



Long-term feeding of vegetable oils to Senegalese sole until market size: Effects on growth and flesh quality. Recovery of fatty acid profiles by a fish oil finishing diet



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ABSTRACT

The present study evaluates the long term (5 months) effects of feeding vegetable oil-based diets to Senegalese sole (*Solea senegalensis*) until market size. Extruded isonitrogenous and isoenergetic diets (56% crude protein and 20 kJ/g) were formulated to substitute 0 (CTRL), 50 (VO50) and 100% (VO100) of FO by a VO blend (rapeseed oil, RO, soybean oil, SO, and linseed oil, LO). A concomitant replacement of 50% FM and FO (VO50PP50) by plant protein sources (pea, soybean meal, potato concentrate, corn and wheat gluten) and oils was also evaluated. After the growth trial (140 days), fish were fed a fish oil-based diet (CTRL) over a period of 26 days and growth, flesh quality and organoleptic properties were determined.

Results show that it seems possible to substitute up to 100% of FO by a VO blend, as well as concomitantly substituting 50% FO and FM by vegetable sources in on-growing Senegalese sole diets, without compromising growth performance and feed utilization. This species selectively retains highly unsaturated fatty acids (ARA and DHA) and seems to adapt well to a low dietary supply of these FAs. At the end of the growth out experiment fish fed VO50 diet showed a similar $n-3$ HUFA profile to the CTRL fed fish. Total FO substitution resulted in a strong reduction of muscle EPA content that was totally recovered after 26 days of re-feeding with a FO based diet. Despite the observed changes in fatty acid composition, sole fillets from fish fed 100% VO are very well accepted by fish consumers and still are good nutritional value end-products for human consumption, providing 1.5 times the RDI level (0.4 g per 100 g of muscle) of EPA + DHA.

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1. Introduction

Fish oil (FO) is the main lipid source in fish diets due to its high level of $n-3$ highly unsaturated fatty acids (HUFA), particularly docosahexaenoic acid (22:6 $n-3$, DHA) and eicosapentaenoic acid (20:5 $n-3$, EPA), which are known to be essential for optimal growth and health of farmed fish. Additionally, fish are the main dietary source of DHA and EPA for humans, with terrestrial sources showing negligible amounts (Turchini et al., 2011). These fatty acids (FA) are extremely important in the prevention and management of type 2 diabetes, cardiovascular and inflammatory diseases in humans (Ruxton, 2004; Simopoulos, 2005; Von Schacky, 2006).

The decrease in FO and fishmeal (FM) production since the mid 1990s was mainly due to dwindling fisheries resources. However, demand continues to increase, especially impelled by aquaculture industry expansion (average annual growth rate of 6.6%) (FAO, 2012), resulting in increased feed cost and, consequently, marginal profits. These facts impose the usage of alternative lipid sources in fish diets. Some of the most promising alternative candidates are vegetable oils (VO) due to their higher availability, and lower and steadier prices than FO (Turchini et al., 2009).

Vegetable oil's fatty acid composition differs from FO because they are devoid of $n-3$ HUFA and show very high content of specific fatty acids or FA classes: soybean oil (SO) is very rich in linoleic acid (18:2 $n-6$, LOA), rapeseed oil (RO) in monounsaturated fatty acids (MUFA), particularly oleic acid (18:1 $n-9$, OA) (Dubois et al., 2007), whereas linseed oil (LO) has a very high content of α -linolenic acid (18:3 $n-3$, ALA). Thus, it is possible to obtain partially balanced VO based diets by fulfilling individual fatty acid deficiency through the blending of different

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oils (Turchini et al., 2009). VO blends are a practical way to offset differences between VO and FO fatty acid composition. Nevertheless, FO replacement is a challenging practice since vegetable sources lack n – 3 HUFA and marine species show a very limited ability to produce these long chain fatty acids from its precursor, ALA (Sargent, 2002; Sargent and Tacon, 1999).

