

# Effects of Replacement Fishmeal with Biofloc on Feed Utilization and Growth Performance, Morphological and Chemical Characteristics of Red Tilapia

## ABDELWAHAB MOHAMMED ABDELWAHAB<sup>1,2\*</sup>, AHMED ALMADANI<sup>1</sup>, IBRAHIM ALMEHSEN<sup>1</sup>

<sup>1</sup>Department Animal and Fish Production, College of Agriculture and Food Science, King Faisal University, Kingdom of Saudi Aarabia; <sup>2</sup>Department Animal Production, Faculty of Agriculture, Fayoum University, Egypt.

Abstract | Biofloc is the aggregates of living and dead particulate organic matter, phytoplankton, bacteria and grazers of the bacteria, which is suspended in ponds and tanks. The current study aimed to determine feed utilization, growth performance, morphological and chemical body features, and blood profile upon dietary biofloc feeding to red Tilapia fingerlings. The diets were control and biofloc (00.0%, 33.0%, 66.0%, and 100.0%). The diets were fed to red Tilapia fingerlings (14.3  $\pm$  0.15 g) thrice a day. The study lasted fourteen weeks. Feed utilization, growth performance, morphological fish characters, chemical fish composition, flesh color, and blood profiles were determined. The obtained results illustrated that fishmeal replacement with 33.0% biofloc resulted in the nearest results if compared to the control diet in feed utilization, growth performance, morphological fish characters, chemical 33.0% of biofloc gave a comparable effect in protein and energy productive values compared to the control diet versus lower values (p<0.05) of 66.0% and 100.0%% biofloc diets. Furthermore, the biofloc diets (66.0 and 100.0%) almost decreased in all the recorded parameters compared to biofloc (33.0%) and control diets except body fat content and flesh color. Therefore, it might be concluded that feeding biofloc up to 33.0% could be promising in feed utilization, growth performance, morphological fish characters, chemical fish composition, and blood profiles.

Keywords | Biofloc, Feed intake, Growth, Morphological, Chemical, Flesh color

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\*Correspondence | Abdelwahab Mohammed Abdelwahab, Department Animal and Fish Production, College of Agriculture and Food Science, King Faisal University, Kingdom of Saudi Aarabia; Email: amsaid@kfu.edu.sa

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# **INTRODUCTION**

A quaculture is considered one of the most important and fastest-growing fields in the production of protein for human consumption and has become one of the most successful industries and activities worldwide (Ekasari et al., 2023). The demand for these products has increased steadily, along with the increase in the demand for feed. It is known that protein is one of the main components of feed for aquatic organisms such as fish and crustaceans

(Abdelwahab et al., 2020). Fish feed is considered more than half of the costs of the fish farming project. Fishmeal is considered one of the most important sources of protein for aquatic organisms. Therefore, replacing fishmeal with an alternative cheap protein source is necessitated to face the increasing demand for fish feed as biofloc and other supplements (Almadani, 2017).

Biofloc technology is considered to be as new blue revolution since nutrients can be continuously recycled and reused in

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the culture medium. Biofloc is the aggregates of living and dead particulate organic matter, phytoplankton, bacteria and grazers of the bacteria, which is suspended in ponds and tanks. Biofloc was used as the replacement for fishmeal in several studies (Xu and Pan, 2012). Different proportions of biofloc (10.0 - 100.0%) were fed to different fish species (Bauer et al., 2012; Valle et al., 2014). Several studies have reported that biofloc helps the growth performance of saltwater shrimp species including Penaeus monodon (Shyne Anand et al., 2013), Litopenaeus vannamei (Wasielesky et al., 2006), Farfantepenaeus paulensis (Ballester et al., 2010), Marsupenaeus japonicas (Zhao et al., 2012). Biofloc diets were fed to fish species because of high-quality protein, growth stimulants, and prebiotics content (Ju et al., 2008), which in turn stimulate digestive enzymes and improve health status (Singh et al., 2005; Xu and Pan, 2012). It was concluded in some studies that the biomass of biofloc is probiotics (Bairagi et al., 2002, 2004). On the other hand, feed utilization, growth performance, meat quality, and cost-effectiveness were investigated with 100g red Tilapia cultured in different biofloc systems for 42 days. The result indicated that the production performance of red tilapia was lower with biofloc whereas the fish quality was similar with other treatments (Ekasari et al., 2023).

