



IJPPR

INTERNATIONAL JOURNAL OF PHARMACY & PHARMACEUTICAL RESEARCH
An official Publication of Human Journals

ISSN 2349-7203



Human Journals

Research Article

October 2017 Vol.:10, Issue:3

© All rights are reserved by Prameela Kandra et al.

Comparison of Compositional Characteristics of Marine and Freshwater Shrimp Waste



IJPPR
INTERNATIONAL JOURNAL OF PHARMACY & PHARMACEUTICAL RESEARCH
An official Publication of Human Journals



ISSN 2349-7203

Haritha Dupetti¹, Venkatesh Kuncham², Prameela Kandra^{3*}

^{1*} *Department of Biotechnology, GITAM Institute of Technology, GITAM University, Visakhapatnam - 530045, Andhra Pradesh, India*

² *Department of Biotechnology, GITAM Institute of Science, GITAM University, Visakhapatnam - 530045, Andhra Pradesh, India*

Submission: 27 September 2017
Accepted: 5 October 2017
Published: 30 October 2017

Keywords: Shrimp waste, astaxanthin, lipid, *Penaeus vannamei*, *Macrobrachium malcolmsonii*.

ABSTRACT

Objective: The aim of the present study was to compare the compositional characteristics of marine and freshwater shrimp waste. **Methods:** Shrimps were peeled by separating meat from head and carapace. The material was homogenized. The moisture and ash content was measured by drying the samples in an oven at 105°C, subjected to muffle furnace treatments respectively and total fat by soxhlet extraction method. The total protein content was measured by microKjeldahl method and total carbohydrate content was measured by using Anthrone method. Macro and micro minerals were determined according to AOAC method. **Results:** The compositional characteristics of marine (*Penaeus vannamei*) and freshwater shrimp waste (*Macrobrachium malcolmsonii*) were compared. The process yield and proximate composition of shrimp waste have to lead to the production of 40% waste and 60% as meat. Nutrients like protein, lipids, carbohydrates, minerals, astaxanthin, and chitin were evaluated for industrial purposes. High protein content (28.45%) was observed in fresh water waste (*M. malcolmsonii*) than marine shrimp waste (27.23%). Twenty three fatty acids were identified with the percentage of saturated, unsaturated and trans fatty acids in marine shrimp waste at 38.08%, 58.36%, and 3.55% respectively. **Conclusion:** The results obtained from this would be useful for further studies on evaluation of various food functionalities and also in shrimp processing industries to supplement formulations for animal feeding.



www.ijppr.humanjournals.com

INTRODUCTION

Shrimp production boosted tremendously across the world in recent years with the production of 3.6 million metric tons per year. Asia alone accounts for 80% of world shrimp production making it a frontier in shrimp farming. India is the second leading producer and supplier of shrimp, in Asia, with an export rate of 63%. Around 150,000 hectares area is underutilization for aquaculture in India, out of which 48% contribution is from nine coastal districts of Andhra Pradesh. Indian shrimp farming is majorly focused on marine and fresh water shrimps, thus making coastal aquaculture diverse in terms of resources used, practices adapted and environmental characteristics. Penaeid shrimps *Penaeus monodon* and *Penaeus vannamei* are two major marine species being cultivated in India. However, there seems an increasing shift in marine shrimp farming from *Penaeus monodon* to *Penaeus vannamei* in coastal districts of Andhra Pradesh, during 2010-15, as *P. vannamei* are more resistant to diseases, tolerant to high stocking densities and have shown high growth rate in low saline waters [1]. The production of *P. vannamei* for the year 2010-11 was 80,717 tonnes [2], whereas, by the end of the year 2014, production surged to as high as 375,000 tonnes, owing to 92% export growth record in quantity terms.

Penaeus vannamei is native to the pacific coast of Mexico and was introduced to Asia in the year 1996. *P. vannamei* can grow throughout the year in areas where water temperatures are normally $> 20^{\circ}\text{C}$ and are known to live in tropical marine habitats. It is observed that *P. monodon* is highly prone to stress-induced diseases compared to *P. vannamei*, thus affecting shrimp farming. However, the stress conditions are actually leading to an increased production of reactive oxygen species. Shrimps (*P. vannamei*) fed with dietary astaxanthin have shown good growth, the better survival rate in low salinity conditions [3] and also shown good pigmentation in their exoskeletons [4]. Except for Plasmodium and Toxoplasma and aphids, other animals are usually not capable of synthesizing carotenoid pigments hence they require the dietary intake of astaxanthin to meet health demands [5].

Advances in shrimp production, in India, have led to the cultivation of fresh water monsoon river prawn *Macrobrachium malcolmsonii*. Initially this was considered as an artisanal culture, now it has entered into the commercial arena [6]. In the year 2005 the production of fresh water shrimp was 42,780 tonnes, now it has increased several folds due to low profile, lower stocking densities, no major diseases, and more potential for rural aquaculture and environmental sustainability.

