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Effects of enriched *Artemia* with n–3 long-chain polyunsaturated fatty acids on growth performance, stress resistance and fatty acid profile of *Litopenaeus vannamei* postlarvae

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Abstract

A 15-day study was conducted to evaluate the effects of *Artemia* metanauplii enriched with two commercial supplements (Easy DHA-Selco and *S. presso*) which contained high levels of n–3 long chain polyunsaturated fatty acids (n–3 LC-PUFA) on growth performance, stress resistance and fatty acid profile of *Litopenaeus vannamei* post larvae (PL). In this regard, PL were fed with three different types of *Artemia* including: (1) newly hatched *Artemia franciscana* nauplii as the control group; (2) *Artemia* metanauplii enriched with Easy DHA-Selco; and (3) *Artemia* metanauplii enriched with *S. presso*. Survival rates did not change among different groups. PL fed *Artemia* enriched with the *S. presso* and the Easy DHA-Selco showed the highest wet and dry weight, respectively (p<0.05). Moreover, PL fed *Artemia* enriched with the commercial emulsions had higher survival rate (~ 10 %) than treatments fed newly hatched *Artemia* (p<0.05).The concentration of n–3 PUFA especially DHA and also n-3 / n-6 PUFA ratios were higher in PL fed with *Artemia* enriched with the commercial emulsions than the control group. Feeding enriched *Artemia* with n–3 LC-PUFA is recommended to improve growth performance in larval stages of *L. vannamei*.

Keywords: *Artemia*, DHA, EPA, Penaidae, Post larvae

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Introduction

The brine shrimp (*Artemia* sp.) is the most convenient and least labor-intensive live food available for the culture of several fish and crustacean species. Because of the ease in production and their suitable biochemical composition, *Artemia* sp. nauplii have resulted in quick and successful developments in the commercial hatchery rearing of several fish and crustacean species (Sorgeloos *et al.*, 1998). Despite the many advantages of *Artemia* sp. nauplii, the concentrations of n–3 long chain polyunsaturated fatty acids (n–3 LC-PUFA) mainly eicosapentaenoic acid (EPA, 20:5n-3) and docosahexaenoic acid (DHA, 22:6n-3) in newly hatched nauplii are extremely low (Sorgeloos *et al*., 1998). In this regard, enrichment techniques, which take advantage of the non-selective feeding habit of *Artemia* sp, with oil emulsions are used for enhancing the n–3 LC-PUFA content of this live food. However, because of high catabolism of n–3 LC-PUFA in *Artemia*, adequate enrichment of these fatty acids is difficult (Evjemo *et al.*, 2001).

Lipids are a source of energy, fat soluble vitamins, cholesterol and essential fatty acids in juvenile and larval crustaceans, and are crucial to elevated growth and molting success (Kattner *et al.*, 2003). Moreover, marine crustaceans have a limited ability to elongate and desaturate n–3 LC-PUFA from their precursor α-linolenic acid (ALA, 18:3n−3), thus these fatty acids should be provided in their diets (Glencross, 2009). It has been reported that, dietary n–3 LC-PUFA has positively affected growth, survival and stress tolerance in the post larval (PL) stage of different crustacean species such as Indian shrimp, *Fenneropenaeus indicus* (Immanuel *et al.*, 2001), black tiger shrimp, *Penaeus monodon* (Immanuel *et al.*, 2004); southern pink shrimp, *Farfantepenaeus paulensis* (Martins *et al.*, 2006), giant freshwater prawn, *Macrobrachium rosenbergii* (Das *et al.*, 2007); spiny lobster, *Panulirus homarus* (Chakraborty *et al.*, 2010) and freshwater crayfish, *Astacus leptodactylus leptodactylus* (Koca *et al.*, 2015). Moreover, it has been reported that n–3 LC-PUFA can influence gill membrane permeability and can modulate osmoregulation in crustacean gill which consequently may affect their survival in a salinity stress test (Palacios and Racotta, 2007).The aim of the present study is to evaluate the effect of feeding enriched *Artemia* metanauplii with the commercial supplements (Easy-DHA and *S. presso*, INVE, Belgium) to *L. vannamei* post larvae (M_1-PL_{12}) on the growth performance, stress resistance and fatty acid profile.

