

Ameliorative effects of vitamin C (L-ascorbyl-2-polyphosphate) on growth, survival rate, feed conversion ratio, coefficient of variation, and hematology of juvenile snubnose pompano *Trachinotus blochii* (Lacepède, 1801)

^{1,2}Lam Son Ho, ¹Hue Thi Nguyet Nguyen, ³Khanh Thi Pham, ^{1,2}Hoang Do-Huu

¹ Institute of Oceanography, Vietnam Academy of Science and Technology, Nha Trang, Vietnam; ² Graduate University of Science and Technology, Viet Nam Academy of Science and Technology, Ha Noi, Viet Nam; ³ Institute of Aquaculture, Nha Trang University, Nha Trang, Vietnam. Corresponding author: H. Do-Huu, dohuuhoang2002@yahoo.com

Abstract. This study sought to determine the effects of vitamin C added to feed on the growth (daily growth rate in length - DLG; weight - DWG; specific growth rate in length - SGR_L, and weight - SGR_w) survival rate (SR), feed conversion ratio (FCR), coefficient of variation (CV), and hematology of juvenile snubnose pompano (*Trachinotus blochii*). Five levels of vitamin C (VC), including 0, 150, 300, 450, and 600 mg VC kg⁻¹, were supplemented to the control diet. Each treatment was repeated 3 times in marine cages (1×1×1 m) at a density of 30 fish per cage. After eight weeks of experiment, the results showed that there were significant differences in the growth parameters of fish ($p < 0.05$) among the VC treatments. Daily growth rate, relative growth rate, and specific growth rate in terms of length and weight were highest in the treatment with 450 mg VC kg⁻¹ of feed and lowest in the control treatment ($p < 0.05$). However, the experiment showed that the added VC to the feed did not affect the SR of snubnose pompano ($p > 0.05$). The FCR values were substantially reduced in fish fed diets supplemented with all VC levels ($p < 0.05$). Fish on a diet featuring 150 or 450 mg VC kg⁻¹ had substantially lower values of coefficient of variation in weight (CV_w) ($p < 0.05$), whereas the addition of 300 or 600 mg VC kg⁻¹ increased the CV_w values ($p < 0.05$). Moreover, the addition of VC to the diet substantially increased hematological parameters, such as red blood cell (RBC), white blood cell (WBC), hemoglobin (Hb), hematocrit (Ht), and serum protein ($p < 0.05$), whereas there was no difference in mean corpuscular hemoglobin (MCH), mean corpuscular volume (MCV) and mean corpuscular hemoglobin concentration (MCHC) among treatments ($p > 0.05$). Thus, dietary vitamin C positively influenced growth and hematological parameters in snubnose pompano in the 450 mg VC kg⁻¹ of feed treatment.

Key Words: blood parameters, dietary vitamin C, growth performance, pompano fish.

Introduction. The snubnose pompano (*Trachinotus blochii*) is a species with high economic value, that has been introduced to Viet Nam since 2004 (Binh & Quan 2011). However, the survival rate (SR) of imported fingerlings for farming is lower than that of domestic production breeds (Binh & Thanh 2008). Thus, fry produced successfully since 2009 can reduce the disadvantages of imported fingerlings, as well as technical protocols. However, the egg quality of the broodstock has been unstable because the SR of fish larvae is low, and the rate of malformation is still high (Hung 2011). Therefore, it is essential to comprehend the nutritional requirements during the early larval stage and to develop a reasonable feeding strategy that can affect the quality of fingerlings. Several studies have reported that an adjusted larval diet can improve the survival, growth and feed utilization ratio of shrimp and fish larvae (Gapasin et al 1998; Cahu et al 2003; Kashani et al 2010; Adel & Khara 2016). Feed supplements are quite diverse in the

market, and include unsaturated fatty acids (HUFAs), immune stimulants (MOS, β -glucan, selenium) (Do-Huu 2014; Sang et al 2015; Nguyen et al 2018; Do-Huu 2019; Do-Huu et al 2019; Do-Huu 2020; Pham et al 2022), pigments (astaxanthin and canthaxanthin) (Lam et al 2016), and vitamins (E, C, and D) (Do-Huu 2023; Do-Huu et al 2024b). Therefore, the quality of seeds can be improved by nutritional supplementation.

Ascorbic acid or vitamin C (VC) is a vital nutritional component that plays an important role in normal growth and physiological functions of fish. Most fish species cannot synthesize AA by themselves because of the lack of the L-gulonolactone oxidase enzyme (Chatterjee 1973). Therefore, dietary vitamin C supplementation is necessary to ensure growth and development of fish. AA, a water-soluble vitamin, plays an important role in the metabolism of collagen, which is a prerequisite for the formation of connective tissue, growth and development of the skeletal fertility system, reduction of stress, and boosting of immunity (Dabrowski et al 1988; Drouin et al 2011; Omoniyi & Ovie 2018). In addition, AA is considered an antioxidant because of its ability to reduce stress in fish (Dawood & Koshio 2018) and its role in immune function and disease resistance (Sobhana et al 2002; Ai et al 2006; Nsonga et al 2010; Adel & Khara 2016; Omoniyi & Ovie 2018). In the immune response, AA stimulates serum complement hemolysis, immune cell proliferation, phagocytosis, the release of signaling substances, and antibody production (Trichet et al 2015). In addition, AA deficiency can cause malformations of the gills and jaw, spinal deformities (curvature, arched back), growth reduction, and hemorrhage in the eyes and fins of fish (Dabrowski et al 1988; Fracalossi et al 1998). The AA demand of fish can support the maintenance of normal development and affect the survival, growth, and immunity of many fish species, such as cobia (*Rachycentron canadum*) (Zhou et al 2012), mrigal (*Cirrhinus mrigala*) (Zehra & Khan 2012), rainbow trout (*Oncorhynchus mykiss*) (Adel & Khara 2016), golden trevally (*Gnathanodon speciosus*) (Do-Huu 2021; Do-Huu et al 2024a), common carp (*Cyprinus carpio*) (Labh 2017), hybrid grouper (Ebi et al 2018), and golden trevally (*Gnathanodon speciosus*) (Do-Huu 2021). Vitamin C has also been reported to affect some hematological parameters in fish, such as sturgeon (*Acipenser ruthenus*) (Tatina et al 2010), common carp (*C. carpio*) (Labh 2017), rohu (*Labeo rohita*) (Tandel et al 2019), and flounder (*Platichthys stellatus*) (Yu et al 2020). The inclusion of 60 mg VC kg⁻¹ to the diet of pompano (*T. ovatus*) affected growth, meat quality, and antioxidant capacity. However, AA had an effect on growth, but did not affect the SR and FCR of this species (Zhang et al 2019).

