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Effect of boric acid in rainbow trout (*Oncorhynchus mykiss*) growth performance

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ABSTRACT

The effects of different levels of boron supplementation to feed on the growth parameters of rainbow trout (*Oncorhynchus mykiss*) were investigated for the first time using four diet variations (0.01%, 0.05%, 0.10%, and 0.20% of boron in feed) in addition to a control diet (without boron). After 120 days of feeding, the final weight (FW), weight gain, condition factor, feed intake, feed conversion rates, specific growth rate, protein efficiency ratio, survival rates, economic conversion rate, and economic profit index were calculated for each treatment and compared with the control as growth parameters. The initial weight was 17.89 ± 1.12 g; at the end of feeding, the FWs were determined as 89.91 ± 2.11 g for the control and 90.56 ± 1.34 g, 99.21 ± 3.76 g, 88.14 ± 2.43 g, and 78.74 ± 1.47 g for 0.01%, 0.05%, 0.10%, and 0.20% of boron feed treatments, respectively. The results of this study show that feeding rainbow trout with 0.05% boron-supplemented feed stimulates the growth parameters, but 0.20% of boron supplementation inhibits the growth of rainbow trout.

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Boron supplementation; rainbow trout; growth performance; feed efficiency; specific growth rate

Introduction

As one of the fastest developing industries in the world, aquaculture has increased its production to cover human protein need. This increase induces new requirements for factors such as aquaculture feed. The feed in aquaculture mainly depends on fish meal and fish oil as the main protein sources, and its consumption has been exponentially increasing in the last decades. Accordingly, alternative proteins and some additives, which improve the efficiency of feed intake, have been studied in the feed industry (Tacon and Jackson 1985; Hardy and Tacon 2002). In particular, the limit of protein source encourages the use of additives in feed rations prepared for fish.

Boron as a trace element generally occurs in sedimentary rocks, soils, coal, seawater, etc. in different forms such as borates, boric acid, and boric oxide (Samman et al. 1998). Turkey, the United States, South America, Russian Federation, and China have commercially important deposits of boron-containing minerals (Woods 1994). Serving at cross purposes, boron has been used many different industries such as glass fibres, ceramics, detergents, fertilizers, wood preservatives, and cosmetics (Scialli et al. 2010). Involved in metabolic, nutritional, and physiological processes, boron is a necessary dietary component for animals (Nielsen 1997). Boron functionally occurs in mineral metabolism, immune response, and the endocrine system in animals. In addition, it participates in bone growth (Hunt 1994; Kabu and Akosman 2013). However, the boron functions in animals have not been thoroughly described. Moreover, higher concentrations than the determined level of boron for certain animal species can be toxic (Goldbach et al. 2007).

Among the studies on different animal species such as rat (*Mus domesticus*) and frog (*Xenopus laevis*) (Fort et al. 1999; Bustos-Obregón and Olivares 2012), there are studies about the boron functions and availability in fish. Emiroğlu et al. (2010) reported that the average boron concentrations in the muscles of *Leuciscus cephalus* varied from 7.34 ± 0.44 to 53.73 ± 21.25 boron mg kg^{-1} dry weight) in sampling stations depending on the distance to the active area of the borate industry. They also showed the differences in accumulation concentrations of boron in the muscle, liver, gill, and head. The necessity of boron in the embryonic development of zebrafish (*Danio rerio*) has been determined. A supplementation of boron concentration of 45 µmol L^{-1} to the water improved the hatching rates of fertilized eggs in comparison to the boron concentration of 0.1 µmol L^{-1} in water (Rowe and Eckhert 1999). Loewengart (2001) reported that the boron concentration in water should be >0.03 mg boron L^{-1} to avoid boron deficiency and support the development of the embryonic and larval stages of rainbow trout. Furthermore, Adhikari and Mohanty (2012) reported that 8.0 mg boron L^{-1} reduced the survival rates, growth, and feed intake of Indian major carp (*Cirrhinus mrigala*). The exposure time and weight of each individual contribute to the effect of boron on fish. As the only currently available feeding experimental study, Ardó et al. (2008) determined that 0.05% boron in feed combination with Chinese herbs (*Astragalus membranaceus* and *Lonicera japonica*) enhanced the immune response of Nile tilapia (*Oreochromis niloticus*) in comparison to that without boron. However, no data were available at the time on the growth parameters of rainbow trout affected by different boron concentrations.

