Oral administration of baker’s yeast (Saccharomyces cerevisiae) acts as a growth promoter and immunomodulator in Labeo rohita (Ham.)

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**Abstract**

The effect of baker’s yeast (Saccharomyces cerevisiae), in the diet of the Rohu (Labeo rohita) innate immune response were investigated. Indian major carps Labeo rohita were fed with four different diets for eight weeks: a formulated diet as control diet and the same diets supplemented with 5%, 7.5% and 10% baker’s yeast as an experimental diets. After every fifteen days interval different growth parameters (such as ADG, SGR, FCR and PER), Serological parameters (such as TSP, TSA, TSG and A:G), different Hematological parameters (such as TLC, TEC, Hct, MCV and MCH) and different Non- specific immunological parameters (such as PR, PI, Respiratory Burst activity) were evaluated during experimental trial. At the end of the experimental period, fishes of all the tanks were challenged with pathogenic bacteria Aeromonas hydrophila. The results show that, yeast cell wall able to enhance the innate immunity and also have a positive co-relation with growth parameters. Through the absorption of yeast wall particle, the immune function and disease resistance of the entire organism is stimulated. These results support the possible use of baker’s yeast as natural immunostimulants in common fish diets.

**Keywords:** Immunostimulant; Labeo rohita; Saccharomyces cerevisiae

**Introduction**

Several whole microorganisms, live or not, such as bacteria, fungi or algae, increase disease resistance in mammals and fish [1,2]. In fish, as in other aquatic organisms, the whole microorganisms administered have mainly been bacterial species, which in the form of feed additives, have been shown to improve the intestinal microbial balance and increase the health status of fish, seemingly by colonising the gut and acting as antagonists to pathogens and so increasing resistance to pathogens [3,4]. More recently, other whole microorganisms have been tested for their possible immunostimulant properties in fish. Thus, the oral administration or injection of the yeasts Saccharomyces cerevisiae or Candida utilis has been shown to increase both humoral (myeloperoxidase and antibody titer) and cellular (phagocytosis, respiratory burst and cytotoxicity) immune responses, and to increase or confer resistance against pathogenic bacteria in channel catfish, rainbow trout or gilthead seabream [5-8]. Possible use of baker’s yeast in fish diets has many advantages. Firstly, they can be produced rapidly, easily and inexpensively and, at the same time, they are very stable and can be recycled from other industries. They are also natural substances so no negative effects may be expected either to the animals or to the environment. Moreover, there is no need to isolate their components, which consists mainly of cell wall sugars (β-glucans, mannanproteins and chitin), all are well-proved immunostimulant compounds.

The yeasts (Saccharomyces cerevisiae) has been used in gilthead seabream was Lyophilized form, which is not easily available to the farmer and also very costly but the baker’s yeast, which is directly used in the bakery industry is low cost material. With this aim, the present paper discusses the effects of the dietary intake of the baker’s yeast as supplementary feed and possible difference with the use of control diets are also established.

**Material and Methods**

Fingerlings of the species were obtained from a carp culture farm at the vicinity of Midnapore town has an initial measurement of 12.0±0.2g. Fish were (12.2 ± 0.22g) released into continuous flow glass aquaria (76 X 41 X 41 cm² area; 200.00 l capacity) after acclimatization for 15 days to prevailing laboratory condition of water temperature (31-33 °C) and pH (7.42 –7.53). Studies were conducted at room temperature for 60 days. The water quality (pH, DO, Alkalinity, Ammonia) of the experimental aquaria was monitored periodically once in a week following the methods of APHA (1998) and maintained at normal level.