Owing to the lack of studies on the effects of VC on snubnose pompano, this study provides information on the effects of VC on growth, survival, and swarm populations as hematological characteristics of snubnose pompano. The results also suggest the necessary amount of VC added to the feed to improve and promote the growth of snubnose pompano. These findings have important implications for economic efficiency in fish production.

Materials and methods

Ethical approval. All experimental procedures followed the regulations for animal care and the use of laboratory animals at VAST.

Diet preparation. The ingredients of the basal feed are listed in Table 1. VC derivatives L-ascorbyl-2-polyphosphate (35% ascorbic acid) were supplemented at 150, 300, 450 and 600 mg VC kg⁻¹ diet in the basal diet, providing 0.0, 51.36, 104.82, 154.09 and 205.45 mg ascorbic acid (AA) equivalent kg⁻¹ diet. The VC analysis was performed using reverse-phase high-performance liquid chromatography (HPLC; Model HP 1100, USA). Vitamin C was dissolved in water and mixed with the basal feed. The ingredients were mixed and pressed into a size suitable for the size of the fish mouth. The final feed was stored in plastic bags until use.

Table 1

Ingredient composition of experimental diets for snubnose pompano (*Trachinotus blochii*)
(g kg⁻¹ dry matter)

<i>Ingredients</i>	<i>g kg⁻¹</i>
Peruvian fish meal	55.20
Soybean meal	7.00
Yeast meal	9.44
Wheat meal	10.00
Squid liver oil	5.50
Soybean oil	2.80
DHA	0.05
Mineral premix*	0.10
Vitamin premix**	0.08
Seaweed powder + others	9.83
Total	100

Note: * - mineral premix (mg kg⁻¹ diet): FeSO₄ H₂O, 31.570 mg; MnSO₄ H₂O, 11.363 mg; CuSO₄ 5H₂O, 30950 mg; ZnSO₄ H₂O, 62.513 mg; CoCl₂ 6H₂O (1%), 2.513 mg; KI, 620 mg; selenium, 250 mg; MgSO₄ 7H₂O, 1200 mg; Ca(H₂PO₃)₂ H₂O, 3000 mg; ** - vitamin premix (mg or g kg⁻¹ diet): thiamin, 850 mg; riboflavin, 45 mg; pyridoxine HCl, 20 mg; vitamin B12, 6.0 mg; vitamin K₃, 350 mg; inositol, 500 mg; pantothenic acid, 60 mg; niacin acid, 200 mg; folic acid, 250 mg; biotin, 1.20 mg; retinol acetate, 32 mg; choline chloride, 2500 mg.

Experimental fish and design. Snubnose pompano were taken from Vung Ngan Marine Farm, Nha Trang University, with an average weight of 81.23±7.65 g, equivalent to 9.32±0.60 cm total length. Healthy fish, without scratches, no deformities, uniform size, and active swimming were selected for the experiment.

The experiment was carried out for eight weeks at the Vung Ngan Marine Farm, Nha Trang. Fish were reared in 15 cages at a density of 30 fish per cage. The experimental cage was made of a PE net with a size of each cage being 1×1×1 m, mesh size of 2 cm, and a net covering the top of the cage to prevent fish from jumping out. There were five diet treatments: 0, 150, 300, 450, and 600 mg VC kg⁻¹ in dry weight were added to the feed, with three replicates per treatment.

Feeding and husbandry. The experimental fish were fed twice a day (at 8 am and 4 pm) for 8 weeks. Mortalities were recorded during the experiment and dead fish were removed daily. The weight and length of all fish in each cage were measured twice per week. Environmental factors including dissolved oxygen (DO), pH, the transparency of water were assessed using the DO 200, pH model HI 991300, and Secchi plates, respectively. The experiment was conducted in offshore cages. Therefore, the environmental parameters depend on the natural conditions of the marine region. The environmental parameters are presented in Table 1. The environmental conditions during the experiment did not fluctuate. Salinity ranged from 32 to 35 ‰, pH was between 8.12-8.20, seawater temperature was 28-29°C, dissolved oxygen (DO) was between 6.8-7.2 mg L⁻¹, and transparency between 5.8-9.0 m. Snubnose pompano in the wild can live at a salinity of 3-33‰, appropriate temperature of 22-28°C, and DO over 2.5 mg L⁻¹ (Hung 2011). Thus, the environmental factors around the experimental cages were suitable for snubnose pompano.

Growth performance parameters

Specific growth rates in length (SGR_L) and weight (SGR_W) in % day⁻¹:

$$SGR_L = (\ln L_2 - \ln L_1) / (t_2 - t_1) \times 100$$

$$SGR_W = (\ln W_2 - \ln W_1) / (t_2 - t_1) \times 100$$

Daily length (DLG) and weight (DWG) growth:

$$DLG=(L_2-L_1)/(t_2-t_1)$$

$$DWG=(W_2-W_1)/(t_2-t_1)$$

Length gain (LG) and weight gain (WG):

$$LG=(L_2-L_1)/L_1 \times 100$$

$$WG=(W_2-W_1)/W_1 \times 100$$

Where: L_1 , L_2 - length of fish (cm) at time t_1 , t_2 (day); W_1 , W_2 - weight of fish (g) at time t_1 , t_2 (day); t_1 , t_2 - time at the start (t_1) and the end of the experiment (t_2) (day).

Fulton's condition factor (K) was also determined (Nash et al 2006):

$$K=W/L^3 \times 100$$

Where: K - condition factor; W - body weight (g); L - body length (cm).

$$FCR\% = W_t / (W_2 - W_1)$$

Where: FCR - feed conversion ratio; W_t - total consumed feed; W_2 - initial body weight (g); W_1 - final body weight (g).

$$SR = F_2 / F_1 \times 100$$

Where: SR - survival rate; F_1 - number of fish at the start of the experiment; F_2 - number of fish at the end of the experiment.

$$CV\% = S \times 100 / X$$

Where: CV - coefficient of variation of fish body length/weight; S - standard deviation of fish body length/weight; X - mean body length/weight.