Fish flesh color is the first parameter evaluated by consumers, and is therefore an important parameter quality relevant to market acceptance (Tareq et al., 2022). Therefore, the effect of biofloc on flesh color was evaluated in addition to blood indicative health status (red blood cells, hematocrit, glucose, and total protein) of red Tilapia (Almadani, 2017). Therefore, the current study aimed to investigate feed utilization, growth performance, morphological fish characters, chemical fish composition, and blood profiles of red Tilapia upon feeding biofloc (33.0, 66.0, and 100.0%).

## **MATERIALS AND METHODS**

The study were carried out in the lab of Animal and Fish Production Department, College of Agriculture and Food Sciences, King Faisal University. Ethics of the scientific research deanship committee of King Faisal University were followed in the current study (Ref. No. KFU-REC-2022-JUN-EA002289). The fishmeal in the study was replaced with 33.0, 66.0 and 100.0% biofloc powder.

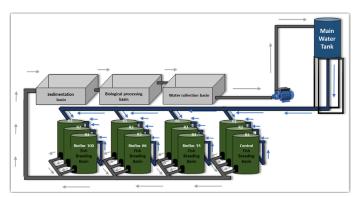
### $\ensuremath{\mathsf{Site}}$ of study and culture management system

The culture system includes 12 basins (three replicates/ treatment) connected together (Figure 1). The basin is made of fiberglass with a size of 1.0 square meter. Ethics of the scientific research deanship committee of King Faisal University were followed in the current study (Ref. No. KFU-REC-2022-JUN-EA002289). The culture system is a closed system enabled to replace water regularly (Aquatic Eco-Systems, Inc. Apopka, Florida 32704 USA). The

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culture controlled system was kept under a cycle of 12.0 h light and 12.0 h dark in addition to 28.0 °C temperature. The red tilapia fingerlings (14.33±0.10 g) were adapted in the culture system for two weeks. The adapted fingerlings were distributed through complete random design to four groups; control diet and three biofloc diet groups (33.0, 66.0 and 100.0%). Ingredients of control and bioflocformulated diets and chemical composition used during the study were shown in Table 1. The biofloc powder were obtained from the College of Marine Sciences, King Abdulaziz University, Jeddah. The biofloc powder were mixed with the diet in proportions 33.0, 66.0 and 100.0%. The study lasted fourteen weeks. Feed utilization, growth performance, morphological fish characters, chemical fish composition, flesh color, and blood profiles were determined in the current study.



**Figure 1:** Schematic diagram of study experimental design R1, R2, R3; numbers of replicates per treatment.

Table 1: Ingredients (g/kg) and chemical composition of	
control and Biofloc formulated diets.	

Parameters	Treatments				
	Control	33.0%	66.0%	100.0%	
Fish meal	169.6	113.6	57.6	0.00	
Soybean	280.8	301.0	323.8	330.8	
Corn gluten	100.0	100.0	100.0	100.0	
Yellow corn	210.0	200.0	200.0	130.5	
Wheat bran	150.0	91.8	21.3	30.0	
Biofloc powder	0.00	90.6	181.3	274.7	
Fish oil	64.6	78.0	91.0	109.0	
Dicalcium Phosphate	5.0	5.0	5.0	5.0	
Minerals and vitamins	5.0	5.0	5.0	5.0	
Sodium chloride	10.0	10.0	10.0	10.0	
Limestone	5.0	5.0	5.0	5.0	
Total	100	1000	1000	1000	
Chemical composition, 9	6				
Dry matter	93.79	94.02	94.23	94.56	
Crude protein	34.0	34.0	34.0	34.0	
Fat	10.5	11.2	11.9	13.1	
Crude fiber	3.40	4.07	4.72	5.65	
Nitrogen free extract	38.80	37.03	35.33	32.75	
Ash	7.11	7.70	8.24	9.08	
Energy	4.50	4.50	4.50	4.50	

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**Table 2:** Effects of Biofloc on growth performance of Red Tilapia.

