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PROXIMATE COMPOSITION, FATTY ACIDS AND CHOLESTEROL CONTENTS OF CRAB *Portunus pelagicus* , FROM NORTH WEST ARABIAN GULF

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ABSTRACT

The Crab *Portunus pelagicus* have been collected in October 1996 from the Northwest Arabian Gulf region. The Biochemical composition of muscle for male and female were studied. From the chemical analysis, it was observed that male contained maximum moisture and protein, while female recorded the minimum. The female contained highest fat, ash, cholesterol, iodine value and free fatty acid contents. Both male and female treatments resulted in high significant differences (P ? 0.01) in protein and ash and significant differences (P ? 0.05) in fat and free fatty acids between male and female. The fatty acid composition of the muscle's lipid was determined by Gas Chromatography (GC). Certain major fatty acids varied widely between male and female. The data presented could be useful in evaluating the nutritional status of populations, and then the chemical composition.

INTRODUCTION

Protein, lipid, minerals and carbohydrates are the basic components of aquatic organisms, and all have distinct roles. The proteins do not seem important in primitive organisms but some invertebrates use amino acids for ionic balance and in moving up the evolutionary scale. (Gurr and Harwood, 1991). Lipids in marine organisms are associated with a variety of functions, reflecting special biochemical and environmental conditions of the marine milieu. Lipids are the major metobolic reserve in most marine animals, including mammals , fish and crustaceans (Napolitano and Ackman, 1992). Saturated and monounsaturated fatty acids are generally not essential, but certain polyunsaturated fatty acids (PUFAs) are essential dietary nutrients for reasons associated with their specific structures (Napolitano, 1990).

Decosahexanoic acid (DHA) may reduce cardiac arrythmia and the role of eicosapentanoic acid (EPA) originally viewed as a precursor of PG_13 and

thus reducing platelet aggregation, has been extended to reduced vasoconstriction, and improved red blood cell deformability which in all factors potentially reducing) cardiac stress (Ackman, 1994 a). The fatty acids composition of the oil was investigated in the late 1950; as part of the interest in relating dietary polyunsaturated fatty acids to serum cholesterol levels in humans (Ackman, *et al.,* 1981). The long chain (C20, C22) monoethylenic fatty acids of the depot fats of marine animals are plausibly derived from their diet (Ratnayaka & Ackman, 1979). In animal tissues sterols are associated with a number of functions and one of the most important is their role as structural components of cellular membranes (Napolitano, *et al.*, 1993). Free fatty acids (FFA) are very minor components which resulted from lipid hydrolysis by micro-organisms enzyme (Hantoush, 1998).

It is clear that at iodine values (I.V.) of 150 or less, there is a relatively low percentage of saturated acids, and correspondingly a high proportion of two specific monoenes, 20: 1 and 22: 1 (Ackman, 1988).

This report presents a study of the biochemical composition to obtain information on the porximate composition, free fatty acids cholesterol, iodine value and the concentrations of the major fatty acids for muscles of Crab *Potunus pelagicus* in order to provide nutritional data for dietary planning and web foods.

MATERIALS AND METHODS

Sample collection:

Ten male and twelve female specimens with (11.7-15.2) cm in length, (5.6–7.8) cm in widith and (114.4-256.9) in weight of Crab *Potunus pelagicus* have been collected during October 1996 from Northwest Arabian Gulf (Fig. 1). The fresh samples were kept in crushed ice in polystyrene cool box and transfered to the laboratory.

Chemical Analysis:

Proximate Composition:

For each male and female crab, the muscles were freeze–dried, ground, sieved and analyzed for moisture, protein, lipid and Ash, in triplicate. One–gram samples were used for moisture determination based on A.O.A.C. (1984) method (7.003). Percent protein was determined from a one-gram portion of dried sample by Lowry, *et al.,* (1951) method. Total lipids were extracted following the method described by I.U.P.A.C (1979). Ash was determined on one gram of the dried sample according to A.O.A.C (1984) method (7.009). All data were expressed on a wet weight basis.

