Growth performance, carcass composition, and immunophysiological indices in juvenile great sturgeon (*Huso huso*) fed on commercial prebiotic, Immunoster

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Received: December 2009   Accepted: April 2010

Abstract

Growth performance, carcass composition, and immunophysiological indices in juvenile great sturgeon (*Huso huso*) fed on commercial prebiotic Immunoster (IS) were investigated. After a four-week acclimatization period, 270 great sturgeon juveniles weighing 95.68 ± 10.05 g were randomly distributed into 9 fiberglass tanks in three replicates and kept at a density of 30 fish per tank for a period of 8 weeks at water temperature 20.55 ± 5.11 °C and dissolved oxygen 6.73 ± 0.35 mg L⁻¹. IS was added at two levels of 1% and 3% to the basal diet in place of cellulose. At the end of the trial, blood sampling and carcass analysis were conducted. Weight, length, body weight increase (BWI), specific growth rate (SGR), feed conversion ratio (FCR), protein efficiency ratio (PER) and condition factor (CF) were significantly affected by IS 3% compared with IS 1% and the control (P<0.05). There was significant difference (P<0.05) in crude protein of carcass between the experimental groups. Mean corpuscular volume (MCV) and mean corpuscular hemoglobin (MCH) showed significantly higher levels in experimental treatments compared with the control (P<0.05). Immunoglobulin M (IgM) level and lysozyme activity in IS 1% group were higher than the control group. It can be concluded that IS can enhance growth performance and improve some immunophysiological indices of great sturgeon.

Keywords: *Huso huso*, Prebiotic, Immunoster, Growth Performance, Immunophysiological indices

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Introduction

Sturgeons, which their evolutionary history goes back to 100 million years ago are anadromous and potamodromous species of the Northern hemisphere (Bahmani, 1998). Great sturgeon, *Huso huso*, is an important aquaculture species in Russia, Eastern Europe, Japan and Iran. This species is good in aquaculture activities (Mohseni et al., 2006). The days of culture in sturgeon fish are too long. The results of such studies can lead to prevent different kinds of diseases.

Prebiotic is expressed as a non-digestible food ingredient that profitably affects the host by selectively stimulating the growth and/or activation of one or a limited number of bacteria in the intestine that can enhance host health status (Gibson and Roberfroid, 1995). Prebiotics are carbohydrates, which can be classified into monosaccharides, oligosaccharides and polysaccharides (RingØ et al., 2010).

Mannan oligosaccharides (MOS) are complex carbohydrates derived from yeast cell walls. These materials contain mannose as the primary carbohydrate element. MOS has beneficial effects on the growth of cattle, swine, and avian species (Moran, 2004 cited in Salze et al., 2008). Data about the effect of MOS on sturgeons is very rare.

Immunostimulants can be divided into biological substances and synthetic chemicals which stimulate and enhance lymphocytes’ activity, antibody production, protection against pathogens and lysozyme activity (Sakai, 1999).

β-glucans are the most important structural polysaccharides in the cell walls of plants, fungi, algae, yeast and bacteria. They show immunostimulatory properties and increase survival rate, disease resistance and modulate innate and acquired immunity responses in fish (Dalmo and Bogwald, 2008).

Immunostem™ (IS) is a commercial prebiotic and an immunostimulant derived from the cell wall of a single source of brewers yeast, *Saccharomyces cerevisiae*. This substance contains MOS and β-glucans.

The effects of MOS on the growth performance, hematological parameters and immune responses have been studied in several aquatic species including gulf sturgeon, *Acipenser oxyrinchus* (Pryor et al., 2003), rainbow trout, *Oncorhynchus mykiss* (Staykov et al., 2007), European sea bass, *Dicentrarchus labrax* (Torrecillas et al., 2007, 2010), channel catfish, *Ictalurus punctatus* (Welker et al., 2007), cobia, *Rachycentron canadum* (Salze et al., 2008), red drum, *Sciaenops ocellatus* (Burr et al., 2008), Nile tilapia, *Oreochromis niloticus* (Sado et al., 2008), Atlantic salmon, *Salmo salar* (Grisdale-Helland et al., 2008) and rohu, *Labeo rohita* (Andrews et al., 2009).

