



Stabilization of linseed oil with vitamin E, butylated hydroxytoluene and lipid encapsulation affects fillet lipid composition and sensory characteristics when fed to rainbow trout

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ARTICLE INFO

Article history:

Received 8 July 2010

Received in revised form 13 July 2011

Accepted 21 July 2011

Keywords:

Rainbow trout

Fatty acids

Linseed oil

Encapsulation

Antioxidants

Sensory analysis

ABSTRACT

An experiment was conducted to determine the effect of adding antioxidants to or encapsulating linseed oil (LO) on its oxidative stability and the growth, fatty acid (FA) composition and sensory properties of rainbow trout. Four pelleted diets differing only in lipid source were prepared: fish oil (FO), LO, LO stabilized with vitamin E (7.5 g/kg) and butylated hydroxytoluene (12.5 g/kg) (stabilized LO) and the stabilized LO encapsulated in hydrogenated palm oil (630 g/kg) (encapsulated LO). The oxidative stability index of the FO and LO diets after 168 days of storage in sealed containers at room temperature were both 0.00 h, whereas those of the stabilized LO and encapsulated LO diets were 9.20 and 11.40 h, respectively. The diets were fed to rainbow trout (22 fish/tank; 7 replicates/treatment; initial weight 31 g) for 168 days. The fish were then euthanized and fillets were analysed for FA composition, malonaldehyde (MDA), colour and sensory attributes. There were no significant differences among treatments on growth performance or MDA levels of fillets. Fish fed FO had higher *a** and *b** levels on the Hunter L, *a*, *b* scale than fish fed any of the LO diets ($P < 0.05$). The total *n*-3 polyunsaturated FA (PUFA) content of trout fed LO was higher ($P < 0.05$) than those fed FO (355 g/kg of total FA vs. 276 g/kg) or encapsulated LO diets (289 g/kg). There were no significant differences in fillet eicosapentaenoic acid or docosahexaenoic acid concentrations due to dietary oil. Trained panelists determined that the aroma intensity of the trout fed FO was higher ($P < 0.05$) than those fed stabilized LO and that fish fed FO had a higher rancid aroma and flavour intensity than those fed the other diets ($P < 0.05$). Fillets from fish fed stabilized LO had higher ($P < 0.05$) aroma desirability and overall acceptability than those from fish fed the FO diet. Consumer panelists found no significant differences between the sensory attributes of fish fed the four diets and exhibited no preference among treatments. Addition of antioxidants to or encapsulation of LO improves its oxidative stability during storage and processing and results in fish fillets with FA composition and sensory characteristics equal or superior to fish fed FO.

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Abbreviations: ALA, Alpha linolenic acid, 18:3n-3; DHA, Docosahexaenoic acid, 20:5n-3; EPA, Eicosapentaenoic acid, 22:6n-3; FA, Fatty acid; FO, Fish oil; LA, Linoleic acid, 18:2n-6; LO, Linseed oil; MDA, Malonaldehyde; OSI, Oxidative stability index; PUFA, *n*-3 Polyunsaturated FA; TBARS, Thiobarbituric acid reactive substances.

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1. Introduction

The world's demand for fish oil (FO) currently exceeds supplies and as aquaculture production is expected to grow at an annual rate of 6.5% until 2025, a replacement for FO in aquafeeds must be developed (Shepherd et al., 2005). However, replacing FO is problematic because of its unique fatty acid (FA) composition. It contains high concentrations of highly unsaturated FA, including eicosapentaenoic acid (EPA; 20:5n-3) and docosahexaenoic acid (DHA; 22:6n-3) (NRC, 1993). Aquaculture species require EPA and DHA for a number of functions, such as formation of anti-inflammatory prostaglandins (Yang et al., 2004) and leukotrienes (Kragballe et al., 1987). Furthermore, the health benefits of consuming EPA and DHA are well established (Van Horn et al., 2008) and fish are an important source of these nutrients in human diets (Schmidt et al., 2005). Most vegetable oils contain relatively low levels of n-3 FA and all are completely devoid of EPA and DHA (Bell et al., 2001; Geurden et al., 2006; Richard et al., 2006). However, many vegetable oils contain alpha linolenic acid (ALA; 18:3n-3). Linseed oil (LO) is a rich source of ALA, comprised of approximately 53% total lipid (NRC, 1993). Biosynthesis of EPA and DHA from ALA requires a series of desaturation and elongation steps and while salmonid fish possess all of the enzymes required to accomplish these conversions, the efficiency of this process is low (Ruyter and Thomassen, 1999; Tocher, 2003). Thus, replacing FO with LO results in reduced EPA and DHA concentrations in fish tissues (Caballero et al., 2002; Tocher et al., 2002; Richard et al., 2006; Drew et al., 2007). The high ALA content of LO makes it susceptible to oxidation during storage and feed processing, which may further reduce the conversion of ALA to EPA and DHA (Boran et al., 2006). It may also lead to off-flavours in fish products, reducing consumer acceptance.

