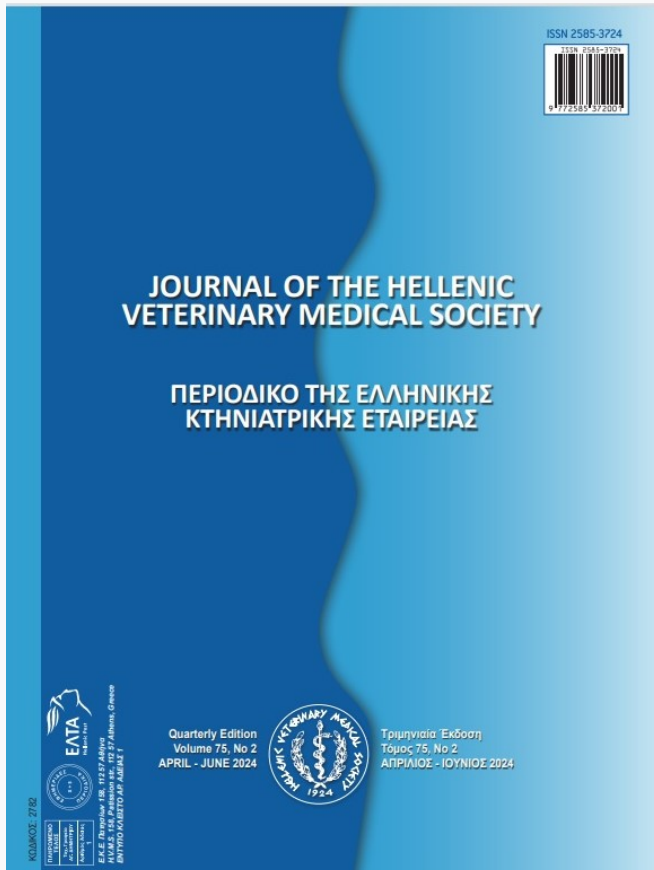


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## Effect of Growth Performance and Survival Rate of Goldfish (*Carassius auratus* L. 1758) Fed Garlic (*Allium sativum* L.) Supplemented Diets

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**ABSTRACT:** This research evaluated the effect of different garlic (*Allium sativum*) levels on the nutritional characteristics, growth, and survival rate of goldfish (*Carassius auratus*). Five different commercial experimental diets were prepared garlic was added at levels of 0 (Control), 5, 10, 15, and 20 g·kg<sup>-1</sup>. The experiment was carried out for 84 days. Diet 4 group, in which 20 g·kg<sup>-1</sup> garlic was added to the trial feed, showed the best growth performance compared to Diet 0 (Control) and other groups (Diet 1, Diet 2, Diet 3) ( $p < 0.05$ ). The best growth in parameters such as body weight gain, percent live weight increase, and specific growth rate was in the Diet 4 group ( $p < 0.05$ ). It was determined that the statistical difference between the groups for the condition factor and survival rate parameters was insignificant. The highest feed conversion rate was determined in the Diet 0 group with the lowest Diet 4 group ( $p < 0.05$ ). As a result, it has been determined that feeding goldfish with the addition of garlic up to 20 g·kg<sup>-1</sup> (2%) in its diet will have a positive effect on growth.

**Keywords:** Growth; *Carassius auratus*; Garlic; Goldfish; Feed utilization; Nutrition

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## INTRODUCTION

In aquaculture, feeding plays an important role in bringing products to early market size. This situation has become a rule in aquaculture over time. For this reason, aquaculture researchers continue to give a high priority to the search for new feed additives (Cho and Lee, 2012). Studies have reported that different extracts obtained from plants and plants improve animal performance (Akbari et al., 2016). One of them is garlic "*Allium sativum*".

It is a bulbous, hollow, upright-stemmed, two-year herbaceous plant from the Liliaceae family. It rarely binds seeds, so it can be produced with bulbs (tooth). The history of garlic, whose homeland is India, is as old as the history of civilization (Dikel, 2015). In 2021, global garlic production reached 48.718.240 tonnes, with China responsible for 20.513.385 tonnes and Türkiye producing 46.454 tonnes, as reported. (Anonymous, 2023). Therefore, in addition to being a medicinal plant that can be grown in many countries, garlic is a plant that has become widespread in aquaculture studies as a feed additive.

