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Nutritional profiling of selected species of edible marine molluscs from the south-west coast of India

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ABSTRACT

The current study determined the nutritional parameters of selected species of edible marine molluscs *viz.*, Indian squid *Uroteuthis* (*Photololigo*) *duvaucelii,* veined octopus *Amphioctopus marginatus*, spineless cuttlefish *Sepiella inermis* and edible oyster *Crassostrea bilineata* (=*madrasensis*) from the Arabian Sea and estuarine systems of the south-west coast of India. The selected species demonstrated balanced essential to non-essential amino acids ratio (1.04-1.52). *U. (P.) duvaucelii* exhibited greater quantities of sulfur comprising amino acids (0.102 g 100 g^{-1} wet weight) and lysine (1.566 g 100 g^{-1}). Among polyunsaturated fatty acids (PUFAs) eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA) were found to be prominent in edible portion of the molluscs studied (7.6-10.3 and 8.7-17.4% respectively). *A. marginatus* exhibited significantly greater *n*-3/*n*-6 PUFA (~7, $p<0.05$) than other molluscs. Lower thrombogenicity and atherogenicity indices (<0.45 and <0.85, respectively) make the mollusc studied during the present investigation, valuable food items for cardio-protection and anti-platelet aggregation. Higher content of vitamin D_3 (489 IU) and vitamin K1 (1.84 µg 100 g^{-1}) in *C. bilineata* signified their importance in preventing osteoporosis. The results reveal that these species are good sources of essential elements and toxic metals were below threshold limits of recommended standards for human consumption.

Keywords: Amino acids, Atherogenicity index, Marine molluscs, Polyunsaturated fatty acids, Thrombogenicity index, Vitamins

Introduction

Molluscs are considered as nutritious seafood and culinary delicacies. The phylum Mollusca represents one of the most diverse groups, comprising about 23% of all named marine organisms. The edible oyster *Crassostrea bilineata* (=*madrasensis*) a dominant molluscan species, is widely distributed in backwaters and estuaries of the coastal regions of the Indian subcontinent. During the past several years, culture of oysters and cephalopods has gained importance due to their delicacy and nutritive value. Several published information are available on the nutritional profiling of certain marine molluscs (Chiou *et al.*, 2001; Salwa *et al*., 2007). They contain relatively low fat and high polyunsaturated fatty acids (PUFAs), some of which cannot be synthesised by human and must be obtained from their diet (Smoothey, 2013). Evaluation of proximate profiles is often essential to guarantee that they meet requirements of commercial specifications and food regulations. The mollusc species *Uroteuthis (Photololog) duvaucelii, Amphioctopus. marginatus, Sepiella. inermis* and *Crassostrea bilineata.* encompasses a major share in the Indian fishery sector contributing to socio-economic development of the country (FAO, 2016, 2018). This paper describes the nutritional profiling of *U. (P.) duvaucelii, A. marginatus, S. inermis* and *C. bilineata* with an objective to enhance its acceptance among users and for future policy formulation for sustainable utilisation of the resources.

Materials and methods

Study area and preparation of sample

U. (P.) duvaucelii, S. inermis and *A. marginatus* (5 kg each) were collected from Cochin Fisheries Harbour (8°48'N; 78°9'E) along the south-west coast of India. After collection, the samples were washed to eliminate the extraneous materials and ink gland was precisely removed as nutritional evaluation of edible parts was intended. *C. bilineata* (5 kg) was harvested from the Ashtamudi Lake of Kollam District (9°3′N; 76°53′E), depurated for 5 h and edible meat was separated from the shells. The tissues were ground and stored at -80°C for further biochemical analyses.

Biochemical analyses

Proximate compositions of mollusc species were determined by standard methods; *viz.,* protein (Lowry