**Preparation of experimental feeds**

The four prepared feeds (Cont., Exp-1, Exp-2, Exp-3) were formulated using locally available ingredients (mustard oil cake, rice polish, fish meal and tapioca powder). Feed formulation was done basically by "Square method" using determined values of protein content of the different ingredients. Proportion of each ingredient required was calculated precisely providing allowance for the premix. Locally available Baker’s Yeast (Saccharomyces cerevisiae) was used as an immune stimulant. Baker’s Yeast were diluted in water and supplemented @ 5%, 7.5% and 10% in the feed Exp-1, Exp-2 and Exp-3 respectively; where as control feed was not supplemented with Saccharomyces cerevisiae. Feeds were pelleted separately with local made (Kolkata, India) hand pelletiser. The pellets were dried in a thermostatic oven (M/S Modern Industrial Corporation, Mumbai, India) at 37°C and less then 10% moist [9, 10] and stored in an air tide jars at room temperature. Proximate composition of the four prepared feeds (Cont., Exp-1, Exp-2, Exp-3) were detailed in (Table 1).

**Growth performance and conversion ratio**

Fishes were fed twice daily at 8.00 and 16.00 h with ration size maintained with 6% of their body weight in two equal portions. The

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net weight was recorded every 15 days with an electronic and feed quality was readjusted after every weighing period of 15 days. For evaluating the dietary performances, the nutritional indices like live weight gain (LWG), Net weight gain (NWG), Average daily growth (ADG), Protein efficiency ration (PER), Specific growth rate (SGR) and Protein efficiency ratio (PER) were used. Two fishes of each group were terminated through overdose anaesthetisation by MS222 (Sigma chemicals, India) [11] at the end of the experiment, and stored at −20 ºC until analysis.

Proximate analysis

Proximate analyses of ingredients, feeds and body carcass were determined following the method provided in AOAC (1990). Moisture content was determined gravimetrically in a hot air oven at 100+ 10 ºC for 24 h. Crude protein contents were determined by the micro-kjeldahl method. Crude lipid contents were determined by the soxhlet extraction method using petroleum ether (Boiling point: 40-60ºC) in the electro-thermal soxlet apparatus. After extraction of lipid the defatted samples were used for the estimation of crude fiber following Patra [12]. Ash content was estimated by inserting samples in a muffle furnace at 500+ 50ºC for 10 h.

Biochemical analysis

DNA (Deoxy-ribo nuclic acid) and RNA (Ribo nuclic acid) contents from liver (hepatopancreas) tissues were estimated as per the scheme given by Munro and Fleck (1969). Tissue was taken from fishes and homogenized with 0.25 m sucrose solutions. 250 µl homogenate and 500 µl 5% TCA mixed thoroughly centrifuged and wait for 15 minutes and then precipitate was dissolved in PCA (several time in different concentration) and centrifuged with 5000 rpm (two times). The ultimate supernatant was used for RNA and DNA estimation by UV spectrophotometer.

Enzyme assay

Two specimen from each of the replicate were sampled both the initial stage and at the end of a 60 days experimental period. The fishes were dissected and the liver was cleaned and removed and pieces of liver was thoroughly washed in ice chilled double distilled water and subsequently collected in ice-cooled Petri dishes, weighed and cut into small pieces. A 5% homogenate was prepared in potter- Elvehjen homogenizer with neutral glass powder and ice cold 0.1 M phosphate into small pieces. A 5% homogenate was prepared in potter- Elvehjen homogenizer with neutral glass powder and ice cold 0.1 M phosphate buffer (pH 7.4) and centrifuged at 2,500 rpm for 15 minutes in a refrigerated centrifuge. The supernatant was used for the GOT and GPT activity following the method of Bernfeld [13].

Initially and after completion of the study, experimental fishes from different feeding trials were separately pooled and liver was dissection out for ACP and ALP activity estimation. After weighting the whole quantity of proportionate quantity, 10 % sucrose and 1 % tritonex mixture were added and the contents homogenized in a tissue homogenizer. The samples were then put in appendrop tube and cold centrifuged (at -4 ºC) at 11000 rpm for 15 minutes. The supernatant was collected as sample for determining the ACP [14] and ALP [15] activity.

