Processing of Canola Meal for Incorporation in Trout and Salmon Diets

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Canola meals (two commercial meals and one low-heat meal) were processed to reduce fiber content, then washed with selected solvents to reduce the content of antinutritional substances and further concentrate protein. The meals, fiber-reduced meals, and washed meals were used to provide 40% of total protein (26–38% of feed) in the diets of 6-g rainbow trout for 3 weeks or 25% of total protein (21–31% of feed) in the diets of 23-g chinook salmon for 11 weeks. Air-desolventized (low-heat) canola meal, as compared to commercial meal, provided no protein quality advantage in trout feeds. Fiber reduction processing of commercial meal increased meal protein content by 11–16% and reduced crude fiber by 23–50%, but did not have any effect on the quality of protein for trout or salmon. Solvent-washing of fiber-reduced meal improved fish response to canola meal, probably due to reduced glucosinolate content, but possibly also due to reduced sinapine content and alterations in protein availability. Protein concentration was increased by 25–40% by washing, and glucosinolate concentration was reduced by 40–90%.

KEY WORDS: Aquaculture, canola meal, feed, fiber reduction, meal processing, solvent washing.

Salmonid species (salmon or trout) require high dietary concentrations of protein and utilize protein and lipid as their principal sources of dietary energy (1). When reared in confinement without access to natural feed, the diet generally supplies protein in the form of fish meals or other by-products of the capture fishery.

Fish meal protein is more expensive than plant protein; for example, if all the value is attributed to the protein component, canola meal protein is approximately U.S. $0.40/kg and menhaden meal protein is approximately U.S. $0.75/kg (prices sourced in November, 1990). Thus, fish nutritionists are investigating plant sources of protein that would be acceptable in salmonid diets. However, plant protein sources, such as oilseed meals, generally have a lower concentration of protein and may also contain constituents undesirable in salmonid feeding.

Soybean meal is used in the feeds of a number of fish species but appears to be unacceptable in chinook salmon diets (2), although it is used in trout formulations. Hardy and Sullivan (3) noted through least cost formulation that replacement of soybean meal with canola meal in trout production diets would reduce feed costs by $19 per ton (U.S.).

The use of canola meal in commercial aquaculture feeds is currently limited to about 7%, due to concerns regarding the presence of undesirable constituents—fiber, glucosinolates, sinapine and phytate. Research reports have suggested that canola meal can be used as 25% of salmonid diets, with use limited by protein availability, antinutritional substances and low protein concentration (4). However, the feed value can be improved through processing.

A number of methods to process canola meal to remove the undesirable constituents have been studied by various groups in Europe and Canada (5). In this project, we adapted some of the previously developed technology for processing canola meal to produce a product that could be used in larger proportions in salmonid feeds. The specific objectives were i) to process commercial and low-heat canola meals to reduce undesirable constituents; and ii) to conduct feeding trials with the processed canola meals in trout and salmon.

EXPERIMENTAL PROCEDURES

The work was conducted in two phases. In Phase 1, a commercial meal and a low-heat meal were processed to reduce fiber. The fiber-reduced meals were washed by one of three solvents (water at pH 4.5, 80% aqueous ethanol, or ammonia/methanol) to produce a total of six processed canola meals for feeding trials. A seventh processed meal was obtained *via* an ammonia/methanol/hexane extraction of whole canola seed. These materials, along with the starting materials and the fiber-reduced meals, were incorporated into the diets of 6-g rainbow trout to provide 40% of the dietary protein in a 3-week feeding trial. In Phase 2, the acid and ethanol washes were consecutively applied to fiber-reduced commercial canola meal. Two variations of the acid-wash treatment were investigated—a hot acid wash and an enzyme-supplemented acid wash. The three treated meals, the starting material meal, and the fiber-reduced meal were incorporated into the diets of 23-g chinook salmon to provide 25% of the dietary protein in an 11-week feeding trial.

Materials. For Phase 1, four samples (90–250 kg each) of commercial canola meal were obtained from four Western Canadian oilseed crushers and blended to form a single, 700-kg lot. Similarly, three samples (70–120 kg each) of commercially produced meal were obtained from three of the same crushers, but these samples were taken just prior to the desolventizer-toaster step. This material was air-desolventized, then blended to form a 250-kg lot of meal, which was termed "low-heat" meal. For Phase 2, 400 kg of commercial meal was obtained locally.

Fiber reduction. Fiber content of the canola meals was reduced by milling the meals, then sieving over a 70-mesh screen (U.S. sieve number designation) and collecting the screen unders.

