

Effects of water exchange and feed quality on carcass composition, ratio, color and growth performance of striped catfish (*Pangasianodon hypophthalmus*)

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Abstract. The technological aspect in fish farming becomes an essential factor associated with meat quality. The present work aimed to investigate the effects of water exchange and protein content on carcass composition, ratio, color and growth performance of striped catfish, *Pangasianodon hypophthalmus*. The experiment was arranged according to a 2 × 2 factorial design: water exchange at 0 and 100% day⁻¹ (treatments W0 and W100, respectively) and feed protein at 16 and 32% (treatments P16 and P32, respectively). The results showed that the water quality fitted the requirement for fish growth: the plankton abundance in W0 was higher than in the treatment W100. The protein content of the flesh was significantly higher in treatment P32 than in treatment P16. Conversely, the fat content of the flesh was significantly higher in treatment P16 than in treatment P32. All the treatments revealed a significant effect on the carcass ratio, in which the highest weight was found in both treatments, P32 and W100. The highest yellowness (b*) score of the fillet was found in treatments P16 and W0. In addition, the redness (a*) value was found higher in treatment P32 than in treatment P16. The protein levels had a significant difference in the weight gain and SGR of the fish, while the water exchange did not significantly affect them. The survival rate did not significantly differ among treatments, while the FCR in treatment P32 was lower than in treatment P16. The water exchange could be applied to improve the *P. hypophthalmus* meat quality by inducing a color improvement.

Key Words: aquaculture system, carcass ratio, color fillet, protein level.

Introduction. Fish is commonly used as one of the protein sources for human demand all over the world and nowadays the amount of catches from aquaculture has continued to improve compared to wild catches (California Environmental Associates 2018). To meet the demand, the fish culture system should not only consider the quantity, but also the quality of the fish including nutritional composition, food safety, appearance, texture, color, taste and flavor. The high quality of striped catfish (*Pangasianodon hypophthalmus*) fillet is identified by its color. The white fillet will contribute to a higher acceptance of the consumers, instead of the red or yellow fillet. Consequently, the quality of the fillet can determine the market (Amaya & Nickell 2015). It seems that the culture system and the fillet process are crucial factors to produce a good quality *P. hypophthalmus* fillet (Ramadhan et al 2016).

In the world, the quantity and quality of fresh water continues to decline. On the other hand, the need for protein continues to increase. The quality freshwater resources of the fish farmers for fish producing in ponds are limited, which can affect the meat quality and the production performance. Aquaculture technology dissimilarities in each area might result in various characteristics of the flesh, particularly a color ranging from yellow to white, also producing a musty flavor (Lehto & Vielma 2018). Nurilmala et al (2015) reported that pangasius fillet obtained from floating net culture with relatively stagnant water tended to have higher yellow intensity compared to that produced in ponds. In addition, Burr et al (2012) investigated the impacts of depuration on the quality of salmon cultured in a flow-through tank and stocked-grow out tank.

The feed is considered as one of the highest expenditures in aquaculture production leading to approximately 70% of production cost. The use of low protein feed (approximately 20%) can be one of the possible economic solutions (Hua et al 2019). This may adversely affect the quality of the fish flesh, i.e. the nutritional content, taste, texture and color (Hardy & Lee 2010). However, the flesh color seemed to be unchanged when treated with different feed compositions, as reported by Johnsen et al (2011). Briefly, the proportion of fish meal (20, 15, 10%) did not cause any impacts on the odor, taste, texture and color of the flesh, as well as on the growth performance of the fish (growth rate, feed consumption, mortality). Hardy & Lee (2010) reported that the feed composition could alter the quality of the fish, including the skin color, due to changes in source and amount of pigment such as astaxanthin in the feed. Furthermore, the replacement of fish meal and fish oil with plant protein was associated with changes in the odor and composition of fatty acids, which in turn alters the taste. Several findings from the field of fish farming relate the limited fresh water sources to the fish meat more yellow color and to the smell of mud, but the cause of the low quality of fish meat has not been determined. On the other hand, the fish raw material for the fillet processing industry affects the fillet quality.

