International Journal of Applied Bioresearch

# Effect of Water Probiotics on Growth Performance of *Channa punctatus*

**R.ArthiManju[#‡], J.Felicitta[#], M.Sakthivel[#], M.A.Haniffa[*], S.Valliammal[#] and G.Chelladurai[#]**

PG. Department of Zoology, Kamaraj College[#], Tuticorin-628003 Tamil Nadu, India, Centre for Aquaculture Research and Extension (CARE), St. Xavier's College (Autonomous), Palayamkottai-627002. TamilNadu, India

Published: 15 November, 2011; Vol. No.1: 25-28; Online: www.bioresjournal.com/documents/ijab0005


Effect of water probiotics for *Channa punctatus* based on growth performance, food conversion ratio and gut microbial load was investigated. *C. punctatus* (2.80-2.83g) was assigned to four treatments, with three replicates each. T1 (0.2mg of *Lactic acid bacillus* / 1000ml of water), T2 (0.4mg of *Lactic acid bacillus* / 1000ml of water), T3 (0.6mg of *lactic acid bacillus* /1000ml of water feed) and T4 (0.8mg of *Lactic acid bacillus* / 1000 ml of water). Fish were fed frequently with a diet of 40% crude protein at a rate of 4% of live body. After 45- days of feeding experiment with control diets, the treated with probiotic showed significantly better results of growth performances and FCR than those with the control diet. There was a difference between the treated fishes and control fishes. The obtained results showed that the weight gain, food conversion ratio, specific growth rate and survival rate were better in higher concentration of probiotics treated in water. The total heterotrophic count in the initial stage was 3.4 X $10^4$ and the final stage was 8.8 x$10^6$. In conclusion that the probiotic treatment in *C. punctatus* of 0.8mg/1000ml water was recommended to stimulate productive performances.

*Channa punctatus*/ Probiotics

In aquaculture water quality plays a vital role in sustaining good health of the fish. Nowadays chemotherapeutic agents usages has led to a search for the alternative remedy of disease control. Use of antibiotics will improve survival of the animal, but they also change the micro biota of the intestine. Such methods may cause the development of resistant bacteria. (Aoki et al. 1985). A novel approach is the use of probiotic bacteria to control the potential pathogens. (Gomez -Gil et al. 2000, Robertson et al. 2000). Nowadays there is a good interest in the use of probiotic bacteria in aquaculture to improve the disease resistance, quality of water and growth of the fish (Verschuere et al. 2000). Aquaculturists face a growing problem of decreasing water quality. Poor water quality is a major stress to all aquatic animals. Stresses are additive and increase the susceptibility of the animals to disease while decreasing their growth rate and feed conversion efficiency. Probiotics create a hostile environment for Pathogens by producing inhibitory compounds (bacteriocins, lysozymes, proteases and hydrogen peroxide).

Author contributions: R.Arthi Manju, J.Felicitta and M.A.Haniffa design of the work; M.Sakthivel-Providing facilities for the study; S. Valliammal performing the work; G.Chelladurai assisting for completion of the work.

The authors declare no conflict of interest.
This article is IJAB direct Email Submission.
Freely available on online through the IJAB open access www.bioresjournal.com.
Received 27, August 2011
Accepted 30, October 2011.

‡To whom correspondence may be addressed.
ram_probiotic@yahoo.co.in
This article contains supporting information online at www.bioresjournal.com/documents/ ijab0005

*Channa punctatus* is the most widespread and common snakehead. Murrels are very much important to promote murrel farming among fish farmers. (Haniffa et al. 2002). Aquatic animals differ from terrestrial animals in the level of interaction between the micro biota and surrounding environment. Trust and Sparrow reported that the different species of gut micro biota diversity can affect the nutrition, growth and susceptibility of disease among fishes. The bacterial load is more in the Gills and intestine. The gut flora can be changed seasonally both qualitatively and quantitatively (Wedemeyer, 1977). *Lactic acid bacteria* are gram positive bacteria. They are mostly found in milk and dairy products. The present experiment was conducted on *C. punctatus* fingerlings treated with water probiotic strain of *lactic acid bacillus* for the growth and adhesion of micro organism in the gut.