The tremendous increase in world shrimp consumption has led to a great growth in cultivating shrimps, both marine and fresh water, which inevitably has led to an increase in waste that is being produced by the seafood processing industries [7]. During shrimp processing, generally, 40-58% is discarded as waste, which includes head, carapace, and tail [8]. However, these byproducts are known to contain many valuable compounds like protein, chitin, lipids, and carotenoids [9,10]. Continuous production of this biomaterial without the corresponding development of utilizing technology has resulted in waste collection, disposal, and pollution problems [11].

Carotenoids are a group of natural pigments that are ubiquitous in nature [12,13,14]. Currently, the nutraceutical industry synthetically manufactures five major carotenoids like lycopene, β - carotene, canthaxanthin, zeaxanthin, and astaxanthin, for use in a range of food products - vitamin supplements and health products, cosmetics and as feed additives for poultry, livestock, fish and crustaceans [15-17]. Astaxanthin is primarily synthesized by marine microorganisms, such as green algae *Haematococcus pluvialis* and accumulates in aquatic animals thus coloring their flesh and waste to red. The main pigment provides reddish-orange color to the shrimp. In humans, Astaxanthin plays many roles including biological, pharmaceutical, antioxidants, UV-photo inhibitory effects, cardio protective, anti-hypertensive, anti- tumorigenic and chemoprevention [18-21]. Hence, there is a great demand for natural carotenoids as a replacement of currently used synthetic products in foods and feeds [22].

Shrimp waste is a natural and cheapest source of biopolymer chitin and chitosan which has been used as a promising tool for nanoscale drug carrier system of poorly absorbed therapeutic drugs in cancer therapy [23- 26]. Small quantities of shrimp waste byproducts are used for animal feed, much of valuable products are being wasted. The studies on the composition of valuable products in the waste are restricted to some varieties of shrimp. Hence, the main purpose of this study is to determine the proximate composition, lipids, astaxanthin, and minerals from marine shrimp (*P. vannamei*) and fresh water shrimp (*M. malcolmsonii*) waste with the intension of assessing total extract yield and comparison of marine and fresh water shrimp discards.

MATERIALS AND METHODS

Preparation and processing of shrimp

About 1000g of marine shrimp *Peanaeus vannamei* was obtained from seafood processing industry, Marikavalasa, Visakhapatnam, Andhra Pradesh, India and 1000g of fresh water shrimp *Macrobrachium malcolmsonii* was acquired from a local sea food market in Visakhapatnam, Andhra Pradesh, India and were transported to the laboratory under frozen conditions. The frozen shrimp were thawed at room temperature. Shrimps were peeled manually (hand) by separating meat from head and carapace. The material was homogenized in a laboratory mixer (local made) and sorted according to particle size. The average particle size diameter (0.4-0.5mm) was maintained (ASAE standards). The material was vacuum packed in polyethylene bags and kept at -20°C until further use. Astaxanthin was purchased from Sigma (97%, SML0982) and all chemicals and solvents used were of analytical grade.

Proximate composition of shrimp waste

The mass and length of the shrimps were analyzed by weighing and measuring individually. Peeled samples were weighed to calculate process yield. The moisture content was measured by drying the samples in an oven at 105°C [27]. Crude ash content was determined by incineration of shrimp waste in a muffle furnace at 550°C [28]. Total fat by soxhlet extraction [29], chitin was estimated as per the method of [30]. The total protein content was measured by microKjeldahl method [31] and total carbohydrate content was measured by using Anthrone method [32]. Macro and micro minerals were determined according to AOAC method [33,34]. Iron, copper, lead, and cadmium were estimated by using Atomic Absorption Spectrometer (AA 7000, Shimadzu). Sodium, potassium, magnesium, phosphorus, calcium were determined titrimetrically and spectrophotometrically [34].

Determination of fatty acid composition by Gas chromatography

The fatty acid composition of the shrimp waste was determined by gas chromatography [34]. 100 mg of sample was weighed and dissolved in 10 ml hexane. 100µl of 2N potassium hydroxide in methanol was added, vortexes for 30 seconds and centrifuged (REMI centrifuge, R-8C, India). The clear supernatant of the test sample (2 ml) taken into individual vials and placed in an auto sampler which robotically shifted them into the auto-injector into GC-MS for analysis. The methyl esters of fatty acids in chloroform were analyzed by using a

Shimadzu GC -FID fitted with a flame ionization detector. The column (Agilent HP-88) used had a length of 100m and 0.25micron film thickness. Injection temperature of 220°C; detection temperature of 230°C; N₂ flow rate of 1 ml min⁻¹; injection volume of 1µl of conditions was maintained. Peaks were identified by co-chromatography by using standard FAME37-Mix (Sigma).

