Original article

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Evaluation of health status and meat quality of dumbo catfish *Clarias gariepinus* maintained using the biofloc system

Evaluasi status kesehatan dan kualitas daging ikan lele dumbo *Clarias gariepinus* yang dipelihara menggunakan sistem bioflok

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ABSTRACT

This research aimed to evaluate the health status, growth performance, and meat quality of African catfish *Clarias gariepinus* maintained in biofloc and nonbiofloc systems. This study applied 3 treatments with 4 replications, i.e. K500 (control, a non-biofloc system at a fish stocking density of 500/m^3), BF500 (biofloc system at a fish stocking density of 500/m^3), and BF700 (biofloc system at a fish stocking density of 700 /m^3). The initial body length and bodyweight of the fish used in this experiment were 10–12 cm and 10–12g, respectively. Feeding was done by using a commercial feed containing 29.76% protein content. The results showed that fish survival in treatments BF500 and BF700 were significantly different (P<0.05) from the control. The growth rate of African catfish in the control (K) was the lowest (3.64 ± 0.56%) among the treatments. The results of the organosensory test showed that the application of biofloc systems produced a higher quality of fish meat with a range value of 7–9 compared to that of the control with a range of 6–7. In conclusion, the application of biofloc systems could improve the fish health status, growth performance, and meat quality of African catfish.

Keywords: biofloc systems, *Clarias gariepinus*, density, health status, meat quality

ABSTRAK

Penelitian ini bertujuan mengevaluasi status kesehatan, kinerja pertumbuhan, dan kualitas daging ikan lele Afrika *Clarias gariepinus* yang dipelihara menggunakan sistem bioflok dan nonbioflok. Penelitian ini menggunakan 3 perlakuan dengan 4 kali ulangan, yang terdiri dari: K500 (kontrol, sistem nonbioflok dengan kepadatan ikan 500 ekor/m^3), perlakuan BF500 (sistem bioflok dengan kepadatan ikan 500 ekor/m^3), dan perlakuan BF700 (sistem bioflok dengan kepadatan ikan 700 ekor/m^3). Ikan yang digunakan berukuran panjang dan bobot awal masing-masing 10–12 cm dan 10–12 g. Pemberian pakan dilakukan dengan menggunakan pakan komersil dengan 29,76% protein. Hasil penelitian menunjukan tingkat kelangsungan hidup antara perlakuan BF500 dan BF700 berbeda nyata (P<0.05) dengan perlakuan K500. Laju pertumbuhan ikan lele pada perlakuan K500 lebih rendah dibandingkan perlakuan lain. Hasil pengujian *organosensory* menunjukkan bahwa ikan yang dipelihara dalam sistem bioflok menghasilkan kualitas daging yang lebih tinggi dengan kisaran nilai 7–9 dibandingkan dengan ikan K500 dengan nilai 6–7. Berdasarkan hasil penelitian ini dapat disimpulkan bahwa aplikasi sistem bioflok dapat meningkatkan status kesehatan, laju pertumbuhan, dan kualitas daging ikan lele Afrika.

Kata kunci: bioflok, *Clarias gariepinus*, kepadatan, kualitas daging, status kesehatan

INTRODUCTION

In order to increase production to meet the demand of aquaculture products, culture intensification become a necessity. As a consequence, however, nitrogen waste generated in the system will increase remarkably (Avnimelech, 2007; Dauda *et al.*, 2019). Ammonia is one of the most common inorganic nitrogenous waste in an aquaculture system, which can be critically threatening the welfare of the cultured species. Thereby, ammonia elimination in the culture system is required (Pujiastuti & Suwartha, 2017). Ammonia reduction in aquaculture system can be conducted by employing heterotrophic bacteria in biofloc systems (BFT). Biofloc system is an alternative method to manage aquaculture waste that was adapted from domestic waste treatment (De Schryver *et al.,* 2008). The main principal of BFT system is ammonia conversion by heterotrophic bacteria into microbial biomass. This microbial biomass could form aggregates that is called biofloc that can be consumed by the cultured organisms (De Schryver *et al*., 2008).