Study of blood parameters

Blood samples were collected by heparinized syringe from caudal vein for haematology; EDTA (Ethylene diamine tetraacetic acid) was used as anticoagulant. 1.0 mg EDTA ml-1 of blood or 1 drop of 1.0% solution 5 ml 3 of blood was used for haematology. Haematological parameters were estimated according to the method of Wintrobe [16]. MCV, MCH, MCHC were calculated by using standard formulae (Decie and Lewis 1991). Blood samples were collected in the laboratory for serological diagnosis by syringe from caudal vein and heart.

Determination of the Total Serum Protein (TSP) and albumin by Gornall’s biuret method [17].

\[ TSG = \frac{TSP \cdot TSA}{TSP - TSA} \]

(Where TSG = Total Serum globulin, TSP = Total Serum Protein, TSA = Total Serum albumin.)

Determination of immunity level

On day 60, blood was collected form fishes of each group. Part of the blood was heparinised and the rest was allowed to clot for serum samples, which were preserved at −20ºC for further analysis. Immediately after collection, the heparinised blood samples of each group were pooled to three aliquots. Part of the blood was analysed for leucocrit value in duplicate per sample [18]. The rest of the heparinised blood was immediately used for the phagocytic assay [8, 19]. Blood was collected from the fish by using a 0.2 ml glass syringe rinsed with an anticoagulant. Then the blood was transferred into the heparinised vial and mix properly. 0.1 ml of freshly prepared NBT solution was added to 0.1 ml of the heparin mixed blood and 15 µl of stimulant solution in the incubating bottle. The bottles were incubated at 37ºC for 10 minutes and at 26ºC for another 10 minutes. 50-70 µl of this blood was transferred onto a clean slide and makes a thick smear with a spreader slide. The slides were air dried and stain with Wright's stain. For staining with the Wright's stain, first flood the slide with 1 ml of the staining solution for 30 seconds then 1 ml of distilled water was added and keep for another 30 seconds. The slide was then pour off the stain and dried. Then the slide was then under oil immersion lens at 100 X. The positive cells had the violet coloured formazan granules in the cytoplasm. The percentage of the positive cells gave the idea about the non-specific immune status of the organism.

Challenge trial

Rohu fishes (Labeo rohita) of medium size (435±28g) were collected from a semi-intensive culture pond at the University campus. The intestine were gently excised & cut open with a pair of sterile scissors. The non-adherent micro floras of the intestine were isolated by three times washed with sterile solution & homogenized with 10 ml distilled water in stomacher bags. The presumptive numbers of micro flora were determined by the spread plate technique using nutrient agar. The pathogenic strain Aeromonas hydrophila (AH1) were isolated by the method of Kaneko [20], which had been cultured and maintained in the Aeromonas selective medium (M884, Hi – Media). After 60 days feeding trial, fishes of each experimental group were released in four aseptic tanks. Different water quality parameters (Temperature pH, Dissolve Oxygen, Alkalinity, Ammonia) were maintained in normal level. Different experimental feeds and control feeds were provided twice daily according to 6 % of their body weight. The fishes in each treatment were challenged with Aeromonas hydrophila (AH1). Fishes in all replicates were immersed in a suspension of Aeromonas hydrophila (AH1), ~ 10⁷ CFU ml – 1 according to Austin et al. [21]. This was followed by a second immersion ~ 10⁸ CFU ml – 1 after 7 days Austin et al. [21]. The survivability of the fishes was recorded against pathogenic strain for last 10 days.

Statistical analyses

As all the above analyses were carried out on pooled samples of a given lot, standard deviations of means were calculated. However, for evaluating the dietary performances, nutritional indices, enzymatic
activity and RNA: DNA ratio, different haematological, serological, immunological parameters and challenge trials; correlation and regression test were performed through SPSS software package. Significant differences between the means of the treatments were tested by Duncan Multiple Range Test [22] through SAS software package [23].

Results

Locally available ingredients (Mustard oil cake, Rich polish and Fish meal) were used for preparing experimental feeds for *Labeo rohita*. Except control feed (Control), other three experimental feeds (Exp-1, Exp-2 and Exp-3) were supplemented with baker's yeast (*Saccharomyces cerevisiae*) and it was replaced with equal amount of rich polish. All the four experimental feeds are isocaloric and isonitrogenous. The average crude protein percentage of the feeds were 32.18± 0.22 and the crude protein percentage were around 7.67±0.09 (Table 1).