The objective of the present study is to evaluate the influence of IS on the growth efficiency, carcass composition, hematological and biochemical indices and immune responses of *Huso huso*.

Materials and methods

The study was conducted in Shahid Dr. Beheshti Sturgeon Fish Propagation and Rearing Center, Rasht, Iran and Dr. Dadman International Sturgeon Research Institute, Rasht, Iran in 2009. Prior to the feeding trials, fish were fed on the basal diet to apparent satiation four times per
day for a 4-week acclimatization period. Then, 270 great sturgeon juveniles with a mean body weight of 95.68 ± 10.05 g were randomly allocated into 9 fiberglass tanks (2 × 2 × 0.53 m) and kept at a density of 30 fish per tank. The tanks were continuously aerated. Water of the tanks (using filtered water from the Sefidroud River) was replaced every 12 h to prevent accumulation of faeces and uneaten food. During the trial, water temperature, oxygen concentration and pH value were kept at 20.55 ± 5.11 ºC, 6.73 ± 0.35 mg L⁻¹ and 7.92 ± 0.09, respectively. The completely randomized design of this study consisted of three treatments (Control, IS 1% and IS 3%) in triplicates. All groups were fed on their respective diets four times daily (08.00, 14.00, 20.00 and 02.00 h) for a period of 8 weeks. The feeding rate was initially 4% of body weight per day and gradually decreased to 2%.

The ingredients of the experimental diets (based on the formulation of Dr. Dadman International Sturgeon Research Institute) are presented in Table 1. IS was supplied by Awill company, Victoria, Australia. IS was added at two levels of 1% and 3% to the basal diet in place of cellulose, except the control. All dry ingredients were thoroughly mixed for 30 min in a food mixer. Then, liquid ingredients were added to the diets and ingredients were mixed again for 20 min. The mixture was placed into a commercial meat grinder for thorough mixing and extruded through a 4 mm diameter strand and dried in a drier at 30 ºC for 24 h. The pellets were packed in sterile bags and sealed and stored at -15 ºC for a week. Analysis of the experimental diets is showed in Table 2. Proximate analysis of the diets was conducted according to the standard methods of AOAC (Association of Official Analytical Chemists) (1995) in the laboratory of veterinary organization, Rasht, Iran. Moisture content was estimated by drying the samples to constant weight at 105 ºC in an oven (Memmert, Germany), crude protein content (N×6.25) was measured using a Kjeldahl system (Buchi, Switzerland), crude lipid content was determined by a Soxhlet system (Buchi, Switzerland) and ash content was measured by weight after incinerating at 550 ºC for 6h in a hotspot furnace (Gallenkamp, England). A bomb calorimeter (Parr, USA) was utilized to measure the energy content. Biometric data was recorded after feeding was ceased for 24h. Fish were individually weighed and measured once every two weeks and ration was adjusted according to new biomass of each tank.

The growth performance of juveniles such as body weight increase (BWI), specific growth rate (SGR), feed conversion ratio (FCR), protein efficiency ratio (PER), condition factor (CF), hepatosomatic index (HSI) and survival rate were calculated based on the standard formulae: 

\[ \text{BWI} = \frac{\text{final body weight} - \text{initial body weight}}{\text{initial body weight}} \times 100 \]
\[ \text{SGR} = \left( \ln \text{final weight} - \ln \text{initial weight} \right) \times 100/\text{days} \]
\[ \text{FCR} = \frac{\text{feed consumption}}{\text{body weight gain}} \]
\[ \text{PER} = \frac{\text{weight gain}}{\text{protein intake}} \]
\[ \text{CF} = \frac{(\text{body weight} \times \text{body length}^3)}{\text{body length}} \times 100 \]
\[ \text{HSI} = \frac{\text{liver weight}}{\text{body weight}} \times 100 \]
\[ \text{survival rate} = \frac{\text{final number of fish}}{\text{initial number of fish}} \times 100 \]

(Hung et al., 1993, 1997, Haghighi et al., 2009; Luo et al., 2010).
Table 1: Ingredients of the experimental diets

