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Research Article

Evaluation of growth development and pigmentation of *Heros severus* cultured in a biofloc system with enriched pigment diets

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ABSTRACT. This study aimed to evaluate the development and the increase in the skin pigmentation of the organism *Heros severus* cultured in a biofloc system with diets enriched with carotenoid pigments. The culture was made in 80 L water tanks with 20 juvenile organisms of *H. severus*; each treatment was made by duplicate. Four experimental diets were used: a) trout feed, El Pedregal®; b) TetraColor®; c) carrot and d) beetroot. The diet that obtained the highest values regarding the growth of the fish was the control diet. Regarding the coloration of the fish, the beetroot diet was the diet that presented the highest values with 9.55 µg of total carotenoids in tissue, presenting significant differences (P = 0.001) concerning the other diets, proving that a diet based on beetroot can be a good option for the culture of ornamental fish. Because it allows the organisms to have a survival rate above 90%, have similar growth to the control group, and a significant improvement in coloration, being a natural carotenoid low-cost source to improve the commercialization of organisms. Nevertheless, it did not present significant differences (P = 0.005) regarding beetroot and TetraColor® diets, while it presented significant differences with the carrot diet.

Keywords: *Heros severus*; ornamental fish; carotenoids; biofloc; pigmentation; aquaculture

INTRODUCTION

Ornamental aquaculture in Mexico is an important economic activity due to the high commercial value that many species can have nationally and worldwide (Carvalho-De Oliveira et al. 2020). In that sense, *Heros severus* stand out as a specie with high potential for the ornamental market due to its calm behavior, quick adaptation to new environments, easy reproduction, and beautiful coloration (Alishahi et al. 2014, Veras et al. 2016). In its natural habitat, *H. severus* is associated with areas with high vegetation and feeds on small invertebrates and vegetal material (Soares et al. 2019). However, commercial food has been the most used in captivity strategy for the culture of the specie. Nevertheless, commercial food does not cover all the nutritional requirements of the organisms, so the use of live diets is required as a supplement to formulations from fish meals, which increases the production costs and environmental impact.

New cultivation methods have been proposed, among which the use of biofloc technology stands out, which allows for the addition of a carbon source and high oxygenation, the development of microbial flocs with an association of microalgae, protozoa, rotifers, and nematodes with a high diversity of nutrients availa-

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to fish during the culture (Avnimelech 2012, Emerenciano et al. 2013). Its protein content varies between 30 to 50% but also provides fatty acids, vitamins, minerals, and probiotic microorganisms that reduce bacterial pathogens in the production system. It helps the organisms to have better nutrient assimilation in the intestine (Monroy et al. 2013), which highly improves fish's nutrition, improving their survival and growth.

On the other hand, one of the main aspects to consider in ornamental aquaculture is the coloration of species for its commercialization. However, fish are incapable of synthesizing carotenoids *de novo*, which is why additives such as astaxanthin or carotenoid pigments are added to diets in aquaculture (Simpson 1978), which increases the pigmentation of cultivated species and contributes to animal welfare. It has been proven that carotenoid pigments have antioxidant properties, are precursors of hormones involved in reproduction processes, improve nutrient assimilation, accelerate growth, and increase larval survival (Meyers 2000).

The objective of this study was to evaluate the effect of using biofloc technology and adding diets enriched with pigments on *H. severus* survival, growth, and coloration.

MATERIALS AND METHODS

The experimental work occurred in the Live food production and Biofloc Laboratory installations in the Universidad Autónoma Metropolitana, Unidad Xochimilco.

Experimental design and culture conditions

Fish were acclimatized for four weeks in 25 L aquariums with a thermostat of 150 W to maintain the temperature of 27 ± 2°C. A filter based on rocks and sponges was placed on maintaining the aquarium's cleanliness, and an aerator stone was used to maintain oxygenation.

After acclimatization, the organisms were moved to a water tank of 80 L (90 cm high × 50 cm diameter) filled with 60 L of water and 20 juvenile *Heros severus*. An aeration system was placed in each water tank with a 25 cm aerator stone, with enough power to move the water column. Also, a thermostat of 150 W to maintain the temperature of 27 ± 2°C was placed. Each treatment was made by duplicate.