To determine the cause of the lower quality of fish meat from ponds with limited freshwater resources, research was carried out in earth ponds. Therefore, the present work aimed to investigate the effects of water and feed quality on carcass composition, color, ratio and growth performance of *P. hypophthalmus*. The results of this study can be used as a reference for further research to solve the issue of the poor fish quality.

Material and Method

Experimental animal and materials. A total of 20 *P. hypophthalmus* with an average weight of 80 g were supplied from the teaching farm of Jakarta Technical University of Fisheries (JFU). Chemicals for analysis included H₂SO₄, NaOH, NH₄OH, Se powder, phenolphthalein (Merck).

Experimental design. The 2 × 2 factorial design was arranged, consisting of two variables and two-way interaction: (1) water exchange, at 0 and 100% day⁻¹, of water volume (W0 and W100, respectively) and (2) feed protein content of 16 and 32% (P16 and P32, respectively). The experiment was carried out in triplicates, during 90 days, at the JTUF, Indonesia. Fish were randomly maintained in 12 ponds (each 1.5 × 3.0 × 1.0 m) filled with water at a height of 80 cm and a density of 20 individuals. The fish were fed twice a day with commercial feed, at a dose of 4% of fish biomass.

Analytical methods. The water quality, including the abundance of plankton, was analyzed. The fish were analyzed for the quality of fillet, including their chemical composition and color.

Analysis of water quality and plankton. Dissolved oxygen was quantified using a DO meter, while other water quality indicators such as pH, alkalinity, TOM, total ammonia nitrogen (TAN) and nitrite were detected using APHA (2017). Plankton abundance was measured using a Sedgwick-Rafter Counting Cell. All these parameters were analyzed at The Research Center for Fresh Water Fish in Bogor.

Production performance analysis. The carcass ratio, weight gain, specific growth rate, feed conversion ratio and survival rate were determined as follows (Robson & Spangler 1978; Weatherley & Rogers 1978):

$$X1 = Y1/Z1 \times 100$$

Where:

X1 - weight gain (%);

Y1 - final body weight (g) – initial body weight (g);

Z1- initial body weight (g).

$$X2 = Y2/Z2 \times 100$$

Where:

X2 - specific growth rate (SGR) (%);
Y2 - in final body weight - in initial body weight;
Z2 - days number of the experiment.

$$X3 = Y3/Z3 \times 100$$

Where:

X3 - carcass ratio (%);
Y3 - carcass weight;
Z3 - body weight.

$$X4 = Y4/Z4 \times 100$$

Where:

X4 - feed conversion ratio (FCR);
Y4 - feed supply (kg);
Z4 - fish biomass increase (kg).

$$X5 = Y5/Z5 \times 100$$

Where:

X5 - survival rate (%);
Y5 - number of fish harvested - number of fish stocked;
Z5 - number of fish stocked.

Chemical composition. The moisture analysis was performed at a drying temperature of 105°C for 5 h, while for the ash analysis the sample was dried at 100°C for 24 h, then ashed at 550°C for 8 h. The lipid content was determined using the soxhlet method, with the extraction of the sample at 60°C for 8 h, then an oven-drying at 105°C for 2 h. The crude protein content was determined by using the distillation method. The sample was destructed at 410°C for 2 h, before distillation and titration, using a Kjeltec 2300 (FOSS) Laboratory Analyzer Unit (AOAC 2005).

Color measurement. The color of the fish flesh was measured according to a method of Shie & Park (1999), using a chromameter (CR-310, Minolta, Japan) previously calibrated on the standard white plate (Markovic et al 2013). The fillet was analyzed for lightness L* (0-100), redness a* (+ = red, - = green), and yellowness b* (+ = yellow, - = blue). The measurement of these 3 color indicators was based on the International Commission on Illumination (CIE) and was immediately performed in triplicates (Shie & Park 1999). The color measurement for the flesh of *P. hypophthalmus* was also performed by Kulawik et al (2016).

