PERFORMANCE EVALUATION IN THE GROWING OF THE STELLATE STURGEON FRY IN 35 DAYS FROM HATCHING UNDER THE CIRCUMSTANCES OF ENRICHENING FOOD WITH PROBIOTICS

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Abstract

The main objective of the present paper was to evaluate the influence of the commercial probiotic BetaPlus® Ultra on the growth performance of the stellate sturgeon after 35 days of hatching, but the specific objective was to evaluate the food retention efficiency in the transition period from feeding with natural feed to that with commercial feed. For this purpose, a 93-day experiment was carried out with 6 experimental variants (5 different probiotic concentrations : V_1-1.28x10^{13} CFU, V_2-2.56 x10^{13} CFU, V_3 -3.84x10^{13} CFU, V_4 – 5.12x10^{13} CFU, V_5 -6.4x10^{13} CFU and V_6- control variant, in duplicates). The experiment started with a number of 6000 exemplars of 35-day post hatched larvae (500 specimens/rearing unit) with a mean individual weight (± SD) of 0.35 ± 0.02 g and a total length between 3-5 cm. According to the rearing technology, sorting was carried out throughout the experimental period which led to the division of the experiment into 9 different experimental stages according to feeding protocol.

Feeding efficiency, expressed by FCR and protein retention, indicated by PER shows an insignificant increase ($p > 0.05$) in the V2 variant and an insignificant decrease in the case where the probiotic (V_6) was not administered. Therefore, according to this experiment, a concentration of 2.56x10^{13} CFU / kg of commercial probiotic BetaPlus® Ultra represents an optimum value for promoting the growth, feeding efficiency and retention of proteins of trout after more than one gram.

Key words: sturgeon, probiotic, FCR, PER

INTRODUCTION

In aquaculture, probiotics are used for a short period of time, but in the recent years they have become an integral part of aquaculture with a positive role for increasing and improving disease resistance.

Numerous probiotic microorganisms have been identified for aquaculture practices, many of which differ significantly in their mode of action. However, there are several common mechanisms of action for probiotic strains. Among the common mechanisms of probiotics, we mention: it helps in efficient food conversion and muscle tissue enhancement ([1], [15]) and offers protection against pathogens by competitive exclusion through adhesion to the substrate, producing organic acids (formic acid, acetic acid, lactic acid), hydrogen peroxide and many other ant bactericidal compounds, lysozyme ([5],[18]). There are several ways of administering probiotics in aquaculture among which we mention the most frequently used: bathing, administering water supplements and enriched food. However, enriched food with probiotics has proven to be the most effective method of colonization and establishment of strains in the intestine ([11 [8],[14]).

The stellate sturgeon, a species of fish from the Acipenseridae family, with a great potential in aquaculture, has a lower

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adaptability to commercial feed compared to other sturgeon species ([13]). Therefore, the addition of probiotics to feed could help producers to reduce the losses recorded during transition from natural feed to commercial feed.

The general objective of the present experiment was to assess the influence of the commercial probiotic BetaPlus® Ultra on the growth performance and survival of the sturgeon larvaein the transition period from feeding with natural feed to that with commercial feed.

MATERIAL AND METHOD

The present experiment was carried out between 19.06.2012 and 18.09.2012 (93 days) in a commercial sturgeon farm located in Horia, Tulcea county, Romania.

The experimental facilities consisted in a flow-through system with 12 rearing units. Tanks of light green color, truncated-cone shape, with a capacity of 330 liters and with reduced dimensions (130 x 120 x 40 cm) are suitable for the postlarvae period of sturgeons. The technological water was supplied mainly from the Lake Horia but when needed it was mixed with water from a deep well in a reservoir. The water flow through the rearing units was 6.7 l / min, ensuring a total water volume exchange in about 45 minutes. Before reaching the tanks water was mechanically filtrated through a Crystal filter, filled with 1-1.5 mm silicium particles, with a filtration capacity of 20 m^3. The water was also biologically controlled by an U.V filter adapted to the used flow.