Feeding of organisms

Four experimental diets were used, two of them were comercial: Trout feed, El Pedregal® with 32% protein and 5% lipids as the control diet, and TetraColor® with 47.5% protein and 6.5% lipids. The other two diets were formulated based on carrots and beetroot respectively. The diets based on carrot and beetroot (carotenoids sources) have approximately 30% protein and 10% lipids provided by chicken gizzards, and 3.5% fiber with apple, banana, and 250 g of oatmeal; also, it was added two mineral and multivitamin supplement pills. These diets were agglomerated with gelatin (50 g). Carrot and beetroot were added at 457 and 561 g, respectively. Both cases provided approximately 14,000 µg of β-carotenes.

The diets were proportionated at 5% of the total fish biomass in the water tank. Food was supplied by 2.5% in the morning and the afternoon, according to the formulas used by (Emerenciano et al. 2011).

Biofloc production

For the formation of the microbial floc, moringa flour was added as the carbon source for TetraColor® treatment. Carrot flour to the treatment diet based on a carrot, and beetroot flour to the treatment based on beetroot, at a proportion of 0.1% of the total biomass of the fish in the water tank, maintaining a C:N relation of 20:1.

Organisms' biometry

The organisms of each treatment, every 15 days, were weighed with a Nimbus® digital scale with a precision of 0.01 g. The biometric variable of length was measured with a digital Vernier with a precision of 0.01 mm.

Fish pigmentation

The method for the extraction of pigments in tissue (Ponce et al. 2016) was used to evaluate the increase of coloration of organisms, for which six organisms from each water tank were randomly selected. It obtained 1 g of body tissue except for the head and digestive tract, and it was placed in a 10 mL vial, where 2.5 g of anhydrous sodium sulfate was added. The sample was pushed against the vial's walls with a glass stirrer, 5 mL of chloroform was added, and it was left to rest for two days at 0°C. When the chloroform formed a clear layer of 1 or 2 cm above the residual, the optical density reading was taken at 500 nm in a spectrophotometer (Spectronic 20 Genesys) for which aliquots of 0.03 mL were taken from the prepared samples and were contrasted with control. The total content of carotenoids was calculated as µg per wet weight of tissue with the following formula:

$$\text{Total content of carotenoids} = \frac{\text{absorption at the peak wavelength}}{(0.25 \times \text{sample weight (g)})} \times 10$$

where: 10 = dilution factor, 0.25 = extinction coefficient.

Data analysis

All obtained values were introduced to a database in Excel 2010 to obtain the descriptive statistic of the information. Also, the growth tendency curves of the measured biometric variables were obtained.

The next formula was used to obtain the gain (G):

G = final value - initial value

For the absolute growth rate (AGR) it was used the next formula:

$$\mathrm{AGR} = \frac{\text{final value} - \text{initial value}}{\text{days of experimentation}}$$

For the instantaneous growth rate (IGR) it was used the next formula:

$$\mathrm{IGR} = \frac{\ln(\text{final value}) - \ln(\text{initial value})}{\text{days of experimentation}} \times 100$$

It was used the formula of Wang et al. (2015) to obtain the survival rate (SR):

SR% = (surviving population / initial population) × 100

The data normality was determined using the Kolmogorov-Smirnov test, and the Bartlett test was applied for the homogeneity of the variance. The statistical comparison of growth and carotenoid concentration between the experimental groups was made through one-way variance analysis. Using the Tukey technique, a multiple means test was performed to identify the groups that obtained significant differences (α = 0.05) using the statistical program Systat 13.0.

RESULTS

Survival

The diet with the highest survival was the beetroot diet, with 95%, followed by the TetraColor® diet, with 90%; the carrot diet had a 70% survival, and the control diet (Trout feed, El Pedregal®) with a 30% survival (Table 1).