Antioxidants may be added to lipids to prevent their oxidation. Phenolic antioxidants, such as vitamin E and butylated hydroxytoluene (BHT) bind with free radicals and are chain-breaking antioxidants that terminate lipid peroxidation (Shahidi et al., 1992; Wanasundara and Shahidi, 1994). Vitamin E not only reduces lipid peroxidation (Chaiyapechara et al., 2003; Lukaszewicz et al., 2004); when fed at levels above nutritional requirement, it also improves the sensory quality of fish products (Waagbø et al., 1993). Encapsulation using hydrogenated palm oil further improves the oxidative stability of LO by protecting it from environmental oxygen (Turchiuli et al., 2005). Based on these observations, we hypothesize that reducing the oxidation of ALA in LO by the addition of vitamin E and BHT or by encapsulation will improve its conversion to EPA and DHA by rainbow trout and improve the sensory quality of rainbow trout fillets.

2. Materials and methods

2.1. Oil products and diets

Four oil products were used in these studies: (1) a commercial mixed-species FO (Danish FO, FF of Denmark, Skagen, Denmark), (2) cold-pressed LO (Bioriginal Food and Science Corp., Saskatoon, SK, Canada), (3) LO with 7.5 g vitamin E and 12.5 g BHT/kg (stabilized LO) and (4) the stabilized LO encapsulated in hydrogenated palm oil (630 g/kg) (encapsulated LO).

Vitamin E was feed grade lutavit E 50% SD/WD (BASF Corporation, Mount Olive, NJ, USA). BHT was purchased from Sigma Chemical Co. (St. Louis, MO, USA). Encapsulated LO was Protected LO 35% obtained from JEFO Nutrition Inc. (St. Hyacinthe, QC, Canada).

The diets differed only in the lipid source and met all other nutrient requirements of rainbow trout (NRC, 1993). The ingredient composition and formulated digestible nutrient value of the diets is shown in Table 1. The digestible nutrient values of all ingredients included in the diets were determined prior to diet formulation. Fish meal was not included in any of the experimental diets to eliminate it as a source of EPA and DHA. The diets were mixed on a Hobart mixer (Model L-800; Troy, OH, USA) and cold extruded on a Hobart grinder (Model 4822; Troy, OH, USA) using a 3-mm die. Following extrusion, the diets were dried in a forced air oven (55 °C, 12 h), chopped and screened to obtain a uniform pellet size. Feed was stored at -20 °C in sealed polyethylene bags until use.

2.2. Fish management

Triploid rainbow trout (*Oncorhynchus mykiss*) were purchased from Wild West Steelhead (Lucky Lake, SK, Canada) and maintained in 360 L tanks that were part of a semi-closed recirculation system filtered biologically at the Prairie Aquaculture Research Centre (University of Saskatchewan), where animal protocols are approved by the University of Saskatchewan Committee on Animal Care and Supply and follow principles established by the Canadian Council on Animal Care (1993, 2005). Twenty-eight tanks were used in this trial, each consisting of 22 (31.3 ± 1.8 g) fish with seven replicates per treatment. Water temperature was maintained at 15 ± 1 °C. Dissolved oxygen (8.9 mg/L), pH (7.5) and temperature were observed and recorded daily. Chlorine (0.0 mg/L), nitrate (11.5 mg/L), nitrite (0.1 mg/L) and ammonia (0.2 mg/L) were monitored on a weekly basis. Photoperiod was a 14 h light per 10 h dark cycle using incandescent lighting at 7 lx. Between feedings, feed was stored in sealed containers in a freezer at -20 °C. The fish were fed to satiety twice daily and the amount of feed consumed by each experimental unit was recorded on a daily basis. Each tank of fish was weighed on days 0, 28, 56, 84, 112, 140 and 168.

2.3. Sample collection

At the end of the 168-day feeding period, fish were euthanized by a blow to the cranium. One fish per tank was gutted and frozen whole at -20 °C for FA analysis. The remaining fish were weighed along with their livers and viscera to determine

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