## Materials and Methods

### Experimental Design

The experimental fish *C. punctatus* were purchased from the private fish farm, Tirunelveli and transported to Kamaraj College. The fish were acclimatized in cement tanks (3m x 1.5m x 1m). During this period of ten days acclimatization, the fish were fed with control diet (Fishmeal, Jawala acetes, Soy flour, Tapioca flour and Wheat flour). Fifteen fingerlings of *C. punctatus* (Length: 5 ± 0.12cm, weight 2.80 ± 0.03gm) were introduced into the plastic troughs. Triplicates were maintained for each experimental diet (Control diet untreated with Probiotics). The experimental diets were fed thrice a day (9a.m, 1p.m and 5p.m) at a rate of 4% of its body weight per day (Haniffa et al. 1987). Water quality parameters, pH, dissolved oxygen temperature were measured periodically by standard methods (APHA, 1998) and water was changed once in four days. Experimental duration is about 45 days.

### Feed Preparation

When compared with live feeds, semi – moist feeds were widely used in energetic experiment of *C. punctatus* because of their high feed conversion efficiency, easy preparation, less consumption and easy digestion (Haniffa et al. 2002). Control diet (without probiotics treated water medium), T1 (0.2mg of *Lactic acid bacillus* /1000ml water), T2 (0.4mg of *Lactic acid bacillus* / 1000ml water), T3 (0.6mg of *lactic acid bacillus* /1000ml water) and T4 (0.8mg of *Lactic acid bacillus* /1000ml water). The composition and formulation of all diets are given in Table -1. The experimental feeds were prepared using known quantities of ingredients. The selected ingredients (fish meal, jawala acetes, soy flour, tapioca, wheat flour) were powdered and sieved to get fine particles of uniform size. Then the ingredients were weighed according to the formulation and hand kneaded by adding sufficient quantity of distilled water and finally made into dough. The dough was autoclaved for 15 minutes and stored in air - tight containers (Song Zeng Fu et al. 2006).

**Table- 1**: Proximate Composition of Selected ingredients

| Ingredients (%) | Diets |
|---|---|
| | Control |
| Fish Meal | 45 |
| Prawn head waste | 20 |
| Soy flour | 14 |
| Wheat Flour | 10 |
| Tapioca Flour | 5 |
| Fish Oil | 4 |
| Vitamin Mix | 1 |
| Mineral Mix | 1 |

### Screening of Total Heterotrophic Bacterial Count

This study to improve the influence of gut microbial count in particular to total heterotrophic bacteria for once in a fort night in *C. punctatus*. After the completion of experiment, the gut was removed from the fish. The gut was ground in mortar and pestle using sterile saline. The aliquot was serially diluted and plated on Tryptic Soy Agar. After 24hrs of incubation at $32^0$C using the spread plate technique. The bacterial load of gut expressed as number of colony forming units / ml (CFU / ml) (Nallathambi et al. 2002).

## Results and Discussion

The result of dissolved oxygen (DO) content was observed to be higher in treatments (9.0mg/l) than that of the control (8.0mg/l).The pH varies between 6.5-7.1. After 45 days, there was a significant difference between the mean weight of groups T (1-4) the highest weight gain (g) was 9.71 ± 0.15g noticed in T4 (Table 2) The mean values of SGR were significantly

different ($P$<0.05) among the different treatment groups. The highest SGR was observed in T4 (3.46 ± 0.01% / day) and the least FCR was observed in the T4 (2.02 ± 0.08). The total heterotrophic count in the initial stage was 3.4 X $10^4$ and the final stage was 8.8 x $10^6$ (Table 3). The survival rate was also higher in T4 (100%) and the least survival was 86% in control. All the Probiotics treated diets resulted in growth performances and feed utilization better than that of the control diets. (Table 2), suggesting that the treatment of Probiotics reduced the culture cost of *C.punctatus.* Similar results were observed by Sharma and Bhukhar (2000) in *Cyprinus carpio.* They also observed the growth and weight gain of *C. carpio* fingerlings was better in Aquazyn treated waters. That also indicates the bacterial strains and substrains present in Aquazyn played a vital role in enhancing weight of fish. This is also supported by Ghosh et al. (2003) and Swain et al. (1996) in Indian carps. Noh et al. (1994) and Bogut et al. (1998) also proved that the commercial probiotic preparation of *Streptococcus faecium* improved the growth and feed efficiency of Israeli Carp. According to the present study, the results indicated that there existed definite difference in various concentration of probiotic. Among different concentrations T4 showed comparatively better growth performance than others. Among all the treatments, the T4 gave maximum weight gain (9.71 ± 0.15g), followed by T3 (8.65 ± 0.13), T2 (9.00 ± 0.13) and T1 (8.1± 0.09) (Table 2) our findings were similar to Haroun et al. (2006). They found that high concentration of Biogen® resulted in the maximum weight gain in *Oreochromis niloticus*. The results of the present study are also supported by Yanbo and Zirong et al. (2006). Gatesoupe (1991) found significant weight gain in *Schopthalmus maximus* larvae when fed with bio encapsulated *Lactic acid bacteria* and *Bacillus toyoi.*

