

http://www.periodicos.uem.br/ojs/ ISSN on-line: 1807-863X

Doi: 10.4025/actascibiolsci.v45i1.64475



**BIOTECHNOLOGY** 

# Lipid profile and metabolic parameters of tilapia in the finishing phase in earth ponds or using biofloc technology

Emerson Giuliani Durigon<sup>1</sup>, Juliano Uczay<sup>2</sup>, Nilce Coelho Peixoto<sup>3</sup>, Samuel Marasca<sup>1</sup>, Thamara Luísa Staudt Schneider<sup>1</sup>, Luana Haselein Maurer<sup>4</sup>, Tatiana Emanuelli<sup>5</sup> and Rafael Lazzari<sup>1,2</sup>

<sup>1</sup>Programa de Pós-Graduação em Zootecnia, Universidade Federal de Santa Maria, Av. Roraima, 1000, 97105900, Bairro Camobi, Santa Maria, Rio Grande do Sul, Brazil. <sup>2</sup>Programa de Pós-Graduação em Agronegócios, Departamento de Zootecnia e Ciências Biológicas, Universidade Federal de Santa Maria, Campus Palmeira das Missões, Palmeira das Missões, Rio Grande do Sul, Brazil. <sup>3</sup>Departamento de Ciências da Saúde, Universidade Federal de Santa Maria, Campus Palmeira das Missões, Palmeira das Missões, Rio Grande do Sul, Brazil. <sup>4</sup>Instituto Federal de Educação, Ciência e Tecnologia Farroupilha, Alegrete, Rio Grande do Sul, Brazil. <sup>5</sup>Centro Integrado de Desenvolvimento de Análises Laboratoriais, Departamento de Tecnologia e Ciência de Alimentos, Centro de Ciências Rurais, Universidade Federal de Santa Maria, Santa Maria, Rio Grande do Sul, Brazil. \*Author for correspondence. E-mail: emersom.durigom@gmail.com

**ABSTRACT.** Tilapia is a species with great growth potential. Its production comes from a semi-intensive system, such as earthen ponds (EP). Recently, biofloc technology (BFT) appears as an option to intensify fish production. The objective of this work was to compare the organosomatic indices, biochemical parameters, and chemical composition of tilapia reared in EP and BFT. Fish were grown for 150 days, with an initial weight of  $\cong 2$  g and a final weight of  $\cong 780$  g. Thereafter, tissues and organs were collected to determine organosomatic indices and analyze biochemical parameters, fatty acid, and proximate composition. The carcass yield was higher for tilapia reared in EP than BFT. The production system did not affect the fish fillet yield. The other organosomatic parameters were higher for tilapia reared in BFT. Tilapia reared in EP showed higher content of crude protein and lipids in the fillet. In both production systems, there was no difference in the body lipid profile. Fish in BFT showed a higher concentration of glucose and ammonia in the muscle and amino acids in the liver. Fish reared in EP showed a higher concentration of lactate in the liver compared to those in BFT. In conclusion, the production system alters the metabolism of fish. The biofloc has a considerable amount of fatty acids, which can be considered in the formulation of diets for tilapia in this system.

Keywords: fish farming; Oreochromis niloticus; polyunsaturated fatty acids; farming system.

Received on July 23, 2022. Accepted on April 20, 2023.

#### Introduction

Nile tilapia (*Oreochromis niloticus*) is the third most produced species in the world, with approximately 4,525 thousand tons, representing 8.3% aquaculture production (FAO, 2020). This fish is consolidated as one of the main species farmed in aquaculture worldwide, as it is rustic, has good growth rates, and absence of intramuscular spines (El-Sayed, 2006). Due to these characteristics, tilapia adapts to intensive production systems, such as the net tank and biofloc technology (BFT).

In Brazil, most fish production (80%) occurs in small properties with less than 2 ha, in earthen ponds (EP) that require water renewal or mechanical aeration (Valenti, Barros, Moraes-Valenti, Bueno & Cavalli, 2021). In this context, it is necessary to develop farming technologies to reduce the use of water and nutrients, thus minimizing environmental impacts, without harming the production indices (Hu et al., 2015).

BFT is a system that has a balanced ratio of carbon and nitrogen to stimulate the development of microorganisms that contribute to the control of water quality and can serve as food for fish (Azim & Little, 2008). This system has minimal or no water renewal (Avnimelech & Kochba, 2009). Recent studies compare BFT with the clear water (CW) system, mainly in the initial phases (1 to 20 g), and show that BFT causes an "anti-stress" effect on fish (Lovera et al., 2017). In addition, these authors showed that BFT provides better survival rates, feed conversion, and water savings compared to CW.

In the growth phase (50 to 200 g), tilapia reared in BFT shows better performance, immune response, and antioxidant status, besides increased activity of digestive enzymes (Azim & Little, 2008; Long, Yang, Li, Guan, & Wu, 2015; Mansour & Esteban, 2017). Luo et al. (2014) evaluated tilapia in a BFT and CW system in the

Page 2 of 9 Durigon et al.

finishing phase (greater than 600 g body weight) and concluded that the BFT system is beneficial for tilapia production. However, it is worth mentioning that the CW system does not contain microalgae, which are food sources present in EP (He et al., 2018).

