Enrichment of *Artemia* nauplii with essential fatty acids and vitamin C: effect on rainbow trout (*Oncorhynchus mykiss*) larvae performance

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Abstract

The effect of essential fatty acids (EFA) and vitamin C-enriched *Artemia* nauplii on growth, survival, and resistance to temperature (high) stress in rainbow trout larvae reared in tanks were investigated. The larvae (average weight 120.43mg±13.5) were fed 6 times daily starting at the onset of exogenous feeding for 1 week. Triplicate groups of fish were offered one of four treatments (1) commercial starter food for rainbow trout، (2) newly hatched *Artemia* nauplii (unenriched)، (3) highly unsaturated fatty acid (HUFA) + vitamin C-enriched *Artemia* nauplii and (4) combination of 10 % HUFA+ vitamin C enriched nauplii and commercial starter food. After 1 week, all groups of fish were switched to the commercial diet for an additional period of 3 weeks. Statistical analysis of growth after the first week and at the end of the experiment, showed that growth of larvae in various treatments was significantly different ($P<0.05$). After 4 weeks, the larvae in treatment 3 with the average weight of 657.50 ± 57.93 mg had the highest body weight ($P<0.05$). The highest percentage of survival (96%) was observed in treatment 3 ($P<0.05$). Proximate compositions of trout larvae after one week feeding with experimental diets showed that the protein in the larvae of treatments 3 and 4 was significantly different compared to other treatments ($P<0.05$). The best result of resistance to temperature (up to 24°C) was observed in larvae reared on treatment 3 with 91.34 ± 1.52 percent ($P<0.05$).

Keywords: Essential fatty acid, vitamin C, *Oncorhynchus mykiss*, stress resistance, growth and survival

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Introduction

Successful rearing of larval fish is the most critical stage in the production cycle for many species. *Oncorhynchus mykiss* has a promising market potential in Europe, East and South Asia. It is also an important aquaculture species in Iran. The high nutritional quality of its flesh encourages investigations on the aquaculture potential of this excellent food fish (Sedgwick, 1990). The consumer demand stimulated the development of intensive aquaculture of this species in Asian countries. The problem in rearing larval fish is that of food supply (Leger et al., 1986). Therefore a readily available, easily acceptable and highly digestible diet with high nutritional value should be used as larval fish starter diet (Girii et al., 2002).

Improvements in larval nutrition are necessary to solve some of these problems to come up with a viable and dependable *Oncorhynchus mykiss* larviculture technology for commercial scale application.

Species reared successfully in aquaculture have generally been large larvae with fully developed digestive systems at the first feeding stage, e.g. salmon, trout and catfish. Live prey organisms, primarily zooplankton, have been used to raise the larvae of fish species which cannot be reared on prepared feeds (Leger et al., 1987). Among zooplankton, brine shrimp (*Artemia* spp.) and rotifers (Brachionus plicatilis) have been used most extensively as live food for rearing marine and freshwater fishes (Leger et al., 1987; Bengtson et al., 1991; Hosseinpour et al., 2010). The use of *Artemia* nauplii is well established due to its many advantages: year- round availability as on-the shelf cysts; good nutritional value for some fish; and relatively easy improvement through simple enrichment techniques (Leger et al., 1987). Nutritional deficiencies have been another concern when using brine shrimp. Some stocks of *Artemia* nauplii have shown a deficiency in eicosapantaenoic acid (EPA; 20:5n-3) and doccosahexanoic acid (DHA; 22:6n-3) (Takeuchi and Watanabe, 1982). The essential fatty acids (EFA) for fish are broadly recognized to comprise polyunsaturated fatty acids (PUFA) with carbon chain lengths of 18 and HUFA with carbon chain lengths of 20 and 22, of both the n-3 and n-6 series. These fatty acids cannot be synthesized by the fish de novo, though the 18- carbon PUFA can be converted by some species to longer-chain, more highly unsaturated fatty acids of the same series (Sargent et al., 1989). On the other hand, ascorbic acid is also an important micronutrient in fish diet. It is needed in the synthesis of collagen necessary in the formation of connective tissues and bone matrix (Dabrowski et al., 1994). Hence, these fatty acids and vitamin C must be provided in the diet to meet the fish requirements.

