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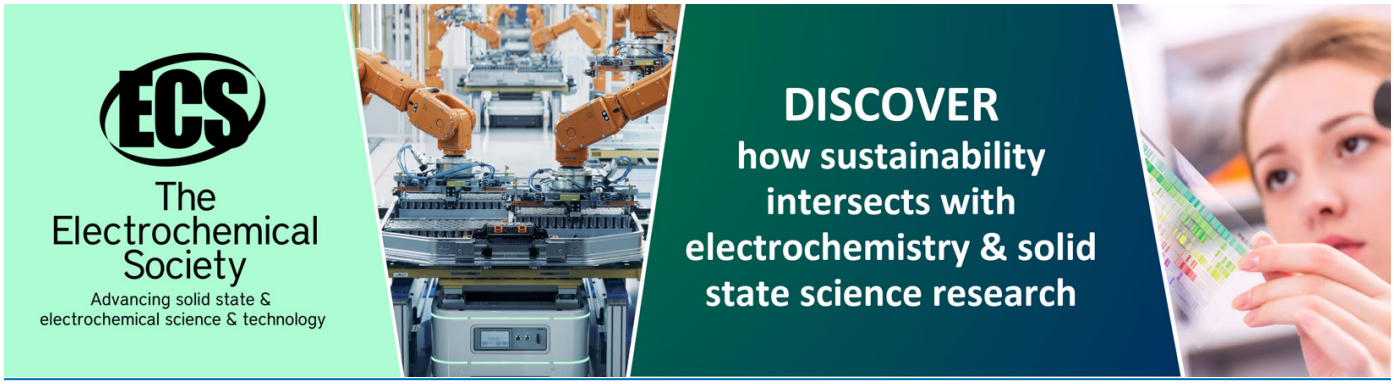

Effect of food additives, Thepax and Labazyme on growth performance and immune response of young common carp *Cyprinus carpio* L.

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Abstract. This study was carried out for 60 day. A total number of 70 fish (Cyprinus carpio L.) with an average of 13.40±1.85 g and a length 11.07 ±1.27 cm were randomly divided into five treatments as a two tank treatment, similar to its contents of conventional ingredients (i.e., fishmeal, soybea soybean meal, corn, barley and wheat bran as well as vitamin mixture premix and carboxy methyle celloluse (CMC)). They differ in their content from food additives. They were commercially ready made in dietary T1 and T2 (Thepax 0.5, 1 gm/ kg), T3 and T4 (Labazyme 1, 2 gm/ kg) and control (0.0 gm/kg) of experimental diets. Data collected included fish weight and feed conversion ratio measured at the end of the study. The results found that the addition of food additives has significantly been affected by weight gain, specific growth rate and feed conversion ratio ($P<0.05$), with the best results obtained in fish food additives, pellets have been supplemented to Thepax 1 gm/ kg to enhance growth and immune responses.

Keywords: Fish, *Cyprinus carpio*, Food additives, Growth

1. Introduction

The probiotic was first described to the secreted of substances by some microorganism that catalyze the growth of another, a development of this qualifier was coined by the propose while the probiotic is substances and organisms, that participate to microbial of intestinal balance, move a role in maintaining health and also innervate the host's immune system [1][2].

Uniform consuming of food contain probiotic is bespoke to institute a positive evenness of inhabitance of beneficial microbes in the intestinal flora, explore on the application of probiotics in animals of aquatic has increase the demand for sustainable cultivation (aquaculture) [4]. The benefit of supplement contain improvement in values of feed, donating to digestion of enzymatic, anti-mutagenic andante-carcinogenic activity, inhibition of pathogenic microorganism, growth-promoting factors and an augmentation of immune response [3][4][5].

The expressed of prebiotic as food of a-non-digestible ingredient, that financial gain effects of the host, by selectively component, that financial gain effects of the host, and by stimulating (selectively) the growth and activation of one (or limited number) of intestine bacteria, in order to can promote host health case [5]. It uses sometimes together (symbiotic) [6].

