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Lima, Eduardo Cesar Rodrigues de; Souza, Rafael Liano de; Girao, Pamela Jenny Montes; Braga, Ítalo Felipe Mascena; Correia, Eudes de Souza

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Culture of Nile tilapia in a biofloc system with different sources of carbon[1]

Cultivo da tilápia do Nilo em bioflocos com diferentes fontes de carbono

Eduardo Cesar Rodrigues de Lima[2]*, Rafael Liano de Souza[2], Pamela Jenny Montes Girao[2], Ítalo Felipe Mascena Braga[2] and Eudes de Souza Correia[2]

ABSTRACT - The aim of this work was to evaluate the effects of using different sources of organic carbon on water quality, growth performance and the acceptability of Nile tilapia (*Oreochromis niloticus*) fillets grown in a biofloc system. The experiment was carried out over 145 days at the Aquaculture Station of the Federal Rural University of Pernambuco, Brazil. Fish of 72.6 ± 6.83 g were stored (35 fish m^{-3}) in 19 circular tanks (800 L) in a completely randomised experimental design with three treatments, including as a source of carbon, sugar (SUG), liquid molasses (MOL) and molasses powder (MOP), each with five replications, and one control treatment (CTL) without bioflocs, with four replications. Dissolved oxygen concentrations were significantly higher ($P \leq 0.05$) in the tanks with no bioflocs due to the absence of bacterial biomass. Total ammoniacal nitrogen (TAN) showed a statistical difference ($P \leq 0.05$) between the SUG treatment and the other treatments with bioflocs, having the lowest concentration of 2.53 mg L^{-1}. Survival was greater than 80%, with no statistical difference between treatments ($P > 0.05$); productivity varied from 9.72 (SUG) to 14.22 kg m^{-3} (CTL) ($P \leq 0.05$). Water consumption in the tanks with bioflocs was 11.8 times lower than in the control (CTL). The tilapia fillets from the bioflocs with sugar were preferred by the evaluators, with a score of 7.77 (like moderately to like very much). The carbon sources used (molasses and sugar) can be employed in the culture of *O. niloticus* tilapia in bioflocs with no damage to the culture water or to productivity.

Key words: Tilapia culture. Sustainability. Chitralada. Molasses. Fillets.

RESUMO - Este trabalho objetivou avaliar os efeitos da utilização de diferentes fontes de carbono orgânico na qualidade da água, desempenho de crescimento e aceitabilidade de filés da tilápia do Nilo (*Oreochromis niloticus*) cultivada em sistema de bioflocos. O experimento foi realizado na Estação de Aquicultura da Universidade Federal Rural de Pernambuco, Brasil, durante 145 dias. Peixes de 72,6 ± 6,83 g foram estocados (35 peixes m^{-3}) em 19 tanques circulares (800 L) em um delineamento experimental inteiramente casualizado, com três tratamentos envolvendo as fontes de carbono açúcar (AÇU), melaço líquido (MEL) e melaço em pó (MEP), com cinco repetições cada e um tratamento controle (CTL) sem bioflocos, com quatro repetições. A concentração de oxigênio dissolvido foi significativamente maior ($P \leq 0,05$) nos tanques sem bioflocos devido à ausência de biomassa bacteriana. O nitrogênio da amônia total (NAT) apresentou diferença estatística ($P \leq 0,05$) entre o tratamento AÇU e os demais com bioflocos, exibindo a menor concentração de 2,53 mg L^{-1}. A sobrevivência foi superior a 80%, não havendo diferença estatística entre os tratamentos ($P > 0,05$), e a produtividade variou de 9,72 (AÇU) a 14,22 kg m^{-3} (CTL) ($P \leq 0,05$). O consumo de água nos tanques com bioflocos foi 11,8 vezes menor que o controle (CTL). Os filés de tilápia oriundos do biofloco com açúcar mostraram ter a preferência dos avaliadores, com nota 7,77 (gostei moderadamente a gostei muito). As fontes de carbono utilizadas (melaços e açúcar) podem ser utilizadas no cultivo da tilápia *O. niloticus* em bioflocos sem prejuízos à água de cultivo e à produtividade.

Palavras-chave: Tilapicultura. Sustentabilidade. Chitralada. Melaço. Filés.

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*Author for correspondence
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[2]Departamento de Pesca e Aquicultura, Universidade Federal Rural de Pernambuco/UFRPE, Rua Dom Manoel de Medeiros, s/n, Dois Irmãos, Recife-PE, Brasil, 52.171-900, educesar19@gmail.com, rafaelliano@hotmail.com, pamjmontesgirao@hotmail.com, italo_96felipe@hotmail.com, escorreia@uol.com.br

INTRODUCTION

Improving productivity is one of the main priorities in the development of aquaculture and in particular tilapia farming. The intensification of production systems is seen as the easiest way to reach this goal (AVNIMELECH *et al*., 2008; PIEDRAHITA, 2003). Fish farming with biofloc technology has some advantages over traditional fish farming, among which are that it requires little or no water exchange, has less environmental impact, includes the recycling of nitrogen compounds, the synthesis of bacterial biomass and the supply of a highly nutritious complementary food (AVNIMELECH, 2012).