The biomass formed in biofloc system consist of various microorganism rich in those beneficial components, i.e. enzyme, protein, lipid, carotenoids, an amino sugar, and vitamin (Mabroke *et al.*, 2018). It is potentially used as an additional feed (Husain *et al*., 2014) and was reported to contribute to the growth performance of the cultured fish. Protein and lipid content in biofloc are considered sufficient to support fish growth, i.e. 24%–50% of protein (Ju *et al.*, 2008; López-Elías *et al.*, 2015) and 0.5%–3.5% of lipid (Kuhn *et al.*, 2009; Maicá *et al.*, 2012). Further, Ekasari *et al.* (2014) and Luo *et al.* (2014) reported that biofloc contributed to enhance the immunity of Pacific white shrimp and tilapia, respectively. In this regard, this study aimed to evaluate the health status and meat quality of African catfish reared in BFT and non-BFT.

MATERIALS AND METHODS

Experimental design

A completely randomized design was applied in this study. There were two biofloc treatments and a control with 4 replications per treatment:

BF500 : stocking density 500 ind/m^3 in biofloc system

BF700 : stocking density 700 ind/m^3 in biofloc system

K500 : stocking density 500 ind/m^3 in nonbiofloc system (control)

Experimental fish rearing

Seven days before the fish were reared, a commercial probiotic containing *Bacillus* sp. was inoculated to the rearing media at a dose of 2 g/m^3. The NH_4Cl was used as the N source and tapioca as the carbon source, using a C: N ratio of 15:1. The daily growth of bacteria was observed until the day before rearing started. The initial body length and bodyweight of the fish used in this experiment were 10–12 cm and 10–12g, respectively and it was reared for 45 days. Feeding frequency was twice a day using 5% of the total biomass (Yusuf *et al.,* 2015). Sampling was done every two weeks and so was the water quality assessment. The observed growth parameters were feed efficiency and daily growth rate. To calculate the later parameters, fish was weighed at the beginning and end of the study, then the average weight was determined.

Challenge test

A challenge test was conducted at the end of the study using 15 tested catfish for each treatment. *Aeromonas hydrophila* was incubated at room temperature for 2×24 hours. A 0.1 mL of the bacteria were injected in a concentration of 10^8 CFU/mL. This challenge test was managed for 14 days. Furthermore, mortality, relative percent survival (RPS), and blood profile after the challenge test were observed. Feeding was still done, except on day 7 and day 14. Apart from the other parameters mentioned before, mortality was calculated every day.

Experimental parameters

Health status

The health status of the tested fish was determined through blood profile changes. The blood profile test was conducted on day 0, day 15, day 30, day 45, and post-challenge test on day 60. The observed parameters were erythrocyte, hemoglobin, hematocrit, and leucocyte differentiation.

Organosensory assessment

Flesh quality was inspected using the organosensory assessment. The parameters consisted of texture, color, smell, and taste. This assessment was professionally done by panel members and laboratory specialists in a total of 30 people. The panel assessment result was tabulated and followed using quality assessment by comparing the result and the standard requirement in the score sheet (SNI 01-2346-2006).

Fish nutritional composition

The fish nutritional composition was assessed by the measurement of proximate composition and glycogen contents. Moisture measurement was done using an oven at a high temperature (110°C) for 6 hours. Kjeldahl and Folch methods were applied in measuring protein and lipid, respectively. Glycogen analysis was done by diluting the sample using 30% KOH, saturated Na_2SO_4, and 95% alcohol. The mixture was heated in a water bath (110°C). After that, it was precipitated. A 0.5 M of NaOH titration was done, then the mixture was added O-toluidine + CH_3COOH. The last step was done using a spectrophotometer in 635 nm of wavelength.

Water quality

The water quality parameters were measured according to the standard procedure by APHA (1998) consisted of temperature, dissolved oxygen (DO), pH, TAN, nitrite, nitrate, total suspended solids (TSS), floc volume (FV), and culturable bacteria density.

Data analysis

All data except meat quality by organosensory assessment was tabulated using Ms. Excell 2007 and the statistical analysis was done using SPSS 16 through analysis of variance in 95% of confidence level. Data homogeneity and normality were analyzed using Levene and Kolmogorov-Smirnov test. The significant results were analyzed using the posthoc Duncan test. Organoleptic assessment was analysed descriptively using scoring method.

RESULTS AND DISCUSSIONS

Results

Health status

On day 30 and 45, there were no significant differences in the blood profile result amongst treatment (P>0.05) (Table 1). However, particularly in day 15 post-challenge test, the leucocyte and haematocrit level was slightly higher in the biofloc treatments compared to that of the control.