Previous studies on tilapia farming in BFT focused on the initial stages, or in specific growth stages. The present study aimed to evaluate tilapia farming in BFT and EP during the entire productive period by assessing organosomatic parameters, chemical composition, biochemical parameters, and fatty acid profile. Therefore, novel data are provided concerning the whole farming process.

#### Material and methods

## **Cultivation system**

Tilapias were reared in two production systems, BFT and EP, for 150 days. Fish were fed commercial feed according to each growth stage (El-Sayed, 2006). From 2 g to 10 g of weight, 1 mm feed with 45% crude protein (CP) was supplied; from 10 g to 40 g, 2 mm feed with 45% CP; from 40 g to 150 g, 3 mm feed with 36% CP; from 150 g to 350 g, 4 mm feed with 32% CP; and after 350 g until the end of the cultivation cycle, a 6 mm feed with 32% CP was supplied, as recommended by the feed maker. Fish were fed to apparent satiety. At the end of the period, twelve tilapias from each system were randomly caught, with an average size of  $780 \pm 22$  g. All experimental procedures were registered by the Animal Research Ethics Committee of the Federal University of Santa Maria, protocol number 2423200420.

The BFT system was not subjected to organic fertilization because it had already been matured with the previous cultivation of tilapia and was composed of  $10 \text{ m}^3$  tanks. Molasses was used as a carbon source when the ammonia value exceeded  $2 \text{ mg L}^{-1}$ . Tanks were lined with geomembrane, covered with greenhouse, under constant aeration to maintain the levels of oxygen and solids in suspension. Solid decantation was carried out whenever the values exceeded  $40 \text{ mL L}^{-1}$ , until reaching  $25 \text{ mL L}^{-1}$ . To correct the pH, agricultural limestone was used whenever the value was less than 7.0. Alkalinity was corrected whenever the value dropped from  $70 \text{ mg CaCO}_3 \text{ L}^{-1}$ , with sodium bicarbonate at a concentration of  $0.025 \text{ g L}^{-1}$ . The stocking density was  $55 \text{ fish m}^{-3}$  (Luo et al., 2014)

EP used for farming (250 m²), had a depth ranging from 80 to 120 cm, the stocking density was 4 fish m $^{-3}$  Whenever ammonia exceeded 2 mg L $^{-1}$ , 20% of the water in the tank was replenished. Emergency aeration was provided by an aerator (Electric fountain aerator, 1.5 hp) whenever the oxygen level was less than 3 mg L $^{-1}$ . Transparency was measured with a Secchi disk, and was kept between 20 and 40 cm. When the value was less than 20 cm, water was replenished. Chemical fertilization (urea) was carried out whenever the transparency value exceeded 40 cm.

# **Organosomatic indexes**

Fish were euthanized to collect data on weight, body, and organ length. Subsequently, the organosomatic parameters were calculated: condition factor (CF) = (total weight x 100/fish length<sup>3</sup>) x 100; hepatosomatic index (HI) = (liver weight/fish weight) x 100; visceral fat index (VFI) = (visceral fat weight/fish weight) x 100; intestinal quotient (IQ) = (length of digestive tract/ fish length) x 100; enterosomatic index (EI) = (digestive tract weight/total weight) x 100; fillet yield (FY) = (fillet weight/total weight) x 100; and carcass yield (CY) = (carcass weight/total weight) x 100.

## Fillet and biofloc composition

The contents of dry mass (DM), CP, and mineral matter (MM) of BFT and fish fillets were determined according to the methodology proposed by AOAC (1995). To determine lipid levels, the Bligh and Dyer (1959) technique was used. Biofloc was collected by decanting the cultivation water (about 400 L) for approximately 30 min, and then eliminating the supernatant. This operation was carried out a few times until obtaining a material with the least amount of water possible, afterwards, biofloc was to dried in a stove with air recirculation (at 55°C for 72 hours) for chemical analysis.

# Fatty acid profile

Fat was extracted from samples using chloroform and methanol, as described by Bligh and Dyer (1959), and used for determination of the fatty acid profile. To prevent lipid oxidation during and after extraction,

0.02% butyl hydroxy toluene was added to the chloroform used. Fat was saponified in methanolic KOH solution and then esterified in methanolic  $H_2SO_4$  solution (Hartmann & Lago, 1973). Methylated fatty acids were analyzed using an Agilent Technologies gas chromatograph (HP 6890N) equipped with a capillary column (DB-23 60 m x 0.25 mm x 0.25  $\mu$ m) and flame ionization detector. The temperature of the injector port was set at 250°C and the carrier gas was nitrogen (0.6 mL min.<sup>-1</sup>). After injection (1  $\mu$ L, split ratio 50:1), the oven temperature was held at 150°C for 1 min. then increased to 240°C at 4°C min.<sup>-1</sup>, and held at this temperature for 12 min. Standard fatty acid methyl esters [37-component FAME Mix, C 22:5 n3 and PUFA (polyunsaturated fatty acid) 2 from Sigma, Saint Louis, MO, USA] were run under the same conditions and the subsequent retention times were used to identify the fatty acids. Fatty acids were expressed as percentage of the total fatty acids identified.

## **Biochemical parameters**

Samples of gills, liver, and muscle were collected. In the gills, the total ammonia content was determined (Verdouw, Van Echteld, & Dekkers, 1978). Liver and muscle tissues were analyzed for total protein (Bradford, 1976), glycogen and glucose (Dubois, Gilles, Hamilton, Rebers, & Smith, 1956), lactate (Harrower & Brown, 1972), total ammonia, thiobarbituric acid reactive substances (TBARS) (Buege & Aust, 1978) and amino acids (AA) (Spies, 1957).