A preliminary laboratory investigation determined that tempering meals to 16% moisture resulted in a sieved product (material passing through 70-mesh) that had a higher protein content, lower crude fiber content and higher dry matter yield than meals with lower moisture

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contents. Thus, for pilot-scale fiber reduction, canola meals were always adjusted to 16% moisture content.

The commercial meals were milled in a disc mill (0.008″ gap, single disc mill Model No. 148-2-8″; Bauer Brothers Co., Ltd., Brantford, ON) at a rate of 75 kg/hr. The disc mill could not be used for the low-heat meal because the meal stuck to the discs and plugged the mill. The low-heat meal was milled in a comminuting chamber mill (Fitzpatrick Co., Elmhurst, IL) equipped with a 50-mesh screen at a rate of 20 kg/hr. Milled meals were sieved through a vibratory screen (Model 111 A-MS, Rotex Inc., Cincinnati, OH).

Solvent washing. For Phase 1, one of three washing treatments was applied to 20 kg (commercial) or 7.5 kg (low-heat) of fiber-reduced meal. In the acid wash process, meals were extracted for 1 hr at 22–23°C with nine times the weight of water acidified to pH 4.5. This pH represents the minimum solubility point of canola meal protein. The washed solids were recovered in a disk centrifuge (Model SA07, Westfalia, Olde, Germany). In the ethanol wash process, meals were extracted twice (1.5, 0.5 hr) with 8.5 times the weight of 80% (w/w) aqueous ethanol at 22–23°C. The washed solids were recovered in a decanter centrifuge (Bird 6″ continuous bowl, Bird Machine Co., Saskatoon, SK). For the ammonia/methanol wash process, ammoniated aqueous methanol (10% ammonia, 5% water, 85% methanol) was produced by sparging a weighed quantity of anhydrous ammonia into 95% aqueous methanol in a sealed reactor vessel. Meals were extracted at 22–23°C for 0.5 hr with 10 parts (by weight) of the ammoniated aqueous methanol followed with 5 parts of 95% methanol. The washed solids were recovered by use of the decanter centrifuge.

For Phase 2, fiber-reduced commercial meal was washed with acidified (pH 4.5) water under three conditions similar to those used for Phase 1 (differences are noted): an ambient temperature wash (identical to acid wash used in Phase 1), a 90°C acid wash, and an enzyme treatment-acid wash. For the latter treatment, fiber-reduced meal was blended with acidified water (0.047 M HCl), then 0.1% of Novo SP311 (Novo Industri A/S, Enzymes Division, Bagsvaerd, Denmark) was added, and the slurry was held at 50°C for 4 hr. Novo SP311 is an experimental, cell wall-degrading enzyme preparation, it is similar to Novo's commercial product Pectinex Ultra SPL.

The solids from each washing process were diluted with water to 13–18% solids content and drum-dried (Double Drum Dryer, Model T5.5, Goudsche Machinefabrik, BV, Gouda, The Netherlands) under a steam pressure of 335 kPa, a drum speed of 0.66 rpm and a rate of 25–30 kg/hr. To further increase the protein content of the acid-washed meals produced for Phase 2, the drum-dried products were washed two times with 80% aqueous ethanol, as described for Phase 1. The ethanol-washed solids were air-dried for seven days to evaporate residual ethanol.

Whole seed processing. During Phase 1, canola protein product was also produced by a simulation of the ammonia/methanol/hexane process for whole canola seed, developed at the University of Toronto (6). The glucosinolate content of this meal is claimed to be below the detection limit, protein content is 50%, and protein recovery is about 90% (7). Because the seed is ground in the presence of the extractant, the hull fragments are large and can be sieved out of the meal after air desolventization.

Whole canola seed and ammoniated aqueous methanol (10:5:85) were simultaneously fed to a Szego mill. The solids were recovered *via* a decanter centrifuge, and washed with methanol to remove excess ammonia, then washed twice with hexane to remove oil. The hexane-extracted meal was air-desolventized for four days, then sieved through a 70-mesh screen to recover a fiber-reduced material in the unders fraction.

Products. Seven processed canola meals were produced during Phase 1 and three in Phase 2. These processed meals plus the three fiber-reduced meals (two commercial and one low-heat) and the three starting material meals were assayed for dry matter, protein, ash, fat, fiber, phytate, sinapine and glucosinolates.