Standard length

Mean values (± standard deviation, SD) of the standard length (SL) of the organisms with the different experimental diets are shown (Table 2). The organisms that obtained the highest SL were the ones of the control diet, followed by TetraColor® and beetroot diets, and the diet that obtained the smallest size was the ones in the carrot diet. ANOVA test showed significant differences (P = 0.005); between the carrot diet and the rest of the diets.

Table 1. Survival rate of *Heros severus* with the four experimental diets. Different superscript letters mean significant differences (P = 0.005).

Experimental diet	Survival rate (%)
Control	30.00 ± 7.63^{a}
TetraColor®	90.00 ± 2.88^{b}
Carrot	70.00 ± 5.00^{c}
Beetroot	95.00 ± 5.00^{b}

The control diet obtained the highest G, AGR, and IGR with 17.77 mm, 0.12 mm d^{-1}, and 0.29% d^{-1}, respectively. In contrast, the diet with the lowest values was the carrot diet, with 13.03 mm, 0.09 mm $d^{-1,}$ and 0.21% d^{-1}, respectively (Table 3). The growth curves of the four experimental diets regarding the SL are shown (Fig. 1).

Weight

Mean values (± SD) of the organism's weight with the different experimental diets are shown (Table 4). The control diet was the one that presented the highest value with 6.92 ± 2.80 g, while the beetroot diet was the one that obtained the lowest value with 5.87 ± 2.37 g. ANOVA test did not show significant differences (P = 0.072) between diets.

The growth curves of the four experimental diets regarding weight are shown (Fig. 2). The diet that obtained the highest G, AGR and IGR was the control diet with 5.52 g, 0.04 g d^{-1}, and 1.07% d^{-1}, while the carrot diet presented the lowest value with 4.21 g, 0.02 g d^{-1}, and 0.89% d^{-1} (Table 5). Nevertheless, the groups had no significant differences (P = 0.072).

Fish pigmentation

Regarding fish coloration, a difference in the optical density and carotenoid content was observed in fish with a beetroot diet which obtained the highest value with 0.238 A and 9.55 µg, respectively. The control diet was the one that presented the lowest values with an optical density of 0.121 A and a carotenoids content of 4.85 µg (Table 6, Fig. 3). ANOVA test presented significant differences (P = 0.001) between all diets, except between TetraColor® and carrot diet, which did not show significant differences (P = 0.959).

DISCUSSION

Regarding the biofloc culture, the system allowed maintaining the organisms for six weeks (after the four weeks of acclimatization). Without water changes and managed to maintain the water quality in the culture

Table 2. Mean values (± standard deviation) of standard length (mm) of *Heros severus* fed with the four experimental diets. Different superscript letters in the same row mean significant differences (P = 0.005).

Days of experiment	Experimental diet			
	Control	TetraColor®	Carrot	Beetroot
0	32.39 ± 1.85	34.23 ± 3.55	34.75 ± 4.37	33.12 ± 2.04
15	33.94 ± 1.90	36.95 ± 3.12	37.74 ± 5.03	36.23 ± 4.03
30	36.70 ± 2.43	39.46 ± 3.88	39.28 ± 4.89	37.76 ± 4.17
45	38.89 ± 3.40	41.89 ± 4.08	41.22 ± 6.77	40.56 ± 4.99
60	41.92 ± 3.82	43.75 ± 4.22	43.67 ± 6.52	43.62 ± 4.70
75	43.91 ± 4.10	44.17 ± 4.38	44.57 ± 6.34	42.87 ± 5.73
90	44.12 ± 4.94	45.44 ± 4.75	45.36 ± 6.65	43.66 ± 5.62
105	46.74 ± 5.75	46.53 ± 5.02	45.82 ± 7.08	44.96 ± 5.22
120	47.93 ± 6.62	48.89 ± 5.45	47.77 ± 7.50	46.74 ± 5.72
135	50.17 ± 7.13[a]	50.00 ± 5.84[a]	47.78 ± 7.75[b]	48.47 ± 6.76[a]

Table 3. Gain (G), absolute growth rate (AGR), and instantaneous growth rate (IGR) values of the standard length of *Heros severus* with the four experimental diets. Different superscript letters in the same row mean significant differences (P = 0.005).

