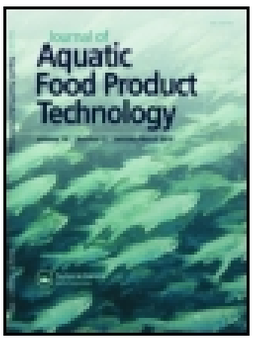

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Nutritional Evaluation of Indian Ocean Swimming Crab, *Charybdis smithii* (Portunidae), an Unconventional Crab Resource from the Indian Coast

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ABSTRACT

Investigations were carried out to determine the nutritional profile of Indian Ocean swimming crab *Charybdis smithii*, which is emerging as an unconventional resource in trawl discards of India. The average protein content was 9.38 g/100 g, fat 0.86 g/100 g, ash 0.34 g/100 g, fiber 0.13 g/100 g, and carbohydrate 1.8 g/100 g. One-way analysis of variance showed no significant variations of constituents except in dry matter and carbohydrate between sexes. Macronutrients, Na (317.1, 327.6/100 g), K (148, 177.40 mg/100 g), Ca (187.90, 285.80 mg/100 g), and Mg (34.31, 41.49 mg/100 g), showed significant variation between sexes. The composition of micronutrients in male and female were Cu (0.28, 0.15 mg/100 g), Fe (0.57, 0.71 mg/100 g), and Zn (1.71, 2.75 mg/100 g). Mineral content showed significant difference between sexes. Amino acid analysis showed that 12.04 and 11.47 g/100 g essential amino acid glycine was present in male and female, respectively, and lysine concentration was 13.96 and 12.65 g/100 g, respectively. The nutritional profile of the species was determined the first time and shows that it is comparable with any other edible crabs and could be exploited as a commercial resource to supplement nutritional demand.

KEYWORDS

Unconventional; *Charybdis smithii*; proximate composition; mineral composition; amino acid; fatty acid

Introduction

Demand for seafood is increasing worldwide due to increasing health consciousness. Apart from its widely accepted taste, crustaceans, especially shrimp, lobster and crab, are recognized as a good source of protein and mineral supplement to human health (Chigozie and Kolade, 2015). The marine fishery has shown signs of depletion during the last two decades, and the quantity of conventional commercial fishery resources have depleted rapidly (Worm et al., 2006). Since seafood is recognized as a healthy food in terms of protein, unsaturated fatty acids, and minerals, the demand for seafood in the global market is increasing. However, a huge quantity of the marine fish catch is being discarded (Alverson et al., 1994; Kelleher, 2005) due to lack of market demand. Among crustaceans, marine crabs are the most diverse group and have great demand as food. In India, conventional edible varieties of crabs contributing to the fishery economy are dominated by the spotted crab *Portunus sanguinolentus* and the blue swimmer crab *Portunus pelagicus. Charybdis lucifera, Charybdis annulata*, and *Charybdis natator* have also been identified as edible varieties in different coastal areas of the country (Radhakrishnan et al., 2007). The cross crab, *Charybdis feriatus*, which is caught as bycatch in trawling, is a preferred seafood. As the demand has increased, the landing of the species has also increased with extension of trawl fishing area (Dineshbabu, 2011). When trawling extended beyond 100 m depth, the Indian Ocean swimming crab, *Charybdis smithii*, emerged as a significant component of the trawl catch. Through exploratory surveys, Silas (1969)

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and Sulochanan et al. (1991) predicted that *C. smithii* has the potential of forming a commercial fishery; and Balasubramanian and Suseelan (2001a) have identified its wide distribution within the Indian Exclusive Economic Zone (EEZ). The biomass estimation showed that in the Indian EEZ, the biomass of the species was as high as 1,740 kg per ha (Balasubramanian and Suseelan, 2001a). Romanov et al. (2009) reported that these crabs aggregate at night in the upper 150-m layer, and the estimated biomass derived from pelagic trawling exceeded 130 kg/km^2. They also reported that abundance of *C. smithii* can reach 15,000 individuals/km^2 during the southwest monsoon along the equatorial Indian Ocean. A recent study from the southwest coast of India showed that a huge quantity of *C. smithii* are found in trawl discards. During 2008–2009, out of estimated discards from trawlers operated off 100-m depth from Mangalore fisheries harbor, *C. smithii* formed about 2.9% of the discards (Dineshbabu et al., 2012).

