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Evaluation of Nutritional status of Penaeid Prawns through Proximate Composition Studies

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Abstract
The proximate composition and biochemical constituents were analysed in the muscle tissue of penaeid prawns selected in the present investigation namely *Peaneus monodon, P. indicus, P. semisulcatus, Litopenaeus vannamei, Metapenaeus monoceros* and *M. dobsoni.* The Crude protein, Carbohydrate, Lipid, Moisture, Ash, Free amino acids, Free Fatty acids including Saturated, Mono-Unsaturated and Poly-Unsaturated Fatty acids quantified in the muscle tissue of Penaeid Prawns clearly demonstrates the nutritional status of edible Portion of prawns i.e., muscle tissue for human consumption due to its possessing of good quality of Proteins, higher amounts of free amino acids, lower amounts of fats. From the results obtained in the present investigation, it inferred that all the Penaeid prawns selected are supposed to have the ideal nutritional ingredients hence are considered as good source of food for human consumption.

Keywords: Penaeid prawns, Proximate composition, Fatty acids, Amino acids

1. Introduction
In India, millions of people suffering from malnutrition. Protein deficiency may be minimized to some extent by making available cheaper fish meal items which are available to local communities. Edible Crustaceans, such as Crab, Prawn, Cray fish and Lobster constitute one of the major sources of nutritious food for human beings. There are much studies encouraging crustacean consumption [5, 7, 15, 24, 27, 33, 34]. The nutritive values of crustaceans depend upon their biochemical composition, such as Protein, Amino acids, Lipids, Fatty acids, Carbohydrates, Vitamins and Minerals. Among the Seafood, Prawns and Shrimps contribute about 20-22% by volume of the world Seafood market [13]. Due to their nutritious nature, apart from the supply of good quality Proteins, Lipids, they also contain several dietary minerals such as Calcium, Iron etc., which are beneficial and essential and play an important role in maintenance of Physiological and Biochemical activities in human beings. Therefore prawns and shrimps are considered to be most popular species as it is a part of almost every nations traditional meal rich in protein and minerals. Penaeid shrimps constitute a major capture fisheries component in the Aquaculture practices. Among the penaeid prawns *Penaeus monodon, P. indicus, P. semisulcatus, Litopenaeus vannamei, Metapenaeus monoceros, M. dobsoni* are considered to be important major commercial prawns species available in India. Among the above prawn species, several are considered to be most prominent promising candidate species for culture activity in India. Till date the primary focus of prawn nutrition research has been evaluate diet quality in terms of growth parameters of the cultured prawn species, but knowledge of the biochemical composition of edible organisms, which is extremely important since the nutritive value is reflected in biochemical contents, was not attempted for tropical penaeid prawns of Andhra Pradesh. Therefore, the present study is aimed to probe into the aspects for the evaluation of proximate composition of basic biochemical constituents including proteins, lipids, carbohydrates, amino acid profiles, fatty acid profiles etc., to assess the nutritional significance of penaeid prawns.

2. Materials and Methods
Penaeid prawns *Penaeus monodon, P. indicus, P. semisulcatus. Litopenaeus vannamei, Metapenaeus monoceros and M. dobsoni* of equal size were collected from local landings (Latitude 15^0 02^1 55^{11} N, Longitude 80^0 02^1 50^{11} E). All the samples were put in crushed ice in insulated containers and brought to the laboratory for preservation prior to analysis. Prawns were wrapped carefully with Aluminium foil and frozen in deep Refrigerator at - 20°C to

facilitate peeling process after thawing when needed as most crustaceans. After defrosting, the prawns were separated into the exoskeleton and endoskeleton i.e., edible muscle portion. The edible portion i.e., Muscle tissue was used for experimentation and was homogenised in Electrical Homogenizer. The grounded samples were then freeze-dried and powdered and stored in Refrigerator for further Biochemical analysis. The Protein content was estimated by following the method of Lowry *et al* [16] using TCA precipitated sample by as per Folin-Ciocalteu method with Bovine Serum Albumin (BSA) as slandered. The carbohydrate content was estimated by the method Roe [28] by using TCA extracted sample. Total Lipid content was extracted with Chloroform-Methonol mixture following the method of Floch *et al.* [14]. 1 g of powdered tissue in a crucible was kept in a Mussle furnace at 60 °C for 4 h. The residue ash weighed and the percentage was calculated. Moisture content was estimated by Hot Air oven method.

Analysis of Profiles of Amino acids

The individual amino acids were determined in Muscle tissue using LKB Automatic Amino acid analyser. Amino acids were extracted into Ethanol medium and were subsequently discovered in Citrate buffer (0.1 M) and 0.5 ml was loaded for quantification suitable standards also run simultaneously. All the conditions pertaining to the quantification were standardized.