Statistical analysis. The variance of data was analyzed, according to the 2 × 2 factorial design. The comparison between means was performed with the least significant difference, followed by Duncan's multiple range tests (for Post hoc analyses). The results were considered statistically significant when p-values were below 0.05 (p<0.05). All statistical analyses were conducted using the Statistical Package for Social Sciences (SPSS, version 16.0 for Windows).

Results

Water quality and abundance of plankton. Table 1 presents the results of the water quality analysis, including DO, pH, TAN and plankton. These parameter levels were at the desirable range for fish growth. In a particular case, some parameters, i.e. TAN, appeared to be higher in W100 than in W0. The treatments resulted in the variability of the planktonic organisms in the water. Obviously, Chlorophyceae and Bacillariophyceae could be noted as the most abundant planktonic organisms in W0 than in W100, followed by rotifers.

Table 1

Water quality parameters abundance of plankton

Treatments	DO (mg L ⁻¹)	pH	TAN (mg L ⁻¹)	Chlorophyceae (cells mL ⁻¹)	Bacillariophyceae (cells mL ⁻¹)	Rotifer (cells mL ⁻¹)
W0xP16	5.10±0.51	6.65±0.37	0.075±0.001	10,436±1739	3,176±405	2,853±491
W0xP32	4.80±0.49	6.68±0.33	0.039±0.001	7,123±326	1,643±295	1,343±205
W100xP16	4.57±0.11	6.53±0.07	0.085±0.002	4,083±526	656±87	146±39
W100xP32	4.80±0.01	6.67±0.07	0.087±0.002	2,883±125	59±14	7±2

Carcass composition and ratio. Table 2 presents the results of protein content, fat content, and carcass ratio of pangasius fillet. The results suggested that the treatment P32 resulted in a higher protein content of the flesh, compared to the treatment P16 (P<0.05). In terms of fat content, the treatment P16 yielded a higher fat content compared to the treatment P32 (P<0.05). The results demonstrate that the treatment P32 resulted in a higher protein content of the fillet than the treatment P16. The highest carcass ratio (47.87%) was achieved for the treatment P32 and W100. Meanwhile, the treatment P32 and W0 yielded a significantly higher carcass ratio (44.59%), compared to the fish treated with P16 and W100 (38.88%) as well as with the treatment P32 and W0 (37.87%). Among the treatments with P32, no significant difference was found in the carcass ratio between W0 and W100 (P>0.05). Carcass ratio reached the highest quantity for the treatment P32 and W100; conversely, the lowest one was found for the treatment P16 in combination with either W0 or W100.

Table 2

Chemical components and carcass ratio of *Pangasianodon hypophthalmus* fillet treated with levels of water exchange and feed protein

Treatments	Protein content (%)	Fat content (%)	Carcass ratio (%)
W0xP16	67.12±0.49 ^a	22.39±0.61 ^a	37.87±1.17 ^a
W0xP32	72.01±1.52 ^b	19.62±1.44 ^c	44.59±1.22 ^b
W100xP16	65.39±1.42 ^a	25.69±1.41 ^b	38.88±1.05 ^a
W100xP32	72.8±1.45 ^b	17.34±1.36 ^c	47.87±1.12 ^c

^{a,b,c}- different letters in the same column indicate significant difference at P<0.05.

Figure 1 shows the qualitative assessment of the fillet color. The results demonstrated that the treatment W100 produced a darker fillet (Figure 1A and 1B), while the fillet of the fish treated with the treatment W0 showed a yellow fillet (Figure 1C and 1D).