et al., 1951), carbohydrate (AOAC, 2005) and lipid (Folch *et al.*, 1957). Moisture content (AOAC, 1990) and crude ash of samples were analysed by established methods (Joy and Chakraborty, 2017). Amino acids were estimated as described by Chakraborty and Joseph (2015) and the amino acid score for essential amino acids was calculated as described earlier (FAO/WHO/UNU, 2007). The total cholesterol was determined spectrophotometrically (Varian Cary 50, Palo Alto, CA, USA) as described by Wanasundara and Shahidi, (1999) with suitable alteration. Samples (10-30 mg) were weighed and mixed thoroughly with 0.3 ml of 33 % (w/v) KOH and 3.0 ml of 95% (v/v) aqueous ethanol. The mixture was heated to 70-80°C in a water bath for 15 min and after cooling, 10 ml of hexane and 3 ml of distilled water were added to the mixture. The resulting solution was allowed to separate into two layers. Aliquots (1 ml) of upper hexane layer were separated and the solvent was evaporated under a stream of nitrogen. Two millilitre of O-phthalaldehyde reagent (50 mg dl^{-1} in glacial acetic acid) and 1 ml of concentrated sulphuric acid was carefully added and then mixed thoroughly. After 10 min, the absorbance of the solutions was read at 550 nm. A standard curve was prepared and the cholesterol contents of the samples were expressed as mg per 100 g wet sample. The fatty acid composition of the total lipids was evaluated as described by Metcalf *et al.* (1966) and Chakraborty and Paulraj (2009). The nutritional indices, such as thrombogenicity index (TI) and atherogenicity index (AI) (Ulbricht and Southgate, 1991) as well as hypocholesterolaemic/hypercholesterolaemic (h/H) ratio were calculated (Santos-Silva *et al*., 2002). Fat soluble vitamins (A, D3, E and K1) were analysed by a modified method of Salo-Vaananen *et al*. (2000). Briefly, the stock solutions (1, 10, 25, 50 and 100 ppm) of vitamin standards (Sigma-Aldrich, USA) were stored at -20°C, except vitamin D3, where the stock solutions were stored at 4°C. The lipids (0.1 g) were extracted utilising the established method (Chakraborty *et al.,* 2014), before being hydrolysed (KOH/ MeOH 0.5 N, 2 ml). The hydrolysed mixture (2 ml) was extracted with petroleum ether (fraction of 40-60°C, 15 ml) and washed with deionised water (2×10 ml) to make it alkali-free. The non-saponifiable portion was concentrated under vacuum using a rotary evaporator (Heidolph Instruments, Germany) at 50°C before being reconstituted in MeOH. The latter was filtered through a syringe filter (0.2 mm) before being injected (20 ml) in the HPLC (Shimadzu LC 20AD). The HPLC system was equipped with a reverse phase column (Phenomenex, C_{18} 250 mm length, 4.6 mm inner dia), that was housed in a column oven (32°C) and connected to a photodiode array detector. The gradient program was as follows: 20% MeOH (HPLC grade) up to 3 min, which was increased to 100% over the next 5 min, and held for 37 min, with a complete run time of 45 min. The flow rate was 1 ml min^{-1}. Water soluble vitamin (C) was evaluated on the basis of quantitative discolouration of 2, 6-dichlorophenol indophenol titrimetric method (AOAC, 2005). Estimation of minerals was carried out by an inductively coupled plasma mass spectrometry (iCAP™ Q, Thermo Fisher). The dry samples of molluscs (0.1 g) were digested with 7 ml HNO_3 and 3 ml HCl. After 1 h of open digestion, the samples were digested with a microwave digester (Anton Paar Multiwave Go), digested samples were diluted with milliQ water and the minerals were analysed.

Statistical analyses

One-way analysis of variance (ANOVA) was carried out with Statistical Program for Social Sciences 13.0 (SPSS, USA) to measure significant differences between the means. The significant differences were represented as $p<0.05$ and values were assigned as mean of triplicates ± standard deviation. The mean variance data set was distinguished using principal component analysis (PCA) and variables were selected nutritional parameters.

Results and discussion

The total carbohydrate content was recorded to be greater in *C. bilineata* (13.39 g 100 g^{-1} wet weight). Bivalves were reported to store carbohydrates in larger quantities during their growing season apparently as a source of energy during the active period of growth and use them over the rest of the year (Salaskar and Naik, 2011; Shafakatullah *et al*., 2013). The samples of *C. bilineata* were collected during post-monsoon period and that appropriately corroborated the theory put forth by previous reports. The moisture content of the mollusc ranged from 79 to 85% of wet tissue (Table 1).