Standard length	Experimental diet			
	Control	TetraColor®	Carrot	Beetroot
G (mm)	17.77 ± 4.82[a]	15.77 ± 4.52[a]	13.03 ± 3.63[b]	15.35 ± 3.68[a]
AGR (mm d^{-1})	0.12 ± 0.03[a]	0.11 ± 0.03[a]	0.09 ± 0.02[b]	0.10 ± 0.02[a]
IGR (% d^{-1})	0.29 ± 0.07[a]	0.25 ± 0.08[a]	0.21 ± 0.06[b]	0.25 ± 0.07[a]

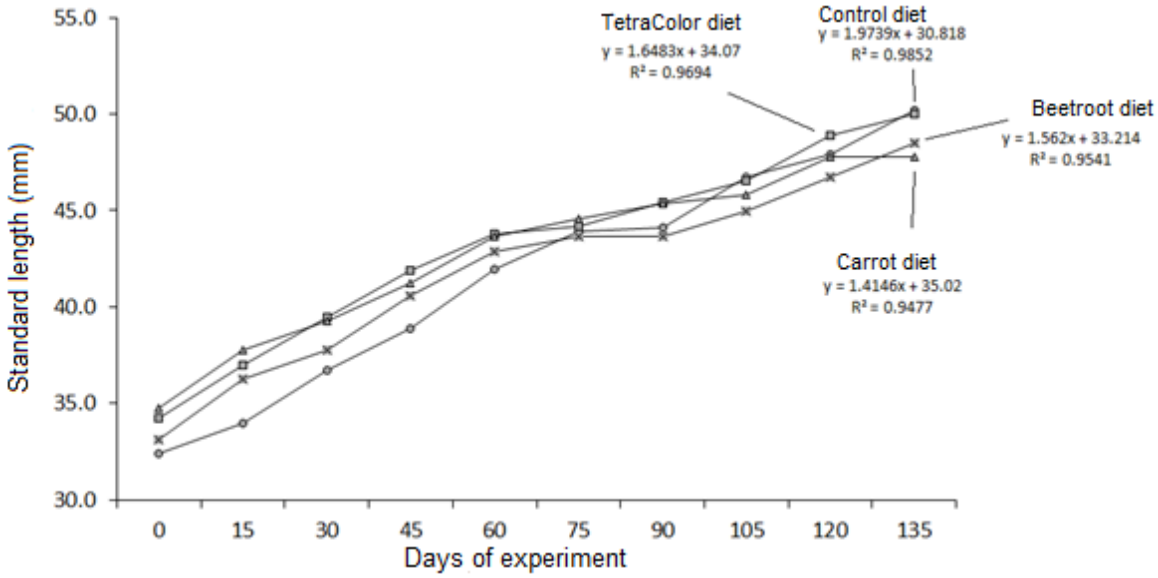

Figure 1. Standard length growth curves of *Heros severus* with the four experimental diets.

medium, this was reflected in the survival of the organisms because after the fifth week, there were no high mortalities. In this regard, the organisms fed with a carrot and beetroot diet maintained survival rates above 90%, having mortalities between 5 to 10%, while the organisms of the control diet had mortalities of 70%. There is an investigation with the ornamental fish *Pterophyllum scalare* about the incorporation of three different pigment sources (*Rhodococcus* sp., *Tagetes erecta*, and *Capsicum annuum*) (Ponce et al. 2016).

Table 4. Mean values (± standard deviation) of weight (g) of *Heros severus* fed with the four experimental diets.

