### LIPIDIC PROFILES OF TISSUE AND LIVER OIL OF BURBOT, Lota lota (L.)

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Background. Burbot, Lota lota (L.), is a freshwater fish that is related to the marine family Gadidae. Its distribution is widespread in northern Canada. This species is not fished commercially at present. The high content of vitamin A and vitamin D in burbot liver oil was first reported in 1930. Since that time, there have been little or no research studies made on the nutritional components of burbot liver oil. The present study was aimed to assess the key nutritional components present in liver oil isolated from burbot caught in two northern Canadian lakes. Materials and Methods. Liver oil extracted from liver isolated from burbot caught in two northern Canadian lakes was tested for n-3 fatty acids and fat-soluble vitamins. Thermal stability of the liver oil was also evaluated. Burbot tissue was also analyzed for comparison purposes.

Results. The contents of n-3 fatty acids, vitamin A and vitamin D of burbot liver oil were found to be comparable to those of the (reference) cod liver oil. Vitamin K content was discovered to be surprisingly high in comparison to known leafy green vegetable food sources.

Conclusion. Burbot liver oil could provide a single source of dietary supplement of n-3 fatty acids, vitamin A, vitamin D, and vitamin K to meet the daily recommended nutritional allowances.

Keywords: burbot, omega-3 fatty acid, nutrition, vitamin A, vitamin D, vitamin K

## INTRODUCTION

belongs to the order Gadiformes along with fishes of marine Gadidae family. Its distribution is widespread in the northern hemisphere northward from 40°N (Scott and Crossman 1973). Burbot is nocturnal and can live up to 15 years. The adult (maturity at 3 to 4 years old) is about 30 to 50 cm in length, weighing in the range of 1 to 7 kg. Burbot spawns from January to March. Most of the fishing under ice cover is made during this period, as burbot tends to congregate over sand or gravel bottom.

There are commercial catches of burbot in northern Europe, viz., Finland, Russia, and Sweden. The reported 1994-2003 catch averaged about 3000 t globally (Anonymous 2005). Finland accounts for about 50% of the commercial catches. Burbot fillet is used for human consumption as well for conversion to pet food.

In northern Canada, burbot is only caught sparingly Burbot, Lota lota (L.), is a freshwater fish that for food use. A part of the problem has been elusiveness of burbot to render easy commercial fishing. Typically, burbot is a by-catch of winter commercial fishing of lake trout, Salvelinus namaycush; common carp, Cyprinus carpio; walleye, Stizostedion vitreus; yellow perch, Perca flavescens; northern pike, Esox lucius; and lake whitefish, Coregonus clupeaformis. The burbot is discarded intentionally when the commercial fish catch is sorted at the ice-fishing site. Exploratory field studies have suggested that the burbot by-catch (during the winter months) could be as much as 20% of the normal commercial catch (Wong unpublished: "An exploratory assessment of burbot (Lota lota L.) catches in 4 northern Canadian lakes").

> As shown by the following proximate analysis in Table 1, there are considerable similarities in the proximate composition between burbot (a freshwater gadiform species) and cod (a saltwater gadiform species).

> > Table 1

Nutritional content of cod and burbot tissues (Anonymous 2008a, b)

	Value per 100 g of edible portion (skinless tissue)				
	energy [kJ]	protein [g]	total lipid [g]	ash [g]	
Atlantic cod, Gadus morhua	343	17.81	0.67	1.16	
Burbot, Lota lota	377	19.31	0.81	1.16	

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The medicinal benefits of consumption of fish and fish oil have been studied for more than 25 years. Cod liver oil is typically isolated from cod liver in commercial cod fishery in the northeastern Atlantic. Unfortunately, the cod stock has been largely depleted which resulted in substantial curtailment of commercial cod fishing. Thus, the supply of cod liver oil is expected to be reduced similarly. Indeed, some commercial manufacturers of cod liver oil in Europe are searching actively for alternative sources. The oil content of liver has been reported to the 6% to 10% of overall body weight of the burbot (Holčík and Mihálik 1972). There is a market opportunity for burbot liver oil to replace cod liver oil. There are no known commercial suppliers of burbot liver oil anywhere in the world today.