**Table - 2**: Growth performance of *C. punctatus* treated with probiotics

| Parameters | T1 | T2 | T3 | T4 | Control |
|---|---|---|---|---|---|
| IW | 2.8 ± 0.03[a] | 2.50 ± 0.04[a] | 2.7 ± 0.03[a] | 2.6 ± 0.03[a] | 2.8± 0.05[b] |
| FW | 10.9 ± 0.08[a] | 11.5 ± 0.09[a] | 11.7 ± 0.07[b] | 12.31 ± 0.03[b] | 10.6 ± 0.07[a] |
| WG | 8.1± 0.09[a] | 9.00± 0.13[a] | 9.00 ± 0.13[b] | 9.71 ± 0.15[b] | 7.8 ± 0.09[a] |
| SGR | 3.02± 0.08[a] | 3.11 ± 0.06[b] | 3.13± 0.06[b] | 3.46 ± 0.01[a,b] | 2.97 ± 0.09[a] |
| FCR | 2.80 ± 0.03[b] | 2.26 ± 0.05[a] | 2.15 ± 0.04[a] | 2.02 ± 0.08[a] | 2.91 ± 0.07[b] |
| Survival | 92% | 97% | 98% | 100% | 86% |

The mean values having different superscripts in the same row are significantly different at P< 0.05% level and to ± indicates the standard derivation.

Table- 3: Total Heterotrophic Bacterial Count in Tryptic Soy Agar

| Initial | Diets | Day 15 | Day 30 | Day 45 |
|---|---|---|---|---|
| 3.4 X $10^4$± 1.1 | Control | 3.3 x $10^3$ ±1.1 | 4.7 x $10^3$ ±1.6 | 4.3 x $10^4$ ±1.0 |
| | T1 | 3.7 x $10^4$ ±1.2 | 6.4 x $10^4$ ±1.3 | 4.6 x $10^4$ ±1.1 |
| | T2 | 5.7 x $10^2$ ±1.6 | 7.2 x $10^3$ ±1.3 | 7.7 x $10^5$ ±1.6 |
| | T3 | 5.3 x $10^2$ ±1.5 | 7.5 x $10^4$ ±1.1 | 8.3 x $10^5$ ±1.1 |
| | T4 | 7.3 x $10^2$ ±1.1 | 8.5 x $10^4$ ±1.3 | 8.8 x $10^6$ ±1.2 |

All values are reported as CFU / gm and ± indicates the standard deviation.

Haroun et al. (2006) observed higher SGR (1.98) optimum FCR (1.77) in commercial probiotic Biogen® incorporated feed in *Oreochromis niloticus*. In the present study the Specific Growth Rate (SGR) was maximum (3.46 ± 0.01) in T4. It was followed by T3 (3.17 ± 0.06), T2 (3.11 ± 0.06) T1 (3.02 ± 0.08) and control (2.97± 0.09). There was a significant difference in SGR observed between the treatments. Similar results were also reported by Hidalgo *et al.*, (2006) who found maximum SGR (4.96 ± 0.24) in *Dentex dentex*. The use of probiotics caused a significant survival increase when compared to the control and other treatments the survival of *C. punctatus* in T4 was (100%) greater than other treatments. In the initial stage of the experiments the total heterotrophic bacterial count was found as 3.4 X $10^4$ CFU / gm. In the final stage the microbial load was found to be maximum (8.8 x $10^6$ CFU / gm). This observation was supported by Lekha (2002) in *C. carpio* fed with yeast. It could be concluded that the treatment of probiotics in medium of *C. punctatus* improved the growth performances and gut microflora. Based on our results, use of 0.8 CFU/g/1000 ml of water in *C. punctatus* was recommended to stimulate productive performances.

**Acknowledgement**

The authors wish to express their gratefulness to Dr.J.Mohanraj, Principal, Kamaraj College, Tuticorin for providing necessary facilities and for the encouragement of the research activities.

**References**

Aoki T, Kanazawa T, Kitao T. 1985. Epidemiological surveillance of drug resistant Vibrio anguillarum strains. *Fish Pathol*., 20.199-208.

Apha. 1998. Standard Methods for the examination of water and wastewater (20th edition). (ed. by L.S.Clesceri, A.E. Greenberg and A.D. Eaton), American Public Health Association (APHA) American Water Works Association (AWWA) and water Environment federation (WEF), Washington DC.