Analytical. Dry matter contents were determined by drying 2-g samples in a forced air oven at 105°C to constant weight. Nitrogen content of dry, ground, defatted samples was determined by an automated Kjeldahl method with the Kjel-Foss Automatic Model 16210 (ref. 8; method 7.021) and crude protein content was obtained by multiplying by 6.25. Lipid content of dried, ground samples was determined by the Butt tube method (ref. 9; method Aa 4-38). Ash was assayed by charring a dried, ground sample at 600°C in a muffle furnace to constant weight. Crude fiber was determined on dried, ground samples by the procedure of Knox *et al.* (10). Acid detergent fiber and lignin were determined on dried, ground samples by AOAC method 7.074-7.077 (8), and neutral detergent fiber (insoluble dietary fiber) by AACC method 32-20 (11). Glucosinolate content of dried, defatted samples was measured by the method of Daun and McGregor (12); total glucosinolates was taken to be the sum of allyl-, 3-butenyl, 4-pentenyl, 2-hydroxy-3-butenyl, and 2-hydroxy-4-pentenyl-glucosinolates. Phytate content of dried samples was assayed *via* the procedure of Latta and Eskin (13). Sinapine content was determined by the method of Legueut *et al.* (14).

In vitro *digestibility and acid solubility. In vitro* digestibility of the 11 canola materials from Phase 1 was estimated by means of a pepsin digestibility test (ref. 8; method 7.053-7.059). The method was modified to take into account the lower body temperature (as compared to poultry and livestock) of cold-water fish (15). Protein solubility also was measured after incubation in 0.075 HCl without the addition of pepsin.

Feeding trials, Phase 1. The eleven canola meal materials obtained in Phase 1 plus a herring meal control were evaluated in short-term (three-week) feeding trials with rainbow trout (6-g average weight) from commercial stock. The fish were distributed at random into groups of 50 fish each in 24 tanks. The water supply was dechlorinated city water heated to a temperature of 13.8–14.5°C. Feeding was to satiation at each of two hand feedings each day, and the amount of feed consumed each day was recorded. The fish were anesthetized (MS-222 and sodium bicarbonate) before individual weighing on day 22. Specific growth rate was calculated as $[(\ln W_2 - \ln W_1)/(T_2 - T_1) \times 100]$, where W_1 and W_2 are the mean body weights at the beginning (T_1) and end (T_2) of the test period, respectively. The efficiency of feed conversion was calculated as [feed consumption (air-dry)/body weight gain (wet)].

The diets in Phase 1 were formulated to contain 40% of crude protein. The control diet contained herring meal

as the principal source of protein. The experimental diets contained the canola materials in amounts to supply 16% of protein (*i.e.,* to supply 40% of the total dietary protein). In this way, comparisons of response were possible on the basis of the available amino acids contained in the 16% of protein contributed by the various canola materials compared to the 16% of protein contributed by the herring meal. The remaining protein in the experimental diets was contributed by herring meal (21.5%) and by wheat (2.5%, also present in the control diet at this level).

The percentage of canola products in the diets varied from 26 to 38%. Because differences existed between the amounts of carbohydrates, minerals and fatty acids contributed by the canola meal materials and by the herring meal, where feasible, dietary adjustments were made to accommodate these differences. For example, canola oil and herring oil were added at different concentrations to the diets to adjust for the amounts of the oils present in herring meal and the canola preparations. Since the fish adjust feed intake to meet their energy requirements, it was considered to have the least confounding effect if fiber content of the diets was not adjusted. The amounts of each diet consumed were adjusted to equivalence with the control diet by correcting for differences in crude fiber content. The approach was validated by the finding that fiber-reduction of canola meals did not improve growth rate. Neither did feed efficiency improve when feed consumption was adjusted for the amount of fiber. The vitamin-mineral supplement included in all of the diets was designed to satisfy the requirements of the fish without dependency on the test materials for any of the components.

Phase 2. The five canola meal materials produced from Phase 2 plus a herring meal control were evaluated in an 11-week feeding trial with chinook salmon. The fish (average weight 23 g) had been transferred to sea water 60 days prior to the experiment. They were distributed into eighteen tanks with 41–42 fish per tank. Water was supplied by a flowthrough system. The temperature of the water ranged from 12.5°C at the beginning of the experiment to 10°C when the experiment was terminated. The fish were fed to satiation once daily, six days per week, and a record was kept of the feed consumption of the fish in each tank. The fish were individually weighed after being fed the experimental diets for 52 and 77 days. Specific growth rates and estimates of the efficiency of feed conversion were calculated for the periods from 0 to 52 days, 53 to 77 days, and from 0 to 77 days.

The total protein concentration in the diets was 46%, with 11.5% of protein derived from the canola products in substitution for an isonitrogenous quantity of the control herring meal. Thus, canola protein provided 25% of the total protein in the diet. The percentage of canola products in the diets varied from 21 to 31%. Similar considerations regarding diet formulations were used in Phases 1 and 2.

