Latin American Journal of Aquatic Research, 47(2): 260-269, 2019
DOI: 10.3856/vol47-issue2-fulltext-6

Research Article

Soybean and linseed oil in replacement of fish oil in diets for female lambari *Astyanax altiparanae* Garutti & Britski, 2000

Marcelo D. Pontes[1], Daniel A.V. Campelo[2], Renato B. Ferraz[3], Jener A.S. Zuanon[3] Wilson M. Furuya[4] & Ana Lucía Salaro[3]

[1]Fundação Instituto de Pesca do Estado do Rio de Janeiro, Rio de Janeiro, RJ, Brasil
[2]Universidade Federal do Pará, Instituto de Estudos Costeiros, Bragança, PA, Brasil
[3]Universidade Federal de Viçosa, Departamento de Biologia Animal, Viçosa, MG, Brasil
[4]Universidade Estadual de Ponta Grossa, Ponta Grossa, PR, Brasil
Corresponding author: Ana Lucia Salaro (salaro@ufv.br)

ABSTRACT. This study aimed to evaluate the effect of different proportions of soybean and linseed oil to replace fish oil in diets for female lambari (*Astyanax altiparanae*). A completely randomized design with seven treatments and five replicates was used. The treatments consisted of a diet containing fish oil and six diets containing different proportions of soybean oil (S) and linseed oil (L), 10S/0L, 8S/2L, 6S/4L, 4S/6L, 2S/8L and 0S/10L. The fish that received the highest proportion of linseed oil (0S/10L) had the lowest crude lipid in their carcasses. The highest linolenic acid (18:3n3) concentrations were detected in the carcasses of fish fed diets with high proportions of linseed oil (4S/6L, 2S/8L, and 0S/10L). The highest n6/n3 ratios corresponded to the carcasses of fish fed diets containing the lowest proportions of linseed oil (10S/0L and 8S/2L). The highest levels of 20:5n3 (eicosapentaenoic acid [EPA]), 22:6n3 (docosahexaenoic acid [DHA]) and ΣEPA+DHA occurred in fish fed diets containing fish oil. Among the fish that received diets with vegetable oils, the ΣEPA+DHA was higher in the fish of the treatments 4S/6L, 2S/8L and 0S/10L. Diets supplemented with a 4S/6L proportion provide adequate deposition of the n3 fatty acids series in the lambari carcass.

Keywords: *Astyanax altiparanae*; vegetable oils; essential fatty acids; arachidonic acid; eicosapentaenoic acid; docosahexaenoic acid

INTRODUCTION

Due to their nutritional quality, fish are of great importance in global food security (Golden *et al*., 2016). In this sense, aquaculture will play an increasing role in the supply of fish to meet the growing worldwide demand (Thilsted *et al*., 2016). However, much of the current aquaculture production is still dependent on fishery products, such as fishmeal and fish oil (Pauly & Zeller, 2017). The use of fishmeal and fish oil to meet the production of food for aquaculture has reached unsustainable proportions (Tacon & Metian, 2015). A good alternative is the use of vegetable oils, due to their wide availability and profitability (Turchini *et al*., 2009). When replacing fish oil by vegetable oils in aquaculture feeds, the profile of fatty acids in the diet should be the central aspect to be considered because there is a strong correlation between the fatty acid profile of fish and the diet provided (Tocher, 2010).

Among the most common vegetable oils used, soybean oil provides competitive prices, high availability in the world market and is rich in linoleic acid (18:2n6; Ng & Wang, 2011). Unlike fish oil, however, it is deficient in fatty acids of the n3 series (Caballero *et al*., 2002). Linseed oil, in contrast to most vegetable oils, contains more than 50% linolenic acid (18:3n3) (Bell *et al*., 2003; Ng & Wang, 2011) but it is still produced on a smaller scale and is, therefore, more expensive compared to soybean oil and other vegetable lipid sources. The use of a soy and linseed oils mixture would be an alternative to fish oil.

Corresponding editor: Mariel Gullian

Although vegetable oils do not present long-chain polyunsaturated fatty acids (PUFAs), some species of tropical freshwater fish can synthesize arachidonic acid (ARA; 20:4n-6) from 18:2n6, and eicosapentaenoic acid (EPA; 20:5n-3) and docosahexaenoic acid (DHA; 22:6n-3) from 18:3n3 (Paulino *et al*., 2018). The synthesis of ARA, EPA, and DHA is performed by desaturation and elongation reactions, catalyzed by the enzymes, Δ5 and Δ6 desaturases (Tocher, 2003, 2010). As this does not result in the predominant formation of one fatty acid over another, adequate amounts of the precursor fatty acids must be supplied in the fish's diet to achieve an appropriate 18:2n6/18:3n3 relationship.

Alteration of the dietary fatty acid profile may influence the lipid metabolism of fish, which may lead to alterations in growth performance, carcass fatty acid profile and proximate composition (Bell *et al.*, 2002; Asdari *et al.*, 2011; Castro *et al.*, 2016). Therefore, it is essential to take into account the growth and body composition when evaluating the alteration of a lipid source of the diet.

Fish of the genus *Astyanax* live in freshwater environments of tropical and subtropical regions of South America (Menezes *et al*., 2007). Already, more than 100 species of this genus, popularly known as lambari *Astyanax altiparanae*, have been described (Lima *et al*., 2003). Lambari is an economically valuable fish, with great potential for culture and their flesh is highly appreciated by the consumer (Salaro *et al*., 2015). These fish are also small, in nature reaches a weight of up to 60 g, with omnivorous feeding habits and good acceptance of processed diets, have a short production cycle, reaching sexual maturity around four months in captivity (Porto-Foresti *et al*., 2010). Lambari has apparent sexual dimorphism, where the male copulatory organ manifests a rough anal fin due to the presence of spicules (Porto-Foresti *et al*., 2010), which facilitate reproduction. This sexual dimorphism enables gender classification and the use of only the females for finishing since they have a higher growth rate than the males.

Due to its small size, rapid growth, short life cycle and ease of production in captivity, this species can be used as an experimental model for the development of new production technologies. Therefore, it is possible to conduce the experiments with adults lambari in an easier and less costly way than large fish species. An example is the recent development of thermal shock techniques can produce sterile triploid females (Adamov *et al*., 2016), promoting growth gain and carcass yield (Nascimento *et al*., 2017).