The molluscs considered in this study were found to be rich sources of protein (12.8-18.3 g 100 g^{-1}), which is essential for human growth and development (Table 2). An earlier report indicated comparable protein content of *Sepia recurvirostra* (13.16-13.51%) and *Crassostrea rhizophorae* (9-10%) in different seasons (Martino and Cruz, 2004; Nurjanah *et al*., 2012). The balanced quantities of essential to non-essential amino acids (1.04-1.52) demonstrated that these molluscs were good sources of protein. The most abundant essential amino acid in *U. (P.) duvaucelii* was found to be arginine (0.43 g 100 g^{-1}), followed by leucine (0.30 g 100 g^{-1}) and lysine (0.287 g 100 g^{-1}) (Table 2). The cholesterolemic properties of proteins were related to their amino acid content and arginine/lysine ratio (Rajmohan and Kurup, 1997). The molluscs exhibited an arginine to lysine ratio of ~2.3, thereby signifying their importance to retain the good cholestrolemic index. The aromatic amino acids are

Table 1. Proximate compositions (g 100 g^{-1} wet tissue) of edible marine molluscs

	U. (P.) duvaucelii	*C. bilineata*	*A. marginatus*	*S. inermis*
Lipid	0.92±0.16[a]	1.19±0.12[b]	1.58±0.19[c]	1.24±0.12[b]
Protein	14.08±0.38[a]	18.36±0.13[b]	13.54±0.24[c]	12.89±0.15[d]
Carbohydrate	10.55±0.16[a]	13.39±0.12[b]	11.72±0.19[c]	9.86±0.12[d]
Moisture	85.21±0.20[a]	82.11±0.11[b]	80.36±0.36[b]	79.11±0.17[b]
Crude ash	0.91±0.11[a]	1.32±0.37[b]	0.74±0.05[c]	0.65±0.06[d]
Cholesterol (mg 100 g^{-1} wet tissue)	175.61±1.10[a]	34.74±0.19[b]	99.43±0.30[c]	148.26±1.26[d]

Data expressed as mean ± standard deviation (n=3)
Means followed by the different superscripts (a-d) within same row indicate significant difference ($p<0.05$)

precursors for the synthesis of catecholamines and thyroid hormone (Logan and Rice, 1987), which was found to be significantly greater in *U. (P.) duvaucelii* (0.324 g 100 g^{-1}) ($p<0.05$). The molluscs exhibited leucine-isoleucine proportion (~1.5) as prescribed by FAO/WHO (FAO/WHO, 1990). Leucine is a branched chain amino acid, which was reported to stimulate protein synthesis (Tipton, 2017); whereas previous reports indicate that excess leucine affected the metabolic pathways of niacin and tryptophan causing niacin deficiency (Gopalan and Srikantia, 1960; Raghuramulu *et al.,* 1965). An earlier report deduced that dietary leucine-isoleucine proportion could be correlated to pellagra, and that supplementing the amino acid isoleucine might alter the irregularities caused by excess contents of dietary leucine (Krishnaswamy and Gopalan, 1971). Sulfur-containing amino acids are recognised as effective modulators of lipid metabolism and beneficial against metabolic syndrome (Oda, 2006). These amino acids were found to be in greater quantities in *C. bilineata* and *U. (P.) duvaucelii* (0.09 and 0.102 g 100 g^{-1} wet weight) than those recorded in other molluscs. This appropriately indicated that the protein from mollusc could effectively supplement the limiting amino acids in daily diets. The amino acid scores of mollusc species regarding essential and non-essential amino acids are shown in Table 2 and the scores were significantly greater in *U. (P.) duvaucelii* as compared to other mollusc species ($p<0.05$).