In addition to the use of garlic as an alternative to chemotherapeutic agents or antibiotics in aquaculture (Erguig et al., 2015), it is available in the literature that it positively affects the growth performance of fish and also has an effect on some parameters (Akbari et al., 2016; Chesti and Chauhan, 2018; Dadgar et al., 2019; Mahmoud et al., 2019; Setijaningsih et al., 2021). Also, they are often used for the cure of several animal diseases (Shalaby et al., 2006; Megbowon et al., 2013). It has the ability to enhance the boost of the immune system. Additionally, it can assist in reducing the adverse effects of pathogens, especially fungi and bacteria (Fazlolahzadeh et al., 2011; Nwabueze, 2012; Setijaningsih et al., 2021)

The rapid growth performance contributes to the early market introduction of fish. Therefore, increasing

the growth performance of goldfish with feed additives can accelerate the market introduction process. Goldfish play a significant role in the global trade and cultivation of ornamental fish, whose significance is growing daily. The total value of all goldfish varieties traded globally has surpassed \$1 billion a year in more than 125 nations. (Anonymous, 2016; Arslan and Özbaş, 2020).

In this study, to accelerate the growth performance and increase the survival rate of goldfish, which have an important share in the global ornamental fish trade, and to bring them to early market size, garlic was used as a feed additive in goldfish feeds and fed.

## MATERIALS AND METHODS

### Preparation of feeds

The diets to be used in the experiment were five different commercial experimental feeds, crude protein (44%), crude fat (7%), and energy (14.90 MJ·kg<sup>-1</sup> digestible energy) being equal were prepared by ArtAkua Fish Food. The different levels of garlic 0 (Control), 5, 10, 15, and 20 g kg<sup>-1</sup> (0.5%, 1%, 1.5%, 2%) were added in powder form to the commercial diets (Rızaei et al., 2022). The nutritional content is shown in Table 1. The rules of the AOAC (Association of Official Analytical Chemists) were used for the chemical analysis of experiment feeds (Anonymous, 1995). The samples representing each group were weighed before being placed in moisture containers for the determination of a matter. Second, they spent about 12 hours in the oven, which was set to 105±2 °C, until they achieved a constant weight, after which they were placed in the desiccator to cool to room temperature. The Kjeldahl technique was then used to analyze the crude protein. Following that, ether extraction using the Soxhlet extraction system was used to ascertain the results of the crude lipid analysis. After burning the samples at 550°C in the ash furnace, the ash analysis was completed. (Anonymous, 1995).

**Table 1.** The proximate compound of diets used in the garlic feeding experiment on *C. auratus* (% wet weight)

Component	Experimental diets %				
	Diet 0 (Control)	Diet 1	Diet 2	Diet 3	Diet 4
Crude protein	42.7±3.64	42.7±3.64	42.7±3.64	42.7±3.64	42.7±3.64
Crude lipid	6.6±0.12	6.6±0.12	6.6±0.12	6.6±0.12	6.6±0.12
Crude cellulose	1.9±0.13	1.9±0.13	1.9±0.13	1.9±0.13	1.9±0.13
Crude ash	6.6±0.22	6.6±0.22	6.6±0.22	6.6±0.22	6.6±0.22
Moisture	5.91±0.24	5.91±0.24	5.91±0.24	5.91±0.24	5.91±0.24
Dietary energy [MJ·kg <sup>-1</sup> ]	14.95	14.87	14.92	14.93	14.90

The values shown here, represent the mean value ± standard deviation of three replicate analyses.; garlic contents in diets [g·kg<sup>-1</sup>]: Diet 0 = 0, Diet 1 = 5, Diet 2 = 10, Diet 3 = 15, and Diet 4 = 20

## Experimental conditions

This research was conducted in the research laboratory of the 'Faculty of Fisheries, Akdeniz University'. Experimental fish were acquired from the Kepez Unit of the "Ministry of Agriculture and Forestry Mediterranean Fisheries Research Production and Training Institute" (Antalya/ Türkiye). The fish were fed for two weeks in a 269 L (110 × 35 × 70 cm) glass tank, adapted to the new conditions, and no signs of disease were detected. At the end of the two weeks, the experiment was conducted in 15 glass tanks measuring 65 L (70 × 30 × 40 cm) and included three replications. The fish were distributed at random into 15 glass tanks, each holding 30 goldfish, with a mean live weight of  $3.90 \pm 0.03$  g and a total length of  $5.86 \pm 0.04$  cm. Fluorescent lighting was used during the experiment for 10 hours each day.