<table>
<thead>
<tr>
<th>Ingredients (%)</th>
<th>Control</th>
<th>IS 1%</th>
<th>IS 3%</th>
</tr>
</thead>
<tbody>
<tr>
<td>Kilka fish meal</td>
<td>42</td>
<td>42</td>
<td>42</td>
</tr>
<tr>
<td>Meat meal</td>
<td>9</td>
<td>9</td>
<td>9</td>
</tr>
<tr>
<td>Soybean meal</td>
<td>19.5</td>
<td>19.5</td>
<td>19.5</td>
</tr>
<tr>
<td>Wheat flour</td>
<td>11</td>
<td>11</td>
<td>11</td>
</tr>
<tr>
<td>Sunflower oil</td>
<td>9</td>
<td>9</td>
<td>9</td>
</tr>
<tr>
<td>Molasses</td>
<td>1.5</td>
<td>1.5</td>
<td>1.5</td>
</tr>
<tr>
<td>Lecithin</td>
<td>0.2</td>
<td>0.2</td>
<td>0.2</td>
</tr>
<tr>
<td>L-Methionine</td>
<td>0.5</td>
<td>0.5</td>
<td>0.5</td>
</tr>
<tr>
<td>L-carnitine</td>
<td>0.1</td>
<td>0.1</td>
<td>0.1</td>
</tr>
<tr>
<td>Salt</td>
<td>1.5</td>
<td>1.5</td>
<td>1.5</td>
</tr>
<tr>
<td>Vitamin C</td>
<td>0.1</td>
<td>0.1</td>
<td>0.1</td>
</tr>
<tr>
<td>Vitamin E</td>
<td>0.1</td>
<td>0.1</td>
<td>0.1</td>
</tr>
<tr>
<td>Cellulose</td>
<td>3</td>
<td>2</td>
<td>0</td>
</tr>
<tr>
<td>Vitamin premix*</td>
<td>1.5</td>
<td>1.5</td>
<td>1.5</td>
</tr>
<tr>
<td>Mineral premix**</td>
<td>1</td>
<td>1</td>
<td>1</td>
</tr>
<tr>
<td>Immunoster</td>
<td>0</td>
<td>1</td>
<td>3</td>
</tr>
</tbody>
</table>

* Vitamin premix (g 100 g⁻¹ vitamin premix except A, 160000 IU and D₃, 40000 IU): E, 4; K₃, 0.2; B₁, 0.6; B₂, 0.8; B₃, 1.2; B₅, 4; B₆, 0.4; B₉, 0.2; B₁₂, 0.8; H₂, 0.02; C, 6; Inositol, 2; BHT (butylated hydroxyl toluene), 2.
** Mineral premix (g 100 g⁻¹ mineral premix): Fe, 2.6; Zn, 1.25; Se, 0.2; Co, 0.048; Cu, 0.42; Mn, 1.58; I, 0.1; Cholin chloride, 1.2.

Table 2: Proximate composition of experimental diets (Dry matter basis)

<table>
<thead>
<tr>
<th>Ingredients (%)</th>
<th>Control</th>
<th>IS 1%</th>
<th>IS 3%</th>
</tr>
</thead>
<tbody>
<tr>
<td>Moisture</td>
<td>6.1</td>
<td>5.9</td>
<td>6.2</td>
</tr>
<tr>
<td>Crude protein</td>
<td>42</td>
<td>41.3</td>
<td>42.2</td>
</tr>
<tr>
<td>Crude lipid</td>
<td>15</td>
<td>15.2</td>
<td>14.8</td>
</tr>
<tr>
<td>Fiber</td>
<td>2.4</td>
<td>2.2</td>
<td>2.3</td>
</tr>
<tr>
<td>Ash</td>
<td>10.1</td>
<td>10.2</td>
<td>10.1</td>
</tr>
<tr>
<td>NFE</td>
<td>30.5</td>
<td>31.1</td>
<td>30.6</td>
</tr>
<tr>
<td>Gross energy (MJ kg⁻¹)</td>
<td>14.65</td>
<td>14.69</td>
<td>14.62</td>
</tr>
</tbody>
</table>

* NFE, nitrogen free extract = 100- (Protein + Lipid + Fiber + Ash)