Lambari is quite popular as live bait for sports fishing and as a snack, and also have a great potential for commercialization as canned food (Dutra *et al*., 2012). For human consumption, these fish have a slaughter weight of 5 to 10 g, and typically, the whole carcass is consumed, removing only the scales and viscera, but still with skeleton and head, which makes the high carcass yield of this species an advantage.

Farmed lambaris presented a PUFA-rich tissue, even fed the commercial low-content ARA, EPA, and DHA, suggesting an ability of these fish to produce these fatty acids from their precursors (Gonçalves *et al*., 2012). EPA levels in farmed *A. bimaculatus* are high relative to other tropical freshwater fish, evidencing the ability of fish to synthesize and accumulate PUFA of this genus (Furuya *et al.*, 2013). However, due to the fatty acid profile of the diets, or the natural food, the n6 content is high concerning the n3 content (Gonçalves *et al*., 2012; Furuya *et al*., 2013). In this sense, the manipulation of the lambari diet could produce a PUFA-rich snack. Feeding lambari with a diet with the appropriate ratio of 18: 2n6/18: 3n3 could maximize PUFA synthesis and fish production for consumption rich in biologically active fatty acids without the need for fish oil. Therefore, this study aimed to evaluate the effect of different proportions of soybean and linseed oil to replace fish oil in diets for female adult lambari *A. altiparanae*, by evaluating the growth performance, proximate carcass composition, and fatty acid profiles.

MATERIALS AND METHODS

This experiment was approved by the Ethics Committee of the Department of Animal Science of the Federal University of Viçosa, Brazil (protocol N°20/2011).

Experimental design and experimental diets

A completely randomized design with seven treatments and five replicates was used. The treatments consisted of a diet containing fish oil and six diets containing different proportions of soybean oil (S) and linseed oil (L): 10S/0L, 8S/2L, 6S/4L, 4S/6L, 2S/8L and 0S/10L. All experimental diets were formulated to be isoenergetic (4372.86 ± 17.73 kJ kg^{-1}) and isonitrogenous (319.82 ± 5.99 g kg^{-1}). The diets were ground in a hammer mill (0.8 mm sieve), manually mixed and pelleted in an electric meat grinder. The pellets were then dried in a forced-air oven at 50°C for 24 h, crushed in a manual mill and manually passed through granulometric sieves (Tecnal, Piracicaba, SP, Brazil) to obtain pellets of 1.0 mm. Tables 1 and 2 provide the formulation, proximate composition and fatty acids analyses of the diets, respectively.

Fish and culture conditions

Female adult lambari with 4.87 ± 0.63 g (mean weight ± SD) were weighed on a precision scale with an accu-

Table 1. Formulation and proximate composition of the experimental diets. [a]Assurance levels per kilogram of product: Vit. A min-2,500,000 UI; Vit. D3 min-600,000 UI; Vit. E min-37,500 UI; Vit. K3 min-3,750 mg; Vit. C min-50,000 mg; Vit. B1 min-4,000 mg; Vit. B2 min-4,000 mg; Vit. B6 min-4,000 mg; Vit. B12 min-4,000 mg; niacin min-22,500 mg; biotin min-15 mg; Ac. Folic min-1,250 mg. [b]Assurance levels per kilogram of product: calcium pantothenate min-12,000 mg; Cu min-2,500 mg; Co min-125 mg: Fe min-15 g; I min-375 mg; Mg min-12.5 g; Se min-87.5 mg; Zn min-12.5 g. [c]Values determined at the Laboratory of Animal Science of Federal University of Viçosa, MG, Brazil. FO: fish oil, S: soybean oil, L: linseed oil.

Ingredient (g kg^{-1})	FO	10S/0L	8S/2L	6S/4L	4S/6L	2S/8L	0S/10L
Soybean meal	535.0	535.0	535.0	535.0	535.0	535.0	535.0
Corn gluten meal	70.0	70.0	70.0	70.0	70.0	70.0	70.0
Cornmeal	140.5	140.5	140.5	140.5	140.5	140.5	140.5
Wheat bran	145.0	145.0	145.0	145.0	145.0	145.0	145.0
L-lysine	3.3	3.3	3.3	3.3	3.3	3.3	3.3
DL-methionine	3.0	3.0	3.0	3.0	3.0	3.0	3.0
Dicalcium phosphate	38.0	38.0	38.0	38.0	38.0	38.0	38.0
Salt	2.5	2.5	2.5	2.5	2.5	2.5	2.5
Vitamin mix[a]	1.5	1.5	1.5	1.5	1.5	1.5	1.5
Mineral mix[b]	1.0	1.0	1.0	1.0	1.0	1.0	1.0
BHT	0.2	0.2	0.2	0.2	0.2	0.2	0.2
Soybean oil	0.0	60.0	48.0	36.0	24.0	12.0	0.0
Linseed oil	0.0	0.0	12.0	24.0	36.0	48.0	60.0
Fish oil	60.0	0.0	0.0	0.0	0.0	0.0	0.0
Proximate composition							
Crude protein (g kg^{-1})	324.9	322.82	314.03	320.48	311.35	317.12	328.04
Crude lipid (g kg^{-1})	88.02	88.46	92.19	88.28	93.22	89.42	93.00
Ash (g kg^{-1})	85.86	88.48	87.99	79.16	86.95	85.46	85.24
Gross energy (MJ kg^{-1})	18.31	18.23	18.30	18.41	18.24	18.26	18.40

Table 2. Fatty acid composition (g kg^{-1}of total identified fatty acids) of the experimental diets. FO: fish oil, S: soybean oil, L: linseed oil, ΣSFA: sum of saturated fatty acids, ΣMUFA: sum of monounsaturated fatty acids, ΣPUFA: sum of polyunsaturated fatty acids, Σn6: sum of the n6 fatty acids series, Σn3: sum of the n3 fatty acids series, n6/n3: ratios of the n6 and n3 fatty acids series.