Extraction of Astaxanthin content from shrimp waste

The total astaxanthin content from shrimp waste was determined by spectrophotometric method [35]. This method involves 5 g of waste with 75ml of the mixture of 60% (v/v) of n-hexane and isopropyl alcohol. The extracts were washed with the equal volume of 0.1% NaCl solution in order to separate the phases and to remove isopropyl alcohol from n-hexane phase. The phase of n-hexane extract was dried by filtration over anhydrous sodium sulfate for 120 minutes. The dry extract was diluted in hexane to the known volume and the absorbance was measured at a wave length of 472nm. This procedure was repeated in triplicate at 20°C. The amount of astaxanthin present can be calculated by using the following formula.

$$\text{The amount of astaxanthin in shrimp waste} = \frac{A_{472} \times \text{Volume of extract (ml)} \times \text{Dilution factor}}{0.2 \times \text{Weight of sample (g)}}$$

Where A= absorbance at 472nm

Determination of total carotenoids

The total carotenoid concentration was determined by using the spectrophotometric modified method from shrimp waste [36]. To prepare standard graph different volumes 0.2, 0.4, 0.6, 0.8, 1.0 and 1.2 ml stock solution of astaxanthin (1%) were prepared and solutions were diluted up to 10ml with n-hexane. The absorbance was measured at 472nm using n-hexane as the blank. The experiment was carried out in triplicate; standard graph for astaxanthin was constructed.

Identification of astaxanthin and carotenoids by thin layer chromatography

The solvent extract from shrimp waste was subjected to thin layer chromatography to identify astaxanthin and carotenoids. Thin layer chromatography was made of Silica-G mixed with water in a proportion that the thick suspension is spread on 20x20 cm glass plates with the

thickness of 0.25mm by using the spreader. The concentrated solvent extract was applied (50µl) to the plates and eluted by using mobile phase of acetone: hexane (25:75 %v/v).

RESULTS

Process yield is an important part of shrimp processing industries. The masses of different parts of the two varieties of shrimp - marine water (*P. vannamei*) and fresh water (*M. malcolmsonii*) were studied. The results on weight and yield of shrimp byproducts were represented in Table 1.

Table 1: Yield of shrimp processing byproducts and their weights

Variety of shrimp	Components	Weight (g) ¹	Yield (g/100g)
Marine shrimp (<i>P. vannamei</i>)	Head	6 ± 1	30 ± 3
	Meat	13 ± 2	60 ± 3
	Shell	2.5 ± 0.3	7 ± 1
	Tail	0.9 ± 0.4	3 ± 1
	Total	22.4 ± 3	100
Fresh water shrimp (<i>M. malcolmsonii</i>)	Head	18.7	46 ± 2
	Meat	13.3	35 ± 3
	Shell	4.1	10 ± 1
	Tail	3.9	9 ± 1
	Total	40 ± 2	100

¹ Values in the brackets are the mean of 10 determination ± the standard deviation

The mass of each marine shrimp (*P. vannamei*) varied from 20.4 to 24.4 g with an average of 22.4 ± 3 g. The average number of pieces of shrimp ranged from 41- 44 per kilogram. The length of shrimp varied from 12.7 to 17.78 cm with an average of 15.24 ± 2 cm. The mass of each fresh water shrimp (*M. malcolmsonii*) varied from 32.92- 47.26 g with an average of 40.02 ± 2 g. The average number of pieces was 23-25 per kilogram and the length ranged from 63-65 cm with an average of 64 cm. Analysis of moisture is very critical as it determines the stability of the residue. Fresh shrimp waste residue was having high moisture content when compared to dried shrimp waste. The results of proximate and chemical analysis of marine and fresh water shrimp waste residues were presented in Table 2.

Table 2: Proximate chemical composition of marine and fresh water shrimp waste

Component (%)	Marine water shrimp waste (<i>P. vannamei</i>)	Fresh water shrimp waste (<i>M. malcolmsonii</i>)
Moisture	68 ± 0.4	69 ± 0.5
Ash content ^b	3 ± 0.2	2 ± 0.3
Carbohydrates ^{a,b}	1 ± 0.4	0.05 ± 0.2
Total protein ^{a,b}	27 ± 0.2	28 ± 0.2
Total lipid ^{s,b}	0.58 ± 0.3	0.95 ± 0.3

^a Values on dry weight basis (n=3)

^b Mean of three determinations ± standard deviation

The total ash content of marine and fresh water shrimp waste was found to be 3.38 and 2.24 respectively. The total protein in fresh and marine shrimp waste was 28.45 % and 27.23% respectively. Higher protein content was observed in fresh water waste (*M. malcolmsonii*) than marine shrimp waste (*P. vannamei*). High protein content was reported in species *P. monodon* and *M. rosenbergii* [40]. Similarly, high protein in the head (47.75%) and shell (47.43%) were reported in *Penaeus* species [41]. Due to high protein, content in waste residue different species waste is directed mainly for the production of flakes and formulation for animal feeds [42]. In addition to this, the residue also consists of very low carbohydrate content in both fresh shrimp and marine shrimp waste. The total lipids have quantified the values found for fresh water shrimp waste was 0.95 and marine shrimp waste was 0.58. Only marginal differences were found in lipid content of fresh and marine shrimp waste. Similarly, the residues also contain different minerals. The results of the composition of some of the major elements and minor minerals were presented in Table 3.