Bogut I, Milakovic Z, Bukvic Z, Brikic S, Zimmer R. 1998. Influence of probiotic Streptococcus faecium M74 on growth and content of intestinal microflora in Carp Cyprinus carpio. *Czech J. Anim. Sci.*, 43, 231-235.

Gatesoupe FJ. 1991. The effect of three strains of lactic bacteria on the production rate of rotifers, *Brachionus plicatilis*, and their dietary value for larval turbot, *Scophthalmus maximus*. *Aquaculture*, 96:335-342.

Ghosh K, Sens K, Ray AK. 2002. Growth and survival of rohu, *Labeo rohita* (Hamilton) Spawn fed diets supplemented with fish intestinal microflora. *Acta Ichthylogica et Piscatoria*., 32(1): 83-92.

Gomez-Gil B, Roque A, Turnbull JF. 2000. The use and selectionof probiotic bacteria for use in the culture of larval aquatic Organisms. *Aquaculture*, 191:259-270.

Haniffa MA, Murugesan AG, Flemming T. 1987. Influence of plant and animal food on food utilization of the freshwater carp Labeo rohita(ham). *Curr sci.*, 16: 846-848.

Haniffa MA, Jesu Arokia Raj A, Sethuramalingam TA, Sridhar S. 2002. Digestibility of lipid in different feeds by striped murrel *Channa striatus*. *J. Aqua. Trop*., 17(3):185-191.

Haroun Erel, Ampi GA, Kabir Chowdhury MA. 2006. Effect of dietary probiotic Biogen Supplementation as a growth promoter on growth performance and feed utilization of Nile tilapia Oreochromis niloticus (L). *Aquaculture Research*, 37(14):1473-1480.

Lekha S. 2002. Studies on the influence exogenous enzyme protease and probiont yeast added diets on growth response food transformations. Protein metabolization and gut microbial diversity in common carp, Cyprinus carpio (Linn). Ph.D., Thesis. Institute of Coastal Area - Studies, Manonmanium Sundaranar University, Nagercoil.

Nallathambi T, Easwar M, Kuberaraj K. 2002. Abundance of indicator and general heterotriphic bacteria in Port Blair bay. Andamans. *Indian J. Marine Sciences*, 31(1): 65-68.

Noh SH, Han K, Won TH, Choi YJ. 1994. Effect of antibiotics, enzyme, yeast culture and probiotics on the growth performance of Israeli carp. *Korean J. Anim. Sci*., 36: 480-486.

Robertson PAW, Odowd C, Burrells C, Williams P, Austin B. 2000. Use Carnobacterium as probiotic for Atlantic salmon (Salmo salar L) and rainbow trout (*Oncorhynchus mykiss*, walbaum). *Aquaculture*, 185:235-243.

Sharma OP, Bhukhar SKS. 2000. Effect of aquazyn-tm-1000, a probiotic on the water quality and growth of *Cyprinus carpio* Var. *Communis*. *Indian. J. Fish.*, 47(3):209-213.

Song Zeng FU, Tian - Xing, Aili-Sheng, Zhaug Li-Jing, Zheng Xiao – Long. 2006. Effect of dietary supplementation with Clostridium bulgaricum on the growth performance and humoral immune response in Miichthys miuy. *J Zheijang Univ Science*., B.7 (7): 596-602.

Swain SK, Rangacharyulu PV, Sakar S, Das KM. 1996. Effect of a probiotic supplement on growth nutrient utilization and carcass composition in mrigal fry. *J. Aquac.*, 4, 29-35.

Verschuere L, Rombaut G, Sorgeloos, Verstracte W. 2000. Probiotic bacteria as biological agents in aquaculture. *Micro and Mole. Bio. Rev.*, 64(4): 655-671.

Wedemeyer GA, Meyer F, Smith L. 1977. Environmental Stress and fish diseases. *Thr Pub., Neptune city, NJ*, 200.

Yanbo W, Zirong X. 2006. Effect of probiotics for common carp (Cyprinus carpio) based on growth performance and digestive enzyme activities. *Anim. Feed Sci. Technol.*, 127:283-292.

Hidalgo MC, Skalli A, Abellan E, Rizcun A, Cardenete G. 2006. Dietary intake of probiotics and masilinic acid in juvenile dentex (*Dentex dentex* L.): effects on growth performance, survival and liver proteolytic activities. *Aquaculture nutrition*, 270.