SHORT COMMUNICATION

EPA and DHA Contents of Encapsulated Fish Oil Products

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Seventeen brands of encapsulated fish oil or fish oil concentrate products, purchased during the period 1984–88 over the counter in the United States, United Kingdom or Canada, were analyzed for their mg contents of eicosapentaenoic (EPA) and docosahexaenoic (DHA) acids per g of capsule contents. The mg contents were determined with respect to methyl tricosanoate internal standard by gas liquid chromatography (GLC) on a SUPELCOWAX-10 flexible fused silica capillary column. The alkyl ester and free fatty acid products showed very high levels of EPA (259–300 mg) and DHA (172–254 mg) whereas in the triglyceride oils EPA ranged from a low of 80 to a high of 250 mg, and DHA ranged from 78 to 156 mg, per g of capsule contents. The mg/g results indicate that the label claims for EPA and DHA for the majority of the products sampled are presented with reasonable accuracy.

Encapsulated fish oil and fish oil concentrate products have recently been criticized for their low contents of omega-3 fatty acids (1). It is known that retail products can be expected to show some variation from batch to batch but the figures given (1), i.e., 38% of claimed EPA (eicosapentaenoic acid) content and 85% of DHA (docosahexaenoic acid) content, are suggestive of deficiencies in analytical technique because in most products EPA ≥ DHA. As we have been monitoring these products for some time, we wish to present our data for a variety of encapsulated fish oil products analyzed by a chromatographic method developed expressly for this purpose.

EXPERIMENTAL

All samples were purchased over the counter in the U.S., the U.K., or Canada and were analyzed with an internal standard as proposed by Einig and Ackman (2). In brief, a sample capsule was opened and approximately 50 mg was weighed out into a screw-capped (Teflon-lined) centrifuge tube (15 ml). To this was added methyl tricosanoate (23:0) in the form of 1.0 ml of a solution in benzene containing 5 mg/ml. Boron trifluoride solution (1.0 ml, 7% BF_3) was added and the tube was flushed with nitrogen, capped, and heated for one hour at 100°C. After cooling the sample, water (3 ml) was added and the benzene solution of methyl esters was recovered. The aqueous layer was then extracted two more times with hexane (2 ml each). The benzene and the hexane extracts were combined and washed once with water, dried over Na_2SO_4, and concentrated slightly for injection into a gas-liquid chromatograph. Cyclohexane or even n-hexane can be substituted for the benzene if desired.

Gas-liquid chromatography was carried out on a flexible fused silica open-tubular column, 30 m × 0.25 mm i.d., coated with SUPELCOWAX-10 (a bonded Carbowax-20M polyglycol) in a Perkin-Elmer Model 8420 GC. The carrier gas was helium at 12 psig with a split ratio of 100:1. The oven temperature for programmed operation was initially 105°C for 8 min; then it was programmed at 3°C/min to 240°C with a final 10 min hold. Area data for whole oil analyses was corrected for flame ionization response (3,4) to give weight percent composition for volatile methyl esters. The EPA-23:0-DHA factors of 0.99–1.00–0.97 (2) were used for these specific ω-3 fatty acids. The majority of the analytical results of Table 1 are averages of two complete analyses of the contents of one capsule. Ethyl tricosanoate was employed with ethyl ester samples.

RESULTS

A whole fish oil chromatogram for this liquid phase is reproduced elsewhere (5) and the essential part of a typical chromatogram has also been published (2). Table 1 includes identification by trade name, distributor/manufacturer, label feature, label claim for EPA and DHA content, and figures from our analyses for EPA and DHA in mg/g of capsule contents and as percentages (w/w) of fatty acids.

DISCUSSION

There are several problems in determining the EPA and DHA contents of encapsulated fish oil products, not the least being their susceptibility to oxidation (6–8). This can take place during any stage of the analysis if due care is not taken. Should it have already occurred, especially during manufacturing or concentration steps, then polymers may be present. The fatty acids which are cross-linked in these polymers do not appear in GLC analyses but their mass reduces the concentrations in mg/g of EPA and DHA per gram of sample. The same dilution effect applies to residual solvents, to natural nonfatty acid components such as sterols, hydrocarbons or waxes, and to any materials such as deliberately added antioxidants. The area percent of methyl esters of fatty acids from GLC analysis is not accurate for w/w% EPA or DHA content of a sample, even when corrected for FID response (9), and gives only the results from volatile esters of fatty acids. The 23:0 (or other suitable internal standard) approach is the only GLC method that gives accurate w/w% data relative to content in the starting sample. This may be conveniently expressed as mg/g since many capsules are of 1 gram capacity.

HPLC is not at present a competitive method for EPA/DHA analyses since each detector must be individually calibrated for detector response with pure fatty acids (or esters)—a relatively expensive and potentially unreliable basis for quantitation.