At the end of the trial, six fish per treatment (two fish per replicate) were randomly selected to carry out the carcass analysis. Proximate analysis of carcass was
performed according to AOAC (1995). Livers were excised and weighed to calculate HSI. At the end of trial, nine fish per treatment (three fish per replicate) were randomly captured and blood samples were collected using a 2–mL syringe from the caudal vein to evaluate immunophysiological indices. The extracted blood was divided in two sets of Eppendorf tubes. One set contained heparin for hematology studies and the other one (non-heparinized) was centrifuged at 3000 rpm for 10 min in order to measure biochemical and immune indices. All sets were stored at 80°C until analyzed. Before the blood samplings, fish were starved for 24 h. Hematocrit (Hct) values were determined using microhematocrit heparinized capillary tubes. The amount of hemoglobin (Hb) was measured according to the cyanmethemoglobin method. Red blood cell (RBC) and white blood cell (WBC) were counted in a Neubauer hemocytometer. Mean corpuscular volume (MCV= [Hematocrit × 10 / RBC]), mean corpuscular hemoglobin (MCH= [Hemoglobin × 10 / RBC]) and mean corpuscular hemoglobin concentration (MCHC= [Hemoglobin × 100 / Hematocrit]) were calculated. To estimate the differential leucocyte count, blood smears were prepared, air-dried, fixed in methanol and stained using Giemsa (Merck, Germany) (Klontz, 1994; Ameri Mahabadi, 2000; Zorriezhahra et al., 2010). Leucocytes in blood smears were categorized into lymphocytes, neutrophils, eosinophils and monocytes. Total serum protein was evaluated using the biuret reaction (Doumas et al., 1981; Soltani et al., 2010). Albumin was measured using the bromocresol green binding method (Doumas et al., 1971; Soltani et al., 2010). A digital freezing osmometer (Roebling, Germany) was utilized to assess osmolarity. Ca²⁺ and Mg²⁺ values were determined using colorimetric method using an autoanalyzer (Technicon RA-1000, USA) according to (Kazemi et al., 2006). Na⁺ and K⁺ concentrations were measured by means of flame photometer (Jenway, England). Immunoglobulin M (IgM) content was estimated according to the method described by Siwicki and Anderson (1993). Lysozyme levels were determined based on the method of Ellis (1990). Levene’s test was used to determine the homogeneity of variance. The means of all parameters such as growth performance, carcass composition and immunophysiological indices were subjected to one-way analysis of variance ANOVA, and comparisons among treatment means were made by Tukey as a post-hoc test using SPSS software (version 17). Statistical significance was accepted at the P<0.05 level. All data in the text are presented as mean ± SD.

Results

Growth indices are summarized in Table 3. Fish fed on IS at 1% and 3% had better growth performance during the 8-week feeding trial. Final weight, final length, BWI, SGR, FCR, PER and CF were significantly affected by IS 3% compared to IS 1% and the control (P<0.05). HSI was insignificantly higher in fish fed on IS 3% (P>0.05). Survival rate was 100% in all experimental treatments.
Carcass proximate composition is presented in Table 4. There was significant difference in crude protein among the experimental groups (P<0.05). Crude protein of fish fed on IS at 1% and 3% was significantly (P<0.05) higher than the fish fed on the control diet. Lipid, moisture and ash content of the whole body were not significant (P>0.05). Table 5 shows the levels of hematological indices of great sturgeon juveniles at the end of the trial. MCV and MCH were significantly higher in IS-fed groups compared to the control diet-fed group (P<0.05). There was an insignificant increase in Hct, WBC and neutrophil in fish fed on IS at 1% and 3% (P>0.05). Significant differences (P<0.05) were observed in lymphocyte, eosinophil and monocyte among the treatments.