Several studies have demonstrated the positive effect of enriched live food on the growth and survival performance of various aquaculture species (Gapasin et al., 1998; Lim et al., 2002; Noori et al., 2005). The effects of supplemental ascorbic acid in enriched live food for *Clarias garipinus* larvae of start feeding (Merchie et al., 1997), labrox and *Clarias gariemus* (Merchie et al., 1995b), *Chanos chanos*

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(Gapasin et al.,1998), *Penaeus monodon* (Merchie et al 1998), *Penaeus vannamei* (Wouters et al.,1999) and *Bidyanus bidyanus* (Smith et al.,2004) has been investigated.

The aim of this study was to test the effect of EFA and ascorbate supplementation in enhancing rainbow trout larval growth, survival and resistance to temperature stress (up to 24 °C).

Materials and methods

Artemia cysts (Urmia Lake, Iran) were hatched following standard procedures (Sorgeloos et al., 1993; Lavens and Sorgeloos., 1996). Newly hatched *Artemia* (Instar I) nauplii (200,000 nauplii / l) were divided in batches in 5 liter plexiglass tanks. The enrichment protocol followed the method of Treece, (2000). 30g of gelatin was dissolved in 800 ml boiled deionized water and was left to cool to 40 °C. 160 ml cod liver oil was mixed with ascorbic acid (16g) and 4 raw egg yolks, it was homogenized using a blender and stored in the refrigerator for 1 week. 0.5 ml of the enrichment suspension (assuming a density of 200 *Artemia* per ml) was added per liter to the incubation water at the onset of the enrichment period. Another 0.5 ml/l of the enrichment diet was added 12 hours before harvesting and nauplii were harvested after 24 hours. Newly hatched *Artemia* nauplii served as the control (Leger et al., 1987; Treece, 2000). Samples of unenriched and enriched *Artemia* were also collected regularly and stored at -20ºC. These samples were later used for the analysis of fatty acid methyl esters. Vitamin C was not analyzed in the present study.

The four treatments (in a completely randomized design with 3 replicates per treatment) were: (1) larvae fed commercial starter food ≠2 or ≠3 (Bioproducts, Inc., France, 58% protein, 15% lipid and 11% ash); (2) larvae fed newly hatched *Artemia* nauplii; (3) larvae fed HUFA+vitamin C enriched *Artemia* nauplii and (4) larvae fed a combination of 10% HUFA+vitamin C enriched nauplii and commercial starter food. Treatment 1 served as control. The fish larvae in all treatments were fed 6 times per day (4, 8, 12, 16, 20 and 24 hours). The daily ration was adjusted according to larvae weight gain after 7 and 14 days of rearing. Trout larvae at the first feeding stage (swim up) were obtained from a local *Oncorhynchus mykiss* hatchery, in Gorgan, Iran. 1200 uniformly sized yolk-sac larvae (120.63mg±13.50 SD) were randomly divided into 12 groups (four treatments, three replicates) of 100 individuals. Fishes of each group were transferred in to a 35 liter tank using flow-through system at the rate of 0.5 L/h. Aeration was applied through a number of narrow pipes terminating to bubbliers. Culture tanks were cleaned daily, and physic-chemical parameters were measured every morning prior to feeding. Water quality was maintained within optimum range: temperature (9.3 ±1.36 °C), dissolved oxygen (7.8-8.6 mg/l), pH (8- 8.2), total ammonia (0.5±0.03 mg/l), residual chlorine (0.05±0.03 mg/l) and the photoperiod was set at 12L: 12D cycle (light period from 8-20 hours) and light intensity was kept at 40 lux at the tank surface. Dead larvae were removed twice daily and counted. Feeding was stopped

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six hours prior to sampling fish for chemical analysis and growth measurement on days 8, 21 and 29. Ten fish from each replicate were randomly harvested at weekly intervals, bulk-weighted and the total length (TL) was taken. The amount of feed fed per group was recorded weekly and used to calculate feed efficiency ratios (Wang et al., 2005). 90 fish per treatment were randomly collected (30 fish per replicate tank) on days 7 and 28. Samples were oven-dried at 60 °C for 24 h then stored at -20 °C. These dried samples were later analyzed for fatty acid methyl esters (Lepage and Roy, 1986), using flame ionization, (DANI-1600 models, Italy). Proximate composition; moisture, crude protein, crude lipid and ash were also measured in duplicate using standard methods (AOAC, 1990). Moisture by drying samples at 105 °C overnight, protein by measuring kjeldahl nitrogen, lipid was analyzed by ether extraction using a soxhlet system, and ash by heating for 5h at 550 °C in a muffle furnace.