RESULTS

Composition. The compositions of the canola meals, the fiber-reduced meals and the washed canola meals from Phases 1 and 2 are shown in Table 1. Disc milling and sieving of commercial canola meal reduced crude fiber content by 50% in Phase 1, but only by 23% for the Phase 2 meal; protein content was increased by 16% and 11%, respectively. For low-heat meal, fiber was reduced by 21% and protein increased by 4%.

Use of acidified water to wash fiber-reduced canola meals slightly increased the protein concentration—an 8–11% increase for Phase 1 meals and a 3–6% increase for Phase 2 meals. The acid-washing treatments increased crude fiber concentration (14–19% for Phase 1 meals and 29–53% for Phase 2 treatments), but decreased glucosinolates (45–75%), phytate (3–42%), and sinapine (40–60%).

In the variations of acid washing applied in Phase 2, the hot acid wash reduced sinapine and glucosinolates more effectively than the room temperature or enzyme-treatment acid wash, but the fiber components were more concentrated by this treatment. The hot-acid wash treatment was less effective than the others in reducing phytate. Since only one replication was possible, it would be useful to verify these results.

The double ethanol wash (Phase 1) of the fiber-reduced meal increased protein content by 18–22%. Ethanol washing also resulted in a concentrating of the fiber components, increasing crude fiber content by about 45% as the ethanol-soluble materials were washed out. Ethanol washing decreased sinapine content by approximately 90% and glucosinolate content by 80%, but phytate content increased by about 25%.

Ethanol washing of the acid-washed, fiber-reduced meals for Phase 2 increased protein content of the meals by 12–15%. The protein contents of room temperature acid-washed and the hot acid-washed meals, which were subsequently ethanol washed, were similar (54–55%); that of the enzyme-treated, acid-washed and ethanol-washed meal was higher (58%). As in Phase 1, ethanol washing was effective in reducing sinapine (80% to 85% reduction) and glucosinolates (75% to 85% reduction). The fiber and phytate components, which are not ethanol-soluble, were concentrated by this step.

Ammonia/methanol washing (Phase 1) of fiber-reduced meals increased crude protein content by 17–31%. Fiber components were concentrated in the meal by the washing process. Ammonia/methanol washing reduced the sinapine and glucosinolate contents, but increased the concentration of phytate.

In comparison to the washing processes, whole seed processing produced a meal that was highest in protein content. Fiber components were lower than in washed meals. Glucosinolate content was intermediate between ethanol-washed and ammonia/methanol-washed meals. Sinapine content was comparable to that of the ammonia/methanol-washed meal. The phytate content was the highest of all the products tested. This was as a result of concentration during processing, the whole-seed starting material was 2.1% phytate.

Process yields. The yields of dry matter and protein observed for the processing of the canola meals are shown in Table 2. There were major differences between the two commercial and the low-heat meals in the yields resulting from the fiber reduction. On an "as is" moisture basis, the yield of fiber-reduced meal from commercial meal was 35–39% and 49.5% from low-heat meal. The products differed in protein concentration. Ethanol washing removed more dry matter and crude protein than the other solvent washes.

Pepsin and acid solubility. The percentage of protein solubilized by pepsin in acid solution and by acid alone

TABLE 1

Composition of Canola Meals (commercial and low-heat), Fiber-Reduced Canola Meals and Washed Canola Meals