Omega-3 Fatty Acids. The current medical consensus is that long-chain unsaturated omega-3 fatty acids, viz., 5,8,11,14,17-eicosapentaaenoic acid (EPA; C20:5) and cis-4,7,10,13,16,19-docosahexaenoic Acid (DHA; C22:6), present in fish and fish oil, are especially effective in reducing the risk of coronary heart diseases (Pigott and Tucker 1987, Connor and Connor 1997, Harris, 1997, Connor 2000, Kris-Etherton et al. 2002). Moreover, hypertriglyceridemia could also be remedied by the onpurpose consumption of omega-3 rich fish oil (Phillipson et al. 1985, Connor 1988, Harris et al. 1990).

**Fat-Soluble Vitamins.** Burbot liver oil has been cited in the literature to be particularly rich in vitamin A and vitamin D (Branion 1930). Unfortunately, there are virtually no modern data available on fat-soluble vitamin content of burbot liver oil.

The present study was undertaken to determine the marketable nutritional characteristics, viz., n-3 fatty acids, vitamin A, vitamin D, and Vitamin K, of liver oil isolated from burbot caught in representative lakes in northern Canada.

# MATERIAL AND METHODS

Burbot was caught in several ice fishing campaigns in Athapapuskow Lake (Manitoba) and Amisk Lake (Saskatchewan), Canada. It may be noted that in the Canadian Shields, the landscape is dotted with numerous

relatively shallow lakes. The first batch (Athapapuskow Lake in February 2005) of liver samples was isolated by local field personnel from 17 fish (average weight: 1.82 kg; weight range: 0.91 to 3.51 kg). The liver sample was shipped by air freight to Arbokem Vancouver Lab in an ice-packed container. The transit time was 3 days.

A new batch of burbot was caught in Athapapuskow Lake in early March 2005. The liver was isolated manually at the Arbokem Vancouver Lab, from 5 whole fish, which were transported by Arbokem personnel from Bakers Narrows (nearest airport) to Vancouver. The transit time was about 8 hours, from freezer (-18°C) to freezer (-18°C). The test results from the processing of this batch of whole burbot are given Table 2.

Additional burbot liver (about 200 kg) was acquired in January 2006 from commercial fishermen operating in Amisk Lake (Saskatchewan), Canada, for the isolation of liver oil.

Composite of liver samples (of each series: mixed Athapapuskow Lake samples of February 2005 and March 2005, and Amisk Lake sample of January 2006) was macerated and extracted with a mixture of chloroform-methanol (1 : 2 v/v ). Typically, 1 g of wet liver composite was extracted twice with 2 g of solvent mixture. The extract was subsequently de-solventized in a rotary evaporator at 65°C under water-jet vacuum. Aliquots of isolated oil were kept under N<sub>2</sub> atmosphere in glass vials for dispatch to contractors for chemical analysis of fatty acids (especially omega-3), vitamin D, vitamin A, and vitamin K. With the exception of sterol analysis, all chemical analyses were performed by four independent contract laboratories located in Canada, Germany, and Ireland. The contract-laboratory approach for chemical analysis was undertaken because of certain concurrent research work on burbot-oil processing in these laboratories. The maximum elapsed time between dispatch of sample vials (from storage at +4°C refrigeration) and sample analysis at a contract laboratory is 10 days.

The first step in the metabolic activation of vitamin  $D_3$  is the hydroxylation of carbon 25, primarily in the liver (Dusso et al. 2005). Although 25-hydroxyvitamin  $D_3$  is rec-

Table 2
Proximate analysis of burbot, *Lota lota*, caught in Athapapuskow Lake and Amisk Lake

	Athapapuskow Lake*			Amisk Lake**		
	sum	fish X- 06705C3	fish X- 06705C4	fish X- 06705C5	average of 3 fish	total of 3402 fish
Total length [cm]		61.0	54.6	61.0	58.9	51.2-61.2
Weight (wet, frozen) [kg]		1.889	1.913	1.876	1.876	3724
Isolated liver (wet) [kg]	0.203				0.0677	217
Isolated liver [% wet fish weight]					3.6	5.8
Dried solids content of isolated liver [%]					51.3	48.7
Oil yield [% oven-dried liver]	44					32.5
Liver oil yield [g per fish]					15.3	20.7
[g per kg fish]					8.1	18.4

<sup>\*</sup>Random sample of 3 fish from a 5-fish lot; liver isolated by Arbokem laboratory staff in Vancouver (March 2005).

<sup>\*\*</sup>Liver isolated in the field by commercial fishermen (January 2006).

ognized to be several times more potent than native vitamin  $D_3$ , its presence in fish has been noted to be extremely low at the level of <0.2  $\mu g$  per 100 g (Anonymous 2001a, Ovesen et al. 2003). It was decided that at these low levels, it is not practical to attempt to measure 25-hydroxyvitamin  $D_3$  in burbot liver oil or burbot tissue.

