Effect Whole and Cell Wall of Saccharomycescervisiae in Immunity Factors on Rainbow Trout (Onhorhynchus mykiss)

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Abstract: In order to investigate effects of diets with probiotic Biosaf®containing whole Saccharomyces cervisiae and prebiotic Sofmannan® containing saccharomyces cervisiae cell wall on humoral immune factors several rainbow trout juveniles were studied. this study was performed during august and September 2012 in the reproduction and rearing complex of salmonidaekoohestantalai, firoozkooh (Iran). overaly 200 juvenile with primary weight 100 gr have feed 60 days 4 treatment and 1 control treatment. 2 treatment was contained 2gr/kg and 4gr/kg probiotic Biosaf and 2 treatment was contained 0.3% and 0.6 % prebiotic Sofmannan. while control treatment had no probiotic and prebiotic added. In the first,15th and 30th days of blood serum of rainbow trout the measured parameters in pro and pre treatment and control had no changes on the level of IgM, IgA, C3, C4 and it was no difference between the level of control and pro and pre treatments (p>0.05) But in the 45th day the sampled blood serum of the rainbow trout in pro and pre and control treatments shown that the level of IgM, IgA, C3, C4 in 0.3 % it was significant difference between 4 gr/kg, 2 gr/kg pro and 0.6%pre and control treatment (p<0.05) and the pro treatment had no different with control. at the end of sampling, the level of complement and immunoglobulin has shown that 2 and4 gr/kg pro and 0.3% and 0.6 % pre treatments had significant difference with control (p<0.05).Based on these results it seems that the optimum amount of yeast cell wall as prebiotic dietary supplements Sacchaomyces cervisiae it could be increase earlier the immune response than whole yeast saccharomyces cervisiae as probiotic. after time probiotic and prebiotic increased complement and immunoglobolin. but prebiotic enhanced immune system earlier than probiotic.

Key words: Rainbow trout • Probiotic Biosaf • Prebiotic Sofmanan • Whole Saccharomyces cervisiae • Cell wall Saccharomyces cervisiae. humoral immune factores • IgM • IgA. C3 • C4

INTRODUCTION

A probiotic is defined classically as a viable microbial dietary supplement that beneficially affects the host through its effects in the intestinal tract. This definition, however, was initially intended for use with animal feed products. For human nutrition, the following definition has been proposed " alive microbial food ingredient that is beneficial to health " [1]. Lactobacteria, befidobactrio, yeast and Saccharomyces cervisiae as a probiotic in aquaculture [2].

A prebiotic is defined " as a Nondigestible food ingredient that beneficially affects the host by selectively stimulating the growth and / or activity of one or a limited number of bacteria in the colon "[3].

Cell wall of Saccharomyces cervisiae are constructed almost entirely of β-1,3-D-glucan, β-1,6-D-glucan, mannoproteins and chitin, bound together by covalent linkages [4] and introduce as a prebiotic. we known in vertebrates, each one of these profied compounds that mentioned above increase innate defence mechanisms and / or disease resistance [5-9]. however, very little data exists concerning the use of whole yeast as immune stimulants [10,11]. although their use would make the administration of such substances in fish farms cheaper and easier compared with the use of commercial isolated compounds.
β-glucans and monnoproteins are the main yeast cell wall components, the former enhance fish humoral (immunoglobulin and complement activity) and cellular (phagocytic and respiratpry burst activities and bacterial killing) immune responses both invivo and invivo [12-21]. in the knowledge that whole Saccharomycyes cervisiae, as well as their single cell wall components, can stimulate the fish immune system [10,11].

Rainbow trout culture is economically important in Iran and bacterial infectious diseases in trout farming seems to be the major reason for decreasing the production level in same farms success and failure of fish culture programs are determing by early life stage conditions [22]. some works have been to evaluate competitive exclusions of potential probiotics on rainbow trout [23-25]. Stimulation of immune system in rainbow trout with several candidate probiotics and prebiotics has also been evaluated by some researchers [23, 26-28].

The aim of the work was to test immunostimulant potential of the dietary administration to rainbow trout Onhorhynchusmykiss by a cell wall Saccharomyces cervisiae and to compare the activity with that of a whole yeast Saccharomyces cervisiae possible differences in the yeast cell wall composition and immunostimulant effects of both strains are considered.

MATERIAL AND METHODS

In this study, effect of Saccharomyces cervisiae as a probiotic and cell wall of Saccharomyces cervisiae as a prebiotic were evaluated on humoral immunity factors (IgM, IgA, C3, C4) in rainbow trout.

Experiment Design: The experiment was carried out with five treatment included two levels of probiotic (2 gr/kg, 4 gr/kg ), prebiotics (0.3%, 0.6%) and control without supplementate in from of complete randomized design (CRD). the evaluation was conducted in closed recycling system for a period of 60 days, at farm in Tehran province. Two hundreds fish with 100gr average weight were used for each treatment.

Probiotic and Prebiotic Supplement: Commercial probiotic (Biosaf®) and prebiotic (safmannan®) were used in this experiment contained whole and cell wall of Saccharomyces cervisiae respectively.

Fish Dissection Diet: the fish were collected from each feed trial, every 15 days after 20 h of starvation at the end of experiment [29].

Sampling and Measurement of Blood Serum: Fish were sampled twice a mount for immune parameters at each sampling for enzymatic analyses, three fish from each tank were taken at random. blood serum was obtained by blood centrifugation 3500 rpm for 5 min. Anephelometery were use following the manufacturers intruductions to determine total factors (IgM, IgA,C3,C4) concentrations in serom. all test were studied in serology labratoar in Iran.

