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EFFECTS OF DIETARY VITAMIN E AND SAGE (*SALVIA OFFICINALIS* L.) ON GROWTH PERFORMANCE OF KOI CARP (*CYPRINUS CARPIO* L., 1758)

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Abstract

The present research aims to evaluate the impact of both dietary vitamin E and dietary sage supplementation on growth performance of koi carp fingerlings, reared in a partial recirculating aquaculture system. Koi carp specimens with an average individual biomass of 5.1 ± 0.4 g were equally distributed, as follows: V1 - diet supplemented with 1% vitamin E (V1), respectively, V2 - diet supplemented with 1% sage. A 5% BW (Body weight) daily feeding ratio was applied. During the 49 days trial, individual biomass measurements were performed at initial, after 8, 15, 25 and 37 days, as well as at the final stage of the experiment. The specific growth rate indicates a superior fish production at V2 (3.30%BW/day), compared to V1 (2.75%BW/day). Also, better FCR values are recorded at V2 (1.28), compared to V1 (1.62). The Protein efficiency ratio (PER) results indicate a better ability of V2 koi carp specimens to utilize proteins (1.39), compared to V1 specimens (1.10). Thus, dietary supplementation with 1% sage has a superior effect on growth performance of koi carp fingerlings, compared to 1% vitamin E dietary supplementation.

Key words: *koi carp, sage, vitamin E, dietary supplementation, growth performance.*

INTRODUCTION

Plants and their extracts are known to possess many bioactive components such as tannin, alkaloids, and essential oils which have both antimicrobial and antioxidant activities. These bioactive components exert their beneficial effects by manipulating the intestinal microflora and improving digestibility (Asheg et al., 2014). Plant-derivatives, or the so-called "phytogenics" and plant extracts "phytobiotics" which are mainly obtained from aromatic plants and their essential oils, have been greatly utilized to enhance the growth performance of fish (Abdel-Latif et al., 2020).
Certainly, the use of immunostimulants as functional additives is acknowledged to improve the non-specific defence mechanism in fish, so giving resistance to infections (Cristea et al., 2012). Among the natural phytobiotic products with the potential of growth-promoting activity is sage (*Salvia officinalis*). The sage is one of the largest genera of the family *Lamiaceae*. It is widely distributed in the temperate, subtropical, and tropical regions all over the world (Sharifi-Rad et al., 2018).
The vitamin E plays a significant role in the health and growth of fish by improving their immune responses as well as their resistance to stress and disease (Naderi et al., 2017).
According to other authors (Cristea et al., 2012), various types of feed additives enhance the digestibility and/or utilization efficiency of nutrients, including exogenous enzymes, stimulators of enzyme secretion, compounds that aid in the digestive process by improving absorption, mobilization and transport of nutrients, feeding stimulants that reduce feed/nutrient waste, prebiotics, probiotics, and botanical extracts that modulate gut microflora.
The aim of the present study is to evaluate the impact of both dietary vitamin E and dietary sage supplementation on growth performance dynamics of koi carp fingerlings, reared in a partial recirculating aquaculture system.

MATERIALS AND METHODS

Experimental design

Two experimental diets were used: V1 - diet supplemented with 1% vitamin E (V1), respectively V2 - diet supplemented with 1%

sage. The feed proximate analysis revealed a value of 56% protein and 15% lipids.

The introduction of vitamin E and sage into fish feed was made by applying the following described protocol:

• a mixture of phytobiotic/vitamin E and gelatin (2% concentration) was made

• the mixture was sprayed uniform over the feed surface, while assuring a continuous shaking,

• the final mixture is dried at 25°C, for 2 hours and administrated to fish biomass as described in the feed administration protocol.

The experiment was conducted at the MoRAS Research Center - Food Science and Engineering Faculty, "Dunarea de Jos" University of Galati, during a period of 49 days. After acclimation, fish were stocked in a partial recirculating system with consists in a series of rectangular rearing units and water conditioning unit (aeration unit, mechanical and biological filtration). In order to maintain the water quality parameters within an optimal range for koi carp growth, a daily exchange rate of 30% was applied. Fish were divided into 2 treatments (with replicates) and fed with the experimental diets, by applying a 5% BW daily feeding rate. Feed was administrated manually, 4 times per day, at 9:00, 11:00, 13:00 and 15 h, during a 7 weeks experimental period.

