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Preliminary study using Biofloc as food for weight and size increase in *Chirostoma jordani* (Wollman, 1849) in laboratory recirculation system.

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ABSTRACT

Populations of *C. jordani* fish known as "charal" in Mexico nowadays are decreasing due to modification in habitat, by human and industries, contamination, overfishing, and especially interactions with nonnative introduced species. As consequence, "charal" require immediate attention to maintain and preserve its natural populations through controlled cultures. Culture of this organism was made in five ponds, in each one 60 fishes were placed with an average size and weight of 0.82 ±0.22g and 4.18 ±0.17cm respectively. They fed daily with a commercial food for tilapia micotek extruded 1mm with 45% of protein. Biofloc system was controlled with carbon input (molasses and rice powder) and nitrogen (commercial food) to guarantee C/N=15:1 relation, adequate for bacterial growth. At the end of the 189 days of culture, *C. jordani* reached a gain of 746.42% in weight and 473.40% in total length. *C. jordani* fish had a daily increase of 0.0332 g achieving an increase per day of 1.345%; the increase per day of total length (AGR) reached values of 0.0471 cm, which is the 0.492 of IGR. Utilization of biofloc as protein source added to *C. jordani* diet is important, because it improves the growth in size and weight, and provides live food that this fishes need in their diet.

Key words: *Chirostoma*, biofloc, weight, size, growth rates.

RESUMEN

Las poblaciones del pez *C. jordani*, conocido como "charal" en México, en la actualidad, están disminuyendo debido a la modificación del hábitat, humano e industrial, contaminación, la sobrepesca, y, en especial, las interacciones con especies introducidas, no nativas. Como consecuencia, el charal requiere atención inmediata para mantener y preservar sus poblaciones naturales a través de cultivos controlados. El cultivo de este organismo se llevó al cabo en cinco tinas circulares, en cada una se colocaron 60 peces con una talla y peso promedio de 0.82 ±0.22g y 4.18 ±0.17cm respectivamente. Se les proporcionó diariamente un alimento comercial para tilapia microtek extruido 1mm con 45% de proteína. El sistema biofloc se controló con el aporte de carbono (melaza y polvillo de arroz) y nitrógeno (alimento comercial) para garantizar la relación C/N= 15:1 adecuado para el crecimiento bacteriano. Al finalizar los 189 días de cultivo, *C. jordani* alcanzó una ganancia 746.42% en peso y 473.40% en longitud total. El pez *C. jordani* tuvo un incremento por día de 0.0332 g alcanzando un incremento por día en porcentaje de 1.345; el incremento por día de la longitud total (TCA) alcanzó los valores de 0.0471 cm, que significa el 0.492 del TIC. La utilización del biofloc como fuente de proteínas que se añaden a la dieta de *C. jordanii* es importante, ya que mejora el crecimiento en talla y peso, y provee el alimento vivo que necesitan estos peces en su dieta.

Palabras clave: *Chirostoma*, biofloc, peso, talla, tasas de crecimiento.

INTRODUCTION

C. jordani fish known as "charal" in Mexico has a wide distribution in central plateau. Historically, it has considered as a resource of high

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economic performance, social, cultural and ecological importance (Álvarez del Villar, 1970; Miller et al. 2005). Many indigenous groups that settled and distributed along the river system Lerma Chapala-Santiago depends almost exclusively of its extraction, which represents an important food and economic source for this human populations (Martínez-Palacios et al. 2002; Arredondo-Figueroa et al. 2012). Nowadays, populations of this organism are decreasing due to habitat modification by human and industries, contamination, overfishing, and especially interactions with nonnative introduced species (Miller et al. 2005).

As consequence, charal require immediate attention to maintain and preserve its natural populations. Persons engaged to aquaculture, especially in countries in development, are searching new technologies that improve production costs and that do not alter the ecosystem where this activity is developed. Most aquaculture farming pour toxic wastes into the water or earth, mostly the systems of sustainable and intensive production, which are expensive to operate.

In last years, in Asian countries, appears a popular technology: "biofloc technology" (BFT) (Avnimelech 2009, 2012). Consist in propitiate floc formation, which is constituted by a heterogeneous mix of microorganisms (fungus, algae, bacteria, protozoa and rotifers) and of 30 to 40% of inorganic matter as colloids, organic polymers, cations and dead cells; these can reach a size up to 1000 µm, are irregularly shaped, highly porous and permeable to fluids (Chu and Lee 2004a,b). Flocs are a rich source of amino acids. BFT has been apply in countries like South Korea, Indonesia, Malaysia, Thailand, China, Australia, Hawaii, Brazil, Ecuador, Peru, Colombia, USA, Mexico, Guatemala and Belize, successfully (Emerenciano et al. 2012b).