With the motivation of boron functions in the metabolism and growth of animals, the present study aims to investigate

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the effect of boron-supplemented feed at the rates of 0.00%, 0.01%, 0.05%, 0.10%, and 0.20% on the growth parameters of rainbow trout (*Oncorhynchus mykiss*) and compare it to the control group without boron supplementation.

Materials and methods

Experimental fish and handling

This study was approved by the Animal Experiments Local Committee in Adana, Turkey (no. 3-2/2016). The feeding activities were performed for 120 days in a commercial rainbow trout fish (*O. mykiss*) farm in Pozantı-Adana, Turkey. In total, 1575 rainbow trout were used in the study. Each treatment and control consisted of three equal replicates (105 fish per replicate); the fish in each replicate were randomly assigned to 15 net cages of 1 m^3 each (10 mm mesh size). At the beginning of the experiment, the average live weight of all experimental fish was 17.89 ± 1.12 g. The water temperature and oxygen content of the pools in the experiment were checked twice daily using an OxyGuard oxygen meter (OxyGuard, Birkerød, Denmark). The water temperature was 8°C, and the oxygen contents were 8.9–13.2 mg/L during the experimental period.

Diet preparation and experimental design

During the experiment and for 2 weeks prior to the beginning of the experiment, the fish were fed a commercial trout feed produced by Skretting (Stavanger, Norway), of which nutritional values are listed in Table 1. The concentration of boron in the initial feed and the water was determined by the curcumin method spectrophotometrically (Dyrssen et al. 1972). A 5 g feed was crushed and extracted with 10 mL of distilled water boiling for 15 minutes under reflux and was filtered as quickly as possible (Porter et al. 1981). The absorbance of the boric acid in the obtained solutions and the water samples with three replicates based on complexing with curcumin was measured at 540 nm. The minimum detection limits for boron were 0.02 mg/g in the feed and 0.1 mg/L in the water (Grinstead and Snider 1967).

Four different boron level diets (0.01%, 0.05%, 0.10%, and 0.20% of boron in feed) were prepared by supplementing boron to the feed, and a boron-free diet was used as the control. The boron levels in feed were adjusted with boric acid (Sigma–Aldrich, Steinheim, Germany). The feed treatments were prepared in 5-kg-weight batches. First, the powdery boric acid was diluted with 500 mL of water and impregnated with the feeds by spraying and impregnation. Then, it was lubricated to prevent boric acid in the feed from passing to the water by washing. The feed batches were dried in the shade and stored in buckets with covers. Similarly, water lubrication was applied to the control group. Hand-feeding to visual satiation was applied to the triplicates by feeding the fish twice daily (08:30 h and 16:30 h), where satiation was defined as the point when the fish did not approach the surface when food was offered. The satiation feeding was applied at the precise time by the same person throughout the feeding experiment to avoid the possibility of 'inter-worker' error.

Table 1. Nutritional values of the trout feed used in the study.

	Average of total feed (%)[a]
Diet composition	
Crude protein	45
Lipid	20
Crude cellulose	2.0
Crude ash	10
Macro elements	
Phosphorus	1.10
Calcium	1.90
Sodium	0.30

[a]Boron was found lower than the detection limit (0.02 mg/g) in the feed.

Growth performance calculations and statistical analysis

The specific growth rate (SGR, % day^{-1}) is 100 × (ln w1 − ln w0)/t, where w1 and w0 are the wet weights at times t1 and t0. The economic conversion ratio (ECR, US$ kg^{-1}) is [feed cost (US$$^{-1}$) + boron cost (US$ kg^{-1})] × feed conversion ratio (kg diet kg^{-1} fish). The feed conversion ratio (FCR) is (Wfinal − Winitial)/consumed feed, where Wfinal and Winitial are the live weights (g) of the fish on the initial (t) and final (T) days, respectively. The economic profit index [EPI (US$ $fish^{-1}$) is: final weight (FW, kg $fish^{-1}$) × fish sale price (US$ kg^{-1}) − ECR (US$ kg^{-1}) × weight increase (kg)], which was developed by Martinez-Llorens et al. (2007). The protein efficiency ratio (PER) is Weight gain (g)/protein intake (g) (Skalli et al. 2004). The condition factor (CF) is calculated as CF = (Body weight (g)/[Fish size (cm)]^3×100) (Martinez and Vázquez 2001).

The mean values and standard deviations of each growth parameter were calculated from the data of the three replicates for each treatment. The growth parameters were statistically tested by ANOVA using SPSS 15.0 for Windows. When only two groups were compared, the differences between the groups were tested using *post hoc* Tukey's HSD tests. The effects with $P < .05$ were considered significant.