Materials and methods

Experimental setup

This study was carried out in a private shrimp hatchery (Kamal Meygo, Choebdeh, Abadan, Iran) for 15 days. Larvae of *L. vannamei* were obtained from domesticated brood stocks, which were induced by eyestalk ablation to spawn in captivity. Larvae were reared in 10-ton rectangular concrete tanks at 28–30 °C, salinity 30 ppt and a density of 200 nauplii L^{-1}. From Zoea I to Mysis I, larvae were fed solely on *Chaetoceros gracilis* in concentration of 10×10^4 (m L^{-1}). Upon reaching Mysis I, larvae were individually counted and transferred to the

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experimental units. For this purpose nine rectangular 25 liter tanks filled with sand-filtered and UV treated seawater were used, and each tank was stocked with 100 Mysis I L^{-1}, with a daily water exchange of 50 %. Tanks were supplied with constant aeration maintaining oxygen near saturation level (6 ppm). Average values for water temperature, salinity, dissolved oxygen, pH and alkalinity were 28.0±1.5 °C, 30.0±0.2 ‰, 6.0±0.3 mg L^{-1},8.1±0.2 and 136.0±12.5 mg L^{-1} respectively, and photoperiod was 12L:12D (light: darkness).

Artemia

Three treatments with three replicates each were established: (1) Newly hatched *Artemia franciscana* nauplii (control group); (2) *Artemia* metanauplii enriched with Easy DHA-Selco; and (3) *Artemia* metanauplii enriched with *S. presso*. The second instar stage *Artemia* nauplii (*A. franciscana*— Salt lake aquafeed, premium grade, USA) were separated from the hatching container by using a 120-µm sieve and transferred to 10-l enrichment containers at a density of 200 nauplii mL^{-1} of sea water at room temperature (28 °C). Strong aeration was provided to the rearing containers to keep the O_2 at optimum level. The nauplii were enriched with an *in vivo* encapsulation method with the commercial supplements Easy DHA-Selco and *S. presso* (INVE, Belgium) (Table 1) at a daily dose of 0.6 g L^{-1} of sea water as recommended by the manufacturer for 24 h. After 24 h, the enriched *Artemia* were harvested and rinsed with water over a 120-µm sieve to remove any remaining emulsion. Feeding was carried out once a day (11:00) and *Artemia* was offered at an initial density of 2 mL^{-1}, which increased gradually to 10 mL^{-1} at the end of the experiment. Daily before water renewal, the number of remaining *Artemia* was estimated in each experimental unit and the amount of *Artemia* was maintained or increased as needed.

Table 1: Easy DHA-Selco and *S. presso* composition according to the manufacturer (INVE Aquaculture, Belgium)

Ingredients	Easy DHA-Selco	*S. presso*
Moisture (%)	30	58
Crude lipid (%)	67	33
Crude protein (%)	-	3
Crude ash (%)	1	1
Crude fiber (%)	1	0.5
Phosphorous (%)	0.2	0.2
Sodium (%)	0.2	0.2
Calcium (%)	0.1	0.1
Vitamin A (IU kg^{-1})	1500000	110000
Vitamin D_3 (IU kg^{-1})	150000	10000
Vitamin E (mg kg^{-1})	3600	5400
Vitamin C (mg kg^{-1})	800	8000
n–3 LC-PUFA (mg kg^{-1})	200	150
DHA / EPA	2.5	9

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Sampling and stress tests

At the end of the feeding trial, survival was estimated by individually counting the number of PL from each experimental unit. For body weight, all the post larvae were collected, rinsed with freshwater and blotted dry and wet weights (mg post larvae^{-1}) were measured by weighing post larvae with an electronic microbalance. Dry weight was determined using an electro balance (±0.01 mg) by placing 20 post larvae on a pre-weighed microscope slide. The slides were then placed in a laboratory oven at 60°C for 24 h and then reweighed to determine dry weights of post larvae. At the end of the trial, three replicates of 100 PL_{12} from each of the experimental treatments were submitted to salinity and formalin stress tests. For the salinity stress test, PL were transferred to 2000-ml plastic containers, filled with fresh water (0 ppt) or brackish water (15 ppt). For the formalin, stress test PL were transferred to 2000-ml plastic containers filled with seawater that contained 100-ppm formalin. Mortality was monitored at 5-min intervals during 2 h. Shrimp presenting no movement of pleopods and no reaction to mechanical stimuli were considered dead.