The aim of this study was to stablish, in laboratory conditions, Biofloc system and analyze the morphology and wealth being grade of *Chirostoma jordani* fishes under this production system. Even though the results are emerging, they are encouraging to continue improvement of technology and use it for a longer period during fish growth.

MATERIAL AND METHODS

Experimental design

Biofoloc system consists of two sections: a 1000L container with vertical aeration from container bottom; five ponds of 250 L each one. Both units are connect with a recirculation system through two electric pumps of 1.0 HP (Fig. 1).

Biofloc production

Tilapia juveniles (75) used for experimental tests have a mean value of length (5.0 cm) and weight (4.2 g). Fishes were placed in 1000 L cylinder beaker and fed daily with a commercial food for tilapia micotek extruded 1mm of company Food High Quality El Pedregal, S.A de C.V., with 45% of protein taking into account the 10% of their body weight and adjusting quantity every 15 days. Biofloc system was controlled with carbon input (molasses and rice powder) and nitrogen input (commercial food) to guarantee relation C/N= 15:1 adequate for bacterial growth.

Chirostoma jordani *(charal) culture*

Culture took place in five ponds, in each pond 60 fishes were placed with an average size and weight of 0.82 ±0.22g and 4.18 ±0.17cm

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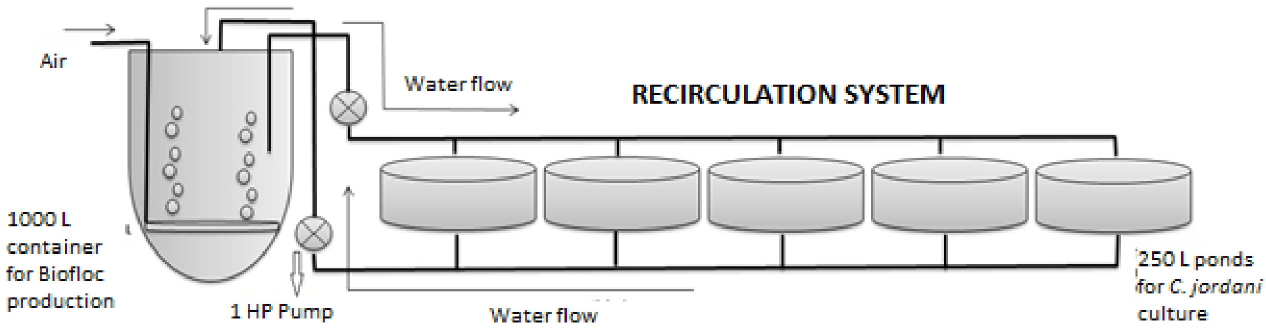

Fig. 1: Used recirculation system in biofoloc production and culture of *C.jordani*

respectively. To these organisms it was daily given, 2.0 g of balanced food for trout microtek extruded 1mm of company Alimentos de Alta Calidad el Pedregal, S.A de C.V.

For measuring and weighing the organisms, they were placed in a glass container with 250 mL of water and 0.75 mL of colloidal solution (®Pentabiocare) during 5 minutes to sedate them and reduce manipulation stress. Organism's size was registered every 21 days using a digital electronic calibrator (®Truper 0/150 mm; 0.01 mm); weight was determined with a digital electronic balance (®Ohaus 400 g; 0.01g). Experiment lasted 189 days.

Values of weight, total length, pattern length, height and width, per sample, were incorporated to a database in Excel 2010, to determine its mean and standard deviation. To determine the absolute rate of growth it was used the next formula:

$$\text{Absolute Growth Rate}$$

$$(\text{AGR}) = \frac{\text{weigth or length}_2 - \text{Weigth or length}_1}{\text{Time}_{\text{final}} - \text{Time}_{\text{inicial}}}$$

To obtain the instantaneous growth rate used following formula:

$$\text{Instantaneous Growth Rate(IGR)}$$

$$\text{IGR} = \frac{\text{LN Weigth or length}_2 - \text{LN Weigth or length}_1}{(\text{Weigth or length}_1)\ \text{x}\ (\text{Time}_{\text{final}} - \text{Time}_{\text{inicial}})}\ \text{x}\ 100$$

RESULTS

In Table 1 the mean obtained values of weight and length are shown with its standard deviation of *C. jordani* during the 189 days of culture, and in Table 2 the weight and length gain of *C. jordani* cultured in biofloc system during culture time. With respect to AGR and IGR in Fig. 2 it is shown that *C. jordani* fish had an increase per day of 0.0332 g during the 189 days of culture, reaching an increase per day of 1.345%; the increase per day of total length (AGR) reached values of 0.0471 cm, which means the 0.492 of IGR.