Statistical Analysis: one way analysis of variance (ANOVA ; SPSS, 20 and Excell ) was used to determine whether significant variation between the treatments excited difference between means were determined and compared by HSD Tukey test. all test used a significance level of (p<0.05 ) data are reported as means ± standard deviation.

RESULT

Natural Complement Activity: The dietary intake of either whole yeast Saccharomyces cervisiae and cell wall of Saccharomyces cervisiae increase the natural complement activity of rainbow trout after 15 and 30 days of treatment, although no significant differences with the control were seem. However, supplementation of the diet with cell wall of s.c produced a significant inhibition of the complement activity after 45 days in 0.3% prebiotic. Tables 1&2(Fig. 1). Our study showed that treatment with cell wall Saccharomyces cervisiae indicate high value than control for factor C3, C4 and significant diffrence (p< 0.05). In addition, supplementation of the diet 4 gr/kg in prebiotic and 0.6 % in prebiotic also produced as significant inhibition of the complement activity after 60 days of treatment (p<0.05). But the level of C3 in probiotic 2gr/kg had no significant different to control (p>0.05). Table 3&4 (Fig. 2).

Blood Serum Immunoglobulin (Igm): The dietary intake of either whole yeast Saccharomyces cervisiae and cell wall of Saccharomyces cervisiae increase the IgM level of rainbow trout after 15, 30 days of treatment, although no significant differences with the control were seem. However, supplementation of the diet with cell wall of Saccharomyces cervisiae produced a significant inhibition of the IgM level after 45 days in 0.3 % prebiotic Table 5(Fig. 3). our study showed that treatment with cell wall Saccharomyces cervisiae indicate high value than control for factor IgMand significant difference (p<0.05).
Table 1: average of C3 in 45 days

Table 2: Average of C4 after 45 days

Table 3: average of C3 60 days

Table 4: Average of C4 after 60 days

Table 5: Average of IgM in 45 days

Table 6: Average of IgM in 60 days

Figure 1: Pro2gr/kg: probiotic: 2gr/kg, pro4gr/kg: probiotic: 4gr/kg, pre 0.3%: prebiotic 0.3%, pre 0.6%: prebiotic 0.6%, nun: control.

Figure 2: Pro2gr/kg: probiotic: 2gr/kg, pro4gr/kg: probiotic: 4gr/kg, pre 0.3%: prebiotic 0.3%, pre 0.6%: prebiotic 0.6%, nun: control.
At the end of sampling the level of IgM in probiotic 2gr/kg, 4gr/kg and prebiotic 0.6 % had significant difference to control. (p<0.05) Table 6 (Fig. 4).

**DISCUSSION**

In aquaculture, stress, nutritional, environmental and quality of water our closely interlinked and these factors can lead to sub_optimum immune and performance and leave the animal susceptible to disease [30]. The results presented here show that a supplement of 0.3 % prebiotic in rainbow trout extruded diets earlier significance increased immune factors and complement system in after 45 days with control (p<0.05). cell wall of *Saccharomyces cerevisiae* are constricted almost entirely of β-1,3-D-glucan, β-1,6-D-glucan mannoproteins and chitin, bound together by covalent linkages [31]. However very little data exists concerning the use of yeast as immune stimulants.although the whole yeast enhances immune factors by production bacteriocins, antibiotics, proxidas. [10, 11].

The complement system is composed of more than 35 solubles plasma proteins that play key roles in innate and adaptive immunity [32]. The complement proteins have multi functional roles in the defence against macroorganisms ranging from the opsonisationlysis and killing of bacterial,chemotaxis and anaphylaxis [33]. In the present study the level of complement system in 0.3 prebiotic after 45 days increased and had significant difference between other treatments and control. this result is similar to observation by Yousefian 2012, that showed effect of Grobiotic–A as prebiotic on (*Rutillusfrisiikutum*) and increased C3, C4 after 8 weeks [34]. This result is similar to observation by staykov 2007 he showed effect of bio_MOS on immune states of rainbow trout and common carp and that showed increased complement system in probiotic 2 gr/kg BIO_MOS(35),at the end of sampling the level of complement in 2 gr/kg probiotic and 4gr/kg probiotic also increased and had significant differences with control.

Serum immunoglobulin are major component of the humoral immunity system and IgM is the main immunoglobulin in present in fish.amount of IgM in the serum was increased in 0.3 % prebiotic and had significant different to control.

This observation is a like to cuesta that showed increased level of IgM in prebiotic on sturgeon by cell wall of yeast [36]. At the end of sampling the level of IgM in the serum was increased in all treatment than to control resembling can et al. [37]. It is known that In the present study, fish feed with 0.3 prebiotic had significantly different to other treatment earlier in (IgM, C3, C4) than fish feed with 0.6 % prebiotic and 2 gr/kg, 4gr/kg probiotic and control.in fish, the innate immune system (Non specific)is the fundamental defence mechanisms against infectious microorganism, improvement of this Non specific disease defence (innate immunity) is particularly relevant to ubiquitous opportunistic bacterial pathogens that can take advantage of fish stocks when stressed.the innate immune system has also been shown to be important in activity in acquired immune response in mammalian research and this is also probably occurring in fish [38]. Due to the out breaks of farms increased with regard to the price of probiotic and prebiotic using this type of economic supplements are recommended.

**REFERENCES**