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TABLE 1

Claimed and Experimental (as Determined by GLC with Internal Standard or by Normalization of Total Area Percent) Contents of EPA and DHA in Some Retail Fish Oil Nutritional Supplements Sold in Capsule Form, on the Basis of 1 g Capsules, with Product Type

Trade name, company and label "features"	Product type	EPA and DHA levels (manufacturers' claim)		EPA and DHA levels (determined by GLC)			
		EPA	DHA	EPA		DHA	
		mg/g	mg/g	mg/g	wt%[a]	mg/g	wt%[a]
Norwegian cod liver oil (TWINLAB)[b]	Oil	66-82	68-84	80	8.7	103	11.1
Biosaumon (Medicorp [France])[c] —Natural and without danger —No physico-chemical processing	Oil	120	160	81	8.4	106	11.2
MaxEPA 300 (Walgreen Labs Inc.) —Hi potency omega-3 fatty acids —Cold water natural fish oil	Oil	180	120	161	15.1	108	10.2
MaxEPA (Solgar Co., Inc.) —Marine lipid concentrate	Oil	180	120	138	15.4	96	10.9
Natural OMEGA-3 (Country Life) —Fish body oils[d]	Oil	180	120	162	16.8	110	11.6
Cardi-Omega 3 (Solar Nutritionals Inc.) —All natural MaxEPA fish oil concentrate	Oil	180	120	138	16.9	98	11.8
Your Life (P. Leiner Nutr. Prod. Inc.) —Natural fish oil concentrate	Oil	180	120	155	16.4	104	11.0
proto-chol (E.R. Squibb & Sons Inc.) —Natural fish oils	Oil	180	120	144	15.4	118	12.7
Natural brand OMEGA-3 (Sonergx Nutr. Prod.) —Fish oil concentrate	Oil	180	120	149	14.4	129	12.6
NATURE'S BEST (Nature's Best Food Suppl.) —Natural fish oil concentrate	Oil	180	120	114	12.3	156	16.9
PROMEGA (Parke-Davis) —Natural fish oil concentrate	Oil	280	120	256	27.3	124	13.2
Super EPA 500 (Walgreen Labs Inc.) —Hi potency omega-3 fatty acids —Cold water natural fish oil	Ethyl Ester	300	200	269	27.5	172	17.6
Cholesterex (Nutr. Prod. of Amer. Inc.) —Omega-3 fish oil concentrate —Purified natural fish oil concentrate[e]	Oil	200	85	167	18.0	78	8.5
OMEGA-'3' EPA SUPER 500 (Schiff Bio-Food Proc.) —Unsaturated fish oils[f]	Methyl Ester	300	200	302	29.0	211	20.6
Healthcrafts EPA-Forte (Booker Health Products [U.K.]) —Selected marine lipid concentrate	Ethyl Ester	310	210	259	27.7	173	18.8
NATURE'S PRIDE EPA PURE/700 (Nature's Products Inc.) —Highest EPA and DHA potency —Marine lipid concentrate[g]	FFA	333	250	265	29.6	254	28.8
PROMEGA PEARLS (Parke-Davis) —Natural fish oil concentrate	Oil	300 "omega-3" fatty acids per pearl[h]		258	25.7	113	11.5

Note: Many products contain 1-10 mg of tocopherols and some have residual cholesterol. These results are not corrected for such materials.

[a]Wt % is g fatty acid per 100 g of total fatty acids volatile in GLC analysis.

[b]Capsule size is 500 mg. Vitamin A—1250 I.U., Vitamin D—130 I.U. are on a capsule basis. EPA and DHA calculated for 1000 mg.

[c]Vitamin A—700 I.U., Vitamin D—70 I.U. per 1 g.

[d]Includes garlic 50 mg and ascorbyl palmitate 2 mg "in a blend of glycerin, soybean oil, lecithin and lemon oil."

[e]Fat <1 g per serving of two capsules, according to carton.

[f]Willow bark (10 mg) included.

[g]Capsule size is 1200 mg. Label claim values are calculated for 1000 mg.

[h]Pearls softgel size is 600 mg. Label claim value is given per softgel and may include fatty acids other than EPA and DHA.

In our experience, the w/w% figures for any one component in an analysis as complex as fish oils are usually within ± 5% for two different GLC units in one laboratory. This range of error takes into account the differences arising from split, column losses and/or peak coincidences, FID geometry and nonlinearity, and type of electronic area integration. It is extraordinarily difficult to achieve an "absolute" figure for any fatty acid as w/w% of total fatty acids, but the superiority of open tubular columns for peak resolution and the use of the 23:0 internal standard lend confidence to our data. That the 23:0 methodology is satisfactory is also confirmed by the evident similar origin of several different oils and of the two lots of ethyl esters. Our results indicate that label claims for EPA and DHA for the majority of the products sampled are presented with reasonable honesty. The deficiencies of Table 1 may be based on the less satisfactory packed-column GLC, or on faulty analytical technology. Products purchased at different times may also have different compositions not reflected in label statements but leading to other laboratories obtaining different results than those in Table 1.

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