Fatty Acid Composition of Lipids from the Contents of Rock Lobster (*Panulirus cygnus*) Cephalothorax

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ABSTRACT: The contents of rock lobster cephalothorax were analyzed for lipid content and fatty acid composition. They contain a diversity of saturated (35.5 ± 0.5%), monounsaturated (26.3 ± 1.7%), and polyunsaturated fatty acids, n-3 highly unsaturated fatty acids (11.5 ± 0.5%) among them. The possibility of using these products as a supplement to fish and food animals' diets is discussed.
JAOCS 73, 259–261 (1996).

KEY WORDS: Cephalothorax, fatty acid composition, fish meal, rock lobster.

Polyunsaturated fatty acids, especially n-3 highly unsaturated fatty acids (HUFA with ≥20 carbon and more than four double bonds) are believed to play an important role as dietary components for marine fish and farm animals (1–4). One source of polyunsaturated fatty acids could be the hepatopancreas and other tissues present in the cephalothorax of rock lobster. These materials are generally discarded during processing of harvested lobsters and may provide a low-cost supplement to commercial aquaculture diets. Australia is the world's largest producer and exporter of rock lobster, with the western rock lobster (*Panulirus cygnus*) accounting for 60% of the total Australian catch. The waste products of the industry, mostly lobster carapaces, are of low commercial value and are generally discarded. The present study was undertaken to determine the distribution of fatty acids in the contents of the western rock lobster cephalothorax, comprising mainly hepatopancreas, to evaluate its value as a supplement to commercial aquaculture diets. The potential of this material as an inexpensive source of HUFA was also studied.

EXPERIMENTAL PROCEDURES

The contents of frozen rock lobster carapaces were removed, thawed, and dehydrated in a domestic dehydrator (Harvest Maid Dehydrator-High Speed Model FD-1000, Upper Hutt, New Zealand). Partly dried material was minced and ground to a fine powder in a blender (Waring Commercial Blender, Finsbury, CT).

Extraction of lipid fraction. The samples of prepared powder were stirred as a 1:2 mixture with methanol/chloroform (1:30, wt/vol) solvent with a magnetic stirrer for one hour. The residues were reextracted twice by the same procedure. Supernatants were pooled, washed first with 0.88% KCl and then with methanol/0.88% KCl solution, and evaporated under vacuum. The residues, which contained the "lipid fraction" of rock lobster waste material, were weighed to establish the yield.

Sample preparation for gas chromatography/mass spectrometry (GC/MS) analysis. Lipid fractions (50 mg), dissolved in 1 mL toluene, were subjected to a methylation process by adding 2 vol of 1% vol/vol sulfuric acid in methanol and allowing it to stand for 12 h at 50°C. Five mL of 5% aqueous sodium chloride solution was added to each sample, followed by 10 mL hexane. The mixtures were thoroughly shaken and allowed to separate. The supernatant from each mixture was removed and stored, and the residue was reextracted with an additional 10 mL hexane. Supernatants for each sample were pooled and washed with 4 mL of 2% aqueous $KHCO_3$ solution. After phase separation, the supernatants of each sample were dried by passing through a small column of anhydrous sodium sulfate. These methylated lipid fractions in hexane were used for GC/MS analyses (5).

GC/MS analyses. The methylated lipid fractions, which contained the methylated fatty acids (FAME), were analyzed with a Hewlett-Packard (Avondale, PA) GC/MS Model 5971. Chromatography was performed on a DB 23 column (i.d. 0.32 mm and film thickness 0.25 μ; J&W Scientific, Folsom, CA) with helium as the carrier gas. The initial column temperature was 50°C for 5 min, followed by an increasing gradient of 5°C/min to a temperature of 240°C, with the latter temperature being held for at least another 20 min. Run times were approximately one hour. Individual fatty acids were identified by means of known retention times, standards, and mass spectra and quantitated by introducing an internal standard (nonadecanoic acid) of known weight before methylation. The added nonadecanoic acid could then provide a known ratio of area integration to weight of standard concentration, allowing the weight of the other fatty acids to be calculated from their area integrations.

RESULTS AND DISCUSSION

Crustacean cephalothorax contains the organs of the respiratory, circulatory, digestive, and excretory systems. These or-

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gans include heart, gills, stomach, hepatopancreas, and excretory organs or green glands. Hepatopancreas takes up more space within the cephalothorax than any other single organ.

The hepatopancreas was considered to be the most important part of the cephalothorax for the purposes of this investigation. However, to facilitate the removal of the waste material, the hepatopancreas was not separated from other cephalothorax components. Such an approach makes the process of obtaining the waste material more practical.

The yield of lipid fraction obtained by the extraction procedure for four samples was 19.4 ± 1.0% w/w of dry weight. The principal fatty acid composition of the lipid fraction of the contents of rock lobster cephalothorax is shown in Table 1.

Total lipid investigated showed a relatively wide range of saturated fatty acids, which comprised 35.5% of total fatty acids. The most abundant of these was palmitic acid, 16:0. Odd-numbered carbon acids were represented by pentadecanoic, heptadecanoic, and nonadecanoic acids, which were found in small amounts. Long-chain monoenoics (20:1 and 22:1) were also present in relatively small amounts. The percentage of polyunsaturated fatty acids was 22.6%, with n-3 HUFA accounting for 50.8% of them. This distribution of fatty acids may be partly due to the diet of the rock lobster, which includes seagrasses and red and green algae as important nutritional components (6).