The nutritive value and biochemical composition of crab meat have been studied in various parts of the world, including edible and non-edible species from Indian Ocean waters (Jeyalakshmi and Chandran, 2014; Kathirvel et al., 2014; Premarathna et al., 2015; Soundarapandian et al., 2013). Preliminary investigations carried out on biochemical composition revealed that there is a potential for the species to be recognized as a safe human food (Balasubramanian and Suseelan, 2001b). The present work determined the proximate nutrition components of the species, in order to supplement the information on the nutritional quality of the species as human food. Recognition of *C. smithii* as a healthy seafood will increase the demand for the species in the seafood market, resulting in additional income for the fishermen and also reduction of resource loss by avoiding/reducing discard of the species. Detailed biochemical composition of *C. smithii* was determined for the first time, and no previous reports are available on this species.

Materials and methods

For the study, samples of *Charybdis smithii* were collected from the landings of multiday operating trawlers from Mangalore fishing harbour during January 2015 to December 2016. A part of *C. smithii* catch was packed in ice immediately after bringing it to the deck. The samples collected from the boat were brought to the laboratory. After sex-wise segregation, the carapace of individual crab was removed, and the crab meat, including the meat from chelate legs, was extracted and transferred to a Petri dish for further analysis. Carapace width (CW), carapace length, and the body weight were determined.

Proximate and mineral analysis

Triplicate samples of 30 males and 30 non-berried females were analyzed for moisture content. Known weight of the sample was kept at 80°C in hot air oven overnight to attain constant weight to ensure that the moisture content of the sample was completely evaporated (Vijayagopal et al., 2015). Dry matter (DM) of the sample was determined by drying at 105°C for 24 h in a hot air laboratory oven with a fan with forced air circulating system (Vijayagopal et al., 2015). Crude protein (CP) (N × 6.25) of the crab meat was estimated by micro-Kjeldahl method (Humphries, 1956), crude lipid (CL) was estimated using petroleum ether (60–80°C boiling point) by Soxhlet apparatus. Crude fiber (CF) was determined by the remaining fraction after refluxing with a standard solution of H_2SO_4 and NaOH under controlled conditions for 30 min (AOAC, 1995). The nitrogen-free extract (NFE) of diets considered as total carbohydrates was derived by subtracting CP, CL, CF, and ash from DM (Vijayagopal et al., 2015).

Minerals

Mineral estimation was carried out by the method of inductively coupled plasma-optical emission spectrometric (ICP-OES) determination of elements using microwave-assisted digestion. Ten micro-liters of nitric acid was used for digestion for 10 min. After digestion, the samples were made up to

100 ml and introduced to ICP-OES machine ICAP 6300 for analysis (Horwitz and W Latimer, 2005; Latimer, 2012).

Amino acid analysis

The amino acid content of crab meat was determined based on the study of Hofmann et al. (1997, 2003) using isotope with gas chromatography–combustion–isotope ratio mass spectrometry (GC-C-IRMS/MS) (GC: Hewlett-Packard 58590 series II, Germany; combustion series II-interface, IRMS MAT 252, Finnigan MAT, Germany; MS: GCQ, Finnigan MAT, Germany). The capillary column of dimension 50 m × 0.32 mm i.d. × 0.5 μm BPX5 (SGE) was connected to gas chromatography. The flow of carrier gas (helium) was maintained at 1.5 ml/min, with the head pressure 13 psi. A weighed amount of crab meat sample was treated with hydrochloric acid (6N, 15 ml, 145°C, 4 h), and oxidized samples were used for sulphur amino acid. For tryptophan analysis, alkaline hydrolysis was followed instead of acid hydrolysis, since it is stable at basic condition. The solutions were evaporated to dryness using a rotary evaporator (Büchi Laboratoriumstechnik AGRE121; Switzerland) connected to a diaphragm vacuum pump (MC2C; Vacuubrand GmbH, Germany) to remove hydrochloric acid. For the process of derivatization, trans-4-(aminomethyl)-cyclohexanecarboxylic acid (purity, 97%; Sigma Aldrich, St. Louis, MO, USA) was added to hydrolysate as internal standard. The standards were treated with dichloromethane and dried to remove moisture by flushing inert helium with passive heating in an oil bath (60°C). The acidified isopropanol (12 ml) (acetyl chloride + 2-propanol) (1:4 v/v) was added following heating (100°C, 1 h). The contents were evaporated to dryness in the oil bath at 60°C by gently flushing helium. The dry residue was evaporated and treated with dichloromethane; this process was repeated to remove traces of isopropanol and water. The final residues were treated with trifluroacetic anhydride of 200 ml volume overnight at room temperature. Then, the fraction of the solution was injected in GC-C-IRMS/MS. The details of temperature program are given in Table 1. Classification of Nelson and Cox (2004) was adopted for classification of amino acids as nutritionally "essential" or "non-essential" or "conditionally essential."