### Table 3: Growth performance of *Huso huso* in the 8-week feeding trial (N=90 per treatment for growth indices and N= 6 per treatment for HSI)

<table>
<thead>
<tr>
<th>Growth index</th>
<th>Control</th>
<th>IS 1%</th>
<th>IS 3%</th>
<th>P-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Initial weight (g)</td>
<td>95.08 ± 10.30a</td>
<td>96.32 ± 9.82a</td>
<td>95.65 ± 10.11a</td>
<td>0.710</td>
</tr>
<tr>
<td>Final weight (g)</td>
<td>290.28 ± 58.23a</td>
<td>306.51 ± 58.18a</td>
<td>354.17 ± 57.28b</td>
<td>0.000</td>
</tr>
<tr>
<td>Initial length(cm)</td>
<td>30.84 ± 1.09a</td>
<td>30.96 ± 0.89a</td>
<td>30.85 ± 1.11a</td>
<td>0.655</td>
</tr>
<tr>
<td>Final length (cm)</td>
<td>42.26 ± 2.57a</td>
<td>42.67 ± 2.20a</td>
<td>44.52 ± 2.34b</td>
<td>0.000</td>
</tr>
<tr>
<td>BWI (%)</td>
<td>207.15 ± 13.85a</td>
<td>219.52 ± 19.78a</td>
<td>271.08 ± 10.68b</td>
<td>0.005</td>
</tr>
<tr>
<td>SGR (% day⁻¹)</td>
<td>1.99 ± 0.87a</td>
<td>2.06 ± 0.11a</td>
<td>2.34 ± 0.51b</td>
<td>0.006</td>
</tr>
<tr>
<td>FCR</td>
<td>1.70 ± 0.10b</td>
<td>1.58 ± 0.11a</td>
<td>1.30 ± 0.49a</td>
<td>0.005</td>
</tr>
<tr>
<td>PER</td>
<td>1.39 ± 0.08a</td>
<td>1.52 ± 0.10a</td>
<td>1.84 ± 0.06b</td>
<td>0.001</td>
</tr>
<tr>
<td>CF</td>
<td>0.38 ± 0.005a</td>
<td>0.39 ± 0.005ab</td>
<td>0.41 ± 0.010b</td>
<td>0.021</td>
</tr>
<tr>
<td>HSI (%)</td>
<td>3.59 ± 0.37a</td>
<td>3.38 ± 0.35a</td>
<td>3.66 ± 0.38a</td>
<td>0.404</td>
</tr>
</tbody>
</table>

Means in the same row with different superscripts are significantly different (P<0.05)

### Table 4: Carcass proximate composition of *Huso huso* in the 8-week feeding trial (N=6 per treatment)

<table>
<thead>
<tr>
<th>Ingredients (%)</th>
<th>Control</th>
<th>IS 1%</th>
<th>IS 3%</th>
<th>P-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Crude protein</td>
<td>14.69 ± 0.61a</td>
<td>14.91 ± 0.68a</td>
<td>15.52 ± 0.20b</td>
<td>0.049</td>
</tr>
<tr>
<td>Crude lipid</td>
<td>9.20 ± 1.04a</td>
<td>9.74 ± 1.96a</td>
<td>9.44 ± 1.35a</td>
<td>0.824</td>
</tr>
<tr>
<td>Ash</td>
<td>1.06 ± 0.04a</td>
<td>1.01 ± 0.04a</td>
<td>1.00 ± 0.04a</td>
<td>0.067</td>
</tr>
<tr>
<td>Moisture</td>
<td>73.84 ± 1.46c</td>
<td>73.48 ± 1.80b</td>
<td>73.06 ± 1.47a</td>
<td>0.682</td>
</tr>
</tbody>
</table>

Means in the same row with different superscripts are significantly different (P<0.05)

