Dietary supplementation with Arthrospira platensis in tambatinga $(\cite{Colossoma\ macropomum} \times \cite{Piaractus\ brachypomus})^1$

Suplementação alimentar com *Arthrospira platensis* na alevinagem de tambatinga ($\bigcirc Colossoma\ macropomum \times \bigcirc Piaractus\ brachypomus)$

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ABSTRACT - The aim of this study was to evaluate different levels of dietary supplementation with dried *Arthrospira* platensis meal during the breeding season of the tambatinga hybrid, using zootechnical performance, haematological parameters and percent composition of the fillet, and by measuring the parameters of water quality. Experimental breeding lasted 64 days. The fry, with an initial weight of 3.56 ± 0.02 g, received diets with two levels of supplementation (20 and 40%) and one control (commercial feed, without the addition of *A. platensis* meal). The water quality parameters over safe limits were total ammonia (NH₃), nitrite (NO₂⁻), and nitrate (NO₃⁻), which increased as the level of supplementation increased, but did not cause any mortalities. The best zootechnical results for final weight, weight gain, feed conversion and feed efficiency were obtained with the treatments that included the addition of 20 and 40% meal. The highest levels of crude protein (CP) in the fillets were found in the treatments with added supplement (20 and 40%). For the haematological parameters, the total number of erythrocytes, haematocrit, haemoglobin concentration, total leukocytes and total plasma protein had higher values for the treatments with added meal, both 20% and 40%, when compared to the control treatment.

Key words: Pisciculture. Nutrition. Zootecnhical performance.

RESUMO - O trabalho teve como objetivo avaliar a suplementação alimentar de farinha seca da *Arthrospira platensis* em diferentes níveis, durante o período de alevinagem do híbrido tambatinga, por meio do desempenho zootécnico, parâmetros hematológicos, composição centesimal dos filés e aferição dos parâmetros de qualidade da água. A criação experimental teve duração de 64 dias. Os alevinos, com peso inicial de 3,56±0,02 g, receberam rações com dois níveis de suplementação (20 e 40%) e um controle (ração comercial, sem a inclusão da farinha de *A. platensis*). Dos parâmetros de qualidade da água, os que se encontraram acima dos limites seguros foram à amônia total (NH₃), nitrito (NO₂⁻), e nitrato (NO₃⁻), aumentando conforme o maior nível de suplementação, porém não ocasionaram mortalidades. Os melhores resultados zootécnicos de peso final, ganho em peso, conversão alimentar e eficiência alimentar foram obtidos nos tratamentos de 20 e 40% de inclusão da farinha. Os maiores teores de proteína bruta (PB) nos filés foram encontrados nos tratamentos que receberam a suplementação (20 e 40%). Com relação aos parâmetros hematológicos, o número total de eritrócitos, hematócrito, concentração de hemoglobina, leucócitos totais e proteína plasmática total apresentaram valores superiores nos tratamentos com inclusão de farinha, tanto 20 como 40%, quando comparado ao tratamento controle.

Palavras-chave: Piscicultura. Nutrição. Desempenho zootécnico.

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INTRODUCTION

Tambatinga is a hybrid fish, the result of crossing females of the tambaqui (*Colossoma macropomum* Cuvier, 1818) with males of the pirapitinga (*Piaractus brachypomus* Cuvier, 1818), whose tropical rheophilic species are native to the Amazon basin, and have been commercially exploited for decades (SOUZA *et al.*, 2012). In 2017, the production of round fish, which include hybrids of the tambaqui, pacu and pirapitinga, was 192.7 thousand tons, representing 38.7% of domestic aquaculture production (CARVALHO FILHO, 2018). Fish farming is an agricultural sector that has grown extremely quickly over the last decade, and improving animal performance and resistance to disease are major challenges faced by producers (DELHI BAI; REDDY; KALARANI, 2014).

In recent years, large investment has been channelled into research aimed at guaranteeing nutritional aspects that would meet the physiological needs for the growth, maintenance, reproduction and health of aquatic organisms. Fish nutrition is a science that is linked to the requirements of each species and to the phase of development, and which seeks greater growth efficiency in order to reduce the costs of aquaculture production (WATTERS *et al.*, 2012). A balanced diet nourishes the animal in addition to boosting the defence system, enabling a better zootechnical response and animal health under stress factors that lead to economic loss for the sector as a whole (SIGNOR *et al.*, 2010).