Biological material and water quality

The koi carp (*Cyprinus carpio* L.) fingerlings (5.1 ± 0.4 g), obtained through artificial reproduction in the spring of 2020 year were acclimated to experimental rearing conditions for 2 weeks, during which fish were fed with commercial diet (56% crude protein).

The water dissolved oxygen (DO) concentration, pH and temperature (°C) were measured daily, by using a HQ40d Portable Multi-Parameter.

Growth performance and feed utilization parameters

A number of four intermediary biometric and biomass measurements were made in order to upgrade the quantity of daily administrated feed. The analysed technological indicators were as follows (Petrea et al., 2019):

Individual biomass gain: IBG = (Bf)-(Bi)/ fish number [g/fish], with Bf – final fish biomass; Bi – initial fish biomass (1);

Relative growth rate: RGR = ((Bf- Bi)/t)/Bi [g/g/day], with Bf - final fish biomass; Bi – initial fish biomass, t - duration of the experiment (2);

Specific growth rate: SGR = 100 x (ln Bf - ln Bi)/t [% fish biomass/day], with Bf - final fish biomass, Bi – initial fish biomass, t - duration of the experiment (3);

Feed conversion ratio: FCR = F/IBG [kg feed intake/kg fish biomass gain], with F - feed intake, FBG – individual biomass gain (4);

Protein efficiency ratio: PER = BG/(F*CP/100) [kg/kg], with FBG - individual biomass gain, F - feed intake, CP - crude protein (5).

Variation coefficient: $CV_{w/L}$ = (Dev. St./Avg w/L) x 100 [%], with Dev. St. - standard deviation, $Avg_{w/L}$ - fish body weight/length

Data analysis

For statistical analysis was used the IBM SPSS Statistics 20 for Windows and statistical differences between treatments were tested using T test ($\alpha = 0.05$) after a normality test (Kolmogorov-Smirnov). Comparisons between variants were assessed using post-hoc Duncan test for multiple comparisons (ANOVA).

RESULTS AND DISCUSSIONS

Water quality

The water parameters were within the optimal range for koi carp growth, as mentioned by Boyd and Tucker (2012), at both experimental variants, as follows: temperature ranged between 19-23.5°C, the DO ranged between 5.2-7.5 mg/L at V_1, respectively 4.7-7.4 mg/L at V_2, and pH ranged between 6.9-8.1 upH at V_1, respectively 6.7 - 7.9 upH at V_2.

The pH and DO variation for V_1 and V_2, respectively, are presented in Figure 1 and Figure 2. Not significant differences ($p>0.05$) were recorded between both experimental variants in terms of DO, pH and temperature. However, the vitamin E diet experimental variant (V1) had registered better values related to technological water quality, compared to sage diet experimental variant (V2).

It is observed that the dissolved oxygen concentration and pH had a decrease in the last part of the experiment period, which indicates the limitations of the production system in terms of maintaining water quality in terms of increasing the amount of food administered.

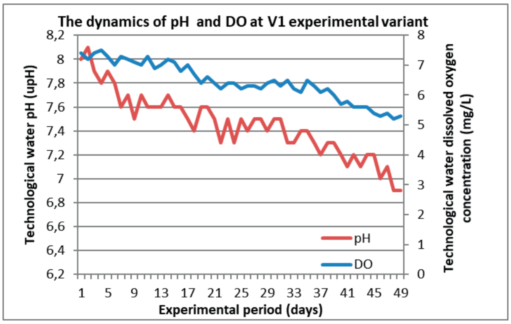

Figure 1. The dynamics of pH and DO for V_1

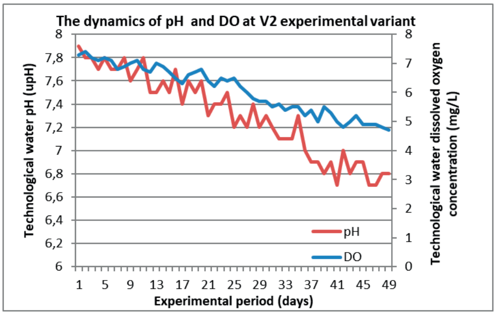

Figure 2. The dynamics of pH and DO at V_2 experimental variant

The effects of experimental diets on the growth performance indices and feed utilization parameters are presented in Table 1.