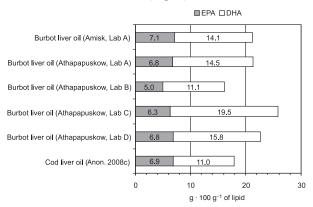
The thermal stability of the nutritional ingredients of burbot liver oil under simulated cooking conditions was evaluated. The test consisted of heating a sample of burbot liver oil in a stirred open beaker at 178 °C, just below the smoke point of the burbot liver oil.

#### RESULTS AND DISCUSSION

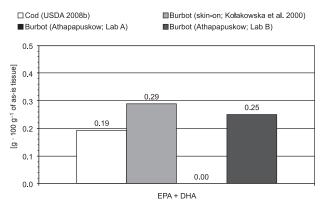
Omega-3 fatty acids. Key n-3 fatty acids (viz., C20:5 and C22:6) in burbot liver oil were analyzed using gas chromatography: **AOAC** Official 41.1.28A-996.06 (Anonymous 2002a); Derivitization of fatty acids to form methyl esters for flame-ionization detection; Detection limit = 0.01%. Fig. 1 shows that the quantity of these n-3 fatty acids compared favourably to that of reference cod liver oil. The larger-than-expected variability in DHA test results, especially those of Lab B and Lab C, might be due in part to the imperfect resolution of n-3 C22:5, n-6 C22:5, and n-3 C22:6 (DHA) peaks in the gas chromatographic analysis. Nevertheless, these results were perhaps not surprising as (freshwater) burbot and (saltwater) cod are from the same order of Gadiformes.

The total n-3 fatty acid (i.e., EPA + DHA) content of burbot tissue was found to be at the same low level as Atlantic cod tissue (Fig. 2). The discrepancy between the test results of the two laboratories could be attributed to the inherently very low level of combined EPA and DHA present in these tissue samples. For a daily intake target of 1 g of combined EPA + DHA, one would need to consume approximately 400 g of burbot tissue.

**Cholesterol.** Cholesterol content of burbot oil samples was analyzed gas chromatography and gas chromatography-mass spectrometry (Sample dissolution in pyridine for flame-ionization detection; Detection limit = 0.01%). Most fish oil contains a small amount of cholesterol. Burbot liver oil was found to have a similar level of cholesterol as cod liver oil (Fig. 3).



**Fig. 1.** Total C20:5 and C22:6 n-3 fatty acids in liver oil of burbot, *Lota lota* 



**Fig. 2.** Total C20:5 and C22:6 n-3 fatty acids in tissue of burbot, *Lota lota* (skin-off, except where noted)

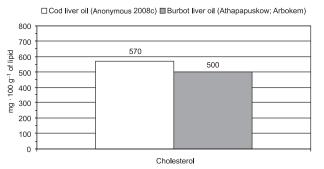


Fig. 3. Cholesterol in liver oil of burbot, Lota lota

Vitamin A. Vitamin A is a fat-soluble vitamin, which is of particular nutritional importance for the visual system in humans (Anonymous 2001b). Several carotenoids are precursors of vitamin A. Yellow and orange vegetables contain significant amounts of carotenoids. Certain fish and fish oil are also noted to contain a high quantity of vitamin A. Cod liver oil is known to contain a high quantity of vitamin A (in esterified form). In commercial preparation of cod liver and other food-use fish oil, only a fraction of the original vitamin A in the fish oil is retained during the deodorization operation.

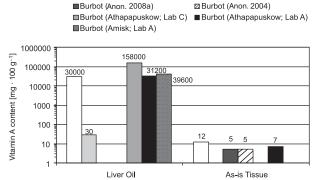
High-performance liquid chromatography method (Anonymous 1993), Chapter 36 modified. Ultra-Violet Detection limit = 10 International Units per 100 g) was used for the analysis of vitamin A. Fig. 4 shows the concentration of vitamin A (as retinol equivalent) in burbot liver oil to be in the range of 500 000  $\mu$ g, about 20 times higher than that reported in cod liver oil. However, the test data from JR Laboratories showed considerable lower value of vitamin A. Further testing of new samples of liver oil is recommended for establishing the actual range of vitamin A concentration.