Days of experiment	Experimental diet			
	Control	TetraColor®	Carrot	Beetroot
0	1.39 ± 0.22	1.73 ± 0.37	1.70 ± 0.63	1.54 ± 0.32
15	1.78 ± 0.29	2.39 ± 0.65	2.76 ± 1.45	2.16 ± 0.76
30	2.24 ± 0.45	2.94 ± 0.89	2.87 ± 1.33	2.58 ± 0.92
45	2.80 ± 0.70	3.55 ± 1.15	3.47 ± 2.00	3.19 ± 1.28
60	3.58 ± 1.00	3.99 ± 1.34	4.12 ± 2.27	3.79 ± 1.53
75	4.13 ± 1.28	4.48 ± 1.54	4.60 ± 2.45	4.13 ± 1.66
90	4.29 ± 1.44	4.69 ± 1.65	4.61 ± 2.48	4.26 ± 1.76
105	4.96 ± 1.78	5.17 ± 1.86	4.87 ± 3.02	4.66 ± 1.99
120	5.93 ± 2.28	5.86 ± 2.15	5.71 ± 3.94	5.66 ± 2.61
135	6.92 ± 2.80	6.37 ± 2.37	5.92 ± 4.52	5.87 ± 2.37

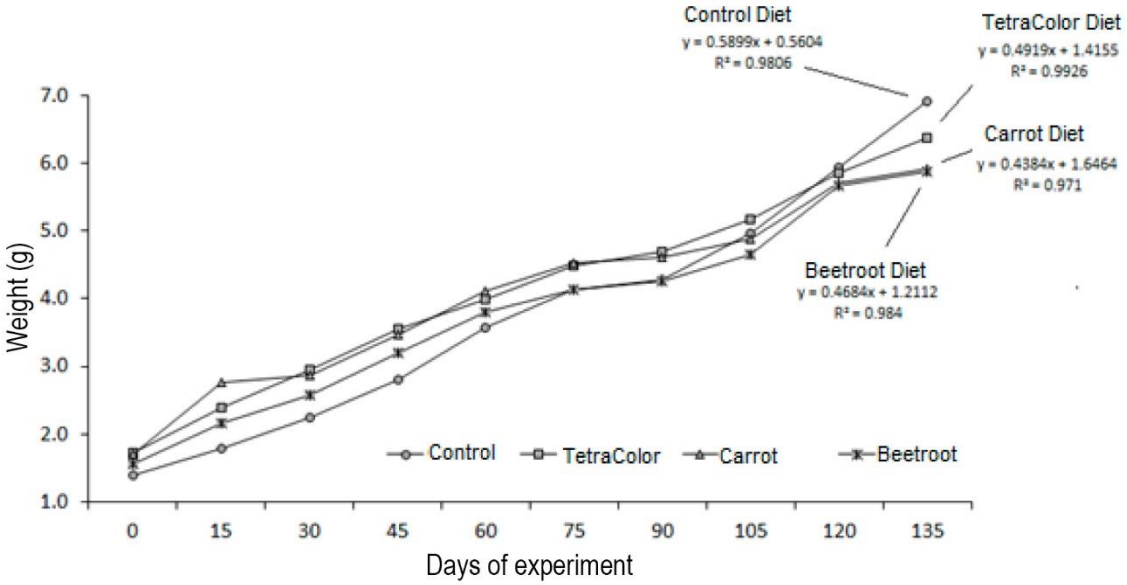

Figure 2. Weight growth curves of *Heros severus* with the four experimental diets.

However, it was not in a biofloc system. The main food source was Wardley® flakes, and they obtained survival rates of 75, 72.22, and 77.8%, respectively, while this investigation obtained survival of 90 and 20% more than the experiment with angel fish without biofloc.

According to the achieved results, the control diet (Trout feed, El Pedregal®) was the one that obtained better results regarding the organism's growth. Nevertheless, the difference was only 1.99 mm for TetraColor®, 2.42 mm for beetroot, and 4.7 mm for the organisms fed with carrot, considering the SL. This difference can be caused to the difference in space the organisms had because control organisms had a bigger space due to their low survival. At the same time, other diets conserved 90% of the initial population. Therefore, they could compete for food, even though it was always given considering the 10% of the organisms' total weight in the culture medium. This condition causes stress in the organisms; therefore, their growth is slower due to food competition (De Sousa et al. 2020).

DeLara et al. (2017) studied *Puntius conchonius* fed a trout-based diet in a biofloc system with 60% protein produced by incorporating molasses and rice flour. When comparing the growth in SL and weight of the *Heros severus* specimens, it is found that they obtained daily increases of 55% (SL), values higher than those obtained in this investigation. However, it is necessary to consider that the specie and food differ from those used in his investigation.