Hematological analysis. Blood was collected from the caudal vein using a heparinized disposable hypodermic needle and transferred to a disposable heparinized tube. The collected blood samples were used for hematological analyses. Blood was drawn from another set of fish (three fish per replicate) for serum collection and frozen at -20°C until use. Red blood cell count (RBC), hemoglobin (Hb), white blood cell count (WBC), hematocrit (Ht), mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH), and mean corpuscular hemoglobin concentration (MCHC) were determined using a hematology analyzer (Sysmax XT-1800i Japan). Total serum protein (TSP) levels were determined using a biochemical immunological analyzer (COBAS 6000).

Statistical analysis. Statistical comparisons were performed by one-way analysis of variance (one-way ANOVA), followed by Duncan post-hoc test using SPSS version 20.0. Statistical significance was considered at $p < 0.05$. Data are presented as the mean \pm standard error.

Results

Effect of dietary ascorbic acid supplementation on the length of juvenile snubnose pompano. The lengths of snubnose pompano fed different levels of vitamin C supplementation for 8 weeks are presented in Table 2 and Figure 1. The experimental results showed that diets supplemented with different concentrations of vitamin C affected the length of fingerlings ($p < 0.05$).

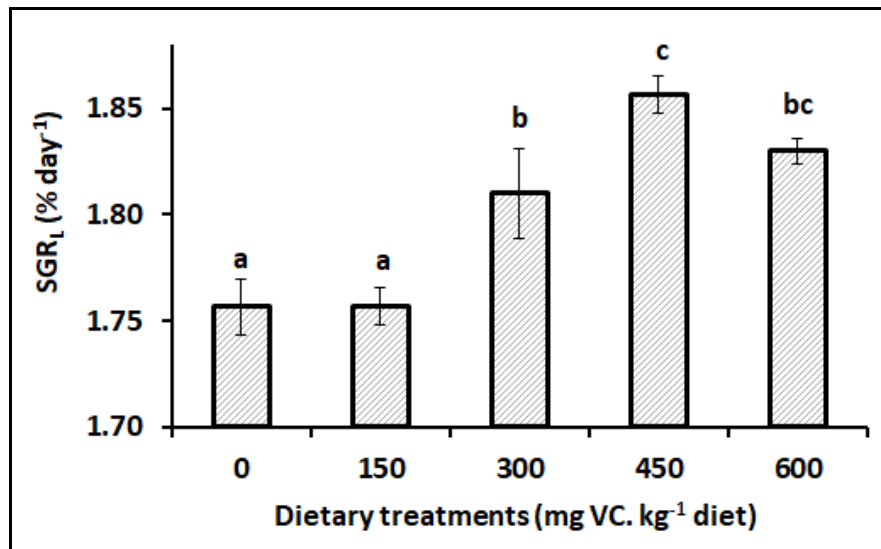


Figure 1. Effect of dietary ascorbic acid supplementation on specific growth rate in length (SGR_L) of juvenile snubnose pompano (*Trachinotus blochii*).

Table 2
Effect of dietary ascorbic acid supplementation on growth of juvenile pompano (*Trachinotus blochii*)

Targets	Experimental (mg VC kg ⁻¹ dry feed)				
	0	150	300	450	600
L ₁ (cm)	9.32±0.60 ^a	9.32±0.60 ^a	9.32±0.60 ^a	9.32±0.60 ^a	9.32±0.60 ^a
L ₂ (cm)	24.94±0.11 ^a	24.89±0.11 ^a	25.70±0.28 ^b	26.37±0.11 ^c	25.98±0.12 ^{bc}
DLG (cm day ⁻¹)	0.28±0.00 ^a	0.28±0.00 ^a	0.29±0.01 ^b	0.30±0.00 ^c	0.29±0.00 ^{bc}
LG (%)	167.33±2.19 ^a	167.33±1.20 ^a	175.67±2.96 ^b	183.00±1.16 ^c	178.67±1.20 ^{bc}

Note: different superscripts in each row show significant differences ($p < 0.05$); L₁ (g) - initial length; L₂ (g) - final length; DLG - daily length gain (cm day⁻¹); LG - length gain; data are presented as mean±standard error.

At the end of the experiments, the mean length of snubnose pompano fed with different concentrations of VC increased, ranging from 24.89-26.37 cm, with the most elevated length of 26.37±0.11 cm in the treatment supplied with 450 mg VC kg⁻¹. The average length of fish fed the 300-600 mg VC kg⁻¹ diet substantially increased, with a peak in fish fed 450 mg VC kg⁻¹ feed (Table 3). Similarly, the growth rates were substantially increased in cohorts of fish provided with 300 and 600 mg VC kg⁻¹ ($p < 0.05$), with the most elevated values of DLG, LG, and SGR_L observed in fish fed 450 mg VC kg⁻¹ feed. Although the experimental results showed the most elevated length growth rate in the treatment supplemented with 450 mg VC kg⁻¹ feed, the statistical analysis showed that there was no significant difference from the treatment with 600 mg VC kg⁻¹ feed ($p > 0.05$).

Effect of dietary ascorbic acid supplementation on body weight of juvenile snubnose pompano. The mean body weight, DWG, WG and SGR_w of snubnose pompano fed with different VC levels are presented in Table 4 and Figure 2. Supplying different VC concentrations affected the body weight of the fish ($p < 0.05$). At the experiment's conclusion, the most elevated weight (248.09±1.64 g) was found in the treatment with 450 mg VC kg⁻¹ of feed and the lowest weight (221.07±2.31 g) was found in the control treatment. The mean weight of fish fed a diet featuring 150-600 mg VC kg⁻¹ was substantially boosted ($p < 0.05$). In the groups of fish fed with vitamin C, the fish fed the diet with 450 mg VC kg⁻¹ had the most elevated mean weight, which was substantially higher ($p < 0.05$) than that of fish fed 150 and 300 mg VC kg⁻¹, but not substantially different from the mean weight of fish fed 600 mg VC kg⁻¹ ($p < 0.05$).

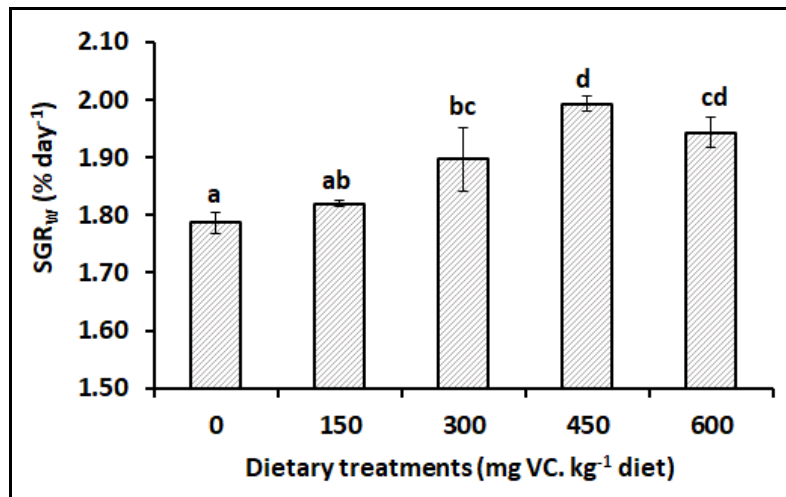


Figure 2. Effect of dietary ascorbic acid supplementation on specific growth rate in weight (SGR_w) of juvenile snubnose pompano (*Trachinotus blochii*).