The lipid content of cephalopods and bivalves were found to be significantly lower (0.92 to 1.58 g 100 g^{-1}) (Table 3) when compared with Nudibranch molluscs (1.42-2.14 g 100 g^{-1}) (Zhukova, 2014). Cholesterol content of *C. bilineata* was lower (34.74 mg 100 g^{-1}) than those in cephalopods (99-176 mg 100 g^{-1}). Fatty acids are principal components required by the body for diverse metabolic functions and total saturated fatty acid (SFA) ranged from 36 to 43%, among which palmitic acid being prominent (20.1-24.7%) (Table 3). *A. marginatus* recorded greater PUFAs (34.37%) and aggregate of EPA and DHA (27.7%) are of great significance as they play an imperative role in prevention of cardiovascular ailments (Breslow, 2006). The *n*-3/*n*-6 ratio is a useful biomedical marker, an increase in ratio is critical to help in the prevention of coronary heart disease and reduce cancer risk (Chakraborty *et al.*, 2014), whereas *A. marginatus* exhibited significantly greater *n*-3/*n*-6 PUFA proportion (~7, $p<0.05$). *A. marginatus* displayed a higher ∑PUFA/∑SFA ratio (0.9) than the suggested value (above 0.45) for healthy diet (HMSO, 2001). Lower levels of TI and AI in the edible parts of molluscs varied from 0.60-0.83 and 0.3-0.45, respectively, which might reduce the potential risk of developing arthrosclerosis (Tonial, 2014). The h/H ratio could provide an appropriate validation of lipids as it is based on the lipid metabolism. The greater value of this ratio is desirable and found to be ≥1.5 in the mollusc species studied.

The correlations among the nutritional parameters of molluscs were statistically evaluated utilising principal component analysis with respect to DHA, EPA, Σ*n*-3, AI, TI and Σ*n*-3/Σ*n*-6 (Fig. 1). The loading plot displayed PC1 as 83.6% variance and PC2 demonstrated a variance of 16.4%. The DHA, EPA, Σ*n*-3 of *A. marginatus* showed positive correlation with AI, TI and Σ*n*-3/Σ*n*-6 while the DHA, Σ*n*-3 of *C. bilineata* exhibited significant positive correlation with its nutritional indices. The aggregate *n*-3 fatty acids of *U. (P.) uvaucelii* and TI were found to be significantly correlated, whereas EPA of *S. inermis* displayed positive correlation with TI.

Vitamin E (lipid-soluble) and vitamin C (water soluble) are major components in cell antioxidant defense system and is specifically obtained from diet. *A. marginatus* was found to possess greater contents of vitamin E (16.61 IU), whereas vitamin C was found to be significantly greater in *C. bilineata* (48.36 µg 100 g^{-1}). Vitamin D3 was found to be greater in *C. bilineata* (489 IU) and provides a protective effect against multiple diseases such as cancer, type-1 diabetes by regulating calcium and phosphorus absorption.

Molluscs are good bio-indicators for macro-minerals and trace elements availability in surrounding environment (Cravo and Bebianno, 2005). The most abundant

Table 2. Amino acid composition (g 100 g^{-1} wet weight) of the edible marine molluscs