Avilés-López et al. (2017) conducted two experiments with different diets to observe the weight gain of

Table 5. Gain (G), absolute growth rate (AGR), and instantaneous growth rate (IGR) values of the weight of *Heros severus* with the four experimental diets.

Weight	Experimental diet			
	Control	TetraColor®	Carrot	Beetroot
G (g)	5.52 ± 1.21	4.64 ± 0.87	4.21 ± 2.60	4.32 ± 1.32
AGR (g d^{-1})	0.04 ± 0.01	0.03 ± 0.01	0.02 ± 0.02	0.03 ± 0.01
IGR (% d^{-1})	1.07 ± 0.20	0.87 ± 0.09	0.83 ± 0.31	0.89 ± 0.20

Table 6. Mean optical density values and total carotenoid content of *Heros severus* fed with the four experimental diets. Different superscript letters in the same column mean significant differences ($P = 0.001$).

Experimental diet	Optical density (500 nm)	Carotenoids content (µg)
Control	0.121 ± 0.07^{a}	4.84 ± 0.28^{a}
TetraColor®	0.190 ± 0.06^{b}	7.60 ± 1.50^{b}
Carrot	0.188 ± 0.02^{b}	7.52 ± 0.93^{b}
Beetroot	0.238 ± 0.02^{c}	9.55 ± 0.89^{c}

Astronatus ocellatus and *Danio rerio*. One with *Daphnia* sp. enriched with microalgae and bacteria produced in a biofloc system and the other with trout feed with 45% protein and using coffee as the carbon source to produce biofloc. Their values of daily weight gain for *A. ocellatus* were 2.70 and 2.02% for *D. rerio*. In both cases, 1% more of what was obtained in this investigation with *H. severus*. It should be noted that the food in Avilés-López et al. (2017) experiment has 15% more protein.

Castro et al. (2018) experimented with *Cyprinus carpio* using trout feed with 40% of protein and placing the organisms in a biofloc system with four different carbon sources: moringa, yucca, macroalgae, and coffee, determining the gain in weight of the organisms. The daily increase of the organisms was 3.54% for moringa, yuca had 3.50%, microalga presented at 3.75%, and coffee had 3.09%. These values are above the ones obtained in this research with *H. severus*, which goes from 0.83 to 1.07%. In this case, the difference in the quantity of protein was 10%.

Ponce et al. (2016), that worked with *Pterophyllum scalare* fed with Wardley flakes as the control group and Wardley flakes with a supply of *Rhodococcus* sp., *Tagetes erecta*, and *Capsicum annuum*, as carotenoids sources, obtained weight values of 0.57, 0.67, 0.97, and 1.06% daily increase. They obtained values of 0.29, 0.23, 0.30, and 0.50% of the daily increase for the length. Comparing those values with this investigation made with *H. severus* obtained higher values for the weight because the carrot and beetroot diet obtained daily increment values of 0.83 and 0.89%. For SL, the increase was similar to those obtained with the *Rhodococcus* sp. diet, 0.23% daily increase, and with control, 0.29%. This investigation obtained values from 0.21 to 0.25% of the daily increase. It should be noted that Wardley flakes have between 28 to 30% of protein, which is the protein value that the carrot and beetroot diets had (30%).

With the above, what was said by De Sousa et al. (2020) is that a diet rich in protein allows substantial growth. However, it can also be mentioned that adding vegetable protein from carrots and beets or other vegetable sources, rich in carbohydrates and pigments, can not only be used for coloring fish but also intervenes in their growth, as mentioned by Amar et al. (2001). The effectiveness of carotenoids in fish growth is specific for every fish because each can have a different metabolic pathway and, therefore, its transformation towards tissue formation (Alishahi et al. 2014). As reported by Wyban et al. (1997) and Supamattaya et al. (2005), better efficiency of reproducers and larvae of *H. severus* was observed.