Senegalese sole is a high value marine flatfish species. In recent years information on its nutritional needs has been attained (Borges et al., 2009; Rema et al., 2008; Silva et al., 2010), making this species a promising candidate for large-scale commercial farming. Senegalese sole ability to deal with diets in which the marine derived ingredients were replaced by vegetable sources was recently demonstrated both in juveniles and market-size sole (Benítez-Dorta et al., 2013; Borges et al., 2014; Cabral et al., 2011, 2013). Furthermore, in Senegalese sole juveniles it seems possible to totally replace supplemental FO by either LO (Benítez-Dorta et al., 2013) or by a VO blend of RO, LO and SO (Borges et al., 2014), without any detrimental effects on growth performance or feed efficiency. In salmonids it was also possible to replace 100% FO by VOs without compromising growth or feed efficiency (Bell et al., 2003, 2004; Torstensen et al., 2005), but in marine carnivorous species such as gilthead seabream (Benedito-Palos et al., 2009; Izquierdo et al., 2005) and European seabass (Mourete and Bell, 2006; Richard et al., 2006) this replacement was only feasible at lower FO substitution levels (up to 60%). In Atlantic halibut Martins et al. (2011) succeeded substituting 70% of FO by LO. Although in Senegalese sole total replacement of dietary supplemental FO by a VO blend could be achieved (Borges et al., 2014), it must be kept in mind the high contribution of FM ($\approx 70\%$) to total dietary fat. The formulation of practical diets able to fulfil this species nutritional needs, together with the low lipid tolerance of this species (Borges et al., 2009, Borges et al., 2013), results in practical constraints concerning the dietary inclusion of higher lipid levels from vegetable sources. Responding to recent trends from the aquafeed industry, higher substitutions of FO in practical diets for Senegalese sole can only be possible if defatted protein sources or if a concomitant replacement of FM and FO are considered.

Vegetable ingredients currently used to substitute FM and FO in fish diets lack n – 3 HUFA. This decrease in dietary essential FA is reflected in muscle fatty acid profile (Benedito-Palos et al., 2009; Fountoulaki et al., 2009; Regost et al., 2003a). However, in Senegalese sole juveniles fed VO diets, the muscle level of DHA was similar to that observed in FO fed fish even when FO was totally substituted by VO (Benítez-Dorta et al., 2013; Borges et al., 2014). Moreover, depending on the species, it is possible to replenish essential fatty acid (EFA) levels in muscle by the usage of finishing diets formulated with 100% FO, or at least partially recover the fatty acid profile to maintain sensory characteristics and fillet nutritional value for human consumption (Izquierdo et al., 2005; Montero et al., 2005; Regost et al., 2003a, 2003b).

The present study focused on the long term (5 months) effect of feeding vegetable oil-based diets to Senegalese sole until market size on growth, flesh quality and organoleptic properties. In order to achieve this purpose, diets were formulated to substitute 50% and 100% of FO by a VO blend during the grow-out period. A concomitant replacement of 50% FM and FO by plant protein sources and oils was also tested to evaluate a lower contribution of dietary marine sources to total dietary fat. After the growth trial (140 days), fish were fed a fish oil-based diet (CTRL) over a period of 26 days to determine the evolution of flesh fatty acid profile and sensory characteristics.

2. Materials and methods

The current study was conducted under the supervision of an accredited expert in laboratory animal science by the Portuguese Veterinary Authority (1005/92, DGV-Portugal, following FELASA category C recommendations), following the European Economic Community animal experimentation guidelines, Directive 2010/63/EU.

2.1. Experimental diets

Feed ingredients and the proximate composition of the dietary treatments are presented in Table 1. Four extruded isonitrogenous and isoenergetic diets (56% crude protein and 20 kJ/g on a dry matter basis) were formulated. Control diet (CTRL) containing 2.5% of supplemental FO was compared with diets where 50% (VO50) and 100% (VO100) FO was replaced by a VO blend. The blend is a mixture of three vegetable oils (15% RO; 20% SO; 50% LO) widely available in the market and commonly used by the feed industry. Finally, diet VO50PP50 with a concomitant replacement of 50% FO by the VO blend referred above and 50% FM by a mixture of plant protein sources (pea, soycomil PC, potato protein concentrate, corn and wheat gluten) was formulated without major changes in dietary protein and fat content. All diets contained hydrolysed protein concentrate (CPSP) and 5% of squid meal to improve palatability. Diet VO50PP50 was supplemented

Table 1
Feed ingredients and proximate composition of the dietary treatments.