Canola material	Moisture %, as is	%, Dry matter basis									Glucosinolates[c] μmol/g, dry basis
		Crude protein	Crude fat	Ash	ADF[a]	NDF[b]	Crude fiber	Lignin	Phytate	Sinapine	
Commercial canola meal											
Phase 1	8.9	40.4	3.3	8.5	20.1	24.8	13.5	17.6	3.5	2.6	18.1
Phase 2	9.3	42.1	2.8	8.1	16.2	23.2	11.3	15.8	3.1	2.1	14.8
Low-heat canola meal (Phase 1)	7.6	40.0	2.1	8.2	21.0	23.6	13.2	15.7	3.8	2.4	17.6
Fiber-reduced commercial canola meal											
Phase 1	9.7	46.7	3.8	9.2	10.2	13.3	6.8	8.7	4.5	2.5	19.7
Phase 2	12.7	47.1	3.9	8.0	12.8	18.2	8.7	11.1	2.6	2.3	16.4
Fiber-reduced low-heat canola meal (Phase 1)	9.3	41.7	2.3	8.8	16.6	21.2	10.4	15.3	3.8	2.5	17.8
Acid-washed, fiber-reduced commercial canola meal											
Phase 1—room temp.	7.2	50.4	2.6	8.9	11.7	17.8	8.1	10.6	3.1	1.5	11.0
Phase 2—room temp.	7.7	48.7	3.2	7.0	15.8	22.1	11.2	13.9	1.5	1.3	5.2
Phase 2—90°C	8.5	48.6	2.8	9.6	19.2	22.5	13.3	16.8	2.5	0.9	4.1
Phase 2—enzyme treatment	6.5	50.1	3.9	8.0	16.8	19.2	12.1	15.5	1.9	1.2	6.3
Acid-washed, fiber-reduced low-heat canola meal (Phase 1)	10.1	46.4	0.5	9.5	18.5	19.6	11.9	17.3	2.8	1.4	9.8
Ethanol-washed, fiber-reduced commercial canola meal (Phase 1)	13.0	57.2	0.6	9.0	13.2	18.5	9.7	12.2	5.5	0.2	4.1
Ethanol-washed, fiber-reduced low-heat canola meal (Phase 1)	12.3	49.2	0.5	8.3	21.8	30.1	15.1	20.4	4.8	0.3	3.5
Ethanol & acid-washed, fiber-reduced meal (Phase 2)	11.2	55.0	1.0	6.3	18.9	25.3	12.8	17.5	2.4	0.2	1.3
Ethanol & hot acid-washed, fiber-reduced meal (Phase 2)	11.5	54.4	0.7	8.0	21.3	27.5	14.3	19.6	3.1	0.2	0.6
Enzyme-treated and ethanol & acid-washed, fiber-reduced meal (Phase 2)	10.4	57.8	1.6	7.3	21.0	24.9	14.4	20.0	2.6	0.2	0.9
Ammonia/methanol-washed, fiber-reduced commercial canola meal (Phase 1)	12.5	58.1	0.1	9.1	14.1	22.3	10.3	13.0	5.8	0.8	1.4
Ammonia/methanol-washed, fiber-reduced low-heat canola meal (Phase 1)	7.8	48.6	0.4	8.3	22.7	27.2	14.5	21.0	4.6	0.7	1.1
Fiber-reduced canola meal produced by U. of T. process (Phase 1)	7.2	60.6	2.6	9.6	10.3	16.9	7.9	9.8	6.1	0.8	2.6

[a] Acid detergent fiber.

[b] Neutral detergent fiber.

[c] Total glucosinolates includes 3-butenyl, 4-pentenyl, 2-OH-3-butenyl, and 2-OH-4-pentenyl glucosinolates.

is shown in Figure 1. (This test was conducted only on Phase 1 materials.) Total solubility of canola protein (between 80 and 90%) was similar for all the samples. However, the proportions of protein solubilized by acid alone and that requiring assistance of pepsin varied among the canola materials.

The acid solubility of canola protein from commercial meal was generally lower than that of protein from low-heat meal. Acid washing of the fiber-reduced meals removed some acid-soluble protein, as indicated by the reduced acid solubility of the protein in the acid-washed meals, compared with that in the corresponding unwashed

TABLE 2

Processing Yields for Dry Matter and Protein During Fiber-Reduction and Solvent Washing of Canola Meal

	Dry matter yield	Protein yield
Canola meal starting materials	100	100
Fiber-reduced meal		
Phase 1—commercial meal	39	45
Phase 1—low-heat meal	49	51
Phase 2—commercial meal	34	37
Acid-washed meal		
Phase 1—room temperature, commercial meal	33	41
Phase 1—room temperature, low-heat meal	40	46
Phase 2—room temperature, commercial meal	27	31
Phase 2—90°C	25	29
Phase 2—enzyme treatment	23	28
Ethanol-washed meal		
Phase 1—commercial meal	28	39
Phase 1—low-heat meal	39	48
Ethanol & acid-washed meal		
Phase 2—room temperature	21	27
Phase 2—90°C	21	27
Phase 2—enzyme treatment	18	25
Ammonia/methanol wash		
Phase 1—commercial meal	32	46[a]
Phase 1—low-heat meal	40	48[a]

[a]Value includes unknown proportion of ammonia nitrogen absorbed during processing.

fiber-reduced meals. The percentage of protein solubilized due to pepsin action was higher in the acid-washed concentrates when compared with the other canola products.

For the ethanol-washed meals, protein solubility was similar to the low-heat fiber-reduced meal. Ammonia/methanol washing of the fiber-reduced meals reduced the proportion of protein soluble in dilute acid to levels similar to those in the acid-washed meals. The protein in the meal from ammonia/methanol/hexane-extracted seed was as soluble as that of low-heat, fiber-reduced meal.