Fatty acid	Experimental diets						
	FO	10S/0L	8S/2L	6S/4L	4S/6L	2S/8L	0S/10L
14:0	2.5	0.1	0.0	0.1	0.1	0.0	0.0
16:0	15.9	11.1	10.3	9.5	8.4	5.9	4.7
18:0	2.6	3.5	3.2	0.3	3.4	2.6	2.3
22:0	0.0	0.5	0.6	0.0	0.0	0.0	0.1
14:1	0.5	0.0	0.4	0.6	0.4	0.2	0.3
16:1	4.8	0.1	0.1	0.1	0.1	0.1	0.1
18:1n9	16.1	26.9	23.4	22.2	23.1	16.4	13.6
20:1	2.0	0.3	0.3	0.2	0.0	0.0	0.0
18:2n6	13.5	49.9	43.6	39.5	20.4	22.6	16.8
18:3n3	1.9	5.7	13.4	18.4	29.8	29.4	32.2
20:4n6 (ARA)	0.9	0.0	0.0	0.0	0.0	0.0	0.0
20:5n3 (EPA)	6.4	0.0	0.0	0.0	0.0	0.0	0.0
22:6n3 (DHA)	19.1	0.2	0.0	0.0	0.0	0.0	0.1
ΣSFA	21.8	15.9	14.6	10.3	12.3	8.8	7.4
ΣMUFA	23.5	27.3	24.2	23.1	23.6	16.7	14.0
ΣPUFA	41.9	55.9	57.4	58.1	50.2	52.1	49.2
Σn6	14.5	50.0	43.9	39.7	20.4	22.7	16.9
Σn3	27.4	5.9	13.4	18.4	29.8	29.4	32.3
n6/n3	0.53	8.47	3.28	2.16	0.68	0.77	0.52

racy of 0.01 g (model MB45, Toledo®, Brazil). The fish were then randomly distributed into 35 aquaria (60 L water) at a density of 27 fish per aquarium, with each aquarium being an experimental unit. The aquaria were maintained in a recirculation system with a mechanical and biological filter and a UV sterilization lamp. The water temperature (27°C), dissolved oxygen (6.5-7.5 mg L^{-1}), pH 6.5-6.8 and total ammonia (0.00-0.03 mg L^{-1}) were maintained throughout the experimental period. Water parameters were measured with a multi-parameter meter (model HI 9828, Hanna Instruments, Barueri, SP, Brazil). The photoperiod was adjusted to 12 h by fluorescent lamps (60 W).

Refuges of PVC pipe refuges were placed at the bottom of the aquarium To ensure the welfare of fish during the experiment, and floating shelters made with nylon lines (similar to roots of aquatic macrophytes) were installed to ensure the welfare of fish during the experiment.

The fish were fed manually four times daily until satiation, for 90 days. The average feed intake was 8.10 ± 0.42 g ind^{-1}. The aquaria were weekly siphoned for water renewal and removal of feces, exchanging 1/3 of the water volume.

Growth performance

At the end of the experiment period, all fish were slaughtered with a lethal dose of benzocaine (100 mg L^{-1}), counted and weighed on a precision scale (accuracy of 0.01 g) to evaluate growth performance by survival rate (SR = (final number of fish / initial number of fish) × 100), weight gain (WG = final mean biomass - initial mean biomass), feed intake (FI = grams of feed offered / number of fish), feed conversion ratio (FCR = dry feed intake / wet weight gain), specific growth rate (SGR = (ln final weight - ln initial weight) / days) × 100), and carcass yield (CY = (eviscerated fish weight / whole fish weight) × 100). The carcass was considered to be a fish without scales and viscera.

Chemical analysis and fatty acid profile of diets and carcass

The diets and carcass chemical composition analyses (dry matter, ash, crude protein, crude lipid, and gross energy content) were determined according to the AOAC (2000), in the Laboratory of Food Analysis, Department of Animal Science of the Federal University of Viçosa, Brazil. The fish were first lyophilized and ground in a ball mill. The moisture was analyzed by oven drying at 110°C until constant weight. The ash was obtained by incinerating the samples in a muffle furnace at 600°C for 3 h. For crude protein, the Kjeldahl method (N×6.25) was used. Assessment of crude lipid was based on the procedure of Folch *et al.* (1957). Gross energy was measured by burning the samples in a bomb calorimeter.

To ascertain the fatty acid profile of the carcasses, fish from each aquarium were pooled and ground in a blender, homogenised and subjected to lipid extraction with a mixture of chloroform, methanol and water (2:2:1.8 v/v/v), according to the Bligh & Dyer (1959) method. The methyl esters of fatty acids were prepared as described by Hartman & Lago (1973) and separated by a gas chromatograph (model GC15-A, Shimadzu) equipped with a flame ionization detector and a fused silica capillary column (320×30m×0.32 mm; Omegawax). The gas fluxes (White Martins) were 1.2 mL min^{-1} for the carrier gas (H_2), 40 mL min^{-1} for the make-up gas (N_2), and 30 and 300 mL min^{-1} for H_2 and the synthetic air flame, respectively. The split ratio of the sample was 1:100. For separation of fatty acids, the injection point was set at 240°C and the detector at 250°C. The initial column temperature was programmed at 165°C, held for 12 min and increased from 165 to 235°C at a rate of 5°C min^{-1} and kept at 235°C for 9 min. Fatty acid identification was made by comparing the relative retention times of the sample peaks and a mixture of fatty acid methyl ester standards and methyl esters containing linoleic acid geometric isomers, *c*9, *t*11 and *t*10, *c*12 (189-19 and O5632, respectively; Sigma), and spiking of the sample with standards. Triplicate injections of 2 μL were done. This methodology was also used in the analysis of the fatty acid profile of experimental diets. The fatty acid profiles of the experimental diets and the carcasses were undertaken at the CBO Laboratory Analysis, Campinas, Brazil.

In experimental diets and carcass, it was considered only the fatty acids that yielded values above 0.5 g kg $diet^{-1}$ at least in one treatment. In experimental diets, it was also presented the sum of saturated fatty acids, the sum of monounsaturated fatty acids, the sum of polyunsaturated fatty acids, the sum of the n6 fatty acids series, the sum of the n3 fatty acids series, and ratios of the n6 and n3 fatty acids series (Table 2). In the carcass it were also calculated the sum of fatty acids 20:5n3 (EPA) and 22:6n3 (DHA), sum of saturated fatty acids, sum of monounsaturated fatty acids, sum of polyunsaturated fatty acids, sum of the n6 fatty acids series, sum of the n3 fatty acids series, and the ratios of the n6 and n3 fatty acids series.

Statistical analysis

The data of productive performance, carcass chemical composition and fatty acid profile were evaluated by a Lilliefors test to verify the assumption of normality of errors, and a Bartlett test to confirm the homogeneity of variances. Afterward, a one-way analysis of variance

(ANOVA) was performed, followed by Tukey's test. All tests used SPSS 10.0 for Windows software (SPSS Inc., Michigan Avenue, Chicago, IL, USA) at 5% significance.