Table 3: Major and minor minerals present in marine and fresh water shrimp waste

Component	Marine shrimp waste (<i>P. vannamei</i>) ^{a,b} mg/1000g	Fresh water shrimp waste (<i>M. rosenbergii</i>) ^{a,b} mg/1000g
Calcium	725±6	925±4
Phosphorus	170±4	286±5
Sodium	1390±5	2500±2
Potassium	884±4	710±1
Magnesium	81±2	81±3
Iron	11±2	18±2
Lead	0.4±0.1	0.4±0.1
Zinc	70±2	65±2
Manganese	10±3	9±0.5
Cadmium	0.03±0.1	0.04±0.1
Copper	6±1	13±1

^a Dry weight basis

^b n=3 Mean of three determinations \pm standard deviation

Most of the major minerals like calcium, phosphorus, sodium, potassium, and magnesium were common in both varieties of wastes. Similarly, good amounts of minor minerals like iron, zinc and manganese and negotiable amount of lead were present in marine and fresh water shrimp waste. Twenty-three fatty acids were identified with the percentage of saturated, unsaturated and trans fatty acids in marine shrimp waste at 38.08%, 58.36%, and 3.55% respectively. Two fatty acids cis-9- tetradecanoic acid and cis-10-pentadecanoic acids were absent in marine shrimp waste. Similarly, in fresh water shrimp waste, the total percentage of saturated, unsaturated and trans fatty acids was 47.1%, 86.15 %, and 3.0% respectively. Four fatty acids palmitic acid, behinic acid, lignoceric acid and nervonic acids were absent in fresh water shrimp waste. Out of the saturated fatty acids, palmitic acid (C16:0) predominated in marine shrimp waste with 20.78%. Among polyunsaturated fatty acids, the most abundant were eicosapentaenoic acid (EPA) (C20:5) and decosahexanoic acid (DHA) (C22:6). These two fatty acids represent 14.97% in marine shrimp waste and 10.9% in fresh water shrimp waste (Table 4).



Table 4: Fatty acid composition (g/100g total fatty acids) of the total lipids extracted from marine and fresh water shrimp waste

Fatty acid	Name of the FAME	Marine water shrimp (<i>P. vannamei</i> g/100g)	Fresh water shrimp (<i>M. malcolmsonii</i> g/100g)
Saturated fatty acids			
C14:0	Myristic acid	1.17	0.8
C15:0	Pentadecanoic acid	0.7	3.8
C16:0	Palmitic acid	20.78	-
C17:0	Heptadecanoic acid	1.25	22.9
C18:0	Stearic acid	10.36	3.4
C20:0	Arachidic acid	0.9	15.8
C21:0	Henecosanoic acid	0.81	0.4
C22:0	Behinic acid	1.51	-
C24:0	Lignoceric acid	0.6	-
Total		38.08	47.1
Unsaturated fatty acids			
C14:1	Cis-9 tetradecanoic acid	-	0.53
C15:1	Cis-10 pentadecanoic acid	-	0.88
C16:1	Oleic acid	2.27	14.4
C18:1	Linoleic acid	15.2	28.9
C18:2	Linolenic acid	15.67	13.3
C18:3	Cis-11 Eicosenoic acid	1.62	3.2
C20:1	Cis-11 Eicosenoic acid	0.93	7.32
C20:2	Cis-11,14 Eicosenoic acid	1.38	0.8
C20:3	Cis-8,11,14 Eicosenoic acid	0.67	0.4
C20:4	Trienoic acid	3.78	2.82
C20:5	Cis-5,8,11,14,17 Eicosapentanoic acid	7.42	5.3
C22:1	Euricic acid	1.32	0.7
C24:1	Nervonic acid	0.55	-
C22:6	Cis-4,7,10,13,16,19 docosahexanoic acid	7.55	5.6
Total		58.36	86.15
Trans fatty acids			
C18:1	Trans 9 octa	3.55	3.0
Total		3.55	3.0
EPA + DHA		14.97	10.9
ω3/ ω6		3.32	1.00
ω3/ ω6/ ω9		1.77	-

EPA: Eicosapentanoic acid

DHA: Docosahexanoic acid

The results also showed that omega- 3 and omega- 6 ratios were at values of 3.32 in marine shrimp waste and 3.0 in freshwater shrimp waste. Similarly, the results also show that omega 3, omega 6 and omega 9 ratios are at a value of 1.77 in marine shrimp waste.

For further studies to evaluate the amount of astaxanthin present in shrimp waste mixture of solvents isopropyl alcohol and hexane was used and the results were compared to marine and fresh water shrimp waste. A standard graph was constructed to know a number of carotenoids (Figure. 1).