Fatty acid analysis

Hot extracted crude fat from crab meat was used for determining the fatty acid methyl esters (FAMEs). The method suggested by Padua-Resurreccion and Banzon (1979) was used for this process. The analytical techniques such as FAMEs were quantified by gas chromatographer (GC-2010, Shimadzu, Japan) equipped with fused silica column (BPX-70) and flame ionization detector (FID). Methylation was performed by the acid-catalyzed method. The trans-esterified samples (100 μl) were made up to 1 ml by *n*-hexane, and the fraction of samples (1 μl) was injected into a gas chromatograph for analysis. The identification of peaks obtained from the lipid profiling was determined by comparing with National Institute of Standards and Technology Library (NIST 11 mass spectrometry library; NIST/EPA/NIH; version # 2011). Further analysis was carried out following the methodology suggested by Nareshkumar (2007).

Table 1. Temperature program for GC-C-IRMS/MS.

Time (min)	Temperature (°C)	Temperature/min
1	50	Start
10	50–100	10°C/min
10	100–175	3°C/min
10	175–200	3°C/min
10	250	Stop

Statistical analysis

Experimental species were subjected to one-way analysis of variance (ANOVA). Independent variables were used to determine the significance level between male and female for the proximate composition, mineral composition, fatty acid, and amino acid analysis using the Statistical Package for Social Sciences, 16.0 software (SPSS Inc., 2007, Chicago, IL, USA).

Results

Description of the species

Charybdis (Goniohellenus) smithii MacLeay, 1838 is a decapod crab belonging to family Portunidae. For the present study, the samples were collected from trawlers operating off Malabar and Konkan coast of Eastern Arabian Sea, at a depth of 100–150 m. Males collected were in the size range of 36–68 mm CW, with a weight range of 4–64 g (mean size 55 mm and mean weight 27 g). In females, the size range and weight were 35–62 mm CW and 4–45 g (mean size of 49 mm CW and mean weight of 21 g), respectively.

Proximate composition

The results of the proximate composition of the males and females *C. smithii* expressed in wet weight are given in Table 2. The dry weight of males and females was 13.02 ± 0.09 g/100 g and 12.07 ± 0.02 g/100 g, respectively, showing more moisture content in females compared to males, whereas the protein and fat content of the males was more than in females. The protein value in males and females was 9.48 ± 0.29 g/100 g and 9.27 ± 0.29 g/100 g, respectively, and fat content was 0.92 ± 0.02 g/100 g in males and 0.79 ± 0.17 g/100 g in females. Similarly, ash content and carbohydrate were higher in males, but fiber content was higher in females. The variation in composition between males and females was not statistically significant ($p < 0.001$), except in DM and carbohydrate.

The mineral composition of copper (Cu), zinc (Zn), manganese (Mn), iron (Fe), magnesium (Mg), sodium (Na), potassium (K), and calcium (Ca) were analyzed during the study, and the results are shown in Table 3. The study showed that both male and female crabs are rich in Na, K, Ca, and Mg, and female crabs contained higher amount of minerals than males (Table 3). Minerals in males and females showed highly significant variation ($p < 0.001$) for sodium, potassium, and calcium, whereas for zinc, the variation was significant at 5% level ($p < 0.05$).