Twenty nine day- old trout larvae were subjected to temperature stress test following the method described by Ako et al., 1994; and Kanazawa, 1995). The test involved immersing fish 10 fish larvae/replicate in 24 °C for a period of one hour. The mortality was recorded at every 5 min intervals. Results were expressed as % total body dry weight

$$SGR\% = \frac{(\mathrm{LnWt2} - \mathrm{LnWt1})}{t2 - t1} \times 100$$ (Koueta et al., 2002)

SGR=specific growth rate Wt2 = final weight (mg) of each fish

Wt1 =Initial weight (mg) of each fish t= duration of the experiment in days

$$BWI\% = \frac{Wt2 - Wt1}{Wt2} \times 100$$ (Koueta et al., 2002)

BWI=body weight Increase Wt1 =Initial weight (g) of each fish

Wt2 = Final weight (g) of each fish

$$IWG\ (mg/day) = \frac{WG}{t} \times 1000$$ (Lara-Flors et al., 2003)

IWG=increased weight gain WG=weight gain (g)

$$CV\ SGR\% = \frac{SD}{mean\ SGR} \times 100$$ (Wang et al., 2005)

CV SGR= coefficient of variation for SGR

SD=standard deviation of SGR

$$FCR = \frac{\text{feed fed(g dry weight)}}{\text{fish weight gain (g wet weight)}}$$ (Lara-Flors et al., 2003)

At the end of the experiment the number of surviving fish was recorded and used for calculating mortality. Diet effects on total length, survival, weight, SGR and temperature stress were analyzed using two- way analysis of variance (ANOVA) (SPSS version 9).

Results

Fatty acid contents of newly hatched and enriched *Artemia* are shown in Table 1.

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The individual fatty acid levels were consistently higher (except for 18: n-9 and 16:0) in the HUFA+ vitamin C-enriched nauplii than in the newly hatched nauplii. The HUFA, EPA and DHA levels (8.99±0.29, 7.72±0.32 and 1.27±0.25 mg/g DW respectively) were highest in treatment 3.

Table 1: Certain fatty acids (mg FA/g DW) of newly *Artemia nauplii* (A), *Artemia* enriched with HUFA +vitamin C (B)

Fatty acid	A	B
14:0	**1.30±0.10**	**1.30±0.20**
16:0	**15.79± 0.39**	**15.50±1.95**
18:0	**3.99±0.78**	**4.53±0.07**
18:1n-9	**18.35±0.47**	**16.67±0.43**
18:2n-6	**1o.12±1.95**	**11.33±0.54**
18:3n-3	**30.48±1.83**	**36.43±0.37**
20:5n-3	**2.80±0.43**	**7.72±0.32**
22:6n-3	**tr**	**1.27±0.25**
SFA	**21.41±0.84**	**22.59±0.30**
USFA	**62.51±0.57**	**67.83±0.37**
PUFA	**27.18±1.84**	**43.42±3.5**
HUFA	**2.80±0.43**	**8.99±0.28**

*Data are mean±SD (n=3), SFA=saturated fatty acid, USFA= unsaturated fatty acid, HUFA= highly unsaturated fatty acid and PUFA=poly unsaturated acid, tr=trace

Fatty acid content of trout larvae is shown in Table 2. The EPA (2.98±0.32 mg/g DW) and DHA (0.36±0.10 mg/g DW) levels in treatment 3 were generally high compared to other treatments. The PUFA level was significantly different in all treatments ($P<0.05$), with highest PUFA observed in treatment 3 ($P<0.05$). The HUFA level was high in treatment 3 and in other treatments it was zero (Table2).