For this to occur efficiently, it is necessary to ensure a C to N ratio of 15:1 to 20:1 by the addition of a source rich in organic carbon (AVNIMELECH, 2009). Sugar, starch, cellulose, glucose, acetate, glycerol and wheat flour are examples of sources rich in organic carbon (AVNIMELECH, 2009; DE SCHRYVER *et al*., 2008). These carbon sources include alcohols, sugars, starches and fibres, and their degradation can take from a few minutes to a few hours. According to Hargreaves (2013), sugar and molasses are rapidly assimilated by bacteria, increasing biofloc production in less time. More-complex carbohydrates, such as maize and wheat, are metabolised more slowly, and have the advantage of providing a structure for fixing the bacteria, in addition to requiring a set of bacterial enzymes for their degradation which, when ingested by fish, aid in digestion. Fibre-rich materials should be avoided, as they are very resistant to degradation (CHAMBERLAIN, 2001).

Tilapia are perfectly adapted to biofloc systems (AVNIMELECH, 2007). The ability to feed by filtering the water allows them to ingest suspended bioflocs, and due to being a robust and fast-growing fish, they are adapted to well-densified systems (AVNIMELECH, 2011). Several studies have been carried out with tilapia in a biofloc system, including studies on stocking density during the fattening phase (LIMA *et al*., 2015; WIDANARI; EKASARI; MARYAM, 2012), the uptake/absorption of microbial flakes by the tilapia (AVNIMELECH, 2007), the effect of C to N ratios on nitrogen removal and tilapia productivity (PÉREZ-FUENTES *et al*., 2016), and a comparison of biofloc and recirculating systems in aquaculture (LUO *et al*., 2014).

The aim of this study therefore, was to evaluate the effects of sources of organic carbon on water quality, growth performance and the acceptance of Nile tilapia (*Oreochromis niloticus*) fillets grown in a biofloc system.

MATERIAL AND METHODS

The study was carried out over 145 days at the Aquaculture Station of the Federal Rural University of Pernambuco, Brazil. Nineteen circular glass-fibre tanks with a capacity of 1000 L and a working volume of 800 L were used, located in an external area with natural illumination and covered by screens to prevent escape of the fish.

The experiment used an aeration system maintained by a radial compressor (7.5 hp), providing aeration at two air-outlet points, both with porous stones. The tanks were supplied in the proportion of 80 L of fresh water with bioflocs (previously matured) to 720 L of clear water. The clear water was filtered (200 μm) and chlorinated to 10 ppm active chlorine using sodium hypochlorite, and dechlorinated by constant aeration for 24 hours. In the tanks with the biofloc treatments, there was no exchange of water, with fresh water only being used to replace losses from evaporation, while in the control tanks (clear water) the water was renewed weekly (87.5%).

Maturation/preparation of the biofloc inoculum lasted for 22 days, and was carried out in six circular glass-fibre tanks with a capacity of 250 L and a working volume of 200 L, which were filled with filtered fresh water and sterilised as described above. Five tilapia (*O. niloticus*) with an average weight of 44.0 g were placed in each tank, giving an initial biomass of 1.1 kg m^{-3}. The fish were fed twice daily with Pirá 36 extruded commercial feed (Guabi®, Brazil) with guaranteed levels of 10% moisture, 36% crude protein, 6.5% ether extract, 11% mineral matter and 6% fibre, which was offered twice daily in an amount equal to 5% of the biomass. To induce the heterotrophic medium during preparation of the bioflocs, the amount of carbon used per treatment was determined by the protein content (%) of the commercial feed being used, assuming that the protein contained 16% nitrogen and that the tilapia excreted 70% of the protein nitrogen. If 1,000 g of feed contains 36% protein (16% nitrogen), there are 57.6 g of nitrogen, of which 30% is the fraction that could be digested and transformed into muscle; 40.32 g of nitrogen will be excreted. To maintain a C to N ratio of 15:1, 604.8 g of carbon were required, with 1,950 g of sugar supplied as per Emerenciano *et al*. (2007) and Wasielesky *et al*. (2006). It should be noted that there was 31% carbon in the sugar and 30% in the molasses. The volume of bioflocs transferred to each experimental unit was the same, and represented 10% of the total tank volume (80 litres).