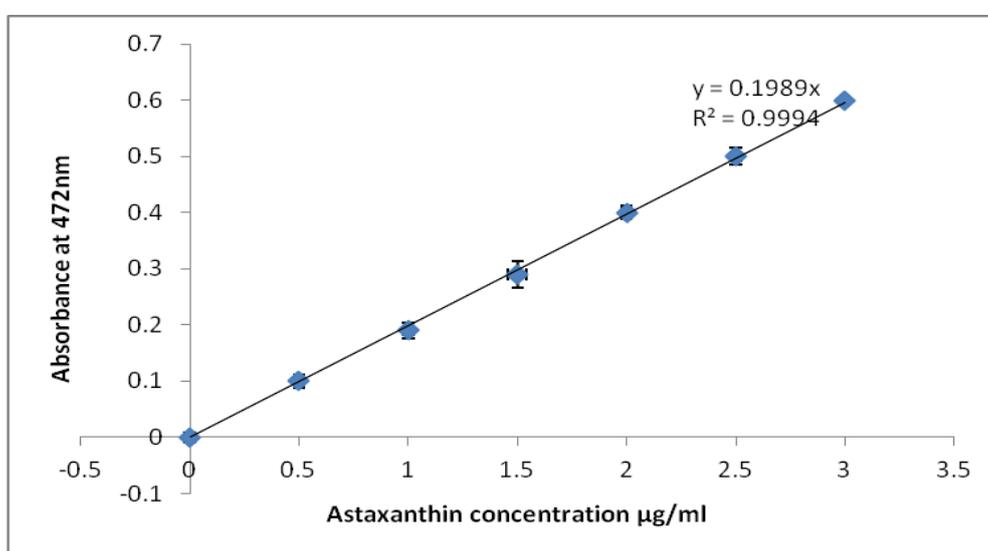


Figure. 1: Calibration graph of astaxanthin at the wavelength of 472nm

All extracts obtained were orange-red pastes. This could be because the mixture of polar and non-polar solvents enhances color and the yield. As shown in Table 5 highest yield was observed in marine head waste 88mg/kg as compared to head component of fresh water shrimp 73mg/kg.

Table 5: Comparison of the yield of astaxanthin and chitin in different body components of fresh and marine water shrimp waste.

Variety of shrimp	Component	Yield of astaxanthin (mg/kg residue) ^{a,b}	% Yield of chitin ^c
<i>P. vannamei</i>	Head	88±3	10.1
	Carapace	68±2	4.4
	Tail	3±1	20.2
<i>M. malcolmsonii</i>	Head	73 ±2	20.0
	Carapace	50 ± 4	5.4
	Tail	4±1	13.8

^a Results expressed on a dry weight basis

^b Mean value of three determinations ± standard deviation

^c Dry weight basis

Similarly, the astaxanthin concentration in carapace of marine shrimp waste was higher 68mg/kg than freshwater shrimp waste 50mg/kg. Minimal differences were observed in the concentration of tail portion. Separation of carotenoids from marine and fresh water shrimp waste extract have shown 9 bands with different Rf values on TLC. The following Rf values 0.264 (orange), 0.339 (orange), 0.396 (orange), 0.446 (orange), 0.528 (orange), 0.61 (orange), 0.64 (orange), 0.691 (orange), 0.735 (yellow) were common bands in both waste residues and 0.786 (yellow) was observed in fresh water shrimp waste. The orange band with an Rf value of 0.339 corresponds to astaxanthin. Orange bands with Rf values 0.528 and 0.691 corresponds to astaxanthin monoester and diester respectively. The amount of chitin on dry weight basis found in *P. vannamei* head, carapace and tail were 10.12%, 4.4%, and 20.2 % respectively. Similarly, the chitin content of *M. malcolmsonii* head has 20.08%, carapace 5.4% and 13.8% found in tail portion. The total chitin content of fresh water shrimp was 39.28 % compared to marine shrimp 34.72%. High chitin content was reported in head part of *M. malcolmsonii* compared to *P. vannamei* (Table 5).

DISCUSSION

Recent advances in the understanding of process yield and proximate composition of shrimp waste have led to a growing interest in nutraceutical production for industrial and nutritional purposes. During processing of marine and fresh water shrimps the non-edible parts such as head, carapace and tail were removed. The residue represents approximately 60% of raw material as meat and 40% of waste. Shrimp process yield is economically important to the

processing industries in India and other countries. The process yield is not only significant in calculating weight loss on shelling procedures but also is significant in the generation of the enormous amount of residue as waste, which contains valuable products. Depending on the type of shelling, the process yield differs. The most representative being the removal of the head (Cephalo thorax) constituting 63% and exoskeleton of shrimp constituting 53.2% [37]. Some authors reported the different amount of process yield for *Penaeus* species [38]. It was reported as - 40-50% meat, 33-45 % head, 22% shell and 15% tail, that also depends on the size of shrimps. Cephalothorax of *Litopenaeus* species found in Brazil was reported at 33% [39]. Our manual (hand) process yield results, however, indicate a good yield in the meat for marine shrimp *P. vannamei* and good yield in head waste for fresh water shrimp *M. malcolmsonii*. This could be due to the size of marine shrimp, representing higher mass in meat and consequently reducing the weight of byproducts.

The proximate composition analysis of shrimp waste varies widely from species to species and with the same species from one individual to another [43]. The chitin content of fresh water shrimp waste was higher than marine shrimp waste. Both waste residues have shown the majority of macro and microelements with adequate amounts of calcium, phosphorus, sodium, potassium, iron, and zinc. Similarly, predominant amount of macro elements in the residue of the northern pink shrimp (*Pandalus borealis*) were reported [10,38,44]. The total lipid content of fresh water was high when compared to marine water shrimp waste. Total lipid content (2.3) was reported in *Pandalus borealis* [45], *Penaeus* species was 0.76% [38], *P. monodon* giant tiger shrimp was 3.58% [40] and *P. subtilis* was 0.94 [46]. High levels of lipid content were found in *P. monodon* (3.50) and sea-bob shrimp (3.79). *Penaeus* species captured from Egypt found to have high total lipid content in cephalothorax (10.5%) and 3.7% in the exoskeleton giving an average of 7.14% in the mixed residues. Skin meat usually contains less than 1% of total lipids [47].