RESULTS

Growth performance

There were no significant differences ($P > 0.05$) in the productive performance parameters of fish fed the different experimental diets (Table 3).

Carcass proximate composition

The fish that received the highest proportion of linseed oil (0S/10L) had lower crude lipid in the carcasses compared to the fish fed diets containing fish oil and the highest proportions of soybean oil (10S/0L, 8S/2L, and 6S/4L). However, the crude lipid content of the fish supplied the diet 0S/10L did not differ from those fed diets 4S/6L and 2S/8L (Table 4).

Carcass fatty acid profiles

Among the treatments, fish fed diets containing fish oil had higher concentrations of myristic (14:0) and palmitoleic (16:1) fatty acids ($P < 0.05$). The same effect was observed for margaric acid (17:0), except for those animals fed the 0S/10L diet, which did not differ from the fish fed diets containing fish oil (Table 5).

The highest linolenic acid (18:3n3) concentrations were recorded in the carcasses of fish fed diets with relatively higher proportions of linseed oil (4S/6L, 2S/8L, and 0S/10L). The lowest Σn3 fatty acids were detected in animals that received the lowest proportions of linseed oil concentration (10S/0L and 8S/2L). However, the Σn3 fatty acids in fish fed with the diets 4S/6L, 2S/8L, and 0S/10L did not differ from those fish fed the 6S/4L and fish oil diets. The highest n6/n3 ratios existed in the carcasses of fish fed diets containing the lowest proportions of linseed oil (10S/0L and 8S/2L), and the lowest values occurred in animals receiving the highest proportions of linseed oils (4S/6L, 2S/8L, and 0S/10L) and the fish fed with fish oil (Table 5).

The highest levels of 20:5n3 (EPA), 22:6n3 (DHA) and ΣEPA+DHA were observed in fish fed diets containing fish oil. Among the fish that received diets with vegetable oils, the higher values for ΣEPA+DHA were found in the fish of the treatments 4S/6L, 2S/8L and 0S/10L (Table 5). The Σn6 fatty acids did not differ among the fish of different treatments. There were no significant differences in ARA contents in the fish carcasses among the different treatments (Table 5).

DISCUSSION

Few studies evaluated the effect of different proportions of vegetable oil to replace fish oil in diets for adult fish, because conducting experiments with fish in this phase it is difficult and costly, ones most of the adult fish produced have a high final weight. Although the reduced growth rates presented by female adult lambari, due to the high energy cost of the sexual maturation processes (Minte-Vera *et al*., 2016), the lack of differences in productive performance parameters indicates that this species efficiently use vegetable oils as a source of fatty acids and energy. Similarly, in previous studies, the use of different vegetable lipid sources did not affect the growth performance of several other species, such as the Nile tilapia *Oreochromis niloticus* (Ng *et al*., 2013), panga *Pangasius hypophthalmus* (Asdari *et al*., 2011), Murray cod *Maccullochella peelii peelii* (Turchini *et al*., 2011) and Florida pompano *Trachinotus carolinus* (Rombenso *et al*., 2016).

The comparatively lower content of lipids in the carcasses of the fish fed with the highest ratios of linseed oil can be related to the fatty acid profile of the diet, since this may affect the synthesis and deposition of various fatty acids in the fish tissues. The use of various sources of vegetable lipids can cause different effects on lipid deposition in different fish species (Bell *et al*., 2002). The lipid source of the diet can regulate gene expression and the activity of enzymes involved in lipid metabolism, influencing the increase or reduction of lipid content in different tissues (Castro *et al*., 2016).

The lower lipid content in the carcasses of the fish fed the highest proportions of linseed oil may be related to the alteration in the fatty acid profile of the diet. The lipid source of the diet can regulate gene expression and the activity of enzymes involved in lipid metabolism, influencing the increase or reduction of lipid content in different tissues (Castro *et al*., 2016). The oxidation or deposition in tissues may be selective, depending on each fatty acid and its function in the organism (Bell *et al*., 2001). The replacement of fish oil by soybean oil increased the deposition of lipids in the liver and muscle of turbot *Scophthalmus maximus* L., and this increase may be associated with alterations in the expression of genes involved in the synthesis and oxidation of fatty acids (Peng *et al*., 2014). Replacement of 50% fish oil by palm oil (Bell *et al*., 2002) or canola oil (Bell *et al*., 2001) caused a reduction in the muscle lipid content of salmon *Salmo salar*. In contrast, the replacement of fish oil by vegetable oils did not affect the lipid content in the muscle of European sea bass *Dicentrarchus labrax* (Montero *et*

Table 3. Growth performance of lambari *Astyanax altiparanae* females fed diets with fish oil and different proportions of soybean and linseed oils. FO: fish oil, S: soybean oil, L: linseed oil. Data are presented as means (standard deviation). Ns: not significant by analysis of variance, F test ($P > 0.05$).

Growth performance	Treatments						
	FO	10S/0L	8S/2L	6S/4L	4S/6L	2S/8L	0S/10L
WG^{ns} (g)	3.03	3.27	3.29	3.12	3.15	3.35	3.50
	(0.28)	(0.23)	(0.36)	(0.23)	(0.17)	(0.24)	(0.48)
FI^{ns} (g ind^{-1})	7.81	8.25	7.99	8.18	8.02	8.09	8.36
	(0.29)	(0.11)	(0.23)	(0.33)	(0.44)	(0.57)	(0.50)
FCR^{ns}	2.59	2.53	2.45	2.63	2.55	2.41	2.41
	(0.17)	(0.18)	(0.29)	(0.22)	(0.22)	(0.15)	(0.19)
CY^{ns} (%)	74.58	71.29	73.27	73.72	75.05	73.94	73.44
	(0.98)	(1.92)	(1.11)	(0.95)	(2.23)	(1.16)	(1.23)
SGR^{ns} (% d^{-1})	0.61	0.66	0.66	0.63	0.63	0.66	0.68
	(0.05)	(0.07)	(0.08)	(0.05)	(0.03)	(0.04)	(0.08)
SR^{ns} (%)	97.78	100.00	98.52	95.56	97.04	97.78	98.52
	(3.31)	(0.00)	(2.03)	(8.03)	(1.66)	(3.31)	(2.03)

Table 4. Carcass proximate composition (g kg^{-1} dry matter) of lambari *Astyanax altiparanae* females fed diets with fish oil and different proportions of soybean and linseed oils. FO: fish oil; S: soybean oil, L: linseed oil, ns: not significant by analysis of variance, F test ($P > 0.05$). *Means (standard deviation) on the lines followed by different letters show significant variation by Tukey test at 5% probability.