The experiment started with a total of 6000 post larvae (500 specimens / rearing unit) with a mean individual weight (± SD) of 0.35 ± 0.02 g and an mean total length ranging between 3-5 cm. Throughout rearing period nine sortings was carried out. Each period between sortings was considered a distinct development stage characterized also by different feeding scheme as follows:

- Stage 1: 19.06-10.07.2012 (22 days), mix Artemia frozen salt and Nutra Pro 3, ad libidum;
- Stage 2: 10.07-17.07.2012 (7 days), 2 tables mix and 4 tables Nutra Pro 3, ad libidum;
- Stage 3: 17.07-24.07.2012 (7 days), Nutra Pro 3 and Nutra Pro 2 in proportion of 3: 1, 7% of biomass;
- Stage 4: 24.07-30.07.2012 (7 days), Nutra Pro 3 and Nutra Pro 2 in proportion of 1: 1, 7% of biomass;
- Stage 5: 30.07-14.08.2012 (15 days), Nutra Pro 2, 5% of biomass;
- Stage 6: 14.08-21.08.2012 (7 days), Nutra Pro 2 and Nutra Pro 0 in proportion of 1: 1, 4% of biomass;
- Stage 7: 21.08-29.08.2012 (8 days), Nutra Pro 0, 4% of biomass;
- Stage 8: 29.08-05.09.2012 (7 days), Nutra Pro and Nutra Pro MPT in proportion of 1: 1, 4% of biomass;
- Stage 9: 05.09.-18.09.2012 (13 days), Nutra Pro MPT, 4% of biomass.

Table 1 Initial data of the experiment

Experimental variant	V_1	V_2	V_3	V_4	V_5	V_6
Concentration g / kg of feed	10	20	30	40	50	0
Concentration (CFU / kg feed *)	$1.28x10^{13}$	$2.56 x10^{13}$	$3.84x10^{13}$	$5.12x10^{13}$	$6.4x10^{13}$	0
Average weight (± SD) (g)	0.34±0.05	0.32±0.03	0.35±0.03	0.38±0.02	0.37±0.03	0.35±0.02
Initial density (kg/m^3)	0.57	0.53	0.58	0.63	0.61	0.58

* colony-forming units

Table 2 Biochemical composition of feed

Composition / granulation	Nutra Pro 3	Nutra Pro 2	Nutra Pro 0	Nutra Pro MPT
Granulation (mm)	0.5-0.7	0.7-1.1	1.1-1.7	1.7
Raw protein (%)	55	54	54	50
Raw fat (%)	16	18	18	20
Digestible energy (MJ/kg)	18.8	19.4	19.4	19.7

The larvae feeding was performed daily, initially only manually and in the last four stages by automated feeders (brand AGK, Germany), 4-6 times/ day. The nutritional information regarding the administered feeds is shown in table 2. Regarding the biochemical composition of the mixture of frozen Artemiasalina feed, it was determined in the laboratory and it was: 55.8% protein, 11.2% lipids, 6.9% carbohydrates and 5.9% ash.

Water quality was daily monitoredand the critical parameters(pH, temperature, O_2, NH_4 –N) were within tolerable requirements of the sturgeons and implicitly for the colonization of the administered probiotic strains. The BetaPlus® Ultra probiotic was purchased from Biochem and represents a commercial probiotic in the form of brown powder whose active component is an equal mixture of dry sporogenic bacteria belonging to the genus Bacillus: *Bacillus licheniformis* (DSM 5749) and *Bacillus subtilis* (DSM 5750) 1: 1 ratio and betaine (nitrogenous substance with attractive role).

The experiment was performed in duplicate 2 x 5 concentrations of probiotic BetaPlus® Ultra. The concentration of the commercial probiotic Beta Plus® Ultra was 1.28×10^{12} colony-forming units (CFU) / g probiotic, which means 6.4×10^{6} CFU *Bacillus licheniformis* and 6.4×10^{6} CFU *Bacillus subtilis*. The amount of probiotic administered in the food ranged from 10 to 50 g / kg feed in the first 5 experimental variants; the last variant was the control (without probiotic). The incorporation of the probiotic into the mixture of frozen *Artemiasalina* was done without any binder, but for comercial feeds a gelatin solution of 2% was used according to the protocol described by [2].

The BetaPlus® Ultra probiotic was chosen because of its beneficial properties: appetite enhancement, increased feed efficiency, enzyme production that increase food digestibility. Withal, bacteria of the genus Bacillus induced significant results in sturgeons ([6]);

The recommended doses of probiotics in aquaculture are within the range of 10^{6}-10^{10} CFU / g of feed ([12]). However, some authors ([10]) achieved considerable results in terms of growth performance using concentrations of 10^{11} CFU / g of feed.

Statistical analysis

All data analysis was performed at Excel and SPSS. Data were presented as mean ± SD (standard deviation). Data were analyzed by one-way analysis of variance (ANOVA), and significant differences between feeding groups were determined by Duncan's multiple-range post-hoc test. Statistical significance was accepted at $P < 0.05$.

