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The effects of camelina oil and solvent extracted camelina meal on the growth, carcass composition and hindgut histology of Atlantic salmon (*Salmo salar*) parr in freshwater

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ABSTRACT

A study was conducted to evaluate the nutritional value of experimentally solvent extracted camelina meal (SECM) and camelina oil (CO) fed to Atlantic salmon parr. Fish (initial weight: 8.4 g) were fed a control diet, or diets containing 5, 10, 15 or 20% SECM, or 50% and 100% CO substituted for fish oil in diets. Fish were reared in freshwater at 11.7 ± 0.5 °C for 16 weeks. Weight gain from week 13 to week 16 decreased in fish fed 10% SECM, and the daily growth coefficient (DGC) was lower in both 10% (1.22) and 15% (1.22) SECM groups (p < 0.05), but was similar between the fish fed 20% SECM (1.23) diet and the control (1.37; p ≥ 0.05) diet. The final body weight was not affected by dietary treatments. Condition factor, feed conversion ratio, protein retention ratio and hepato-somatic index were similar among all groups (p ≥ 0.05). Carcass protein, fat, ash and moisture were also unaffected. The length, width and area of simple villi in the distal intestine were unaffected by treatments (p ≥ 0.05), however, increased size of lamina propria was observed in fish fed 15 and 20% SECM diets (p < 0.05). Results suggest that fish performance was not affected by the inclusion of 5% SECM in the diet, and 50 to 100% replacement CO by fish oil.

This study investigated novel feed ingredients from *Camelina sativa* applied in aqua-feed for Atlantic salmon. Camelina oil is as good as fish oil, and it contains health-promoting factor for fish intestine. Solvent extracted camelina meal can be used at 5% in the diet. These findings can reduce reliance on fishmeal and fish oil in the future.

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1. Introduction

Fish meal and fish oil are important components of fish feeds, however, the increasing demands of the world's aquaculture production upon the finite quantity of these feed ingredients require that fish feeds become increasingly comprised of alternative protein and lipid sources of plant and/or animal origin that support similar fish performance. Numerous studies have investigated the potential of alternate plant proteins, particularly soybean meal, corn gluten meal and canola meal or their concentrates and vegetable oils and animal fats in salmonid fish diets (Carter and Hauler, 2000; Kaushik et al., 2004; Bell et al., 2005; Turchini et al., 2009). Applied research conducted in the past two decades has established the maximum amounts of some plant protein and lipid sources that carnivorous fish can tolerate anti-nutritional factors (ANFs) or toxicants (depending on the source, some or most of the following may be present i.e., protease inhibitors, lectins, phytic

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acid, saponins, phytoesterogens, alkaloids, tannins, cyanogens, glucosinolates) that adversely affect digestion, absorption and physiological utilization of protein and amino acids (Krogdahl et al., 2010). Recent nutritional strategies to replace fish meal in salmonid feeds with a major proportion of plant protein have shown marked improvement in growth and protein utilization. However, the growth performance of fish fed diets based extensively on plant protein sources generally has remained below that of fish meal-based diets.

Camelina sativa, a flowering oilseed plant in the family Brassicaceae, also called false flax, is grown in Europe and North America and shows potential as an animal feed ingredient (Waraich et al., 2013). Camelina oil, rich in α -linolenic acid (18:3n-3) and linoleic acid (18:2n-6), can replace all fish oil in the diet without inhibiting fish growth (Morais et al., 2012; Hixson et al., 2014a,b). High oil residue camelina meal (HOCM), the remaining meal after conventional oil, is removed from camelina seed by mechanical pressing using an expeller. HOCM contains 33.9% crude protein and 12% crude lipid (NRC, 2011). The crude protein level is elevated to 39.2–47.4% on a fat-free dry matter basis (Zubr, 2003), when HOCM is further processed by solvent extraction to produce solvent extracted camelina meal (SECM). SECM contains several





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ANFs, such as glucosinolates, phytic acid, tannins, sinapine, mucilage, and non-starch polysaccharides (Matthaus, 1997; Zubr, 2010). The concentration of ANFs in defatted meal should be higher than in HOCM. The degraded products from glucosinolates have a bitter taste (Mithen et al., 2000), and depress thyroid function in rainbow trout (Burel et al., 2000, 2001). Phytic acid reduces the availability of minerals, such as zinc, magnesium and phosphorus (Denstadli et al., 2006). Tannins inhibit protease, lipase and amylase activities (Mandal and Ghosh, 2010). Sinapine reduces feed palatability due to its bitter taste (McCurdy and March, 1992). In previous studies, the growth of rainbow trout was not inhibited by 16–20% HOCM (Pan et al., 2011; Bullerwell and Anderson, 2012).