A



B

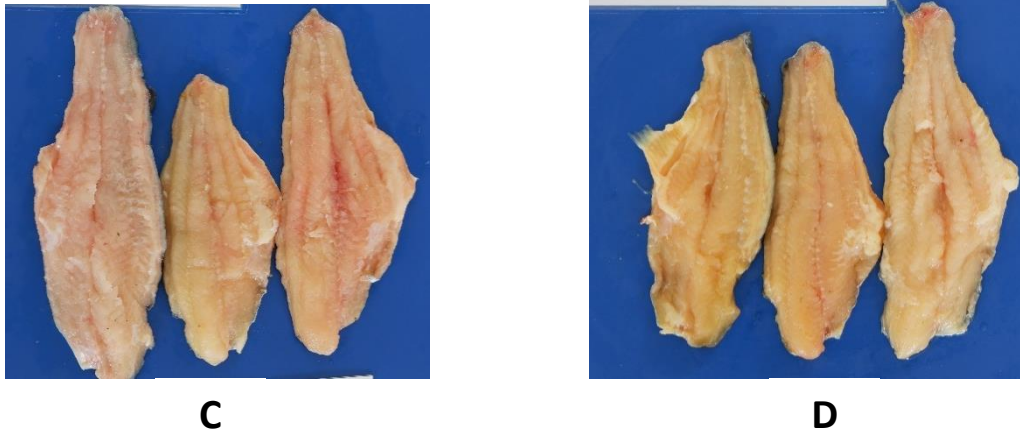


Figure 1. Appearance of fillet under different combined treatments of water exchange with feed protein: A=W100xP32; B=W100xP16; C=W0xP32; D=W0xP16.

Meanwhile, Table 3 presents the results of the *P. hypophthalmus* fillet's color profile. The results of the fillet's color measurement showed the variety of the color profile as represented by b, a and L values. Fillet yellowness (b^*) is significantly different for the treatment W0 compared to the treatment W100 ($P<0.05$). Likewise, for the treatment P16, the meat was more yellow than for the treatment P32 and W0 ($P<0.05$). It is argued that the feed protein significantly influenced the redness (a^*) value ($P<0.05$), being higher in the sample treated with P32 than for the treatment P16, regardless of the water exchange treatments. Additionally, the lightness value (L^*) differed significantly ($P<0.05$) among the treatments. The highest L value, 64.18, was found in the treatment P16 and W100.

Table 3
Color profile of *Pangasianodon hypophthalmus* fillet treated at different levels of water exchange and feed protein

Treatments	Yellowness(b^*)	Redness (a^*)	Lightness (L^*)
W0xP16	28.48±2.39 ^a	9.37±1.88 ^a	60.73±0.86 ^a
W0xP32	21.10±4.06 ^b	11.46±1.96 ^b	58.56±2.29 ^a
W100xP16	14.04±1.08 ^c	7.99±1.84 ^a	64.18±1.17 ^b
W100xP32	12.21±1.13 ^c	12.15±0.61 ^b	58.05±1.06 ^a

a,b,c- different letters in the same column indicate significant difference at $P<0.05$.

Growth performances. Table 4 presents the weight gain, specific growth rate (SGR) and FCR. The results showed that fish fed with a higher level of protein (the treatment P32) yielded a more significant increment of weight gain ($P<0.05$) than those fed with lower protein levels (the treatment P16).

Table 4
Growth performances of *Pangasianodon hypophthalmus* treated at different levels of water exchange and feed protein

Treatments	Weight gain (%)	SGR ($\% \text{ day}^{-1}$)	Survival rate (%)	FCR
W0xP16	86.46±24.69 ^a	0.669±0.130 ^a	91.67±7.64 ^a	3.44±0.08 ^a
W0xP32	303.61±13.27 ^b	1.530±0.047 ^b	96.67±5.77 ^a	1.51±0.05 ^b
W100xP16	70.93±6.67 ^a	0.647±0.114 ^a	90.00±0.00 ^a	3.34±0.01 ^a
W100xP32	298.45±6.70 ^b	1.534±0.073 ^b	96.67±2.89 ^a	1.63±0.04 ^b

a,b,c- different letters in the same column indicate significant difference at $P<0.05$.