After 60 days feeding trial initial and final carcass composition of *Labeo rohita* in relation to various feeds was presented in (Table 2). The carcass composition of the test animals revealed an apparent increase in the final carcass protein and lipid over the initial carcass protein and lipid. Significantly highest carcass protein (12.01±1.29) and lipid (3.16± 0.31) was recorded in feed Exp-1 fed fishes as compare to other experimental and control feeds. This was clearly indicating that, the enhancement of carcass composition with the increasing supplementation of yeast at a specific level (5% supplementation). Perhaps it was due to the enzymatic activity in the gut and there by nutrients are spread for the growth and it make overall well being for the fishes.

Maximum weight gain of 135.18±1.21(of 10 fishes) was obtained in feed Exp-1 fed fishes followed by Exp-2 (109.88±1.36) and Exp-3 (102.86±1.96) and control (92.82±1.29) respectively (Table 3, Figure 1). There were significant differences (P≤0.05) in the different growth parameters among the different experimental feed fed fishes (Table 3). Significantly lowest (P≤0.05) FCR (3.54±0.033) was observed in Exp-1 fed fishes; it was also observed that significantly higher SGR was obtained from feed Exp-1 fed fishes (1.27±0.04). It indicates that Exp-1 feed shows better utilization of nutrients than the other feeds. Similarly significantly (P≤0.05) highest PER values (6.46±0.11) was obtained

### Table 1: Proximate composition (%) of different experimental diets* for *Labeo rohita.*

| Proximate composition of feeds | Control | Exp –1 | Exp –2 | Exp –3 |
|-------------------------------|---------|--------|--------|--------|
| Moisture(%)                   | 7.18± 0.86  | 7.23± 1.05  | 7.19± 1.29  | 7.28± 1.05  |
| Crude protein (%)             | 32.83± 0.13  | 32.18± 0.22  | 31.85± 0.32  | 31.53± 0.32  |
| Crude lipid (%)               | 7.92± 0.07  | 7.67± 0.09  | 7.54± 0.11  | 7.41± 0.23  |
| Ash (%)                       | 11.69± 0.16  | 10.62± 0.08  | 10.08± 0.06  | 9.55± 0.19  |
| Energy (KJ/g)                 | 9.35± 0.004  | 9.43± 0.002  | 9.46± 0.003  | 9.50± 0.008  |

*On dry matter basis.
Results are means of five separate determinations (Mean ± SEM). Figures having same alphabets in the different rows are significantly different (p ≤ 0.05).

### Table 2: Initial and final carcass composition of *Labeo rohita* after 60 days experimental trial on four different dietary treatment.

| Carcass Composition (%) | Initial | Experimental tanks |
|-------------------------|---------|--------------------|
|                         | Control | Exp –1 | Exp –2 | Exp –3 |
| Crude protein (%)       | 7.75± 0.86  | 9.96± 1.05  | 12.01± 1.29  | 10.40± 0.30  |
|                         | ±0.09  | ±0.10  | ±0.19  | ±0.65  |
| Crude lipid (%)         | 2.93± 0.71  | 2.90± 0.29  | 3.16± 0.31  | 2.93± 0.93  |
|                         | ±1.21  | ±0.12  | ±0.03  | ±0.12  |
| Ash (%)                 | 12.63± 0.089  | 12.55± 0.089  | 12.89± 0.073  | 12.68± 0.071  |
|                         | ±0.062  | ±0.062  | ±0.062  | ±0.062  |

Results are means of five separate determinations (Mean ± SEM). Figures having different alphabets (superscribed) in the same row are significantly different (p ≤ 0.05).