Fatty acid analysis

For fatty acid profile assessment, fatty acid methyl esters were prepared by acidic methanolysis of lipid extracts using sulfuric acid in methanol (Christie, 1993). In this regard, the lipid sample (up to 50 mg) was dissolved in 2.5 % sulfuric acid in methanol (2 ml) in a test tube. The mixture was left for 1 h at 80° C, and then the samples were cooled to room temperature. After that, sodium salt buffer (0.9 %) was added to samples and the required esters extracted with n-hexane, using a Pasture pipette to separate the supernatant. The solution was centrifuged (4000 g, 50 min at 4° C) and the supernatant, which contained fatty acid methyl esters was separated, and then evaporated under a stream of nitrogen. Finally, the remaining dry fatty acid methyl esters were dissolved in isooctane (1 ml) and determined by gas chromatography, The fatty acid composition of *Artemia* (n=1) and shrimp post larvae (n=3) were determined by an auto sampler gas chromatography (GC, Agilent technologies 7890 N, USA), equipped with a flame ionization detector (FID) and a cyanopropyl–phenyl capillary column (DB-225MS, 30 m×0.250 mm ID ×0.25µm Film thickness, USA according to Agh *et al.*(2014).

Statistical analyses

Data were analyzed using SPSS ver.15.0 (Chicago, Illinois, USA), and data presented as mean±standard error of the mean. Arcsine transformations were conducted on all data expressed as percentages. One way ANOVA was performed at a significance level of 0.05 following confirmation of normality and homogeneity of variance. Tukey's procedure was used for multiple comparisons when statistical differences were found among groups by the one-way ANOVA.

Results

Growth performance, survival and stress resistance

In the present study, survival rate did not change among different groups, ranging between 30.6 and 33.9% in the control

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group and PL fed with *Artemia* enriched with the Easy DHA-Selco, respectively (p>0.05). PL fed *Artemia* enriched with the *S. presso* had the highest wet weight (Fig. 1a). However, PL fed *Artemia* enriched with the Easy DHA-Selco and the control group showed the highest and the lowest dry weight, respectively (Fig. 1b; p<0.05).