## Statistical analysis

Data were subjected to the Shapiro-Wilk normality test. Results with normal distribution were tested by analysis of variance (ANOVA). For variables that showed a significant difference (p < 0.05), their means were compared by Student's t-test. All analyses were run using the  $R^{\circ}$  statistical package.

## **Results**

## **Organosomatic indexes**

The FY showed no difference between the cultivation systems (BFT and EP). CY was higher in fish reared in EP. The CF, HS, VFI, IQ, and EI indices were higher in fish reared in the BFT system (Table 1).

Table 1. Organosomatic parameters of tilapia reared in biofloc (BFT) system or earthen ponds system (EP) (mean ± standard deviation).

Parameters	BFT	EP	P
CF	$2.60 \pm 0.14^{a}$	$2.27 \pm 0.07^{\rm b}$	<0.001
HI (%)	$2.26 \pm 0.47^{a}$	$1.52 \pm 0.43^{b}$	0.012
VFI (%)	$4.34 \pm 0.98^{a}$	$2.24 \pm 0.44^{b}$	< 0.001
IQ (%)	$9.29 \pm 1.14^{a}$	$7.79 \pm 1.12^{b}$	0.046
EI (%)	$3.07 \pm 0.40^{a}$	$2.32 \pm 0.40^{b}$	0.022
FY (%)	$32.20 \pm 1.69^{a}$	$33.86 \pm 2.72^{a}$	NS (0.22)
CY (%)	$86.67 \pm 1.15^{b}$	$90.51 \pm 1.46^{a}$	< 0.001

Means followed by different letters, in the same row, are significantly different by Student's t-test (p < 0.05). NS: non-significant; CF: condition factor; HI: hepatosomatic index; VFI: visceral fat index; IQ: intestinal quotient; EI: enterosomatic index; FI: fillet yield; CY: carcass yield.

#### Fillet and biofloc composition

There was no difference for DM and MM. The levels of CP and lipids were higher in fillets of fish reared in EP. In the biofloc system, fish showed higher levels of MM and CP, 23.84 and 21.34%, respectively, while the lipid content was 2.39% (Table 2).

**Table 2.** Chemical composition of biofloc and fillet of tilapia reared in biofloc system (BFT) or earthen ponds (EP) (mean ± standard deviation).

Parameters (%)	BFT	EP	P	Biofloc
DM	$21.45 \pm 2.38^{a}$	$21.35 \pm 0.13^{a}$	NS (0.18)	93.14 ± 0.99
MM	$2.38 \pm 0.34^{a}$	$2.31 \pm 0.54^{a}$	NS (0.82)	$23.84 \pm 2.51$
CP	$18.30 \pm 0.95^{b}$	$19.61 \pm 0.48^{a}$	0.004	$21.34 \pm 1.12$
Lipids	$1.67 \pm 0.49^{b}$	$2.79 \pm 0.76^{a}$	0.047	$2.39 \pm 0.42$

Means followed by different letters, in the same row, are significantly different by Student's t-test between the two production systems. Mean values of the biofloc meal were not considered in this statistical analysis (p < 0.05). NS: non-significant; DM: dry matter; MM: mineral matter; CP: crude protein.

Page 4 of 9 Durigon et al.

#### **Fatty acid composition**

The composition of fatty acids in the fillet and body of tilapia was not influenced by the production system. The fatty acids found at greatest amounts were C 16:0, C 18:0, C 16:1 n-7 and C 18:1 n-9, C 18:2 n-6 (Table 3).

**Table 3.** Fatty acid composition (%total identified fatty acids) of the biofloc, fillet and whole body of tilapia reared in biofloc system (BFT) or earthen ponds (EP).

Composition (%)	Fillet		Whole	Whole body	
	BFT	EP	BFT	EP	Biofloc
C 12:0	0.07	0.09	0.10	0.08	0.14
C 14:0	2.50	2.78	2.68	2.59	1.59
C 14:1 n-5	0.15	0.17	0.24	0.19	0.00
C 15:0	0.19	0.20	0.18	0.19	0.58
C 15:1 n-5	0.05	0.05	0.05	0.07	0.60
C 16:0	24.79	23.59	21.03	23.10	25.03
C 16:1 n-7	4.71	5.02	5.99	5.77	5.05
C 17:0	0.29	0.33	0.23	0.32	0.77
C 17:1 n-5	0.19	0.19	0.23	0.30	0.42
C 18:0	6.37	6.20	4.45	5.73	12.09
C 18:1 n-9	34.11	33.90	37.72	36.13	19.72
C 18:1 n-7	2.60	2.80	2.62	2.47	7.26
C 18:2 n-6	15.36	15.88	15.90	15.87	20.51
C 18:3 n-6	0.84	0.99	1.64	0.98	0.18
C 18:3 n-3	1.17	1.46	1.12	1.30	2.30
C 20:0	0.00	0.07	0.09	0.06	0.00
C 20:1 n-9	1.61	1.69	1.51	1.69	0.70
C 20:2 n-6	0.26	0.25	0.00	0.19	0.00
C 20:3 n-6	0.84	0.78	1.01	0.65	0.20
C 20:4 n-6	1.59	1.27	1.34	0.74	0.58
C 20:3 n-3	0.18	0.20	0.17	0.21	0.00
C 20:5 n-3	0.08	0.10	0.00	0.15	0.00
C 22:4 n-6	1.05	0.92	0.61	0.36	0.33
C 24:0	0.05	0.07	0.00	0.05	1.27
C 22:5 n-6	0.12	0.14	0.09	0.12	0.00
C 22:5 n-3	0.29	0.28	0.25	0.20	0.16
C 22:6n3	0.54	0.57	0.75	0.48	0.51
SFA	34.26	33.32	28.77	32.12	41.46
UFA	65.74	66.68	71.23	67.88	58.54
MUFA	43.44	43.83	48.35	46.62	33.76
PUFA	22.30	22.85	22.88	21.26	24.78
UFA/SFA ratio	1.92	2.00	2.48	2.11	1.41
n-6	20.06	20.23	20.59	18.92	21.81
n-3	2.25	2.62	2.29	2.34	2.97
n-3/n-6 ratio	0.11	0.13	0.11	0.12	0.14