### Table 3: Different growth parameters, haematological parameters and serological parameters after 60 days feeding trial on *L. rohita* with yeast supplemented diets.

| Diets | NWG (g) | ADG (g) | SGR (%) | FCR | PER | Hb (g%) | TLC (×10^9/mm^3) | TEC (×10^9/mm^3) | Hct (%) | Leucocrit value |
|-------|---------|---------|---------|-----|-----|--------|------------------|------------------|---------|----------------|
| Control | 92.82± 1.29  | 1.54± 0.002  | 0.95± 0.30  | 4.64± 0.18  | 4.35± 0.14  | 7.1± 0.91  | 13.8± 0.76  | 1.30± 0.15  | 27.21± 1.02  | 37.10± 2.05  |
| Exp –1 | 135.18± 1.21  | 2.25± 0.001  | 1.27± 0.41  | 3.54± 0.33  | 6.66± 0.11  | 8.2± 0.17  | 22.2± 0.57  | 1.76± 0.11  | 35.34± 1.25  | 59.20± 0.33  |
| Exp –2 | 109.88± 1.36  | 1.83± 0.007  | 1.13± 0.39  | 4.39± 0.34  | 5.30± 0.32  | 7.9± 0.21  | 18.3± 0.76  | 1.52± 0.06  | 28.32± 1.41c | 47.50± 1.34  |
| Exp –3 | 102.86± 1.96  | 1.71± 0.011  | 1.10± 0.30  | 4.36± 0.44  | 5.02± 0.19  | 7.5± 0.20  | 16.7± 0.87  | 1.43± 0.14c | 32.18± 1.30c | 42.60±2.13c |

Figures having different alphabets (superscribed) in the same column are significantly different (p ≤ 0.05).
from Exp-1 fed fishes, which indicate better utilization of protein for growth and metabolism.

Significantly (P<0.05) highest Alkaline phosphatase (ALP) (8.62 ± 0.04) and Acid Phosphatase (1.48 ± 0.02) activity was observed in Exp-1 feed fed fishes (Table 4) following Exp-2 and Exp-3 feed fed fishes. Similarly highest GPT (0.068±0.003) and GOT (0.042 ± 0.002) values were registered in feed Exp-1 fed fishes, where as lowest in feed control fed fishes (0.055±0.001 and 0.032 ±0.003). Significantly (P≤0.05) higher RNA: DNA ratio (2.10 ± 0.003) was observed in fishes fed with Exp-1 feed and least was recorded (1.54 ±0.001) in control feed treated fishes.

Table 4: Activities of ALP, ACP, GOT and GPT in the intestine of Labeo rohita after 60 days feeding trial.

| Treatment | ALP  | ACP  | GOT  | GPT  |
|-----------|------|------|------|------|
| Control   | 1.08±0.01 | 0.40±0.001 | 0.03±0.02 | 0.055±0.001 |
| Exp –1    | 8.62±0.04  | 1.48±0.02  | 0.04±0.06  | 0.068±0.005 |
| Exp –2    | 5.29±0.02  | 1.01±0.001 | 0.039±0.01  | 0.062±0.002 |
| Exp –3    | 5.10±0.03  | 0.73±0.003  | 0.035±0.04  | 0.059±0.006 |

Results are means of three separate determinations (Mean ± SEM), Values with the same superscript in the different rows are not significantly different (p < 0.05) from each other.

RNA/DNA : Initial value = 1.02±0.010

Table 5: Initial and final RNA/DNA ratio in the muscle of Labeo rohita after 60 days feeding trial.

| Treatment | Initial | Final |
|-----------|---------|-------|
| Control   | 1.54±0.001 | 1.54±0.001 |
| Exp –1    | 2.10±0.003 | 2.10±0.003 |
| Exp –2    | 1.98±0.005 | 1.98±0.005 |
| Exp –3    | 1.82±0.004 | 1.82±0.004 |

Results are means of five separate determinations (Mean ± SEM), Figures having different alphabets (superscribed) in the same column are significantly different (p ≤ 0.05)

Table 6: Effect of Yeast (Saccharomyces cerevisiae) on A:G ratio & non specific immunity levels of Labeo rohita after 60 days feeding trial.