	U. (P.) duvaucelii	*C. bilineata*	*A. marginatus*	*S. inermis*
Essential amino acids				
His	0.09±0.003^{a}	ND	0.01±0.00^{c}	0.04±0.00^{d}
Arg	0.43±0.003^{a}	0.34±0.00^{b}	0.35±0.00^{c}	0.43±0.00^{a}
Thr	0.14±0.001^{a}	0.12±0.00^{b}	0.07±0.00^{c}	0.11±0.00^{d}
Val	0.17±0.002^{a}	0.15±0.01^{b}	0.01±0.00^{c}	0.11±0.00^{d}
Met	0.10±0.001^{a}	0.09±0.00^{a}	0.05±0.00^{b}	0.07±0.00^{c}
Ileu	0.18±0.002^{a}	0.14±0.00^{b}	0.06±0.00^{c}	0.12±0.00^{d}
Leu	0.30±0.005^{a}	0.22±0.00^{b}	0.09±0.00^{c}	0.18±0.00^{d}
Phe	0.16±0.003^{a}	0.11±0.01^{b}	0.09±0.00^{c}	0.12±0.00^{b}
Lys	0.29±0.002^{a}	0.16±0.00^{b}	0.14±0.00^{c}	0.17±0.01^{d}
∑ EAA	1.85±0.02^{a}	1.34±0.00^{b}	0.87±0.00^{c}	1.34±0.01^{d}
Non-essential amino acids				
Asp	0.39±0.002^{a}	0.19±0.00^{b}	0.09±0.01^{c}	0.18±0.00^{d}
Glu	0.59±0.002^{a}	0.43±0.00^{b}	0.17±0.00^{c}	0.32±0.00^{d}
Ser	0.10±0.002^{a}	0.09±0.01^{a}	0.05±0.00^{b}	ND
Gly	0.20±0.002^{a}	0.18±0.00^{a}	0.11±0.03^{b}	0.03±0.00^{c}
Ala	0.21±0.005^{a}	0.15±0.00^{b}	0.06±0.00^{c}	0.11±0.01^{d}
Pro	0.13±0.004^{a}	0.16±0.00^{b}	0.14±0.00^{c}	0.11±0.00^{d}
Tyr	0.07±0.003^{a}	0.07±0.00^{a}	0.04±0.00^{b}	0.06±0.00^{c}
Cys	ND	ND	ND	ND
∑ NEAA	1.71±0.03^{a}	1.28±0.00^{b}	0.66±0.00^{c}	0.81±0.00^{d}
Ratio of amino acids				
∑ AA	3.55±0.02^{a}	2.62±0.00^{b}	1.53±0.00^{c}	2.15±0.00^{d}
∑ EAA/∑ AA	0.52±0.01^{a}	0.51±0.01^{a}	0.57±0.02^{b}	0.62±0.01^{c}
∑ NEAA/∑ AA	0.48±0.01^{a}	0.49±0.01^{a}	0.44±0.02^{b}	0.43±0.01^{b}
∑ EAA/∑ NEAA	1.08±0.01^{a}	1.04±0.02^{a}	1.32±0.02^{b}	1.60±0.01^{c}
∑ ArAA	0.32±0.00^{a}	0.22±0.01^{b}	0.14±0.01^{c}	0.61±0.01^{d}
∑ SCAA	0.10±0.00^{a}	0.09±0.01^{a}	0.05±0.00^{b}	0.07±0.00^{c}
Arg/Lys	1.47±0.02^{a}	2.15±0.03^{b}	2.50±0.02^{c}	2.40±0.01^{c}
Leu/Ileu	1.69±0.01^{a}	1.55±0.02^{b}	1.55±0.01^{b}	1.52±0.01^{b}
Essential amino acid score				
Lys	1.57±0.00^{a}	1.04±0.00^{b}	0.23±0.00^{c}	0.41±0.00^{d}
Met+Cys	1.15±0.00^{a}	1.27±0.01^{b}	0.66±0.01^{c}	0.83±0.01^{d}
Thr	1.48±0.01^{a}	1.59±0.00^{b}	0.81±0.01^{c}	1.21±0.01^{d}
Ileu	1.45±0.01^{a}	1.42±0.01^{b}	0.58±0.01^{c}	0.98±0.00^{d}
Leu	0.65±0.00^{a}	1.11±0.00^{b}	1.06±0.02^{c}	0.76±0.01^{d}
Val	1.08±0.00^{a}	1.12±0.01^{b}	0.46±0.01^{c}	0.75±0.00^{d}
Phe+Tyr	1.48±0.01^{a}	1.71±0.01^{b}	0.95±0.01^{c}	1.18±0.01^{d}
His	1.56±0.00^{a}	0.07±0.00^{b}	0.25±0.01^{c}	0.75±0.01^{d}

∑ EAA-Total essential amino acids; ∑ NEAA-Total non-essential amino acids; ∑ AA-Total amino acids; ∑ ArAA-Total aromatic amino acids; ∑ SCAA-Total sulfur containing amino acids

Tryptophan was not determined. Data are expressed as mean±standard deviation (n=3); ND: Non-detectable. Means followed by different superscripts (a-d) within same row indicate significant difference ($p<0.05$)

macro-elements in the species were potassium (K), calcium (Ca), sodium (Na) and phosphorus (P) (Table 4). Ca and Mg were found in significant quantities in *C. bilineata* (17.9 and 16.5 mg kg^{-1}, respectively). Calcium and phosphate is crucial for maintaining normal physiological condition and an imbalance of this regulation leads to heart and chronic kidney disease (Blaine *et al*., 2015). The mollusc species considered in the present study possessed greater (>95 mg kg^{-1}) content of Ca+P. The K contents of the molluscs were found to be greater than 15 mg kg^{-1} dry weight, which is essential to regulate various electrochemical and catalytic functions for enzyme systems (Suhail, 2010). The balanced sodium-potassium proportion (≤1.0) of mollusc species proved to be safer for individuals having cardiovascular ailments. Copper and zinc are firmly associated with numerous protein functions and found to

