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Published in:
Journal of Applied Ichthyology

Link to article, DOI:
10.1111/jai.13153

Publication date:
2016

Document Version
Publisher's PDF, also known as Version of record

Received: 27 October 2015 | Accepted: 27 May 2016

DOI: 10.1111/jai.13153

ORIGINAL ARTICLE

WILEY Journal of Applied Ichthyology DWK

Effects of dietary prebiotic GroBiotic®-A on growth performance, plasma thyroid hormones and mucosal immunity of great sturgeon, *Huso huso* (Linnaeus, 1758)

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Summary

The present study was conducted to evaluate the effects of Grobiotic®-A, a commercial prebiotics, when administered in feed on the growth performance, plasma thyroid hormones and mucosal immunity of great sturgeon (*Huso huso*). The commercial prebiotic mixture was supplemented in the diets at four different levels (i.e. 0.0% as control, 0.5%, 1% and 2%, in three replicates, 20 fish per replicate) and fed to the fish for an 8-week period wherein 240 fish were cultured in 1,800-L fiberglass tanks that formed part of a flow-through system. Water temperature was maintained at 20.4 ± 1.5°C. Significant changes in growth performance parameters were observed, but only in those groups fed with 1% and 2% prebiotics. Specifically, marked improvements relative to the control group were observed in percentage weight gain, body weight gain, feed conversion ratio and specific growth rate in prebiotic-fed fish. The levels of plasma thyroid hormones, specifically thyroxine and thyroid stimulating hormones were significantly elevated in the group receiving 2% prebiotics. Activities of lysozyme and alkaline phosphatase in skin mucus were significantly enhanced in prebiotics-fed groups, particularly at an inclusion level of 1% and higher (2% group compared to the control). Inhibitory activity of the skin mucus against pathogens, particularly *Streptococcus iniae* and *Yersinia ruckeri*, was significantly improved following prebiotic feeding. Taken together, dietary inclusion of GroBiotic®-A promoted growth, modulated thyroid hormones, and enhanced mucosal immunity of *H. huso*. This prebiotic mixture has the potential for use in improving the growth performance and health status of farmed great sturgeon.

1 | INTRODUCTION

In the 1990s, the great sturgeon (*Huso huso*) was already listed in the CITES (Convention on International Trade in Endangered Species) as an endangered species (Nazari, Sohrabnejad, & Ghomi, 2009). To address the declining wild stocks, the potential of the great sturgeon for culture under farmed conditions has been actively explored in the last two decades. Some of their features for potential domestication are a fast growth rate, high adaptability to controlled environmental conditions, and their high value for meat and caviar (Jalali, Ahmadifar, Sudagar, & Azari Takami, 2009).

Under culture conditions, sturgeons are easily infected by several opportunistic pathogenic bacteria (e.g. *Streptococcus* sp., *Aeromonas* sp., *Yersinia* sp., and *Vibrio* sp.), in addition to stressful conditions such as poor water quality and high stocking density, all of which continue to be perennial challenges in many intensive aquaculture farms (Yang & Li, 2009). The increasing resistance to antibiotics, medication, and other side effects from drugs associated with the overdependence of

the aquaculture industry on synthetic antimicrobials to combat diseases necessitates the search for alternative strategies to improve health and minimize dependence on these drugs (Lazado & Caipang, 2014; Nayak, 2010). In recent years, significant attention has been paid towards dietary supplements such as probiotics, prebiotics and synbiotics in order to improve growth performance as well as the health and welfare status of farmed aquatic animals (Caipang & Lazado, 2015; Cerezuela, Meseguer, & Esteban, 2011; Yousefian et al., 2012).