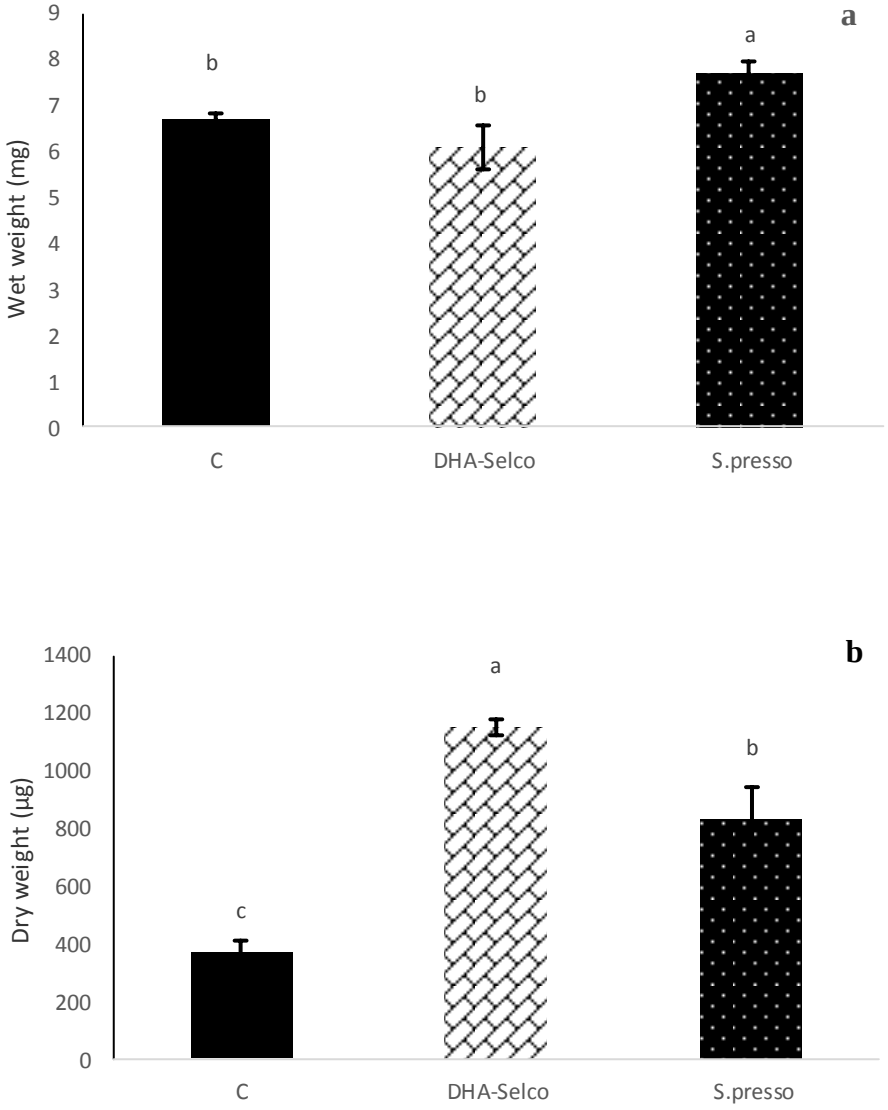

Figure 1: wet weight (a) and dry weight (b) of PL in different experimental groups. A different superscript on the bars denotes statistically significant differences (P < 0.05)

Moreover, survival rate of PL in different experimental groups did not change after exposing to formalin stress test (Fig. 2a) and brackish water stress test (Fig. 2b) and was over 90%. Regarding the freshwater stress test (Fig. 2c) survival rate drastically reduced to 68.67, 59.0 and 60.33% in the control and PL fed *Artemia* enriched with the Easy DHA-Selco and the *S. presso*, respectively. However, PL fed *Artemia* enriched with the commercial emulsions had higher survival rate (~ 10 %) than PL fed newly hatched *Artemia* (p<0.05).

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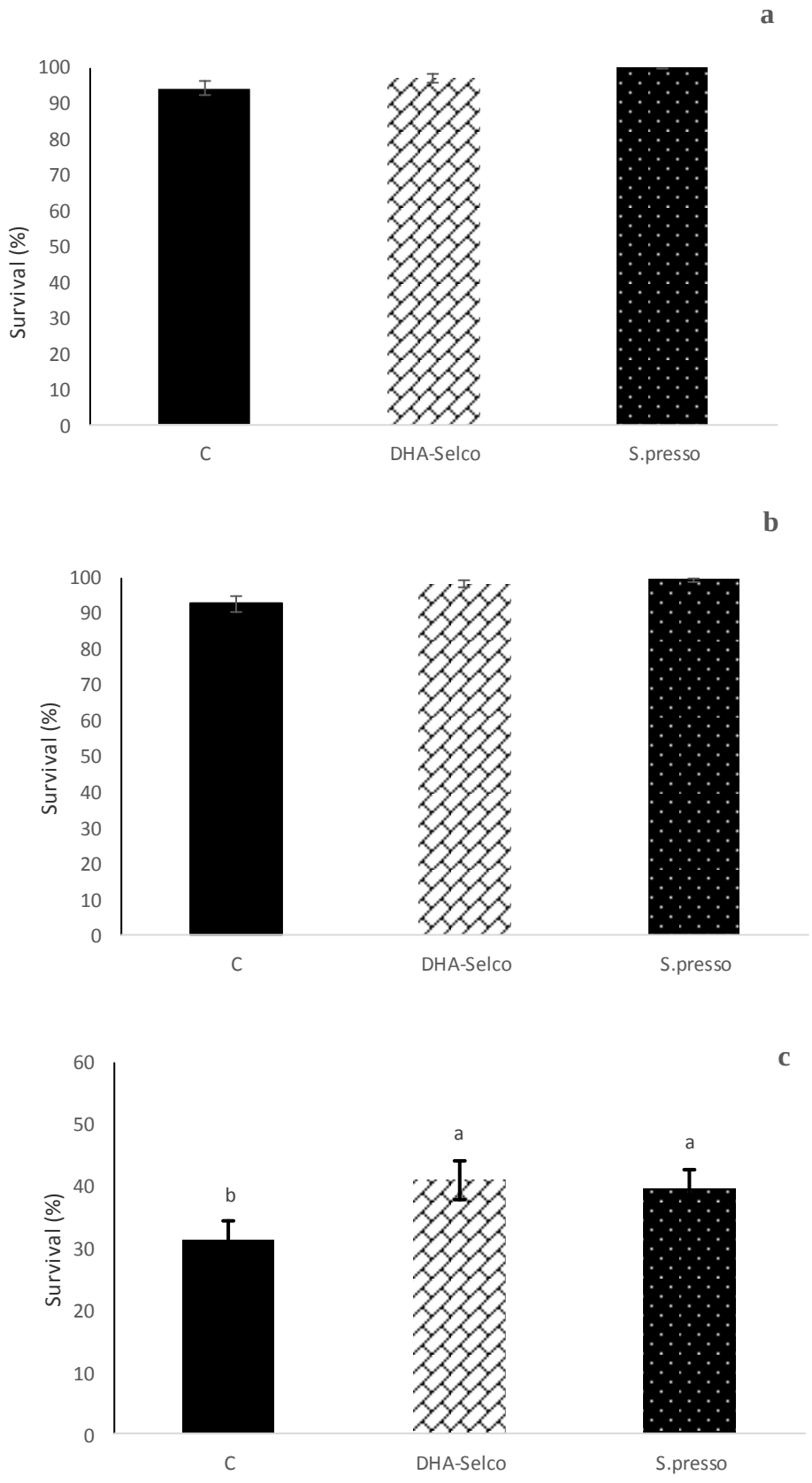

