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Annals of Biological Research, 2013, 4 (2):327-329
(http://scholarsresearchlibrary.com/archive.html)

Annals of Biological Research
Scholars Research Library
ISSN 0976-1233
CODEN (USA): ABRNBW

Increasing resistance of larvae of oncorhynchus mykiss to environmental stresses such as temperature and Ph using the probiotic protexin

Mahshid Rezaei[1], Leila Nateghi[2]

[1]*Department of medicinal plant, Varamin-Pishva Branch, Islamic Azad University, Varamin, Iran*
[2]*Department of Food Science and Technology, Varamin-Pishva Branch, Islamic Azad University, Varamin, Iran*

ABSTRACT

In this study the role of the probiotic protexin in tolerance of larvae of rainbow trout (oncorhynchus mykiss) to physicochemical stresses of water was investigated. In this regard protexin at four different concentrations of 0.15, 0.5, 1 and 1.5 g/kg dried food was used. Larvae with average weights of 2.0±90 mg were fed for 60 days. The experiment was carried out at three replications with 530 larvae in each tray. Feeding varied from 4% to 7% live mass in breeding period. Except larvae of the first treatment, larvae of all other treatments showed higher resistance to heat stress compared with control ($p < 0.05$). Also all treatments survived acidic Ph compared to control treatment ($p < 0.05$). No significant difference in tolerance to alkaline Ph stress was observed between experimental and control treatments ($p > 0.05$).

Key words: larvae of rainbow trout, probiotic, protexin, stress, concentrate

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INTRODUCTION

In recent years aquaculture has been one of the most fast- growing parts of food production. However this industry has faced with problems such as variations of water quality, outbreak of some disease and etc. Outbreak of disease as the major problem in aquaculture industry affects economic expansion of this part in many countries of the world, and currently is regarded as a confounding factor in subdivision of shrimp breeding [1]. To solve these problems, solutions have been suggested including disease control and use of antibiotics which led to environmental issues, antibiotic resistance and etc.

In recent years, use of probiotic have many benefits such as enhancement of production, improving water quality, biological control, enhancement of growth, inhibition of disease and reducing detrimental effects of stress [2]. Regarding the role of probiotics in offsetting negative influences of stress, finding the relationship between use of these substances for aquaculture feeding and their effects on stress full conditions is of profound importance. Several studies investigated the effects of probiotics on aquaculture. Gatesoupe et al., (1994) [3] stated that consumption of ratified enriched with lactic acid bacteria led to enhanced resistance to vibrio pathogen (as ahio lgicol stresses en turbot fish).

Gomez- Gill (2000) [4], reported that use of bacterium pseudomonas flurescens AHz reduced rate of mortality in trout infected by V.anguillarun pathogen. Based on Nikoskelainen et al., (2001) [5], using probiotic lactobacillus rhomnosus in food feeding of trout increased its resistance to aeromonas salmonicida. Flores et al., (2003) [6] reported that use of a kind of livestock probiotic in tilapia feeding resulted in growth and resistance of tilapias larvae to stressful conditions caused by ration variation and stock density compared with witness. Regarding scarcity of studies in the field of the effects of probiotics on stressed aquaculture in terms of physicochemical factors of water, the present research was

performed to investigate the role of probiotic protexin in enhancement of resistance of rainbow trout to environmental stresses of Ph and temperature stresses.

MATERIALS AND METHODS

This experiment was carried out a full randomized design with three replications using trays at dimensions of 42.5×42.5 cm and water flow rate 10-15 l/min. The experiment was done at shahid bahonar workshop of free fish proliferation. Each tray was divided into two equal parts by a plastics wall. For each treatment two trays were placed at one trough. 265 larvae after the biometry measurement of weights and length of 90± 0.2 mg were randomly placed each either-part of trays. Before that survival rate, weight and length of larvae were measured. Feeding varied from 4% to 7% biomass throughout the experimental period. Tank bed was siphoned to remove residual food on a two- day's basis. At the end of the experiment i.e. after 60 days, larvae were exposed to temperature stress as well as to acidic and alkaline Ph stress. Temperature shock included high temperature in the range of 20- 24˚c and low temperature of 1˚c [7]. From each replication 30 larvae were randomly removed and transferred to small baskets inside the aquarium of a given temperature. Survival rate of larvae was then measured during a 15 min interval, and Ph shock included basic Ph values of 10.8 and 11.8 as well as acidic Ph values of 3.8 and 5.8 [7]. In order to carry out basic and acidic Ph stress experiments, from each replication 30 and 20 larvae were removed respectively followed by transferring to small baskets into the aquarium of a given Ph. Survival rate was determined during a 15 min interval. This experiment was done in fall of 2007.

RESULTS

A – Results related to resistance of larvae to temperature stress.
After 15 minutes of exposing larvae to temperatures of 20˚c and 1˚c as high and low values of temperature shock no loss was observed. Therefore temperature was increased to 24˚c and 30˚c larvae from each replication were exposed to these conditions for data recording.

Results related to measurement of survival rate of larvae stressed under 24 ˚c are shown at Table 1. As shown in Table 1, the treatments 2, 3 and 4 showed significantly ($p < 0.05$) higher resistance to high temperature than control treatment. No significant difference was observed between treatment 1 and control treatment in resistance to temperature stress ($p > 0.05$).