Table 3
Effect of dietary ascorbic acid supplementation on some growth parameters of juvenile pompano (*Trachinotus blochii*)

Target	Treatments (mg VC kg ⁻¹ dry feed)				
	0	150	300	450	600
W ₁ (g)	81.23±7.65 ^a	81.23±7.65 ^a	81.23±7.65 ^a	81.23±7.65 ^a	81.23±7.65 ^a
W ₂ (g)	221.07±2.31 ^a	225.33±0.73 ^b	235.33±7.22 ^b	248.09±1.64 ^c	241.34±3.52 ^c
DWG (g day ⁻¹)	2.49±0.41 ^a	2.57±0.12 ^{ab}	2.75±0.13 ^{bc}	2.98±0.29 ^c	2.86±0.64 ^c
WG (%)	172.00±2.65 ^a	177.67±0.88 ^{ab}	189.67±8.95 ^{bc}	205.67±2.03 ^c	197.00±4.36 ^{bc}
K	1.35±0.01 ^a	1.39±0.01 ^b	1.39±0.01 ^b	1.38±0.00 ^{ab}	1.35±0.01 ^a

Note: different superscripts in a row show significant differences ($p < 0.05$); W₁ (g) - initial weight; W₂ (g) - final weight; DWG - daily weight gain (g day⁻¹); WG - weight gain; K - condition factor; data are presented as mean±standard error.

The growth parameters of the experimental snubnose pompano in terms of SGR_w, DWG, and WG tended to be proportional to the levels of VC added to the feed. The SGR_w ranged between 1.79-1.99% day⁻¹, DWG ranged between 2.49-2.98 g day⁻¹ and WG ranged between 172.00-205.67%. Fish receiving a diet enriched with 300-600 mg VC kg⁻¹ had significantly higher growth rates ($p < 0.05$), but fish fed 150 mg VC kg⁻¹ did not substantially change their weight growth in comparison to growth of fish in the control. The most elevated growth in weight was in the treatment with 450 mg VC kg⁻¹ dry feed (SGR_w of 1.99% day⁻¹, DWG of 2.98 g day⁻¹ and WG of 205.67%) and the lowest was in the control treatment (Table 3, Figure 2). In addition, growth in weight did not substantially differ between fish fed 300-600 mg VC kg⁻¹ feed ($p > 0.05$). The most elevated increase in weight was documented in fish fed with 450 mg VC kg⁻¹ of feed. In addition, K of snubnose pompano were considerably higher in the groups of fish that were fed all levels of VC supplementation.

Effect of dietary VC supplementation on FCR, SR and CV. The FCR, SR, and CV of snubnose pompano are presented in Table 4. A significant difference in CV values was observed between fish fed VC and the control treatment diets in terms of food consumption ($p < 0.05$; data not shown). FCR in all groups of fish fed VC supplementation was substantially lower than that in the control ($p < 0.05$).

Table 4

Food conversion ratio (FCR) and survival rate (SR) of snubnose pompano (*Trachinotus blochii*)

Target	Experiments (mg VC kg ⁻¹ of feed)				
	0	150	300	450	600
FCR	1.82±0.01 ^b	1.77±0.12 ^a	1.76±0.11 ^a	1.77±0.01 ^a	1.76±0.01 ^a
SR (%)	100±0.00	100±0.00	100±0.00	100±0.00	99.0±1.00

Note: different superscripts in each row show significant differences ($p < 0.05$); data are presented as mean±standard error.

The CV of pompano fed different levels of vitamin C after 8 weeks is presented in Figure 3. There was a statistically significant lower coefficient of variation in length (CV_L) value in fish fed 150 mg of VC kg⁻¹ and a substantially higher CV_L value in fish fed a diet featuring 300 mg VC kg⁻¹ compared with the CV_L value of fish fed the control diet ($p < 0.05$). However, the diets containing 450-600 mg VC kg⁻¹ did not modulate the CV_L value of snubnose pompano ($p > 0.05$). In addition, fish fed a diet featuring 150 or 450 mg VC kg⁻¹ had substantially lower coefficient of variation in weight (CV_W) values than those fed the control diet ($p < 0.05$). In contrast, the addition of 300 or 600 mg VC kg⁻¹ feed increased the CV_W values compared to CV_W in fish fed the basal diet ($p < 0.05$).

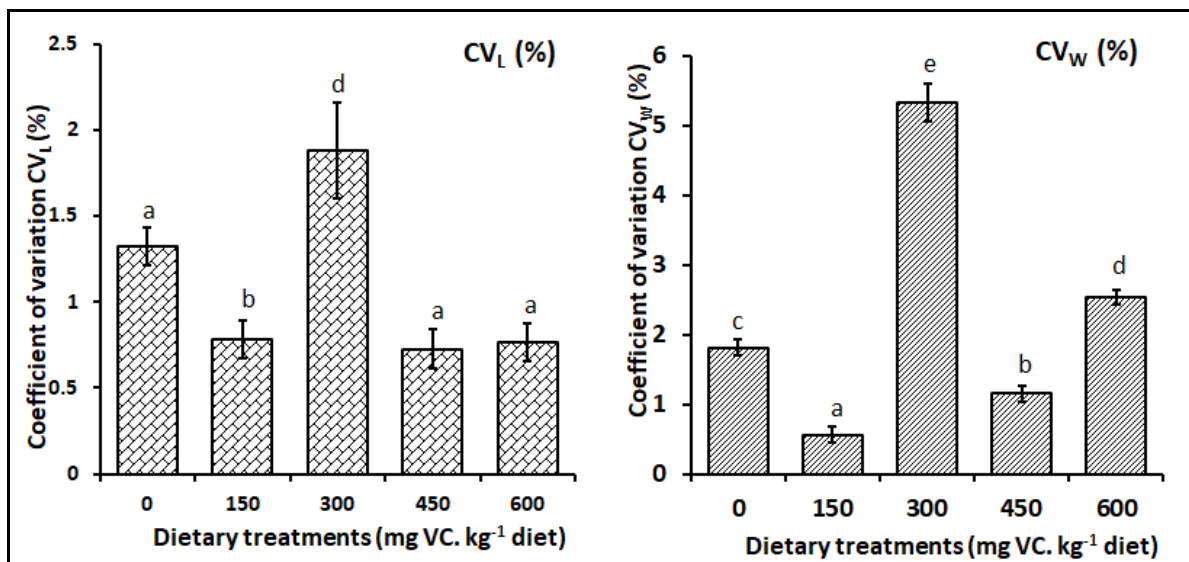


Figure 3. Coefficient of variation of length (CV_L %) and coefficient of variation of weight (CV_W %), (mean±standard error) of snubnose pompano (*Trachinotus blochii*).