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Table 1: Mean vales (±D.S.) with weight and length of *C. jordani* cultured in biofloc system during 189 days of experimentation.

Sample (days)	Weight (g)	Total length (cm)	Pattern length (cm)	Height (cm)	Width(cm)
0	0.84 ±0.22	1.88 ±0.22	1.98 ±0.17	0.87 ±0.06	0.34 ±0.09
21	2.71 ±0.140	2.38 ±0.25	2.71 ±0.28	0.92 ±0.06	0.87 ±0.14
42	3.91 ±0.18	3.50 ±0.14	3.84 ±0.14	1.14 ±0.08	1.18 ±0.11
63	4.17 ±0.17	4.58 ±0.13	5.41 ±0.23	1.78 ±0.05	1.46 ±0.14
84	5.31 ±0.23	5.63 ±0.14	5.97 ±0.16	2.05 ±0.07	1.76 ±0.14
105	5.71 ±0.18	6.67 ±0.27	7.01 ±0.18	2.32 ±0.05	1.84 ±0.14
126	6.11 ±0.02	7.71 ±0.18	8.05 ±0.24	2.65 ±0.07	1.98 ±0.09
147	6.51 ±0.02	8.73 ±0.17	9.08 ±0.23	2.89 0.08	2.07 ±0.09
168	6.81 ±0.02	9.76 ±0.27	10.15 ±0.23	3.08 ±0.05	2.52 ±0.12
189	7.11 ±0.27	10.78 ±0.2	11.35 ±0.16	3.17 ±0.07	2.84 ±0.13

Table 2: Weight and length gain of *C. jordani* cultures in biofloc system during the 189 days of experimentation.

	Weight (g)	Total length (cm)	Pattern length (cm)	Height (cm)	Width (cm)
Innitial	0.84 ±0.22	1.88 ±0.22	1.98 ±0.17	0.87 ±0.06	0.34 ±0.09
Finnal	7.11 ±0.27	10.78 ±0.20	11.35 ±0.16	3.17 ±0.07	2.84 ±0.13
Gain	6.27	8.90	9.37	2.36	2.50
Gain (%)	746.42	473.40	473.23	264.36	735.29

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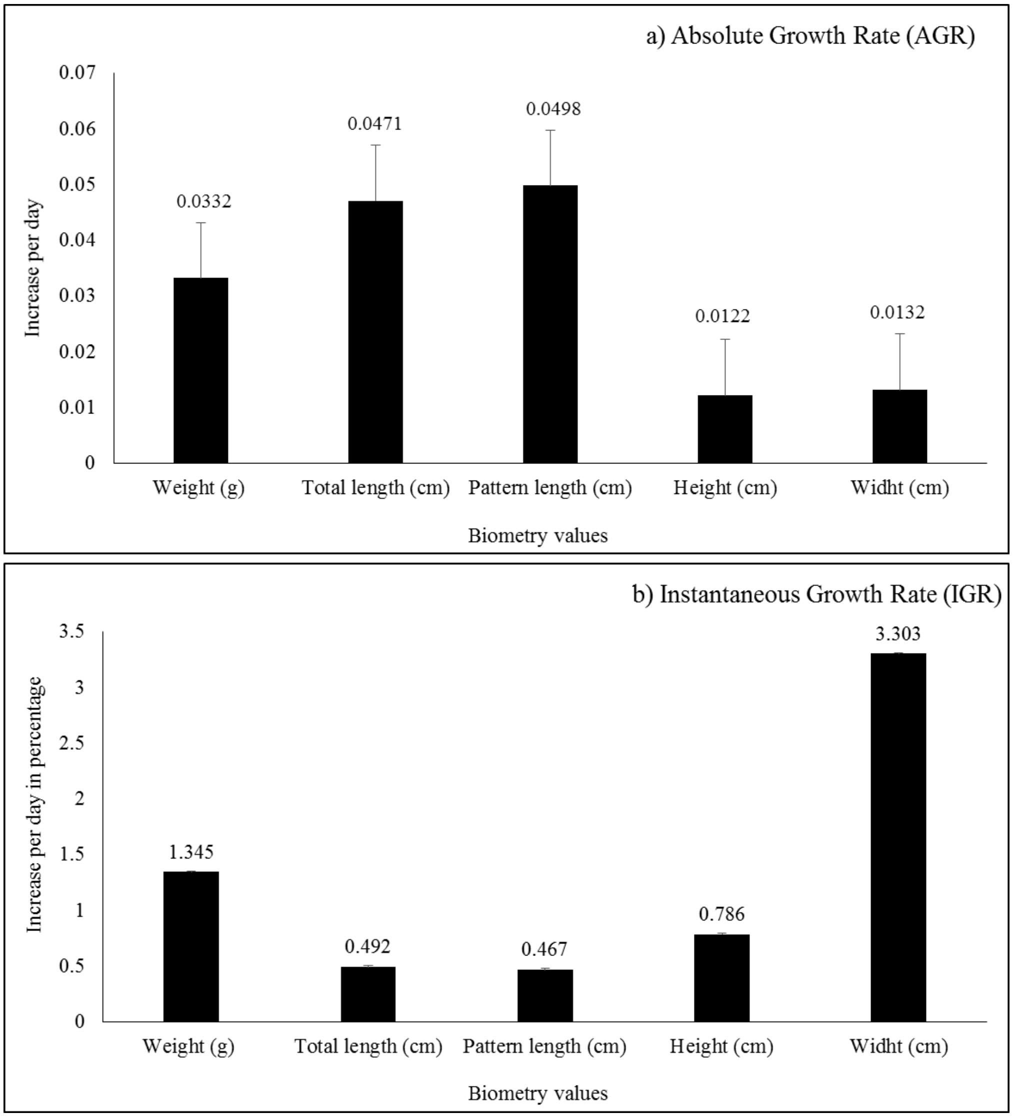