### Table 5: Hematological indices of *Huso huso* in the 8-week feeding trial (N= 9 per treatment)

<table>
<thead>
<tr>
<th>Hematological index</th>
<th>Control</th>
<th>IS 1%</th>
<th>IS 3%</th>
<th>P-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Hct (%)</td>
<td>23.00 ± 1.73a</td>
<td>24.55 ± 2.00a</td>
<td>24.55 ± 3.20a</td>
<td>0.302</td>
</tr>
<tr>
<td>Hb (g dL⁻¹)</td>
<td>5.35 ± 0.61a</td>
<td>5.27 ± 0.76a</td>
<td>5.57 ± 0.62a</td>
<td>0.624</td>
</tr>
<tr>
<td>RBC (×10⁶ mm⁻³)</td>
<td>0.79 ± 0.08a</td>
<td>0.72 ± 0.10a</td>
<td>0.69 ± 0.14a</td>
<td>0.187</td>
</tr>
<tr>
<td>WBC (×10⁹ mm⁻³)</td>
<td>64.05 ± 15.58a</td>
<td>66.66 ± 15.08a</td>
<td>70.66 ± 16.63a</td>
<td>0.674</td>
</tr>
<tr>
<td>MCV (Fl)</td>
<td>292.27 ± 22.05a</td>
<td>342.22 ± 51.62ab</td>
<td>364.68 ± 56.83b</td>
<td>0.009</td>
</tr>
<tr>
<td>MCH (pg)</td>
<td>67.83 ± 5.77a</td>
<td>73.13 ± 11.67ab</td>
<td>84.01 ± 19.48b</td>
<td>0.042</td>
</tr>
<tr>
<td>MCHC (%)</td>
<td>23.20 ± 1.37a</td>
<td>21.38 ± 1.89a</td>
<td>22.82 ± 2.58a</td>
<td>0.153</td>
</tr>
<tr>
<td>Lymphocyte (%)</td>
<td>47.44 ± 8.29a</td>
<td>46.00 ± 4.84ab</td>
<td>37.33 ± 11.22a</td>
<td>0.040</td>
</tr>
<tr>
<td>Neutrophil (%)</td>
<td>22.67 ± 6.81a</td>
<td>28.77 ± 6.26a</td>
<td>25.55 ± 8.07a</td>
<td>0.209</td>
</tr>
<tr>
<td>Eosinophil (%)</td>
<td>26.00 ± 5.36ab</td>
<td>22.22 ± 6.18b</td>
<td>35.66 ± 14.39b</td>
<td>0.018</td>
</tr>
<tr>
<td>Monocyte (%)</td>
<td>3.89 ± 1.90b</td>
<td>2.77 ± 2.22ab</td>
<td>1.44 ± 1.13a</td>
<td>0.029</td>
</tr>
</tbody>
</table>

Means in the same row with different superscripts are significantly different (P<0.05)
Biochemical indices are given in Table 6. No significant differences (P>0.05) were recorded in biochemical indices. Table 7 shows immune indices of *Huso huso*. IgM concentration and lysozyme level in the diet containing IS 1% were higher than the control diet.

### Table 6: Biochemical indices of *Huso huso* in the 8-week feeding trial (N= 9 per treatment)

<table>
<thead>
<tr>
<th>Biochemical index</th>
<th>Control</th>
<th>IS 1%</th>
<th>IS 3%</th>
<th>P-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Total protein (g dL$^{-1}$)</td>
<td>1.50 ± 0.10$^a$</td>
<td>1.56 ± 0.16$^a$</td>
<td>1.51 ± 0.24$^a$</td>
<td>0.742</td>
</tr>
<tr>
<td>Albumin (g dL$^{-1}$)</td>
<td>0.60 ± 0.03$^a$</td>
<td>0.62 ± 0.03$^a$</td>
<td>0.59 ± 0.10$^a$</td>
<td>0.638</td>
</tr>
<tr>
<td>Osmolarity (mOsmo L$^{-1}$)</td>
<td>314.88 ± 10.55$^a$</td>
<td>314.66 ± 9.24$^a$</td>
<td>318.55 ± 7.71$^a$</td>
<td>0.611</td>
</tr>
<tr>
<td>Na$^+$ (meq L$^{-1}$)</td>
<td>131.00 ± 2.06$^a$</td>
<td>131.66 ± 2.29$^a$</td>
<td>130.00 ± 2.95$^a$</td>
<td>0.368</td>
</tr>
<tr>
<td>K$^+$ (meq L$^{-1}$)</td>
<td>1.96 ± 0.35$^a$</td>
<td>2.04 ± 0.24$^a$</td>
<td>2.12 ± 0.28$^a$</td>
<td>0.554</td>
</tr>
<tr>
<td>Ca$^{2+}$ (mg dL$^{-1}$)</td>
<td>5.09 ± 0.60$^a$</td>
<td>5.20 ± 0.53$^a$</td>
<td>4.96 ± 0.95$^a$</td>
<td>0.793</td>
</tr>
<tr>
<td>Mg$^{2+}$ (meq L$^{-1}$)</td>
<td>1.22 ± 0.21$^a$</td>
<td>1.15 ± 0.17$^a$</td>
<td>1.14 ± 0.20$^a$</td>
<td>0.705</td>
</tr>
</tbody>
</table>