An ingredient becomes viable due to certain attractive properties, such as availability and competitive price, in addition to easy handling, transport and storage. Several protein sources have been tested, including animal by-products and single-cell proteins, such as microalgae and bacteria (ABDULRAHMAN, 2014). The genus *Arthrospira* is important, a freshwater, blue-green, filamentous cyanobacterium, which contains about 60-70% proteins, vitamins and minerals, and essential fatty acids, such as palmitic, linolenic and linoleic acid (DELHI BAI; REDDY; KALARANI, 2014; MOREIRA *et al.*, 2013).

Arthrospira platensis Gomont, 1892 is recognised as a nutritional supplement due to the various benefits to human and animal health, improving immunity and the ability to absorb nutrients (KRISHNAVENI; PALANIVELU; VELAVAN, 2013; PROMYA; CHITMANAT, 2011). It may be an appropriate dietary supplement in feeding aquatic organisms, as it has been shown that cyanobacteria, besides being an alternative source of protein, can also be used to improve the colour, taste and quality of the meat (TONGSIRI; MANG-AMPHAN; PEERAPORNPISAL, 2010).

When used in aquaculture, studies have reported the success of supplementation with A. platensis in feeding catfish, (Claria gariepinus), which showed favourable growth with greater weight gain and specific growth rate (PROMYA; CHITMANAT, 2011). Oral administration in tilapia (O. niloticus) increased specific antibodies against different antigens and maximised survival when challenged by Aeromonas hydrophila infection (RAGAP; KHALIL; MUTAWIE, 2012). Ibrahem, Mohamed, and Ibrahim (2013) used the dry biomass in feeding tilapia, and saw a positive improvement in health conditions, in addition to enhanced non-specific immunity and resistance to challenges from Pseudomonas fluorescens. However, the use of A. platensis biomass in fish diets is still open to question regarding the optimal level to be added.

The aim of this study therefore, was to evaluate different levels of dietary supplementation with the dry meal of *A. platensis* during the breeding season of the tambatinga hybrid, by means of zootechnical performance, haematological parameters, percent composition of the fillet and the parameters of water quality.

MATERIAL AND METHODS

The research was carried out at the Indoor Cultivation Laboratory of the Centre for Applied Biotechnology in Aquaculture, Department of Fisheries Engineering, Federal University of Ceará (UFC).

One thousand fingerlings of the tambatinga hybrid $(0.5 \pm 0.01~g)$ were purchased from a commercial fish farm located in the city of Tutóia, in the state of Maranhão. In the laboratory, the fingerlings were transferred to two polyethylene water tanks (500 L) with a supply of oxygen, where they remained for one week for acclimatisation and were fed four times a day on commercial feed including 40% crude protein (CP). The fingerlings were then transferred to a brick tank (2,000 L) under constant aeration, where they were fed three times a day with the same commercial feed for 30 days, until reaching the ideal experimental weight.

Nine rectangular polyethylene reservoirs ($56 \times 36 \times 32$ cm), with a working volume of 60 L were used, with oxygen induction by means of an air compressor via a distribution system comprising hoses and porous stones. The experimental design was completely randomised, with two treatments (T-01 and T-02) and one control (C), each carried out in triplicate. For the treatments, the fish received commercial feed with 28% CP supplemented with 20 or 40% *A. platensis* meal respectively; while for the control the animals were given the feed only, as per El-Sheekh *et al.*, (2014).

The density (8 fish reservoir⁻¹) of the experiment gave a total of 72 fish, with an initial mean weight of 3.56 ± 0.02 g and length of 5.78 ± 0.06 cm. The feeding frequency was three times a day, at a rate of 10% of the stored biomass, as per Silva (2015). The experiment lasted 64 days.

Biometrics were carried out weekly to monitor zootechnical development and adjust the feed. The fish were weighed on a digital balance (0.001 g) and measured individually with a conventional calliper. Eugenol, prepared in absolute alcohol, was used as an anaesthetic for handling the fish, as per Rotili *et al.* (2012), at a concentration of 0.5 to 0.9 mL L⁻¹, based on the weight of the animals (0.5 mL L⁻¹ for fish up to 10 grams, 0.6 mL L⁻¹ for fish up to 20 grams, 0.7 mL L⁻¹ for fish up to 30 grams, 0.8 mL L⁻¹ for fish up to 40 grams, and 0.9 mL L⁻¹ for fish up to 50 grams).