Effect of dietary VC supplementation on the hematological parameters of snubnose pompano. The effects of VC supplementation on the hematological characteristics of snubnose pompano are presented in Table 5. VC supplementation at 300-600 mg VC kg⁻¹ substantially increased the red blood cells (RBC), white blood cells (WBC), and hematocrit (Ht). However, the ratio of RBC to WBC was not substantially affected by vitamin C supplementation ($p > 0.05$). In addition, hemoglobin (Hb) and total protein levels in the serum (TSP) were substantially increased in groups of fish fed a diet supplemented with 150-600 mg VC kg⁻¹ ($p < 0.05$). In contrast, there was no effect on mean erythrocyte volume (MCV), mean hemoglobin (MCH), and hemoglobin concentration (MCHC) ($p > 0.05$) (Table 5).

Table 5

Effect of dietary ascorbic acid supplementation on the hematological parameters of snubnose pompano (*Trachinotus blochii*)

Target	Experiments (mg VC kg ⁻¹ of feed)				
	0	150	300	450	600
RBC (×10 ⁶ cells mm ⁻³)	3.90±0.02 ^a	3.91±0.04 ^a	4.57±0.06 ^b	4.74±0.03 ^c	4.66±0.05 ^{bc}
WBC (×10 ⁴ cells mm ⁻³)	13.12±0.74 ^a	15.14±2.39 ^{ab}	16.78±0.41 ^{bc}	18.49±0.10 ^c	17.55±0.62 ^{bc}
RBC/WBC (%)	29.67±1.33	26.33±2.33	27.33±0.33	26.00±0.00	26.67±1.20
Hb (g dL ⁻¹)	7.23±0.15 ^a	7.73±0.38 ^b	8.10±0.32 ^{bc}	9.37±0.03 ^d	8.70±0.20 ^{cd}
Ht (%)	56.87±0.15 ^a	57.10±0.21 ^a	61.67±1.13 ^{bc}	63.27±1.04 ^c	60.37±0.57 ^b
MCV (fL)	123.53±3.24	120.53±1.69	123.30±6.45	124.33±4.47	123.03±3.88
MCH (pg)	18.77±0.09	18.70±0.46	19.30±0.38	18.93±0.55	18.33±0.35
MCHC (g dL ⁻¹)	15.57±0.54	15.10±0.58	15.50±0.66	16.03±0.26	16.00±0.25
TSP (mg mL ⁻¹)	42.63±0.19 ^a	44.50±0.29 ^b	50.40±0.31 ^c	50.63±0.32 ^c	46.63±0.17 ^d

Note: red blood cells (RBC), white blood cells (WBC), hemoglobin (Hb), hematocrit (Ht), mean erythrocyte volume (MCV), mean hemoglobin (MCH), mean hemoglobin concentration (MCHC) and total protein in the serum (TSP); different superscripts in each row show significant differences ($p < 0.05$); data are presented as mean±standard error.

Discussion. The positive effect of VC on the growth of snubnose pompano observed in this study is similar to previously published results. However, the VC content in this study may be higher than that in other published studies because the VC requirement depends on the fish species and size, VC types, and culture conditions (Zhou et al 2012). The optimum VC diet for growth in mrigan is 35 mg VC kg⁻¹ feed (Zehra & Khan 2012). Zhou et al (2012) reported that the supplementation of VC in the diet of cobia resulted in a higher SGR_w value than that of fish not supplemented with VC. In rainbow trout, supplementation with 150-250 mg VC kg⁻¹ of feed could affect fish growth with the most elevated WG (33±1.2%) and SGR_w (2.36±0.32% day⁻¹) obtained by the addition of 250 mg VC kg⁻¹ of feed (Adel & Khara 2016). 4.8-303 mg VC kg⁻¹ added to the feed helped improve bioavailability and growth of groupers (*Epinephelus fuscoguttatus* and *Epinephelus lanceolatus*), with the most elevated SGR_w of 2.83-3.24% day⁻¹ at 156.2 mg VC kg⁻¹ feed (Ebi et al 2018). In juvenile hybrid groupers, the optimal dietary VC requirement for the most elevated WG% of fish is 148 mg VC kg⁻¹ (Cai et al 2022). The weight of the golden trevally (*G. speciosus*) increased with the supply of 200 mg VC kg⁻¹ feed (Do-Huu 2021; Do-Huu et al 2024a). In juvenile golden pompano (*T. ovatus*), supplying 60 mg VC kg⁻¹ feed resulted in the most elevated growth rate (Zhang et al 2019). In common carp (*C. carpio*), the inclusion of VC at doses of 1000-2000 mg VC kg⁻¹ feed demonstrated a substantial positive trend in growth performance, with the highest WG and SGR achieved at 2000 mg VC kg⁻¹ feed (Labh 2017). However, some studies have indicated that the addition of VC to feed might not affect the growth of fish, such as for sea bream (*Sparus aurata*), with a supplement of 25-200 mg VC kg⁻¹ (Henrique et al 1998), or yellow croaker (*P. crocea*), with a supplement of 12.5-500 mg VC kg⁻¹ feed (Ai et al 2006).

Our results showed that the survival rate (SR) of fish in different treatments ranged from 99 to 100%. Statistical analyses indicated that VC supplementation did not substantially affect the SR of juvenile snubnose pompano ($p > 0.05$). In contrast, Ai et al (2006) found that the SR of yellow croaker improved with the addition of 25 mg VC kg⁻¹ of feed. Similarly, the SR of fish, such as Cobia (*R. canadum*) (Zhou et al 2012) and *C. mrigala* (Zehra & Khan 2012), was also affected by the addition of VC to feed. Zehra & Khan (2012) found that without adding VC to feed, the SR could reach 64.61±0.67%, whereas adding 35-95 mg VC kg⁻¹ feed could increase the SR to 100%.