The optimal level of SECM for Atlantic salmon has not been investigated. Plant protein sources like soybean meal cause enteritis in the distal intestine of Atlantic salmon (Van den Ingh et al., 1991; Van den Ingh et al., 1996; Baeverfjord and Krogdahl, 1996; Bakke-McKellep et al., 2007; Knudsen et al., 2007; Uran et al., 2008a; Uran et al., 2009). Soybeaninduced enteritis causes decreased height of mucosal folds/villi, disappearance of supranuclear vacuoles (SNV) in the intestinal epithelium, widening of lamina propria (LP) in the center of mucosal folds, infiltration of inflammatory cells in the lamina propria (Baeverfjord and Krogdahl, 1996), increased number of goblet cells (GC), and thicker sub-epithelial mucosa (SM) (Uran et al., 2008b). Whether camelina induces these symptoms in Atlantic salmon is not documented. The present study was conducted to evaluate the nutritional value of camelina by-products fed to Atlantic salmon parr in terms of growth performance, carcass composition, as well as morphology and histology of the distal intestine.

2. Materials and methods

2.1. Experimental diets

Camelina (Calena cultivar) was grown by Lyndhurst Farms Ltd. in Canning, Nova Scotia, Canada. Camelina oil was pressed from seeds using a KEK 0500 press at Atlantic Oilseed Processing Ltd., in Summerside, Prince Edward Island, Canada. Solvent-extracted camelina meal (SECM) was made in our laboratory (Faculty of Agriculture, Dalhousie University) at room temperature (20 °C) from high oil residue camelina meal by petroleum ether (Fisher Scientific product# E139-4) extraction at a ratio of 1 g meal/3 mL ether. Petroleum ether was composed of 50–60% pentane and isomers of hexane according to the specifications received by the manufacturer. The analysis of residual camelina meal showed 39% protein and 3% lipid (Table 1).

Each of the seven diets was formulated to be isonitrogenous (44% crude protein) and isocaloric (4400 kcal/kg digestible energy) to meet NRC (2011) requirements for Atlantic salmon. All ingredients used in the diets were typical of those used in commercial salmon feed. A control diet based on fish meal and fish oil, four diets including SECM at 5%, 10%, 15% and 20%, and two diets where 50% or 100% of the fish oil was replaced by CO were prepared. The fish oil level in SECM diets was the same as the control diet. If extra lipid was required to balance digestible energy, it was added as camelina oil. Feed formulation and analyzed amino acid content of the diets are summarized in Tables 2 and 3 respectively.

All feeds were mixed in a Hobart Mixer (Model L600), steam pelleted (2.5 mm) using a California Pellet Mill (San Francisco, USA), dried at 54 °C in an oven (JPW Design & Manufacturing), then cooled to room temperature, and stored at -20 °C until needed. A portion of each feed was crumbled to a suitable size of 0.75–1.0 mm particles and sifted to feed smaller fish for the first four weeks. The size of feed increased to 1.4–2.0 mm from week 5 to week 8, then intact 2.5 mm pellets were fed for the remaining period of the experiment.

2.2. Fish rearing conditions

Fish (2012 year class of St. John River stock) were transported from Big Falls Fish Grower Ltd. (Wolfville, Nova Scotia, Canada). Atlantic

Table 1

The nutritional composition of solvent extracted camelina meal (% as fed basis).