| Treatment | Albumin: Globulin | Phagocytic Ratio | Phagocytic Index | NBT cells |
|-----------|------------------|------------------|-----------------|-----------|
| Control   | 3.00 ± 0.04      | 15 ± 0.49        | 1.52 ± 0.12     | 48.5±0.32 |
| Exp –1    | 2.34 ± 0.03      | 63 ± 1.22        | 2.34 ± 0.09     | 57.0±0.61 |
| Exp –2    | 2.52 ± 0.06      | 42 ± 1.27        | 2.15 ± 0.09     | 52.5±0.24 |
| Exp –3    | 2.49 ± 0.08      | 18 ± 0.86        | 1.49 ± 0.20     | 49.5±0.23 |

Results are means of five separate determinations (Mean ± SEM), Values with the same superscript in the different rows are not significantly different (p < 0.05) from each other.

Discussion

In the last two decades, many substances have proved their...
usefulness in fish culture because of their properties to stimulate the immune system & increase disease resistance. Among these immunostimulants the role of isolated β-glucans, chitin or vitamins is well documented [24-34]. However, the use of whole organisms instead of their isolated components has hardly been evaluated. In this way whole yeast cells (mainly S. cerevisiae), Which represent a major commercial source of β-glucans, have recently been described as good immunostimulants in fish [6,8].

Charlon and Bergot [35] achieved more than 89% survival and good growth of carp larvae; feed exclusively on dry diets having yeast powder and pork / beef freeze dried liver. Alami, Durante et al [36] reported 87% survival and FCR of 0.62 in common carp larvae reared on liver, yeast and commercial trout starter feed. Singh et al. [37] also found that yeast increased the rate of feed intake and conversion efficiency. Mohanty et al., reported 100% survival using a liver based diet containing goat liver, Cod liver oil, Vitamin and mineral mixture with average growth of 133.3 mg / fry in Rohu (Labeo rohit) raised from spawn to fry.

In the present study, S. cerevisiae are also found to stimulate the digestion through the supply of digestive enzymes and certain essential nutrients to the animals. Immunostimulants, particularly Saccharomyces cerevisiae producing several enzymes, which is not, produces by the host. Similar observation was also reported by Swain et al. [38]. A complex polysaccharide including cellulose are better utilized by the host in the presence of direct feed microbes like Aspergillus oryzae, Saccharomyces cerevisiae.

Furthermore, it was reported that yeast in the diet improves feed efficiency, organic phosphorus (phytic acid) utilization and fibre digestion [38]. It also reported that supplementation of immunostimulants in feed improves the nutrition by hydrolytic enzymes including amylase and proteases, the production of vitamins such as biotin and vitamin B12 [39-44] and the host immunity [45]. It indicate that a given amount of immunostimulant elicit more than one protective response by the host.

Dietary intake of immunostimulants by fish has definite advantages and it is a useful method of exposure in large scale fish culture [8], particularly hypopolysaccharides of yeast cell wall was evaluated by many investigators like Oruto et al. [6]; Sakai [28]; Sahoo and Mukherjee [46]. It is recognized that immunostimulants enhance the host defence system against pathogens by increasing phagocytosis, antibody production, leucocit level and reduced A/G (Sakai, 1999). A comparative study done by Oruto et al. [6] revealed that oral administration of whole yeast Saccharomyces cerevisiae enhances the cellular innate immune response. Notably that all kinds of cellular parameter such as phagocytic index, respiratory burst activity, number of erythrocytes, lymphocytes were also enhanced but not the humoral one.

Blood is a pathophysiological reflector of the whole body and therefore, blood parameters are important in diagnosing the status of fish health (Pecie and Lewis, 1991), particularly when some additives used in the feed. In this present study it was observed that all the blood parameters in all the treatments were similar to standard (Banerjee et al., 2002) and Exp-1 fed fishes showed superior as compare to others, indication of the blood parameters revealed the positive impact, but also demonstrated a stable physiological reflection of the whole body [47]. In this study, a superior growth performance in terms of weight gain percentage, specific growth rate was recorded in L. rotila fishes fed with Exp-1 feed as compare to other feed fed fishes. These observations determine the optimum doses of yeast supplementation in feed. This might be helpful for optimum dietary utilization.