Prebiotics are non-digestible food ingredients that selectively stimulate the growth and/or activate the metabolism of certain health-promoting bacteria in the gut, thereby improving the host intestinal balance (Fooks, Fuller, & Gibson, 1999; Ghaedi, Keyvanshokooh, Mohammadi Azarm, & Akhlaghi, 2015). Some of the prebiotic mixtures widely used in aquaculture at experimental and farming levels include inulin, oligofructose, mannanoligosaccharide (MOS), galactooligosaccharide (GOS), β- 1, 3 glucan, ImmunoWall™ and Grobiotic®-A (Burr, Hume, Ricke, Nisbet, & Gatlin, 2010; Caipang & Lazado, 2015; Cerezuela, Cuesta, Meseguer, & Esteban, 2008). Previous studies on the use of some of these dietary supplements revealed growth performance and increased immunity of great sturgeon (Ahmadifar, Akrami, Ghelichi, & Mohammadi Zarejabad, 2011; Akrami, Hajimoradloo, Matinfar, & Abedian Kinari, 2009; Akrami et al., 2013; Hoseinifar et al., 2010; Ta'ati, Soltani, Bahmani, & Zamini, 2011), hence affirming their potential in improving farming conditions.

Grobiotic®-A is a commercial prebiotic mixture composed of partially autolyzed brewer's yeast, dairy ingredient components, and dried fermentation products (Li & Gatlin, 2004). The beneficial properties of this probiotic mixture include growth promotion, efficient nutrient utilization, immunomodulation and conferment of disease resistance, among others (Hoseinifar et al., 2010; Ta'ati et al., 2011). Dietary supplementation of Grobiotic®-A has been evaluated in several aquaculture species such as rainbow trout *Oncorhynchus mykiss* (Sealey et al., 2007), hybrid striped bass *Morone chrysops* × *M. saxatilis* (Li & Gatlin, 2004), goldfish *Carassius auratus* (Raggi & Gatlin, 2012), red drum *Sciaenops ocellatus* (Buentello, Neill, & Gatlin, 2010), Caspian kutum *Rutilus frissi* (Yousefian et al., 2012), and juvenile Nile tilapia *Oreochromis niloticus* (Vechklang et al., 2012). To the best of our knowledge, this prebiotic mixture has not been tested on farmed *Huso huso*. This study was therefore designed to evaluate the effects of dietary inclusion of Grobiotic®-A on the growth performance, thyroid and cutaneous innate immunity of the great sturgeon, to determine if it would be a good supplement for use in aquaculture.

2 | MATERIALS AND METHODS

2.1 | Fish

The experiment was carried out at the Sturgeon Culture Center (Samandak, Sari), Mazandaran Province, northern Iran. Apparently healthy fish with an average body weight of 40.8 ± 6 g (mean ± *SD*, *n* = 240) were randomly selected from the holding tanks. The fish were distributed among the 12 1,800-L fiberglass tanks (2.0 × 2.0 × 0.5 m), with a stocking density of 20 fish per tank. Water parameters (mean ± *SD*) were monitored daily in each tank and maintained as follows: average dissolved oxygen content (5.7 ± 0.4 mg/L), pH (7.76 ± 0.4), temperature (20.4 ± 1.5°C), salinity (2.4 ± 0.11 g/L) and electrical conductivity (5826.4 ± 159.3 MM/cm). In addition, a daily 14L:10D photoperiod (light intensisty 180 lux during the feeding experiment) was provided to the cultured fish, which were acclimated to these conditions for approximately 2 weeks before the start of the feeding experiment. During this period, the fish were fed three times daily at a ration of 3%–4% body weight.

2.2 | Bacterial pathogens

Bacterial pathogens including *Streptococcus iniae* (ATCC29178), *Yersinia ruckeri* (KC291153), *Listeria monocytogenes* (ATCC1143) and *Escherichia coli* (PTCC 1037), were obtained from the Persian Type Culture Collection.

2.3 | Preparation of experimental diets

The prebiotic used in the study is commercially known as GroBiotic®-A (International Ingredient Corporation, Fenton, Missouri, USA) consisting of partially autolyzed brewer's yeast, dairy ingredient components

TABLE 1 Dietary formulation and proximate composition of basal diet used in the study

Ingredient	Composition (%)
Kilka fishmeal	58.0
Wheat flour	19.0
Fish oil	5.2
Soybean oil	5.8
Vitamin premix[a]	3.0
Mineral premix	2.5
Cellulose	2.5
Binder[b]	2.0
Salt	1.0
Anti-fungi[c]	0.4
Antioxidant[d]	0.25
Proximate composition (% dry matter)	
Dry matter	98.84
Crude protein	40.32
Crude lipid	18.86
Ash	9.6
Fiber	2.8
Moisture	8.1
NFE[e]	18.8
P/E ratio (mg protein/kJ)	19.68
Gross energy (kJ/g diet)[f]	21.84