Parameters		Treatments			
	Control	33.0%	66.0%	100.0%	
Initial body weight (g/kg)	12.41±0.16	12.40±0.18	12.47±0.13	12.34±0.14	
Final body weight (g/kg)	41.40±0.07ª	$37.35 \pm 0.06^{b}$	32.66±0.11°	30.17±0.03°	
Body weight gain (g/fish)	28.99±0.14ª	$24.94 \pm 0.15^{b}$	20.19±0.16°	17.83±0.13°	
Body weight gain, %	233.60±3.09ª	$201.01 \pm 4.15^{b}$	161.91±2.75°	144.50±3.67°	
Daily body weight gain (g/fish)	$0.35 \pm 0.002^{a}$	$0.30 \pm 0.002^{b}$	$0.24 \pm 0.002^{\circ}$	$0.21 \pm 0.004^{\circ}$	
Specific growth rate, %	$1.43 \pm 0.01^{a}$	$1.31 \pm 0.02^{b}$	1.15±0.01°	1.06±0.45°	
Survival rate, %	91.11±1.92ª	85.56±5.09 <sup>ab</sup>	$80.00 \pm 5.77^{ab}$	77.78±5.18 <sup>b</sup>	

a, b, c; Values with different superscripts between biofloc and control groups significantly differed at < 0.05.

Table 3: Effects of biofloc on feed efficiency of Red Tilapia.

Treatments				
Control	33.0%	66.0%	100.0%	
43.97±0.68ª	$40.17 \pm 1.04^{b}$	38.10±0.50°	$36.02 \pm 0.29^{d}$	
41.24±0.64ª	$37.67 \pm 0.98^{b}$	35.74±0.48°	$33.79 \pm 0.27^{d}$	
$2.22\pm0.0^{3a}$	$2.19 \pm 0.05^{b}$	2.30±0.02°	$2.30\pm0.02^{d}$	
65.94±1.05ª	$62.13 \pm 1.94^{b}$	$52.99 \pm 1.10^{b}$	$49.50 \pm 0.50^{\rm b}$	
$1.42 \pm 0.02^{b}$	1.51±0.05ª	1.77±0.04ª	1.89±0.02ª	
1.94±0.03ª	1.83±0.06 <sup>ab</sup>	$1.56 \pm 0.03^{b}$	$1.47 \pm 0.01^{b}$	
29.28±1.32ª	24.80±1.67 <sup>ab</sup>	$20.51 \pm 2.60^{b}$	$19.31 \pm 1.17^{\text{b}}$	
$14.65 \pm 0.23^{a}$	13.83±0.43 <sup>ab</sup>	$11.84 \pm 0.25^{b}$	$11.09\pm0.11^{\mathrm{b}}$	
$22.27 \pm 0.74^{a}$	22.31±0.50ª	17.82±1.29 <sup>b</sup>	$18.95 \pm 0.58^{\text{b}}$	
	$\begin{array}{c} 43.97 \pm 0.68^{a} \\ 41.24 \pm 0.64^{a} \\ 2.22 \pm 0.0^{3a} \\ 65.94 \pm 1.05^{a} \\ 1.42 \pm 0.02^{b} \\ \end{array}$ $\begin{array}{c} 1.94 \pm 0.03^{a} \\ 29.28 \pm 1.32^{a} \\ \end{array}$ $14.65 \pm 0.23^{a} \\ 22.27 \pm 0.74^{a} \end{array}$	Control33.0% $43.97\pm0.68^{a}$ $40.17\pm1.04^{b}$ $41.24\pm0.64^{a}$ $37.67\pm0.98^{b}$ $2.22\pm0.0^{3a}$ $2.19\pm0.05^{b}$ $65.94\pm1.05^{a}$ $62.13\pm1.94^{b}$ $1.42\pm0.02^{b}$ $1.51\pm0.05^{a}$ $1.94\pm0.03^{a}$ $1.83\pm0.06^{ab}$ $29.28\pm1.32^{a}$ $24.80\pm1.67^{ab}$ $14.65\pm0.23^{a}$ $13.83\pm0.43^{ab}$ $22.27\pm0.74^{a}$ $22.31\pm0.50^{a}$	Control33.0%66.0% $43.97\pm0.68^a$ $40.17\pm1.04^b$ $38.10\pm0.50^c$ $41.24\pm0.64^a$ $37.67\pm0.98^b$ $35.74\pm0.48^c$ $2.22\pm0.0^{3a}$ $2.19\pm0.05^b$ $2.30\pm0.02^c$ $65.94\pm1.05^a$ $62.13\pm1.94^b$ $52.99\pm1.10^b$ $1.42\pm0.02^b$ $1.51\pm0.05^a$ $1.77\pm0.04^a$ $1.94\pm0.03^a$ $1.83\pm0.06^{ab}$ $1.56\pm0.03^b$ $29.28\pm1.32^a$ $24.80\pm1.67^{ab}$ $20.51\pm2.60^b$ $14.65\pm0.23^a$ $13.83\pm0.43^{ab}$ $11.84\pm0.25^b$	