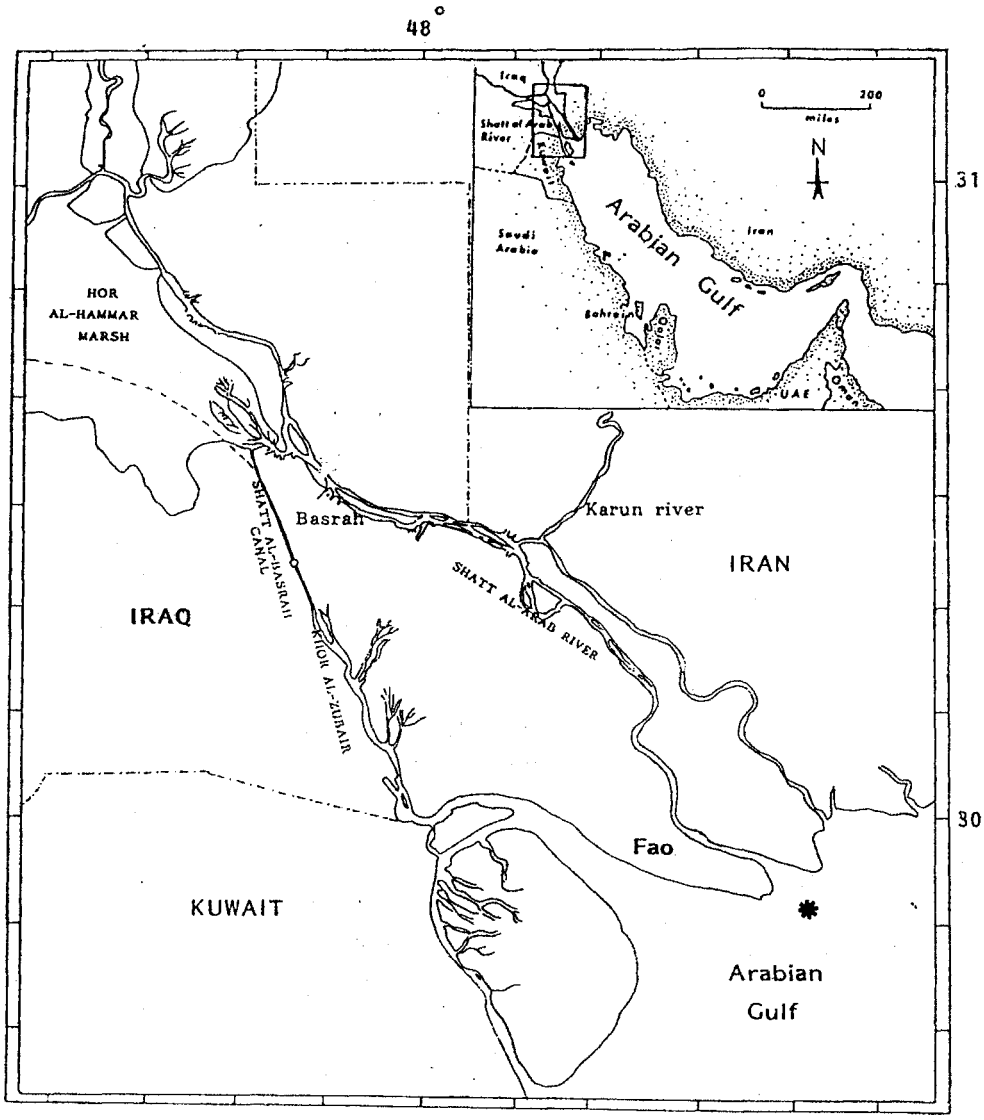

Fig. 1. Map of southern part of Iraq showing the sampling station*

The Fatty Acids and Iodine Value Analysis:

The extracted total lipids in the sub – samples were also analyzed for fatty acids. Fatty Acid Methyl Esters (FAMEs) were prepared using 14 % BF_3 in methanol according to a modified method described by Al-Saad and Al-Timari (1993). Analysis of FAMEs were carried on perkin – Elmer Sigma 300 Capillary Gas Chromatography with a flame ionization detector (FID) using a SE 30 WCOT column 50 m with He as a carrier gas (1.5 ml min^{-1}). Temperature program from 70 °C for 4 min to 300 °C for 30 min at rate 4 °C min^{-1}. Injector and detector port temperatures were (300 °C and 320 °C) respectively. Fatty acids were identified by comparing retention times with those of known standards.