B – Results obtained from evaluation of resistance to Ph stress alkaline stress
In order to investigate effect of different concentrations of protexin on resistance of larvae to stressful condition, Ph 8.10 was first regarded as alkaline shock, during which no loss was observed. Therefore Ph value was increased to 11.8 and 20 larvae from each replication was exposed to this condition. Results related to survival rate of larvae stressed under Ph 8.11 are indicated at Table 2. Table 2, demonstrated that there is no significant difference between all treatments.

Acidic stress
In order to consider effect of different concentrations of protexin on resistance of larvae to acidic stress, Ph 8.5 was used leading to no losses between treatments therefore Ph 8.3 was selected as acidic shock followed by exposing 30 larvae to this condition.

Table 3, shows respected results. based on following Table survival rates in all treatments were higher than those in control treatment significantly ($p < 0.05$).

DISCUSSION

Incorporation of different concentrations of protexin into the fish diet led to an increase in resistance of rainbow trout to Ph and temperature stresses, with protexin resistance of rainbow trout to Ph and temperature stresses, with protexin concentration of 1 g/kg giving the best results. This difference was significant ($p < 0.05$). Results obtained in this study are in agreement with findings of Gatesoupe et al., (1994) [3], Gomez- Gill et al., (2000) [4], Nikoskelainen et al., (2001) [5] and Flores et al., (2003) [6]. In general stress affects aquatics through weakening immune system [8]. Olafsen (1984) [9], reported that in cod fish and larvae of herring, bacteriophagy followed by alkaline intracellular excretions caused by nucleic lactic acid led to stimulation of immune system. Based on Gatesoupe (1994) [3] autochthonous microbes stimulate immune response of aquatics to pathogens. Furthermore Nikoskedlainen et al., (2003) [10] found that use of lactic acid bacterium, lactobacillus rhamnosus, improved immune levels in trout.
Many other studies indicated positive effects of probiotics on aquatics of which we can refer to Rengpipat et al., (1998) [11], Robertson et al., (2000) [12].

Table 1. Survival rate of larvae exposed to high temperature (24˚c).

Treatments	Formula feeding	Percent survival
1	15.0 gr/kg	66.70±12.01^{bc}
2	5.0 gr/kg	3.33^{ab}33.93±
3	1.0 gr/kg	66.96±0.00^{a}
4	5.1 gr/kg	88.98 ±1.92^{a}
5	Witness	33.63±17.6^{c}

Different letter in a row show significant difference ($p < 0.05$).

Table 2. Survival rate of larvae shock to Alkaline Ph

Treatments	Formula feeding	Percent survival
1	15.0 gr/kg	66.96±5.77^{a}
2	5.0 gr/kg	33.98±2.88^{b}
3	1.0 gr/kg	33.98±2.88^{b}
4	5.1 gr/kg	95.00 ±5.00^{c}
5	Witness	66.96±2.88^{a}

Different letters in a row show significant difference ($p < 0.05$).

Table 3. Survival rate of larvae shock to Acidic Ph

Treatments	Formula feeding	Percent survival
1	15.0 gr/kg	44.94± 9.62^{a}
2	5.0 gr/kg	66.96±3.33^{a}
3	1.0 gr/kg	77.97±1.92^{a}
4	5.1 gr/kg	77.87±10.71^{a}
5	Witness	88.68±6.93^{b}

Different letters in a row show significant difference ($p < 0.05$).

CONCLUSION

Evaluation of effects of four different concentrations of probiotic protexin on resistance of larvae of rainbow trout to environmental stresses such as temperature and Ph showed that probiotic treatments had more resistance than control treatment. Therefore it is recommended to use 1g/kg food protexin when occurring stresses due to physicochemical factors of water.

REFERENCES

[1]. S. Ziyaeiynejad, MSc Thesis, Tehran University, (Tehran, Iran, **2004**).
[2]. H. Mohammadi, Nikotech Company (Husbandry Consulting), (Tehran, Iran, **2002**).
[3]. F.J. Gatesoupe, *Aquat. Living Resour*., **1994**, 7, 227-282.
[4]. B. Gomez-Gill, A. Roque, J. F. Turnbull, *Aquaculture.*, **2000**, 191, 259-270.
[5]. S. Nikoskelainen, A. Ouwehand, S. Salminen, G Bylund, *Aquaculture.*, **2001**, 198, 229-230.
[6]. M. L. Flores, M.A.O. Novoa, B. E. G. Mendez, W. L. Madrid, 2003, *Aquaculture.*, **2003**, 216, 193-201.
[7]. M. K, Mirzakhani, MSc Thesis, Tarbiat Modarres University, (Tehran, Iran, **2005**).
[8]. B. Jalali e Jafari, Parasites and Parasitic Diseases of Freshwater Fishes, Reproduction and Aquaculture Department - Department of Education and Publication, **1999,** P. 564.
[9]. J.A. Olafsen, Ingestion of bacteria by cod ŽGadus morhua L.. larvae. In: Dahl, E., Danielsen, D.S., Moksness, E., Solemdal, P. ŽEds, The Propagation of Cod, Gadus morhua L. Bergen, Institute of Marine Research, **1984**, pp. 627–643.
[10]. S. Nikoskedlainen, A. Ouwehand, S. Salminen, M. E. Lilius, G. Bylund, *Fish and Shell Immunology.*, **2003,** 15, 443-452.
[11]. S. Rengpipat, W, Phianphak, S, Piyatitivorakul, P, Menasveta, *Aquaculture.*, **1998**, 167, 301-313.
[12]. P.A.W. Robertson, C. Dowd, C. Burrells, P. Williams, B. Austin. *Aquaculture.*, **2000**, 158, 235-243.