a, b, c, d; Values with different superscripts between biofloc and control groups significantly differed at < 0.05.

#### FEED UTILIZATION AND GROWTH PERFORMANCE

The red tilapia fingerlings of control and biofloc groups were fed daily at 7:00, 11:00 a.m., and 2:30 p.m. Survival rate (%) is calculated; survival rate= final # of fish/ initial # of fish \* 100. Body weight and feed intake were biweekly recorded using digital balance (Trooper China). Feed intake (g/fish) is the amount of feed given during the experimental period/fish (g). Fish were anesthetized and dried to record body weight. Weight gain (WG g) is calculated using the following equation WG g = Wf - Wi. Specific growth rate (DGR) is calculated according to the formula; DGR = (Wf - Wi). t-1, where Wf and Wi are the final and the initial body weight, and t - is the duration of the experimental period. Dividing feed intake to body weight gain is done to calculate feed efficiency. Protein efficiency, protein productive value, energy efficiency, energy productive value were calculated to Nose (1971) and El-Dahhar et al. (2016).

### DIETS AND FISH SAMPLES FOR CHEMICAL ANALYSIS

Diets and fish samples of control and biofloc groups were dried in an oven at 70°C. Thereafter, the samples were ground and chemically analyzed for the determination of a

dry matter, organic matter, crude protein, crude fibers, and ether extract values (Tables 1 and 4) (AOAC, 2005).

### **COLLECTION AND ANALYSIS OF BLOOD SAMPLES**

One blood sample were collected from control and biofloc groups from the caudal vein of four fish at the end of the study. The determined hematological values were red blood cells, hematocrit, total protein, and glucose (King, 2012; Almadani, 2017; Mohammed et al., 2018; Abdelwahab et al., 2020).

#### COLOR OF FISH FLESH MINCES MEASUREMENT

The color of fish flesh minces of control and biofloc groups were determined using Hunter (Almadani, 2017; Abdelwahab et al., 2020). Mince color examination of L, a , and b values are used to determine color of fish flesh minces of control and biofloc groups.