Furthermore, the water exchange treatment showed no effects on both parameters, weight gain and SGR. In addition, the SGR was higher in fish treated with the treatment

P32 than in those exposed to the treatment P16 ($P < 0.05$), while the water exchange did not cause significant effects on the SGR ($P > 0.05$). Also, the FCR in the treatment P32 was significantly lower compared with the treatment P16 ($P < 0.05$). The survival rate did not significantly differ among the treatments ($P > 0.05$). In terms of survival rate, there was no significant difference between treatments.

Discussion. Water quality parameters widely affect aquaculture. Table 1 shows that all the chemical water quality parameters are in line with to the *P. hypophthalmus* growth requirements. The condition of the ponds in this study is the same as in the semi-intensive pond for *P. hypophthalmus* cultivation, whose water quality parameters are as follows: pH of 6.8 to 7.4, total ammonia of 0 to 5 mg L⁻¹ and DO of 2 to 6 mg L⁻¹ (Abedin et al 2017). The recirculation system in cultivation system of *Pangasius hypophthalmus* produced an optimal water quality, with a decreased ammonia concentration (Zidni et al 2017). Bacillariophyceae or diatoms are microalgae containing carotenoid pigments like diadinoxanthin and diatoxanthin (Kaas et al 2017).

Table 2 shows that the treatment P32 resulted in a higher protein content and in a lower fat content of the fish meat, compared to the treatment P16. Tešić et al (2014) tested different fish meals, with no significant influence on the trout meat chemical composition, except for the fat content, which was significantly higher. Fish were fed with a combined meal consisting of 75% fish food pellets and 25% sardine, then it was fed with a mixture containing 30% fish meal, 35% soybean meal, 30% sardines, 5% fish oil and eventually it was fed with standard complete pellet feed. Additionally, Ragaza et al (2015) reported that fish fed diets supplemented with seaweed *Euclima denticulatum* exhibited higher fatty acid accumulation in dorsal muscle when compared with those of fish fed fishmeal-soy protein. The carcass ratio reached the highest quantity for the treatment P32 and W100; conversely, the lowest one was found for the treatment P16, in combination with either of the two treatments W0 or W100. This suggests that the protein content predominantly contributes to the carcass ratio.

Figure 1 shows that the treatment W100 produced a darker fillet, while the fillet of the fish treated with W0 produced a yellow fillet. Table 3 shows a variety of color profile as represented by b* (yellowness), a* (redness), and L* (lightness) values. Regardless of the water exchange condition, the treatment P16 yielded a high score for b* value (yellow). In this work, the water exchange is an essential factor towards a yellow color of the fillet. Noticeably, the plankton abundance in the treatment W0 was higher than in the treatment W100, which may also affect the color of fish meat. The abundance of phytoplankton, especially Bacillariophyceae, for the treatment W0 was higher than for the treatment W100. This type of phytoplankton contains carotenoids (fucoxanthin) which is a yellow pigment (Takaichi 2011). Xanthophyll in the environment is absorbed through the gills and/or phytoplankton is eaten by fish causing the fish meat to turn yellow. Promya & Chitmanat (2011) demonstrated that the carotenoid content in catfish fed with 5% *Cladophora* + basal diets were significantly than in those fed with 5% *Spirulina* + basal diets, 3% *Spirulina* + basal diets and 0% algae + basal diets. Bano et al (2020) suggested that the carotenoid pigment component of the marigold flower powder can stimulate the skin pigmentation of *Trichogaster fasciata*.