The fatty acid composition of waste residue has shown the good amount of unsaturated fatty acids starting from oleic acid to decosa hexanoic acid (DHA). The ratio of omega-3: omega 6 was 3.32 in marine shrimp waste showing that this residue was a potential source of omega fatty acids. Currently, most of the western countries are consuming high levels of omega-6 and omega-3 fatty acids in the ratio of 20:1. Foods with high omega-6 fatty acids will cause inflammation. Carcinogenic eicosanoids will increase the risk of chronic and degenerative diseases such as cancer, high blood pressure, vascular and cardiac diseases. Intake of omega-

3 fatty acids may cause anti-inflammatory, anti-thrombosis and anti-anything [48,49,50]. Thus the fatty acids present in fresh and marine shrimp waste being the adequate source of omega 3 fatty acids offers benefits to human health and enrichment of animal feeds.

Solvent extraction of astaxanthin and its recovery (carotenoids) from marine and fresh water shrimp waste was studied. Different solvents like hexane, acetone, isopropyl alcohol have been used [35] but the mixture of polar and nonpolar solvents enhanced the extraction yield from marine and fresh water waste. Table 6 compares the values found for proximate composition, total lipids and astaxanthin in marine and fresh water shrimps collected in Andhra Pradesh, India with those from other parts of the world. In data comparison for the astaxanthin content in some species, *M. malcolmsonii* have shown 159 μ g/g in waste residue. Similar results were obtained when astaxanthin was extracted with supercritical CO₂ with co-solvent [51]. However, greater levels (206 μ g/g) were reported in *Xiphopenaeus kroyeri* of West Indies. Very low amount of astaxanthin (3.4 μ g/g) was reported in *Pandalus borealis* of Canada (Table 6). *Penaeus monodon* and *Penaeus indicus* of America and Indo West Pacific have shown similar quantity of astaxanthin in their waste residue (59.8 μ g/g). Carotenoids were separated by thin layer chromatography. Carotenoids from marine and fresh water shrimp waste have shown different R_f values of 0.264 to 0.786 with orange and yellow color confirms the presence of free astaxanthin, astaxanthin monoester and astaxanthin diester as their main pigments. These results were supported by red spot shrimp (*Farfantepenaeus paulensis*) and marine crab (*Charybdis cruciater*) [52]. Similar results have been reported about various carotenoids in Antarctic krill shrimp (*Euphasia superb*), by TLC [53]. Hence the presence of astaxanthin varies from species to species and from one to another in same species when they grow at different locations with different nutrients. Chitin is one of the important exoskeleton components of crustaceans. In most of the dried crustaceans chitin content ranges from 20 -50% [54]. Low chitin content (2.6 to 3.6%) was reported in *Metapenaeus* species [55]. The difference in chitin content is due to the difference in body components of the species.

Numerous studies also have been performed to understand more precisely the beneficial effect of various carotenoids on human health. In this context, astaxanthin has been found to act as the important protector against cancer, cardiovascular and other degenerative diseases [56]. Therefore, today much interest exists in information regarding proximate composition, lipid and astaxanthin contents of shrimp waste. The data obtained from marine and fresh

water shrimp waste would be more easily compared with each other and would be useful for further studies on evaluation of various food functionalities. Such data was needed for industries and supplement formulations for animal feeding. However, the bioavailability of astaxanthin is still not fully understood. Further research is needed in the near future to get more accurate knowledge of bioavailability of products from various shrimp wastes in order to develop food-based strategies for long-term supplement of nutraceuticals.

CONCLUSION

In recent years shrimp production has increased tremendously from 30 % to 80% in catch and aqua culture respectively. Depending upon the species, size and shelling procedure as the production increase a number of waste increases from 40% to 50%. Evaluation of process yield and proximate composition of fresh and marine water shrimp wastes will help to know the compositional characteristics for industrial purposes. Further research is needed to develop food additives from shrimp waste.

ACKNOWLEDGEMENT

This work was financially supported by the Science and Engineering Research Board (SERB), Department of Science and Technology (No.SB/FTP/ETA-0053/2014).