### **S**TATISTICAL ANALYSIS

Data were statistically analyzed using the general linear model of the SAS Program (2008). Comparison among groups of control and biofloc were done for feed efficiency, body weight, morphological and chemical composition

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characteristics, flesh mince color, and blood values using the Duncan test. The statistical model:

$$Yij = \mu + Ti + Eij$$

Where; Yij= the experimental observation ij;  $\mu$  =the overall mean; Ti= the effect due to control or biofloc diets; Eij= the experimental error.

# **RESULTS AND DISCUSSION**

The results of the current study represent diets and biofloc

**Table 4:** Effects of Biofloc on morphological characteristics of Red Tilapia.

chemical composition and their feeding effects on feed efficiency and growth performance, morphological and chemical body features, and blood profiles of red Tilapia (Tables 1-7). The chemical composition of diets and biofloc is presented in Table 1. Biofloc contain protein (34.83%), crude fiber (5.65%), fat (13.10%), ether extract (32.75%) and ash (9.08%) as indicated in other studies (Khanjani et al., 2023; Gullian-Klanian et al., 2023). The results indicated the higher the level of biofloc in the diet of fish, the lower feed efficiency and growth performance, morphological and chemical body features, and blood profiles.

Parameters	Treatments			
	Control	33.0%	66.0%	100.0%
Total length, cm	11.55±0.87	11.57±0.77	11.16±0.65	11.09±0.54
Standard length, cm	9.44±0.75	9.56±0.69	9.21±0.54	9.16±0.45
Body depth, cm	3.21±0.19	3.23±0.22	3.08±0.15	3.03±0.15
Body thickness,	1.64±0.16	1.65±0.13	1.61±0.13	1.57±0.12
Conditional factor	1.63±0.18	1.62±0.22	1.56±0.28	1.57±0.15

## **Table 5:** Effects of Biofloc on chemical body composition of Red Tilapia.

Parameters	Treatments				
	Control	33.0%	66.0%	100.0%	
Moisture, %	72.43±0.42	72.14±0.98	73.07±0.32	71.87±0.91	
Dry matter, %	27.57±0.98	27.86±0.32	26.93±0.91	28.13±0.27	
Protein, %	15.34±0.63	14.31±0.34	14.16±1.00	14.27±0.52	
Fat,%	7.42±0.18 <sup>c</sup>	$8.72 \pm 0.36^{bc}$	$8.12 \pm 0.39^{ab}$	9.39±0.40ª	
Ash, %	$4.82 \pm 0.25$	4.83±0.12	4.65±0.26	4.48±0.07	
Energy, Calorie	$1.57 \pm 0.05$	1.63±0.02	$1.57 \pm 0.05$	1.69±0.02	
Energy, KJole	6.50±0.21	6.77±0.09	6.50±0.22	7.02±0.09	

a, b, c; Values with different superscripts between biofloc and control groups significantly differed at < 0.05.

Table 6: Effects of Biofloc on blood parameters of Red Tilapia.

Parameters	Treatments				
	Control 33.0%		66.0%	100.0%	
Red blood cell, 10 <sup>6</sup>	2.75±0.28	2.51±0.19	2.87±0.19	2.63±0.16	
Packed cell volume, %	29.83±3.25	27.40±2.16	30.73±2.19	29.00±3.00	
Glucose, mg/100 ml	46.67±5.51	47.00±1.73	42.33±7.57	44.33±7.37	
Total protein, g/100 ml	4.30±0.44	4.40±1.06	4.53±1.14	4.87±0.46	

## Table 7: Effects of biofloc on flesh coloration of Red Tilapia.

Parameters	Treatments	Treatments					
	Control	33.0%	66.0%	100.0%			
Lightness	57.61±1.35	55.64±1.52	55.94±2.39	57.43±2.71			
Redness	$6.90 \pm 0.78^{b}$	$7.20 \pm 1.68^{ab}$	8.23±2.42ª	8.58±1.18ª			
Yellowness	$16.64 \pm 0.40^{ab}$	15.53±0.65 <sup>b</sup>	17.09±1.69ª	17.95±1.01ª			
ab T7 1 1.1.0°		1 . 0 1 . 1	· · · · · · · · · · · · · · · · · · ·	0.05			

 $^{a,b}$ ; Values with different superscripts between biofloc and control groups significantly differed at < 0.05.