Sexually inverted male fingerlings of the Chitralada strain of *O. niloticus*, were aquired from the Integrated Centre for Fisheries and Aquaculture of CODEVASF at

Porto Real do Colégio, in the State of Alagoas, and stored in two masonry tanks (3 x 10 x 1.5 m) until reaching a weight of 72.6 ± 6.83 g. They were transferred to the culture tanks at a density of 35 fish m^{-3}, in a completely randomised design, with three treatments including as a source of carbon, sugar (SUG), liquid molasses (MOL) and molasses powder (MOP), each with five replications, and one treatment, the control (CTL), in a culture system with clear-water and no bioflocs, with four replications.

The fish were initially fed on Pirá 36 extruded commercial feed (Guabi®, Brazil) and, after reaching an average weight of 250 g, on Pirá 32 (Guabi®, Brazil), with guaranteed levels of 8% moisture, 32% crude protein, 6.5% ether extract, 10% mineral matter and 7% fibre. The feed was offered three times a day, at 08:00, 13:00 and 17:00. Biometrics were taken weekly with the aim of evaluating fish growth and adjusting the amount of feed offered according to the biomass (g) in each tank. This monitoring was carried out by weighing and measuring 28.5% of the population of each experimental unit, using a digital balance (± 0.01 g) and an ichthyometer. The feeding rate varied from 4.0 to 2.7% of the live weight day^{-1} throughout the experiment.

Water quality was estimated based on physical and chemical variables: temperature (ºC), dissolved oxygen (mg L^{-1}) and pH, measured twice a day at 08:00 and 17:00, using a YSI 550-A Dissolved Oxygen Meter and a YSI pH100 pH meter (YSI Inc., Yellow Springs, OH, USA). Samples of water from each tank were collected weekly for analysis of the total ammonia nitrogen ($N\text{-}NH_3$ + $N\text{-}NH_4$), nitrite nitrogen ($N\text{-}NO_2$) and alkalinity ($CaCO_3$). Nitrates (NO_3), orthophosphates (PO_4^{-3}) and total suspended solids were measured every two weeks. The samples were analysed with a Hach DR 2800 digital spectrophotometer (Hach Company, Colorado, USA). Sodium bicarbonate ($NaHCO_3$) was added weekly to correct the alkalinity of the culture water.

Maintenance of the bioflocs was carried out daily, applying the organic-carbon sources to the respective tanks as substrate to develop the bacteria and control the ammonia levels. The amount was calculated based on a C to N ratio of 6:1 considering the ammonia nitrogen dissolved in the culture water, as per Avnimelech (1999) and Samocha *et al*. (2007).

The volume of the sedimentable solids (mL L^{-1}) was analysed weekly, when one-litre samples of water from each experimental unit were transferred to Imhoff cones. After being left to settle for 40 minutes, the corresponding volume for these solids was measured. Around 30 mL L^{-1} was adopted as the ideal level of sedimentable solids (AVNIMELECH, 2012). When necessary, this level was controlled using sedimentation tanks connected to the culture tanks (RAY *et al*., 2010). Water consumption was recorded throughout the culture period. These results were expressed in cubic meters (m^3) and the ratio between water consumption and produced biomass (m^3 kg^{-1}) was calculated.

At the end of the experiment, all the fish were rendered insensible and killed by thermal shock using water and ice (~ 4 ºC). They were then immediately counted and weighed to determine the final weight, weight gain, daily weight gain, specific growth rate, survival rate, feed conversion factor and productivity. All the fish were filleted and the fillets labelled, packed, refrigerated and stored at 7 °C. After 30 days, the fillets were submitted to the sensory evaluation test for acceptance of the appearance attribute (MEILGAARD; CIVILLE; CARR, 1999). Two steaks from the same treatment (one sample) were served separately in random order. The test was performed by 30 untrained tasters who evaluated colour, smell and overall appearance using a nine-point hedonic scale (1 - dislike extremely to 9 – like extremely). Samples of the bioflocs and fish fillets from each treatment were sent to CBO Laboratory Analyses (Rio de Janeiro) to determine the centesimal composition of the following items: moisture, crude protein, lipids, ash and crude fibre.

The Shapiro-Wilk normality test and Bartlett's homoscedasticity test were employed at 5% significance. Where normality of the sample and homogeneity of the variances were found, one-way analysis of variance (ANOVA) was applied to the physical and chemical variables of water quality and fish performance. When a statistical difference was found, ANOVA was complemented by the Tukey means comparison test at 5%. The nonparametric Kruskal-Wallis test was used on the sensory-analysis data. The data for survival were transformed to arcsine $x^{0.5}$ before analysis (ZAR, 1996). All the data were analysed using the BioEstat 5.0 software (AYRES *et al*., 2007).

RESULTS AND DISCUSSION

The results of the physical and chemical variables of the water are shown in Table 1. Temperature and pH presented differences between treatments (P<0.05). The water temperature was within the ideal thermal comfort range for tilapia; according to Kubitza (2011), for optimum tilapia growth, the ideal thermal comfort range is between 27 and 32 ºC, and the pH should be kept between 6.00 and 8.50.