REFERENCES

- [1] Briggs M, Funge-Smith S, Subasinghe R, Phillips M. Introduction and movement of two penaeid shrimp species in Asia and the Pacific, FAO Fisheries Technical Paper. No. 476. Rome, FAO 2005; pp.78.
- [2] SEAI 2013, 42nd Annual Report of Seafood Exporters Association of India, 2011-12, pp.10.
- Flores M, Diaz F, Medina R, Re AD, Licea A. Physiological, metabolic and hematological responses in white shrimp *Litopenaeus vannamei* (Boone) juveniles fed diets supplemented with astaxanthin acclimated to low-salinity water. *Aquac Res* 2007; 38: 740–747
- [3] Vernon-Carter J, Ponce-Parafox JJ, Pedroza-Islas R. Pigmentation of pacific white shrimp (*Penaeus vannamei*) using Aztec marigold (*Tagetes erecta*) extracts as carotenoid source. *ALAN* 1996; 46: 243-246.
- [4] Tonhosolo R, Alexandri FL, de Rosso VV, Gazarini ML, Matsumura MY, Peres VJ, Merino EF, Carlton JM, Wunderlich G, Mercadante AZ, Kimura EA, Katzin AM. Carotenoid biosynthesis in intraerythrocytic stages of *Plasmodium falciparum*. *J Biol Chem* 2009; 284: 9974–9985.
- [5] MPEDA. An Overview. Marine Products Export Development Authority, India, Ministry of Commerce & Industry, GoI, Kochi, India, 2004.63pp.
- [6] Subasinghe S. Chitin from shellfish waste-health benefits over-shadowing industrial uses. *Info. fish International* 1999; 3: 58-65.
- [7] Sachindra NM, Bhaskar N, Mahendrakar NS. Carotenoids in different body components of Indian shrimps. *J Sci Food Agric* 2005; 85:167–172.
- [8] Shahidi F, Metusalach, Brown JA. Carotenoid pigments in seafood and aquaculture. *Cri Rev Food Sci* 1998; 38: 1-67.

- [9] Rodde RH, Einbu A, Varun KM. A seasonal study of the chemical composition and chitin quality of shrimp shells obtained from northern shrimp (*Pandalus borealis*). Carbohydr Polymers 2008; 71: 388-393.
- [10] Prameela K, Murali Mohan CH, Hemalatha KPJ. Efficient use of shrimp waste: present and future trends. Appl Micro Bio 2012; 93: 17-29.
- [11] Goodwin TW, Srisukh S. The carotenoids of the Locust Integument. Nature 1948; 161: 525-526.
- [12] Manuta C. Astaxanthin in insects and other terrestrial arthropods. Nature 1948; 162: 298.
- [13] Mann V, Harker M, Pecker I, Hirschberg J. Metabolic engineering of astaxanthin production in tobacco flower. Nature 2000; 18: 888-892.
- [14] Romer S, Fraser PD, Kiano JW, Shipton CA, Misawa N, Schuch W, Bramley PM. Elevation of the pro-vitamin A content of transgenic tomato plants. Nat Biotechnol 2000; 18: 666-669.
- [15] Del Campo JA, García-González M, Guerrero MG. Outdoor cultivation of microalgae for carotenoid production: current state and perspectives. Appl Micro Bio 2007; 74: 1163-1174.
- [16] Jackson H, Braun CL, Ernst H. The chemistry of novel xanthophylls carotenoids. Am J Cardiol 2008; 101: 50D-57D.
- [17] Peto R, Doll R, Buckley JD, Sporn MB. Can dietary β -carotene materially reduce human cancer rates? Nature 1981; 290: 201-208.
- [18] Tanaka T, Morishita Y, Suzui M, Kojima T, Okumura A, Mori H. Chemopreservation of mouse urinary bladder carcinogenesis by naturally occurring carotenoid astaxanthin. Carcinogenesis 1994; 15: 15-19.
- [19] Sporn MB, Suh N. Chemoprevention of cancer. Carcinogenesis 2000; 21: 525-530.
- [20] Guerin M, Huntley Mark E, Olaizola M. *Haematococcus astaxanthin*: applications for human health and nutrition. Trends Biotechnol 2003; 21: 210-216.
- [21] Paine JA, Shipton CA, Chaggar S, Howells RM, Kennedy MJ, Vernon G. Improving the nutritional value of Golden Rice through increased pro-vitamin A content. Nat Biotechnol 2005; 23: 482-487.
- [22] Allen TM, Ligand-targeted therapeutics in anticancer therapy. Nat Rev Cancer 2002; 2: 705-763.
- [23] Ferrari M. Nanogeometry: beyond drug delivery. Nat Nanotechnol 2008; 3: 131-132.
- [24] Haun JB, Devaraj NK, Hilderbrand SA, Lee H, Weissleder R. Bioorthogonal chemistry amplifies nanoparticle binding and enhances the sensitivity of cell detection. Nat Nanotechnol 2010; 5: 660-665.
- [25] Prameela K, Hemalatha KPJ. Current understanding of the synergistic interplay of chitosan nanoparticles and anticancer drugs: merits and challenges. Appl Micro Bio 2015; 99: 2055-2064.
- [26] AOAC. Official Methods of Analysis. 18th Ed. Association of Analytical Chemists, Gaithersburg MD; 2006.
- [27] AOAC International in Official Methods of AOAC International, method, 1995; 920.153, (39.1.09).
- [28] AOAC Official Method of Analysis, Method 991.36. Fat (Crude) in meat and meat products. 18th Ed. AOAC Int., Gaithersburg, MD, 2006.
- [29] Spinelli J, Lehman L, Weig D. Composition, processing and utilization of red crab (*Pleroncodes planipes*) as an aquaculture feed ingredient. J Fish Res Bd Can 1974; 31:1025-30.
- [30] AOAC 928.08 Alternative II, Nitrogen in Meat, Kjeldahl Method, Final Action, 1974.
- [31] Morris DL. Quantitative determination of carbohydrates with Dreywood's anthrone reagent. Science 1948; 107: 254-255.
- [32] AOAC. Official Methods of Analysis, Association of Official Analytical Chemists, 15th ed. Washington, DC.1990.
- [33] AOAC. Fatty acids in oils and fats, preparation of methyl esters, boron trifluoride method. AOAC Method; 1995. 969.33
- [34] Sachindra NM, Bhaskar N, Mahendrakar NS. Recovery of carotenoids from shrimp waste in organic solvents. Waste Manag 2006; 26: 1092-1098.
- [35] Tolasa S, Cakli S, Ostermeyer U. Determination of astaxanthin and canthaxanthin in salmonid. Eur Food Res Technol 2005; 221: 787-791.
- [36] Holanda HD, Netto FM. Recovery of components from shrimp (*Xiphopenaeus kroyeri*) processing waste by enzymatic hydrolysis J Food Science 2006; 71: 298-303.
- [37] Heu MS, Kim JS, Shahidi F, Jeong YH, Jeon YJ. Characteristics of protease from shrimp processing discards J Food Biochem 2003; 27: 221-236.
- [38] Ogawa M, Maia EL, Fernandes AC, Nunes ML, Oliveira MI, Freitas ST. Waste from the processing of