Means in the same row with the same superscript indicate no significant difference (P>0.05)

### Table 7: Immune indices of *Huso huso* in the 8-week feeding trial (N= 9 per treatment)

<table>
<thead>
<tr>
<th>Immune index</th>
<th>Control</th>
<th>IS 1%</th>
<th>IS 3%</th>
<th>P-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>IgM (mg dL$^{-1}$)</td>
<td>10.13 ± 4.65$^a$</td>
<td>11.41 ± 3.46$^a$</td>
<td>9.71 ± 4.84$^a$</td>
<td>0.694</td>
</tr>
<tr>
<td>Lysozyme (µg mL$^{-1}$)</td>
<td>0.38 ± 0.78$^a$</td>
<td>1.81 ± 3.65$^a$</td>
<td>0.45 ± 1.37$^a$</td>
<td>0.348</td>
</tr>
</tbody>
</table>

Means in the same row with the same superscript indicate no significant difference (P>0.05)

### Discussion

Results indicate that IS at 1% and 3% improved growth indices. Crude protein of carcass composition of fish fed on both levels of IS was significantly higher than the fish fed on the control diet. These results are in agreement with the following similar studies: Staykov et al. (2007) declared that a supplement of 0.2% MOS in rainbow trout, *Oncorhynchus mykiss* diet significantly enhanced body weight and reduced the FCR and mortality in comparison with the control diet. Torrecillas et al. (2007) reported a significant increase in body weight and total length as well as a positive correlation between the MOS levels and feed intake in European sea bass, *Dicentrarchus labrax*, fed on MOS at two levels of 2% and 4%. Enrichment of live feeds such as rotifers and Artemia with 0.2% MOS caused a greater ability to endure hyposaline stress in larval cobia, *Rachycentron canadum* (Salze et al., 2008). In another report, Andrews et al. (2009) evaluated that diets supplemented with 1%, 2% and 4% MOS improved WG, SGR and FCR in rohu, *Labeo rohita* fingerlings.

In contrast, Pryor et al. (2003) observed no differences in final weight, fork length, CF, SGR and FCR between control and 0.3% MOS supplemented groups in gulf sturgeon, *Acipenser oxyrinchus*. Grisdale-Helland et al. (2008) demonstrated that supplementing the diet with 1% MOS had no significant effects on digestibility, feed intake and growth of Atlantic salmon, *Salmo salar* and causing a decrease in the body protein concentration.
The use of MOS as prebiotic to improve growth indices in different species of fish still needs further surveys to explain contradictory results. It may be because of the different basal diets, inclusion levels, characteristics of the animal under study (species and age), period of trial, and circumstances of culture. Sado et al. (2008) declared that the complexity of carbohydrate structure in yeast's cell wall, yeast's various strains, fermentation, and processing procedures can modify their functions.

IS is considered as an immunostimulant for its potential (having β-1, 3 glucans). The use of immunostimulants is an important strategy in aquaculture. Some food components such as vitamins e.g. C and E, chitin, chitosan and several types of glucans like yeast glucan, peptide-glucan, and β-1, 3 glucan have been utilized as immunostimulants in fish (Sakai, 1999).

The analysis of blood parameters is a precious indicator in evaluating the conditions of aquatic animals. For instance, in response to stress, different sorts of pollutants, nutrition, ecological and physiological changes are involved (Bahmani et al., 2001). Leucocytes are one of the most important cells that can stimulate immune responses of fish. These cells produce antibody and can perform macrophagus activity (Jalali et al., 2009). In the current study, the WBC was higher in the IS 3% but no significant difference was seen. The increase in leucocyte count is due to glucan, which can detect particular receptors on WBCs. When β-1, 3 glucans settle on the receptors, the cells start to swallow bacteria and secrete cytokines that stimulate the establishment of new WBCs (Raa, 1996 cited in Andrews et al., 2009). In L. rohita, the leucocyte count in fish treated with MOS at 1%, 2% and 4% was higher than the control (Andrews et al., 2009). Welker et al. (2007) revealed that WBC counts in channel catfish, Ictalurus punctatus fed on Bio-MOS™ at 2 g kg⁻¹ were insignificantly higher compared to fish fed on the control diet.