The concentration of dissolved oxygen in the tanks varied from 1.07 to 7.71 mg L^{-1}. For the MOP, MOL, SUG and CTL treatments, the respective mean

values were 4.56, 4.49, 4.82 and 5.25 mg L^{-1}. The control treatment had significantly higher concentrations of dissolved oxygen ($P \leq 0.05$) than the treatments with a carbon source, possibly due to the absence of bacterial biomass, which is present in the tanks with bioflocs, in addition to greater photosynthetic activity, albeit to a lesser extent in the culture medium. This large variation in oxygen concentration (1.07 to 7.71 mg L^{-1}) was mainly due to the drop in yield of the aeration system, which lasted for approximately fifteen days and was later resolved by replacing the compressor. Avnimelech (2011) suggests that the minimum concentration of dissolved oxygen for the farming of tilapia in bioflocs should be 4 mg L^{-1}.

Total alkalinity varied from 24.0 to 272.8 mg L^{-1} $CaCO_3$, with mean values for CTL, MOP, MOL and SUG of 101.46, 128.8, 93.49 and 99.33 mg L^{-1} $CaCO_3$ respectively. There was no statistical difference between the SUG, MOL and CTL treatments, however there was a significant difference between MOP and the other treatments with bioflocs (SUG and MOL) ($P<0.05$). Ebeling, Timmons and Bisogni (2006) recommend an alkalinity of 150 mg L^{-1} $CaCO_3$ with biofloc technology. The same authors affirm that the consumption of alkalinity by heterotrophic bacteria as a source of carbon (3.57 g g^{-1} ammonia nitrogen), albeit moderate, is an important aspect in systems with limited water exchange, the addition of carbonates being necessary to maintain alkalinity at acceptable levels.

Azim and Little (2008), when comparing water quality in a system with and without bioflocs, obtained a variation in alkalinity from 8 to 250 and from 18 to 27 mg L^{-1} $CaCO_3$ respectively, indicating that a biofloc system reduces the buffering capacity of the water, requiring the constant addition of correctives.

The concentrations of the dissolved nitrogen compounds (TAN, $N\text{-}NO_2$ and NO_3) and of the orthophosphate for the 145 days of culture are shown in Figure 1. The total ammonia nitrogen (TAN) had mean values of 7.17, 4.33, 2.53 and 2.84 mg L^{-1} respectively in the MOP, MOL, SUG and CTL treatments. The lowest concentration for TAN was obtained in the SUG treatment, which differed significantly from MOP and MOL ($P \leq 0.05$) but did not differ from the control ($P>0.05$). The maximum concentration for TAN (Figure 1A) was 14.9 mg L^{-1} in the treatment with molasses powder, equal to a concentration of un-ionised toxic ammonia of 0.54 mg L^{-1} $N\text{-}NH_3$, below the lethal concentration (mg L^{-1} $N\text{-}NH_3$) estimated by El-Sherif, Feky and Amal (2008). Also in Figure 1A, it can be seen that from the 3rd week of culture, the mean concentration of ammonia in the SUG treatment remained stable and well below the other treatments with bioflocs.

Nitrite is an intermediate product in the process of nitrification and denitrification, its accumulation being common in intensive aquaculture systems. During the culture this variable showed mean values of 1.49, 0.82, 0.74 and 2.30 mg L^{-1} $N\text{-}NO_2$ in the CTL, MOP, MOL and SUG treatments respectively, with no statistical difference between treatments ($P>0.05$). The nitrite remained at low levels, with the highest concentrations during the fourth and the eighth weeks. The first peak in nitrite concentration corresponds to the fourth week in all treatments (Figure 1B), which suggests the action of *Nitrosomonas* bacteria that converted the ammonia accumulated during the second and third weeks. The second peak, recorded during the eighth week, was an isolated case in only one tank of the SUG treatment, due to the rise in nitrite concentration in one of the tanks to 48.8 mg L^{-1} $N\text{-}NO_2$. It should be noted that this maximum

Table 1 - Mean values ± standard deviation (minimum - maximum) of water quality variables for the culture of *Oreochromis niloticus* in bioflocs with different sources of carbon