Analysis of Fatty acid Profiles

The Profiles of Fatty acid were estimated by following Gas Chomatography (GC) method [20]. Fatty acids were obtained from Lipids by saponification using Sodium hydroxide dissolved in Methanol-Water mixture (Hydrolysis with alkali). They were methylated into Fatty acid methyl ester using Hydrochloric Acid and Methanol mixture, which can be easily identified by Gas Chromatography. The Fatty acid methyl ester was separated using the mixture of Hexane and anhydrous diethyl ether. For the Organic Phase aqueous Sodium Hydroxide was used as base wash and the upper organic layer was separated. 3µl sample was injected and analysed using Chemito 8610 Gas Chromatography, with BPX 70 capillary column and Frame ionization dictator. Nitrogen was used as carrier gas. The Chromatogram was used for calculation. Slandered Fatty acids were analysed simultaneously. Based on the retention time and peak area, the standard Fatty acids, each Fatty acid in the unknown sample was identified.

The data obtained was subjected to Statistical Analysis in SPSS Version 20.

3. Results and Discussion

Morphometric data obtained for Penaeid prawns *Penaeus monodon, Litopenaeus vannamei, Penaeus indicus, Penaeus semisulcatus, Metapenaeus monoceros and Metapenaeus dobsoni* were recorded and presented in Table. 1 & Figure.1. Total weight of the prawns and Muscle weight (Edible Portion) were recorded and presented in Table. 1. Muscle Somatic Index was calculated for all the Penaeid prawn species and found to be in the range of approximately around 80% (79.24 to 80.91%), clearly indicates the major portion of edible part of the total body. The Proximate composition of Muscle tissues of Penaeid prawns were analysed and presented in Table. 2. Crude Protein, Crude Carbohydrate, Crude Lipid, Moisture content and Ash of Muscle tissue were quantified and presented in Table 2. The profiles of individual amino acids of Muscle tissue of penaeid prawns were analysed and presented in Table 3 & Figures. 2 & 3. Amino acids (Essential) including Arginine, Histidine, Isoleucine, Leucine, Lysine, Methionine, Tryptophan, Valine, Phenylalanine, Threonine and Non-Essential amino acids including Alanine, Asparaginine, Aspartic acid, Cysteine, Glutamic acid, Glycine, Proline, Serine and Tyrosine were quantified and presented in Table. 3. The quantity of Essential Amino acids are relative high compared to Non-Essential Amino acids are statistically significant (Table.4). The Fatty acid types represented by Saturated Fatty acids, Monounsaturated Fatty acids and Polyunsaturated Fatty acids were analysed and presented in Table. 5 and Figure. 4. Anova test result are presented in Table.6.