[a]Premix detailed by Jalali et al. (2009).
[b]Amet binder (MehrTaban-e-Yazd, Iran).
[c]ToxiBan antifungal (Vet-A-Mix, Shenandoah, IA).
[d]Butylatedhydroxytoluene (BHT; Merck, Germany).
[e]Nitrogen-free extracts (NFE) = dry matter − (crude protein + crude lipid + ash + fibre).
[f]Gross energy (kJ/g diet) calculated according to 23.6 kJ/g forprotein, 39.5 kJ/g for lipid and 17.0 kJ/g for NFE.

and dried fermentation products. Four experimental diets were prepared for the study: the basal diet (Table 1) as the control, plus three prebiotic test diet groups supplemented at levels of 0.5%, 1% and 2%. All diets were prepared at the same time and stored in sterile plastic bags at 4°C until used. GroBiotic®-A was replaced with cellulose in the basal diet (control diet, Table 1). Proximate composition of the basal diet was according to the standard methods described by the Association of Official Analytical Chemists (AOAC, 2005): moisture was determined by oven-drying the samples at 105°C (Behr, Germany); crude lipid was determined by chloroform methanol extraction (2:1, v/v); crude protein was determined (Kjeldahl procedure: N × 6.25) using an automatic Kjeldahl system; ash was measured by incineration in a muffle furnace at 500°C for 6 hr.

2.4 | Feeding

The feeding trial lasted for 8 weeks. The basal and supplemented feeds were manually administered at three designated times of day, with a feeding ration of 3% body weight per day. Water parameters as enumerated above were monitored daily to ensure that they were within the range of the biological requirements of the fish throughout the feeding experiment.

2.5 | Sampling strategies

No feed was given 24 hr prior to weighing and sampling the fish. Fish were anesthetized with clove oil (100 mg/L) and thereafter circa 5 ml of blood was collected from the caudal vein with heparinized syringes. The collection tubes with blood samples were immediately placed in ice. Plasma was separated by centrifugation at 3,000 × *g* for 15 min, divided into aliquots and stored at −80°C until used (Adel, Abedian Amiri, Zorriehzahra, Nematolahi, & Esteban, 2015).

At the end of the experiment mucus samples were collected from nine fish per replicate tank following previously described protocol (Balasubramanian et al., 2013). Briefly, mucus from an individual fish was scraped in an anterior to posterior direction of the dorsal body surface using a sterile spatula. The collected mucus was then thoroughly mixed with an equal volume of sterile Tris-buffered saline (TBS, 50 mmol/L Tris HCl, pH 8.0, 150 mmol/L NaCl) and centrifuged at 30,000 g at 4°C for 15 min. The supernatant was then collected, filtered and kept at −80°C until further analysis.

2.6 | Evaluation of growth performance

The following calculations were undertaken at the end of feeding trial, based on the measurements described above:

$$\text{Weight gain} = W_2\,(\text{g}) - W_1\,(\text{g});$$

$$\text{Protein efficiency ratio (PER)} = \text{WG (g)}/\text{protein intake (g)};$$

$$\text{Specific growth rate (SGR\%)} = 100\,(\ln W_2 - \ln W_1)/T.$$

where W_1 is the initial weight, W_2 is the final weight and T is the number of days in the feeding period.

$$\text{Feed conversion ratio (FCR)} = \text{Feed intake (g)}/\text{Weight gain (g)},$$

$$\text{Survival rate\%} = (\text{Final number of fish}/\text{initial number of fish}) \times 100.$$

2.7 | Quantification of thyroid hormones

At the end of the experiment, the levels of plasma thyroid hormones were analyzed by commercially available ELISA kits (Delaware Biotech Inc., Heidelberg, Germany). The hormones quantified based on the manufacturer's standardized protocols were: thyroxine (T4; Delaware Biotech Inc., Heidelberg, Germany), tri-iodothyronine (T3) and thyroid stimulating hormone (TSH; DiaMetra Co., Milano, Italy).