Variable	Treatment				ANOVA
	MOP	MOL	SUG	CTL	(F value)
Temperature (ºC)	27.98 ± 1.39 ab (24.0-32.0)	28.08 ± 1.36 a (24.0-31.0)	27.85 ± 1.26 bc (24.0-30.0)	27.82 ± 1.26 c (25.0-31.0)	23.1608*
Dissolved oxygen (mg L^{-1})	4.56 ± 1.08 a (1.3-7.12)	4.49 ± 0.98 a (1.07-7.68)	4.82 ± 0.95 b (1.39-6.93)	5.25 ± 0.85 c (2.46-7.71)	220.7917*
pH	7.46 ± 0.29 a (6.4-8.2)	7.27 ± 0.41 b (5.86-8.23)	7.41 ± 0.41 b (5.61-8.42)	7.60 ± 0.39 c (6.25-8.74)	246.7909*
Alkalinity (mg L^{-1})	128.80 ± 38.83 a (44.0-272.0)	93.49 ± 27.05 b (40.0-196.0)	99.33 ± 27.18 b (32.0-200.0)	101.46 ± 30.89 ab (24.0-200.0)	4.1627*
TAN (mg L^{-1})	7.17 ± 2.75 a (0.25-14.9)	4.33 ± 2.04 ac (0.10-13.7)	2.53 ± 1.28 b (0.00-9.2)	2.84 ± 1.41 bc (0.08-11.0)	33.3903*
$N\text{-}NO_2$ (mg L^{-1})	0.82 ± 0.81 a (0.02-5.69)	0.74 ± 0.48 a (0.03-5.65)	2.30 ± 3.52 a (0.01-48.8)	1.49 ± 2.74 a (0.01-15.0)	2.0572^{ns}
NO_3 (mg L^{-1})	145.07 ± 78.55 a (2.4-294.0)	127.37 ± 56.94 a (9.0-272.0)	113.27 ± 64.54 a (0.8-256.0)	27.66 ± 20.89 b (0.8-224.0)	7.7748*
Orthophosphate (mg L^{-1})	60.28 ± 35.68 a (4.65-260.0)	67.19 ± 49.84 a (2.1-271.0)	66.13 ± 51.21 a (1.1-272.0)	26.38 ± 24.86 a (2.95-149.0)	2.1327^{ns}
SS (mL L^{-1})	43.16 ± 19.84 a (7.0-120.0)	39.45 ± 18.65 a (2.0-98.0)	35.88 ± 14.84 a (7.0-85.0)	-	0.7847^{ns}
TSS (mg L^{-1})	895.89 ± 489.86 a (276.54-1486.1)	783.57 ± 456.57 a (280.60-1479.9)	616.38 ± 320.98 a (249.69-1121.81)	-	1.1327^{ns}

MOP - molasses powder; MOL - liquid molasses; SUG - sugar; CTL - control; BFT - biofloc; TAN - Total ammonia nitrogen; $N\text{-}NO_2$ - Nitrite nitrogen; NO_3 - Nitrate; SS - Sedimentable solids; SST - Total suspended solids. Values on the same line with different letters show a statistically significant difference between treatments ($P \leq 0.05$). ^{ns} - not significant, * - significant at 5% by F-test

concentration is equal to approximately 160.5 mg L^{-1} NO_2, five times higher than the lethal concentration for Nile tilapia estimated by Yanbo *et al*. (2006). These authors found that concentrations greater than 28.1 mg L^{-1} NO_2 could cause 50% mortality in tilapia fingerlings after 96 hours of exposure.

According to Azim and Little (2008), nitrite and nitrate accumulation during the first weeks is caused by processes of nitrification, which are very common in biofloc systems. Nitrate accumulation started from the second week of culture in the tanks with bioflocs (MOP, SUG and MOL), and was greater and statistically different to the concentrations in the CTL treatment ($P \leq 0.05$). It should be noted that the water in the tanks of the CTL treatment was renewed weekly, which prevented the nitrate from accumulating (Figure 1C).

Studies of phosphorus dynamics in freshwater systems have revealed that most of the phosphorus from the feed is unusable by the fish and that a relatively large fraction (80-90%) is excreted (BARAK *et al*., 2003). With the increase in biomass, there was a greater accumulation of orthophosphate in the system, this increment apparently being smaller in the CTL treatment due to renewal of the water. Concentrations of orthophosphate varied from 1.1 to 272 mg L^{-1} PO_4^{-3} (Figure 1D) with a mean value of 26.38, 60.38, 67,19 and 66,13 mg L^{-1} PO_4^{-3} in the CTL, MOP, MOL and SUG treatments respectively, with no significant difference between them ($P > 0.05$).

The concentrations of sedimentable solids and total suspended solids varied throughout the 145 days of the culture, reaching maximum values of 120 mL L^{-1} and 1,480 mg L^{-1} respectively, and showing no significant difference between carbon sources ($P > 0.05$). The mean concentrations of the solids agrees with Avnimelech (2012), where the maximum levels for total suspended and sedimentable solids in fish production should be 1,000 mg L^{-1} and 100 mL L^{-1} respectively. There was a need to install settling tanks, operating for approximately 8 hours, during the seventh and fifteenth weeks to reduce the concentration of solids.