During the experiment, around 50% of the working volume of water was exchanged every morning by siphoning, to remove leftover feed and waste.

A. platensis biomass was obtained from the Prof. Raimundo Saraiva da Costa Aquaculture Station, of the Department of Fisheries Engineering, UFC. Cultivation of the cyanobacteria was carried out in an external environment of 1,000-L tanks in a recirculating system integrated with Nile tilapia at 10% salinity. Upon reaching the end of the exponential growth phase, the biomass was collected. The wet biomass was filtered through a 60-μm screen, washed with deionised water to remove the salt, and then dried in an oven at 60 °C. The green biomass contained 54.25% CP, 5.54% moisture, 12.75% ash and 3.20% lipids.

The commercial feed used in the fish diet contained 120 g kg⁻¹ moisture, 280 g kg⁻¹ (28%) CP, 35 g kg⁻¹ ether extract, 125 g kg⁻¹ mineral matter, 120 g kg⁻¹ crude fibre, 35 g kg⁻¹ calcium, and 6000 mg kg⁻¹ phosphorus. To prepare the feed, the pellets were ground and sieved to the required particle size, and the *A. platensis* meal was then added, using 5% powdered gelatine (colourless and flavourless) as a binder, diluted in 500 mL of warm water, and finally added to the dry ingredients. The resulting paste was pressed and then dehydrated in an oven at 60 °C for 36 hours. After this time, the formulated material was stored in plastic containers (-20 °C) until required.

Water quality parameters were analysed once a week during the morning. The dissolved oxygen (mg L⁻¹) and the temperature were checked using a HANNA oximeter. The pH was measured with a benchtop pH meter. The total concentrations of ammonia-N (mg L⁻¹), nitrite-N (mg L⁻¹) and nitrate-N (mg L⁻¹) were determined using a HACH DR 2700 spectrophotometer, following the manufacturer's instructions.

With the data obtained from the weekly biometrics (weight and length), the following zootechnical parameters were calculated: Survival-S (%), Weight Gain-GP (g), Mean Growth-MG (cm), Specific Growth Rate-SGR (%), Apparent Feed Conversion-AFC, Feed Efficiency-FE (%) and Protein Efficiency Ratio-PER (%) as per Moraes *et al.* (2009), using the following equations:

$$S(\%) = Nf* 100/Ni$$
 (1)

where:

S = survival rate (%);

Nf = final number of fish;

Ni = initial number of fish.

$$WG(g) = MWf - MWi (2)$$

where:

WG = weight gain (g);

MWf = final mean weight (g);

MWi = initial mean weight (g).

$$MG(cm) = MGf - MGi \tag{3}$$

where:

MG = mean growth (cm);

MGf = final mean growth (cm);

MGi = initial mean growth (cm).

$$SGR(\%) = LnWf - LnWi/T*100$$
 (4)

where:

SGR = specific growth rate (%);

Ln = neperian logarithm;

Wf = final mean weight (g);

Wi = initial mean weight (g);

T = period of the experiment (d).

$$AFC = QF/GB \tag{5}$$

where:

AFC = apparent feed conversion (g feed.g fish⁻¹);

QF = quantity of feed consumed (g);

GB = gain in biomass (g).

$$FE(\%) = WG*100/QF \tag{6}$$

where:

FE = feed efficiency (%);

WG = weight gain (g);

QF = quantity of feed consumed (g).

$$PER = WG/QF \tag{7}$$

where:

PER = protein efficiency ratio;

WG = weight gain(g);

QF = quantity of feed consumed (g).

The percent composition of the fillets was analysed in four individuals from each treatment. The fish (with a length of 13.39 cm and mean weight of 35.50 g) were killed by thermal shock; the skin was then removed and the filleting process carried out. The fillets were vacuum packed and stored at -18 °C until analysed. Twenty-five g of each sample were used, the same as for the feed. The samples were analysed as per the Association of Official Analytical Chemists (2000) for moisture, lipids, ash and proteins, with the analyses carried out at the Laboratory for Fish Technology, Department of Fisheries Engineering, UFC.