The Iodine value determined by the Hubl method as described by I.U.P.A.C (1979).

The Cholesterol Analysis:

For each sample, 0.5 gm of the total lipid collected was weighted for cholesterol analysis with Liebermann – Burchard Reaction using the method, which described by (Wootton, 1974). The samples absorption was reading in a spectrophotometer (Shimadzu UV – 150 – 02) at 620 nm wavelength.

The Free Fatty Acids Analysis:

Total free fatty acids were determined according to the method of Windsor & Barlow (1981) using ethanol: diethyl ether (1:1 v/v) mixture as extracting solvent.

The Statistical Analysis:

Data were treated statistically by Complets Randomized Design (CRD). Revised Least Significant Differences (RLSD) was employed to detect the significant differences between means (Steel & Torrie, 1960).

RESULTS AND DISCUSSION

Results of the proximate chemical composition , cholesterol free fatty acids and iodine value for male and female crabs and their significant differences are shown in Table (1). Protein of male (14.090 %) was generally higher than that of the female (13.367 %), and it's content was differ significantly (p? 0.01). The overall mean protein for the muscle is less than the (`18.2 %) value for muscle of Crab (*Portunus trituberculatus*) reported by Iwasaki and Harada (1985). The lowest amount of ash in male (1.698 %) is perhaps

Table (1). Mean values and statistical analysis of Proximate Composition, Tree Fatty Acids, Cholesterol and Iodine Value in the muscles of male and female Crab.

Biochemical Constituents	Male		Female		(F)	Revised Least Significant Differences	
	Mean	SD ±	Mean	SD ±		5 %	1 %
Moisture %	81.216	0.547	80.386	0.757	0.52 NS	-	-
Protein %	14.090	0.140	13.367	0.067	43.41**	0.246	0.365
Ash %	1.698	0.118	2.329	0.028	53.75**	0.194	0.286
Fat %	3.227	0.164	3.699	0.112	11.35*	0.327	-
Free Fatty Acids % of lipid	1.359	0.148	1.993	0.239	10.17*	0.464	-
Cholesterol mg/100 g lipid	174.311	8.515	185.158	5.017	2.41NS	_	_
Iodine Value	93.31	4.79	105.36	5.68	5.26 NS	_	_

N.S. : Not Significants Differences.
** : Significant Differences at ($P<0.01$).
* : Significant Differences at ($P<0.05$).

related to higher moisture, as explained by Mustafa and Medeiros (1985). Ash was affected significantly (p?0.01) by sexes treatment.

Both crab sexes are high in moisture content, it is highest in male (81.216 %) and lowest in female (80.386 %), with no significant differences (p>0.05). Because of the relationship between moisture and fat which is in a good agreement with that of Al-Aswad, *et al.*, (1980); Al-Habbib, *et al.*, (1986) and Hantoush, *et al.* (1999), the fat content in female (3.699 %) was higher than that in male (3.227 %). The lipid content in both muscles showed significant difference (p?0.05).

In crab cholesterol was at relatively higher levels in female (185.158 mg/100g lipid) than in male (174.311 mg/100 g lipid), but the differences was not significant. Many studies have given different results for the same or similar species, partly due to the problem of sampling process, but primarily because of differences in the methods for the cholesterol determination (Walton, *et al.*, 1989).

The variation in sterol composition may be reflects differences in the quality of available food (Napolitano, *et al.* 1992).

The values for the moisture and cholesterol in muscle of crab reported here were higher than that determined by Iwasaki and Harada (1985), but the lipid content was at relatively good agreement with that, reported (3.9 %) in previous literature.