Proximate composition	Treatments						
	FO	10S/0L	8S/2L	6S/4L	4S/6L	2S/8L	0S/10L
Dry matter^{ns}	262.64	265.93	261.85	262.46	260.36	273.50	262.42
	(13.71)	(9.02)	(3.22)	(6.60)	(5.55)	(20.93)	(8.73)
Ash^{ns}	147.47	155.54	152.72	149.51	142.95	141.50	145.06
	(7.16)	(12.88)	(8.56)	(7.89)	(6.72)	(8.56)	(16.49)
Crude lipid*	218.65^{a}	238.58^{a}	206.09^{a}	207.80^{a}	193.74^{ab}	185.66^{ab}	148.03^{b}
	(26.50)	(27.39)	(30.57)	(31.70)	(24.39)	(17.19)	(19.74)
Crude protein^{ns}	572.59	550.97	555.53	564.79	544.90	547.46	540.23
	(50.37)	(16.37)	(24.77)	(28.38)	(16.83)	(41.08)	(16.60)

al., 2005), surubim *Pseudoplatystoma corruscans* (Martino *et al*., 2002), panga *P. hypophthalmus* (Asdari *et al*., 2011) and tilapia *Oreochromis* sp. (Ng *et al*., 2013), making clear that the use of different lipid source can cause different effects on lipid deposition in different fish species (Bell *et al*., 2002).

Fish fed diets with the highest levels of n3 fatty acids, as well as the highest n6/n3 ratio, presented these same relationships in their carcass fatty acid profile. For the same species, a similar effect was observed when lambari were fed with conjugated linoleic acid and showed efficient incorporation of this fatty acid into carcasses (Campelo *et al*., 2014). These results corroborated those found in other fish species, confirming that the fatty acid profile of fish has a high correlation with the fatty acid profile of the diet consumed (Regost *et al*., 2003; Asdari *et al*., 2011; Zakeri *et al*., 2011).

The competition among the enzymes responsible for the synthesis of ARA and EPA (Ling *et al*., 2006) can be a contributing factor to the absence of ARA in the carcasses of the fish. Lambari can synthesize PUFAs in muscle tissue from the supply of 18:3n3 and 18:2n6 in the diet (Campelo *et al*., 2015). In the synthesis of ARA and EPA, the precursor fatty acids are substrates for the same enzyme, which has a higher affinity for n3 fatty acids series (Tocher, 2010), leading to greater accumulation of EPA instead of ARA (Glencross, 2009).

Another point that may have contributed to the lack of difference in n6 fatty acids and the absence of ARA in fish carcasses of all treatments may be the differen-

Table 5. Carcass fatty acid profile (g kg^{-1} of total identified fatty acids) of lambari *Astyanax altiparanae* females fed diets with fish oil and different proportions of soybean and linseed oils. FO: fish oil, S: soybean oil, L: linseed oil. *Means (standard deviation) on the lines followed by different letters show significant variation by Tukey test at 5% probability. ΣEPA-DHA: sum of fatty acids 20:5n3 (EPA) and 22:6n3 (DHA); ΣSFA: sum of saturated fatty acids; ΣMUFA: sum of monounsaturated fatty acids; ΣPUFA: sum of polyunsaturated fatty acids; Σn6: sum of the n6 fatty acids series; Σn3: sum of the n3 fatty acids series; n6/n3: ratios of the n6 and n3 fatty acids series.

Fatty acid	Experimental diets						
	FO	10S/0L	8S/2L	6S/4L	4S/6L	2S/8L	0S/10L
14:0*	1.24^{a}	0.26^{b}	0.30^{b}	0.34^{b}	0.21^{b}	0.27^{b}	0.39^{b}
	(0.09)	(0.04)	(0.11)	(0.06)	(0.04)	(0.03)	(0.13)
16:0	17.52	13.34	16.11	15.83	11.00	13.00	15.66
	(1.40)	(1.44)	(4.41)	(2.30)	(0.31)	(0.38)	(3.37)
17:0*	0.29^{a}	0.17^{b}	0.18^{b}	0.18^{b}	0.13^{b}	0.17^{b}	0.21^{ab}
	(0.01)	(0.05)	(0.04)	(0.02)	(0.03)	(0.04)	(0.05)
18:0	4.87	4.89	5.89	6.22	4.65	5.42	6.30
	(0.48)	(0.46)	(1.53)	(0.70)	(0.40)	(0.33)	(1.17)
16:1*	3.24^{a}	0.78^{b}	0.86^{b}	1.01^{b}	0.76^{b}	0.94^{b}	1.18^{b}
	(0.43)	(0.16)	(0.25)	(0.14)	(0.09)	(0.15)	(0.32)
18:1n9	23.90	24.45	28.73	28.34	22.04	25.95	27.56
	(3.80)	(2.53)	(6.78)	(1.38)	(0.74)	(1.53)	(2.61)
18:2n6	10.22	15.88	12.14	11.75	15.40	15.39	10.52
	(3.78)	(3.37)	(4.78)	(7.22)	(2.03)	(0.64)	(5.74)
20:2	0.51	0.47	0.40	0.39	0.51	0.65	0.75
	(0.32)	(0.34)	(0.15)	(0.18)	(0.03)	(0.05)	(0.07)
18:3n3*	0.54^{b}	0.59^{b}	0.83^{b}	1.78^{b}	5.44^{a}	5.92^{a}	5.38^{a}
	(0.40)	(0.33)	(0.92)	(2.09)	(1.16)	(0.48)	(4.37)
20:5n3*	0.39^{a}	0.07^{c}	0.08^{c}	0.08^{c}	0.12^{c}	0.07^{c}	0.22^{b}
	(0.04)	(0.03)	(0.04)	(0.02)	(0.05)	(0.03)	(0.05)
22:6n3*	2.68^{a}	0.22^{c}	0.18^{c}	0.19^{c}	1.13^{b}	0.92^{b}	0.89^{b}
	(0.18)	(0.04)	(0.03)	(0.05)	(0.04)	(0.04)	(0.22)
∑EPA- DHA	3.07^{a}	0.29^{c}	0.25^{c}	0.26^{c}	1.24^{b}	0.99^{b}	1.12^{b}
	(0.22)	(0.06)	(0.01)	(0.06)	(0.02)	(0.05)	(0.26)
∑SFA	24.81	19.52	23.41	23.37	16.71	19.63	23.36
	(2.13)	(1.79)	(6.13)	(3.03)	(0.78)	(0.76)	(4.78)
∑MUFA	28.29	25.73	30.44	30.21	23.20	27.41	29.63
	(4.60)	(2.85)	(7.50)	(1.70)	(0.79)	(1.74)	(3.33)
∑PUFA	14.77	17.93	14.09	14.69	23.56	23.81	18.51
	(4.74)	(4.00)	(6.13)	(9.99)	(3.07)	(1.14)	(10.60)
∑n6	10.62	16.56	12.57	12.19	16.20	16.08	11.02
	(3.99)	(3.70)	(5.09)	(7.63)	(2.17)	(0.68)	(6.03)
∑n3*	3.64^{ab}	0.90^{b}	1.12^{b}	2.11^{ab}	6.86^{a}	7.08^{a}	6.74^{a}
	(0.46)	(0.29)	(0.93)	(2.18)	(1.19)	(0.51)	(4.66)
n6/n3*	3.33^{a}	18.87^{a}	16.55^{a}	9.44^{b}	2.39^{c}	2.27^{c}	1.89^{c}
	(0.07)	(2.01)	(0.28)	(1.28)	(0.35)	(0.07)	(0.56)