All groups were hand-fed until they were satiated twice a day (08:00 h and 17:00 h) for 84 days (Hasaan et al. 2013; Gümüş et al., 2016; Arslan and Özbaş, 2020; Monge-Ortiz et al., 2020).

Each glass tank water was aerated with by air stone using a central air pump and, was heated with a thermostat heater at 22 °C. Some water parameters (pH, temperature, and dissolved oxygen) were measured daily. During the experiment, the pH level was measured as  $7.58 \pm 0.021$ . The dissolved oxygen concentration was also measured as  $5.25 \pm 0.041$  mg·L<sup>-1</sup>. After the last feeding of the day, the fish feces and the unused feed accumulated on the bottom of the glass tank were removed by siphoning. Then the glass tank was added approximately 1/3 of the water was completed. Fish were measured over 21-day intervals in this experiment.

Before every measurement, the fish were anesthetized using clove oil (v/v, 1/20) to decrease the stress on the fish (Gümüş et al., 2016).

## Growth parameters

At the end of the experiment, survival, feed uti-

lization, and growth parameters such as weight gain (WG), condition factor (CF) (Le Cren, 1951), feed conversion ratio (FCR) (Halver and Hardy, 1982), specific growth rate (SGR) (Brown, 1957), and survival rate (SR) (Çelikkale, 1994) were calculated as follows (Gümüş et al., 2016):

$$WG = \text{final weight (g)} - \text{initial weight (g)}$$

$$WG (\%) = (\text{final weight} - \text{initial fish weight}) / \text{initial weight} \times 100$$

$$SGR (\%) = [(\log [\text{final weight}] - \log [\text{initial weight}]) / t (\text{time interval in days})] \times 100$$

$$FCR = \text{Feed intake (g)} / WG$$

$$\text{Condition Factor (CF)} = 100 \times [(\text{final fish weight (g)} / \text{final fish length (cm)})^3];$$

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

## Statistical analysis

Using the package application SPSS 20.0 (SPSS INC. Chicago, IL, USA), the experiment's results were statistically evaluated. To ascertain the impact of different garlic ratios on weight and length, variance analysis (ANOVA) was conducted after applying variance homogeneity tests to all data. Duncan's multiple comparison tests were applied to see differences in weight, height, condition factor (CF), feed conversion ratio (FCR), and specific growth rate (SGR) between the groups.

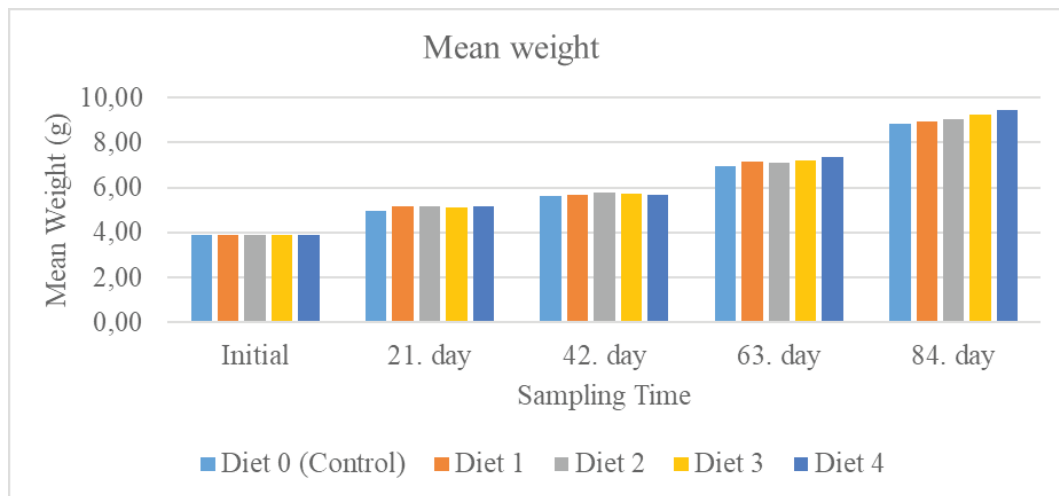
## RESULTS AND DISCUSSION

The results obtained from the experimental groups in the study conducted with garlic added to the experimental feeds at the rates of 0, 5, 10, 15, 20 g·kg<sup>-1</sup> (Diet 0, Diet 1, Diet 2, Diet and Diet 4) were given in Table 2.