Table 2: Whole- body fatty acid composition (mg FA/g DW) of 29-day old trout larvae fed different diets

Fatty acid	Treatment 1	Treatment 2	Treatment 3	Treatment 4
14:0	3.49 ± 0.17^{a}	0.69 ± 0.03^{c}	0.85 ± 0.094^{c}	3.12 ± 0.06^{b}
16:0	23.86 ± 0.25^{b}	22.68 ± 0.15^{c}	19 ± 0.19^{d}	28.44 ± 0.11^{a}
18:0	1.8 ± 0.16^{d}	6.23 ± 0.25^{a}	5.82 ± 0.11^{b}	4.22 ± 0.18^{c}
18:1n-9	14.99 ± 0.08^{c}	14.67 ± 0.08^{d}	16.61 ± 0.14^{a}	15.48 ± 0.19^{b}
18:2n-6	18.07 ± 0.11^{b}	13.95 ± 0.21^{c}	12.23 ± 0.12^{d}	19.55 ± 0.26^{a}
18:3n-3	1.36 ± 0.069^{d}	3.26 ± 0.20^{b}	5.31 ± 0.08^{a}	2.6 ± 0.15^{c}
20:5n-3	-	-	2.98±0.032	-
22:6n-3	-	-	0.36±0.10	-
SFA	19.14 ± 0.11^{b}	17.84 ± 0.06^{d}	21.01 ± 0.032^{a}	18.84 ± 0.20^{c}
USFA	39.15 ± 0.19^{b}	34.10 ± 0.19^{d}	37.91 ± 0.032^{c}	41.15 ± 0.12^{a}
PUFA	1.29 ± 0^{d}	3.30 ± 0.21^{b}	8.14 ± 0.26^{a}	2.91 ± 0.16^{c}
HUFA	-	-	3.44±0.23	-

*Values in each rows with different superscripts are significantly different ($P<0.05$)

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Fish in treatment 2 had the lowest unsaturated fatty acid (HUFA) significantly lower than other treatments (P<0.05). Trout fed *Artemia* enriched with HUFA+ vitamin C (treatment 3) exhibited significantly higher (P<0.05) growth compared to other treatments after 29 days of culture (Table 3). Growth trends were consistent in all 4 treatments and observed to be relatively better in treatment 3 or treatment 4 compared with treatment 1 and 2. Different treatments, showed significant difference in the CV for SGR over the course of experiment (P<0.05). Larvae fed commercial starter food showed significant increase in CV for SGR while the lowest was observed in treatment 3 (Table 4).

Table 3: Average weight and total length [a], percent weight gain of fish fed various dietary treatments. Values are mean ± standard deviation (n=3)

Treatment	Time (day)	Average weight (mg)	Average total length (mm	%weight gain
1	8	180.1±7.6^{d}	21.07±1.1^{b}	8.63±1.50^{d}
2	8	202.76±8.3^{c}	27.72±0.4^{b}	11.82±1.1^{c}
3	8	219.4±20.5^{a}	28.44±0.9^{a}	13.63±1.8^{a}
4	8	210.7±19.1^{b}	28.16±0.8^{a}	12.73±1.5^{b}
1	21	362±22.2^{b}	33.44±0.7^{c}	11.53±0.4^{c}
2	21	358.86±26^{b}	34.02±0.7^{b}	11.37±0.6^{c}
3	21	428.13±28.2^{a}	35.42±0.81^{a}	14.48±0.9^{a}
4	21	417.33±19^{a}	35.07±0.5^{a}	14.08±0.9^{a}
1	29	568.3±20.7^{c}	38.7±0.4^{c}	15.47±0.2^{c}
2	29	560.63±27.3^{c}	38.75±0.6^{c}	15.19±0.5^{c}
3	29	657.5±57.9^{a}	40.74±1.1^{a}	18.39±1.7^{a}
4	29	596.5±39.2^{b}	40.01±0.7^{b}	16.37±1^{b}

*Within columns values with different superscripts are significantly different (P<0.05)

[a] Initial weights and lengths of trout larvae 120.63(mg) ±13.50 SD and 23.26 (mm)± 0.90 SD respectively