2.8 | Cutaneous innate immune responses

Total protein concentration of skin mucus was measured colorimetrically at 540 nm using bovine serum albumin as a standard (Lowry, Rosebrough, Farr, & Randall, 1951).

Alkaline phosphatase activity was quantified using a Pars Azmoon kit (Tehran Company, Iran) according to the manufacturer's instructions, and absorbance was read at 405 nm with a spectrophotometer (Sanchooli, Hajimoradloo, & Ghorbani, 2012).

Lysozyme activity was determined as earlier described by Ellis (1990), with minor modifications. Briefly, 25 µl mucus was added to 1 ml of a suspension of Micrococcus lysodeikticus (0.2 mg/ml in a 0.05 mol/L sodium phosphate buffer [pH 6.2]) and absorbance was measured at 450 nm after 0.5 and 3 min.

At the end of the experiment, antimicrobial activities of the skin mucus (from nine fish per replicate tank) against known pathogens were determined by disc diffusion assay (Bauer, Kirby, Sherris, & Turck, 1966). From overnight cultures in tryptic soy broth of each bacterial pathogen, 0.1 ml (containing 1.5 × 10^6 CFU/ml bacteria) was individually cultured on Mueller Hinton agar plates. Sterile filter paper discs containing 10 µl mucus samples were placed above the seeded plate agar and then incubated at 25°C for 48 hr (Turker, Birinci Yildırım, & Pehlivan Karakaş, 2009). After the incubation period, the zone of inhibition was measured. All plates were performed in triplicate.

Minimum inhibitory concentration (MIC) of skin mucus against bacterial pathogens was determined by serial dilution method as described by Wei, Xavier, and Marimuthu (2010). Different concentrations of mucus extract (25, 50, 75, 100, 125, 150, 200, 250 and 300 µl) were added into a tube containing 1.5 × 10^6 CFU/ml of a specific bacterial pathogen and incubated at 37°C for 24 hr. The MIC is defined as the lowest concentration of the mucus extract at which the pathogen does not demonstrate visible growth (Turker et al., 2009).

2.9 | Statistical analysis

The data were subjected to statistical analysis using the SPSS software version no. 18 (SPSS Inc., Chicago, IL, USA). After satisfying the assumptions of normality and equal variance, the data were analyzed by one-way analysis of variance (ANOVA) followed by Duncan's multiple range tests. Significance was determined at α = .05.

3 | RESULTS

3.1 | Effects on growth performance

The growth performance of juvenile beluga fed diets supplemented with different levels of dietary GBA are presented in Table 2. All studied parameters were affected by prebiotic feeding, but only in groups with 1% and 2% in-feed prebiotic supplements. Distinctively clear effects were noted with body weight gain where the 1%-GA group increased by 30% relative to control, while an almost 50% increment was observed in the 2%-GA group. FCR and SGR also improved significantly in groups receiving 1% and 2% prebiotics in their diets. PER and %SR were significantly improved but only in the group receiving 2% prebiotics.

3.2 | Effects on plasma thyroid hormones

The effects of dietary inclusion of GroBiotic®-A on three thyroid hormones of *H. huso* are detailed in Table 3, whereby significant increases in thyroid hormones were observed only in the 2%-GA group. The remaining inclusion levels did not show any significant alterations in the level of thyroid hormones. The level T4 hormone level increased significantly—by almost 80% in the 2%-GA group. On the other hand, the plasma TSH level was elevated by a significant 8% in the same prebiotic-fed group.