In this study, although all the feeds were isonitrogenous but the concentration of immunostimulant i.e. Baker's yeast (S. cerevisiae) in Exp-1 feed might be helpful for proper nutrient utilization. Whole body carcass composition was higher in Exp-1 feed fed fishes as compare to control one, which could have also noted to the overall low feed utilization level. RNA: DNA is known to provide dependable indication of growth trend [48-50]. The ratio was greatest in the fish fed Exp-1 feed with higher dietary utilization and best growth. Bazaz and keshavnath (1993) found higher RNA: DNA in better growing fish fed with oil supplemented diets using equal level of crude protein. The present study also reports such a finding, where all the feeds are isocaloric and isonitrogenous but 5% supplementation of Baker's Yeast (S. cerevisiae) incorporated feed (feed Exp-1) exhibit better growth as well as better RNA:DNA ratio. The highest level of GOT and GPT as well as ALP and ACP were found in Exp-1 fed fishes and lowest in control. Most of the amino acids normally found in protein undergo transamination reaction and transaminases are localized in both cytosol and mitochondria (Wada and Marino, 1964), which is induced by high protein diet [51], thus a positive correlation between the immunostimulant concentration at a specific level and the GOT, GPT level in the liver could be observed.

The result obtain in this study not only support the use of Baker's yeast for better growth, and proper nutrient utilization but also it act as an immunostimulant by stimulating the immune response. The activation mechanisms involved are known to be related to the carbohydrates, derived from the yeast cell wall. glucans/whole yeast added to feed stimulates the phagocytic activity, respiratory burst activity and increase protection after challenge with pathogenic bacteria, similar finding were reviewed by Robertson, 1999.

In this study oral administration of Baker's Yeast (S. cerevisiae) stimulate the non specific immunity level as measured through enhanced phagocytic activity, leucocit level, respiratory burst activity and reduced A: G. Although enhanced leucocit value does not necessarily relate to an immunostimulatory action of Baker's Yeast. The possible role of yeast as an immunostimulant due to its cell wall which composed of Lipopolysaccharide, such as glucan, which enhanced phagocytic activity of macrophages and globulin level as observed in the present experiment, the phagocytic indices seemed to be reliable indices of a heightened immunostimulatory response. Similar observation also observed by Swicki et al. [8]; Esteban et al. [31]; Oruto et al. [6]; Rodiguezetal [7] in Rainbow trout andgilhead seabream. Albumin − Globulin ratio is a measurable humoral component at the non-specific defences. The reduction of A: G, might be due to the increase of Total serum globulin level with Significance protective mechanisms for fish [46]. Fish produce reactive oxygen species, which are considered to be toxic for fish bacterial pathogens [52,53] and are generated by phagocytes after stimulation by a variety of agents. In this experiment higher respiratory burst activity (O2-production) was increased in all the experimental diets as compare to control diets, but highest number of NBT positive cells were observed in Exp-1 feed fed fishes, an effect also seen in rain bow trout (Swicki at al,1994), Seabream (Oruto et al., 2002) and turbot [54]. After challenge trial with Aeromonos hydrophila, highest survivability observed in Exp-1 fed fishes, the enhanced protection conferred by glucan is not surprising, similar report has been reported to induce resistance in carp (Cyprinus carpio) against E. tada [35]. Evidence suggests that glucan enhance disease resistance by stimulating non-specific components of fish immune system [56,57].

To conclude the present results provide evidences that Baker's yeast
(S. cerevisiae) added in a common fish diet, exhibit better growth, better nutrient utilization and activate the innate immunity, as well as increase the survivability of L. rohita. Optimal doses and administration time have been established in an attempt to provide a useful approach for protecting culture fish against infectious diseases.

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