To euthanise the fish at the end of the experiment, some of the practices recommended by the National Council for the Control of Animal Experimentation were used, following the Guidelines for the Practice of Euthanasia - Normative Resolution 37 (BRAZIL, 2013). The procedure was carried out by surgical anaesthesia, using eugenol at a concentration of 0.9 mL L⁻¹ water for collecting the blood, and the physical method of cooling (thermal shock) for the euthanasia.

Haematologic analysis of the blood collected from the fish was performed as per Ranzani-Paiva *et al.* (2013). Four individuals, anesthetised with eugenol (0.9 mL L^{-1} water), from each experimental unit/treatment underwent blood collection from the caudal peduncle by cross-sectional cut with the aid of a scalpel. From the collected blood (100 μ L) containing sodium heparin (5,000 IU mL = 1.0 mL in 50.0 mL of 0.65% saline solution), 0.3 mL was used in the haematologic evaluation: Hematocrit (%), Haemoglobin level (g dL⁻¹), and Total erythrocyte and leukocyte count, with the samples also analysed on slides (panoptic staining), using no anticoagulant and light microscopy (400x magnification).

The total erythrocyte count (cells \times $10^6~\mu L^{\text{--}1})$ was quantified in a Neubauer haematocytometer. Using 20 μL of blood diluted in 4 mL of Hayem's solution (1:200), the erythrocytes were counted under a microscope at 400x magnification. To determine the haematocrit (%), glass microcapillaries were used, filled to 2/3 of their total volume with homogenised blood and one end sealed with paraffin. The microcapillaries were centrifuged at

10,000 rpm for five minutes, and the separate layers determined with the help of a special ruler to establish the erythrocyte volume. The haemoglobin level (g dL⁻¹) was determined using Drabkin's reagent (Labtest), with 20 µl of blood diluted in 5 mL of reagent; after dilution, readings were taken in a spectrophotometer at an absorbance of 540 nm using distilled water as a blank and the haemoglobin reagent (10 g dL⁻¹, labtest) as the standard.

After the blood had been centrifuged for the haematocrit analysis, 20 μL of the plasma were removed from the centrifuged capillary for an analysis of total plasma proteins. The samples were diluted in 1 mL of Biuruet reagent (Labtest) and placed in a spectrophotometer, previously zeroed with distilled water, for reading at 545 nm; a total protein standard (4 g dL-¹, Labtest) was used to determine the total plasma protein content. The total leukocyte count (cells \times 104 μL -¹) was determined by microscopy using the haemocytometer method after diluting 20 μL of blood in 400 μL of Turk's solution (1:20); a microscope with 400× magnification was used.

The statistical analysis was carried out using the Excel 2013 (Microsoft Corp.) and BioEstat 5.0 software (AYRES *et al.*, 2007), where the data for zootechnical performance, water quality, percent composition of the fillet and feed, and haematological parameters were submitted to single factor variance analysis (ANOVA), with the mean values \pm standard deviation (SD) being determined for the three replications. In cases where there was a significant difference, the mean values of the treatments were compared by Tukey's test (p<0.05).

RESULTS AND DISCUSSION

For the percent composition of the feed, the mean values for moisture and ash showed no difference between treatments (p>0.05); there was however a difference between treatments for the lipid and crude protein (CP) content of the diets supplemented with *A. platensis*, as can be seen in Table 1.

The increased protein in the feed is due to the source used as the supplement being highly proteic. Diets with an insufficient protein content may retard growth, compromise feed efficiency, or even cause immunodepression by mobilising protein from some of the tissue to maintain other vital functions (LIMA; SILVEIRA; TUESTA, 2015). Excessive levels of protein in the diet are neither economically nor environmentally healthy, since protein is the most expensive dietary

Table 1 - Percent composition of the diets used in breeding fingerlings of tambatinga fed different concentrations of *Arthrospira* platensis added to commercial feed (control)

PARAMETER	TREATMENT		
PARAMETER	Control	T-01 (20%)	T-02 (40%)
Moisture (%)	6.16 ± 1.54 a	$5.75 \pm 0.22 \text{ a}$	$5.80 \pm 0.20 \text{ a}$
Ash (%)	10.45 ± 1.40 a	10.72 ± 0.21 a	9.49 ± 0.04 a
Lipids (%)	3.52 ± 0.06 a	$4.12 \pm 0.06 \text{ b}$	$4.58 \pm 0.12 \text{ c}$
Crude Protein (%)	28.11 ± 0.37 a	$32.59 \pm 0.97 \text{ b}$	39.03 ± 0.44 c

Values represent mean values ± standard deviation. Different letters on the same line represent a significant statistical difference (p<0.05)

component, and in excess, increases the excretion of nitrogenous residues (GATLIN, 2010).