Table 3. Fatty acid composition (% total fatty acids) of edible marine molluscs

Fatty acids	*A. marginatus*	*C. bilineata*	*U. (P.) duvaucelii*	*S. inermis*
Saturated fatty acids				
12:0	0.29±0.05[a]	1.03±0.03[b]	0.56±0.04[c]	0.72±0.03[d]
14:0	2.92±0.07[a]	2.64±0.06[b]	2.15±0.07[c]	2.75±0.08[ab]
15:0	1.64±0.14[a]	0.46±0.03[b]	1.83±0.05[c]	2.61±0.06[d]
16:0	21.29±0.13[a]	24.7±0.05[b]	20.1±0.05[c]	22.38±0.07[d]
17:0	2.18±0.02[a]	2.36±0.08[b]	1.15±0.04 [c]	1.82±0.07[d]
18:0	9.63±0.10[a]	8.52±0.07[b]	8.38±0.04[b]	9.92±0.13[c]
20:0	0.18±0.04 [a]	1.37±0.03[b]	0.25±0.04[a]	0.41±0.01[c]
22:0	0.11±0.04[a]	0.28±0.06[b]	0.14±0.02[a]	0.19±0.06[ab]
24:0	0.08±0.003[a]	1.65±0.04[b]	1.29±0.04[c]	1.15±0.15[c]
∑SFA*	38.32±0.37[a]	43.01±0.45[b]	35.85±0.24[c]	41.98±0.48[b]
Monounsaturated fatty acids				
14:1*n*-7	3.25±0.05[a]	2.56±0.06[b]	2.16±0.06[c]	2.23±0.04[c]
15:1*n*-7	2.84±0.09[a]	0.12±0.03[b]	1.62±0.05[c]	1.31±0.02[d]
16:1*n*-7	4.87±0.11[a]	3.48±0.07 [b]	3.63±0.05[b]	3.39±0.03[b]
18:1*n*-7	0.15±0.004[a]	0.07±0.005[b]	0.13±0.02[a]	0.07±0.004[b]
18:1*n*-9	13.61±0.09[a]	18.56±0.10[b]	19.72±0.08[c]	19.98±0.04[d]
20:1*n*-9	0.42±0.04[a]	0.28±0.02[b]	0.05±0.005[c]	0.09±0.002[c]
22:1*n*-9	0.49±0.06[a]	1.3±0.095[b]	1.68±0.04[c]	0.58±0.03[a]
24:1*n*-9	0.29±0.04[a]	0.42±0.03[b]	0.19±0.01[c]	0.18±0.01[a]
∑MUFA**	25.92±0.36 [a]	26.79±0.41[a]	29.18±0.30[b]	27.83±0.12[c]
Polyunsaturated fatty acids				
16:2*n*-4	0.04±0.00[a]	0.12±0.04[b]	0.06±0.01[a]	0.12±0.01[c]
16:3*n*-4	0.04±0.00[a]	0.08±0.002[b]	0.08±0.00[b]	0.05±0.00[c]
18:2*n*-6	1.35±0.05[a]	1.26±0.03[a]	2.42±0.06[b]	1.63±0.02[c]
18:3*n*-6	0.32±0.06[a]	0.59±0.04[b]	0.35±0.02[a]	0.69±0.02[b]
18:3*n*-3	1.19±0.07[a]	3.34±0.10[b]	0.96±0.03[c]	1.21±0.02[a]
20:2*n*-6	2.14±0.04 [a]	1.92±0.05[b]	1.96±0.04[b]	1.85±0.04[b]
20:3*n*-6	0.41±0.05[a]	1.12±0.08[b]	1.16±0.01[b]	0.63±0.03[c]
20:4*n*-6	0.29±0.02[a]	1.05±0.05[b]	0.72±0.03[c]	0.98±0.03[b]
20:5*n*-3 EPA	10.27±0.03[a]	7.57±0.07[b]	8.29±0.05[c]	7.63±0.06[b]
22:5*n*-3	0.89±0.05[a]	0.62±0.02[b]	0.74±0.01[c]	0.58±0.02[b]
22:6*n*-3 DHA	17.43±0.03 [a]	8.68±0.20[b]	14.7±0.05[c]	12.33±0.29[d]
∑PUFA***	34.37±0.37[a]	26.35±0.47[b]	31.44±0.20[c]	27.7±0.45[d]
∑*n*-3	29.78±0.18[a]	20.21±0.02[b]	24.69±0.10[c]	21.75±0.1[d]
∑*n*-6	4.51±0.11[a]	5.93±0.070[b]	6.61±0.07[c]	5.78±0.08[b]
∑*n*-3/∑*n*-6	6.61±0.15[a]	3.40±0.050[b]	3.73±0.05[c]	3.76±0.04[c]
18:1*n*-7/*n*-9	0.01±0.00[a]	0.0037±0.00[a]	0.0066±0.00[a]	0.0035±0.00[a]
DHA+EPA	27.7±0.11[a]	16.25±0.12[b]	22.99±0.06[c]	19.96±0.06[d]
EPA/AA	35.41±0.22[a]	7.21±0.07[b]	11.51±0.04[c]	7.78±0.04[d]
∑PUFA/∑SFA	0.9±0.03[a]	0.611±0.04[b]	0.88±0.02[c]	0.66±0.02[b]
AI	0.71±0.04[a]	0.83±0.02[b]	0.61±0.05[c]	0.78±0.03[ab]
TI	0.3±0.04[a]	0.45±0.01[b]	0.32±0.03[a]	0.41±0.04[bc]
h/H ratio	1.86±0.06[a]	1.50±0.05[b]	2.14±0.04[c]	1.76±0.05[a]