Results

The boron concentrations of the water and initial feed used in this study were found lower than the detection limit. In addition to the initial fish weights, the growth parameters of rainbow trout calculated after 120 days of feeding are shown in Table 2. The FW of the feed treatment that contained 0.05% of boron was significantly the highest value (99.21 ± 3.76 g) compared to the other feed treatments ($P < .05$). However, the highest boron treatment, i.e. 0.20% of boron in feed treatment, has the significantly lowest FW value (78.74 ± 1.47 g). Similarly, the highest weight gain (WG) was found in the 0.05% boron treatment as 81.32 ± 3.76 g ($P < .05$). There were significant differences in WG between the 0.05% boron treatment and the other groups ($P > .05$). The feed intakes (FIs) of the 0.01% and 0.05% boron feed treatments (80.88 ± 0.86 g and 80.45 ± 1.42 g, respectively) were lower than the FIs of the control and the 0.10% and 0.20% boron treatments. The 0.05% boron treatment had the lowest FCR score (0.99 ± 0.02), whereas the scores were 1.15 ± 0.03 in the control and 1.30 ± 0.05 in the 0.20% boron treatment. The 0.05% boron feed treatment also had a higher SGR than the other treatments

Table 2. Effect of boron (B) in diet on growth parameters of rainbow trout after 120 days of feeding ($n = 105$ for each replicate) and the initial fish weight ($n = 1575$).

		B Levels in Feed*			
Growth parameters	Control	0.01%	0.05%	0.10%	0.20%
IFW (g)	17.75 ± 1.01^{a}	17.89 ± 1.19^{a}	18.03 ± 1.26^{a}	17.73 ± 1.09^{a}	18.04 ± 1.06^{a}
FW (g)	89.91 ± 2.11^{b}	90.56 ± 1.34^{b}	99.21 ± 3.76^{a}	88.14 ± 2.43^{b}	78.74 ± 1.47^{c}
WG (g)	72.02 ± 2.11^{b}	72.67 ± 1.34^{b}	81.32 ± 3.76^{a}	69.26 ± 2.43^{b}	60.85 ± 1.46^{c}
CF	$1.47 \pm 0{,}02^{a}$	1.32 ± 0.01^{c}	1.24 ± 0.02^{d}	1.39 ± 0.01^{b}	1.32 ± 0.02^{c}
FI	82.97 ± 1.51^{a}	80.88 ± 0.86^{ab}	80.45 ± 1.42^{ab}	79.31 ± 1.44^{b}	78.82 ± 2.33^{b}
FCR	1.15 ± 0.03^{b}	1.11 ± 0.03^{b}	0.99 ± 0.02^{c}	1.15 ± 0.05^{b}	1.30 ± 0.05^{a}
SGR	1.35 ± 0.19^{b}	1.35 ± 0.01^{b}	1.43 ± 0.03^{a}	1.32 ± 0.02^{b}	1.23 ± 0.01^{c}
PER	2.02 ± 0.05^{b}	2.09 ± 0.05^{b}	2.35 ± 0.06^{c}	2.03 ± 0.08^{b}	1.80 ± 0.07^{c}
SUR (%)	100	100	100	100	100
ECR ($/kg)	1.64 ± 0.04^{b}	1.58 ± 0.04^{b}	1.41 ± 0.04^{c}	1.63 ± 0.07^{b}	1.84 ± 0.07^{a}
EPI	0.18 ± 0.01^{b}	0.18 ± 0.01^{b}	0.21 ± 0.01^{a}	0.17 ± 0.01^{b}	0.15 ± 0.01^{c}

Notes: Each value indicates the average ± standard deviation. The averages expressed using different letters in each row are significantly different ($P < .05$).
*Boron (B) supplementation to the diet at rates of 0.01%, 0.05%, 0.10%, and 0.20 in feed and control (without B). The difference between the averages which is remarked in the same line with different letters (a–e) is significant ($P < .05$).
IFW: Initial fish weight.

and the control. The 0.20% boron treatment had the lowest SGR value (1.23 ± 0.01). The average PERs of all treatments were between 1.80 ± 0.07 and 2.35 ± 0.06, the 0.20% boron feed treatment had the minimum average value, and the 0.05% boron feed treatment had the maximum average value. All treatments including the control had 100% survival rate (SUR) values. The maximum and minimum ECR were calculated for 0.20% and 0.05% boron (1.84 ± 0.07 and 1.41 ± 0.04), respectively. There was no significant difference in EPI of the control, 0.01%, and 0.10% boron feed treatments. However, the 0.05% boron treatment had the highest EPI of 0.21 ± 0.01.