For the water quality parameters, the dissolved oxygen, temperature and pH showed no difference between treatments (p>0.05) (Table 2).

The ideal value for dissolved oxygen should be over 4.0 mg L⁻¹. The ideal pH range for proper development, allowing the fish to express all their genetic potential for growth and reproduction, can vary from 6.5 to 9.0 (SÁ, 2012).

The values for total ammonia (NH₃) also showed no difference between treatments (p>0.05); however, the values were above those considered safe for fish farming. Continuous exposure of the fish to concentrations of toxic ammonia above 0.02 mg L⁻¹ may cause irritation and inflammation of the gills, retard growth and increase susceptibility to handling and disease; values above 0.05 mg L⁻¹ can result in mortality (KUBITZA, 2013). Despite the high values, there were no mortalities during the experimental period. This can be attributed to temperature values around 26 °C and a pH in the range of 8.3, which, as seen in Table 2, are within ideal limits, since the toxic form is dependent on these parameters. As such, the higher the pH and temperature, the higher the percentage of total ammonia present as NH₃, the

deionised form, which is the most toxic. When this concentration exceeds $0.5~\text{mg}~\text{L}^{-1}$ it is recommended that the water be exchanged and aeration increased, which also reduces the total through nitrification and water movement (SÁ, 2012). Fifty percent of the water in the breeding structures was exchanged every morning.

The values for nitrite and nitrate were higher in the treatments that received the supplement, showing a difference between treatment T-02 with 40% *A. platensis*, being over the safe limit, and the control (p<0.05). However, the water used in the daily exchanges had a salinity of 0.26, avoiding problems related to nitrite. Chloride ions, when present in sufficient amounts in the water, associate with nitrite receptors in the cells of the fish gills, preventing the absorption of this toxic compound (KUBITZA, 2007).

Dietary supplementation with *A. platensis* had a positive effect on the zootechnical performance of the tambatinga fingerlings, as shown in Table 3.

The final mean weight (g fish-1) was higher in the treatments that received the supplement, with a difference (p<0.05) between treatment T-02 and the control. Weight gain, total growth in length, and specific growth rate showed no difference between

Table 2 - Quality parameters of the water used in breeding fingerlings of tambatinga fed different concentrations of *Arthrospira platensis* added to commercial feed (control)

PARAMETER -	TREATMENT		
	Control	T-01 (20%)	T-02 (40%)
Dissloved oxygen (mg L-1)	$4.80 \pm 0.52 a$	$4.75 \pm 0.56 a$	4.62 ± 0.47 a
Temperature (°C)	$26.46 \pm 0.38 \ a$	26.48 ± 0.33 a	$26.60 \pm 0.36 a$
pH	$8.35 \pm 0.15 a$	8.36 ± 0.23 a	$8.36 \pm 0.26 \ a$
Ammonia-NH ₃ (mg L ⁻¹)	$2.67 \pm 1.95 a$	2.91 ± 2.51 a	$3.08 \pm 3.35 \ a$
Nitrite-NO ₂ - (mg L ⁻¹)	0.07 ± 0.56 a	$1.66 \pm 2.80 \text{ ab}$	$3.88 \pm 3.29 \text{ b}$
Nitrate-NO ₃ - (mg L ⁻¹)	2.38 ± 2.58 a	$3.58 \pm 2.57 \text{ ab}$	$7.65 \pm 5.39 \text{ b}$

Values represent mean values ± standard deviation. Different letters on the same line represent a significant statistical difference (p<0.05)

Table 3 - Zootechnical performance of tambatinga fingerlings fed different concentrations of *Arthrospira platensis* added to commercial feed (control)

