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Effect on length and weight growth of *Pangasius hypophthalmus* (Sauvage 1878) fed with two probiotics.

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ABSTRACT

Sixty juveniles *Pangasius hypophthalmus* fishes were fed with an inert diet enriched with two probiotics: *Bacillus laterosporus* (extracted from *Cherax quadricarinatus* gut) y *Bacillus* sp (La$_3$) (extracted from *Pterophyllum scalare*), during 24 culture weeks. Two acrylic containers (1 m diameter and 0.40 m depth) with 200 L of water were used to introduce 30 organisms each one. The temperature was maintained at 25± 2°C. Every week the organisms were weight with Ohaus digital balance (0-400 ± 0.01 g) and total length measured with a digital Vernier (0.01 mm precision). With these two biometrical data was obtained de Absolute Growth Rate (AGT) and Good Health Factor (GHF). Also every week the organisms were counted to obtain the survival. Initially the organisms have a weight of 1.215 ± 0.219 g and a length of 5.129 ± 0.083 cm. At the end of culture test, with both enriched diets the length were similar (20.225 ± 4.558 cm), but at final weight, the data was significantly different each other. Final weight with *B. laterosporus* was 90.279 ± 26.705 g and with *Bacillus* sp. (La$_3$) was 24.163 ± 6.833 g. Enriched diet with *B. laterosporus* had shown better GHF. Survival is better (47%) with *B. laterosporus*. It´s important use probiotics in diets for this fishes, but is important to consider obtaining their three specific probiotic obtained from their own intestinal gut and make bioassays to saw their qualities in the survival, length and weight growth and why not the reproductive rates of this important ornamental fish or as human food in Mexico.

Key words: *Pangasius hypophthalmus*, enriched diet, probiotics, absolute growth rate, good health factor.

INTRODUCTION

Pangasius hypophthalmus, common name "cat fish" is benthopelagic specie who lives at 6.5 to 7.5 pH and 22 to 26°C temperature ranges. Their food habits are omnivorous (macro algae, zooplankton, crustaceans and some fishes) (FAO 2013). Recently, this fish was an unexpected successful like aquaculture specie. Their productions levels and international market distribution was similar whit other species who leadership the aquaculture market like tilapias, shrimps and trout's (McGee 2010).

Currently in Mexico, you can find this specie principally at ornamental market, but there is a great interest to produce them in biggest ponds. Mexico ranked fifth who importer more *Pangasius* globally (McGee 2010).

Probiotics used as component for fish diet has demonstrate their benefits to increase length and weight of organisms, bacterial control diseases, nutrients source, essential enzymes for better food digestion, elimination of organic matter and increase of immune response (Irianto y Austin 2002; Burr et al. 2005; Balcazar et al. 2006), against pathogen organisms, reducing diseases risks and use of chemical drugs who pollute water habitat (BioMar 1994). The group of probiotics conformed by Gram-negative facultative anaerobia bacteria used at aquaculture generally were founded at intestine gut of fishes and mollusks; the facultative anaerobia bacteria Gram-positive dominate the human and terrestrial animal intestinal gut (Gatesoupe 1999).

The common probiotics used in aquaculture include a wide taxa range, since lactic bacteria (*Lactobacillus, Lactococcus, Bifidobacterium, Pediococcus, Carnobacterium*); bacilli bacteria (*Bacillus, Paenibacillus, Brevibacillus*) and different genus like *Flavobacterium, Cytophaga, Pseudomonas, Alteromonas, Roseobacter, Aeromonas, Nitrosomonas, Nitrobacter* and *Vibrio*; and yeast like *Debaryomyces, Saccharomyces* (Nikoskelainen et al. 2001).

The mean goal of this research was to probe

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two probiotics: Bacillus sp. (La_3) extracted from "angel fish" *Pterophyllum scalare* intestinal gut and *Bacillus laterosporus* extracted from "Australian red lobster" Cherax quadricarinatus intestinal gut, for length and weight growth, good health and survival of *Pangasius hypophthalmus*.

MATERIAL AND METHODS

Sixty juvenile *Pangasius hypophthalmus* of 5.129 ± 0.083 length and 1.215 ± 0.196 g weight were introduced in two acrylic rounded containers (1.0 m diameter x 0.40 m depth), with 200 L of tap water, with constant air supply and 25± 2 °C water temperature. The containers were covered with a mesh to avoid organisms "stress". Thirty organisms were placed for each container.

An inert diet was prepared and the ingredients were shown at Table 1.

Table 1. Ingredients for preparation diet (500 g).