Figure 2: Survival rate (%) after stress tests with formalin (a), brackish water (b) and freshwater (c) of PL in different experimental groups. A different superscript on the bars denotes statistically significant differences ($P < 0.05$).

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Fatty acid profile of enriched Artemia and post larvae

The fatty acid composition of the *Artemia* nauplii is shown in Table 2. *Artemia* enriched with the Easy DHA-Selco had higher lipid content (4%) than other groups. The concentration of ALA sharply decreased in *Artemia* enriched with the commercial emulsions to 24.3% in the newly hatched *Artemia* (control) and 13.9% in *Artemia* enriched with the *S. presso*. However, the level of n–3 LC-PUFA (mainly EPA and DHA) increased in *Artemia* enriched with the commercial emulsions to 3.2% in the control and 13.9% in *Artemia* enriched with the Easy DHA-Selco. The fatty acid profile of PL was significantly affected by the fatty acid profile of enriched *Artemia* with different commercial emulsions (Table 3). PL fed with *Artemia* enriched with the Easy DHA-Selco had higher lipid content than other groups (p<0.05). Total saturated fatty acids (SFA) gradually decreased to 28.8±0.5 in the control and to 24.8±0.3 in PL fed *Artemia* enriched with the Easy DHA-Selco (p<0.05). Total MUFA (mainly oleic acid [OA, 18:1n-9]) was higher in PL fed with *Artemia* enriched with the Easy DHA-Selco than other groups. In addition, the level of n–6 LC-PUFA (mainly arachidonic acid [20:4n-6]) was higher in the control than PL fed with *Artemia* enriched with the commercial emulsions (p<0.05). In contrast, the concentration of n–3 LC-PUFA especially DHA and n-3 / n-6 ratios were higher in PL fed with *Artemia* enriched with the commercial emulsions than the control.

Table 2: Fatty acid profile (%) of *Artemia* enriched with n–3 LC-PUFA (n = 1).

		***Artemia* enriched with**	
Fatty acid profile	**C**	**DHA-Selco**	***S. presso***
Lipid content (%)	0.9	4.0	0.5
14:0	0.1	0.4	1.8
16:0	9.4	12.5	17.2
18:0	8.4	11.3	7.1
20:0	4.1	1.5	0.9
22:0	0.9	0.9	0.2
24:0	0.2	0.3	0.6
SFA	23.1	26.9	27.8
14:1n-5	0.1	0.7	1.2
16:1n-7	1.6	1.3	2.7
18:1n-7	8.6	7.0	6.4
18:1n-9	20.9	13.9	21.1
20:1n-9	0.7	0.4	-
MUFA	31.9	23.3	31.4
18:2n-6	5.8	5.0	7.2
20:2n-6	0.4	6.4	0.2
20:4n-6	0.8	2.1	0.5
n–6 PUFA	7.0	13.5	7.9
18:3n-3	24.3	15.5	13.9
20:3n-3	0.8	1.1	0.2
20:5n-3	2.3	11.2	5.9
22:5n-3	-	0.6	-
22:6n-3	0.1	2.1	7.2
n–3 PUFA	27.5	30.5	27.2
Total	89.5	94.2	94.3
n-3 LC-PUFA	3.2	13.9	13.3
n-3 / n-6	3.9	2.3	3.4