PARAMETER —	TREATMENT		
PARAMETER —	Control	T-01 (20%)	T-02 (40%)
Final mean weight (g fish-1)	$31.84 \pm 8.24 a$	35.92 ± 10.08 ab	$38.83 \pm 6.76 \text{ b}$
Weight Gain (g fish-1)	28.01 ± 6.91 a	32.37 ± 4.06 a	35.27 ± 3.13 a
Final growth (cm)	13.12 ± 1.11 a	13.45 ± 0.59 a	13.60 ± 0.51 a
SGR (% dia ⁻¹)	3.43 ± 0.24 a	$3.67 \pm 0.26 a$	3.79 ± 0.22 a
Survival (%)	100 a	100 a	100 a
Protein Efficiency Ratio	0.32 ± 0.04 a	$0.39 \pm 0.03 \text{ ab}$	$0.40\pm0.02\;b$
Apparent Feed Conversion (g g ⁻¹)	3.18 ± 0.29 a	$2.54 \pm 0.15 \text{ b}$	$2.49 \pm 0.12 b$
Feed Efficiency (%)	$32.60 \pm 4.0 \text{ a}$	39.50 ± 2.40 ab	$40.22 \pm 1.88 \ b$

Values represent mean values ± standard deviation. Different letters on the same line represent a significant statistical difference (p<0.05)

treatments (p>0.05). During the experimental period there was no mortality in any of the treatments, with 100% survival by the end of the experiment. There was a difference (p<0.05) in the protein efficiency ratio between treatment T-02 and the control. According to the results the potential of *A. platensis* as a supplement to be included in fish diets can be seen through better animal development. The protein efficiency ratio shows how much of the CP in the diet was converted to body weight (ROSSATO *et al.*, 2014).

The values for AFC (g.g⁻¹) showed a difference (p<0.05) between the treatments that received the supplement and the control, which demonstrates the feed conversion efficiency of the diets receiving *A. platensis*. These results are still considered high, since the ideal value for breeding fish is in the range of 0.9-1.5:1, however, for species of round native fish, there is still a difficulty in finding formulations that might reduce this value. According to Kubitza (2012), feed conversion in tambaqui and its hybrids for diets containing 28% CP is 3.7, and if the feed is supplemented with vitamin C, this value drops to 1.76:1.0.

There was difference in the mean value for food efficiency (%) between treatment T-02 and the control (p<0.05). In a study on the effect of replacing fish meal with *A. platensis* in feeding red tilapia, which evaluated growth and carcase composition, the results showed that growth performance (final weight, weight gain and specific growth rate) increased with the increase in substitution up to 75%, and may have been due to improved feed intake and nutrient digestibility (EL-SHEEK *et al.*, 2014). Research on the use of *A. platensis* as a nutritional supplement or as a substitute for fishmeal in commercial fish feed has shown satisfactory results in several areas, such as immunostimulation, and an improvement in feed efficiency and growth rate.

The mean values for percent composition found in the tambating fillets are shown in Table 4.

The moisture content of the fillet showed values within those found in other fish, which vary between 60 and 85% (OGAWA; MAIA, 1999). There was a difference between treatment T-02 and the control (p<0.05), where the percentage moisture of T-02 was lower. There was

Table 4 - Percent composition of the fillets of tambatinga fingerlings fed different concentrations of *Arthrospira platensis* added to commercial feed (control)

DAD AMETED	TREATMENT		
PARAMETER	Control	T-01 (20%)	T-02 (40%)
Moisture (%)	80.76 ± 0.49 a	80.53 ± 0.04 ab	$79.96 \pm 0.14 \text{ b}$
Ash (%)	$1.16 \pm 0.05 a$	1.12 ± 0.4 a	1.16 ± 0.20 a
Lipids (%)	2.81 ± 0.24 a	2.10 ± 0.16 a	2.24 ± 0.66 a
Crude protein (%)	15.39 ± 0.24 a	16.03 ±0.20 b	17.00 ± 0.25 c

Values represent mean values ± standard deviation. Different letters on the same line represent a significant statistical difference (p<0.05)

no statistical difference in the values for ash or lipids between treatments. The values for lipids showed percentages that are considered low, and are therefore an advantage when consuming and processing fillets of this species. According to Lima, Mujica and Lima (2012), knowing the percent composition or chemical composition of the feed is an essential factor in dietary decision-making.

There was a difference between treatments in the protein content of the fillets (p<0.05), where the percentage was higher as the level of *A. platensis* in the diet increased. The mean values for protein in the fillet show that the increases in dietary protein were used for the deposition of body protein and not as an energy source for the deposition of fat. Similar results for research into replacing fishmeal with *A. platensis* meal at different concentrations (0, 5, 10 and 100%) in the diet of *Pangasianodon gigas*, showed increases in protein in the fish carcase as supplementation increased, with the best result for a supplement of 10% (TONGSIRI; MANG-AMPHAN; PEERAPORNPISAL, 2010).