TABLE 2 Effects of dietary inclusion of GroBiotic®-A on growth performance parameters of *H. huso*. Values are mean ± *SD* of 10 individual fish per replicate tank

Parameters	Control	0.5%-GA	1%-GA	2%-GA
Weight gain (%)	57.96 ± 5.6^{a}	56.5 ± 4.6^{a}	94.79 ± 6.32^{b}	98.68 ± 7.14^{b}
BW gain (g)	23.64 ± 1.29^{a}	23.06 ± 1.23^{a}	30.22 ± 1.86^{b}	34.75 ± 1.16^{c}
PER	0.82 ± 0.05^{a}	0.83 ± 0.06^{a}	0.88 ± 0.08^{a}	0.96 ± 0.1^{b}
FCR	2.09 ± 0.2^{a}	2.16 ± 0.4^{a}	1.81 ± 0.3^{b}	1.72 ± 0.2^{c}
SGR (%/day)	1.84 ± 0.16^{a}	1.76 ± 0.12^{a}	2.04 ± 0.24^{b}	2.22 ± 0.32^{b}
SR%	88 ± 1.6^{a}	93 ± 2.2^{a}	96 ± 1.1^{ab}	98.2 ± 1.3^{b}

BW gain, Body weight gain; PER, Protein efficiency ratio; FCR, fed conversion ratio; SGR, specific growth rate; %SR, percentage survival rate.
Values in a row with different superscripts show significant differences ($p < .05$; $n = 10$).

TABLE 3 Plasma thyroid hormones of *H. huso* fed GroBiotic®-A for 8 weeks. Values are mean ± *SD* of 10 fish per replicate tank

Thyroid hormones	Control	0.5%-GA	1%-GA	2%-GA
T4 (μg/dl)	0.87 ± 0.09^{a}	1.08 ± 0.13^{a}	1.26 ± 0.18^{ab}	1.58 ± 0.22^{b}
T3 (μg/dl)	0.64 ± 0.02^{a}	0.74 ± 0.06^{a}	0.68 ± 0.09^{a}	0.61 ± 0.04^{a}
TSH (μg/dl)	0.58 ± 0.04^{a}	0.52 ± 0.08^{a}	0.50 ± 0.07^{a}	0.63 ± 0.05^{b}

Values in a row with different superscripts show significant differences ($p < .05$; $n = 10$).

3.3 | Effects on skin mucus innate immune responses

The 0.5%-GA and 1%-GA groups showed no significant changes in their skin mucus protein levels after 8-week exposure (Fig. 1). However, the group that received 2% prebiotics in the diet exhibited a significant elevation of approx. 20% in skin mucus protein.

Prebiotic feeding resulted in significantly elevated levels of skin mucus lysozyme, especially in the 1%-GA and 2%-GA groups (Fig. 2). Mean lysozyme activity in the 1%-GA group was 43.6 (±0.23) IU/mg, which was 34% higher compared to the control. On the other hand, lysozyme activity in the 2%-GA group was 50.08 ± 0.36 (mean ± *SD*, $n = 10$) IU/mg, equal to an almost 55% increase relative to the control group.

The activity of skin mucus alkaline phosphatase was influenced in almost the same manner with that observed in lysozyme activity (Fig. 3). Significant changes were observed only in groups receiving an inclusion level of 1% and higher (Fig. 3). Skin mucus from 1%-GA and

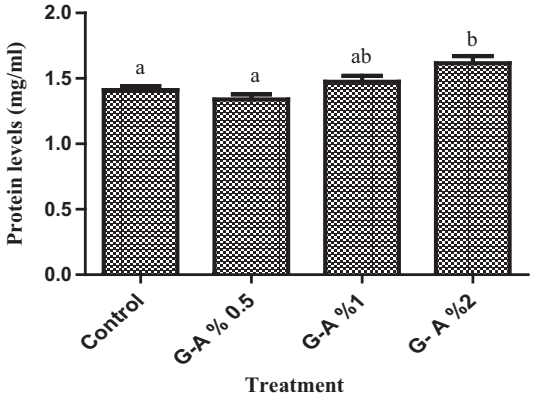

FIGURE 1 Protein levels in skin mucus of *Huso huso* fed with GroBiotic®-A for 8 weeks. Values presented here are mean ± *SE* of 10 individual fish per replicate tank. Different letter notations indicate significant differences at $p < .05$