C 12:0: lauric acid; C 14:0: myristic acid; C 14:1 n-5: myristoleic acid; C 15:0: pentadecanoic acid; C 15:1 n-5: 10-pentadecenoic acid; C 16:0: palmitic acid; C 16:1 n-7: palmitoleic acid; C 17:0: heptadecanoic acid; C 17:1 n-5: 10-heptadecenoic acid; C 18:0: stearic acid; C 18:1 n-7: 11-octadecenoic; C 18:1 n-9: oleic acid; C 18:2 n-6: linoleic acid; C 18:3 n-6: gamma-linolenic acid; C 18:3 n-3: alfa-linolenic acid; C 20:0: arachidic acid; C 20:1 n-9: 11-eicosenoic acid; C 20:2 n-6: dihomolinoleic acid; C 20:3 n-6: dihomopinolenic acid; C 20:4 n-6: arachidonic acid; C 20:3 n-3: dihomolinolenic acid; C 20:5 n-3: eicosa-5,8,11,14,17-pentaenoic acid; C 22:4 n-6: adrenic acid; C 24:0: lignoceric acid; C 22:5 n-6: docosa-4,7,10,13,16-pentaenoic acid; C 22:5 n-3: docosa-7,10,13,16,19-pentaenoic acid; C 22:6 n-3: docosa-4,7,10,13,16,19-pentaenoic acid; C 22:6 n-3: docosa-4,7,10,13,16,19-pentaenoic acid; C 20:2 n-3: docosa-4,7,10,13,16,19-pentaen

# **Biochemical parameters**

In the muscle, there was a difference in the concentration of ammonia and glucose, with higher concentrations observed in fish reared in BFT. In the liver, a higher lactate concentration was found in fish reared in EP, and a higher AA content in fish in BFT. Ammonia, total protein, glucose, glycogen, and TBARS showed no difference between fish reared in BFT and EP (Table 4).

Table 4. Biochemical parameters of tilapia reared in biofloc (BFT) system or earthen ponds (EP) (mean ± standard deviation).

Tissue		BFT	EP	P
Muscle	Ammonia (µmol g <sup>-1</sup> )	$0.44 \pm 0.08^{a}$	$0.27 \pm 0.07^{b}$	< 0.01
	Lactate (g <sup>-1</sup> )	$146.85 \pm 41.99^{a}$	$133.63 \pm 27.78^{a}$	NS (0.75)
	Total proteins (g g <sup>-1</sup> )	$35.66 \pm 6.79^{a}$	$31.64 \pm 5.85^{a}$	NS (0.16)
	Glucose (mg g <sup>-1</sup> )	$16.46 \pm 4.79^{a}$	$9.40 \pm 1.30^{b}$	0.01

	Amino acids (µmol g <sup>-1</sup> )	$4.11 \pm 0.82^{a}$	$5.55 \pm 1.79^{a}$	NS (0.36)
	Glycogen (mg g <sup>-1</sup> )	$11.10 \pm 5.02^{a}$	$17.35 \pm 7.35^{a}$	NS (0.09)
	TBARS (µmol MDA g <sup>-1</sup> )	$0.61 \pm 0.29^{a}$	$0.35\pm0.13^a$	NS (0.131)
	Ammonia (µmol g -1)	$0.50 \pm 0.19^{a}$	$0.67 \pm 0.23^{a}$	NS (0.12)
	Lactate (mg g <sup>-1</sup> )	$9.80 \pm 3.13^{a}$	$16.82 \pm 4.68^{b}$	0.02
	Total protein (g g <sup>-1</sup> )	$28.61 \pm 8.69^{a}$	$31.78 \pm 8.49^{a}$	NS (0.52)
Liver	Glucose (mg g <sup>-1</sup> )	$21.17 \pm 4.73^{a}$	$28.65 \pm 6.99^{a}$	NS (0.06)
	Amino acids (µmol g <sup>-1</sup> )	$3.97 \pm 0.83^{a}$	$2.94 \pm 0.47^{b}$	0.03
	Glycogen (mg g <sup>-1</sup> )	$50.30 \pm 14.92^{a}$	$50.59 \pm 16.68^{a}$	NS (0.97)
	TBARS (µmol MDA g <sup>-1</sup> )	$1.25 \pm 0.28^{a}$	$1.06 \pm 0.21^{a}$	NS (0.175)
Gills	Ammonia (µmol g -1)	$3.69 \pm 0.27^{a}$	$3.92 \pm 0.36^{a}$	NS (0.15)

Means followed by different letters, in the same row, are significantly different by Student's t-test (p < 0.05). NS: non-significant; MDA: malondialdehyde.

