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Presence of pharmacologically active fatty acids in selected edible bivalve species from coastal lagoons of Sri Lanka.

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Abstract

KEY WORDS: EDIBLE BIVALVES; OMEGA-3 POLYUNSATURATED FATTY ACIDS, CORONARY HEART DISEASES; DHA; EPA

Consumption of seafood, more precisely fish lipids has been recognized to provide pharmacologically active fatty acids known as omega - 3 fatty acids that exert a number of beneficial health effects in human beings. The present investigation was undertaken (in year 2001) to determine the fatty acid profiles of four edible bivalve mollusks (*Crassostrea* sp., *Anadara* sp., *Meretrix* sp. & *Gaffrarium* sp.) inhabiting abundantly in four major lagoons in Sri Lanka namely Bolgoda, Negombo, Chilaw and Kalpitiya.

The fatty acid profiles of the bivalve meat were analyzed using Gas Chromatography. The predominant fatty acids found in the bivalve meat of all four species were Palmitic acid (16:0), Palmitoleic acid (16:1), Stearic acid (18:0), Oleic acid (18:1 n-9), Vaccenic acid (18:1 n-7), Eicosapentaenoic acid (20:5 n-3, EPA) and Docosahexaenoic acid (22:6 n-3, DHA). *Anadara* sp. from Negombo lagoon recorded the highest percentage of saturated fatty acids (43%) whereas *Gaffrarium* sp. from Kalpitiya lagoon had the highest amount of monounsaturated fatty acids (24%). *Crassostrea* sp. collected from Bolgoda lagoon was reported to have the highest content of omega-3 polyunsaturated fatty acids (PUFA) which was 36.5% of the total fatty acids and the highest amount of total PUFAs (47.5%) as well. The two most important pharmacologically active fatty acids, EPA and DHA were also recorded in highest amounts in the *Crassostrea* sp., which accounted for more than 70% of its total omega-3 PUFAs. The EPA levels ranged from 5-13% and DHA levels from 6-15% of the total fatty acids among the four bivalve species analyzed. The results of the present study indicate that the bivalve species studied can be considered a potential source of pharmacologically active fatty acids, particularly belonging to the Omega-3 series.

Introduction

Bivalves remain as an under-exploited aquatic resource in Sri Lanka due to low consumer demand. However, consumption of bivalves is increasingly becoming popular world over due to its high nutritional quality and extreme delicacy. In Sri Lankan coastal lagoons, there are several edible bivalve species that inhabit abundantly in semi-muddy bottoms in the shallow waters. Even though they are not yet fully utilized as a human food, they contribute to other industrial purposes such as in kilns and in the shrimp farming industry as natural feed (Joshep, 1993).

Lipids in seafood are considered as the main source of health related fatty acids (omega-3 fatty acids) that exert number of beneficial health effects in the human body (Ackman, 1988, Edirisinghe *et al.*, 1998, 2001). Of all the fatty acids, omega 3 (n-3) polyunsaturated fatty acids (PUFA) are high in lipids of seafood and are increasingly being used as pharmaceutical drugs for a number of pathophysiologies. These fatty acids are reported to have the ability to decrease cholesterol, triacylglycerols and very low density lipoproteins and low density lipoproteins in blood. Of the omega-3 PUFAs, especially eicosapentaenoic acid (EPA) and decosahexaenoic acid are reported to be responsible for these effects minimizing the development of a number of chronic degenerative diseases (Ackman, 1998; Kinsella, 1998). Even though the fatty acid profiles of various edible fish species in Sri Lanka have been documented by some authors in previous studies (Edirisinghe *et al*, 1998, 2001; Jayasinghe *et al*, 1992), the information on the fatty acid profiles in the edible bivalve species is scanty. Hence, the present study was carried out to assess the lipid content and the fatty acid profiles in the meat of selected edible bivalve species which are abundant in Sri Lankan lagoon waters.