Ingredients	Quantity
Carrot*	2
Broccoli*	1
Spinach*	1 bunch
Tilapia's fillet	300 g
Apple	1
Kiwi	2
Banana	2
Vitamin complex	2000 µg
Vitamin C	0.5 g
Complex B	200 µg
Bacteriological agar	20 g dissolved in 800 mL of tap water

*Previously boiled

All ingredients were mixed with a blender: First, wet ingredients were mixed with 100 mL of tap water and then incorporate the dry ingredients (vitamins). The final mixture was placed in a container to mix with the 800 mL of bacteriological agar. This mixture were divided in two portions (1000 mL) and one portion was added with 1.5 mL of *Bacillus* sp. (La_3) and the other portion with 1.5 mL of *Bacillus laterosporus*. The mix was placed in ice cubs plastic containers, disinfected previously with 96% alcohol to obtain 12 diets cubs (28 ± 1 g each one).

Every four weeks (24 weeks total experiment duration), the total length and weight were taken. Total length with a Digital Vernier (0.01 mm precision) was taken. Weight with Ohaus' digital balance (0-400 ±0.01 g). Daily the containers were siphoned to eliminate organic matter and food not consumed.

With biometric data (length and weight) an Excel 2010 data base was made to obtain mean ±S.D. values, also Absolute Growth Rate (AGR) and Good Health Factor (GHF). The used formulas were:

Absolute Growth Rate (AGR)

$$AGR = \frac{(\text{Length or Weight})_2 - (\text{Length or Weight})_1}{\text{Time}_2 - \text{Time}_1}$$

Good Health Factor (HGF)

$$GHF = \frac{\text{Weight}}{\text{Length}^3} \times 10^5$$

Statistical analysis

With Excel 2010 a database was made using total length and weight values. A descriptive analysis was made to obtain mean ±S.D. values. To obtain significant differences between diet treatments, a one-way ANOVA test was made with SYSTAT 12.0 program.

RESULTS

The mean values ±S.D. are shown in Table 2. The tendency curve of growth increase I total length and weight are shown in Fig.1 and 2.

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Table 2. Mean values of length (cm), weight (g) and ±S.D. (Standard deviation) of *P. hypophthalmus* with different probiotics (24 week culture test).

Week	Length (cm)	Weigth (g)	
		Bacillus laterosporus	***Bacillus* sp. (La_3)**
0	5.129	1.215[a]	1.215[a]
D.S.	± 0.083	± 0.219	± 0.174
4	5.341	1.739[a]	1.643[a]
D.S.	± 0.186	± 0.384	± 0.159
8	5.809	2.666[a]	2.038[a]
D.S.	± 0.486	± 2.128	± 0.609
12	7.032	7.990[a]	3.566[b]
D.S.	± 0.982	± 5.445	± 1.421
16	9.510	21.704[a]	7.149[b]
D.S.	± 1.675	± 10.341	± 2.598
20	13.741	47.803[a]	13.707[b]
D.S.	± 2.566	± 16.815	± 4.139
24	20.225	90.279[a]	24.163[b]
D.S.	± 4.558	± 26. 705	± 6.833

Letras iguales en filas en el peso, señalan no diferencias significativas (P> 0.05) entre los probióticos.

With respect AGR, the diet enriched with *B. laterosporus* obtained an increment of 1.416 g day^{-1}, meanwhile *Bacillus* sp. diet only gain 0.349 g day^{-1}. The significant difference (P<0.05), between both diets were shown at eight week culture test.

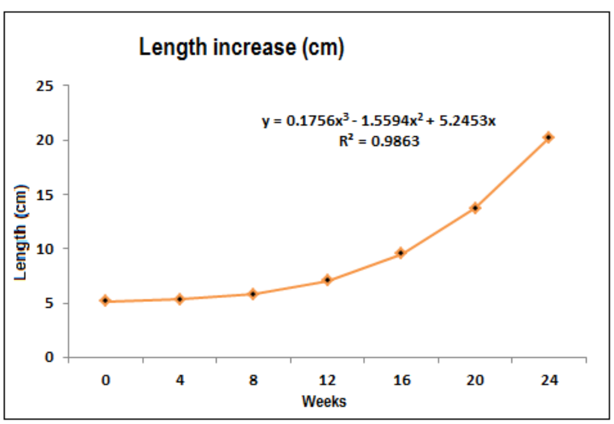

Fig.1. Length increased during 24 culture test weeks of *Pangasius hypophthalmus*.

The weight gain for *B. laterosporus* was 89.064 g, meanwhile *Bacillus* sp. diet gain only 22.948 g.

The GHF for each diet were shown at Fig. 3. *B laterosporus* enriched diet values always shown upper GHF initial value during all 24 culture test weeks, although it began to show a decrease until week 16, because the organism began to lose length and weight asymmetry. With respect *Bacillus* sp. (La_3) diet, the GHF values began to decrease since week four and finished below GHF initial.