- farmed shrimp: a source of carotenoid pigments. *Ciência e Tecnologia de Alimento* 2007; 27: 333–337.
- [39] Nargis A, Ahmed KN, Ahmed GM, Hossain MA, Rahman M. Nutritional value and use of shrimp head waste as fish meal. *Bangladesh J Sci Ind Res* 2006; 41: 63–66.
- [40] Ibrahim HM, Salama MF, El-Banna HA. Shrimp's waste: chemical composition, nutritional value and utilization. *Food / Nahrung* 1999; 43: 418–423.
- [41] Freitas AS, Lopes AB, Stephan MP, Cornejo FEP, Furtado AAL. Chemical composition and molecular protein of waste of shrimp (*Xiphopenaeus kroyeri*). *Boletim do CEPPA. Curitiba* 2002; 20: 111–120.
- [42] Stansby ME. Proximate composition of fish. In *Fish in nutrition* (Heen E, Kreuzer R, ed.), Fishing News (Books) Ltd, London, 1962, pp. 55-60.
- [43] Covaci A, Voorspoels S, Schepens P, Jorens P, Blust R, Neels H. The Belgian PCB/dioxin crisis—8 years later: An overview *Environ Toxicol Pharmacol* 2008; 25, 164–170.
- [44] Shahidi F, Synowiecki J. Isolation and characterization of nutrients and value added products from snow crab (*Chionoectes opilio*) and shrimp (*Pandalus borealis*) processing discards. *J. Agric. Food Chem* 1991; 39: 1527-1532.
- [45] Assunção AB, Pena RDS. Hygroscopic behavior of the dry residue of pink shrimp. *Ciencia e Tecnologia de Alimentos*.2007; 27: 786–793.
- [46] Bragagnolo N, Rodriguez-Amaya D. Optimization of cholesterol determination by HPLC and levels of cholesterol, total lipids and fatty acids of the pink shrimp (*Penaeus brasiliensis*). *Ciência e Tecnologia de Alimentos*.1997; 17: 275–280.
- [47] Sahena F, Zaidul ISM, Jinap S, Saari N, Jahurul HA, Abbas KA, Norulaini NA. PUFAS in fish: extraction, fractionation, importance in health. *Compr Rev Food Sci Food Saf* 2009; 8: 59–74.
- [48] Simopoulos AP. Evolutionary aspects of diet, essential fatty acids and cardiovascular disease. *Eur Heart J Suppl* 2001; 3: D8–D21.
- [49] Simopoulos AP. Omega-3 fatty acids and cancer. *Indoor Built Environ* 2003; 12: 405–412.
- [50] Nobre B, Marcelo F, Passos R, Beirão L, Palavra A, Gouveia L, Mendes R. Supercritical carbon dioxide extraction of astaxanthin and other carotenoids from the microalga *Haematococcus pluvialis*. *Eur Food Res Technol* 2006; 223: 787–790.
- [51] Sachindra NM, Mahendrakar NS. Process optimization for extraction of carotenoids from shrimp waste with vegetable oil. *Biores Technol* 2005; 96: 1195-1200.
- [52] Yamaguchi K, Murakami M, Nakano H, Konosu S, Kokura T, Yamamoto H, Kosaka M, Hata K. Supercritical carbon dioxide extraction of oils from Antarctic krill *J Agr Food Chem* 1986; 34: 904.
- [53] Ornum JV. Shrimp waste- must it be wasted? *INFOFISH International* 1992; 6:48-51.
- [54] Ariyani F, Buckle KA. Ensilaging of prawn heads. *ASEAN Food J* 1991; 6: 58-63.
- [55] Fassett RG, Coombes JS. Astaxanthin: a potential therapeutic agent in cardiovascular disease. *Marine Drugs* 2011; 9: 447–465