Crude lipid Crude protein Moisture	3.0 39.0 7.5	Acid detergent fiber Neutral detergent fiber Digestible energy (kcal/kg)	25.4 39.9 2172
Amino acids		% protein	% As-is
Aspartic acid		8.21	3.2
Threonine		4.12	1.61
Serine		4.83	1.89
Glutamic acid		16.93	6.60
Proline		5.32	2.08
Glycine		4.92	1.92
Alanine		4.26	1.66
Cysteine		2.08	0.81
Valine		4.45	1.73
Methionine		1.60	0.62
Isoleucine		3.11	1.21
Leucine		6.21	2.42
Tyrosine		2.48	0.97
Phenylalanine		3.88	1.52
Histidine		2.88	1.12
Lysine		4.55	1.78
Arginine		8.37	3.27
Tryptophan		0.96	0.37

Fatty acid composition of camelina oil can be found in another study (Hixson et al., 2014a).

salmon parr (initial mean weight 8.4 ± 0.2 g) were divided randomly among 21 tanks (210 L) with 50 fish/tank. Each tank was supplied with 3 L/min flow-through freshwater with temperature and oxygen

Table 2

Formulation and analyses of experimental diets fed to Atlantic salmon parr (% as fed basis).

Ingredient (%)	Control	Camelina oil		Solvent extracted camelina meal			
		50%	100%	5%	10%	15%	20%
Fish meal	32.1	32.1	32.1	30.3	28.4	26.5	24.6
SECM	0	0	0	5	10	15	20
Empyreal 75® ^a	15	15	15	15	15	15	15
Wheat gluten meal	5	5	5	5	5	5	5
Poultry byproduct meal	5	5	5	5	5	5	5
Pregelatinized starch	4	4	4	4	4	4	4
Whey	2	2	2	2	2	2	2
Ground wheat	19.7	19.7	19.7	15	10.3	5.7	1
Vitamin and mineral premix ^b	0.2	0.2	0.2	0.2	0.2	0.2	0.2
Special premix ^c	0.25	0.25	0.25	0.25	0.25	0.25	0.25
DL-Methionine	0.17	0.17	0.17	0.17	0.17	0.17	0.17
Fish oil	16.1	8	0	16.1	16.1	16.1	16.1
Camelina oil	0	8	16.1	1.5	3.1	4.6	6.1
Dry matter (%) ^d	92.8	91.5	92.1	93.2	91.6	89.4	91.4
Crude protein (%) ^d	43.6	43.5	43.2	42.9	43.4	43.1	42.8
Crude lipid (%) ^d	19.9	20.6	20.1	21.8	22.5	25.1	25.1
Calcium (%) ^d	1.9	1.9	1.9	1.8	1.7	1.6	1.5
Phosphorus (%) ^d	1.2	1.2	1.2	1.2	1.2	1.1	1
Sodium (%) ^d	0.4	0.4	0.4	0.3	0.3	0.3	0.3
Potassium (%) ^d	0.4	0.4	0.4	0.5	0.5	0.5	0.6
Magnesium (%) ^d	0.1	0.1	0.1	0.1	0.1	0.1	0.2
Manganese (mg/kg) ^d	128	120	126	132	133	136	137
Copper (mg/kg) ^d	7	7	11	9	10	8	12
Zinc (mg/kg) ^d	134	122	132	132	138	139	132
Glucosinolates (µmol/g) ^e	0	0	0	1.8	3.3	5.5	7.4

^a Cargill Corn Milling, Nebraska, USA.

^b Vitamin and mineral premix (per kg of diet): vitamin A, 5000 IU; vitamin D₃, 4000 IU; vitamin K, 2 mg; thiamin, 8 mg; riboflavin, 18 mg; pantothenic acid, 40 mg; niacin, 100 mg; folic acid, 4 mg; biotin, 0.6 mg; pyridoxine, 15 mg; vitamin B_{12} , 4 µg; inositol, 100 mg; zinc, 77.5 mg; manganese, 125 mg; iron, 84 mg; copper, 2.5 mg; iodine, 7.5 mg; ethoxyquin, 42 mg; and wheat shorts, 1372 mg.

^c Special premix (per kg/diet): selenium, 0.22 mg; vitamin E, 250 mg; ascorbic acid, 200 mg; astaxanthin, 60 mg; and wheat shorts, 1988 mg.

^d Analyzed by Nova Scotia Agriculture Quality Evaluation Division Laboratory Services, Truro, NS, Canada.

e Analyzed by Agriculture and Agri-Food Canada, Saskatoon, SK, Canada.

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