The control treatment (with no bioflocs) consumed 102.4 m^3 of water, equivalent to 25.6 m^3 per tank and 8.99 m^3 kg^{-1} of produced fish, whereas the treatment with bioflocs consumed only 0.76 m^3 kg^{-1} (Table 2). This ratio (m^3 kg^{-1}) in the control treatment corresponds to 11.8 times that found in the tanks with bioflocs. This low water consumption in the tanks with bioflocs, i.e. the high efficiency of the sustainable production of biomass, saving water, was also seen by Luo *et al*. (2014). These authors carried out an experimental culture of tilapia in two closed culture systems (bioflocs and recirculation), and obtained a water consumption of 1.67 and 1.0 m^3 kg^{-1} respectively.

Figure 1 - Variation in nitrogen compounds (A- total ammonia nitrogen, B- nitrite nitrogen, C- nitrate) and orthophosphate (D) over 145 days of *O. niloticus* tilapia culture in bioflocs with different sources of carbon

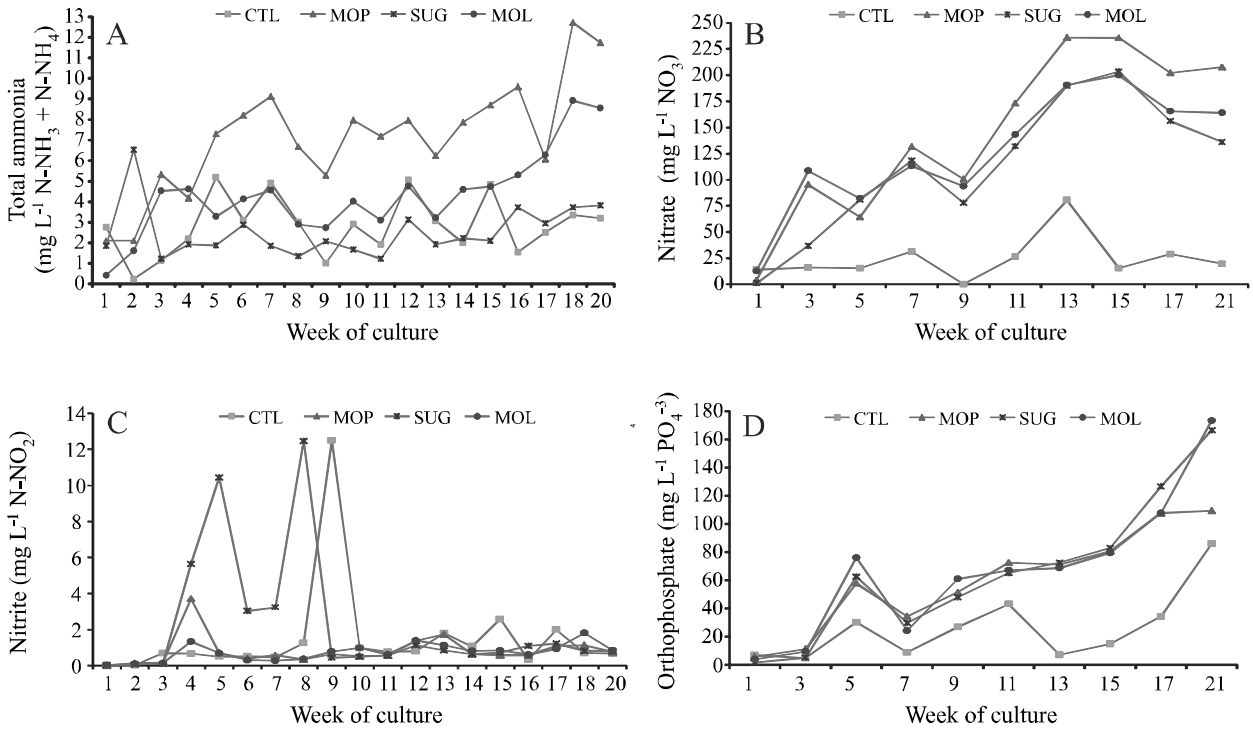

Table 2 - Ratio of water consumption to biomass production for 145 days of *O. niloticus* culture in bioflocs with different sources of carbon

Item	BFT			CTL	BFT[1]
	MOP	MOL	SUG		
Biomass production (Kg)	9.66	9.3	7.78	11.38	26.74
Water consumption (m^3)	7.3	7.3	5.8	102.4	20.4
Ratio (m^3 Kg^{-1})	0.75	0.78	0.74	8.99	0.76

MOP - molasses powder; MOL - liquid molasses; SUG - sugar; CTL - control; BFT - biofloc; [1] - Represents the three treatments with bioflocs

Evaluation of fish growth was carried out from the variables shown in Table 3. Final growth, weight gain, daily weight gain and specific growth rate showed no difference (P>0.05) between the biofloc treatments (MOP, MOL and SUG), however the CTL treatment differed (P≤0.05) in relation to the MOL only. Fish from the control treatment (with no bioflocs) had a higher mean final weight (409.84 g) when compared to those grown in bioflocs in the MOL treatment (339.21 g). This result is the opposite of that found by Azim and Little (2008), where fish from the treatment with bioflocs grew more (140.72 g) when compared to the treatment with no bioflocs (127.51 g).