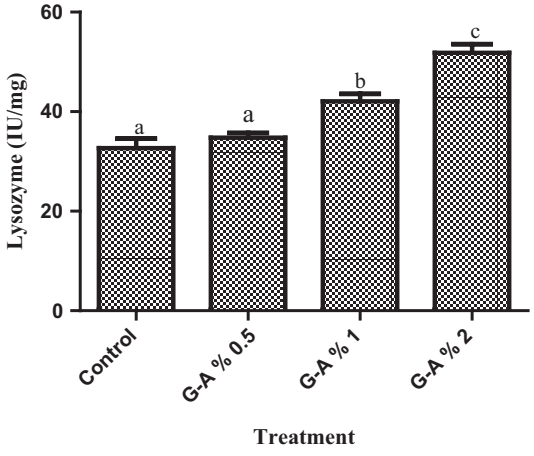

FIGURE 2 Lysozyme activity (IU/mg) in skin mucus of *Huso huso* fed with GroBiotic®-A for 8 weeks. Values presented here are mean ± *SE* of 10 individual fish per replicate tank. Different letter notations indicate significant differences at $p < .05$

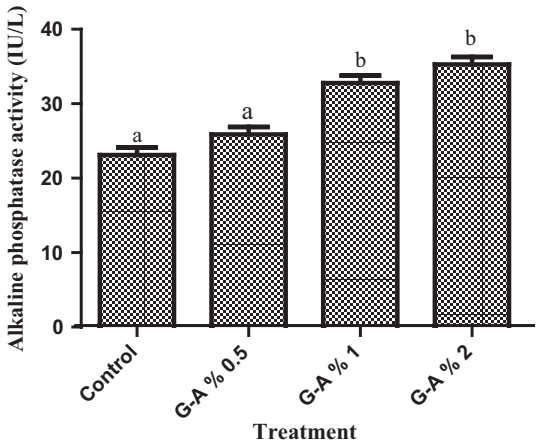

FIGURE 3 Alkaline phosphatase (IU/L) activity in skin mucus of *Huso huso* fed GroBiotic®-A for 8 weeks. Values presented here are mean ± *SE* of 10 individual fish per replicate tank. Different letter notations indicate significant differences at $p < .05$

2%-GA groups displayed an elevated alkaline phosphatase activity of at least 40% relative to the control group.

Marked enhancement in the antibacterial activity of *H. huso* skin mucus against different bacterial pathogens was observed in groups receiving 1% and 2% in-feed prebiotics (Table 4). In these two prebiotic-fed groups, for instance, inhibitory activity against *S. iniae* was promoted by at least 44% while inhibition of *Y. ruckeri* improved by no less than 33%. MIC of skin mucus from control fish performed at highest concentration against *S. iniae* and at lowest concentration with *Y. ruckeri*. The MICs of skin mucus from fish groups receiving 1% and 2% prebiotics were generally lower compared with the 0.1%-GA group. The MIC of skin mucus against *S. iniae* and *Y. ruckeri* for 1%-GA and 2%-GA groups was identical.

4 | DISCUSSION

The present study demonstrated the benefits from Grobiotic®-A, a commercial prebiotics, which consist of partially autolyzed brewer's yeast, dairy ingredient components, and dried fermentation products (Li & Gatlin, 2004, 2005; Zheng, Wang, Gatlin, & Ye, 2011). Inclusion of the prebiotics in the diet significantly impacted growth performance, plasma thyroid hormones and mucosal immunity of *Huso huso*.

The application of Grobiotic®-A in other fish species resulted in increased weight gain, improved feed efficiency, and better nutrient digestibility as well as enhanced disease resistance (Buentello et al., 2010; Li & Gatlin, 2004, 2005; Vechklang et al., 2012). Th present study corroborated those earlier findings on the beneficial effects of growth performance. In particular, 2% Grobiotic®-A in the diet significantly influenced the growth performance parameters of the fish, specifically in weight gain. However, in a study with hybrid striped bass, Li and Gatlin (2004) did not observe such an increase in weight gain. This apparent difference could be due to the biological peculiarities between fish species and/or the duration of prebiotic feeding.