The propertion of free fatty acids (FFA) was relatively low in both male and female (<2.0 % of the total lipid). There was significant differences (P?0.05) in FFA between male and female. The overall mean (FFA) for both sexes are less than the (3.8 %) value for muscle of the Queen Crab (*chionocetes opilio*) reported by Addison, *et al.*, (1972).

The fatty acid concentration per gram of lipid is presented in Table (2). The major fatty acids in the lipid of carb's muscle were palmitic (16:0), stearic (18:0), palmitoleic (16:1), gadoleic (20:1), eicosapentenoic (20:5) and docosahexanoic (22:6) representing 0.31, 0.26, 0.40, 0.28, 0.84 and 0.33 % of the lipid in male, and 0.35, 0.15, 0.32, 0.35, 1.12 and 0.50 % of the lipid in female, respectively. The crustacea are generally good sources of PUFAs and are also low in saturated fatty acids, but are not low in cholesterol (Ackman, 1995). The lower temperatures of the environment may require more highly unsaturated molecules in an important lipids (Ackman, 1982). The polyunsaturation level of these poly-lipid fatty acids is inversely correlates with the ambient temperature, and an increased unsaturation of fatty acids may result in more disordered lipid phase in the membrane, and an increase in the degree of unsaturation in the acyl chains results in a lowering of transition temperature (Napolitano *et al.*, 1992).

Table (2). Fatty Acids concentrations (per gram of lipid) in the muscles of male and female Crab *Portunus pelagicus.*

Fatty Acids	Common Name	Chemical structure	Sex	
			Male	Femal
14:0	Myristic acid	$CH_3 (CH_2)_{12} COOH$	0.09	Trace
16: 0	Palmitic acid	$CH_3 (CH_2)_{14} COOH$	0.31	0.35
18:0	Stearic acid	$CH_3 (CH_2)_{16} COOH$	0.36	0.15
Total of Saturated Fatty Acids (SFAs)			0.66	0.55
16:1	Palmitoleic acid	$C_{15} H_{29} COOH$	0.40	0.32
18:1	Oleic acid	$C_{17} H_{33} COOH$	0.07	0.13
20: 1	Gadoleic acid	$C_{19} H_{37} COOH$	0.28	0.35
22:1	Cetoleic acid	$C_{21} H_{41} COOH$	0.23	0.11
Total of Mono Unsaturated Fatty Acids (MUFAs)			0.98	0.91
18:2	Linolenic acid	$C_{17} H_{31} COOH$	0.10	0.10
18:3	Linolenic acid	$C_{17} H_{29} COOH$	0.12	0.17
20:5	Eicosapentanoic acid	$C_{19} H_{29} COOH$	0.84	1.12
22:6	Docosahexanoic acid	$C_{21} H_{31} COOH$	0.33	0.50
Total of Polyunsaturated Fatty Acids (PUFAs)			1.39	1.89
Total Fatty Acids (% of Lipid)			93.90	89.21

C2O and C22 polyunsaturated fatty acid could accumulate in marine animal depot fats directly from absorption of long – chain fatty acids of planktonic algae (Ackman, 1994b). The values for precent fatty acid content agree with those reported by Ackman and McLeod (1988) as explained in Table (3).

The mean iodine value (105.36) for female greater than that for male (93.31) with not significant differences ($P>0.05$) between both sexes. Because of the fatty acid interrelationships with iodine values, it was possible to develop an empirical formula:

PUFAs % = 1.668 + 0.042 (Iodine value oil – 100)

REFERENCES

Ackman, R.G. 1982. Fatty acid composition of fish oils. In "Nutrition Evaluation of Long – chain fatty acids in fish oils" . S.M. Barlow and M.E. Stansby , Eds. Academic press, London, pp: 25-88.

Ackman, R.G. 1988. Oils and fats group international lecture, the year of fish oils. Chemistry and Industry, March 7: 139-144.

Ackman, R.G. 1994 a. Animal and Marine lipids. In "Technological Advances in improved and alternative sources of lipids". B.S. Kamel and Y. Kakuda, Eds. Blackie Academic and Professional, an Imprint of Chapman and Hall. London, pp: 292-328.