Regarding growth efficiency in terms of their weight, better instantaneous growth rates were obtained than in the investigation conducted by Alishahi et al. (2014). They used *D. salina* flour as a supplement. They found that 100 to 200 mg kg^{-1} significantly affects growth efficiency, including the specific growth rate, food conversion rate, and average and daily weight and size gain of *H. severus*. Nevertheless, the best IGR they obtained was 0.50% d^{-1}, while in this experiment, IGR was obtained above 0.80% d^{-1} with the carrot and beetroot diets. However, the growth rates obtained in this experiment were lower than those presented by Vesal & Vosooghi (2017), which worked with juveniles of *H. severus* with fish oil and soy diets. These authors obtained weight IGR up to 25.57% d^{-1} and a minimum of 10.85% d^{-1}, which can be due to the quantity of raw protein that the used diets had, which was above 64%, while commercial diets used in this

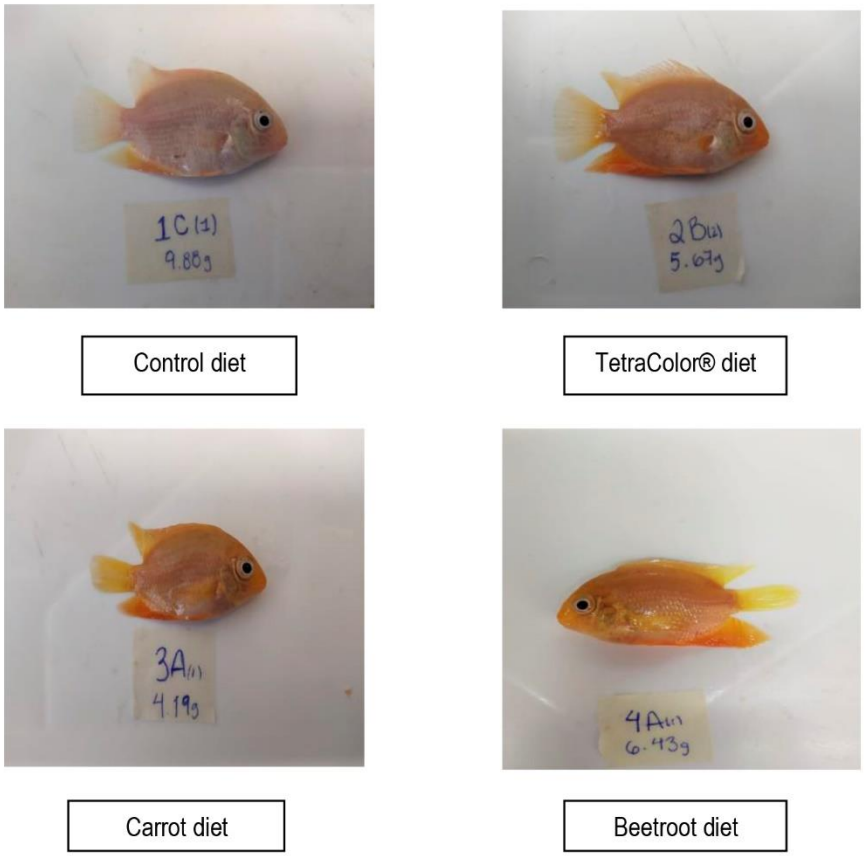

Figure 3. Color differences of *Heros severus* fed with the four experimental diets.

experiment had 40% of raw protein and carrot and beetroot diets had 30%.

It should be noted that there are studies such as Paixão et al. (2019), which worked with optimal feeding frequency for larvae and juveniles of *H. severus*. Juveniles with a feeding frequency of twice a day (similar to this experiment) obtained an IGR in weight of de 9.13 ± 0.07% d^{-1}. These authors mention that feeding frequency does not affect the survival and uniformity of the organisms. As mentioned before at the beginning of the discussion, variations can occur due to competition or the species' hierarchical behavior, which is accentuated when the experiments do not start with organisms of the same size or weight. When the organisms grow evenly, the survival rate is high; it is also due to the quality of the feed supplied, balancing the number of proteins, carbohydrates, and pigments, such as the carrot and beetroot diets. Also, Carvalho-De Oliveira et al. (2020) worked with *H. severus* fry fed with *Artemia* nauplii at different feeding rates, obtaining a weight IGR of 4.5 to 6.2 % d^{-1}. Values below those obtained in this investigation.