There was a difference in the total erythrocyte count between treatments (p<0.05), showing an increase for increased levels of *A. platensis* in the diet (Table 5). Knowledge of haematological characteristics is an important tool that can be used as a sensitive and effective index for monitoring physiological and pathological changes in fish (SATHEESHKUMAR *et al.*, 2011). Red blood cell counts are almost always part of the analysis, and this test can help diagnose anaemia and other conditions that affect the red blood cells.

Haematocrit values were higher as the supplement of *A. platensis* in the diet increased, where there was a difference (p<0.05) between treatment T-02 relative to the control. This reflects the ratio of erythrocytes in the blood to leukocytes, thrombocytes and blood plasma, and is one of the most reliable haematological

parameters due to its low variability and low margin of error when determined (RANZANI-PAIVA *et al.*, 2013).

There was a difference (p<0.05) in haemoglobin levels between the treatments that received the supplement and the control. The reduction in haemoglobin may indicate a lower capacity for transporting gases within the body (ARAÚJO *et al.*, 2011). The haematocrit, haemoglobin level and total red blood cell count can indicate the capacity of the fish for transporting oxygen, and is related to the concentration of oxygen available in the breeding environment (SILVA; LIMA; BLANCO, 2012).

There was a difference (p<0.05) in the values for total plasma protein and total leukocyte count between the treatments that received the supplement and the control. An increase in leukocytes can be seen during the start of stress in most fish species, and is considered an attempt to recover from homeostatic imbalance; whereas a decrease can be attributed to weakening of the immune system (SILVA; LIMA; BLANCO, 2012). The effectiveness of *A. platensis* in fish lies in its ability to promote growth, survival and the non-specific immune function against pathogens, as well as in its chemoprotective action (IBRAHEM; MOHAMED; IBRAHIM, 2013).

The results show that the addition of *A. platensis* to fish diets is efficient for improving zootechnical, haematological and bromatological parameters. The results may have been due to the nutritional and immunostimulatory properties of cyanobacteria. *A. platensis* is one of the richest sources of protein and iron, containing eight of the ten essential amino acids, which makes this cyanobacterium an alternative against anaemia and malnutrition in aquatic organisms (ALENCAR *et al.*, 2011).

Table 5 - Haematological parameters in tambating fingerlings fed different concentrations of *Arthrospira platensis* added to commercial feed (control)

PARAMETER -	TREATMENT		
	Control	T-01 (20%)	T-02 (40%)
Erythrocytes (cells \times 106 μ L ⁻¹)	1.02 ± 0.26 a	$1.34 \pm 0.20 \text{ b}$	$1.89 \pm 0.40 \text{ c}$
Haematocrit (%)	27.24 ± 5.52 a	31.81 ± 3.10 ab	$35.05 \pm 5.83 \text{ b}$
Haemoglobin level (g dL ⁻¹)	7.51 ± 2.47 a	$9.42 \pm 1.52 \text{ b}$	$10.12 \pm 1.48 \ b$
Total Plasma Protein (g dL ⁻¹)	3.00 ± 0.31 a	$3.39 \pm 0.36 \text{ b}$	$3.70 \pm 0.27 \text{ b}$
Leukocytes (cells \times 10 ⁴ μ L ⁻¹)	2.15 ± 0.34 a	2.69 ± 0.68 a	$3.71 \pm 1.41 \text{ b}$

Values represent mean values ± standard deviation. Different letters on the same line represent a significant statistical difference (p<0.05)

CONCLUSIONS

- 1. The concentrations of the nitrogenous compounds were higher in treatments T-01 (20% *A. platensis*) and T-02 (40% *A. platensis*), but this did not cause any mortalities:
- 2. The indices of zootechnical performance, final weight gain (g), protein efficiency ratio, apparent feed conversion and feed efficiency, increased with the level of supplementation (20 and 40%);
- 3. The highest crude protein (CP) content in the fillets was found in the treatments that received the supplement (20 and 40%);
- 4. The values for the haematological parameters were higher in the treatments with added meal, at both 20 and 40%, when compared to the control treatment.

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