## Discussion

# **Organosomatic indexes**

BFT contains highly digestible nutrients for fish (Emerenciano, Martínez-Córdova, Martínez-Porchas, & Miranda-Baeza, 2017). This is due to microorganisms that live in this system, such as microalgae, rotifers, ciliates, and nematodes (Monroy-Dosta, Lara-Andrade, Castro-Mejía, Castro-Mejía, & Emerenciano, 2013). Tilapia can use the BFT substrate, due to its filtering feeding behavior (El-Sayed, 2006). In the present study, the constant availability of food may have contributed to raising the organosomatic indices in fish reared in BFT. The constant presence of food results in increased metabolic activity involving the breakdown and absorption of nutrients, mainly protein (Baldisserotto, 2018). In this study, BFT had an approximate 21% CP content. A similar result was reported in other studies (Silva et al., 2018) The CP of the diet together with the protein of bioflocs may have caused an increase in the routes of anabolism and catabolism (glycogenesis and deamination) and therefore there was an increase in the liver size.

Fish reared in BFT had a higher CF compared to fish reared in EP. This index is used as a comparison, as it provides information on the physiological state of the animals. Based on this assumption, individuals with greater mass in a given length, are in a better condition (Lima-Junior, Cardone, & Goitein, 2002).

The increase in EI in the BF system can be directly related to the consumption of substrates, which results in an increase in the surface of the intestine, enabling greater contact and, consequently, greater absorption of nutrients (Moreira et al., 2012). These same authors observed an increase in this parameter when fish were fed diets containing natural feed, compared to diets based on feed.

#### Fillet and biofloc composition

Fish reared in BFT system had a lower content of CP and fat in the fillet. This may be related to the different food constituents between the production systems. In BFT, tilapia can absorb the CP present in the biofloc (Poli, Legarda, Lorenzo, Martins, & Vieira 2019), which may have caused an imbalance in the energy/protein ratio of the animal diet, thus leading them to deposit less CP and lipids (NRC, 2011).

In this study, tilapia reared in EP had a higher lipid content in the fillet. This result may be related to the presence of microalgae, rich in protein and lipids, with 23.16% CP and 36.58% lipids in the DM (He et al., 2018). Because it is a filter-feeding species, tilapia can obtain a large part of nutrients from microalgae, this may also have contributed to a greater amount of CP in fillets of tilapia reared in EP. On the other hand, tilapia reared in BFT had lower content of lipids, which was also observed by Lovera et al. (2017).

#### Lipid profile

In EP, there is production of microalgae, which are sources of PUFA, such as eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA). In closed systems, these nutrients come from the diet (Toledo, Silva, Vieira, Mouriño, & Seiffert, 2016). However, BFT has been considered an important source of fatty acids that contributes to fish feeding as documented by Azim and Little (2008), Ekasari, Crab, and Verstraete (2010), and Toledo et al. (2016). The consumption of biofloc can contribute to animal growth and health (Haridas et al., 2017), since microorganisms can represent 30 - 50% nutrient intake (Avnimelech, 2007). Some fatty acids have been found in large quantities in the biofloc, for example, C 20:4 n-6, which can represent 5 times more quantity compared to feed, in addition to being an important source of PUFA (Toledo et al., 2016). These fatty acids present in the biofloc may have been incorporated into the fillet and carcass, as in this study a 20% greater amount was observed for C 20:4 n-6 in the fillet and 44% greater in the body composition of fish reared in BFT.

Page 6 of 9 Durigon et al.

The linoleic acid (C 18:2 n-6) was the most abundant PUFA in the biofloc, whereas palmitic acid (C 16:0) was the most abundant SFA. A similar result was observed by Ekasari et al. (2010). Also in the biofloc, large amounts of C 16:1 n-7, C 18:1 n-7, and C 18:1 n-9 were also observed, in which the sum of n-6 was greater in BFT (21.81 mg g<sup>-1</sup>). This increase may be related to the time of the BFT system, because the longer the cultivation time, the greater the development of dinoflagellates (Durigon et al., 2020), which are the main responsible for the presence of fatty acids in the BFT.

The BFT fatty acid profile can vary for several reasons, including the carbon source used, the composition of microorganisms, the salinity of the water (Ekasari et al., 2010), and the lipid composition of the diet (Toledo et al., 2016). In addition, microorganisms can synthesize new fatty acids, elongating and desaturating the carbon chain (Ekasari et al., 2010). The significant presence of PUFA n-3 and n-6 in the biofloc, such as  $\alpha$ -linolenic (C 18:3 n-3), can be elongated and desaturated to produce EPA and DHA, however, the process occurs in a low percentage (Suárez-Mahecha et al., 2002). In the present study, this fatty acid was not found with significant amounts in fish reared in BFT. The lack of a specific diet for fish reared in this system can cause the consumption of this fatty acid.