*Total saturated fatty acids; **Total monounsaturated fatty acids; ***Total polyunsaturated fatty acids. Data are presented as mean values of three samples (mean±SD). Different superscripts (a-d) within same row indicate significant difference ($p<0.05$)

EPA- Eicosapentaenoic acid, DHA - Docosahexaenoic acid, AA - Arachidonic acid, AI - Atherogenicity Index, TI - Thrombogenicity Index, h/H- hypocholesterolaemic/ hypercholesterolaemic ratio

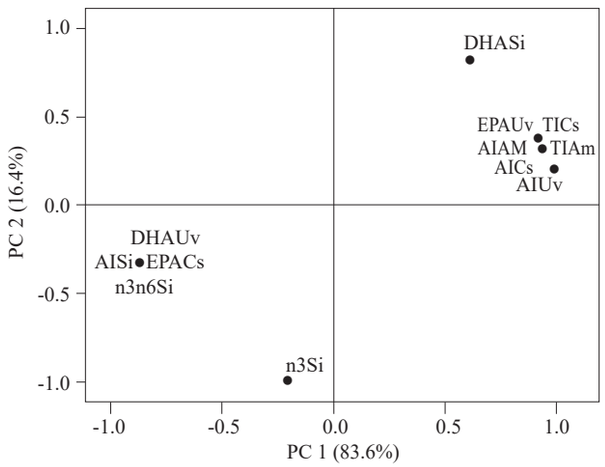

Fig. 1. Loading plot diagram representing the correlation of total *n*-3 fatty acids, EPA, DHA and fatty acid indices of the edible marine molluscs.

Am - *A. marginatus;* Cs - *C. bilineata;* Uv - *U. (P.) duvaucelii;* Si - *S. inermis; n*3-*n*-3 Fatty acids; EPA - Eicosapentaenoic acid; DHA - Docosahexaenoic acid; AI - Atherogenicity index; TI - Thromrbogenicity index; n3n6-*n*-3/*n*-6

be efficiently absorbed and retained in *S. recurvirostra* both from the food and seawater pathways (Nurjanah *et al*., 2012). *C. bilineata* displayed significantly greater content of Fe (2.44 mg kg^{-1}) than other mollusc species studied ($p<0.05$) (Table 4). Selenium is critical for normal functioning of endocrine tissues and is a part of amino acids, such as seleno-cysteine and seleno-methionine with antioxidant functions (Venugopal and Gopakumar, 2017) and was found to be significantly higher ($p<0.05$) in *A. marginatus* (0.06 mg kg^{-1}) than other mollusc species.

Notably, the digestive gland of cephalopods is capable of retention of high atomic weight elements, such as mercury (Hg), cadmium (Cd) and lead (Pb), likely as chelating agents with proteins (Venugopal and Gopakumar, 2017). The EU directive of 1881/2006 and 629/2008 set maximum level of trace metal contaminants in cephalopods (without viscera) species as, Pb - 1 mg kg^{-1}, Cd - 1 mg kg^{-1} and Hg - 0.5 mg kg^{-1} in wet weight basis (Jinadasa, 2014). In the present study, the mollusc species displayed Cd (<1 mg kg^{-1}) and Pb (<1 mg kg^{-1}) level lower than the set maximum level of contaminants.