Post challenged survival

After 14 days of challenge test, the fish survival in biofloc treatments were significantly higher compared to the K500 treatment (P<0.05) (Figure 1).

Meat quality

The result of the organosensory assessment showed that the average value of the eye, gill,

Table 1. Erythrocyte, leucocyte, hemoglobin (Hb), and hematocrit (Hc) African catfish flesh reared in a biofloc system and nonbiofloc system with different stock density (before and after challenge test).

Day−	Treatment	Erythrocyte ($\times10^6$ cell/mm^3)	Leucocyte ($\times10^4$ cell/mm^3)	Hb (g/dL)	Hc (%)
0	BF500	1.25 ± 0.08^a	7.73 ± 0.82^a	5.40 ± 0.67^a	21.20 ± 3.89^b
	BF700	1.43 ± 0.14^a	8.95 ± 1.79^a	6.10 ± 0.48^a	26.48 ± 5.16^{ab}
	K500	1.37 ± 0.25^a	6.25 ± 2.04^a	5.30 ± 1.32^a	17.63 ± 3.17^a
15	BF500	1.35 ± 0.19^a	8.09 ± 1.48^b	4.80 ± 0.99^a	26.60 ± 4.33^b
	BF700	1.64 ± 0.34^a	8.63 ± 0.30^b	5.45 ± 1.24^a	30.28 ± 4.67^b
	K500	1.33 ± 0.08^a	6.29 ± 0.83^a	4.60 ± 0.59^a	16.83 ± 5.53^a
30	BF500	1.60 ± 0.44^a	8.53 ± 1.17^a	6.90 ± 0.58^a	25.16 ± 1.62^a
	BF700	1.56 ± 0.14^a	8.01 ± 1.25^a	6.40 ± 0.16^a	25.68 ± 4.77^{ab}
	K500	1.66 ± 0.26^a	7.81 ± 0.97^a	8.25 ± 0.41^b	30.68 ± 2.18^b
45	BF500	1.66 ± 0.31^a	7.31 ± 0.27^a	6.00 ± 0.75^a	25.66 ± 2.94^a
	BF700	1.99 ± 0.14^a	8.22 ± 1.27^a	5.60 ± 0.16^a	26.30 ± 4.99^a
	K500	1.78 ± 0.11^a	6.98 ± 1.06^a	5.60 ± 0.63^a	21.46 ± 4.27^a
60 (after challenge test)	BF500	1.21 ± 0.22^a	8.44 ± 0.63^a	5.60 ± 1.64^b	16.03 ± 2.81^a
	BF700	1.37 ± 0.09^a	8.69 ± 0.61^a	6.95 ± 0.81^{ab}	25.77 ± 3.46^b
	K500	1.15 ± 0.14^a	7.83 ± 0.44^a	4.95 ± 0.82^a	17.33 ± 2.61^a

Note: *Different superscript in the same column indicates significant difference (P<0.05). BF500 = stock density 500 ind/m^3 in biofloc system; BF700 = stock density 700 ind/m^3 in biofloc system; K500 = stock density 500 ind/m^3 in non biofloc system (control).

body mucus, smell, appearance, taste, and texture of the sample was higher in the BF500 and BF700 treatment. The result ranged from 7–9. On the contrary, the K500 treatment showed lower assessment results which ranged from 6–7.

Fish protein

The protein level retained in the fish in the biofloc treatment was higher and significantly different from K500 ($P<0.05$). On the other hand, the fat level of K500 was higher than those of biofloc treatments. The result of the nitrogen-free extract showed a significant difference ($P<0.05$). However, the ash level did not show any significant difference amongst treatments ($P>0.05$).

Growth performance

The African catfish reared in the biofloc system presented a higher result compared to the kontrol (K500) (Figure 2). Unlike the growth result, the feed conversion ratio in the biofloc system was lower (Figure 3).

Water quality

Water quality during the rearing period were shown below in Table 4. There were no significant differences amongst treatment. The dominant bacteria density was also identified and counted. *Kurthia* sp., *Enterobacteria* sp., and *Bacillus* sp. were primarily predominantly found in the biofloc treatment. Meanwhile, *Kurthia* sp., and *Bacillus* sp. were considered to be minor in the control treatment.