Regarding fish coloration, the organisms with a beetroot diet were the ones that obtained better pigmentation, with a total carotenoid content of 9.55 µg. In contrast, control and TetraColor® groups obtained 7.52 and 7.60 µg, respectively. The latter is of great importance. With the carrot and beetroot diet, natural sources of carotenoid pigments are used, leaving aside synthetic carotenoids. According to Gil et al. (2015), its only contribution to the diet pigments at a considerably high price and represent 15 and 20% of their cost, in addition to representing a possible toxicity factor for organisms in culture. By being a stereoisomer different from the natural carotenoid, they are difficult to incorporate into the fish diet (Ako & Tamaru 1999).

In the same way, the organisms with a beetroot diet obtained a higher coloration than the one obtained by Alishahi et al. (2014), where the natural pigment source was *D. salina* given to *H. severus*, the organisms reached a concentration of carotenoids in the skin of 1.0 to 1.2 µg. Another example of the use of natural carotenoids sources to increase the color in fish is the work made by Ponce et al. (2016), which evaluated the

use of *Artemia franciscana* enriched with *Rhodococcus* sp. to increase the growth, survival, and coloration of *Puntius conchonius*. The fish fed with the enriched diet presents a carotenoid content in the skin of 7.04 µg, a value lower than the one obtained in this experiment with organisms fed with the beetroot diet. On the other hand, Chatzifotis et al. (2005) used a diet with Rovimix β-caroteno® at the end of the experiment to improve the coloration of *Pargus pargus*, and the organisms presented a carotenoid concentration in the skin of 8.6 µg, a value below the obtained with the beetroot diet in this experiment.

The search for natural sources of pigments for aquatic organisms has increased investigations to find natural diets that help color the organisms. An example of this is the investigation of Gil et al. (2015), which used diets with red prawn oil (*Pleuroncodes planipes*) to improve the pigmentation, growth, and survival of Koi carp hatchlings (*C. carpio* var. *haematopterus*). Also, Chatzifotis et al. (2005) used enriched diets with *Hematococcus pluvialis*, improving the coloration of *P. pargus*. In addition, there is an investigation made by Nhan et al. (2019), which used a diet based on sweet potatoes and another based on *Ulva intestinalis* to increase the coloration of clownfish (*Amphiprion ocellaris*). These investigations agree on the use of diets with natural sources of carotenoids for the culture of aquatic organisms, mostly used in the aquarium because it represents having a better cost-benefit for the improvement of coloration, which is one of the fundamental characteristics of determining the quality of ornamental fish.

Thanks to this, the biofloc system allowed an economically more profitable culture. Due to low water consumption and having to use less food than traditional culture, only 5% of the total biomass of each culture was supplied in food. The floc's nutritional properties are used, considering that using different carbon sources such as carrot and beetroot flour also allows a good biofloc system.

This research is another example of ornamental fish culture being made in a biofloc system and having a more sustainable aquaculture. Works have been done with *Poecilia reticulata* (Sreedevi & Ramasubramanian 2011), *Scatophagus argus* (Liu et al. 2014), *Carassius auratus* (Faizullah et al. 2015), *Pseudotropheus saulosi* (Harini et al. 2016), *Xiphophorus maculatus* (Boaventura 2016), *Puntius conchonius* (De Lara et al. 2017), *Danio rerio* and *Archocentrus nigrofasciatus* (Da Silva 2018), *Melanochromis* sp. (Castro-Castellón et al. 2020a) and *Astronotus ocellatus* (Castro-Castellón et al. 2020b).

CONCLUSIONS

According to the obtained results in this investigation, it can be concluded that the fish *H. severus* can be cultivated in biofloc using beetroot and carrot as additives to enrich diets with pigments and as the carbon source to promote the biofloc system. These, especially beetroot, helped to obtain survival rates above 90% and growth parameters similar to commercial diets with and without pigments.

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