Fish response. The mean specific growth rate and feed conversion efficiency for the canola meals, the fiber-reduced canola meals, and the washed, fiber-reduced meals are shown in Tables 3 (trout) and 4 (salmon). Fiber-reduction processing of canola meal did not significantly improve the response of either trout or salmon to canola protein, as measured by specific growth rate or feed conversion for the three meal sources tested (Tables 3 and 4). Acid-washing significantly improved the specific growth rate response of trout to commercial and low-heat meals and improved the feed conversion response of trout in the case of commercial meal. Ethanol washing significantly improved the specific growth rate response to commercial meal and improved feed conversion response to both commercial and low-heat meals. Ammonia/methanol-washing significantly improved specific growth rate and feed conversion efficiency responses for both meals (Table 3).

The acid-washed, fiber-reduced commercial canola meal supported the best mean specific growth rate response of trout of all the Phase 1 meals tested, including the control, herring meal diet. The ethanol-washed, fiber-reduced

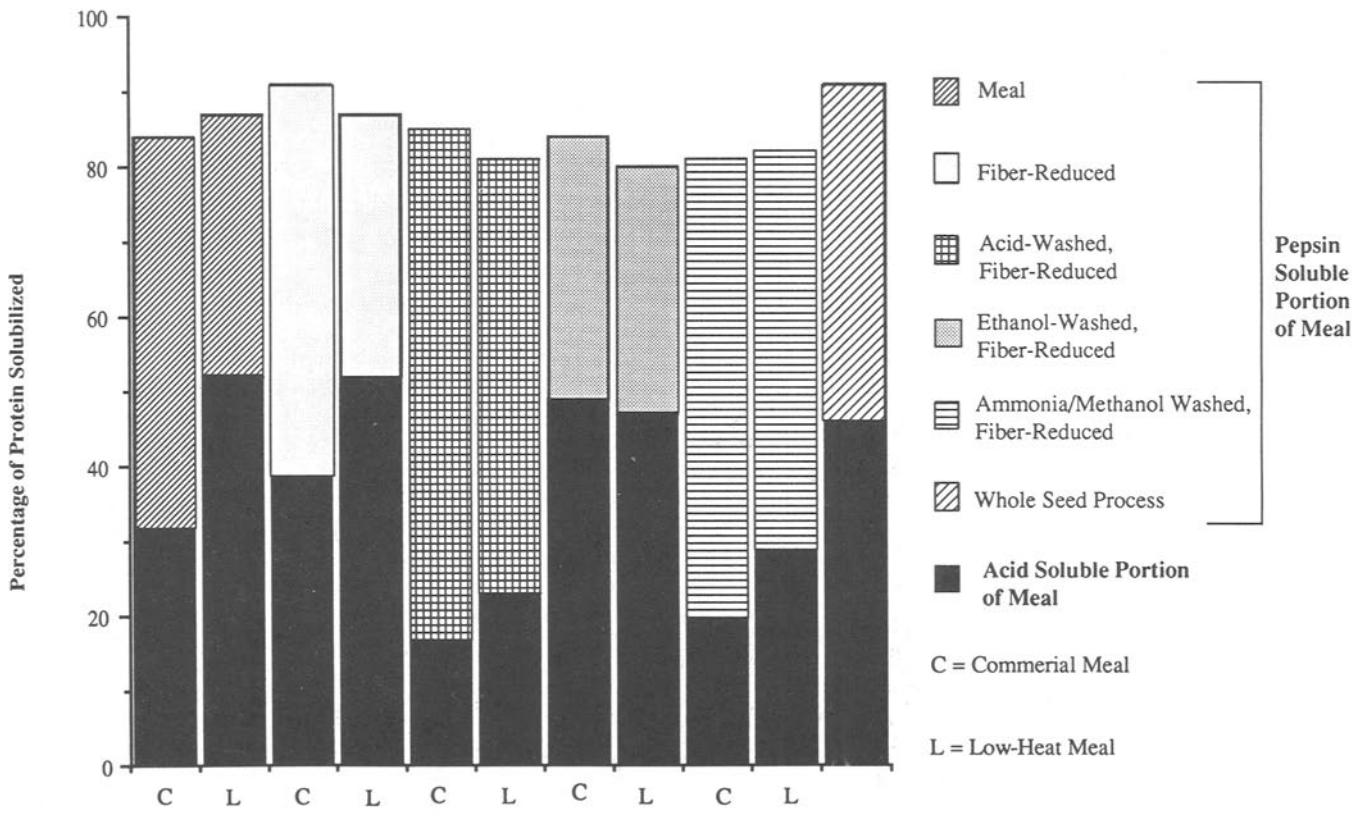

FIG. 1. Percentage of protein solubilized by acid and pepsin in canola products.