Fig. 2: Absolute growth rate (a) and Instantaneous growth rate (b) of organisms of *C. jordani* cultured in biofloc system during 198 days of experimentation.

In Fig. 3 it is observed the tendency growth curves of weight and length of *C. jordani* during 189 days of culture.

DISCUSSION

Culture of fishes of Atherinidae family is based in its totality in live food (Martinez-Palacios et al. 2004), and balanced diets that are used in controlled cultures of this organism must have a high protein value (Martinez-Palacios et al. 2007). Quantity and quality of supplied protein in fish

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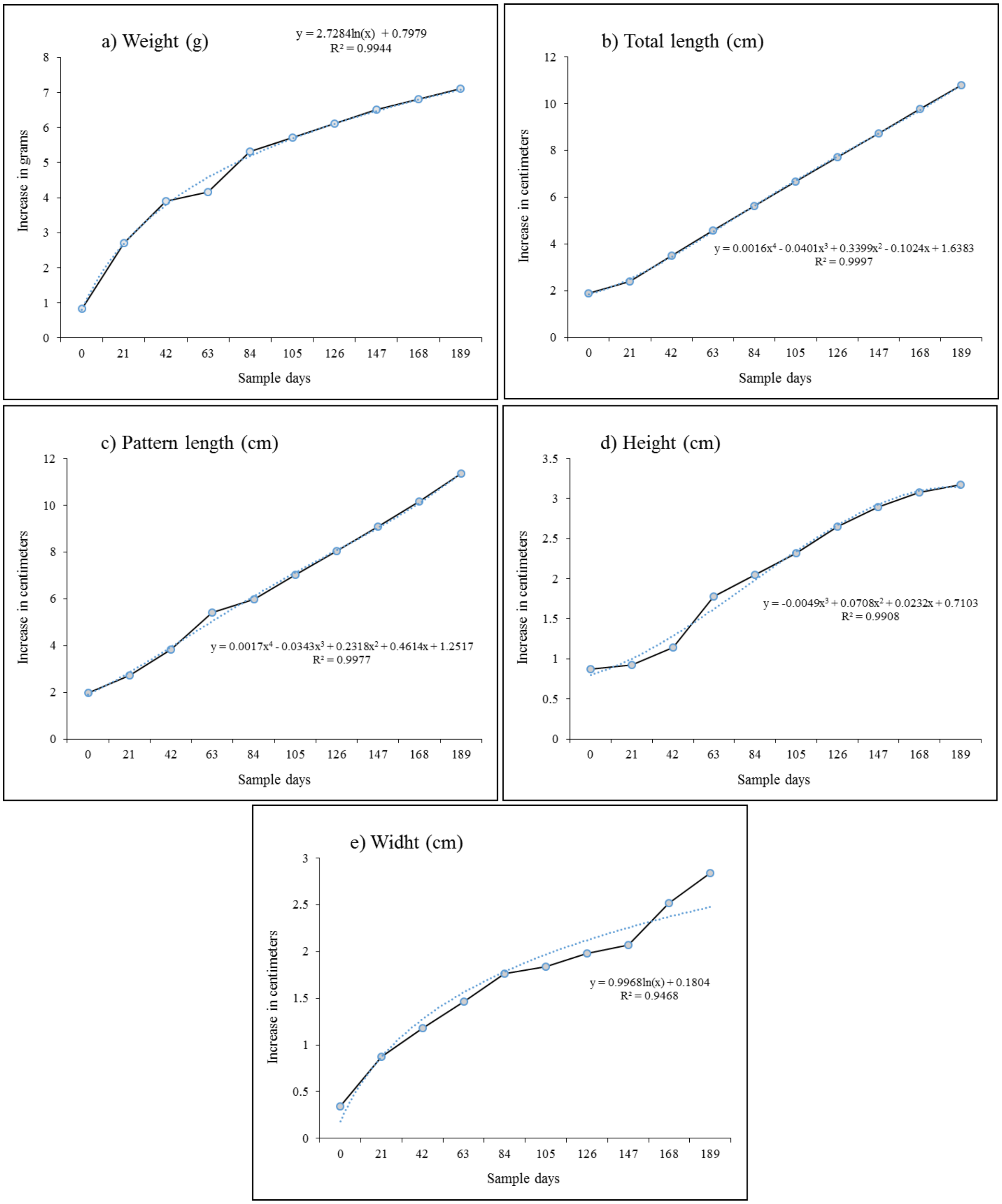