The raised level of interest in HUFA is a result of studies on the relationship between HUFA consumption and prevention of some human diseases (7,8). The main source of n-3 fatty acids in human diets is fatty fish. However, many farm-raised fish are low in n-3 fatty acids because their diets are formulated mainly from agricultural products (4). The use of fish meal in aquaculture diets increases the content of these fatty acids. Not only does aquaculture production have a great demand for fish meal in feeds, but fish meal is also used in feeds of poultry, pigs, cattle, and other farm animals. The use of fish meal in poultry feeds results in the increased poultry n-3 fatty acid content, which enhances their nutritional value (4).

It is interesting to compare the fatty acid composition of lipids from waste material from rock lobster with that of lipids from menhaden, an Atlantic fish caught primarily for production of fish meal and oil. Menhaden oil is not as rich in saturated fatty acids as rock lobster's cephalothorax contents. Saturated fatty acids content is 20.3–33.7% (9,10) as com-

TABLE 1
Fatty Acid Composition of the Contents of the Western Rock Lobster Cephalothorax

Fatty acid	% of Total fatty acids[a] (mean ± SEM)	mg/1 g of Dry sample (mean ± SEM)	mg/1 g of Lipid extract (mean ± SEM)
14:0	2.94 ± 0.029	6.42 ± 0.407	36.05 ± 1.046
15:0	1.00 ± 0.018	2.22 ± 0.128	12.36 ± 0.206
16:0	17.47 ± 0.208	38.20 ± 2.488	214.65 ± 6.330
16:1n-7	6.40 ± 0.080	13.97 ± 0.918	78.57 ± 2.390
16:1n-5	0.31 ± 0.010	0.67 ± 0.035	3.82 ± 0.094
16:2n-4	0.50 ± 0.012	1.07 ± 0.103	6.01 ± 0.350
17:0	1.15 ± 0.022	2.52 ± 0.196	14.17 ± 0.633
18:0	11.76 ± 0.081	25.74 ± 1.844	144.53 ± 5.110
18:1n-9	11.99 ± 0.785	25.91 ± 0.971	146.47 ± 5.496
18:1n-7	2.89 ± 0.231	4.55 ± 0.354	26.80 ± 2.364
18:2n-6	1.58 ± 0.032	3.46 ± 0.258	19.45 ± 0.808
19:0	0.57 ± 0.023	1.26 ± 0.094	7.06 ± 0.328
18:3n-3	1.09 ± 0.028	2.38 ± 0.180	13.35 ± 0.591
18:4n-3	0.55 ± 0.017	1.21 ± 0.094	6.78 ± 0.300
20:1n-9	2.49 ± 0.289	4.62 ± 0.442	26.56 ± 1.329
20:1n-7	1.32 ± 0.013	2.86 ± 0.175	16.12 ± 0.403
20:4n-6	7.02 ± 0.191	15.46 ± 1.439	86.59 ± 4.927
20:4n-3	0.38 ± 0.032	0.80 ± 0.008	4.54 ± 0.179
20:5n-3	5.62 ± 0.178	12.38 ± 1.204	69.27 ± 4.257
22:0	0.58 ± 0.054	1.28 ± 0.009	7.76 ± 0.635
22:1n-11	0.57 ± 0.074	1.23 ± 0.115	6.89 ± 0.728
22:1n-9	0.33 ± 0.003	0.72 ± 0.063	4.09 ± 0.181
22:5n-3	1.69 ± 0.065	3.73 ± 0.382	20.83 ± 1.388
22:6n-3	4.17 ± 0.170	9.20 ± 0.966	51.40 ± 3.550
Totals			
Saturates	35.47 ± 0.237	77.64 ± 0.448	436.58 ± 1.180
Monoenes	26.30 ± 0.876	54.53 ± 0.184	309.32 ± 0.829
n-6	8.60 ± 0.195	18.92 ± 0.731	106.04 ± 2.497
n-3	13.50 ± 0.257	29.70 ± 0.267	166.17 ± 0.959
n-3 HUFA[b]	11.48 ± 0.257	25.31 ± 0.530	141.50 ± 1.905

[a]Unidentified fatty acids are not included in the table.
[b]n-3 Highly unsaturated fatty acids (HUFA) are part of n-3 value and include 20:5n-3, 22:5n-3, and 22:6n-3.

pared with 35% in rock lobster (Table 1). The percentage of linoleic acid is similar in both lipids, whereas the relative content of highly unsaturated fatty acids in menhaden oil is more than twice as much as in rock lobster waste material (13.4–14.3% of 20:5n-3 and 9.6–12.4% of 22:6n-3 as compared with 5.62 and 4.17%, respectively) (Refs. 9,10; Table 1). However, menhaden fish meal has 9.13% lipids, whereas cephalothorax contents of rock lobster provides 19.4% lipids.

The presence of 11.5% n-3 HUFA in the lipids from rock lobster cephalothorax contents makes this readily available waste product valuable as a source of essential fatty acids for farmed species.

The use of this product in dietary formulations for a cultured prawn species, *Penaeus monodon,* is currently being studied. Prawn farming and other aquaculture industries in Australia have generated a significant demand for locally-produced aquaculture feeds. The volume of rock lobster waste that could be available for aquaculture feed production, approximately 200 tons p.a, would be sufficient to help satisfy the demand.

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[Received January 16, 1995; accepted November 16, 1995]