TABLE 3

Mean Specific Growth Rate and Feed Conversion for Rainbow Trout in Response to Inclusion of Canola Meals, Fiber-Reduced Canola Meals and Washed, Fiber-Reduced Canola Meals to Supply 40% of Dietary Protein*

	Mean specific growth rate				Mean feed conversion			
	Control	Commercial meal	Low-heat meal	Canola seed meal	Control	Commercial meal	Low-heat meal	Canola seed meal
Control diet (herring meal)	3.7 bc				1.20 de			
Canola meal		3.5 cde	3.2 fg			1.26 f	1.24 ef	
Fiber-reduced canola meal		3.4 ef	3.4 def			1.25 f	1.20 de	
Acid-washed, fiber-reduced canola meal		4.0 a	3.7 bc			1.13 bc	1.15 cd	
Ethanol-washed, fiber-reduced canola meal		3.7 bc	3.5 cde			1.07 a	1.08 ab	
Ammonia/methanol-washed fiber-reduced meal		3.9 ab	3.8 ab			1.14 c	1.13 c	
University of Toronto process				3.7 bcd				1.19 de

*Means within a biological response followed by different letters are significantly different.

TABLE 4

Mean Specific Growth Rate and Feed Conversion for Chinook Salmon in Response to Inclusion of Canola Meal, Fiber-Reduced Canola Meal and Washed, Fiber-Reduced Meals to Supply 25% of Dietary Protein*

	Mean specific growth rate			Mean feed conversion		
	0–52 days	53–77 days	0–77 days	0–52 days	53–77 days	0–77 days
Control diet	0.55 b	0.74 b	0.61 c	2.7 b	2.7 ab	2.7 b
Canola meal	0.30 c	0.60 b	0.40 d	4.9 c	4.1 b	4.5 c
Fiber-reduced canola meal	0.22 c	0.62 b	0.35 d	6.5 c	3.9 b	5.1 c
Ethanol and acid-washed, fiber-reduced meal	0.81 a	1.06 a	0.89 a	2.0 a	2.0 a	2.0 a
Ethanol & hot acid-washed, fiber-reduced meal	0.64 b	0.84 ab	0.70 b	1.9 a	2.0 a	2.0 a
Enzyme-treated and ethanol & acid-washed, fiber-reduced meal	0.56 b	0.70 b	0.60 bc	2.3 ab	2.5 a	2.4 ab

*Means within a biological response followed by different letters are significantly different.

canola meals supported the best feed conversion efficiency. Acid-plus ethanol-washing of fiber-reduced meal (Phase 2) improved both the specific growth rate and the feed conversion response of salmon, as compared to the commercial meal or the fiber-reduced, commercial meal and, in some cases, as compared to the herring meal control diet (Table 4). In general, the room temperature, acid-washed meal produced an improved response of salmon to canola protein as compared to the other two acid wash treatments.

DISCUSSION

Commercial meal vs. *low-heat meal.* Commercial canola meal receives a heat treatment during desolventization that might result in excessively heat-denatured protein that is less available to the fish. Thus, commercial canola meal and a low-heat canola meal were compared in Phase 1. The tempered low-heat meal was more difficult to mill as compared to the commercial meal. In the disc mill, the more soluble protein in this meal coagulated and plugged the grooves on the discs. Fiber-reduction processing of low-heat meal was less efficient than that for the commercial meal—crude fiber was reduced by 50% in the commercial meal but only by 20% in the low-heat meal.

A higher proportion of the protein present in low-heat meal was soluble in water without the use of pepsin, as compared to the commercial meal. Low-heat meal and the canola products produced from it gave equivalent or poorer growth response in trout as compared to the commercial meal and its products. Thus, there was no advantage (and some disadvantage) to use of low-heat canola meal in salmonid diets.

Fiber reduction. Since salmonids cannot utilize cellulose and require high dietary concentrations of protein, canola meal was processed to reduce fiber and simultaneously increase protein content. Hull and cotyledon material tend to become tightly bound during the heat, pressure, and shear conditions generated in expeller processing, so economical fiber and protein shifts in commercial meal are difficult to achieve.

It was determined that tempering commercial meal to 16% moisture prior to milling enhanced the shifting of fiber and protein. It was hypothesized that this effect was

related to a "toughening" of the hull at higher moisture, increasing its resistance to milling.

Fiber and protein shifting in the composite commercial canola meal used in Phase 1 (50% fiber reduction, 16% increase in protein content) was more efficient than for the commercial meal procured for Phase 2 (23% fiber reduction, 11% increase in protein content). This observation requires further investigation before such processing could be recommended. The yield of fiber-reduced meal was 34–39% on a dry-matter basis. Fiber reduction processing did not appear to alter protein quality, although it was useful in increasing protein concentration in canola meal. The response of rainbow trout and of chinook salmon to unprocessed canola meals (two commercial meals and one low-heat meal) and to the corresponding fiber-reduced meals showed no differences in protein quality.