Palmitic acid was one of the most abundant fatty acids in the composition of the fillet and in the body composition of the fish, varying between 21.03 and 24.79. Values very similar to those found here were also observed by Duarte et al. (2021), who evaluated the fatty acid profile in the muscle of tilapia fed different levels of fish meal.

## **Biochemical parameters**

The concentration of glucose and ammonia increased in the muscle of fish reared in BFT. In the liver, there was a reduction in lactate content and an increase in AA. These changes in metabolism may be related to the imbalance of nutrients present in the diet. BFT contains microorganisms with high digestibility that serve as natural food (Nelson & Cox, 2006). Nutrients with high digestibility can improve the absorption rate and increase its availability (Nelson & Cox, 2006). The increase in the amount of AA in the liver indicates an improved absorption (Uczay et al., 2019). Fish reared in BFT have a lower protein demand from the feed, as they can meet part of their requirement from microorganisms present in this system (Mansour & Esteban, 2017; Silva et al., 2018; Green, Rawles, Schrader, Gaylord, & McEntire, 2019; Durigon et al., 2020). The intake of the BFT protein fraction and the diet may have caused an increase in AA intake, modifying the elevated metabolic parameters (Teodósio, Engrola, Colen, Masagounder, & Aragão, 2020).

Imbalance in the protein fraction and energy of the diet can lead to AA catabolism and its carbon chain can go to energy production routes (NRC, 2011). The carbon chain, on the other hand, can go to gluconeogenesis for conversion to glucose. This is because excess AA cannot be stored. The amino group is transformed into ammonia and excreted. Excessive ammonia, in the body and in water, can cause fish mortality, cross cell membranes and cause cell depolarization, leading to apoptosis (Baldisserotto, 2018). Balanced diets are necessary, especially in systems with little water renewal, since AA imbalance can affect fish metabolism, and such results have already been evidenced (Uczay et al., 2019; Battisti, Rabaioli, Uczay, Sutili, & Lazzari, 2020).

In the present study, fish reared in EP showed lower glucose concentration in the muscle and higher lactate concentration in the liver. This is due to the increase in anaerobic glycolysis in the muscle and gluconeogenesis in the liver; the cycle that involves glucose and lactate is called the Cori cycle (Nelson & Cox, 2006). The glucose present in the muscle is converted to lactate, transported to the liver, through the bloodstream, and again converted to glucose; this route is a way of producing energy in situations of hypoxia or anoxia and the Cori cycle takes place mainly in conditions of increased muscle activity when there is intense exercise (Nelson & Cox, 2006), which was possibly due to the fish farming system. In the EP, fish have more space to move around, consuming more oxygen. In BFT, the space available is smaller, and therefore the liver's lactate production was lower and the glucose levels in the muscle were higher.

## Conclusion

In conclusion, the production system influences the fillet composition and the metabolism of tilapia. BFT has an excess amount of CP and fatty acids, which improves organosomatic indexes, fillet composition, and fish metabolism compared to those reared in EP. However, studies on formulations of diets that consider the nutrients present in the biofloc are necessary to maximize the production of tilapia in BFT.

# Acknowledgments

This work was conducted with the support of the *Coordenação de Aperfeiçoamento de Pessoal de Nível Superior* (Finance Code 001). T.E. is recipient of the *Conselho Nacional de Desenvolvimento Científico e Tecnológico* research fellowship [303654/2017-1] and R.L [process 312849/2020-6]. This work also has support from the project for the Center of Technological Innovation in Agribusiness: a proposal for entrepreneurship, research and regional development contemplated in Public Notice 16/2022.