In aquaculture, use of antibiotics has advanced numerous questions on the expansion of resistance in persistence of the drugs, pathogenic organisms and in food chain [6]. The utilize of prebiotics, probiotics and seems to fashion an alternative wherewithal of disease monitoring in the cultivation fishery. The current study aimed to evaluate the effect of different diets supplemented with Thepax (prebiotic) and Labazyme (synbiotics) on the growth performance and immune response of young common carp *Cyprinus carpio C. carpio*.

2. Materials and Methods

In the growth experiment a group of young common carp was brought at an average weight 13.40 ± 1.85 gm and average total length 11.07± 1.27 cm from fish farming station (Marine Science Center) in Basrah University to the laboratory in the Department of Fisheries and Marine Resources placed in tank and were given a prophylactic treatment with 0.5 NaCl dip for 2 minutes then they were bathed in 0.5 ppm methylene blue for 24 hours. The fish were acclimatized for 20 days with experimental condition and diets before running the experiment at 3% /body weight/ day, after acclimatization process, 70 fish were distributed into 10 round tanks its capacity 30 liters were used with a density of 5 individuals per, (two replications for each treatment). Feeding frequency was twice a day at 07.00 am and 15.00 pm.

The water temperature ranged between 22-26 ºC during the experimental period from July 2018 to August 2019. Before formulating the diet, the dietary ingredients were subjected to proximate analysis [8].

The composition of the feed was fishmeal, soybean meal, corn, barley and wheat barn as major source of dietary protein and lipid as well as carboxyl methyl cellulose (CMC) and it premix (sabra company Turkey) chromic oxide, all these ingredients were mixed at required amount so that the final diet contained 35% crude protein as shown in Table (1), food additives 0.5 gm/ kg and 1 g/ kg Thepax, 1 g/ kg and 2 g/ kg Labazyme diet as well as control diet without any additive (Table 2), cooked in autoclave for 30 minute at a temperature of 70 C° then left to dry in a laboratory atmosphere ,then grinded and added to the mixture of vitamins and food additives, then formed a 60 cc medical syringe and dried up, shaped, cut off and preserved in the refrigerator.

Table 1: Formulation of the experimental diet**.**

Ingredients	%	Proximate composition	Dry weight basis %
Fishmeal	25	Crude protein	35.27
Soybean meal	40	Crude lipid	10.23
Corn meal	10	Ash	8.14
Barley meal	10	Moisture	9.39
Wheat bran	10	Dry matter	20
Carboxy methyl cellulose	2	Carbohydrate	17
Vit premix*	3		

***** Sebro Company Turkey

For growth data all replicate of different treatments weighed at the beginning and every 15 days by electronic balance (Digital counting Balance 1000 g/ 0.01g).

The parameters measured were weight gain, specific growth rate and feed conversion ratio. Fish growth was calculated based on the formula applied [9] as follows:

Weight gain, WG (g) = Wt - Wo

Specific growth rate, SGR (%/day) = Wt- ln W0/t ×100

Where t: feeding duration (days)

Feed conversion ratio, FCR=Total feed given (g)/ WG

Where: Wt: final weight of fish (g) and Wo: initial weight of fish (g)

Table 2: Composition of food additives.

Additive	Composition
Thepax (prebiotic)	Cell wall of yeast *Saccharomyces cerevisiae*, mannan and glucan
Labazyme (synbiotics)	1. Probiotic*: Lactobacillus acidophilus*, *Streptococcus faecium* and *Bacillus Subtilis*. 2. Prebiotic: Amylase, Cellulose and Protease.

2.1. Measurement of blood parameters

At the end of the experiment, the immune response of common carp measured: Hematocrit (Ht), Hemoglobin (Hb), Red blood cells (RBC), White blood cells (WBC), Mean corpuscular volume (MCV), Mean corpuscular hemoglobin (MCH) and Mean corpuscular hemoglobin concentration (MCHC) by [10]0-1dependent the gauge protocols.