tiated synthesis and accumulation of fatty acids in some tissues (Castro *et al*., 2016). As only females were used in this study, it is possible that there was a greater deposition of fatty acids in the gonads of the fish rather than the muscles, as observed by Gonçalves *et al.* (2012) in females of the same species, even in fish fed diets with low levels of ARA. In the European eel *Anguilla anguilla*, females were found to have increased accumulation of ARA in the gonads relative to other tissues (Stottrup *et al*., 2013). The same was observed in trout *Oncorhynchus mykiss*, with a large accumulation of ARA in the ovary during the final maturation phase (Ribeiro *et al*., 2012). Both ARA and the other n6 fatty acids series may have accumulated in the reproductive tissues of lambari because ARA is the precursor of eicosanoids, which act on the control of ovulation (Bruce *et al*., 1999; Astuarino *et al*., 2001).

Although fish fed diets containing fish oil presented the highest levels of EPA and DHA, the presence of these fatty acids in the carcasses of fish fed with different proportions of soybean oil and linseed oil confirms the ability of lambari to synthesize EPA and DHA, since their precursors are included in the diet. The lack of a significant effect on the productive performance of the fish indicates that the vegetable oils used met the nutritional requirements for essential fatty acids. It is likely that under conditions of challenge, the supply of vegetable oils may not be sufficient to meet the nutritional needs of the fish since the PUFAs are precursors of the synthesis of eicosanoids, leukotrienes, and prostaglandins (Montero *et al*., 2003), which can alleviate stress responses in fish (Ganga *et al*., 2011).

Even if the synthesis of EPA and DHA is less in fish fed with vegetable oils, the fish oil replacement for vegetable oils (with higher proportions of linseed oil) may represent improvements in the carcass quality of lambari, for human consumption. Thus, to promote a higher deposition of EPA, a 0S/10L ratio is indicated, whereas, a 4S/6L ratio is recommended for DHA and ΣEPA+DHA since linseed oil and soybean oil have greater availability in the market with a more stable lipid profile and are less expensive than fish oil.

Among the fish fed with vegetable lipid sources, the animals fed diets containing higher levels of linseed oil had the largest Σn3 fatty acids in the carcasses. The intake of fish rich in n3 fatty acids series is beneficial in the prevention and treatment of coronary heart disease, reduces the risk of immune dysfunction and contributes to normal brain development (Simopoulos, 2008). Consequently, it is highly attractive to offer the consumer a fish with an abundance of the n3 fatty acids series (Karakatsouli, 2012). These results demonstrate that lambari can be fed diets consisting exclusively of plant origin products, provided they contain linseed oil, to produce carcasses with a beneficial fatty acids profile for human nutrition.

The possibility of producing a fish rich in the n3 fatty acids series using vegetable oils contributes to the sustainability of aquaculture chain by eliminating the dependence on fish oil and products coming from extraction fishing, which can have their supply compromised due to the seasonality of the products.

ACKNOWLEDGMENTS

We would like to thank the National Council of Technological and Scientific Development (CNPq), Ministry of Science and Technology (MCT) and the Ministry of Fisheries and Aquaculture (MPA), for the award of the scholarship of the announcement EditalMCT/CNPq/CTAgronegócio/MPANº25/2010, Human Resources Training in Fisheries and Aquaculture.

REFERENCES

Adamov, N.S.D.M., Nascimento, N.F.D., Maciel, M., Pereira-Santos, E.C.S., Senhorini, J.A., Calado, L.L., Evangelista, M.M., Nakaghi, L.S.O., Guerrero, A.H.M., Fujimoto, T. & Yasui, G.S. 2016. Triploid induction in the yellowtail tetra, *Astyanax altiparanae*, using temperature shock: tools for conservation and aquaculture. Journal of the World Aquaculture Society, 48(5): 741-750.

Asdari, R., Aliyui-Paiko, M., Hashim, R. & Ramachadram, S. 2011. Effects of different dietary lipid sources in the diet for *Pangasius hypophthalmus* juvenile on growth performance, nutrient utilization, body indices, and muscle and liver fatty acid composition. Aquaculture Nutrition, 17: 44-53.

Astuarino, J.F., Sorbera, L.A., Carrillo, M., Zanuy, S., Ramos, J., Navarro, J.C. & Bromage, N. 2001. Reproductive performance in male European sea bass fed two PUFA-enriched experimental diets: a comparison with females fed a wet diet. Aquaculture, 194: 173-190.

Bell, J.G., McEvoy, J., Tocher, D.R., McGhee, F., Campbell, P.J. & Sargent, J.R. 2001. Replacement of fish oil with rapeseed oil in diets of Atlantic salmon (*Salmo salar*) affects tissue lipid compositions and hepatocyte fatty acid metabolism. Journal of Nutrition, 131: 1535-1543.

Bell, J.G., Henderson, R.J., Tocher, D.R., Mcghee, F., Dick, J.R., Porter, A., Smullen, R. & Sargent, J.R. 2002. Substituting fish oil with crude palm oil in the diet of Atlantic salmon (*Salmo salar*) affects tissue fatty acid compositions and hepatic fatty acid metabolism. Journal of Nutrition, 132: 222-230.