After ascertaining the richness in protein content, the samples were further analyzed for amino acid composition. The results of the study are given in Table 4 and expressed in g/100 g protein. Non-essential amino acids, such as alanine, asparate, glutamate, and serine, were analyzed in both sexes. It was found that the crab meat was rich in non-essential amino acids, and meat of male crabs was comparatively richer in these components (Table 4). In the case of essential amino acid, such as histidine, isoleucine, leucine, lysine, methionine, phenylalanine,

Table 2. Proximate composition of crab *C. smithii* in g/100 g wet weight.

Proximate composition	Male	Female
Dry matter	13.02 ± 0.09	12.07 ± 0.02
Moisture	86.98 ± 0. 21	87.93 ± 0.19
Protein	9.48 ± 0.29	9.27 ± 0.29
Fat	0.92 ± 0.02	0.79 ± 0.17
Fiber	0.11 ± 0.03	0.14 ± 0.02
Carbohydrate	2.1 ± 0.00	1.53 ± 0.01
Ash	0.37 ± 0.04	0.30 ± 0.03

Table 3. Mineral composition of crab *C. smithii* in mg/100 g wet weight.

Mineral composition	Male	Female
Copper	0.28 ± 0.01	0.15 ± 0.02
Zinc	1.714 ± 0.56	2.75 ± 0.22
Manganese	0 ± 0	0.02 ± 0
Iron	0.574 ± 0.21	0.71 ± 0.01
Magnesium	34.31 ± 1.2	41.49 ± 0.29
Sodium	317.1 ± 0.86	327.6 ± 0.37
Potassium	148 ± 0.72	177.4 ± 0.49
Calcium	187.9 ± 0.55	285.8 ± 0.37

Values are given as mean ± SD from triplicate determinations.

Table 4. Amino acid profile of crab *C. smithii* in g/100 g protein.

Amino acid profile in g/100 g	Male	Female
Non-essential amino acids		
Asp	4.06 ± 0.42	3.92 ± 0.36
Ser	2.84 ± 0.03	2.95 ± 0.25
Glu	9.73 ± 0.55	8.80 ± 0.25
Ala	8.02 ± 0.37	8.56 ± 0.84
Total NEAA	24.65	24.23
Conditionally essential amino acids		
Arg	8.46 ± 0.21	5.28 ± 2.59
Cys	0.25 ± 0.03	0.15 ± 0.07
Gly	12.03 ± 1.42	11.47 ± 0.12
Pro	5.81 ± 0.59	6.63 ± 0.59
Tyr	3.12 ± 0.25	3.3 ± 0.11
Total CEAA	29.67	26.83
Essential amino acids		
His	1.82 ± 0.23	2.32 ± 0.47
Ile	5.36 ± 0.13	5.52 ± 0.49
Leu	8.17 ± 0.30	8.15 ± 0.96
Lys	13.95 ± 0.29	12.65 ± 0.57
Met	2.66 ± 0.19	2.97 ± 0.02
Phe	3.83 ± 0.03	3.99 ± 0.36
Thr	3.59 ± 0.02	4.53 ± 1.05
Val	5.71 ± 0.26	6.14 ± 0.41
Trp	ND	ND
Total EAA	45.09	46.27

Classification of AAs as nutritionally "essential" or "nonessential" or "conditionally essential" is as per Nelson and Cox (2004). Values are reported as mean ± SD of three replicates; ND: not detected.

threonine, valine, and tryptophan, amounts were higher in females (Table 4.) The variation of amino acids glutamate, lysine, and methionine between the sexes showed statistical significance at 5% level ($p < 0.05$); for the other amino acids, the variation noted between the sexes was not statistically significant.

The present study showed that *C smithii* possess high fat content (males, 0.92 ± 0.02 g/100 g and females, 0.79 ± 0.17 g/100 g). Fatty acid profile of the species was performed in order to understand the composition of these fats; results are given in Table 5. Among the saturated fatty acids (SFA), palmitic acid (C16), stearic acid (C18), palmitoleic acid (C16:1 Cis), and margaric acid (C17) were the dominant ones detected from the crab meat. Out of these, palmitic acid was the most dominant SFA, constituting 53.2% of the total SFA. High levels of docosahexaenoic acid (DHA) (9.18 g/100 g) and eicosapentaenoic acid (EPA) (10.35 g/100 g) were also found in the crabs (Table 5). The ratio of EPA/DHA was found to be 1.12 (Table 6). For polyunsaturated fatty acids (PUFA), linoleic acid 3.67 g/100 g was the dominant constituent, followed by EPA at 10.35 g/100 g and DHA at 9.18 g/100 g (Table 5).