Fig. 3: Tendency growth curves of weight (a), total length (b), pattern length (c), high (d) and width (e) of *C. jordani* cultured with Biofloc.

culture is very important, as reflected in growth tissue, maintenance, reproduction and metabolic functions of fish (Tacon 1989).

Martinez-Palacios et al. (2008), cultivating *C. estor* obtained with a balanced diet rich in protein (400 g kg^{-1}) a gain of weight (%) of 354.60, in experiment with biofloc, it was obtained a better weight gain (%) with 756.42 during the 189 days of culture. Increasing protein levels (450-550 g kg^{-1}) in diet content not showed better growth values for this fishes. Martinez-Palacios et al. (2007) results, suggest that excess of proteins in diet increases additional energy costs for deamination and consequent reduction of energy for growth, as it has observed in many other species of fish (Cho et al, 1985; Tacon and Cowey 1985; Shiau and Huang 1989).

Azin and Little 2008; Crab et al. (2009, 2012); and Monroy-Dosta et al. (2013), mentioned that biofloc increase protein levels in diet because generated flocs, show an adequate proteins, lipids, carbohydrates and ashes content for its use as food in aquaculture, which can be observed in biometric values percentage gains of *C. jordani* cultured in this experiment.

IGR is an indicator of nutritional status, as a good indicator of protein quality, and under controlled conditions the Gain (%) is proportional to essential amino acids that are provided in balanced diet (Tacon, 1989). This is reflect in IGR (1.29-2.69) of *M. estor* that reached the highest values of proteins in the diet used by Martinez-Palacios et al. (2007). Similar levels reached with *C. jordani* fed with biofloc, as it reached an IGR of 1.345.

According to Emerenciano et al. (2012a,b) nutritional quality of biofloc can variate substantially from 12 to 49 and 13 to 46% of raw protein and lipids respectively. Same tendency may occur with levels of PUFA and HUFA (Azim and Litle 2008, Ekasari et al. 2010), this variations can be the result of a different C:N relation, light intensities, salinity and above all, microbiota conformation. Proliferation of bacterial colonies and microorganisms, generate an increase in flocs, this increase must have a density between 10 and 15 mL L^{-1}, to maintain the proper functioning of the systems. The surplus can be used as protein source for organisms, sometimes in a direct way and other time in form of flour or feed (De Schiver et al. 2008 and 2012; Emerenciano et al. 2011; Castro-Nieto et al. 2012. The nutritional value that bioflocs have for culture animals will depend on preference of food, as its capacity to ingest and digest particles in suspension (Azim and Little, 2008).

Because of all this, biofloc utilization as source of proteins that add to *C. jordani* diet is important, as it improves the growth in weight and size, improves survival and provides the live food that these fish need to avoid high mortality and low survival which occurs in controlled cultures.

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