Total serum protein values in fish fed on IS at 1% and 3% and albumin value in IS 1% group were higher than the control group but no significant difference was found. The increase in total serum protein and albumin concentrations can be due to stronger non-specific responses in fish (Wiegertjes et al., 1996 cited in Andrews et al., 2009).

Immunization of sturgeons against pathogens has not been developed as it has for cyprinids and salmonids (Khoshbavar-Rostami et al., 2007). IgM is an important part of the humoral immune system. Administration of vitamin A, chitin, yeast cells and levamisole as immunostimulants to the diet of Sparus aurata, increased IgM values (Cuesta et al., 2004). In this study, IS 1% insignificantly enhanced IgM concentration in great sturgeon juveniles. Administration of IS 1% (because of having MOS and β-1, 3 glucans) resulted in an increase in serum lysozyme which can contribute to the enhancement in the non-specific immunity. Staykov et al. (2007) reported that O. mykiss treated with MOS at an inclusion rate of 0.2% showed significant differences in lysozyme levels. However, in S. salar, lysozyme concentration was lower in the MOS-fed group compared with the control diet-fed group (Grisdale-Helland et al., 2008).
MCV and MCH were significantly (P<0.05) affected by IS at 1% and 3% compared with the control. Sado et al. (2008) demonstrated that supplementing the diet with 0.2%, 0.4%, 0.6%, 0.8% and 1% MOS had no significant effects on MCV and MCH in Oreochromis niloticus.

There is no record on the effect of MOS on osmolarity and blood serum ions in fish. The effects of MOS on osmolarity and Na⁺, K⁺, Ca²⁺ and Mg²⁺ were studied for the first time in this survey. Results indicate that IS had no significant effects on the above mentioned parameters in H. huso. Studies on the use of MOS in fish hematology are few and involve immunological factors including lysozyme concentration, antibody titer, bactericidal activity, and alternative complement pathway (Torrecillas et al., 2007; Staykov et al., 2007).

In the present study, the effects of IS did not show significant differences in some hematological and biochemical parameters of H. huso. Welker et al. (2007) found no significant differences in the hematological variables such as RBC, Hb, Hct and WBC in I. punctatus fed on MOS at 2 g kg⁻¹. Sado et al. (2008) declared that diets supplemented with 0.2%, 0.4%, 0.6%, 0.8% and 1% MOS had no significant effects on RBC, Hb, Hct, WBC and total protein in O. niloticus. On the contrary, Andrews et al. (2009) observed a significant improvement in WBC, RBC, Hb, serum protein, albumin, and globulin in L. rohita fed on the MOS-supplemented diet in comparison with those fed on the control diet. It appears that fluctuations in hematological and biochemical variables may be associated to characteristics of species, inclusion rates of MOS, ingredients of diets, rearing period, etc.

In conclusion, the results of this trial indicate that IS can enhance growth performance, body composition and affect some immunophysiological variables such as WBC, MCV, MCH, total protein, albumin, IgM and lysozyme in H. huso. Further research is needed to clarify the action mechanisms of MOS, as well as the appropriate inclusion dose and feeding period in great sturgeon.

Acknowledgements
The authors would like to appreciate staff of Shahid Dr. Beheshti Sturgeon Fish Propagation and Rearing Center and Dr. Dadman International Sturgeon Research Institute. The authors also express their sincere thanks to Mr. Mohammad Pourdehghani and Mr. Mehdi Maleki for their assistance during blood analysis.

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Growth efficiency, body composition, survival and hematological changes in great sturgeon (*Huso huso*) juveniles fed diets supplemented with different levels of Ergosan. *Aquaculture Research*, 40, 804-809.