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### FEED EFFICIENCY AND GROWTH PERFORMANCE Feed efficiency and growth performance of red Tilapia fingerlings due to replacement 33.0, 66.0, and 100.0% biofloc diets were represented in Tables 2-3. The results revealed a decrease (p<0.05) in feed intake, feed efficiency, and growth performance values of biofloc groups if compared to control one. Biofloc diet (33.0%) group gave similar results in protein and energy productive values if compared to the control group whereas 66.0 and 100.0% biofloc diets decreased the aforementioned parameters. Feed utilization and growth performance, meat quality, and cost-effectiveness were investigated with 100g red Tilapia cultured in biofloc system. The result revealed that biofloc decreased the production performance whereas the fish quality was similar to control (Ekasari et al., 2023). The negative effects of 33.0%, 66.0%, and 100.0% biofloc diets compared to the control diet might be owing to palatability problems (Walker and Berlinsky, 2011) in addition to imbalances in diet components. On the other hand, several studies indicated higher feed intake, feed efficiency, and growth performance due to biofloc feeding (Mahanand et al., 2013; Shyne et al., 2014). The differences compared with our study might be attributed to the culture conditions, diet formulation, and the percentage of biofloc and composition.

# MORPHOLOGICAL AND CHEMICAL COMPOSITION CHARACTERISTICS

Biofloc diets (33.0, 66.0, and 100.0%) gave similar results as the control diet in the morphological characters concerning total length, body depth and thickness (Table 4). The chemical composition characters (moisture, dry matter, protein, fat, ash and energy) of biofloc (33.0, 66.0, and 100.0%) and control diet groups were shown in Table 5. Similar results were obtained in values of dry matter, protein, ash and energy among biofloc and control groups, whereas 100.0% biofloc diet gave the highest values of fat (p<0.05) compared to other groups. This could be owing to the diet fat contents, which were respectively 11.20%, 11.90%, 13.10%, and 10.50% of 33.0%, 66.0%, 100.0% biofloc, and control diets.

## **B**LOOD PROFILES

Blood profiles (red blood cells, hematocrit, glucose, and total protein) of 33.0, 66.0, and 100.0% biofloc and control groups were represented in Table 6. The results showed that replacement biofloc (33.0, 66.0, and 100.0%) did not give any change in blood values when compared to control diet. This is an indicative of biofloc effects on fish health.

## Color of fish flesh minces

Colors of flesh minces of biofloc and control groups were represented in Table 7. The result revealed that biofloc diets (66.0% and 100.0%) caused a change (p<0.05) in term

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of redness and yellowness flesh minces. The significant improvement of fish flesh color due to 66.0 and 100.0% biofloc diets compared to control and 33.0% biofloc diets were obtained in this study as indicated in earlier studies (Hende et al., 2014), which might be owing to pigment content in biofloc. Biofloc pigment is attributed to the presence of algae in biofloc contents (Venkataraman and Becker, 1985). The alage contain pigments as carotenoids and chlorophylls (Roy and Ruma, 2014), which constitute about 3–5 % of the dry algae biomass (Venkataraman and Becker, 1985).

# CONCLUSIONS

Biofloc inclusion in the fingerlings red Tilapia diet could be promising concerning feed efficiency and growth performances, chemical body composition at level 33.0% or low due to the adverse effects of the higher levels.

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## NOVELTY STATEMENT

Biofloc inclusion in the fingerlings red Tilapia diet could be promising at level 33.0% or low due to the adverse effects of the higher levels.

# **AUTHOR'S CONTRIBUTION**

Authors contribute equally in Conceptualization, writing and editing manuscript. All authors have read and agreed to the published version of the manuscript.

## **CONFLICTS OF INTEREST**

The authors have declared no conflict of interest.

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