In terms of color, the results showed that the treatment P32, combined with either of the two treatments W0 or W100, yielded a higher value of a* (red) compared to the treatment P16. High levels of feed protein will trigger the formation of myoglobin, which is a protein with a spherical structure that stores oxygen. Redfish meat contains a lot of myoglobin. Nisa et al (2016) stated that *P. hypophthalmus* fillet tends to be more reddish due to a greater level of myoglobin. Another research suggested that the chemical profile of feed contributed to the meat color of *P. hypophthalmus* fed with β -carotenoid-rich feed (Gopan et al 2018). Hardy & Lee (2010) asserted that feed composition could affect the quality of fish, including a skin color modification due to the carotenoid (i.e. astaxanthin). Meanwhile, the substitution of fish meal and fish oil with plant proteins could change the odor and composition of the fatty acids. Tzanova (2018) explained that the asxtaxanthin and canthaxanthin (two xanthophylls) are more concentrated in the cardiac muscle than in the skeletal muscle.

The treatment W100xP16 yielded a high L* score compared to other treatments, but the score does not differ significantly from treatment W0xP16 and W100xP32. The high L* value indicates that stripped catfish fillet has a lighter color. The L* score has a range between 0 and 100. The higher the L* score, the higher the lightness level. L* scores are closely related to the presence of oxymyoglobin. When oxymyoglobin is oxidized, metmyoglobin is formed. The latter is of a dark brown in color, which is indicated by a lower L* score. Oxymyoglobin oxidation occurs in fish meat of a lower quality, the L* score indicating the level of the fish freshness (Chaijan & Panpipat 2014). The higher the L score, the higher the freshness of the fish (Ünal et al 2019). It is important to note that color is a key indicator for the consumer preference (Rathod et al 2018).

Table 4 reveals that feed containing high protein (the treatment P32) showed more satisfying effects on the weight gain and SGR of the fish than feed containing low protein (the treatment P16). According to Khan et al (2018), feed rich in proteins (40%) produced the greatest protein content observed in Pangasius fish, compared to other treatments. Protein concentration in feed contributed positively to the growth rate and to the protein efficiency ratio, but negatively related to the feeding conversion ratio (FCR). Opiyo et al (2014) explained that diets with different crude protein levels (32.7, 28.0, 16.0%) determine significantly different mean weights, specific growth rates and feed conversion ratio, with a better performance measured for the diet with 32.7% protein. The optimum level of protein incorporated to the feed may vary, depending on the source of the protein (Ahmed & Maqbool 2017). In this experiment, the treatments W0 and W100, regardless of protein level, seemed too insignificant for altering the weight gain and SGR. Water quality is not directly responsible for the growth performances, but it determines the quality of fillet in such aspects as taste. As also previously reported by Burr et al (2012) in two trials using running water and a recirculating system a water exchange for up to 10-15 days is needed to alleviate the residues of geosmin and 2-methylisoborneol (MIB). In the flow-through system, the presence of MIB dominates over geosmin; in a running water system, geosmin and MIB account for off-flavors of the fillet.

Regarding the consumer acceptance, the *P. hypophthalmus* fillet with a yellow color is less accepted and cheaper compared to the meat of pink and white color. The economic aspect needs to be considered when implementing a technology. *P. hypophthalmus* farmed in a recirculating aquaculture system is considered more "eco-friendly", but it also costs more (Ngoc et al 2016).

Conclusions. The results of the study showed that the protein content of the flesh was significantly higher in treatment P32 than in treatment P16. Conversely the fat content of the flesh was significantly higher in treatment P16 than in treatment P32. The highest carcass ratio was found in treatment P32 and W100. The treatment P16 and W0 reached the highest score for b* (the yellowness), in the fillet of *P. hypophthalmus*. This shows that fish fed with feed containing a higher protein (treatment P32) causes an increment of weight gain, SGR, and lower FCR in comparison with P16. Based on this study, the water exchange method could be applied to improve the fillet quality by inducing a color improvement.

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Conflict of interest. The authors declare no conflict of interest.

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