There were no significant differences between the fish specimens in terms of initial body weight (IBW), at the start of the feeding trial ($p>0.05$). The growth performance indicators (Table 1) reveled no mortalities during V_2 and 10% in varianta V_1, therefore confirming the good results registered in terms of water quality. The average specific growth rate indicates a superior fish production at V_2 (3.30% BW/day), compared to V_1 (2.75% BW/day) experimental variant (Table 1).

However, from the perspective of feeding strategy efficiency, the average food conversion ratio (FCR) indicates better values for V_2 experimental variant (1.28 g feed/g biomass gain), compared to V_1 (1.62 g feed/g biomass gain) (Table 1). The average protein efficiency ratio (PER) registered higher values at V_2 experimental variant (1.39), compared to V_1 (1.10), revealing the ability of fish organism to utilize proteins, which positively affects growth rate (Table 1).

Table 1. Growth performance indicators for each of the experimental variants

Nr. crt.	Indicator	Period	V_1 (vit. E)	V_2 (Sage)
1.	Experimental period (days)	Initial - INT.1	8	8
		INT.1-INT.2	7	7
		INT.2 - INT.3	10	10
		INT.3 - INT.4	12	12
		INT.4 - Final	12	12
		Initial - Final	49	49
2.	Survival (%)	Initial - Final	90	100
3.	Individual average biomass (g/fish)	INITIAL	5.1	5.2
		INT.1	6.7	6.9
		INT.2	8.2	8.8
		INT.3	10.7	12.3
		INT.4	14.3	17.6
		FINAL	19.6	26.1
4.	Individual average length (cm/fish)	INITIAL	7.0	7.2
		INT.1	7.6	8.1
		INT.2	8.3	8.9
		INT.3	9.4	10.3
		INT.4	10.1	11.1
		FINAL	11.6	13.6
5.	Individual biomass gain (g/ex)	Initial - INT.1	1.6	1.7
		INT.1-INT.2	1.6	1.9
		INT.2 - INT.3	2.4	3.5
		INT.3 - INT.4	3.6	5.3
		INT.4 - Final	5.3	8.4
		Initial - Final	14.5	20.9
6.	Relative growth rate - RGR (g/g/day)	Initial - INT.1	0.039	0.042
		INT.1-INT.2	0.034	0.04
		INT.2 - INT.3	0.030	0.039
		INT.3 - INT.4	0.029	0.036
		INT.4 - Final	0.031	0.04
		Initial - Final	0.015	0.016

Nr. crt.	Indicator	Period	V_1 (vit. E)	V_2 (Sage)
7.	Specific growth rate (%BW/day)	Initial - INT.1	3.36	3.63
		INT.1-INT.2	3.03	3.52
		INT.2 - INT.3	2.59	3.31
		INT.3 - INT.4	2.45	3.01
		INT.4 - Final	2.62	3.26
		Initial - Final	2.75	3.30
8.	Feed conversion ratio - FCR (g feed / g biomass gain)	Initial - INT.1	1.29	1.19
		INT.1-INT.2	1.48	1.25
		INT.2 - INT.3	1.69	1.28
		INT.3 - INT.4	1.75	1.38
		INT.4 - Final	1.62	1.26
		Initial - Final	1.62	1.28
9.	Protein efficiency ratio - PER (g/g)	Initial - INT.1	1.38	1.50
		INT.1-INT.2	1.20	1.43
		INT.2 - INT.3	1.06	1.40
		INT.3 - INT.4	1.02	1.30
		INT.4 - Final	1.10	1.42
		Initial - Final	1.10	1.39
10.	Feed protein (%)	Initial - Final	56	56
11.	Daily feeding ratio (% BW)	Initial - Final	5	5
12.	Weight variation coefficient - CVw (%)	FINAL	6.52	7.25
13.	Length variation coefficient - CVL (%)	FINAL	2.89	3.42

By analyzing the variation coefficients, it can be stated that both experimental variants had registered a high homogeneity degree in the first part of the experimental period (Figures 3 and 4).