When 29- days-old trouts were subjected to temperature stress, mortality rates of the HUFA+ vitamin C-treated (treatment 3) and 10% HUFA+ vitamin C+ commercial starter food treated fish (treatment 4) were significantly lower (P<0.05) than other treatments (Table 5). The highest mortality was observed in treatment 1. After 29 days of culture, survival significantly differed among the treatments (P<0.05). The highest survival was observed in treatment 3 (Table 6). The chemical composition analysis of trout larvae of each treatment on days 8 and 29 (Table 7) showed that the crude protein level changed after either 1 week or 4 weeks of sampling. The lowest level of protein (65.60 %) was in treatment 1 after 1 week and 68.70% after 4 weeks. The highest protein level (67.71% and 71.82%) after 1 and 4 weeks respectively was in treatment 3. The highest level of crude lipids in the carcass of larvae 14.64% was found in treatment 1 while the lowest (13.67%) was in treatment 2.

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Table 4: Specific growth rate (SGR), food conversion ratio, body weight increase (BWI) per day and coefficient of variation for SGR of fish fed various dietary treatments. Values are mean ± standard deviation (n=3)

Treatment	Time (day)	SGR%/day	%BWI	CV of SGR %	Feed conversion ratio
1	8	5.95±1.52^{b}	52.57±17.1^{b}	26.98±6^{c}	0.7±0.05^{c}
2	8	7.59±1.3^{a}	70.9±16.8^{a}	18.10±2.8^{b}	0.48±0^{b}
3	8	8.12±0.5^{a}	76.72±6.3^{a}	6.30±0.4^{a}	0.42±0.01^{a}
4	8	7.83±0.5^{a}	73.23±6.6^{a}	7.18±0.5^{a}	0.46±0^{ab}
1	21	25.30±0.4^{b}	205.79±27.7^{b}	7.77±0.5^{d}	0.69±0.04^{a}
2	21	5.24±0.32^{b}	201.41±21.3^{b}	6.12±0.34^{b}	0.69±0.01^{a}
3	21	5.90±0.11^{a}	245.47±8.3^{a}	1.91±0.03^{a}	0.74±0.03^{a}
4	21	5.88±0.18^{a}	244.08±13.5^{a}	1.82±0.03^{a}	0.77±0.03^{b}
1	29	5.40±0.^{3bc}	381.37±51.5^{b}	6.50±0.4^{c}	0.62±0.03^{a}
2	29	5.34±0.3^{c}	372.42±45.2^{b}	5.82±0.3^{b}	0.79±0.01^{c}
3	29	5.74±0.11^{b}	429.82±18^{a}	1.91±0.03^{a}	0.74±0.02^{c}
4	29	5.48±0.1^{a}	391.12±14.9^{b}	1.82±0.03^{a}	0.69±0.02^{b}

*Within columns values with different superscripts are significantly different (P<0.05)

Table 5: Results of temperature stress (24°C) on mortality rates of different treatments in 29- day-old trout

Treatment	1	2	3	4
Mortality%	**66±1^{c}**	**85±1^{b}**	**99.33±1.5^{a}**	**84±1^{b}**

*Within rows values with different superscripts are significantly different (P<0.05)

Table 6: Survival of trout larvae fed different diets during rearing period .Values are mean ± standard deviation (n=3)

Treatment	Survival%			
	Day 1-7	Day 7-21	Day21-29	Day 1-29
1	**86±3.51^{c}**	**87±2.99^{b}**	**90±0.81^{b}**	**67±1^{d}**
2	**96±1.15^{ab}**	**94±2.12^{a}**	**93±3.43^{ab}**	**84±2^{c}**
3	**98±1.15^{a}**	**99±0.64^{a}**	**99±0.6^{a}**	**96±1^{a}**
4	**93±1.52^{b}**	**95±2.77^{a}**	**95±2.77^{ab}**	**88±2.08^{b}**

*Within columns values with different superscripts are significantly different (P<0.05)

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Table 7: Proximate analyses expressed in percent dry weight (mean ±SD) of trout larvae carcass sampled at first week and at the end of the experiment. Within columns values with different superscripts are significantly different (P<0.05)