2.2. Statistical analysis

According to application of SPSS (version 22), the data were subjected to One way analysis of variance (ANOVA).

3. Results and Discussion

Probiotics, prebiotics and symbiosis have been nutrition on farm animals for a long time, the advantages of these stimuli over traditional antibiotic outgrowth promoters are: 1 - No retreat time, 2 - No residual active, 3 - No cause for bacterial mutation [6].

Infectious diseases were cost the aquaculture manufacture each year millions of dollars, thus there are great attention in developing alternate to traditional methods of prevailing disease with chemotherapy [11]. Traditional strategie for disease prevent and treatment, like the use of drugs and vaccines, and have limitation like regulatory restriction or unsuitable means of surrender, this interest in using diet additives that may influence aquatic species, disease resistance and immunity has increase [12][13][14]. Though the concept of practical, feed was relatively modern in the aquaculture manufacture, there is prospect for diets of developing, so as to requirements of meet minimum nutritional, resistance to stress and disease causing organisms, improve health [15].

Average water temperature was 24 ±0.5 °C, dissolved oxygen concentration was 6.4±0.3 mg/l, while the pH was 8.1±0.11 and salinity was 2.1 ppt. The environmental factors in the study were within the reasonable limits for the common carp growth and survival [16].

The amelioration in growth may however, be related to the amelioration in the microbial flora balance of intestinal [13; 14]. In the fish given prebiotic with lower concentration (T1 and T3), fish growth tended to be lower slower than the growth of fish in higher concentration (treatment T2 and T4). This occurred because of the prebiotic and synbiotics concentration added had probably not equaled the required dose. This condition occurred because the administration of an ingredient such as prebiotic was highly influenced by the duration and dose of administration [14], and prebiotic is not an organism and has less influence in natural environment. Oral administration of baker's yeast increased the growth rate of *Labeo rohita*, where the amount of yeast was 5%, 7.5% and 10% in three experimental diets [17].

A similar result was observed in the survival rate of *Oreochromis niloticus* [18], also in study [7] use live *S. cerevisiae* demonstrates that probiotic based diets have a more positive influence on the growth, the five supplemented diets included three prebiotics (0.2% β-glucan, 1% galacto-oligosaccharides, 0.5% mannan-oligosaccharides and two probiotics (1% live yeast S. cerevisiae and 0.01% *Lactobacillus acidophilus*) powder.

The supplementation of trout beginning diet with the proper consistency of probiotic (commercial) could be useful for growth of fry rainbow trout [19]. The considerable refinement of growth parameters con be attribute to the influence of prebiotic and probiotic, that improve assimilation of depressed, nutrients and harmful bacteria cause growth (expression) [20; 21].

In currant study, the Thepax treatments (T2 ,1 gm/ kg) (Table 3 and 4) had higher significant ($P<0.05$) growth performance (final weight (g), WG (g), SGR (%) and FCR) and immune response (RBC (million/mm3), WBC (103.µ-l), Hb (g/dl), Ht (%), MCV (µm3), MCH (µg) and MCHC (%) than control.

The modulation of immune system is one of the generally purported benefits of the prebiotics and their potency to stimulate the systemic and local immunity under in vitro and in vivo conditions is noteworthy. Different prebiotics supplementation can eventually elevate phagocytic, lysozyme, RBC and Hb [22][23][24][25].

Table 3: Effect of different food additives on the growth performance of common carp fish.

Parameter	T1 (0.5 gm/ Kg) Thepax	T2 (1 gm/ kg) Thepax	T3 (1 gm/ kg) Labazyme	T4 (2 gm/ kg) Labazyme	T5 Control
Final weight (g)	19.13±8.43 a	18.61±7.89 a	15.03±5.60 b	16.61±2.46 b	16.08±0.60 b
WG (g)	28.00±0.02 a	32.60±0.72 b	19.25±0.36 c	23.96±1.06 d	27.45±0.67 a
SGR (%)	0.58±0.04 a	0.73±0.09 b	0.49±0.04 ac	0.57±0.03 ac	1.04±0.47 ad
FCR	7.71±0.20 a	7.93±0.09 a	8.69±0.08 a	7.68±0.00 a	11.36±0.45 b

Different letters in the same row are significantly different ($p<0.05$).