# References

- Association of Official Analytical Chemists [AOAC]. (1995). *Official Methods of Analysis of AOAC International* (16th ed.). Arlington, US: AOAC International.
- Avnimelech Y. (2007). Feeding with microbial flocs by tilapia in minimal discharge bio-flocs technology ponds. *Aquaculture*, *264*(1-4), 140-147. DOI: https://doi.org/10.1016/j.aquaculture.2006.11.025
- Avnimelech, Y., & Kochba, M. (2009). Evaluation of nitrogen uptake and excretion by tilapia in biofloc tanks, using <sup>15</sup>N tracing. *Aquaculture*, *287*(1-2), 163-168. DOI: https://doi.org/10.1016/j.aquaculture.2008.10.009
- Azim, M. E., & Little, D. C. (2008). The biofloc technology (BFT) in indoor tanks: water quality, biofloc composition, and growth and welfare of Nile tilapia (*Oreochromis niloticus*). *Aquaculture*, *283*(1-4), 29-35. DOI: https://doi.org/10.1016/j.aquaculture.2008.06.036
- Baldisserotto, B. (2018). Fisiologia de peixes aplicada à piscicultura (3. ed.). Santa Maria, RS: Editora UFSM.
- Battisti, E. K., Rabaioli, A., Uczay, J., Sutili, F. J., & Lazzari, R. (2020). Effect of stocking density on growth, hematological and biochemical parameters and antioxidant status of silver catfish (*Rhamdia quelen*) cultured in a biofloc system. *Aquaculture*, *524*, 1-6.
- DOI: https://doi.org/10.1016/j.aquaculture.2020.735213
- Bligh, E. G., & Dyer, W. J. (1959). A rapid method of total lipid extraction and purification. *Canadian Journal of Biochemistry and Physiology*, *37*(8), 911–917. DOI: https://doi.org/10.1139/o59-099
- Bradford, M. M. (1976). A rapid and sensitive method for the quantitation of microgram quantities of protein utilizing the principle of protein-dye binding. *Analytical Biochemistry*, 72(1-2), 248-254. DOI: https://doi.org/10.1006/abio.1976.9999
- Buege, J. A., & Aust, S. D. (1978). Microsomal lipid peroxidation. *Methods in Enzymolgy*, *52*, 302-310. DOI: https://doi.org/10.1016/s0076-6879(78)52032-6
- Duarte, F. O. S., Paula, F. G., Prado, C. S., Santos, R. R., Minafra-Rezende, C. S., Gebara, C., & Lage, M. E. (2021). Better fatty acids profile in fillets of Nile tilapia (*Oreochromis niloticus*) supplemented with fish oil. *Aquaculture*, *534*, 1-7. DOI: https://doi.org/10.1016/j.aquaculture.2020.736241
- Dubois, M., Gilles, K. A., Hamilton, J. K., Rebers, P. A., & Smith, F. (1956). Colorimetric method for determination of sugars and related substances. *Analytical Chemistry*, *28*(3), 350-356. DOI: https://doi.org/10.1021/AC60111A017
- Durigon, E. G., Lazzari, R., Uczay, J., Lopes, D. L. A., Jerônimo, G.T., Sgnaulin, T., & Emerenciano, M. G. C. (2020). Biofloc technology (BFT): adjusting the levels of digestible protein and digestible energy in diets of Nile tilapia juveniles raised in brackish water. *Aquaculture and Fisheries*, *5*(1), 42-51. DOI: https://doi.org/10.1016/j.aaf.2019.07.001
- Ekasari, J., Crab, R., & Verstraete, W. (2010). Primary nutritional content of bio-flocs cultured with different organic carbon sources and salinity. *HAYATI Journal of Biosciences*, *17*(3), 125-130. DOI: https://doi.org/10.4308/hjb.17.3.125
- El-Sayed, A. F. M. (2006). *Tilapia culture*. London, UK: CABI Publishing. DOI: https://doi.org/10.1079/9780851990149.0000
- Emerenciano, M. G. C., Martínez-Córdova, L. R., Martínez-Porchas, M., & Miranda-Baeza A. (2017). Biofloc technology (BFT): a tool for water quality management in aquaculture. In H. Tutu (Ed.), *Water Quality* (p. 91-109). London, UK: IntechOpen. DOI: https://doi.org/10.5772/66416
- Food and Agriculture Organization of the United Nations [FAO]. (2020). *The state of world fisheries and aquaculture 2020. Sustainability in action*. Rome, IT: FAO. DOI: https://doi.org/10.4060/ca9229en

Page 8 of 9 Durigon et al.

Green, B. W., Rawles, S. D., Schrader, K. K., Gaylord, T. G., & McEntire, M. E. (2019). Effects of dietary protein content on hybrid tilapia (*Oreochromis aureus* × *O. niloticus*) performance, common microbial off-flavor compounds, and water quality dynamics in an outdoor biofloc technology production system. *Aquaculture*, *503*, 571-582. DOI: https://doi.org/10.1016/j.aquaculture.2019.01.034