Discussion

The growth experiment was performed to reveal the effects of boron in the diets at 0.01%, 0.05%, 0.10%, and 0.20% on the growth parameters after 120 days of feeding. The growth parameters were FW, WG, CF, FI, FCR, SGR, PER, SUR, ECR, and EPI.

The 0.05% and 0.01% boron feed treatment had the best and second best growth parameter results, respectively. The FW was 99.21 ± 3.76 g and 89.91 ± 2.11 g for the 0.05% boron treatment and the control, respectively. All calculated parameters had a negative tendency, particularly in the 0.20% boron feed treatments. The FW in the 0.20% boron treatment was 78.74 ± 1.47 g. In all treatments, the SUR values were 100% with no mortalities during the feeding period. The ECR and EPI values indicate that the boron supplementation to the feed has economic benefits in rainbow trout aquaculture, which is one of the most common aquaculture practices. These results encourage the usage of boron-supplemented feed in other aquaculture species.

Both embryonic development and growth of fish are under the influence of diet composition as well as different growing conditions (Ozogul et al 2013; Durmus et al 2014; Oz 2016; Tasbozan et al 2016). For instance, a low boron concentration in water (up to 0.50 mg/L) provokes the embryonic development and increases the growth rate in fish. However, higher levels (>4.0 mg/L) reduce the growth (Loewengart 2001; Adhikari and Mohanty 2012). It has also been reported that 100 and 1000 mg concentrations of boric acid have toxic effects on tissues of juvenile rainbow trout within 96 hours (Topal et al. 2016). Similar to the results of waterborne boron, food-borne boron positively affects the fish growth at defined levels. The results of our study show that <0.05% of boron in the feed caused an increase in WG values and a decrease in FCR values, but >0.05% of boron in the feed reduced the growth parameters.

Two main reasons are assumed to cause the positive effects of boron supplementation on growth. First, boron has a role in the transportation of cell membranes (Takano et al. 2008). NaBC1, which is a boron transporter in cell membrane, serves as an electrogenic Na^{+}-coupled borate transporter, which is essential for cell growth and proliferation (Park et al. 2004). This transporter is related to the integrity and healthiness of the cell membrane (Park et al. 2005). The cell membrane has a cell division, which is mainly responsible for the growth of an organism and can be the main reason for the growth stimulation effect of boron. Second, boron attends to the pathways of numerous enzymatic reactions in animal metabolism (Howe 1998; Adhikari and Mohanty 2012). The boron compounds in animal metabolism protect against the development and decrease in triglyceride, notably low-density lipoprotein, total cholesterol, insulin, and non-esterified fatty acids in serum (Basoglu et al. 2002; Kabu and Civelek 2012). Moreover, boron plays a part in the oxidative stress reducing mechanisms such as the glucose–alanine cycle and methionine metabolism, and boron positively affects the lipid profile (Basoglu et al. 2011). Additionally, boron has many effects related to the bone metabolism in animals. The boron supplementation increases the bone abnormalities and magnesium concentrations in bones and enhances the absorption and retention of calcium and phosphorus (Hegsted et al. 1991; Bai and Hunt 1996). The relationship between boron and these cations induces the development of chickens (Hunt et al. 1994). Boron that was added to the diet of chickens at a level of 3 mg/kg promotes the growth (Hunt and Nielsen 1981). Nevertheless, boron can be toxic when its concentrations are more than the requirements of the animal species, although it is an essential element for organisms (Goldbach et al. 2007). The current study has shown that 0.20% boron in feed inhibits the growth of rainbow trout. This inhibition could have resulted from histopathological damage in fish tissues. Especially, the high concentrations of boron caused interstitial oedema, degenerative and atrophic changes in the myofibrils of muscle tissues of rainbow trout (Topal et al. 2016).

Conclusion

The present study indicates that adding 0.05% boron to the feed improves the growth of rainbow trout at unit time and lower cost. Higher boron concentrations than this level cannot enhance the growth parameters of rainbow trout. We suggest that using boron as an additive in the feed is beneficial for growth in rainbow trout. Moreover, further studies are required to examine the effects of the boron-supplemented feed on the fish meat quality, gamete quality, blood parameters, and resistance to diseases. The results of the current study demonstrate the utility of boron mineral for the feed industry as a new field.

Disclosure statement

No potential conflict of interest was reported by the author(s).

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