Ackman, R.G. 1994 b. Seafood lipids. In " Sea foods: Chemistry, Processing Technology and Quality ". F. Shahidi and J.R. Botta, Eds. Blackie Academic and Professional, an Imprint of Chapman and Hall. London, pp: 34-49.

Ackman, R.G. 1995. Composition and Nutritive value of fish and shellfish lipids. In "Fish and Fishery Products. Composition, Nutritive Properties and Stability". A. Ruiter, Ed. Cab. International, UK., pp: 156-177.

Ackman, R.G. and McLeod, C. 1988. Total lipids and nutritionally important fatty acids of some Nova Scotia fish and shellfish food products. Can. Inst. Food Sci. Technol. J. 21(4): 390-398.

Ackman, R.G. Ratnayake, W.M.N. and Eaton, C.A. 1981 . Considerations of fatty acids in Menhaden from the Northern limits of the species. Proc. N.S. Inst. Sci., 31: 207-215.

Addison, R.F., Ackman, R.G. and Hingley, J. 1972. Lipid composition of the Queen Crab (*Chionocetes opilio*). J. Fish. Res. Bd. Can., 29: 407-411.

Al-Aswad, M.B., Abo-Alnaja, I.J., Salman, A.J. and Ahmed, N.H. 1980. Chemical and Bacteriological study on some commercial important fish in Dukan Lake. I. Chemical study . Zanco, 6(3): 81-98.

Table (3). Fatty Acids concentrations (per gram of lipid) in the muscles of different crab species (Ackman & McLeod, 1988).

Fatty Acids	Crab		
	(Jonah) *Cancer borealis*	(Queen) *Chionoecetes opilio*	(Rock) *Cancer lrroratus*
14:0	0.01	Trace	0.10
16:0	0.12	0.09	0.14
18:0	0.05	0.02	0.05
? SFAs	0.18	0.011	0.20
16:1	0.04	0.04	0.04
18:1	0.19	0.14	0.19
20:1	0.03	0.02	0.06
22:1	Trace	Trace	0.05
? MUFAs	0.26	0.20	0.34
20:5	0.28	0.20	0.27
22:6	0.10	0.09	0.15
? PUFAs	0.38	0.29	0.42
Cholesterol mg/100g	78.4	76.0	70.9

Al-Habbib, A.M., Salih, W.A. and Hamed, K.M. 1986. Seasonal variation in the biochemical composition of the skeletal muscle of the freshwater fish *Barbus barbulus*. JBSR, 17(1): 219-225.

Al-Saad, H.T. and Al-Timari, A.A. 1993. Sources of hydrocarbons and faty acids in sediment from Hor Al-Hammar Marsh, Shatt Al-Arab and North west Arabian Gulf. Mar. Poll. Bull., 26(10): 559-564.

A.O.A.C 1984. Official methods of analysis. 14 th ed. Association of official Analytical Chemists, Inc. S. Williams, Ed. U.S.A. 1141 p.

Gurr, M.I. and Harwood, J.L. 1991. Lipid biochemistry – an introduction, 4 th ed. London, Chapman and Hall.

Hantoush, A.A. 1998. Seasonal variations in the biochemical constituents for muscles of some freshwater and marine fishes from Shatt Al-Arab river and NW Arabian Gulf. M.Sc. thesis, Science College, Basrah University, 93p.

Hantoush, A.A., Al-Saad, H.T. and Abdul-Hassan, E.A. 1999. Seasonal variations of some biochemical aspects of the muscles of some freshwater and marine fishes from Shatt Al-Arab river and NW Arabian Gulf. Marina Mesopotamica, 14(2): 427-453.

I.U.P.A.C 1979. Standard methods for the analysis of oils, fats and derivatives . 6 th ed. International Union of Pure and Applied Chemistry. Pergamon Press. C. Paquot, U.K., 170 p.

Iwasaki, M. and Harada, R. 1985. Proximate and amino acid composition of the Roe and Muscle of selected marine species. J.Fd. Sci., 50 (6): 1585-1587.