In the present study, daily weight gain varied from 1.84 to 2.36 g, these results being higher than those found by Pérez-Fuentes *et al*. (2016), who obtained mean values between 0.95 and 1.24 g day^{-1} studying the effects of C to N ratios on the culture of *O. niloticus* in bioflocs. Survival was greater than 80% in all treatments, with no significant difference between them (P>0.05). Although not statistically different from the others, the SUG treatment had the lowest value for mean survival (80.35%) and a higher standard deviation in one of the four plots, since this variable was equal to 53.6%. However, by disregarding the low survival rate in this one plot, the mean value for the SUG treatment rises to 89.3%. Due to the constant contribution of carbon sources to controlling the ammonia nitrogen, mortality occurred in one replication of the SUG treatment after the addition of 125g of sugar m^{-3}. That amount of carbon added to the tank was enough to consume almost all of the oxygen dissolved in the water. Consequently, there was a sudden drop in pH (7.6 to 4.5), which resulted in acidification of the culture water and contributed to the total mortality of that one plot.

In general, the survival rates in this study are similar to those found by Luo *et al*. (2014) (100%), who evaluated the growth, enzyme activity and well-being of Nile tilapia grown in a recirculation system and in bioflocs. Previous studies have confirmed that bioflocs contributed substantially to the growth and production of tilapia, which are known to use *in situ* particles of food, such as bacteria in suspension (AVNIMELECH, 2007; AZIM; LITTLE, 2008; LITTLE *et al.*, 2008; YUAN *et al*., 2010).

The final biomass varied from 7.78 to 11.38 kg between treatments, resulting in a productivity of around

Table 3 - Mean values ± standard deviation for the growth variables of Nile tilapia *O. niloticus* grown in bioflocs with different sources of carbon

Variable	Treatment				ANOVA
	MOP	MOL	SUG	CTL	(F value)
Final weight (g)	350.08 ± 6.6 ab	339.21 ± 20.67 b	353.26 ± 63.71 ab	409.84 ± 23.69 a	3.7801*
Weight gain (g)	274.6 ± 8.18 ab	264.91 ± 17.61 b	284.29 ± 58.43 ab	340.28 ± 20.25 a	8.5807*
DWG(g dia^{-1})	1.91 ± 0.06 ab	1.84 ± 0.12 b	1.97 ± 0.41 ab	2.36 ± 0.14 a	8.5176*
SGR (% dia^{-1})	1.07 ± 0.08 ab	1.05 ± 0.02 b	1.13 ± 0.09 ab	1.23 ± 0.06 a	9.126*
Survival (%)	98.57 ± 1.96 a	97.86 ± 3.19 a	80.35 ± 18.78 a	99.10 ± 1.79 a	8.4897[ns]
FCR	1.75 ± 0.06 a	1.74 ± 0.05 a	1.89 ± 0.36 a	1.61 ± 0.09 a	5.5656[ns]
Final biomass (kg)	9.66 ± 0.27 ab	9.3 ± 0.74 ab	7.78 ± 1.51 b	11.38 ± 0.73 a	12.293*
Productivity (kg m^{-3})	12.08 ± 0.33 ab	11.63 ± 0.92 bc	9.72 ± 1.88c	14.22 ± 0.92 a	11.2635*

MOP - molasses powder; MOL - liquid molasses; SUG - sugar; CTL - control; BFT - biofloc; DWG - Daily weight gain; SGR - Specific growth rate; FCR - Feed conversion ratio. Values on the same line with different letters show a statistically significant difference between treatments (P≤0.05). [ns] - not significant, * - significant at 5% F-test

9.72 and 14.22 kg m^{-3}. The CTL treatment showed higher productivity than the other treatments (MOP, MOL and SUG), differing from the MOL and SUG ($P \leq 0.05$), but not from the MOP ($P > 0.05$). However, it should be noted that to obtain this high productivity in the CTL treatment (14.22 kg m^{-3}), a volume of water 11.8 times greater than that for the biofloc tanks was used. These results agree with those of Lima *et al*. (2015), who found values for productivity of 6 to 16.5 kg m^{-3} when studying the effects of stocking density on the culture of tilapia in bioflocs using liquid molasses as the source of carbon.

Pérez-Fuentes *et al*. (2016), farming tilapia in bioflocs in circular tanks of 3 m^3 at an initial biomass of 2.85 kg m^{-3}, obtained a productivity of 16.28 to 18.03 kg m^{-3}, similar to that found in the present study. However, the values for productivity obtained by Luo *et al*. (2014) of 28.87 (recirculation system) and 36.95 kg m^{-3} (biofloc system) were much higher than those in the present study. These data agree with Avnimelech (2005), where he states that a productivity of 10 to 40 kg fish m^{-3} can be obtained in tanks with biofloc technology.