Bell, J.G., Mcghee, F., Campbell, P.J. & Sargent, J.R. 2003. Rapeseed oil as an alternative to marine fish oil in diets of post-smolt Atlantic salmon (*Salmo salar*): changes in flesh fatty acid composition and effectiveness of subsequent fish oil "wash out". Aquaculture, 218: 515-528.

Bligh, E.G. & Dyer, W.J. 1959. A rapid method of total lipid extraction and purification. Canadian Journal of Biochemistry and Physiology, 37: 911-917.

Bruce, M., Oyen, F., Bell, G., Asturiano, J.F., Farndale, B., Carrillo, M., Zanuy, S., Ramos, J. & Bromage, N. 1999. Development of broodstock diets for the European sea bass (*Dicentrarchus labrax*) with particular emphasis on the importance of n-3 and n-6 highly unsaturated fatty acid to reproductive performance. Aquaculture, 177: 85-97.

Caballero, M.J., Obach, A., Rosenlund, G., Montero, D., Gisvold, M. & Izquierdo, M.S. 2002. Impact of

different dietary lipid sources on growth, lipid digestibility, tissue fatty acid composition and histology of rainbow trout, *Oncorhynchus mykiss*. Aquaculture, 214: 253-271.

Campelo, D.A.V., Oliveira, K.R.B., Batiston, W.P., Zuanon, J.A.S., Furuya, W.M., Matsushita, M. & Salaro, A.L. 2014. Conjugated linoleic acid in diets for lambari (*Astyanax altiparanae*) (Garutti & Britski, 2000). Aquaculture Nutrition, 21(6): 788-796.

Castro, C., Corraze, G., Firmino-Diógenes, A., Larroquet, L., Panserat, S. & Oliva-Teles, A. 2016. Regulation of glucose and lipid metabolism by dietary carbohydrate levels and lipid sources in gilthead sea bream juveniles. British Journal of Nutrition, 116(1): 19-34.

Dutra, F.M., Machado, W.J., Caetano, M.S. & Gobbo, A.A. 2012. Avaliação sensorial do processamento em conserva, utilizando-se as espécies: tilápia (*Oreochromis niloticus*), lambari (*Astianax* spp.) e pacu (*Piaractus mesopotamicus*). Revista Brasileira de Produtos Agroindustriais, 14: 239-244.

Furuya, V.R.B., Furuya, W.M., Michelato, M., Salaro, A.L., Matushita, M. & Batiston, W.P. 2013. Composição proximal e perfil de ácidos graxos do lambari-do-rabo-vermelho ("*Astyanax fasciatus*") de diferentes classes de peso. Revista Brasileira de Saúde e Produção Animal, 14(4): 820-830.

Ganga, R., Montero, D., Bell, J.G., Atalah, E., Ganuza, E., Vega-Orellana, O. & Izquierdo, M. 2011. Stress response in sea bream (*Sparus aurata*) held under crowded conditions and fed diets containing linseed and/or soybean oil. Aquaculture, 311: 215-223.

Glencross, B.D. 2009. Exploring the nutritional demand for essential fatty acids by aquaculture species. Reviews in Aquaculture, 1: 71-124.

Golden, C.D., Allison, E.H., Cheung, W.W., Dey, M.M., Halpern, B.S., Mccauley, D.J. & Myers, S.S. 2016. Fall in fish catch threatens human health. Nature, 534(7607): 317-320.

Gonçalves, L.U., Parisi, J., Bonelli, A., Sussel, F.R. & Viegas, E.M.M. 2012. The fatty acid compositions of the total, neutral and polar lipids in wild and farmed lambari (*Astyanax altiparanae*) (Garutti & Britski, 2000) broodstock. Aquaculture Research, 45: 195-203.

Hartman, L. & Lago, R.C.A. 1973. Rapid preparation of fatty acid methyl ester from lipids. Laboratory Practice, 22: 475-473.

Karakatsouli, N. 2012. An overview of the use of fatty acids in fish farming research during the last decade, with particular emphasis on fish quality. Journal of the World Aquaculture Society, 43: 291-320.

Lima, F.C.T., Malabarba, L.R., Buckup, P.A., Silva, J.F.P., Vari, R.P., Harold, A., Benine, R., Oyakawa O.T., Pavanelli, C.S., Menezes, N.A., Lucena, C.A.S., Malabarba, M.C.S.L., Lucena, Z.M.S., Reis R.E., Langeani, F., Cassati, L., Bertaco, V.A., Moreira, C. & Lucinda, P.H.F. 2003. Genera incertae sedis in Characidae. In: Reis, R.E., Kullander, S.O. & Ferraris, C. (Eds.). Check list of the freshwater fishes of South and Central America. Edipucrs, Porto Alegre, pp. 106-169.

Ling, S., Kuah, M.K., Tengku, T.S., Kolkovski, S. & Shu-Chien, A.C. 2006. Effect of dietary HUFA on reproductive performance, tissue fatty acid profile and desaturase and elongase mRNAs in female swordtail *Xiphophorus helleri*. Aquaculture, 261: 204-214.

Matsushita, M., Justi, K.C., das Graças, P.R., Milinsk, M.C., Hayashi, C., Marques, S.T.G., Visentainer, J.V. & de Souza, N.E. 2006. Influence of diets enriched with different vegetable oils on the performance and fatty acid profile of Nile tilapia (*Oreochromis niloticus*) fingerlings. Acta Scientiarum Technology, 28: 125-131.

Menezes, N.A., Weitzman, S.H., Oyakawa, O.T., de Lima, F.C.T, Castro, R.M.C. & Weitzman, M.J. 2007. Peixes de água doce da Mata Atlântica: lista preliminar das espécies e comentários sobre conservação de peixes de água doce neotropicais. Museu de Zoologia da Universidade de São Paulo, São Paulo.

Menoyo, D., Lopez-Bote, C.J., Bautista, J.M., Obach, A. 2003. Growth, digestibility and fatty acid utilization in large Atlantic salmon (*Salmo salar*) fed varying levels of n-3 and saturated fatty acids. Aquaculture, 225(1-4): 295-307.

Minte-Vera, C.V., Maunder, M.N., Casselman, J.M. & Campana, S.E. 2016. Growth functions that incorporate the cost of reproduction. Fisheries Research, 180: 31-44.

Montero, D., Robaina, L., Caballero, M.J., Ginés, R. & Izquierdo, M.S. 2005. Growth, feed utilization and flesh quality of European sea bass (*Dicentrarchus labrax*) fed diets containing vegetable oils: a time-course study on the effect of a re-feeding period with a 100% fish oil diet. Aquaculture, 248: 121-134.