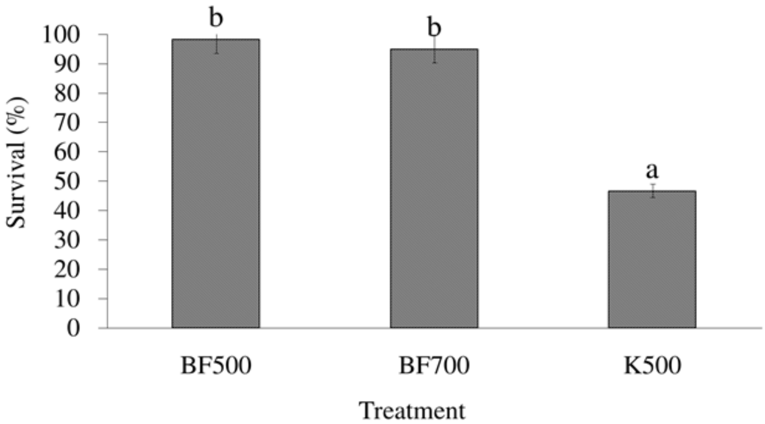

Figure 1. The survival rate of African catfish reared in a biofloc and nonbiofloc system with different stock densities before and after the challenge test towards *A. hydrophila*. The different superscript indicates a significant difference ($P<0.05$). BF500 = stock density 500 ind/m^3 in biofloc system; BF700 = stock density 700 ind/m^3 in biofloc system; K500 = stock density 500 ind/m^3 in non biofloc system (control).

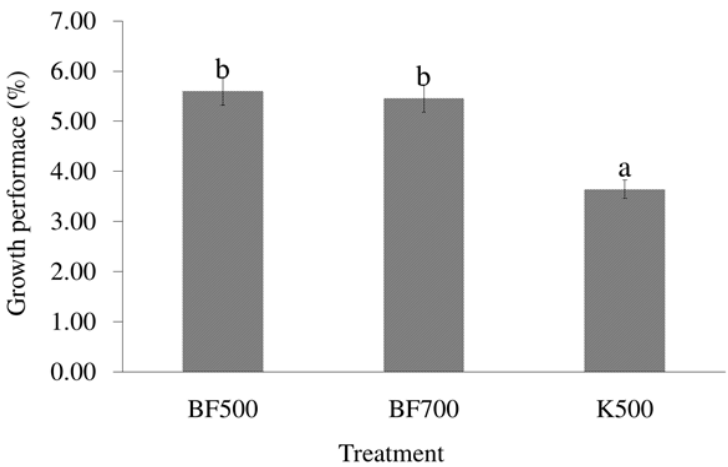

Figure 2. Growth performance of African catfish reared in a biofloc and nonbiofloc with different stock density. The different superscript indicates a significant difference ($P<0.05$). BF500 = stock density 500 ind/m^3 in biofloc system; BF700 = stock density 700 ind/m^3 in biofloc system; K500 = stock density 500 ind/m^3 in non biofloc system (control).

Table 2. Organosensory analysis of African catfish reared in a biofloc system and nonbiofloc system with different stock density (fresh meat and after being steamed).

Parameter	BF500	BF700	K500
Fresh			
Eye	9	9	7
Gill	9	9	7
Mucus	9	9	6
Meat	8	8	7
Smell	8	8	6
Texture	8	8	6
Steamed			
Appearance	7	7	6
Scent	8	9	7
Taste	8	8	6
Texture	8	8	6
Colour	8	8	6

Note: BF500 = stock density 500 ind/m^3 in biofloc system; BF700 = stock density 700 ind/m^3 in biofloc system; K500 = stock density 500 ind/m^3 in non biofloc system (control). Assessment criteria 1-9.

Table 3. Meat proximate composition of African catfish reared in a biofloc system and nonbiofloc system with different stock density.

Treatment	Proximate composition (% dry weight)				
	Ash	Protein	Lipid	Crude fiber	NFE
BF500	14.53 ± 0.78^a	59.13 ± 0.71^b	19.14 ± 0.47^a	3.22 ± 0.53^a	6.44 ± 0.66^a
BF700	14.26 ± 0.60^a	58.91 ± 0.55^b	19.27 ± 0.65^a	2.83 ± 0.46^a	4.58 ± 0.33^b
K500	14.93 ± 0.55^a	53.94 ± 0.46^a	21.09 ± 0.40^b	4.56 ± 0.13^b	8.78 ± 0.43^c

Note: Different superscript indicates significant difference (P<0.05). BF700 = stock density 700 ind/m^3 in biofloc system; K500 = stock density 500 ind/m^3 in non biofloc system (control); NFE (Nitrogen-free extract)