Table 4. Vitamin and mineral compositions of edible marine molluscs

	U. (P.) duvaucelii	*C. bilineata*	*A. marginatus*	*S. inermis*
Vitamins				
Retinol (A) (IU)	107.33±1.22^{a}	8.09±0.06^{b}	99.42±0.32^{c}	181.14±1.05^{d}
Cholecalciferol (D3) (IU)	71.24±1.14^{a}	489.21±1.12^{b}	93.68±0.34^{c}	81.51±0.82^{d}
α- Tocoferol (E) (IU)	2.15±0.26^{a}	0.21±0.06^{b}	16.61±0.21^{c}	14.45±0.23^{d}
Phylloquinone (K1) (µg 100 g^{-1})	1.01±0.11^{a}	1.84±0.17^{b}	1.48±0.43^{b}	0.73±0.06^{a}
Ascorbic acid (C) (µg 100 g^{-1})	33.26±0.75^{a}	48.36±0.15^{b}	42.62±0.22^{c}	43.17±0.18^{c}
Minerals composition (mg kg^{-1} dry tissue)				
Macrominerals				
Ca	11.13±0.04^{a}	17.92±0.10^{b}	13.32±0.37^{c}	17.16±0.40^{b}
Na	5.11±0.09^{a}	16.57±0.10^{b}	7.65±0.35^{c}	2.84±0.62^{d}
P	118.01±0.50^{a}	92.71±0.35^{b}	83.72±0.23^{c}	78.09±0.30^{d}
K	15.42±0.26^{a}	48.25±0.30^{b}	20.05±0.10^{a}	17.46±0.30^{a}
Mg	12.92±0.20^{a}	16.54±0.60^{b}	12.55±0.50^{a}	14.61±0.36^{c}
Microminerals				
Zn	0.76±0.05^{a}	4.99±0.50^{b}	1.06±0.10^{a}	1.38±0.11^{a}
Mn	0.04±0.00^{a}	1.38±0.38^{b}	0.04±0.00^{a}	0.20±0.08^{a}
Cu	0.15±0.01^{a}	0.86±0.10^{b}	0.26±0.12^{a}	0.53±0.07^{c}
Fe	0.42±0.03^{a}	2.44±0.22^{b}	2.21±0.20^{b}	0.64±0.07^{a}
Se	0.02±0.00^{a}	0.02±0.00^{a}	0.06±0.00^{b}	0.03±0.00^{a}
Toxic elements				
Cd	0.06±006^{a}	0.04±0.00^{a}	0.07±0.00^{a}	0.18±0.04^{b}
Pb	0.041±00^{a}	0.074±0.00^{b}	0.041±0.00^{a}	0.045±0.00^{c}
Mineral indices				
Na/K	0.33±0.03^{a}	0.34±0.05^{a}	0.38±0.08^{a}	0.16±0.07^{b}
Ca+P	129.13±0.90^{a}	109.72±0.60^{b}	96.04±0.60^{c}	95.26±0.7^{c}
Ca/P	0.09±0.03^{a}	0.19±0.07^{b}	0.16±0.05^{c}	0.22±0.06^{b}

Data are expressed as mean ± standard deviation (n=3)

Means followed by different superscripts (a-d) within same row indicate significant difference ($p<0.05$)

The present study comprehensively assessed the biochemical profile of commonly available edible molluscs from the south-west coast of India. Our results suggest that they are valuable sources of protein with more prominent content of essential/non-essential amino acid ratio. They constituted prominent levels of DHA, EPA and *n*-3/*n*-6 fatty acid proportion (3.7-6.6), which supported their utilisation as a balanced diet. The ideal AI, TI, h/H ratio and amino acid based health indicators signified these species as a potential healthy diet. Furthermore, they are rich source of vitamins and numerous crucial macro and micro minerals, which are essential for the metabolic functioning of the human body. The present study could help to promote consumer acceptance of the studied mollusc species as potent health food alternatives to overexploited fish resources.

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