In a study on hybrid groupers, the FCR of fish supplemented with 47.2 mg VC kg⁻¹ feed also had a lower FCR and was substantially different from that of the control group (Ebi et al 2018). The results of this study were similar to those of previous studies (Nsonga et al 2010; Zehra & Khan 2012; Labh 2017; Do-Huu 2021; Do-Huu et al 2024a). In addition, our results indicated that the size variation of snunose pompano,

presented as CV value was not affected by the vitamin C content in the diet. Contrary to our study, Pham et al (2021) indicated that dietary VC affected the CV of eels (*Monopterus albus*) at 120 mg VC kg⁻¹ feed, whereas supplementation with 200 mg VC kg⁻¹ feed resulted in substantially lower CV in golden trevally (*G. speciosus*) (Do-Huu 2021).

Hematological parameters (RBC, hematocrit, and hemoglobin) were used as indicators of fish health. RBCs are one of the major producers of free radicals, whereas some RBC, Ht, and Hb undergo peroxidation degradation of saturated fatty acids in phospholipid membranes due to changes in their quality (integrity, size) and quantity (Kiron et al 2004). Ren et al (2007) stated that blood parameters serve as indicators of fish health, with elevated Ht values reflecting the influence of VC and iron distribution, while the absence of VC in the diet impairs haemoglobin production. Our results showed that adding VC produced a marked positive effect on select hematological parameters of pompano (RBC, WB, Ht, Hb, and total serum protein). These results are similar to those of other studies. Affonso et al (2007) reported that RBC, WBC, Ht, and Hb were substantially increased in *Brycon amazonicus* supplemented with 600 and 800 mg VC kg⁻¹ of feed ($p < 0.05$). Nsonga et al (2010) also found the most elevated values of RBC, WBC, and Ht in *O. karongae* supplemented with 60 mg VC kg⁻¹ of feed. For Mekong catfish (*Pangasianodon gigas*), RBC and Ht substantially increased when supplemented with 750 mg VC kg⁻¹ of feed (Pimpimol et al 2012). Also, RBC, Ht and Hb increase in *C. mrigala* fed VC supplemented feed (Zehra & Khan 2012). In carp (*C. carpio*), VC supplementation had a positive effect on RBC, WBC, Ht, and Hb, but did not affect MCV, MCH, and MCHC (Labh 2017). RBC, WBC, Ht, Hb, MCV, and MCH of *Labeo rohita* increased substantially with the addition of VC to the feed (Tewary & Patra 2008; Tandel et al 2019). According to Yu et al (2020), the VC supplementation of 200-800 mg VC kg⁻¹ of feed in the diet for starry flounder (*P. stellatus*) could increase the number of RBC, Ht, Hb, for 2-4 weeks of the experiment. However, the VC supplementation in the diet did not affect WBC and serum protein in Mekong giant catfish (*P. gigas*) (Pimpimol et al 2012), and the Hb and total blood protein in red pacu (*Piaractus brachypomus*) (Hosseini et al 2015).

In addition, Adham et al (2000) demonstrated that insufficient VC supply in the diet can induce macrocytic anemia by decreasing Hb, RBC, and Ht levels. Changes in serum protein concentrations have been used as clinical indicators of health and stress in terrestrial and aquatic organisms (Riche 2007). Immunostimulants can increase immune function by affecting blood cells. Therefore, VC supplementation not only affects growth, but also affects the immune system of fish, especially non-specific immunity (Nsonga et al 2010; Trichet et al 2015; Adel & Khara 2016; Omoniyi & Ovie 2018). Our results, based on growth, FCR, and hematological parameters, indicate that the maximum concentration of vitamin C added to the snubnose pompano should be necessarily higher than 450 mg VC kg⁻¹ of feed.

Conclusions. After 8 weeks of the experiment on the effects of VC added to the feed for snubnose pompano, the results showed changes in growth rate, SR, CV, and hematological parameters. The addition of VC had a possible impact on growth, some hematological parameters, and the health of the snubnose pompano. Therefore, VC supplementation in the diet of snubnose pompano is necessary to promote the growth of commercially cultured fish to meet technical, environmental, and economic criteria. The recommended level of dietary VC supplementation for snubnose pompano is 450 mg VC kg⁻¹ of feed.

Acknowledgements. The authors are grateful for Project KC06.DA20-Nha Trang University and the Institute of Oceanography, Vietnam Academy of Science and Technology, for supporting the funding and material conditions to complete this research. We thank Tran Hua Thanh Van for caring for the fish. Special thanks to Mr. Thu Minh Phan and Dr. The Van Ho for providing insightful comments on the manuscript.

Conflict of Interest. The authors declare that there is no conflict of interest.