Hormones play pivotal roles in the regulation of growth and nutrient intake in fish, and are sensitive to changes in feed intake (MacKenzie, Van Putte, & Leiner, 1998). Thyroid hormones such as thyroxine and tri-iodothyronine, which are the principal thyroid hormones secreted from the hypothalamic-pituitary-thyroid axis, are involved in many physiological processes during growth, development, behavior, and stress in fish (Peter, 2011; Power et al., 2001). In the present study, dietary supplementation of this prebiotic at 2% level was found to have an enhancing effect on thyroid hormones. Metabolic functions of thyroid hormones have been documented in a number of studies and which display a strong correlation with growth-promoting activities (Garg, 2007; Leatherland, 1994). It is plausible that the indirect influence of prebiotics on thyroid hormones is related to the growth-promoting features of this feed additive. However, the mechanism behind such a benefit has yet to be unraveled in future studies.

While the role of prebiotics on the growth, nutrition and physiological responses of fish has been widely demonstrated in a number of studies, the involvement of prebiotics in stimulating fish immunity has been documented less frequently (Buentello et al., 2010; Cerezuela et al., 2008; Sealey et al., 2007). In the present study, we explored the effects of dietary prebiotics in mucosal immunity of *H. huso*. Lysozyme is an important component of the innate immunity in fish and which catalytically hydrolyzes the bond between N-acetyl muramic acid and N-acetyl glucosamine in the cell wall of bacteria (Shailesh & Sahoo, 2008). On the other hand, alkaline phosphatase has been

TABLE 4 Inhibitory activity of skin mucus of *H. huso* fed GroBiotic®-A for 8 weeks. Values are mean ± *SD* of nine fish per replicate tank

	Control		0.5%-GA		1%-GA		2%-GA	
Pathogens	Inhibition zone size (mm)	MIC (μl/ml)	Inhibition zone size (mm)	MIC (μl/ml)	Inhibition zone size (mm)	MIC (μl/ml)	Inhibition zone size (mm)	MIC (μl/ml)
Streptococcus iniae	5.4 ± 0.4^{a}	250	5.8 ± 0.2^{a}	250	8.5 ± 0.4^{b}	200	7.8 ± 0.2^{b}	200
Yersinia ruckeri	8.1 ± 0.32^{a}	75	7.9 ± 0.6^{a}	75	10.8 ± 0.5^{b}	50	12.3 ± 0.6^{c}	50
Escherichia coli	5.8 ± 0.4^{a}	100	6.2 ± 0.2^{a}	75	10.8 ± 0.3^{b}	>50	9.8 ± 0.5^{b}	75
Listeria monocytogenes	6.3 ± 0.3^{a}	125	5.9 ± 0.2^{a}	>100	11.2 ± 1.2^{b}	100	10.6 ± 0.9^{b}	75

Values in a row with different superscripts show significant differences ($p < .05$; $n = 9$).

demonstrated as a potential stress indicator in the epidermal mucus of fish and has a protective role during the first stages of wound healing (Iger & Abraham, 1994). Our results demonstrated that prebiotic feeding enhances these cutaneous immune components.

The fish mucus is known to contain a number of anti-microbial substances, mainly attributed to the presence of lysozyme, agglutinins, thermolabile complement factors, or immunoglobulin (Hellio, Pons, Beaupoil, Bourgougnon, & Le Gal, 2002). In the present study, prebiotic-fed groups showed enhanced inhibitory activities with least MIC values against all tested pathogens under in vitro conditions. This enhancing effect will therefore be helpful in preventing the adherence and multiplication of pathogens in fish, specifically in the mucus, which has an intimate contact with the immediate environment.

In conclusion, the results of the present study revealed that prebiotic Grobiotic®-A could be a potential dietary additive for farmed great sturgeon. In particular, its dietary inclusion appears to improve the growth performance and health status of fish. The changes identified following prebiotic feeding accounted for the response only in the short-term, experimental set-up. Future studies will be directed toward looking into the effects of administering prebiotics during the entire production cycle and/or at the long-term effects of short-term prebiotic feeding. The findings discussed here add to the growing evidence on the prospects of using prebiotics as health-promoting feed additives in aquaculture, which in turn is currently fostering more sustainable eco- and consumer-friendly dietary supplements.

ACKNOWLEDGEMENTS

Financial support from the Iranian Fisheries Research Organization (Tehran, Iran) and the Sari University of Agricultural Sciences and Natural Resources (Sari, Iran) is gratefully acknowledged.

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