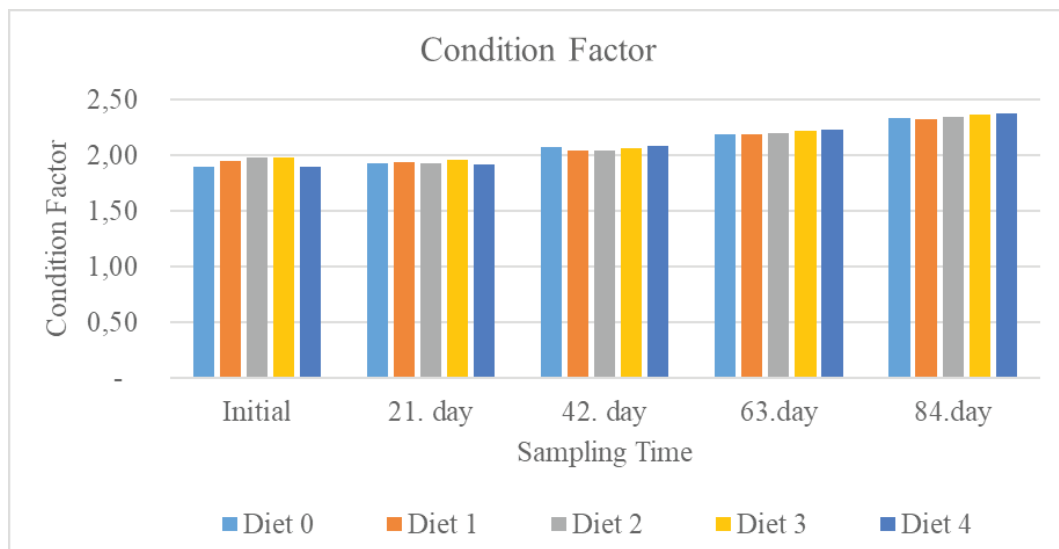
**Table 2.** The experimental fish's growth parameters, survival rate, and feed utilization (Mean ± SD) (n=3)

Parameters	The Experiment Groups				
	Diet 0 (Control)	Diet 1	Diet 2	Diet 3	Diet 4
IW <sup>1</sup> (g)	3.91±0.02	3.91±0.02	3.91±0.02	3.91±0.02	3.91±0.02
FW <sup>2</sup> (g)	8.84±0.23 <sup>c</sup>	8.92±0.18 <sup>bc</sup>	9.05±0.18 <sup>bc</sup>	9.26±0.13 <sup>ab</sup>	9.45±0.19 <sup>a</sup>
WG <sup>3</sup>	4.94±0.23 <sup>c</sup>	5.01±0.16 <sup>c</sup>	5.13±0.17 <sup>bc</sup>	5.35±0.12 <sup>ab</sup>	5.54±0.18 <sup>a</sup>
WG <sup>4</sup> (%)	126.41±5.84 <sup>c</sup>	127.92±3.60 <sup>c</sup>	131.09±4.45 <sup>bc</sup>	136.81±2.86 <sup>ab</sup>	141.54±4.65 <sup>a</sup>
SGR <sup>5</sup> (%·day <sup>-1</sup> )	0.97±0.03 <sup>c</sup>	0.98±0.02 <sup>c</sup>	1.00±0.02 <sup>bc</sup>	1.03±0.01 <sup>ab</sup>	1.05±0.02 <sup>a</sup>
FCR <sup>6</sup>	1.67±0.04 <sup>a</sup>	1.63±0.07 <sup>ab</sup>	1.60±0.06 <sup>abc</sup>	1.54±0.07 <sup>bc</sup>	1.51±0.05 <sup>c</sup>
CF <sup>7</sup>	2.34±0.12	2.33±0.10	2.35±0.13	2.37±0.12	2.37±0.07
SR <sup>8</sup> (%)	100	100	100	100	100