Montero, D., Kalinowskia, T., Obach A., Robaina, L., Tort, L., Caballero, M.J. & Izquierdo. M.S.J. 2003. Vegetable lipid sources for gilthead seabream (*Sparus aurata*): effects on fish health. Aquaculture, 225: 353-370.

Mourente, G., Good, J.E. & Bell, J.G. 2005. Partial substitution of fish oil with rapeseed, linseed and olive oils in diets for European sea bass (*Dicentrarchus labrax* L.): effects on flesh fatty acid composition, plasma prostaglandins E2 and F2α, immune function and effectiveness of a fish oil finishing diet. Aquaculture Nutrition, 11: 25-40.

Nascimento, N.F., Pereira-Santos, M., Piva, L.H., Manzini, B., Fujimoto, T., Senhorini, J.A., Yasui, G.S.

& Nakaghi, L.S.O. 2017. Growth, fatty acid composition, and reproductive parameters of diploid and triploid yellowtail tetra *Astyanax altiparanae*. Aquaculture, 471: 163-171.

Ng, W.K. & Wang, Y. 2011. Inclusion of crude palm oil in the broodstock diets of female Nile tilapia, *Oreochromis niloticus*, resulted in enhanced reproductive performance compared to broodfish fed diets with added fish oil or linseed oil. Aquaculture, 314: 122-131.

Ng, W.K., Chong, C.Y., Wang, Y. & Romano, N. 2013. Effects of dietary fish and vegetable oils on the growth, tissue fatty acid composition, oxidative stability and vitamin E content of red hybrid tilapia and efficacy of using fish oil finishing diets. Aquaculture, 372: 97-110.

Paulino, R.R., Pereira, R.T., Fontes, T.V., Oliva-Teles, A., Peres, H., Carneiro, D.J. & Rosa, P.V. 2018. Optimal dietary linoleic acid to linolenic acid ratio improved fatty acid profile of the juvenile tambaqui (*Colossoma macropomum*). Aquaculture, 488: 9-16.

Pauly, D. & Zeller, D. 2017. Comments on FAO's state of world fisheries and aquaculture (SOFIA 2016). Marine Policy, 77: 176-181.

Peng, M., Xu, W., Mai, K., Zhou, H., Zhang, Y., Liufu, Z., Zhang, K. & Ai, Q. 2014. Growth performance, lipid deposition and hepatic lipid metabolism-related gene expression in juvenile turbot (*Scophthalmus maximus* L.) fed diets with various fish oil substitution levels by soybean oil. Aquaculture, 433: 442-449.

Porto-Foresti, F., Castilho-Almeida, R.B., Senhorini, J.A. & Foresti, F. 2010. Biologia e criação do lambari-do-rabo-amarelo (*Astyanax altiparanae*). In: Baldisserotto, B. & Gomes, L.C. (Eds.). Espécies nativas para piscicultura no Brasil. Editora UFSM, Santa Maria.

Regost, C., Arzel, J., Robin, J., Rosenlund, G. & Kaushik, S.J. 2003. Total replacement of fish oil by soybean or linseed oil with a return to fish oil in turbot (*Psetta maxima*): growth performance, flesh fatty acid profile, and lipid metabolism. Aquaculture, 217: 465-482.

Ribeiro, C.S., Gomes, A.D., Vieira, V.A.R.O., Tabata, Y.A., Takahashi, N.S. & Moreira, R.G. 2012. The effect of ploidy on the fatty acid profile during the reproductive cycle of female rainbow trout (*Oncorhynchus mykiss*). Aquaculture International, 20: 1117-1137.

Received: 30 July 2018; Accepted: 30 October 2018

Rombenso, A.N., Trushenski, J.T. & Schwarz, M.H. 2016. Fish oil replacement in feeds for juvenile Florida pompano: composition of alternative lipid influences degree of tissue fatty acid profile distortion. Aquaculture, 458: 177-186.

Salaro, A.L., Campelo, D.A.V., Pontes, M.D., Zuanon, J.A.S., Furuya, V.R.B. & Furuya, W.M. 2015. Avanços na nutrição e produção de lambaris. In: Tavares-Dias, M. & Mariano, W. (Eds.). Aquicultura no Brasil: novas perspectivas. Vol. 2. Editora Pedro & João Editores, São Carlos.

Simopoulos, A.P. 2008. The importance of the omega-6/omega-3 fatty acid ratio in cardiovascular disease and other chronic diseases. Experimental Biology and Medicine, 233(6): 674-688.

Stottrup, J.G., Jacobsen, C., Tomkiewicz, J. & Jarlbaek, H. 2013. Modification of essential fatty acid composition in broodstock of cultured European eel *Anguilla anguilla* L. Aquaculture Nutrition, 19: 172-185.

Tacon, A.G. & Metian, M. 2015. Feed matters: satisfying the feed demand of aquaculture. Reviews in Fisheries Science & Aquaculture, 23(1): 1-10.

Thilsted, S.H., Thorne-Lyman, A., Webb, P., Bogard, J.R., Subasinghe, R., Phillips, M.J. & Allison, E.H. 2016. Sustaining healthy diets: The role of capture fisheries and aquaculture for improving nutrition in the post-2015 era. Food Policy, 61: 126-131.

Tocher, D.R. 2003. Metabolism and functions of lipids and fatty acids in teleost fish. Reviews in Fisheries Science, 11: 107-184.

Tocher, D.R., 2010. Fatty acid requirements in the ontogeny of marine and freshwater fish. Aquaculture Research, 41: 717-732.

Turchini, G.M., Torstensen, B.E. & Ng, W.K. 2009. Fish oil replacement in finfish nutrition. Reviews in Aquaculture, 1: 10-57.

Turchini, G.M., Francis D.S., Senadheera, S.P.S.D., Thanuthong, T. & De Silva, S.S. 2011. Fish oil replacement with different vegetable oils in Murray cod: evidence of an "omega-3 sparing effect" by other dietary fatty acids. Aquaculture, 315(3): 250-259.

Zakeri, M., Kochanian, P., Marammazi, J.G., Yavari, V., Savari, A. & Haghi, M. 2011. Effects of dietary n-3 HUFA concentrations on spawning performance and fatty acids composition of broodstock, eggs, and larvae in yellowfin sea bream, *Acanthopagrus latus*. Aquaculture, 310: 388-394.