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Table 2 continued:			
EPA / ARA	2.9	5.3	11.8
DHA / ARA	0.1	1.0	14.4
DHA / EPA	0.04	0.2	1.2

Abbreviations: C: *Artemia* without enrichment; SFA: saturated fatty acids; MUFA: monounsaturated fatty acids; LA: linoleic acid; ARA: arachidonic acid; PUFA: polyunsaturated fatty acids; LNA: linolenic acid; EPA: eicosapentaenoic acid; DHA: docosahexaenoic acid; LC-PUFA: long chain-polyunsaturated fatty acids.

Table 3: Fatty acid profile (%) of *Litopenaeus vannamei* post larvae fed with *Artemia enriched* with n–3 LC-PUFA (mean ± SEM, n=3).

		Diets		
Fatty acid profile	***Mysis***	**C**	**DHA-Selco**	**S.presso**
Lipid content (%)	0.4 ± 0.0	0.9 ± 0.1^{b}	2.2 ± 0.4^{a}	1.2 ± 0.2^{ab}
14:0	1.2 ± 0.1	0.6 ± 0.0^{a}	0.5 ± 0.1^{ab}	0.4 ± 0.0^{b}
16:0	17.4 ± 0.3	14.8 ± 0.4^{a}	13.7 ± 0.1^{ab}	14.4 ± 0.0^{b}
18:0	8.0 ± 0.3	11.6 ± 0.2^{a}	8.8 ± 0.1^{c}	10.4 ± 0.2^{b}
20:0	1.3 ± 0.1	0.6 ± 0.0^{c}	1.1 ± 0.0^{a}	0.9 ± 0.1^{b}
22:0	0.2 ± 0.0	0.7 ± 0.0^{a}	0.5 ± 0.0^{b}	0.7 ± 0.1^{a}
24:0	0.3 ± 0.0	0.5 ± 0.0^{a}	0.2 ±0.0^{b}	0.2 ± 0.1^{b}
SFA	28.4 ± 0.5	28.8 ± 0.5^{a}	24.8 ± 0.3^{b}	27.0 ± 0.9^{ab}
14:1n-5	0.7 ± 0.0	0.4 ± 0.0	0.5 ± 0.0	0.5 ± 0.1
16:1n-7	2.4 ± 0.0	0.8 ± 0.0^{b}	1.2 ± 0.0^{a}	0.9 ± 0.1^{b}
18:1n-7	5.7 ± 0.1	7.3 ± 0.3^{a}	5.8 ± 0.3^{b}	5.7 ± 0.3^{b}
18:1n-9	17.6 ± 0.1	14.9 ± 0.8^{b}	18.8 ± 1.0^{a}	14.6 ± 0.9^{b}
MUFA	26.4 ± 0.6	23.4 ± 0.7^{b}	26.3 ± 0.1^{a}	21.7 ± 0.6^{b}
18:2n-6, LA	9.4 ± 0.1	5.3 ± 0.1	5.9 ± 0.3	6.0 ± 0.3
20:2n-6	0.7 ± 0.0	0.7 ± 0.0	0.8 ± 0.0	0.8 ± 0.0
20:4n-6, ARA	1.4 ± 0.1	7.4 ± 0.3^{a}	3.5 ± 0.1^{b}	4.4 ± 0.2^{b}
n–6 PUFA	11.5 ± 0.8	13.4 ± 0.5^{a}	10.2 ± 0.2^{b}	11.2 ± 0.3^{b}
18:3n-3, LNA	14.3 ± 0.5	12.8 ± 0.5^{a}	9.2 ± 0.1^{b}	10.1 ± 0.3^{b}
20:3n-3	0.6 ± 0.0	0.5 ± 0.1^{b}	1.5 ± 0.1^{a}	0.4 ± 0.0^{b}
20:5n-3, EPA	8.9 ± 0.5	13.2 ± 0.1^{a}	10.8 ± 0.2^{b}	11.7 ± 0.4^{b}
22:6n-3, DHA	7.4 ± 0.1	5.3 ± 0.1^{b}	11.8 ± 0.6^{a}	13.3 ± 0.3^{a}
n–3 PUFA	31.2 ± 0.9	31.8 ± 0.8^{b}	33.3 ± 1.0^{a}	35.5 ± 0.6^{a}
Total	97.5 ± 1.1	97.4 ± 1.3	94.6 ± 0.9	95.4 ± 2.0
n-3 LC-PUFA	16.9 ± 0.1	19.0 ± 0.6^{b}	24.1 ± 0.8^{a}	22.2 ± 0.6^{a}
n-3 / n-6	2.7 ± 0.1	1.4 ± 0.1^{b}	3.3 ± 0.2^{a}	3.2 ± 0.1^{a}
EPA / ARA	6.4 ± 0.1	1.8 ± 0.0^{c}	3.1 ± 0.1^{a}	2.7 ± 0.1^{b}
DHA / ARA	5.3 ± 0.5	0.7 ± 0.1^{b}	3.4 ± 0.1^{a}	3.0 ± 0.1^{a}
DHA / EPA	0.8 ± 0.1	0.4 ± 0.0^{b}	1.1 ± 0.1^{a}	1.1 ± 0.1^{a}