	Dietary treatments			
	CTRL	VO50	VO100	VO50PP50
<i>Feed ingredients (%)</i>				
Fishmeal 70 LT ^a	24.50	24.50	24.50	8.00
Fishmeal 60 ^b	27.00	27.00	27.00	13.00
CPSP ^c	5.00	5.00	5.00	5.00
Squid meal	5.00	5.00	5.00	5.00
Pea	–	–	–	11.50
Soycomil PC ^d	–	–	–	4.00
Soybean meal ^e	12.50	12.50	12.50	9.80
Potato concentrate	–	–	–	2.50
Wheat gluten	–	–	–	4.30
Corn gluten	–	–	–	7.50
Aquatex G2000 ^f	11.00	11.00	11.00	8.90
Wheat meal	10.00	10.00	10.00	8.80
Fish oil	2.50	1.25	–	3.00
Rapeseed oil	–	0.37	0.75	0.90
Soybean oil	–	0.25	0.50	0.60
Linseed oil	–	0.63	1.25	1.50
Soy lecithin	0.50	0.50	0.50	0.50
Vit ^g & Min ^h Premix	1.00	1.00	1.00	1.00
Di-calcium phosphate	–	–	–	2.50
L-Lysine	–	–	–	0.50
DL-Methionine	–	–	–	0.20
Binder ⁱ	1.00	1.00	1.00	1.00
<i>Proximate composition</i>				
Dry matter (DM, %)	94.85	94.82	94.92	93.54
Ash (% DM)	14.35	14.39	14.26	11.12
Crude protein (% DM)	55.55	56.05	55.18	55.66
Crude fat (% DM)	8.42	8.54	8.71	10.21
Total phosphorus (% DM)	1.88	1.87	1.91	1.49
Gross energy (kJ/g DM)	19.70	19.97	20.06	21.04

^a Peruvian fishmeal LT (71% crude protein, 11% crude fat, EXALMAR, Peru).

^b Fair Average Quality (FAQ) fishmeal (62% crude protein, 12% crude fat, COFACO, Portugal).

^c Soluble fish protein hydrolysate (87% crude protein, 6.5% crude fat, Sopropêche, France).

^d Soycomil-P (soy protein concentrate, 65% crude protein, 0.7% crude fat, ADM, The Netherlands).

^e Dehulled solvent extracted soybean meal (micronized).

^f Aquatex G2000 (Dehulled, grinded pea grits, 24% crude protein, 0.4% crude fat, SOTEXPRO, France).

^g Vitamins (mg, mcg or IU/kg diet): Vitamin A (retinyl acetate), 2–0 UI; vitamin D3 (DL-cholecalciferol), 2000 UI; vitamin E (Lutavit E50), 100 mg; vitamin K3 (menadiolone sodium bisulfite), 25 mg; vitamin B1 (thiamine hydrochloride), 30 mg; vitamin B2 (riboflavin), 30 mg; calcium pantothenate, 100 mg; nicotinic acid, 200 mg; vitamin B6 (pyridoxine hydrochloride), 20 mg; vitamin B9 (folic acid), 15 mg; vitamin B12 (cyanocobalamin), 100 mcg; vitamin H (biotin), 3000 mcg; vitamin C (Lutavit C35), 1000 mg; inositol, 500 mg; colin chloride, 1000 mg; and betaine (Betafin S1), 500 mg.

^h Minerals (mg or %/kg diet): Co (cobalt carbonate), 0.65 mg; Cu (cupric sulphate), 9 mg; Fe (iron sulphate), 6 mg; I (potassium iodide), 0.5 mg; Mn (manganese oxide), 9.6 mg; Se (sodium selenite), 0.01 mg; Zn (zinc sulphate) 7.5 mg; Ca (calcium carbonate), 18.6%; KCl, 2.41%; NaCl, 4.0%.

ⁱ Diatomaceous earth: Kielseguhr: LIGRANA GmbH, Germany.

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