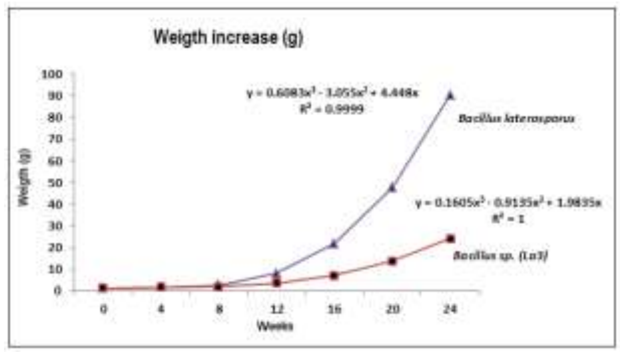

Fig. 2. Weight increased during 24 culture test weeks of *Pangasius hypophthalmus* enriched with *Bacillus laterosporus* and *Bacillus* sp. (La_3).

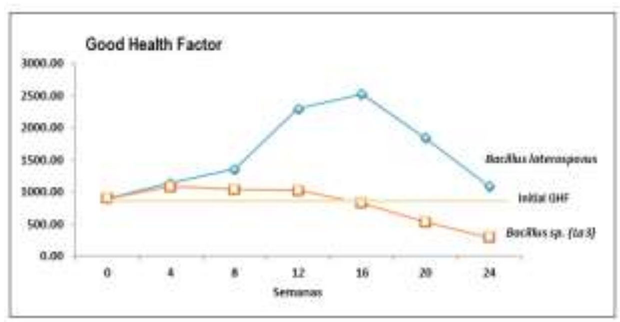

Fig.3. Good Health Factor of *Pangasius hypophthalmus* during 24 culture test weeks.

The organisms fed with *Bacillus laterosporus* shown 47% survival¸ meanwhile *Bacillus* sp. (La_3) diet only has 13% survival.

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DISCUSSION

The diet composition was chosen for the omnivorous habits of *P. hypophthalmus*. It knows that juvenile organism fed from algae, plants, zooplankton and insects and some big adult animals fed fruits, crustaceans and fishes (FAO 2013). That's why is adequate the elaboration and quality of this diet to saw growth in length and weight of organisms, and most important, the diet enrichment with probiotics allows to increase de growth rate of organisms.

At this experiment *Bacillus* sp. (La3) and *B. laterosporus* diets shown significant differences between them until week eight in weight gain, AGR and GHF. The differences doesn't show at length values, in both diets de mean initial value was 5.129 ± 0.083 cm and mean final data was 20.225 ± 4.558 cm. In nature habitat, these organisms can reach maximum total length of 130 cm and 44 kg weight (FAO 2013).

It's probably that these *Bacillus* probiotic diets adequately adhere to the digestive tract of the fish (Vine et al. 2004). Authors like Gildberg et al. (1995) mentioned that these bacteria used as probiotics, in fact, suffer a competitive exclusion in the digestive tract of the host for food or space, so the inclusion of these beneficial bacteria in diets for aquatic animals shown different results on growth animal (Himabindu et al. 2004). The implementation and application of probiotics in diets for aquatic animals is suggested as a prevention measure of diseases (Lara-Flores and Aguirre-Guzmán 2009), and may increase the growth rate (length and weight) or increase of immune response to allow better survival when illness were shown (Aboagye 2008; Mesalhy et al. 2008).

In this research better results were shown when the organisms were fed with *B. laterosporus*. The fishes shown better weight gain, compared with *Bacillus* sp. (La3) diet. The organisms fed with this diet always shown the lowest values. The GHF and AGR were better in organisms fed with *B. laterosporus* that organisms fed with *Bacillus* sp. (La_3) who shown values bellow optimum. This characteristic similarly shown in experiments with tilapia and freshwater shrimp that were fed with a diet enriched with *B. subtilis* (Günther y Jiménez-Montealegre 2004).

According to this, the diet enrichment with probiotics and principally a beneficial bacterium can show different effects on length or weight gains, and consequently good health organisms. That's why is necessary found a specific fish probiotic who allow increase in both cases and consequently good health condition (Verschuere et al. 2000; Geiger 2001).

With respect temperature factor, the mean value was maintained at 22-26°C range (FAO 2013). Because these organisms were poikilothermic, the temperature variation may cause organisms stress and produced a growth decrease due to their impossibility to maintain constant their temperature regulation. Another effect were slow egg development, reduce growth at juvenile stages, decrease food absorption and increase the illness and disease vulnerability (Collección 2013).