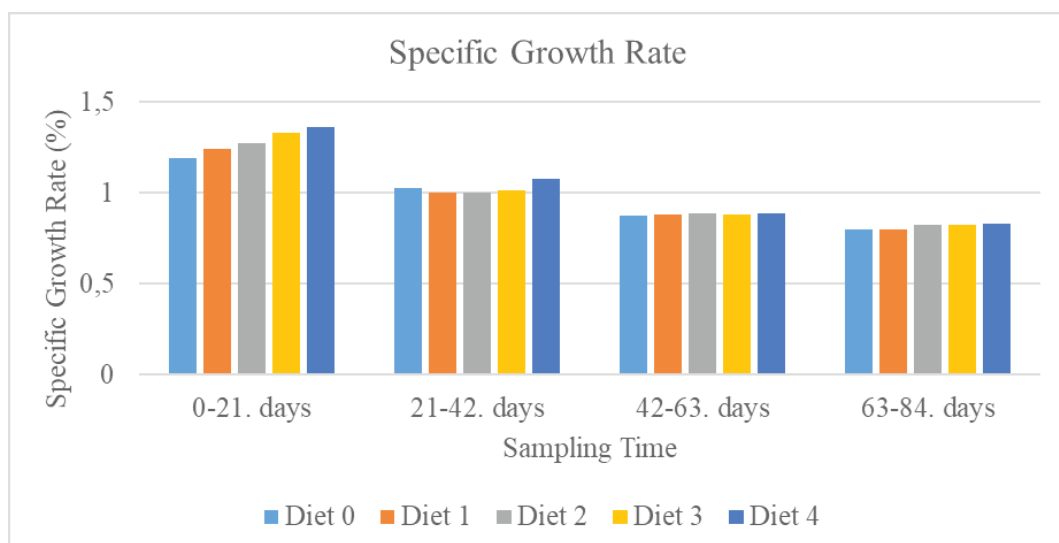
<sup>1</sup>IW: Initial weight, <sup>2</sup>FW: Final weight, <sup>3</sup>WG: Weight gain, <sup>4</sup>WG: Weight gain (%), <sup>5</sup>SGR: Specific growth rate, <sup>6</sup>FCR: Feed conversion rate, <sup>7</sup>CF: Condition factor, <sup>8</sup>SR: Survival rate, garlic contents in diets [g·kg<sup>-1</sup>]: Diet 0 = 0 g·kg<sup>-1</sup>, Diet 1 = 5 g·kg<sup>-1</sup>, Diet 2 = 10 g·kg<sup>-1</sup>, Diet 3 = 15 g·kg<sup>-1</sup>, and Diet 4 gr = 20 g·kg<sup>-1</sup>



**Figure 1.** Growth values by weight of *Carassius auratus* used garlic-supplemented diets feeding experiment



**Figure 2.** Condition factor values of *Carassius auratus* used garlic-supplemented diets feeding experiment



**Figure 3.** Specific growth rate values of *Carassius auratus* used garlic-supplemented diets feeding experiment

It was determined that statistically significant differences between the weight gain, weight gain (%), final weight, feed conversion rate, and specific growth rate values of the fish in the experiment groups ( $p < 0.05$ ) (Figure 1,4). But it was observed no significant statistical differences between the condition factor of the experimental groups in this study (Figure 2).

With the statistical analysis made, the similarities between the groups were shown in Table 2 with letters. An increase in weight values was observed as the garlic level increased. The group fed the diet containing 20 g kg<sup>-1</sup> of garlic (Diet 4) was determined to be the most effective in terms of weight gain (Figure 1).

It was determined that the highest final weight value with 9.45±0.19 in the Diet 4 group and was determined that the lowest value with 8.84±0.23 in Diet 0 (control). The highest value for the weight gain (%), weight gain, feed conversion rate, and specific growth rate was determined in the Diet 4 group. The lowest value for the weight gain (%), weight gain, feed conversion rate, and specific growth rate was determined in Diet 0 (control) and Diet 1. Hence, statistically significant differences were determined between the values of the fish in the experimental groups ( $p < 0.05$ ). The rate of fish in all experimental groups surviving is one hundred percent.

In this study, it was determined that garlic has a positive effect on growth parameters, especially on live weight gain in goldfish. As the garlic level increased, an increase in the weight gain, weight gain (%), final weight, feed conversion rate, and specific growth rate was observed (Figure 1, 3).