References

- Adel A., Khara H., 2016 XML The effects of different dietary vitamin C and iron levels on the growth, hematological and immunological parameters of rainbow trout *Oncorhynchus mykiss* fingerlings. Iranian Journal of Fisheries Sciences 15(2):886-897.
- Adham, Hashem, Abu-Shabana, Kamel, 2000 Vitamin C deficiency in the catfish *Clarias gariepinus*. Aquaculture Nutrition 6(2):129-139.
- Affonso E. G., Silva E. d. C., Tavares-Dias M., de Menezes G. C., de Carvalho C. S. M., Nunes É. d. S. S., Ituassú D. R., Roubach R., Ono E. A., Fim J. D. I., Marcon J. L., 2007 Effect of high levels of dietary vitamin C on the blood responses of matrinxã (*Brycon amazonicus*). Comparative Biochemistry and Physiology Part A: Molecular & Integrative Physiology 147(2):383-388.
- Ai Q., Mai K., Tan B., Xu W., Zhang W., Ma H., Liufu Z., 2006 Effects of dietary vitamin C on survival, growth, and immunity of large yellow croaker, *Pseudosciaena crocea*. Aquaculture 261(1):327-336.
- Binh T., Thanh T., 2008 [The primary results of the study on intensive culture of snubnose pompano (*Trachinotus blochii* Lacepede. 1801) in ponds with industrial feed]. Presentation at the National Young Scientific Conference on Aquaculture, Vietnam Journal of Science and Technology, Institute of Aquaculture Research I, Bac Ninh, Vietnam, 18 p. [In Vietnamese].
- Binh T. T., Quan N. V., 2011 [Techniques for the production of fry and commercial farming of snubnose pompano]. Agricultural Publishing House, Vietnam, 52 p. [In Vietnamese].
- Cahu C., Zambonino Infante J., Takeuchi T., 2003 Nutritional components affecting skeletal development in fish larvae. Aquaculture 227(1-4):245-258.
- Cai Q., Wu X., Gatlin D. M., Zhang L., Zhai H., Zhou Z., Yin H., Geng L., Irm M., 2022 Dietary vitamin C affects growth, antioxidant status and serum immune parameter of juvenile hybrid grouper (*Epinephelus fuscoguttatus* ♀ × *Epinephelus lanceolatus* ♂) fed low fishmeal diets. Aquaculture 556:738285.
- Chatterjee I. B., 1973 Evolution and the biosynthesis of ascorbic acid. Science 182:1271-1272.
- Dabrowski K., Hinterleitner S., Sturmbauer C., El-Fiky N., Wieser W., 1988 Do carp larvae require vitamin C? Aquaculture 72(3-4):295-306.
- Dawood M. A. O., Koshio S., 2018 Vitamin C supplementation to optimize growth, health and stress resistance in aquatic animals. Reviews in Aquaculture 10(2):334-350.
- Do-Huu H., 2021 Effects of dietary vitamin C supplementation on fingerling golden trevally, *Gnathanodon speciosus* (Carangidae). International Journal of Fisheries and Aquatic Studies 9(6):92-95.
- Do-Huu H., 2023 Effects of dietary vitamin E supplementation on growth performance, body composition of golden trevally, *Gnathanodon speciosus*. Asian Journal of Fisheries and Aquatic Research 24(6):37-45.
- Do-Huu H., 2019 [Efficiency of β -glucan utilisation in fish culture]. Vietnam Journal of Marine Science and Technology 19(4A):167-174. [In Vietnamese].
- Do-Huu H., 2020 Influence of dietary β -glucan on length-weight relationship, condition factor and relative weight of pompano fish (*Trachinotus ovatus*, family Carangidae). International Journal of Fisheries and Aquatic Studies 8(2):85-91.
- Do-Huu H., 2014 [The use of mannan oligosaccharide on aquaculture fish]. Science and Technology Journal of Agriculture and Rural Development 14:83-88. [In Vietnamese].
- Do-Huu H., Hue N. T. N., Hich T. V., 2019 Effects of dietary β -glucan supplementation on growth, innate immune, and capacity against pathogen *Streptococcus iniae* of juvenile pompano (*Trachinotus ovatus*). The Israeli Journal of Aquaculture – Bamidgeh 71, 10 p.
- Do-Huu H., Huynh S. M., Nguyen T. N. H., Vo H. T., Tran T. M. H., Huynh T. N. D., Nguyen T. V., 2024a Effects of single or combination of dietary vitamin C and β -

- glucan supplementation on golden trevally, *Gnathanodon speciosus* (Carangidae). Vietnam Journal of Marine Science and Technology 24(1):73-80.
- Do-Huu H., Thi Thanh Thuy N., Ky P. X., 2024b The ameliorative roles of dietary mannan oligosaccharides and vitamin E on growth performance, intestinal microbes, and structure, flesh quality, nutrient efficacy and stress resistance of pompano, *Trachinotus ovatus*. Aquaculture Reports 37:102233.
- Drouin G., Godin J. R., Pagé B., 2011 The genetics of vitamin C loss in vertebrates. Current Genomics 12(5):371-378.
- Ebi I., Yong A. S. K., Lim L. S., Shapawi R., 2018 Dietary ascorbic acid requirement for the optimum growth performances and normal skeletal development in juvenile hybrid grouper, *Epinephelus fuscoguttatus* × *Epinephelus lanceolatus*. Journal of King Saud University - Science 30(4):493-499.
- Fracalossi D. M., Allen M. E., Nichols D. K., Oftedal O. T., 1998 Oscars, *Astronotus ocellatus*, have a dietary requirement for vitamin C. The Journal of Nutrition 128(10):1745-1751.
- Gapasin R. S. J., Bombeo R., Lavens P., Sorgeloos P., Nelis H., 1998 Enrichment of live food with essential fatty acids and vitamin C: effects on milkfish (*Chanos chanos*) larval performance. Aquaculture 162(3-4):269-286.
- Henrique M. M. F., Gomes E. F., Gouillou-Coustans M. F., Oliva-Teles A., Davies S. J., 1998 Influence of supplementation of practical diets with vitamin C on growth and response to hypoxic stress of seabream, *Sparus aurata*. Aquaculture 161(1-4):415-426.
- Hosseini S. H., Sourinejad I., Ashori S., 2015 [Effect of dietary supplementation of Vitamin C on growth performance, survival rate and some hematological parameters in red pacu (*Piaractus brachypomus*)]. Journal of Animal Environment 7(2):197-204. [In Persian].
- Hung L., 2011 [Experimental production of snubnose pompano (*Trachinotus blochii* lacepède, 1801) in Khanh Hoa]. Final Report of Provincial Project, Nha Trang University, Nha Trang, Viet Nam. [In Vietnamese].
- Kashani Z., Imanpoor M., Shabani A., Gorgin S., 2010 Effect of dietary vitamin C, E and highly unsaturated fatty acid on growth and survival of goldfish (*Carassius auratus*). AACL Bioflux 3(4):281-288.
- Kiron V., Puangkaew J., Ishizaka K., Satoh S., Watanabe T., 2004 Antioxidant status and nonspecific immune responses in rainbow trout (*Oncorhynchus mykiss*) fed two levels of vitamin E along with three lipid sources. Aquaculture 234(1-4):361-379.
- Labh S., 2017 Effects of feeding higher level of vitamin C l-ascorbate-2-triphosphate calcium (l-tp) on growth and haemato-biochemical indices of common carp (*Cyprinus carpio* L). World Journal of Pharmaceutical Research 6(6):904-921.
- Lam H. S., Vy N. T., Ngoc P. T., 2016 [Effect of dietary astaxanthin on growth, survival rate and pigmentation of commercial clownfish, *Amphiprion ocellaris*]. Vietnam Journal of Marine Science and Technology 16:321-327. [In Vietnamese].
- Nguyen V. Q., Luong C. T., Do-Huu H., Cao V. N., Huynh S. M., 2018 [Effects of dietary b-glucan on growth and survival of pompano fish, (*Trachinotus ovatus*, Linnaeus 1758)]. Journal of Tropical Science and Technology 17:49-55. [In Vietnamese].
- Nsonga A., J o., W M., Soko C., Mtethiwa A., 2010 Effect of varying levels of dietary vitamin C (ascorbic acid) on growth, survival and hematology of juvenile tilapia, *Oreochromis karongae* (Trewavas 1941) reared in aquaria. Brazilian Journal of Aquatic Science and Technology 13(2):17-23.
- Nash R. D. M., Valencia A. H., Geffen A. J., 2006 The origin of Fulton's condition factor - Setting the record straight. ESSAY: Fisheries History 31(5):236-238.
- Omoniyi A. D., Ovie I. A., 2018 Vitamin C: An important nutritional factor in fish diets. Journal of Agriculture and Ecology Research International 16(2):1-7.
- Pham H. D., Le M. H., Dinh K. V., Siddik M. A. B., Do-Huu H., Ngo M. V., 2022 Effects of enrichment *Artemia* with organic selenium and essential fatty acids on growth performance and fatty acid composition of barramundi (*Lates calcarifer*) larvae. Regional Studies in Marine Science 55:102595.