- Haridas, H., Verma, A. K., Rathore, G., Prakash, C., Sawant, P. B., & Rani, A. M. B. (2017). Enhanced growth and immuno-physiological response of genetically improved farmed tilapia in indoor biofloc units at different stocking densities. *Aquaculture Research*, *48*(8), 4346-4355. DOI: https://doi.org/10.1111/are.13256
- Harrower, J. R., & Brown, C. H. (1972). Blood lactic acid a micromethod adapted to field collection of microliter samples. *Journal of Applied Physiology*, *32*(5), 709-711. DOI: https://doi.org/10.1152/jappl.1972.32.5.709
- Hartman, L., & Lago, R. C. A. (1973). Rapid preparation of fatty acid methyl esters from lipids. *Laboratory Practice*, 22(6), 475-476.
- He, Y., Lin, G., Rao, X., Chen, L., Jian, H., Wang, M., ... Chen, B. (2018). Microalga *Isochrysis galbana* in feed for *Trachinotus ovatus*: effect on growth performance and fatty acid composition of fish fillet and liver. *Aquaculture International*, *26*, 1261-1280. DOI: https://doi.org/10.1007/s10499-018-0282-y
- Hu, Z., Lee, J. W., Chandran, K., Kim, S., Brotto, A. C., & Khanal, S. K. (2015). Effect of plant species on nitrogen recovery in aquaponics. *Bioresource Technology*, *188*, 92-98. DOI: https://doi.org/10.1016/j.biortech.2015.01.013
- Lima-Junior, S. E., Cardone, I. B., & Goitein, R. (2002). Determination of a method for calculation of allometric condition factor of fish. *Acta Scientiarum*, *24*(2), 397-400. DOI: https://doi.org/10.4025/actascibiolsci.v24i0.2311
- Long, L., Yang, J., Li, Y., Guan, C., & Wu, F. (2015). Effect of biofloc technology on growth, digestive enzyme activity, hematology, and immune response of genetically improved farmed tilapia (*Oreochromis niloticus*). *Aquaculture*, 448, 135-141. DOI: https://doi.org/10.1016/j.aquaculture.2015.05.017
- Lovera, K. P. Z., Brito, L. O., Lima, P. C. M., Arana, L. A. V., Galvez, A. O., & Cárdenas J. M. V. (2017). Cultivo de alevines de tilapia en sistema biofloc bajo diferentes relaciones carbono/nitrógeno. *Boletim do Instituto de Pesca*, *43*(3), 399-407. DOI: https://doi.org/10.20950/1678-2305.2017v43n3p399
- Luo, G., Gao, Q., Wang, C., Liu, W., Sun, D., Li, L., & Tan, H. (2014). Growth, digestive activity, welfare, and partial cost-effectiveness of genetically improved farmed tilapia (*Oreochromis niloticus*) cultured in a recirculating aquaculture system and an indoor biofloc system. *Aquaculture*, 422-423, 1-7. DOI: https://doi.org/10.1016/j.aquaculture.2013.11.023
- Mansour, A. T., & Esteban, M. A. (2017). Effects of carbon sources and plant protein levels in a biofloc system on growth performance, and the immune and antioxidant status of Nile tilapia (*Oreochromis niloticus*). *Fish & Shellfish Immunology*, *64*, 202–209. DOI: https://doi.org/10.1016/j.fsi.2017.03.025
- Monroy-Dosta, M. C., Lara-Andrade, R., Castro-Mejía, J., Castro-Mejía, G., & Emerenciano M. G. C. (2013). Composición y abundancia de comunidades microbianas asociadas al biofloc en un cultivo de tilapia. *Revista de Biología Marina y Oceanografía*, 48(3), 511-520. DOI: https://doi.org/10.4067/S0718-19572013000300009
- Moreira, R. L., Silveira, L. P., Teixeira, E. G., Moreira, A. G. L., Moura, P. S., & Farias, W. R. L. (2012). Growth and gastrointestinal indices in Nile tilapia fed with different diets. *Acta Scientiarum. Animal Sciences*, 34(3), 223-229. DOI: https://doi.org/10.4025/actascianimsci.v34i3.13327
- Nelson, D. L., & Cox, M. M. (2006). Lehninger princípios de bioquímica (6. ed.). São Paulo, SP: Sarvier.
- National Research Council [NRC]. (2011). *Nutrient Requirements of Fish and Shrimp*. Washington, DC: National Academies Press.
- Poli, M. A., Legarda, E. C., Lorenzo, M. A., Martins, M. A., & Vieira, F. N. (2019). Pacific white shrimp and Nile tilapia integrated in a biofloc system under different fish-stocking densities. *Aquaculture*, 498, 83–89. DOI: https://doi.org/10.1016/j.aquaculture.2018.08.045
- Silva, M. A., Alvarenga, E. R., Alves, G. F. O., Manduca, L. G., Turra, E. M., Brito, T. S., ... Teixeira, E. A. (2018). Crude protein levels in diets for two growth stages of Nile tilapia (*Oreochromis niloticus*) in a biofloc system. *Aquaculture Research*, *49*(8), 1-11. DOI: https://doi.org/10.1111/are.13730

- Spies, J. R. (1957). Colorimetric procedures for amino acids. *Methods in Enzymology*, *3*, 467-477. DOI: https://doi.org/10.1016/S0076-6879(57)03417-5
- Suárez-Mahecha, H., Francisco, A., Beirão, L. H., Block, J. M., Saccol, A., & Pardo-Carrasco, S. (2002). Importância de ácidos graxos poliinsaturados presentes em peixes de cultivo e de ambiente natural para a nutrição humana. *Boletim do Instituto de Pesca*, *28*, 101-110.
- Teodósio, R., Engrola, S., Colen, R., Masagounder, K., & Aragão, C. (2020). Optimizing diets to decrease environmental impact of Nile tilapia (*Oreochromis niloticus*) production. *Aquaculture Nutrition*, *26*(2), 422-431. DOI: https://doi.org/10.1111/anu.13004
- Toledo, T. M., Silva, B. C., Vieira, F. N., Mouriño, J. L. P., & Seiffert, W. Q. (2016). Effects of different dietary lipid levels and fatty acids profile in the culture of white shrimp *Litopenaeus vannamei* (Boone) in biofloc technology: water quality, biofloc composition, growth and health. *Aquaculture Research*, *47*(6), 1841-1851. DOI: https://doi.org/10.1111/are.12642
- Uczay, J., Battisti, E. K., Lazzari, R., Pessatti, M. L., Schneider, T. L. S., Hermes, L. B., ... Fabregat, T. E. H. P. (2019). Fish meal replaced by hydrolysed soybean meal in diets increases growth and improves the antioxidant defense system of silver catfish (*Rhamdia quelen*). *Aquaculture Research*, *50*(5), 1438-1447. DOI: https://doi.org/10.1111/are.14019
- Valenti, W. C., Barros, H. P., Moraes-Valenti, P., Bueno, G. W., & Cavalli, R. O. (2021). Aquaculture in Brazil: past, present and future. *Aquaculture Reports*, 19, 1-18. DOI: https://doi.org/10.1016/j.aqrep.2021.100611
- Verdouw, H., Van Echteld, C. J. A., & Dekkers, E. M. J. (1978). Ammonia determination based on indophenol formation with sodium salicylate. *Water Research*, *12*(6), 399-402. DOI: https://doi.org/10.1016/0043-1354(78)90107-0