Materials and Methods

Four selected bivalve species namely *Crassostrea madrasensis* (Bolgoda lagoon), *Anadara antiquata* (Negombo lagoon), *Meretrix casta* (Chilaw lagoon) and *Gaffrarium tumidum* (Kalpitiya lagoon) were used for the study. Samples were collected from the natural bivalve beds and they were transported to the laboratory in polyethylene bags under chilled conditions. Immediately after being taken to the laboratory, specimens were cleaned with tap water to remove the mud and dirty particles and then the total lengths and weights of the bivalves were measured. Using a knife, bivalves (30 individuals) were de-shelled, and sucked into clean glass containers separately and kept in a freezer at –20 ^{0}C until analysis.

The lipids in the bivalve meat were extracted and measured by modified Bligh and Dyer method (Hanson and Olly 1963). The fatty acid methyl esters (FAME) of the lipids were prepared by base hydrolysis followed by transesterification. These FAMEs were separated by packed- column gas- chromatography on a Shimadzu GC-14A Gas Chromatograph using hydrogen flame ionization detector and helium as a carrier gas at a flow rate 35ml/min at 175 ^{0}C. The temperature programming used was as follows: 20 minutes at 175 ^{0}C, then increasing up to 240 ^{0}C at a rate of 3 ^{0}C/minute and 10 minutes of holding time at 240 ^{0}C. Comparing with retention times of methyl esters in a standard lipid mixture, the peaks obtained from the methyl esters were identified. The absolute amounts of fatty acids were determined using heptadecaenoic acid (C 17) as an internal standard. Duplicate analyses were carried out for each sample of the bivalve species analyzed. The data were statistically analyzed using the MINITAB Version 10.2 computer software programme.

Results and Discussion

Table 1 shows the fatty acid profiles of the four edible bivalve species analyzed during the present study while a summary of major groups of fatty acids (% of the total fatty acids) in the bivalves are given in table 2.

The four bivalve species tested were reported to contain low lipid level (<2%) on wet weight basis. The most abundant fatty acids were Palmitic acid (C 16:0), Palmitoleic acid (C 16:1), Stearic acid (C 18:0), Oleic acid (C 18:1), eicosapentaenoic acid (C 20: 5 n-3, EPA) and Docosahexaenoic acid (C 22:6 n-3, DHA) but the most predominant acid was Palmitic acid, which contributed 19 to 25% of total fatty acids.

In the present study, saturated fatty acids (SFAs) contributed the major part of the fatty acid profile of the bivalve species except in *Crassostrea* sp. and ranged from 32.7 (in *Crassostrea* sp) to 43.1% (in *Anadara* sp.) of the total fatty acids. This group consisted of myristic acid (C 14:0), pentadecanoic acid (C 15:0), palmitic acid (C 16:0), heptadecanoic acid (C 17:0) and Stearic acid (C 18:0) with Palmitic acid contributing the largest proportion (19-25%). Monounsaturated fatty acids (MUFAs) consisted mainly of Palmitoleic acid (C 16:1) and Oleic acid (C 17:1) and minor quantities of eicosenoic (C 20:1) and erucateic acid (C 22:1). The highest percentage (24.2%) of MUFAs was recorded in *Gaffrarium* sp. (Cockle), whereas the lowest percentage was from *Meretrix* sp. The percentage of PUFAs ranged from 33% to 48%. The highest percentage of PUFAs was recorded from *Crassostrea* sp and the lowest percentage from the *Gaffrarium* sp. All the species contained more than 25% of Omega-3 PUFAs of the total fatty

acids but, they contained comparatively low levels of Omega-6 PUFAs. The amounts of Omega-3 PUFAs in *Crassostrea* sp. (Oyster) and *Meretrix* sp. (Clam) were significantly higher ($p<0.05$) than that recorded from the *Gaffrarium* sp. (Cockle). The Omega-3 PUFAs mainly consisted of C 18: (n-3), C 20:4 (n-3), C 20:5 (n-3) and C 22:6 (n-3). In all four species, the two most biologically active PUFAs, EPA and DHA contributed more than 45% of Omega-3 PUFAs. The highest percentage of EPA and DHA was recorded in the *Crassostrea* sp. collected from the Bolgoda lagoon.