Abbreviations: C: *Artemia* without enrichment; SFA: saturated fatty acids; MUFA: monounsaturated fatty acids; LA: linoleic acid; ARA: arachidonic acid; PUFA: polyunsaturated fatty acids; LNA: linolenic acid; EPA: eicosapentaenoic acid; DHA: docosahexaenoic acid; LC-PUFA: long chain-polyunsaturated fatty acids.

Discussion

Several studies in different crustacean species indicated that the main factor affecting the nutritional value of *Artemia* for the larval stage of these species was the content of n–3 LC-PUFA (Coutteau and Mourente, 1997; Martins *et al.,* 2006; Das *et al.,* 2007; Chakraborty *et al.,* 2010; Koca *et al.,* 2015). The results of the current study showed that, despite improved growth performance of PL, the survival was not greatly varied between control and n–3 LC-PUFA enriched diets. In contrast, several studies reported that *Artemia* enriched with n–3 LC-PUFA emulsions increased survival rate in different crustacean species such as *P. monodon* (Rees *et al.,* 1994), crab *Eriocheir sinensis* (Naihong *et al.*, 1999), mysid shrimp, *Mysidopsis almyra*

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(Domingues *et al*., 2001) and the spider crab, *Maja brachydactyla* (Andres *et al*., 2007). In this regard, some studies analyzing different phases of PL development showed the beneficial effects of n–3 LC-PUFA on survival or growth in more advanced stages of PL (Kontara *et al.*, 1997; Wouters *et al.*, 1997). In the present study, PL fed *Artemia* enriched with the *S. presso* and the Easy DHA-Selco had a higher wet and dry weight than PL fed newly hatched *Artemia*. The higher whole body lipid content in PL fed Easy DHA-Selco may have led to the higher dry weight in this group than in the other experimental treatments. The variation in growth of PL between control and enrichment groups may be due to higher levels of n–3 LC-PUFA mainly EPA and DHA in *Artemia* enriched with Easy DHA-Selco and *S.presso*, respectively than newly hatched *Artemia*, which contained high levels of ALA. This result obviously indicated that, n–3 LC-PUFA have more nutritive value than ALA for growth performance of *L. vannamei* post larvae as also reported in other penaeid species (Immanuel *et al.*, 2001, 2004; Martins *et al*., 2006).On the other hand, higher n-3 / n-6 ratio in PL fed *Artemia* enriched with commercial emulsions may result in increasing growth pattern in these groups. In this context, Das *et al.* (2007) reported that *M. rosenbegii* post larvae fed *Moina micrura* enriched with a high n-3 / n-6 ratio lipid emulsion, had better growth performance and survival rates than other groups. Previous studies in different crustacean species revealed that, n–3 LC-PUFA especially DHA is more important than n–6 PUFA for various physiological functions including, survival, growth and metamorphosis (Immanuel *et al.*, 2001, 2004; Martins *et al.*, 2006; Das *et al.*, 2007). In this study, *Artemia* enriched with the Easy DHA-Selco had a higher lipid content than other groups, which may have led to higher lipid content in PL fed this diet and consequently dry weight elevation in this group. Similarly, rapid increase in lipid level of *Artemia* during the enrichment process with lipid emulsions was reported in other studies (Dhert *et al*., 1990; Velazquez, 1996; Hafezieh *et al.*, 2008).