Vitamin D. Vitamin D is a fat-soluble vitamin which is essential for the maintenance of normal blood levels of calcium and phosphate that are in turn required for, among other things, the normal mineralization of bone (Anonymous 2001b, 2002b, Trivedi et al. 2003), alleviation of certain mood and neurological disorders (Lansdowne and Provost 1998, Gloth et al. 1999, Garcion et al. 2002) and prevention of cancers (Schwartz et al.

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1997, Holick 2004, Townsend et al. 2005). Supply of vitamin  $D_3$  in humans is provided by its synthesis through the action of sunlight (ultraviolet light) on cholesta-5,7-dien-3-ol (7-dehydrocholesterol; provitamin  $D_3$ ) present in human skin, and by the consumption of certain foods, which are enriched with natural or synthetic vitamin D. Vitamin D is found naturally in very few foods. The common sources are fatty fish and fish liver oils. Egg yolk with reported high content of vitamin D arises from the feeding of synthetic vitamin D to laying hens.

□Cod (Anon. 2008b, c)



☐ Cod (Belitz and Grosch 1982)

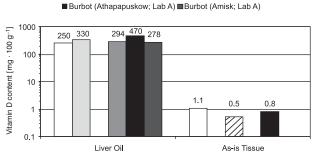
**Fig. 4.** Vitamin A content of burbot, *Lota lota* (1 μg vitamin A, retinol equivalent = 3.33 International Units)

The high level of vitamin D in burbot liver was first cited by Branion in 1930. There has been little or no studies reported on the presence of vitamin D in burbot liver oil during the past 70 years.

High-performance liquid chromatography (Anonymous 1993; Chapter 38; Ultra-Violet Detection limit = 10 International Units per 100 g) was used for the analysis of vitamin D. As given in Fig. 5, preliminary analysis showed the prevalence of vitamin D in burbot liver oil to be comparable to that reported in cod liver oil. It may be noted that in commercial preparation of cod liver and other fish oil for medicinal purposes, most of the natural vitamin D is removed by steam used in the oil deodorization process (Pigott 1996). Synthetic vitamin D (made commonly from the irradiation of ergosterol) is customarily added back into the deodorized fish oil for subsequent retail sale. Thus, the reported content of vitamin D in commercial fish oil products may be somewhat mislead-

□ Cod (Anon. 2008a)

Burbot (Anon. 2004)



□Cod (Belitz and Grosch 1982)

■Burbot (Athapapuskow; Lab C)

**Fig. 5.** Vitamin D content of burbot, *Lota lota* (1 μg vitamin A, retinol equivalent = 40 International Units)

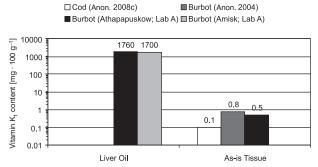


Fig. 6. Vitamin K<sub>1</sub> content of burbot, *Lota lota* 

ing, without a clear description of the processing history of the specified fish oil (Anonymous 1999).

Although synthetic vitamin  $D_2$  is generally assumed to have the same biological activity as natural vitamin  $D_3$ , in humans, there is recent evidence to suggest that vitamin  $D_2$  is less effective than vitamin  $D_3$  in increasing the circulating level of 25-hydroxyvitamin D (Trang et al. 1998, Armas et al. 2004). For the nutritional therapeutic market, it would be advantageous to retain the natural vitamin  $D_3$  found abundantly in burbot liver oil.

**Vitamin K.** Vitamin K is a fat-soluble vitamin that plays an important role in blood clotting and bone metabolism pertaining to the prevention of osteoporosis (Anonymous 2001b, Peterson et al. 2002, Ryan-Harshman and Aldoori 2004) and carotid artery elasticity (Cranenburg et al. 2007). The dietary intake of vitamin K by humans is generally through the consumption of certain leafy green vegetables (Anonymous 1994).

High-performance liquid chromatography (Ultra-Violet Detection limit = 0.1 mg per 100 g) was used for the analysis of vitamin K. Both Atlantic cod and burbot tissues have been reported previously to contain a small amount of vitamin  $K_1$  (Fig. 6). In the presently reported study, burbot liver oil was detected to contain a surprisingly high amount of vitamin  $K_1$  (~1.7 mg per 100 g of oil). Vitamin  $K_1$  is normally found in green leafy vegetables at the level of 500 to 1400  $\mu g$  per 100 g of edible portion (Anonymous 1994). As shown in Table 3, the content of vitamin  $K_1$  of burbot liver oil was 1 to 3 orders of magnitude greater than that found in non-Gadiformes fish components reported by Udagawa (2000). The exact origin of vitamin  $K_1$  in burbot liver oil is not well understood presently.

