were appropriate and in the normal range for comet fish. Based on the results of water quality measurements during the study, it was found that the range of values in all treatments was 24.7-26.4°C, DO 5.6-5.8 mg/l and pH 6.67-7.53. It can be concluded that the quality of comet fish juvenile water during the study was in a suitable condition for raising comet fish. The quality of comet fish water is temperature 26-30°C, DO >5 mg/l and pH 6.5-8.5 [51].

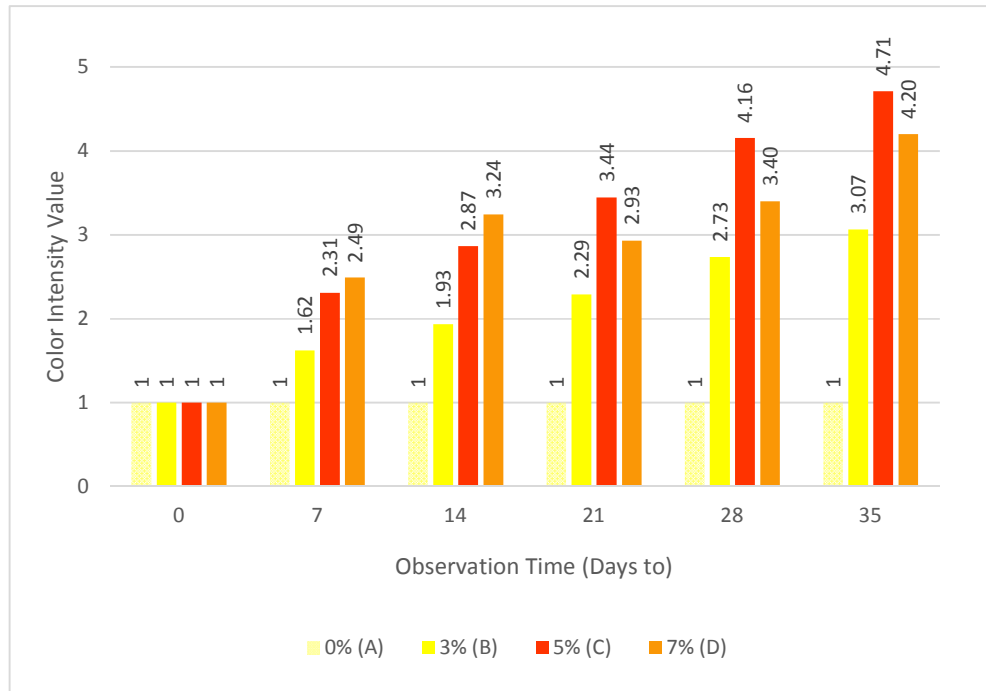


Fig. 1. Graph of average color score increase in comet fish

Table 1. Color intensity of comet fish during research


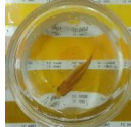
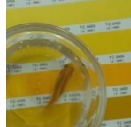






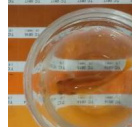

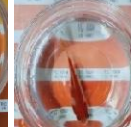





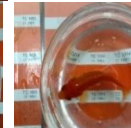


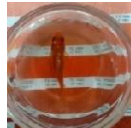
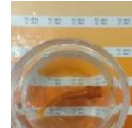


Treatment	Day-					
	0	7	14	21	28	35
A (0%)						
B (3%)						
C (5%)						
D (7%)						

Table 2. Average color intensity value at the end of the comet fish research

Treatments	Increase in color Intensity value
A. No red paprika extract	0 ^a ±0
B. Addition of 3% red paprika extract	2,07 ^b ±0,25
C. Addition of 5% red paprika extract	3,71 ^c ±0,46
D. Addition of 7% red paprika extract	3,20 ^c ±0,40

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

Table 3. Observation results of comet fish water quality

Parameter	Results	Reference (SNI 8110 : 2015)
Temperatur (°C)	24,7-26,4	26 - 30
DO (mg/l)	5,6-5,8	>5
pH	6,67-7,57	6,5 - 8,5

4. CONCLUSION

The addition of paprika extract to commercial feed by 5% was able to increase the best color value in comet fish with an increase in the color intensity value of 4.71. The survival rate of comet fish in all treatments was 100% with the water quality during the maintenance period in the optimal range and meeting standards. The average temperature value was 24.7-26.4°C, DO 5.6-5.8 mg/L and pH 6.67-7.53.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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