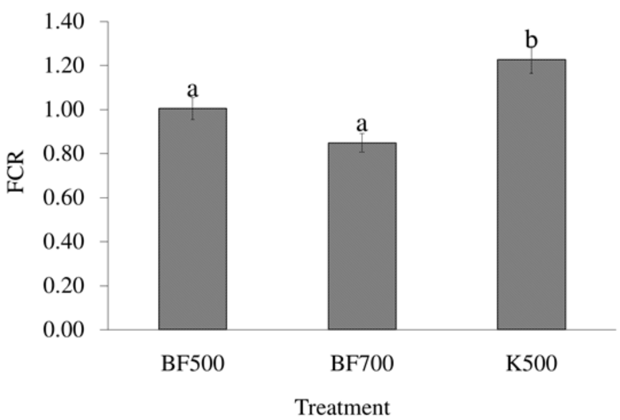

Figure 3. The feed conversion ratio of African catfish reared in a biofloc and nonbiofloc with different stock density. The different superscript indicates a significant difference (P<0.05). BF500 = stock density 500 ind/m^3 in biofloc system; BF700 = stock density 700 ind/m^3 in biofloc system; K500 = stock density 500 ind/m^3 in non biofloc system (control).

Discussion

The health status of K500 treatment during a 45-day of rearing was considered normal. Bakhshi *et al.* (2018) stated that the erythrocyte and leucocyte levels of catfish reared in a biofloc system were 0.99–0.1×10^6 cell/mm^3 and 5.9–6.4×10^3 cell/mm^3, respectively. The hemoglobin level was at a normal level both in kontrol and biofloc treatment. After the challenge test, the hemoglobin level in the control treatment decreased, while the biolfoc treatment remained normal compared to the previous study by Hastuti and Subandiyono (2018).

In general, the blood profile of the African catfish presented that those which reared in biofloc treatment potentially more resistant to disease. It is confirmed by the challenge test using *A. hydrophila.* The survival rate after the challenge test was 98.32% and 95% for both densities, respectively. Various biofloc studies showed similar results. Aquaculture technology based on biofloc was able to improve immunity in shrimp (Ekasari *et al.*, 2014; Xu & Pan, 2014). Ekasari *et al*. (2014) explained that bacteria found in biofloc similarly functioned as probiotics. The poly-β-hydroxybutyrate content contributed to the fish immunity system hence it promoted greater immunity towards pathogens (Aalimahmoudi & Azarm, 2017). The availability of protozoa, rotifer, and heterotrophic bacteria also contributed to the high survival rate in the biofloc system since it lowered cannibalism amongst individuals

Table 4. Water quality of African catfish rearing in a biofloc system and nonbiofloc system with different stock density.

Parameter	BF500	BF700	K500
DO (mg/L)	4.95−5.68	5.15−5.85	5.65−6.35
pH	6.55−7.92	6.64−7.83	7.26−7.31
Temperature (^{o}C)	28.5−29.0	28.2−28.8	27.8−28.3
TAN (mg/L)	0.15−0.81	0.16−0.82	0.16−0.84
Nitrite (mg/L)	0,.28−0.81	0.26−0.81	0.34−0.88
Nitrate (mg/L)	0.33−0.93	0.31−0.88	0.46−0.93
TSS (mg/L)	367−715	364−733	215−660
Floc volume (mL/L)	70−79	71−80	−

Note : BF500 = stock density 500 ind/m^3 in biofloc system; BF700 = stock density 700 ind/m^3 in biofloc system; K500 = stock density 500 ind/m^3 in non biofloc system (control). DO = dissolved oxygen, TAN = total amonia nitrogen, TSS= total suspended solids.

Table 5. Bacteria composition in the biofloc and nonbiofloc system.

Treatment	Bacteria genera	Bacteria colony (CFU/mL)
BF500	*Kurthia* sp., *Plesiomonas* sp.	3.50×10^6
	Enterobacteria sp.	1.49×10^6
	Bacillus sp.	9.52×10^6
BF700	*Enterobacteria* sp.	4.34×10^6
	Bacillus sp.	5.54×10^6
	Kurthia sp.	4.47×10^6
K500	*Enterobacteria* sp.	1.05×10^5
	Micrococcus sp.	0.91×10^5
	Campylobacter sp.	1.18×10^5

Note: BF500 = stock density 500 ind/m^3 in biofloc system; BF700 = stock density 700 ind/m^3 in biofloc system; K500 = stock density 500 ind/m^3 in non biofloc system (control).