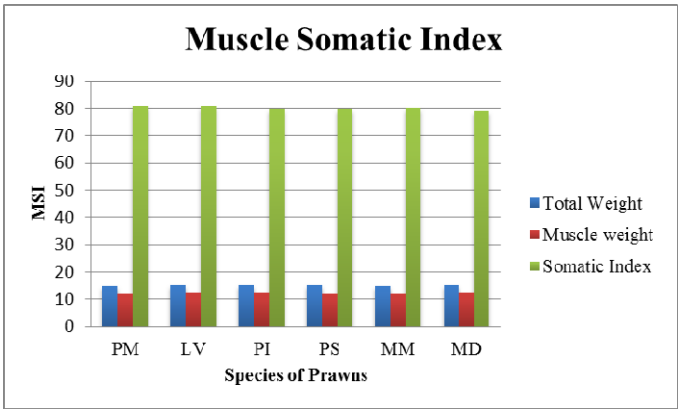

Fig 1: Muscle Somatic Index in selected penaeid prawns

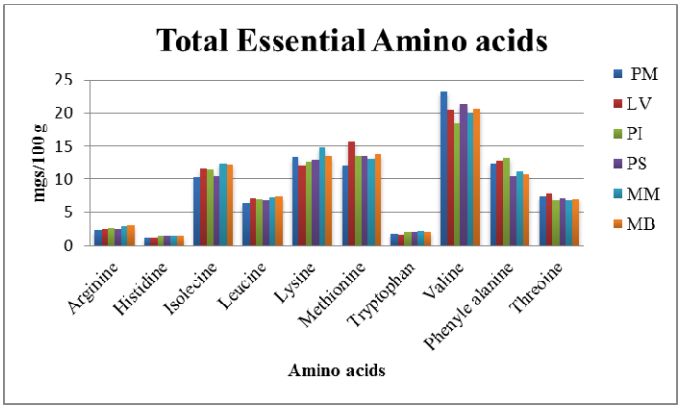

Fig 2: Selected Essential Amino acids in the Muscle tissue of penaeid prawns

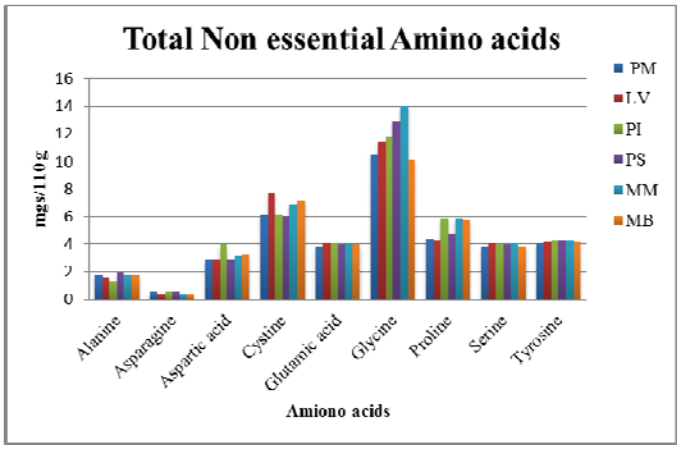

Fig 3: Non-essential Amino acid content of Muscle tissue of penaeid prawns

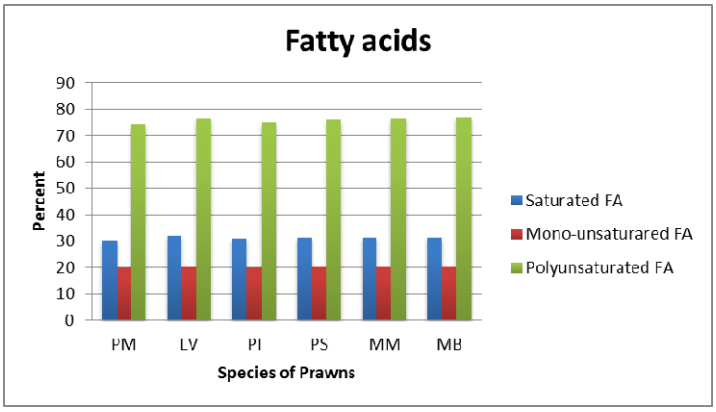

Fig 4: Fatty acid profiles in the Muscle tissue of penaeid prawns.

PM: Penaeus monodon; LV: Litopenaeus vannamei; PS: Penaeus monodon; LV: Litopenaeus vannamei; PI: Penaeus indicus; PS: Penaeus semisulcatus; MM: Metapenaeus monoceros; MD: Metapenaeus dobsoni.

Table 1: Somatic Index of Muscle tissue of selected penaeid prawns

	PM	LV	PI	PS	MM	MD
Total Weight (g)	14.85±0.85	15.22±0.79	15.21±0.78	15.14±0.75	14.95±0.85	15.32±0.76
Muscle weight (g)	12.04±0.48	12.28±0.49	12.13±0.45	12.08±0.53	11.99±0.48	12.14±0.49
Somatic Index	80.91	80.68	79.75	79.78	80.2	79.24

All values Mean ± SD of Six individual observations

Table 2: Proximate Composition of Muscle tissue of selected penaeid prawns

Parameter	Percent in Body weight					
	PM	LV	PI	PS	MM	MD
Crude Protein	36.88±0.92	35.35±0.86	35.13±1.39	34.37±0.88	30.74±0.99	30.48±0.94
Crude Carbohydrate	3.34±0.28	3.18±0.21	3.12±0.18	3.04±0.17	2.85±0.21	2.82±0.22
Crude Lipid	15.34±0.75	15.12±0.68	16.18±0.84	16.72±0.78	17.14±0.82	17.45±0.84
Moisture	76.38±1.34	75.14±1.38	76.18±1.74	75.84±1.75	75.82±1.94	76.14±1.49
Ash	1.39±0.23	1.42±0.28	1.63±0.24	1.49±0.25	1.54±0.24	1.48±0.21

All values Mean ± SD of Six individual observations.

PM: *Penaeus monodon*; LV: *Litopenaeus vannamei*; PS: *Penaeus monodon*; LV: *Litopenaeus vannamei*; PI: *Penaeus indicus*; PS: *Penaeus semisulcatus*; MM: *Metapenaeus Monoceros*; MD: *Metapenaeus dobsoni*.

Table 3. Amino acid content in the Muscle tissue of selected penaeid prawns.