Treatment	Time(day)	Moisture %	Protein %	Lipid %	Ash %
1	8	81.61±0.10^{a}	65.60±0.25^{d}	12.05±0.56^{a}	7.07±0.40^{d}
2	8	81.02±0.44^{b}	66.99±0.13^{b}	11.3±0.07^{b}	11.3±0.07^{b}
3	8	81.06±0.54^{b}	67.71±0.49^{a}	12.01±0.31^{a}	6.98±0.39^{a}
4	8	81.05±0.18^{b}	67.30±0.15^{c}	11.33±0.46^{b}	6.87±0.54^{c}
1	29	83.61±0.53^{a}	68.70±0.55^{c}	14.64±0.46^{a}	7.31±0.79^{ab}
2	29	83.61±0.53^{a}	70.08±0.77^{a}	13.67±0.30^{d}	7.61±0.29^{ab}
3	29	81.5±0.4^{b}	71.82±0.28^{ab}	14.15±0.14^{c}	7.32±0.55^{a}
4	29	81.25±0.34^{b}	69.25±0.34^{b}	14.36±0.22^{b}	7.39±0.47^{b}

Discussion

Several studies have demonstrated the positive effect of enriched live food on the growth performance of various species. HUFA- enriched *Artemia* nauplii fed to *fenneropenaeus indicus* (Taiebi, 2001), *Sepia oficinalis* (Koueta et al., 2002), and *Chanos chanos* (Gapasin et al.,1998) exhibited better growth and survival. Gilthead sea bream larvae also grow better if fed rotifers enriched with highly unsaturated n-3 HUFA (Mourente et al., 1993). Similar to the finding of Tamaru et al., 1993 and Hosseinpour et al., 2010, in the present study significant differences were found in the growth of 8- days– old trout larvae fed different diets, larvae fed Artemia enriched with HUFA+vitamin C (treatment 3) exhibited significantly higher growth than larvae fed commercial food (treatment1) after 29 days of culture (Table3 and 4). On the other hand, survival of 29 days-old trout fed various diets were significantly different (Table 6) supporting the results of Ako et al., (1994); Gapasin et al., (1998) and Taebi, (2001). In the first week, the *Oncorhynchus mykiss* larvae fed unenriched *Artemia* nauplii, obtained higher growth rates compared to those fed the commercial diet (treatment1) (table3). In our study the proximate composition of *Artemia* nauplii was 61.7% protein; 11.44% lipid and 6.78% ash on a dry weight basis (DW). It may be explained that the higher dietary protein level can meet the requirements of body protein synthesis in early stages and then support fast growth of larvae (Watanabe et al.,1987b). The proteolytic enzymes in *Artemia* may play a significant role in contributing to the digestion process, in addition to digestion brought about by the proteolytic enzymes of the fry itself (Bengeston et al., 1991).

Milkfish larvae given *Artemia* enriched with HUFA+ vitamin C showed better growth and higher survival after a stress test (Gapasin et al., 1998). Ako et al., 1994; Gapasin et al., 1998 observed no or few mortalities among fish fed *Artemia* enriched with menhaden oil (high DHA:

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EPA ratio) compared to high mortalities among fish fed unenriched *Artemia*. Red sea bream (*Pagrus major*) and marble sole *(Euryglossa orientalis*) larvae given diets containing DHA and lecithin tolerated temperature and salinity changes, low oxygen and air exposure better than the larvae given DHA and lecithin- free diets (Kanazawa, 1995). Furuita et al., (1995 a, b) reported that yellowtail larvae and red sea bream juvenile fed *Artemia* enriched with DHA exhibited higher survival in the stress test than those fed *Artemia* enriched with EPA. In the present study, trout larvae fed *Artemia* enriched with HUFA+ vitamin C (treatment3) showed better growth and increased resistance to temperatures than those given unenriched diet (treatment 2) and commercial diet (treatment1). This result is similar to Chanos chanos (Gapasin et al., 1998) Sepia officicinalis (Koueta et al., 2002).