RESULTS AND DISCUSSIONS

Growth performance and nutrient utilization are the two basic criteria that determine aquaculture production and profitability [10]. Food supplementation with probiotics is, according to some authors ([17],[3], [7], [4],[18], [9]), a means to reduce costs production because probiotics favor the growth and efficiency of feed use. At the end of the experimental period, the highest mean value for individual weight gain was recorded in variant V_2while the lowest mean individual weight was observed in V_6 variant (Fig. 1).

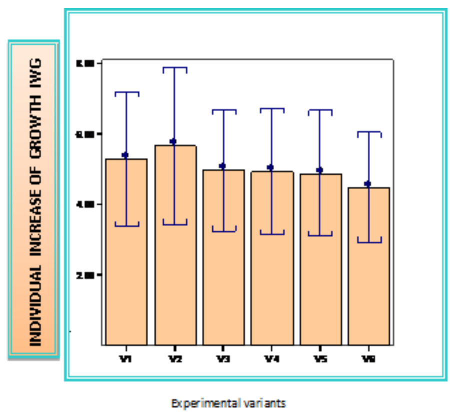

Fig. 1 The average of the individual weight gain (± SD.) on experimental variants

The values of the average individual weight gain (IWG) did not show significant ($p>0.05$) differences between the variants excepting the last experimental stage when the mean value of IWG was significantly higher in V2 comparing with other variants. Regarding the conversion factor it did not show statistical differences ($p>0.05$) between the experimental variants in any of the stage of the experiment.

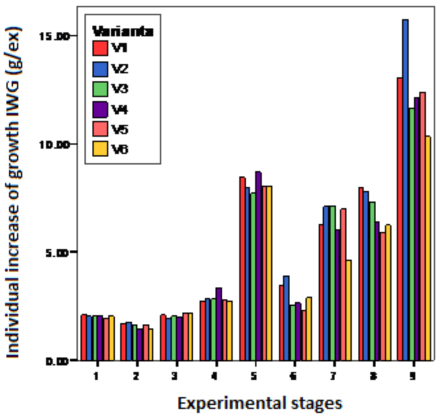

Fig. 2 The average of the individual growth increase (IWG) for the variants experienced in stages

From figure 3 it can be seen that the best (lowest) average value of the FCR was recorded in V_2 while the feeding efficiency was lower in variant V_6; however there were not registeredsignificant differences among variants, $p>0.05$. If we analyse FCR values for every experimental stage though, we can observed that the best valuesfor feed convertion ratio were recorded in the first experimental stages, while in the final stages the efficiency of the food conversion diminished (figure 4). A possible explanation for recording a higher FCR in the first two stages of the experiment is that in those periods natural food was administered in combination with comercial feed.