- Pham T. A., Nguyen T. S., Le H. N., 2021 [Effects of dietary vitamin C on growth, survival rate and coefficient of variation of swamp eel (*Monopterus albus* Zuiew, 1793) from 15 to 60 days]. *Journal of Fisheries Science and Technology* 1:2-8. [In Vietnamese].
- Pimpimol T., Phoosamran K., Chitmanat C., 2012 Effect of dietary vitamin C supplementation on the blood parameters of Mekong giant catfish (*Pangasianodon gigas*). *International Journal of Agriculture and Biology* 14:256-260.
- Ren T., Koshio S., Ishikawa M., Yokoyama S., Micheal F. R., Uyan O., Tung H. T., 2007 Influence of dietary vitamin C and bovine lactoferrin on blood chemistry and non-specific immune responses of Japanese eel, *Anguilla japonica*. *Aquaculture* 267(1-4):31-37.
- Riche M., 2007 Analysis of refractometry for determining total plasma protein in hybrid striped bass (*Morone chrysops* x *M. saxatilis*) at various salinities. *Aquaculture* 264(1-4):279-284.
- Sang H. M., Thuy N. T. T., Do-Huu H., 2015 Effects of dietary organic selenium on growth, survival, physiological and hematology conditions of snub-nose dart (*Trachinotus blochii* Lacepede, 1801). *Israeli Journal of Aquaculture - Bamidgeh* 67, 9 p.
- Sobhana K. S., Mohan C. V., Shankar K. M., 2002 Effect of dietary vitamin C on the disease susceptibility and inflammatory response of mrigal, *Cirrhinus mrigala* (Hamilton) to experimental infection of *Aeromonas hydrophila*. *Aquaculture* 207(3-4):225-238.
- Tandel R., Sharma A., Aadil R., Bhat H., Dash P., Bhat A., Deo A., 2019 Dietary vitamin C supplementation on growth and haemato-biochemical parameters of *Labeo rohita* (Ham.) fingerlings. *Journal of Entomology and Zoology Studies* 7(1):72-79.
- Tatina M., Bahmani M., Soltani M., Abtahi B., Gharibkhani M., 2010 Effects of different levels of dietary vitamins C and E on some of hematological and biochemical parameters of sterlet (*Acipenser ruthenus*). *Journal of Fisheries and Aquatic Science* 5(1):1-11.
- Tewary A., Patra B. C., 2008 Use of vitamin C as an immunostimulant. Effect on growth, nutritional quality, and immune response of *Labeo rohita* (Ham.). *Fish Physiology and Biochemistry* 34(3):251-259.
- Trichet V. V., Santigosa E., Cochin E., Gabaudan J., 2015 The effect of vitamin C on fish health. In: *Dietary nutrients, additives, and fish health*. Wiley-Blackwell, pp. 151-171.
- Yu Y. B., Park H. J., Kang J. C., 2020 Effects of dietary ascorbic acid on growth performance, hematological parameters, antioxidant and non-specific immune responses in starry flounder, *Platichthys stellatus*. *Aquaculture Reports* 18:100419.
- Zehra S., Khan M. A., 2012 Dietary vitamin C requirement of fingerling, *Cirrhinus mrigala* (Hamilton), based on growth, feed conversion, protein retention, hematological indices, and liver vitamin C concentration. *Journal of the World Aquaculture Society* 43(5):648-658.
- Zhang G., Wang S., Chen C., Ma Y., Xie D., Wang Y., Sun L., You C., Li Y., 2019 Effects of dietary vitamin C on growth, flesh quality and antioxidant capacity of juvenile golden pompano *Trachinotus ovatus*. *Aquaculture Research* 50(10):2856-2866.
- Zhou Q., Wang L., Wang H., Xie F., Wang T., 2012 Effect of dietary vitamin C on the growth performance and innate immunity of juvenile cobia (*Rachycentron canadum*). *Fish & Shellfish Immunology* 32(6):969-975.

Received: 25 August 2024. Accepted: 27 September 2024. Published online: 30 October 2024.

Authors:

Lam Ho Son, Institute of Oceanography, Vietnam Academy of Science and Technology, 01 Cau Da St., Nha Trang 650000, Viet Nam; Graduate University of Science and Technology, Viet Nam Academy of Science and Technology, 18 Hoang Quoc Viet St, Ha Noi 100000, Viet Nam, email: hslamqt@gmail.com

Hue Nguyen-Thi Nguyet, Institute of Oceanography, Viet Nam Academy of Science and Technology, 01 Cau Da St., Nha Trang 650000, Viet Nam, email: huenguyen82@gmail.com

Khanh Thi Pham, Institute of Aquaculture, Nha Trang University, 02 Nguyen Dinh Chieu St., Nha Trang 650000, Viet Nam, email: khanhpt@ntu.edu.vn

Hoang Do-Huu, Institute of Oceanography, Vietnam Academy of Science and Technology, 01 Cau Da St., Nha Trang 650000, Vietnam; Graduate University of Science and Technology, Viet Nam Academy of Science and Technology, 18 Hoang Quoc Viet St, Ha Noi 100000, Viet Nam, email: dohuuhoang2002@yahoo.com

How to cite this article:

Lam H. S., Hue N. T. N., Khanh T. P., Do-Huu H., 2024 Ameliorative effects of vitamin C (L-ascorbyl-2-polyphosphate) on growth, survival rate, feed conversion ratio, coefficient of variation and hematology of juvenile snubnose pompano *Trachinotus blochii* (Lacepède, 1801). *AAFL Bioflux* 17(5):2324-2336.