The feed conversion ratio (FCR) was between 1.61 and 1.89 and showed no difference ($P > 0.05$) between treatments, achieving better results than Azim and Little (2008), who used respectively a system with and without bioflocs in the culture of tilapia (3.51 and 4.97), and by Rakocy *et al*. (2004), who found an FCR of 1.9 and 2.2 for fish with a mean weight of 678 and 912 g at densities of 25 and 20 fish m^{-3} respectively. These data agree with studies by Avnimelech (2007, 2009), who states that bioflocs contribute positively to fish growth, since microbial flakes are composed of bacteria, zooplankton, protozoa and microalgae, and contain approximately 61% crude protein.

The centesimal composition of the bioflocs can vary according to the species produced, the environmental conditions, feeding habits, duration of the culture and the presence of specific microorganisms (AVNIMELECH, 2007; CHAMBERLAIN *et al*., 2001). According to Azim and Little (2008), bioflocs that contain more than 38% crude protein, 3% lipids, 6% fibre and 12% ash (based on the dry matter) are considered suitable for the production of tilapia. The amount of protein found in the bioflocs in this study (Table 4) varied from 31.5 to 33.4% and was slightly lower than that suggested by Azim and Little (2008), however these values are higher than those found (23.7-25.4%) by Elías *et al*. (2015).

The chemical composition of the tilapia fillets is shown in Table 4. The results of the present study were close to the data found by Simões *et al*. (2007) for fillets of *O. niloticus*, with 77.13% moisture, 19.36% protein, 2.60% lipids and 1.09% ash. The minimum and maximum values for the centesimal composition of the Nile tilapia are moisture (76.8-79.1%), protein (17.0-21.0%), lipids (0.99-2.07%) and ash (0.65-1.09%) (GRYSCHEK; OETTERER; GALLO, 2003; MOREIRA, 2005; VILA NOVA; GODOY; ALDRIGUE, 2005).

The results of the sensory analysis of the tilapia fillets are shown in Table 5. When the colour attribute of the fillets was analysed, the tasters preferred those from the SUG treatment, giving a score of 7.57 (like moderately to like very much). In fact, the fillets grown in the biofloc with sugar had a pale white colour, different to those from the liquid molasses and molasses powder, which had a slightly brownish colour. The overall appearance had the lowest score (5.4) in the MOP treatment and the highest (7.77 and 6.8 respectively) in the SUG and CTL treatments.

Table 4 - Centesimal composition (%) of tilapia fillets and of bioflocs formed during the 145 days of *O. niloticus* culture with different sources of carbon

Item (%)	Fillet[1]				Biofloc[2]		
	MOP	MOL	SUG	CTL	MOP	MOL	SUG
Moisture	79.61	76.47	79.98	79.20	-	-	-
Protein	17.3	20.28	17.58	18.13	31.55	33.22	33.42
Lipids	1.14	1.24	0.81	0.95	1.16	1.25	1.65
Ash	1.18	1.32	1.31	1.46	19.06	13.68	13.37
Fibre	<0.02	<0.02	<0.02	<0.02	3.50	4.84	6.95

[1 - 2] Values expressed as wet and dry matter respectively

Table 5 - Mean values (minimum - maximum) for the sensory analysis of *O. niloticus* tilapia fillets from culture in bioflocs with different sources of carbon

Variable	Treatment				Kruskal-Wallis (H test)
	MOP	MOL	SUG	CTL	
Color	5.30 a (1 - 8)	5.87 a (1 - 8)	7.57 b (1 - 9)	6.50 a (1 - 9)	33.3115*
Odor	6.13 a (2 - 9)	6.10 a (1 - 9)	6.87 a (4 - 9)	6.33 a (4 - 9)	3.9886^{ns}
Overall appearance	5.4 a (1 - 8)	6.27 ac (1 - 9)	7.77 b (5 - 9)	6.80 bc (2 - 9)	30.3523*

[1]- Refers to the nine-point hedonic scale (1 - dislike extremely 9 - like extremely). Values on the same line with different letters show a statistically significant difference between treatments ($P \leq 0.05$). ^{ns} - not significant, * - significant at 5% by H test

CONCLUSIONS

1. The sources of carbohydrates used (molasses powder, liquid molasses and sugar) were efficient in controlling ammonia and the production of microbial flakes;
2. The culture of tilapia in bioflocs proved to be an environmentally sustainable activity, since it was possible to use a volume of water that was 11.8 times less than compared to the system in clear water;
3. Values for productivity greater than 9.7 kg *O. niloticus* m^{-3} and for survival of over 90% can be achieved using a biofloc system;
4. Sugar as a source of carbon has a positive effect on the visual aspect of tilapia fillets and on their acceptability in the consumer market.

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