Takeuchi and Watanabe (1987); Takon (1990) reported that a dietary deficiency of the essential fatty acids (EFA) is manifested as poor growth, increased water content of the muscle, high liver lipid content and poor feed efficiency that were similar to our study (Table 7).

In the current investigation, the larvae fed by HUFA+ vitamin C for 1 week exhibited more resistance to temperature stress (Table5), compared to treatment 1. The lowest mortality value consistently occurring in fish treated with HUFA+ vitamin C (treatment 3) may indicate that ascorbic acid supplementation enhanced resistance to stress. When subjected to salinity stress test, 20 day old *Clarias gariepinus* larvae fed ascorbate-supplemented diet exhibited significantly low mortality than those larvae fed an ascorbate free diet (Merchie et al., 1995). Although it is possible that HUFA alone may have improved growth performance in trout (as reported in *chonos chonos* by Gapasin et al., 1998), the synergistic effect of vitamin C cannot be neglected. Better growth was observed among tilapia finger lings (Anadu et al., 1990) and plaice (Rosenlund et al., 1990) fed diets supplemented with ascorbic acid.

The preceding studies attest the importance of EFA and/or vitamin C in fish growth and development. Trout fed with EFA + vitamin C exhibited significantly ($P<0.05$) higher growth than those given unenriched live food after 29 days of culture. When subjected to temperature stress, mortality of the EFA + vitamin C- treated fish was significantly lower ($P<0.05$) among the treatment groups. Optimum requirements of these nutrients in trout, however, are not yet known.

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غنی سازی ناپلئوس آرتمیا با اسیدهای چرب ضروری و ویتامین C:اثر روی لارو قزل آلای رنگین کمان

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چکیده

در این تحقیق اثر ناپلئوس آرتمیای غنی شده با اسیدهای چرب ضروری و ویتامین C روی میزان رشد، بقاء و مقاومت بـه اسـترس دما(دمای بالا)در لاروهای قزل آلای رنگین کمان پروش یافته در حوضچه ها مورد بررسی قرار گرفته شد. لاروهای تازه به تغذیـه افتاده(با متوسط وزن۱۳/۵±۱۲۰/۴۳میلی گرم) روزانه ۶ بار تغذیه شدند. به مدت یک هفته ۴ تیمار هر کدام با سه تکرار کـه تیمـار ۱) غذای آغازین تجاری برای قزل آلای رنگین کمان، تیمار ۲) ناپلئوس آرتمیا تازه تخمه گشایی (غنی نشده) ،تیمار ۳) نـاپلئوس آرتمیای غنی شده با اسیدهای چرب بلند زنجیره + ویتامین Cو تیمـار ۴) ترکیـب ۱۰ درصـد از نـاپلئوس آرتمیـای غنـی شـده بـا اسیدهای چرب بلند زنجیره + ویتامین C به همراه غذای آغازین تجاری ماهی قزل آلا است. بعد از یک هفته ،همه تیمارهای ماهی از غذای تجاری به مدت ۳ هفته استفاده کردند. آنالیز آماری از رشد بعد از یک هفته و پایـان دوره آزمـایش نشـان داد کـه میـزان رشـد در تیمارهـای مختلـف دارای اخـتلاف معنـی دار بـود($P<۰/۰۵$) . بعـد از ۴ هفتـه لاروهـای تیمـار ۳ بـا متوسـط وزن ۵۷/۹۳±۶۵۷/۵۰میلی گرم بیشترین وزن را نشان دادند($P< ۰/۰۵$). بیشترین درصد بقا (۹۶درصد) در تیمار ۳ مشاهده شد. بررسی از ترکیبات تقریبی بدن لارو قزل آلای رنگین کمان در هفته اول نشان داد که میزان پروتئین در لاروهای تیمار ۳ و ۴ اختلاف معنـی دار با بقیه تیمارها داشته است($P< ۰/۰۵$). بهترین نتیجه از میزان مقاومت لارو در مقابل استرس دما (بالای ۲۴ درجه سانتی گـراد) در تیمار ۳ به میزان ۱/۵۲±۹۱/۳۴ درصد مشاهده شد

واژگان کلیدی:اسید های چرب ، ویتامین C، قزل آلای رنگین کمان، استرس، رشد و بقاء

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