(Apriyani *et al.*, 2016). Biofloc is also able to boost antioxidant activity because of the carotenoids, vitamin C, and essential fatty acids content (Xu & Pan, 2012).

The fish proximate composition of the BF500 treatment were 59.13% of protein, 19.27% of lipid, 6.44% of NFE, 14.53% of ash, and 3.22% of crude fiber. Those results complied with the past study by Salamah (2014). Yusuf *et al.* (2015) also stated that the protein and lipid contents were around 59.04% and 21.98%, respectively. Protein content in the catfish flesh was fairly high. It explained that the biofloc system provided a protein source to support metabolism and health status.

Water quality parameters in the biofloc treatment presented a better result to maintain a decent environment. Nitrite and nitrate were lower compared to K500 treatment. *Bacillus* sp. in the biofloc treatment diminished excess ammonia to form a protein. Thus it eliminated toxic substances and formed nutrients at once (VanWyk & Avnimeleh, 2007). Dissolved oxygen in all treatments ranged from 4.95–6.35 and is considered normal. It is supported by Schveitzer *et al.* (2013) who stated that the DO content in biofloc systems ranged from 4.2–5.9 mg/L. The pH level varied between 6.55–7.92. According to Ray *et al.* (2011), pH could vary between 6.3–8.5. Total ammonia nitrogen (TAN) result was 0.15–0.84 mg/L. A decrease in TAN is caused by nitrogen conversion by heterotrophic bacteria (Martinez-Porchasa *et al.*, 2020). Total suspended solids (TSS) ranged from 215–732.5 mg/L. De Schryver *et al.* (2008) stated that the recommended range of TSS was 200–1000 mg/L. Sumitro (2017) described that the maximum floc volume in catfish rearing was 80 ml/L. The floc volume in this study which varied between 70–80 mg/L was in line with the Sumitro (2017).

Organosensory analysis observed the appearance of eye, gill, body mucus, smell, appearance, taste, and texture. The results showed that the average score of the biofloc treatment varied between 7–9. Meanwhile, the nonbiofloc treatment presented a lower score which ranged from 6–7. Referring to SNI 01-2729.1.2006, the minimal score to fulfill the quality and food safety standard of organoleptic analysis was 7. A lower score will be categorized as unsafe (Nurjanah, 2011). The microbial activity also contributed to the flesh quality changes (Wijayanti & Lukitasari, 2008). Probiotic in biofloc system was assumed to prevent decomposition process. Thus the appearance of fish reared in the biofloc system was better than the one reared in the nonbiofloc system. Wibowo *et al.* (2014) stated that texture change was characterized by a dented area around the body surface after being pressed. Meat softening happens because protein decomposition becomes smaller molecules such as polypeptides, amino acids, and ammonia. Biofloc application was assumed to slower protein decomposition so that the quality was well-maintained.

Based on the bacteria abundance analysis, *Bacillus* sp. dominated the population. As many of 9.52×10^6 CFU/mL was counted in the BF500, while 5.54×10^6 CFU/mL was detected in the BG700. It was supported by Yusuf *et al.* (2015) who stated that *Bacillus* sp. abundance in biofloc system varied between 10^6–10^8 CFU/mL. *Bacillus* sp. is known for its beneficial activity in the digestive tract. It releases the exogenous enzyme to support expenditure energy (Ogello *et al.*, 2014). The energy gap from the previous mechanism will be utilized to support growth.

Biofloc is abundant in various bioactive molecules, such as carotenoids, chlorophyll, phytosterol, bromophenols, and amino sugar (Ju *et al.,* 2008). Biofloc recycles excess waste to maintain the C/N ratio and induces heterotrophic bacteria in converting ammonia into microbe biomass (Ogello *et al.*, 2014). It indicates that biofloc can be one of a nutrition source to support fish growth. Biofloc also enhances feed efficiency by the declining feed conversion ratio. *Bacillus* sp.. as biofloc former, contributes significant growth in catfish rearing and improve feed efficiency. Therefore, feed utilization could be saved up to 10–20% (De Schryver *et al.,* 2008).

CONCLUSION

The application of biofloc system could support the health status and improve meat quality of African catfish.

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