It has been reported that n–3 LC-PUFA can increase tolerance in young stages of crustaceans and fish when exposed to different stress tests such as salinity (Palácios *et al.*, 2004; Palácios and Racotta, 2007), temperature (Chim *et al*., 2001) and total ammonia (Cavalli *et al*., 2000; Martins *et al*., 2006) and physical stress (Ako *et al*., 1994). In the present study, PL fed *Artemia* enriched with commercial emulsions showed higher survival rates than the control group when exposed to freshwater stress test. In this context, Palácios *et al* (2004) demonstrated that, the beneficial effect of n–3 LC-PUFA supplementation in the diet on survival of *L. vannamei* post PL to low salinity stress test is related to modification of fatty acid composition of gills and to a larger gill area which increases the surface of ion transport and the number of Na^+/K^+-ATPase pumps. In fact, n–3 LC-PUFA possibly promotes an increase in the synthesis of new membranes in gills that would result in an increase in the surface, and can incorporate the most suitable fatty acid composition to counteract the effect of salinity changes

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that modify either permeability or functional enzymes such as Na^+/K^+-ATPase and carbonic anhydrase activities (Palácios *et al*., 2004; Palácios and Racotta, 2007).

In the present study, the fatty acid profile of newly hatched *Artemia* showed high concentrations of OA and ALA, but negligible levels of EPA and DHA, which is in line with previous reports (Cavalli *et al.*, 2000; Martins *et al.*, 2006). In addition, the fatty acid composition of the metanauplii reflected to some extent the fatty acid profile of the commercial emulsions used in the enrichment process, which resulted in a drastic increase in n–3 LC-PUFA. In this study, EPA increased from 2.3% in the control group to 11.2 and 5.9% in *Artemia* enriched for 24 h with Easy DHA-Selco and *S. presso*, respectively. Regarding DHA, this fatty acid increased from 0.1% in newly hatched *Artemia* to 2.1 and 7.2% in *Artemia* enriched for 24 h with Easy DHA-Selco and *S. presso*, respectively. In this context, Immanuel *et al.* (2001) reported that in *A. franciscana* nauplii enriched with different levels of lipid the concentration of EPA and DHA were increased considerably from 2.45 to 5.1% and from 0.3 to 1.9%, respectively after 6 h enrichment period. Moreover, Immanuel *et al.* (2004) reported that these two fatty acids increased from 2.68 to 5.43% and 0.53% to 2.23%, respectively in *A. franciscana* nauplii enriched with different levels of lipid after 12-h enrichment period. In addition, higher lipid content in Easy DHA-Selco than *S.presso* led to an increase in *Artemia* total lipid content that consequently increased PL dry weight in this group. In the current study, the trend of total SFA and MUFA in PL whole body to some extent was the same as that of *Artemia* in different groups. PL fed newly hatched *Artemia* had higher levels of ALA than other groups; however, total n–3 LC-PUFA, DHA and n–3 / n–6 ratio were higher in PL fed *Artemia* enriched with the commercial emulsions than the control group. These results agree with previous studies in different crustacean species fed *Artemia* enriched with emulsions containing high levels of n–3 LC-PUFA (Wouters *et al.*, 1997; Immanuel *et al*., 2001, 2004; Palácios *et al*., 2004; Martins *et al.*, 2006; Das *et al*., 2007).

In conclusion, feeding *Artemia* enriched with n–3 LC-PUFA resulted in better growth performance and stress resistance in *L. vannamei* PL. Moreover, n–3 LC-PUFA especially DHA and n–3 / n–6 increased in PL fed *Artemia* enriched with the commercial emulsions which may consequently have positive effects on their physiological functions such as growth performance, osmoregulation and stress resistance.

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