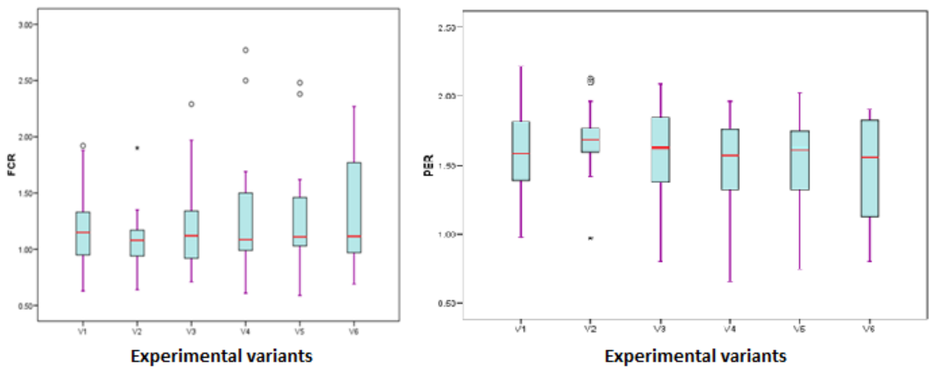

Fig. 3 FCR and PER variation (minimum, medium, max) on experimental variants

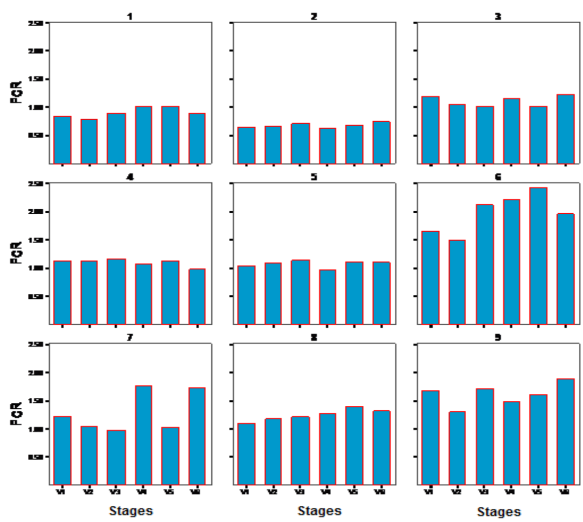

Fig. 4 FCR variation on variants and stages experiment

Regarding protein efficiency ratio (PER), the mean values from V_2 variant -1,70±0,26 (mean of all stages) were slightly higher, but notstatistically significant (p> 0.05), in the second experimental variant compared to the other variants (V_1 -1,58±0,31; V_3-1,59±0,37;V_4 -1,35±0,36;V_5-1,53±0,35; V_6 -1,45±0,38). Similar results were also reported by [10] in the case of Labeorohita (4.5g), which for high concentrations of probiotic (10^{12} CFU / kg) recorded values of the protein efficiency factor between 0.8-1.52.

Regarding the evolution of PER during the experimental stages we can say that this indicates a better protein retention in the first stages. The relationship between the protein level of the food and PER is a directly proportional; decrease of feedprotein induced a lower protein efficiency factor.

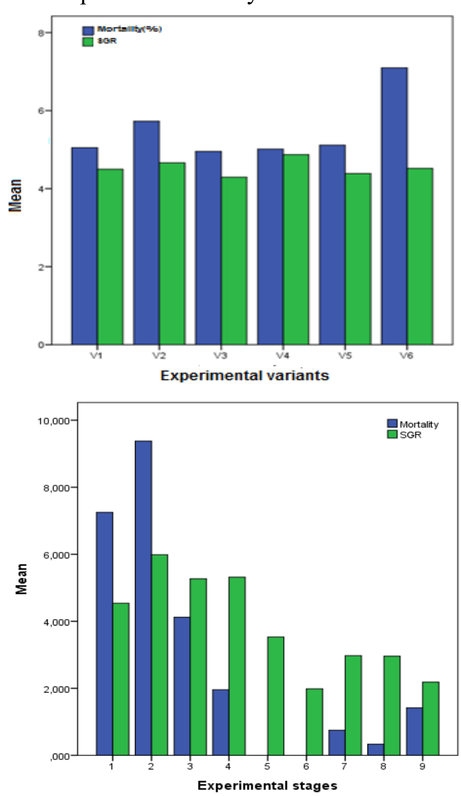

Fig. 5 Mortality and SGR mean variation for different variantsduring experimental stages

The specific growth rate recorded lower values in the case of the control variant compared to the other experimental variants; however no significant difference among variants were observed (p>0.05).

Regarding the evolution of the specific growth rate in time, over the experimental stages, it can be seen from the figure 5 the same tendency as the other indicators analyzed so far of better growth in the first stages compared to the last ones.

Mortality recorded during the experimental period did not vary significantly (p>0.05) between the five experimental variants but was significantly (p<0.05) higher in the control variant (V_6) However, of all the experimental variants, control variant V_6 recorded the highest mortality during all stages, the second stage having the highest mortality due to critical period that larvae have to endure during switching from mixed feed to commercial feed only. Therefore, in the case of the present experiment we can say that the administration of probiotics favors survival during larval and post larval period.

As with other articles regarding the effect of probiotics on sturgeons ([6], [16]) it can be said that even in this experiment, the increase of the concentration of probiotics is not directly proportional to the survival.

CONCLUSIONS

The biomass growth dynamics followed the same trend as technological performance indicators, the best values being registered in the second experimental variant (2.56 x10^{13} CFU / kg feed). The lowest values in terms of growth performance indicators were recorded in variant V_6, control variant, where no probiotic was administered. The sturgeon larvae grew better in the early stages of the experiment compared to the final stages, given the more rapid growth rate of the earlier lifestages. The IWG and SGR did not show significant differences between experimental variants, recording however slightlyhigher values in V_2. The control variant differed significantly (p>0.05) from the experimental variants from both perspective of growth performance and survival rate.

Therefore, according to this experiment, probiotics administered to sturgeon larvae and juveniles significantly improve survival rate.

Likewise, it demonstrated that concentration of 2.56x10^{13} CFU / kg of the commercial probiotic BetaPlus® Ultra represent a possible optimal value for promoting growth, feeding efficiency and retention of proteins of the earlier stages of sturgeon juveniles.However, in order to have a more clear perspective, more tests, including challenge tests, should be performed.

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