Table 4: Effect of different food additives on the immune response of common carp fish.

Parameter	T1 (0.5 gm/ Kg) Thepax	T2 (1 gm/ kg) Thepax	T3 (1 gm/ kg) Labazyme	T4 (2 gm/ kg) Labazyme	T5 Control
RBC (million/mm3)	1.41±0.25 ^{a}	2.72±0.33 ^{b}	1.59±0.28 ^{ac}	2.07±0.30 ^{c}	1.02±0.20 ^{d}
WBC (103.µ-l)	3.14±0.23 ^{a}	5.46±0.83 ^{b}	4.14±0.43 ^{a}	5.2±0.48 ^{b}	2.84±0.31 ^{a}
Hb (g/dl)	9.01±0.26 ^{a}	24.79±2.67 ^{b}	12.13±0.32 ^{c}	14.12±0.44 ^{c}	7.02±0.42 ^{a}
Ht (%)	17.10±1.79 ^{a}	36.24±3.53 ^{b}	21.56±3.10 ^{c}	23.53±2.42 ^{c}	13.24±1.05 ^{a}
MCV (µm3)	110.06±2.31^{a}	132.20±1.77 ^{b}	121.60±1.55 ^{c}	125.62±2.14 ^{c}	104.35±5.14 ^{a}
MCH (µg)	59.08±6.41 ^{a}	88.18±1.26 ^{b}	68.92±5.48 ^{c}	72.60±4.54 ^{bc}	54.02±3.25 ^{a}
MCHC (%)	5.23±0.35 ^{a}	10.27±0.11 ^{b}	5.40±0.32 ^{a}	5.43±0.27 ^{a}	5.06±0.13 ^{a}

Different letters in the same row are significantly different ($p<0.05$).

The cell wall of yeast *S. cerevisiae* is one of the effective adsorbents which is rich in crude protein (40-45%) with high biological value, is administered through the digestive tract, have a positive impact on the hosts health through its direct nutritional effect, *S. cerevisiae* contains enzymes alpha-amylase and protease that breakdown the starch and protein molecules, respectively, and help in digestion and efficient utilization [21].

References

[1] Lilly D M and Still W R H 1965 Probiotics: Growth-Promoting Factors Produced by Microorganisms. *Science*, **147**: 747-748.
[2] Parker R B 1974 Probiotics, the other half of the antibiotic story. *Animal Nutrition and Health*, **29**: 4-8.
[3] Delbert M *et al* 2012 Prebiotics and probiotics definitions and applications. *SRAC Publication*, **14**(4711): 219–29.
[4] Burr G 2007 Effects of prebiotics on nutrient digestibility of a soybean-meal-based diet by red drum (*Sciaenops ocellatus*). *Aquaculture Research*, **39**: 1680-6.
[5] Gibson GR and Roberfroid MB 1995 Dietary modulation of the human colonic microbiota: introducing the concept of prebiotics. *Journal of Nutrition*, **125**: 1401-12.