It was determined that it can be used in goldfish feed up to 20 g·kg<sup>-1</sup> (2%) within the scope of this experiment for 84 days. Similar results with this study were found by Akbary et al. (2016) reported that it has a positive effect on *Mugil cephalus* and can be used up to 0.3%. Akbary et al. (2016) reported that 0.3% garlic extract in the diet increased final weight and length, body weight increase, specific growth rate, protein production ratio, and protein efficiency ratio in larvae. In the present study, the statistical difference between the groups was significant for the final weight, weight gain, and specific growth rate parameters ( $p < 0.05$ ). The highest final weight was determined in the Diet 4 group (9.45±0.19) with the lowest Diet 0 (Control) group ( $p < 0.05$ ). Maniat et al. (2014) reported the best final weight, weight gain rate, food conservation ratio, and specific growth rate were

observed in the fish fed 10 g·kg<sup>-1</sup> garlic powder in the diet for Benni fish (*Mesopotamichthys sharpeyi*). It was concluded that 10 g·kg<sup>-1</sup> garlic powder had a good effect on the growth performance and body composition of Benni fish (Maniat et al., 2014). The present research determined that garlic can be used in goldfish feed up to 20 g·kg<sup>-1</sup>.

It was determined that like these studies in *Cyprinus carpio haematopterus* (Chesti and Chauhan, 2018) in Ecotype Cichlid (Megbowon et al., 2013), in *Acipenser ruthenus* (Dong-Hoon et al., 2012) had also been reported. Samson, (2019) reported that adding garlic powder at a rate of 1-1.5% to the diet of red tilapia (*Oreochromis sp.*) increased feed utilization and fish survival rate and the addition of garlic to the diet may be used as an alternative to synthetic or chemical supplements to promote fish growth, feed utilization, and survival rate. In this study, it was determined that can be used in goldfish (*C. auratus*) feed up to 20 g·kg<sup>-1</sup> (2%).

Samson, (2019) determined that the highest feed conversion rate was control (1.92 ± 0.24) and 2% (1.81 ± 0.05) but we determined that just Diet 0 (control) (1.67±0.04). Abdel-Hakim et al. (2010) reported that, in order to improve growth performance and feed utilization, also to lower the costs of production of one kg/gain in weight, supplementing growing Nile tilapia diets with dried garlic at 5g·kg<sup>-1</sup> (0.5%) level.

On the other hand, the results of the presently reported study are not consistent with the results of studies conducted on *Labeo rohita* fingerlings in Sahu et al. (2007), where they had reported feeding effects of garlic on feed conversion ratio and specific growth rate values of the treated group not significantly different from each other. However, we determined statistically significant differences between the specific growth rate and feed conversion rate values of the fish in the experiment groups ( $p < 0.05$ ). We think that this situation may be caused by species differences. Pashaki et al. (2018) observed no significant effects on the growth parameters of *Cyprinus carpio* after adding different levels of garlic to diets. But they reported garlic extract at 5 g·kg<sup>-1</sup> supplemented diet can improve some parameters of the growth, blood, and immune of Common carp fingerling in these study result. Moreover, Thanikachalam et al. (2010) and Nobahar et al. (2014) also observed no significant effects on the growth parameters of *Clarias gariepinus* and *Huso huso* after adding different levels of garlic to their diets. However, it was determined that

statistically significant differences between the final weight, weight gain, weight gain (%), feed conversion rate, and specific growth rate values of the fish in the experiment groups ( $p < 0.05$ ).

## CONCLUSION

The results acquired in this research showed that garlic meal as a natural feed additive represents an alternative additive as a growth promoter. When the level of garlic added to the diet increased, it was determined that there was an increase in some parameters of fish. In the experiment, garlic added to fish feed at a rate of  $20 \text{ g} \cdot \text{kg}^{-1}$  (2%) had a positive effect on growth performance and feed conversion ratio data of goldfish. Therefore, supplemental dietary garlic in the fish diet at  $20 \text{ g} \cdot \text{kg}^{-1}$  (2%) concentration will be advantageous for use in aquaculture to develop the feed performances and growth of goldfish. The effects of herbal additives on animal nutrition continue to be determined by many researchers. We think that determining the positive aspects of alternative

herbal feed additives and expanding their use can provide high-quality and maximum products in the field of aquaculture in a short time. Future research with different levels of garlic-added feeds should focus on different parameters such as carcass quality, hematological, and serum parameters.

## ACKNOWLEDGMENTS

The authors would like to thank the cooperation provided by Nektar Yem Co. Ltd. (ArtAkua Fish Food).

## CONFLICT OF INTEREST

None declared

## ETHICAL STATEMENT

The study was conducted with the approval of the Ethics Committee of the 'Faculty of Fisheries, Akdeniz University' (AÜ-SÜFHADYEK). Approval Number: 2018.12.05 - (27.12.2018/009)

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