The present investigation provides valuable information on the lipids of some edible bivalve species inhabiting Sri Lankan coastal lagoons especially in terms of fatty acid profiles. All four species studied contain omega 3 polyunsaturated fatty acids over 25% of their total fatty acids and over 45% biologically active PUFAs (DHA & EPA) of the total omega-3 PUFAs. Of the four species studied, *Cassotrea* sp. (oyster) had the highest content (36% of the total fatty acids) of omega-3 PUFAs where EPA & DHA are predominant (over 70% of Omega-3 PUFAs). Edirisinghe *et al* (1998) also observed a similar trend in the levels of total omega-3 PUFAs in 14 edible small pelagic fish species caught from Sri Lankan coastal waters.

Conclusion

The results of the present investigation show that the edible bivalve species studied could be considered as a potential source of important omega-3 fatty acids which play a significant role in human health preventing coronary heart diseases and other pathophysiologies. Therefore, encouragement of consumption as well as culturing of these edible mollusk varieties will be extremely beneficial to developing countries like Sri Lanka as they provide a rich source of health related nutrients at a low cost and as a delicacy to seafood consumers.

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Table 1: Average value of fatty acids (% of the total fatty acids) in the bivalve species analyzed (n=30).

Fatty acid %	Oyster (*Crassostrea* sp.)	Clam (*Meretrix* sp.)	Cockle (*Gaffrarium* sp.)	Blood Cockle (*Anadara* sp.)
C 14:0	0.86	1.41	2.27	3.74
C 15:0	0.41	ND	1.08	0.57
C 16:0	20.92	19.04	25.40	24.19
C 17:0	2.13	2.27	2.73	2.42
C 18:0	7.91	6.95	8.09	11.09
C 20:0	0.23	2.99	ND	0.43
C 22:0	ND	3.06	ND	ND
C 23:0	0.24	1.55	2.16	0.69
C 14:1	ND	ND	ND	ND
C 16:1	2.09	3.40	8.65	6.63
C 17:1	0.25	ND	ND	ND
C 18:1 (n-9)	11.89	7.66	5.50	3.37
C 18:1 (n-7)	3.32	2.09	5.41	4.29
C 20:1 (n-9)	0.62	1.68	3.68	3.78
C 22:1 (n-9)	ND	2.14	0.97	4.69
C 16:2 (n-4)	0.34	ND	1.20	0.51
C 16:2 (n-6)	0.23	ND	2.35	1.23
C 18:2 (n-6)	10.05	1.72	0.99	2.29
C 20:2 (n-6)	ND	ND	2.22	1.63
C 20:3 (n-6)	ND	ND	ND	ND
C 20:4 (n-6)	0.75	3.62	1.64	0.50
C 16:4 (n-3)	1.96	7.20	2.89	3.24
C 18:4 (n-3)	0.91	ND	ND	0.46
C 18:3 (n-3)	1.35	1.69	1.14	0.94
C 20:4 (n-3)	3.08	4.96	4.50	5.10
C 20:5 (n-3)	12.89	4.64	7.16	7.08
C 22:4 (n-3)	0.39	2.92	1.06	1.36
C 22:5 (n-3)	1.10	2.31	2.34	1.44
C 22:6 (n-3)	14.82	12.26	6.49	10.23

ND = Not detected

Table 2: Summary of the major groups of fatty acids (% of the total fatty acids) in the four edible bivalve species analyzed.

	Oyster (*Crassostrea* sp.)	Clam (*Meretrix* sp.)	Cockle (*Gaffrarium* sp.)	Blood Cockle (*Anadara* sp.)
SATURATES	32.71	37.27	41.73	43.13
MONOENES	18.19	16.98	24.21	22.77
POLYENES	47.55	41.30	31.85	35.49
n-6 PUFA	11.04	5.33	7.21	5.66
n-3 PUFA	36.52	35.97	25.65	29.84
EPA	12.89	4.64	7.16	10.23
DHA	14.82	12.26	6.49	5.10

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