Meal washing. Washing processes are required to reduce the antinutrient content of canola meal. Glucosinolate content of the meal is of primary concern, since it causes growth depression and deformities in trout. Sinapine content, due to its bitterness and potential for decreasing palatability, and phytate content, due to its ability to tie up essential minerals, are also of conern. Substantial changes in canola meal composition occurred with processing and the extent of change was affected by the meal source.

Acid-washing did not increase protein content as well as did washing with ethanol or ammoniated methanol. Acid-washing removed about 40% of the glucosinolates and sinapine. The ethanol wash removed up to 90% of the sinapine and 80% of the glucosinolates, while the inverse was true for the ammonia/methanol wash, which removed 90% of the glucosinolates and 70% of the sinapine. Ammonia is destructive to some glucosinolates. Acid-washing removed 10–50% of the phytate, but this compound is not soluble in alcohol, and thus was concentrated by this processing, increasing 20–30%. The combination of acid-washing and ethanol-washing (Phase 2) did not improve protein content as much as alcohol (ethanol or methanol) washing alone (Phase 1). However, reduction of the antinutrient components was more effective with a combination of acid-washing and ethanol-washing.

Whole seed processing. The meal from the ammonia/methanol/hexane processing of whole seed had the highest protein content, its glucosinolate content was intermediate between the Phase 1 ammonia/methanol-washed meal and the ethanol-washed meal, while sinapine content was similar to ammonia/methanol-washed meal.

Nutritive quality. The high concentration of protein used in salmonid diets makes the assessment of protein digestibility of particular importance. Pepsin digestibility and acid solubility were used as a measure of protein quality in the canola products, in addition to feeding trials to measure fish response on the basis of the growth rate and the efficiency of feed utilization. The experimental diets were formulated to assure that any differences in response would be attributable only to differences in the quality of protein and associated antinutrients contributed to the diets by the various canola products.

Washing of fiber-reduced canola meal with various solvents improved fish response for specific growth rate and feed conversion. The reason for the enhanced responses to washed meal cannot be conclusively stated from this work, but the relationships between growth responses and concentration of selected components in the tested meals were identified and are shown in Table 5.

Relationships were noted between enhanced specific growth rate and decreasing solubility of canola protein in dilute acid and an increased proportion of pepsin-soluble protein (Table 5). There was no relationship between feed conversion and protein solubility. There were also positive relationships between improved specific growth rate and feed conversion and reduced glucosinolate and sinapine contents. It should be noted that sinapine and glucosinolates are co-extracted by alcohol ($r = 0.95$ and 0.99 for Phases 1 and 2, respectively).

These experiments show that washed, fiber-reduced canola meals are a good source of dietary protein for trout and salmon, when included in the feed to provide up to 25% (salmon) and 40% (trout) of the dietary protein. Higher fish meal protein replacement levels may be possible; 25% and 40% were the levels tested in this work. Fish growth response to the washed, fiber-reduced canola meals equaled or surpassed that of the control (herring meal) diet. The improved growth response to washed canola meal (as compared to unprocessed meal) is likely due to reduced levels of glucosinolates, and possibly reduced levels of sinapine, although other experiments would be required to confirm this.

Although a detailed economic evaluation was not conducted, examples from the soybean crushing industry suggest that the application of fiber reduction and wash processing to canola meal may be economically feasible.

TABLE 5

Correlation Coefficients Between Fish Response and Composition of Canola Meal Products

	Phase 1 (n = 11)		Phase 2 (n = 5)	
	Specific growth rate	Feed conversion	Specific growth rate	Feed conversion
Proportion of protein soluble in dilute acid	0.73	0.15	NM[a]	NM
Proportion of protein solubilized by pepsin	0.67	0.00	NM	NM
Glucosinolate content	0.68	0.79	0.87	0.98
Sinapine content	0.63	0.86	0.88	0.98
Phytate content	0.41	0.11	0.64	0.61

[a]NM, not measured.

The soy industry produces two meals—high protein (49%)/low fiber (4%) and lower protein (44%)/higher fiber (7%). The same approach may be applied in the canola crushing industry where meal could be processed to prepare analogous high protein (42%)/lower fiber (6%) and lower protein (33%)/higher fiber (15%) meals. Ethanol-washing of soybean meal is presently being investigated to reduce oligosaccharide content to improve its feed value for poultry. Similarly, ethanolic or acidic extraction of fiber-reduced canola meal to remove antinutrients would produce a meal with enhanced value for inclusion in fish feeds.

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