Lowry, O.H., Rosebrough, N.J., Farr, A.L. and Randall, R.J. 1951. Protein measurment with the Folin phenol reagent. J. Biol. Chem., 193: 265-275.

Mustafa, F.A. and Medeiros, D.M. 1985. Proximate composition, mineral content, and fatty Acids of Catfish (*Ictalurus punctatus*, Rafinesque) for Different seasons and cooking methods. J. Fd. Sci., 50: 585-588.

Napolitano, G.E. 1990. Fatty acid composition of three cultured algal species (*Isochrysis galbana, Chaetoceros gracilis* and *Chaetoceros calcitrans*) used as food for bivalve larvae. J. World Aquacult Soci., 21(2): 122-130.

Napolitano, G.E. and Ackman, R.G. 1992. Anatomical distributions and temporal variations of lipid classes in Sea Scallops *Placopecten magellanicus* (Gmelin) from Georges Bank (Nova Scotia). Comp. Biochem. Physiol., 103 B (3): 645-650.

Napolitano, G.E., Ackman, R.G. and Silva - Serra, M.A. 1993. Incorporation of dietary sterols by the Sea Scallop *Placopecten magellanicus*(Gmelin) fed on microalgae. Mar. Biol., 117: 647-654.

Napolitano, G.E., MacDonald, B.A., Thompson, R.J. and Ackman, R.G. 1992. Lipid composition of eggs and adductor muscle in gaint Scallops (*Placopecten magellanicus*) from different habitats. Mar. Biol., 113: 71-76.

Ratnayake, W.N. and Ackman, R.G. 1979. Fatty alcohols in capelin, Herring and Mackerel oils and muscle lipids: I. Fatty Alcohol details linking dietary copepod fat with certain fish depot fats. Lipids, 14(9): 795-803.

Steel, R.G.D. and Torrie, J.H. 1960. Principles and procedures of statistics . McGraw – Hill Book Co., Inc. New York. 481 p.

Walton, C.G., Ratnayake, W.M.N. and Ackman, R.G. 1989. Total sterols in seafoods : Iatroscan TLC/FID versus the kovacs GLC/FID method. J.Fd. Sci., 54: 793-795, 804.

Windsor, M. and Barlow, S. 1981. Introduction to fishery by products. Fishing News Books, Ltd. Farnham. Surrey. England, 187 p.

Wootton, I.D.P. 1974. Microanalysis in Medical Biochemistry. 5 th ed. Churchill Livingston. Edinburgh and London, 307 p.

المحتوى الكيميائي الحيائي والاحماض الدهينة والكولسيترول في السرطان البحري *Portunus pelagicus* من شمال غرب الخليج العربي

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الخلاصـــــة

تم اصطياد السرطان البحري *Portunus pelagicus* خلال شهر تشـــرين الاول لسنة 1996، من بيئة شمال غـــرب الخليـــج العربـــي. درســـت المكونات الكيميائية الحياتية المختلفة في عضلات كـــل مـــن الذكـــور والاناث. من التحاليل الكيميائية يلاحظ ان الذكور احتوت على نسبة من الرطوبة والبروتين اعلى من الاناث. بينما الاناث احتوت علـــى نســـبة اعلى من الدهن والرماد والكولسيترول والرقـــم اليـــودي والاحمـــاض الدهنية الحرة. اظهـــــر التحليل الاحصائي فروقـــــات عالية المعنويـــة (p? 0.01) في البروتين والرماد وفروقات معنوية (p?0.05) في الدهـــن والاحماض الدهنية الحرة بين جنسي الذكور والاناث. حـــددت نوعيـــة الاحماض الدهنية وكمياتها باستخدام جهاز كرومـــــاتوغرافي الغـــاز اذ لوحظ تباين واسع في تراكيز الاحماض الدهنية الرئيسية بيـــن الذكـــور والاناث. تمثل البيانات المستحصلة فـــي الدراســـة اهتمامـــاً واضحـــاً بالتركيب الكيميائي والقيمة الغذائية للسرطان البحري.