Table 5. Fatty acid profile of crab *C. smithii* in g/100 g lipid.

	Fatty acid profile	g/100 g
	Saturated fatty acid	
C16:0	Palmitic acid	27.33 ± 0.08
C16:1 Cis	Palmitoleic acid	4.11 ± 0.01
C17:0	Margaric acid	3.89 ± 0.01
C18:0	Stearic acid	15.98 ± 0.05
	Monounsaturated fatty acid	
C18: 1 Cis	Oleic acid	25.46 ± 0.07
	Polyunsaturated fatty acid	
C18: 2 Cis	Linoleic acid	3.67 ± 0.01
C20: 5 Cis	Eicosapentaenoic acid (EPA)	10.35 ± 0.07
C22:6	Docosahexaenoic acid (DHA)	9.18 ± 0.02

Table 6. Sum of various classes of fatty acids and their ratio.

Fatty acid	%
Total	100
SFA	51.31
MUFA	25.46
PUFA	23.2
PUFA/SFA	0.45
EPA/DHA	1.12

Discussion

The only reported work on the biochemical composition of the species gave a moisture content of 86–90%, which is comparable to the present result (Balasubramanian and Suseelan, 2001a). While calculating wet weight, protein and CL content are estimated as 9.27–9.48 g/100 g and 0.79 g/100 g to 0.9 2 g/100 g, respectively (which is equivalent to 73.07–76.99 g/100 g protein and 6.60–7.13 g/100 g fat in dry weight). The results of the present study are slightly higher than those reported by Balasubramanian and Suseelan (2001b) with dry weight calculation (59.8–71.1% protein and 6.2–8.2% fat). This is comparable with shrimp *Penaeus monodon*, using wet weight for comparison (Premarathna et al., 2018). Carbohydrate composition in the present study (2.1 g/100 g and 1.53 g/100 g) is similar to results reported in earlier studies (0.4 and 0.3 g/100 g of wet weight) (Musaiger and Al-Rumaidh, 2005). Comparing the studies on protein and carbohydrate content of other tropical portunid crabs (Badawi, 1971; Radhakrishnan and Natarajan, 1979; Thomas, 1985; Oramadike and Oluwakemi, 2015), *C. smithii* was found to have more or less similar concentrations. Richness in Na, K, Ca, and Mg is identified as one of the major qualities of edible seafood (Chakraborty et al., 2016; Soundarapandian et al., 2014) and these constituents were found in considerably good quantity in the meat of *C. smithii*. Mineral content was comparable to that found by Musaiger and Al-Rumaidh (2005). EPA and DHA concentration in the Malaysian edible crab, *Liocarcinus vernalis*, was estimated to be 5 and 7%, respectively (Rosli Wan et al., 2012), whereas *C. smithii* showed 9 and 10%, respectively. The rich composition of amino acids and unsaturated fatty acids (omega-3 and omega-6 fatty acids) are considered as major criteria to qualify for a recommendation as healthy food (Dey Snigdha et al., 2017; Kris-Etherton et al., 2002; Swanson et al., 2012). The present study found crab meat of *C. smithii* to have a wide amino acid profile and higher levels of linoleic acid (3.67 g/100 g), DHA (9.18 g/100 g), and EPA (10.35 g/100 g). High omega-6 concentration is reported to have physiological importance; because it accumulates in adipose tissues of the body (Jandacek, 2017), it can be projected as a favorable factor for recommending *C. smithii* meat as a healthy food.

Conclusion

Based on the nutritive value of the species derived from this study, the popularization of *C. smithii* as human food can be pursued with awareness programs, and methods can be found to extract the various nutritive components from the crab meat to be used as food additives. In regard to fishery economics, increase in market demand of these crabs will result in the landing of the crabs in fresh and preserved form, thereby increasing the income of the fishermen as well as effectively reducing resource loss due to discarding.

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Disclosure statement

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