Thermal stability. Fig. 7 shows the apparent loss of n-3 fatty acid components of burbot liver oil to be somewhat greater than that of reference flax oil. This level of reduction may be compared to that reported by Kołakowska and Bienkiewicz (1999) for the 13% loss of EPA and DHA in a Baltic herring tissue matrix, upon microwaving-oven heating for 40 min.

The degradation of the fat-soluble vitamins was found to be substantially greater than that of n-3 fatty acids (Fig. 8).

**Commercial outlook.** Burbot belongs to a higher taxon of finfish of which its oil is rich in certain nutritional lipids. The production of burbot liver oil could afford a means of delivery of a single natural product containing the daily recommended intakes of n-3 fatty acids (i.e.,

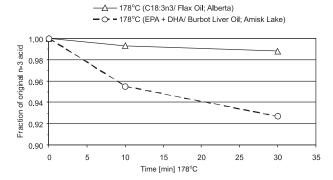
Table 4

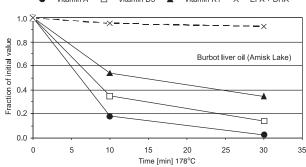
Table 3 Vitamin K<sub>1</sub> content of selected fish species

	Reference	Component	Vitamin K <sub>1</sub>
	Reference	Component	content [µg]*
Blue mackerel, Scomber australasicus	Udagawa 2000	Liver tissue**	9
Pacific saury, Cololabis saira	Udagawa 2000	Gonad tissue**	0.05
Marbled flounder, Pseudopleuronectes yokohamae	Udagawa 2000	Kidney tissue**	0.2
Sand sillago, Sillago ciliata	Udagawa 2000	Liver tissue**	0.2
Ayu, Plecoglossus altivelis	Udagawa 2000	Liver tissue**	2
Burbot, Lota lota	This work	Liver oil	1700-1760

<sup>\*</sup>µg per 100 g of oil (or wet weight).

<sup>\*\*</sup> highest content of Vitamin K<sub>1</sub> reported in all tissues studied.





**Fig. 7.** Thermal degradation of n-3 fatty acid components in liver oil of burbot, Lota lota

Fig. 8. Rate of disappearance of selected fat-soluble vitamins of liver oil of burbot, Lota lota

Nutritional content of one serving (5 g) of liver oil of burbot, Lota lota

Nutritional Component	Burbot Liver Oil*	USA Recommended Nutritional Allowance**	USA Maximum Upper Limit of Daily Intake	Reference
EPA + DHA [mg per day]	1000	500-1800	Probably >> 3000	Kris-Etherton et al. 2002
Vitamin A [µg per day]	1800	1000-1500	60 000-100 000	Anonymous 2001b
Vitamin D [µg per day]	20	5–15	50	Anonymous 1997
Vitamin K [µg per day]	85	60-120	Not determinable	Anonymous 2001b

<sup>\*</sup> representative data from this work;

EPA and DHA), vitamin A, vitamin D, and vitamin K, for Vieth et al. (2001) had previously suggested a safe intake blending into processed foods.

There are however vitamin safety considerations. If the set point was the delivery of 1 g of combined EPA + DHA per day, then the vitamin A and vitamin D contents of burbot liver oil may be exceeded with the present American (USA) "Recommended Nutritional Allowance - RNA" (Anonymous 1997, 2001b) limits. The problem is illustrated in Table 4, for a daily intake of 5 g of burbot liver oil to provide 1 g of combined EPA + DHA.

A practical strategy to overcome this RNA problem would be to blend burbot liver oil with a small amount of flax oil, which contains little or no vitamin A and vitamin D. It is interesting to note, however, that many researchers are now advocating the substantial increase of the recommended daily nutritional allowance intake of vitamin D (Hathcock et al. 2007, Vieth et al. 2007, Holick 2007).

level of 100 µg of vitamin D<sub>3</sub>.

#### **CONCLUSIONS**

The preliminary findings of the presently reported study suggest that burbot liver oil could offer similar nutritional attributes, viz., n-3 fatty acids, vitamin A and vitamin D, as Atlantic cod liver oil. The surprisingly high concentration of vitamin K<sub>1</sub> found in burbot liver oil was previously unknown.

#### **ACKNOWLEDGEMENT**

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<sup>\*\*</sup> For persons at least 9 years old.

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