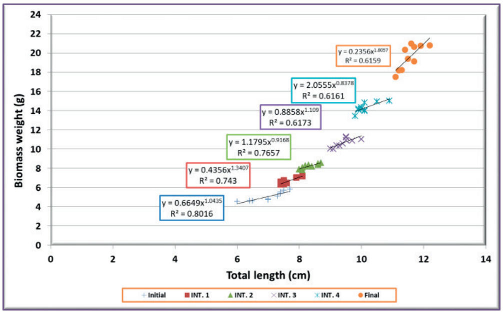

Figure 3. Total Length-Weight relation for V_1 biomass, during the experimental period

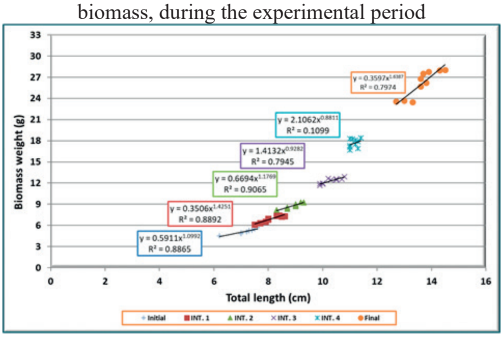

Figure 4. Total Length-Weight relation for V_2 biomass, during the experimental period

However, the V_1 experimental variant revealed a significant decrease on the homogeneity degree after the first 15 experimental days, compared to V_2 experimental variant, which continued to register high homogeneity until the last part of the experimental period (first 37 days) (Figures 2 and 3).

The condition status of biological material was evaluated by using the allometric condition factor F ($F = W/L^b$, where b = allometric exponent, experimentally determined).

The allometric exponent "b" has its values under three units at both variants, during the entire experimental stage (Figures 2 and 3), fact which reveals a faster growth in length rather than weight.

Also, the K condition factor registered lower average values at the end of the trial at V_1 (K = 0.236), compared to V_2 (K = 0.315).

Nowadays, there is an increasing interest in the usage of dietary phytobiotics as natural growth promotors and immunostimulants in practical fish diets to improve growth, health status, immune responses, and protection against bacterial diseases (Abdel-Latif et al., 2020).

Furthermore, phytoadditives appear to be reasonable alternative solutions to substitute synthetic antimicrobials used in aquaculture without any undesired effects upon recent related studies (Daood, 2011).

The present study confirms the beneficial effects of sage use, as observed by Salomon et al. (2020) who evaluated the growth response in juvenile gilthead sea bream (*Sparus aurata*) fed with a functional diet containing a medicinal plant leaf extract from sage (*Salvia*

officinalis) and lemon verbena (*Lippa citriodona*) and subliniated the beneficial effects of the dietary administration.
According to other authors (Cristea et al., 2012), sustainable aquaculture depends on perfectly balance between health and growth condition of fish. Thus, according to other study (Abd-El-Rhman, 2009), certain phytobiotics as propolis-ethanolic-extract were found to promotes the growth of intensive aquaculture fish species, as Nile tilapia. Therefore, the used of phytobiotics and vitamins promotes growth and therefore, maximize productivity and income, especially in intensive and semi-intensive production fish farms. However, a proper period for the administration must be identified and suitable phytobiotics should be selected since, some authors (Kono et al., 2000) reported experienced a reduction growth performance when green and ground tea extracts were added in fish diet.

CONCLUSIONS

The partial recirculating aquaculture system used in this present study managed to maintain water quality within the optimal range, even if a 5% BW feeding ratio was applied. However, if feeding ratio will increase, the water conditioning units must be upgraded, or water exchange rate must increase. The results presented in current research revealed that the supplementation with 1% sage has a superior effect on growth performance of koi carp fingerlings, compared to 1% vitamin E dietary supplementation. Also, the use of sage in fish diet can help define the concept of circular economy given that sage biomass could be obtained in an integrated aquaponic production system that uses nutrients provided by aquaculture effluents as main source for plant biomass growth.
However, further research is required for better understanding the mechanisms of improving fish health and productivity by dietary sage administration. Also, the optimal period for the administration of sage into koi carp diet must be identified in order to integrate this information into a long-time rearing technology.

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