[6] Mohammed AH *et al* 2010 Effect of probiotics on growth performance of Koi Carp *Cyprinus carpio Journal of Applied Aquaculture*, **22**(3): 202-9.
[7] Tejpal D *et al* 2012 Effect of probiotics on the haematological parameters of Indian magur (*Clarius batrachus* L.). *Journal Fisheries and Aquatic Science*, 279-90
[8] Howrwitz W 1984 *Official methods of analysis of the association of Official Analytical Chemists*. 14th edition. (USA:Association of Official Analytical Chemists Washington D.C.). pp1018.
[9] AOAC 1984 *Official methods of analysis*. 14th edition. (Arlington:Association of Official Analytical Chemists), VA: 1141 pp.
[10] Baker D *et al* 2005 Haematology of juvenile *Acipenser oxyrinchus* and *Acipenser brevirostrum* at rest and following forced activity. *Journal of Fish Biology*, **66**: 208–21.
[11] Abdel-Hakim *et al* 2010. Effect of fresh or dried garlic as a natural feed supplement on growth performance and nutrients utilization of the Nile Tilapia (*Oreochromis niloticus*). *Egyptian Journal of Aquatic Biology and Fisheries***, 14**: 19-38.
[12] Gatesoupe FJ 1999 The use of probiotics in aquaculture. *Aquaculture*, **180**: 147-65.
[13] Ringø E 2010 Prebiotics in aquaculture- a review. *Aquaculture nutrition*, **16**(2): 117–36.
[14] Yang S D, Liu F G and Lion C H 2012 Effects of dietary L-carnitine, plant proteins and lipid levels on growth performance, body composition, blood traits and muscular carnitine status in juvenile silver perch *Bidyanus bidyanus*. *Aquaculture*, 342: 48-55.
[15] Kühlwein H 2014 Effects of dietary (1, 3) (1, 6) - D-glucan supplementation on growth performance, intestinal orphology and haemato-immunological profile of mirror carp (*Cyprinus carpio* L.). *Journal of Animal Physiology and Animal Nnutrition*, **98**(2): 279-89.
[16] Horváth L *et al Carp and pond fish culture*. 2nd edition, (Oxford: Blackwell Sci. Ltd.), p170.
[17] Tewary A *et al* 2011 administration of baker's yeast (*Saccharomyces cerevisiae*) acts as a growth promoter and immunomodulator in *Labeo rohita* (Ham.) *Aquaculture Research and Development journal*, **2**: 109-115.
[18] Osman HAM 2010 Influence of dietary commercial Baker's yeast, *Saccharomyces cerevisae* on growth performance, survival and immunostimulation of *Oreochromis niloticus* challenged with *Aeromonas hydrophila*. *Natural Sciences***, 3**(8): 96-10**.**
[19] Wache YF *et al* 2006 Cross effects of the strain of dietary *Saccharomyces cerevisiae* and rearing conditions on the onset of intestinal microbiota and digestive enzymes in rainbow trout, *Onchorhynchus mykiss* fry, *Aquaculture*, **258**: 470-8.
[20] Tukmechi A 2011 Dietary administration of beta-mercapto- *Saccharomyces cerevisiae* enhanced the growth, innate immune response and disease resistance of the rainbow trout, *Oncorhynchus mykiss*. *Fish Shellfish Immunology*, **30**: 923-8.
[21] Shabanzadeh S *et al* 2016 Growth performance, intestinal histology, and biochemical parameters of rainbow trout (*Oncorhynchus mykiss*) in response to dietary inclusion of heat-killed *Gordonia bronchialis*. *Physiology and Biochemistry*, **42**: 65–71
[22] Taher MM *et al* 2018 Effect of bay laurel (*Laurus nobilis*) extract on growth and food conversion of common carp (*Cyprinus carpio*). *Iraqi Journal of Aquaculture*, **1**(15): 17-30.
[23] Mustafa A A A, Salah M N and Al-Niaeem K S 2018 Effect of dietary prebiotic vegetable wastes on serum biochemical parameters and intestinal microflora of juvenile common carp *Cyprinus carpio*. *Biological and Applied Environmental Research*, **2**(2): 162-70**.**
[24] Al-Niaeem KS 2019 Effect of dietary biogen as probiotic on health status in juveniles of the common carp *Cyprinus carpio. Life Science Archives*, **5**(4): 1651-6.
[25] Taher MM and Al-Niaeem KS 2020 Effect of the leaf extract of *Nerium indicum* on health status of the common carp *Cyprinus carpio*. *Biological and Applied Environmental Research*, **4**(2): 229-36.