With respect survival condition was better with *B. laterosporus* enriched diet (47%) than *Bacillus* sp. (La_3) who only has 13% survival. The low survival values may be due to stress biometry manipulation organisms every week

CONCLUSIONS

B. laterosporus and *Bacillus sp.* (La_3) enriched diet allows the same length gain.

B laterosporus enriched diet allows better weight gain, GHF, AGR and survival values in *Pangasius hypophthalmus*.

It's necessary to obtain their three specific *Pangasius hypophthalmus* probiotics for their own intestinal gut and make length, weight and survival bioassays.

BIBLIOGRAPHY

Aboagye D. 2008. Evaluation of the commercially available probiotic lymnozyme® as an effective control of bacterial infections in channel catfish. Master Degree. Auburn University.

Aguirre-Guzmán G, Ricque D, Cruz-Suárez IE. 2002. Survival of *Saccharomyces cerevisiae* in pelleted

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and extruded shrimp feeds. Aquaculture 208: 125-135.

Ahilan B, G. Shine y R. Santhanam. 2004. Influence of probiotics on the growth and gut microflora load of juvenile Gold fish (*Carassius auratus*). Asian Fisheries Science 17: 271-278.

Balcazar JL, I. de Blas, I. Ruíz-Zarzuela, D. Cunningham, D. Vendrell y JL. Múzquiz. 2006. The role of probiotics in aquaculture. *Veterinary Microbiology* 114: 173-186.

BioMar 1994. Biomar lanzará el primer alimento con probióticos autorizado para su uso en la acuicultura. Chile.

Burr G, D Gatlin y S Ricke. 2005. Microbial ecology of the gastrointestinal tract of fish and the potential application of prebiotics and probiotics in finfish acuaculture. Journal of the World Aquaculture Society 36(4): 425-436.

Collección FAO Capacitación. 2013. http://ftp.fao.org/fi/CDrom/FAO_Training/FAO_Training/General/x6709s/x6709s02.htm

FAO Organización de las Naciones Unidas para la Alimentación y la Agricultura. 2013. Pesca y Departamento de Acuicultura. http://www.fao.org/fishery/culturedspecies/Pangasius_hypophthalmus/en

Gatesoupe FJ. 1999. The use of probiotics in aquaculture. Aquaculture 180:147-165.

Geiger F. 2001. Untersuchungen ueber die wirkung oral verabreichter β-glukane und probiotika bei zierfischen gegenueber infektionen durch fakultativ pathogene bakterien. Tesis de Doctorado, Escuela Veterinaria Hannover.115 p.

Gildberg A, A. Johansen y J. Bogwald. 1995. Growth and survival of Atlantic salmon (*Salmo salar*) fry given diets supplemented whit fish protein hydrolysate and lactic acid bactéria during a challenge trial with *Aeromonas salmonicida*. Aquaculture 138: 23-24.

Günther J y R. Jiménez-Montealegre. 2004. Efecto del probiótico *Bacillus subtilis* sobre el crecimiento y alimentación de tilapia (*Oreochromis niloticus*) y langostino (*Macrobrachium rosenbergii*) en laboratorio. Escuela de Ciencias Biológicas, Universidad Nacional, Heredia, Costa Rica. AP.86-3000.

Irinato A. y B. Austin. 2002. Probiotics in aquaculture. Journal of fish Diseases 25: 633-642.

Lara-Flores, M., Aguirre-Guzmán, G. 2009. The use of probiotic in fish and shrimp aquaculture, A review: Chapter 4. In: N. Pérez-Guerra, L. Pastrana-Castro (eds.), *Probiotics: Production, Evaluation and Uses in Animal Feed.* Research Signpost, Kerala, India.

McGee M. 2010. *Pangasius* for Western Aquaculture. Global Aquaculture The Advocate. The Global Magazine for Farmed Seafood. Vol. 13 No 6. Puerto Rico.

Mesalhy SA, MM Fathi y G John. 2008. Effect of probiotic on the survival, growth and challenge infection in Tilapia nilótica (*Oreochromis niloticus*). Aquaculture Research 39: 647-656.

Nikoskelainen S, S Salminen, G Bylund, AC Ouwehand. 2001. Characterization of the properties of human- and dairy-derived probiotics for the prevention of infectious diseases in fish. Applied and Environmental Microbiology 67 (6): 2430–2435.

Verschuere L, G Rombaut, P Sorgeloos y W Verstraete. 2000. Probiotic bacteria as biological control agents in aquaculture. Molecular Microbiology 64:651-671.

Vine NG, WD Leukes, H Kaiser. 2004. In vitro growth characteristics of five candidate aquaculture probiotics and two fish pathogens grown in fish intestinal mucus. FEMS Microbiol. Lett. 231: 145–152.

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