Amino acids	PM	LV	PI	PS	MM	MB
	mgs/100 g					
Essential Amino acids						
Arginine	2.38±0.12	2.42±0.14	2.59±0.14	2.45±0.16	2.89±0.18	2.95±0.19
Histidine	1.12±0.14	1.18±0.14	1.45±0.18	1.35±0.19	1.34±0.15	1.45±0.12
Isolecine	10.43±1.14	11.55±1.15	11.49±1.15	10.45±1.49	12.38±1.48	12.25±1.54
Leucine	6.34±0.56	7.18±0.59	7.05±0.64	6.83±0.65	7.34±0.68	7.38±0.72
Lysine	13.35±1.12	12.14±1.13	12.72±1.34	12.88±1.24	14.75±1.18	13.44±1.15
Methionine	12.14±1.45	15.72±1.49	13.42±1.58	13.44±1.72	13.08±1.65	13.74±1.68
Tryptophan	1.83±0.24	1.72±0.21	2.04±0.18	2.08±0.21	2.14±0.19	2.09±0.21
Valine	23.24±1.58	20.42±1.78	18.43±1.74	21.42±1.84	19.94±1.95	20.63±2.05
Phenyle alanine	12.38±1.14	12.74±1.32	13.13±1.14	10.49±1.04	11.13±1.03	10.73±1.06
Threoine	7.42±0.54	7.89±0.63	6.77±0.52	7.18±0.43	6.74±0.49	7.04±0.42
Total	90.63	92.96	89.09	88.57	81.73	91.7
Non-Essential Amino acids						
Alanine	1.84±0.12	1.54±0.12	1.32±0.18	1.93±0.15	1.83±0.14	1.75±0.14
Asparagine	0.52±0.05	0.38±0.04	0.53±0.03	0.49±0.03	0.42±0.04	0.41±0.05
Aspartic acid	2.88±0.12	2.92±0.16	3.94±0.18	2.95±0.21	3.14±0.22	3.22±0.24
Cystine	6.18±0.79	7.74±0.85	6.13±0.88	6.04±0.89	6.93±0.92	7.18±0.94
Glutamic acid	3.84±0.28	4.12±0.35	4.04±0.38	3.95±0.42	4.05±0.39	3.98±0.41
Glycine	10.53±1.12	11.45±1.14	11.74±1.05	12.94±1.08	13.99±0.98	10.13±1.04
Proline	4.34±0.42	4.31±0.58	5.83±0.64	4.72±0.74	5.84±0.68	5.75±0.71
Serine	3.89±0.24	4.03±0.29	3.95±0.32	3.94±0.28	4.05±0.34	3.89±0.38
Tyrosine	4.03±0.45	4.18±0.54	4.32±0.58	4.21±0.59	4.22±0.68	4.18±0.72
Total	38.05	40.67	41.8	41.17	44.47	40.49

All values Mean ± SD of Six individual observations

Table. 4. ANOVA Result between Total Essential and Non-Essential Amino Acids

ANOVA					
Score					
	Sum of Squares	df	Mean Square	F	Level
Between Groups	6913.440	1	6913.440	689.220	Significant
Within Groups	100.308	10	10.031		
Total	7013.748	11			

Table 5. Saturated and Unsaturated Fatty acids in the Muscle tissue of selected penaeid prawns.

Saturated Fatty acids	**Percent**					
	PM	**LV**	**PI**	**PS**	**MM**	**MB**
Palmitic Acid	8.89±0.23	9.02±0.28	8.93±0.29	9.14±0.32	9.22±0.44	9.18±0.31
Margaric Acid	2.78±0.12	2.88±0.10	2.91±0.15	2.74±0.12	2.89±0.14	2.88±0.14
Stearic Acid	18.75±0.89	20.05±1.12	19.14±1.11	19.38±1.12	19.34±1.12	19.35±1.21
Total	**30.42**	**31.95**	**30.98**	**31.26**	**31.45**	**31.41**
Mono-unsaturated FA						
Oleic Acid	19.68±1.14	20.12±1.18	19.75±1.25	20.33±1.31	20.28±1.19	20.34±1.22
Polyunsaturated FA						
Linoleic Acid	20.18±1.24	20.75±1.28	20.44±1.35	20.38±1.42	21.05±1.38	21.42±1.34
Alpha-Linolenic Acid	16.34±1.25	17.05±1.21	16.75±1.22	16.88±1.31	16.72±1.28	16.82±1.24
Stearic Acid	0.89±0.05	1.02±0.10	0.94±0.08	1.08±0.12	1.04±0.11	0.95±0.08
Eicosatrienoic Acid	7.33±0.28	7.04±0.34	6.73±0.38	6.84±0.49	6.92±0.54	6.82±0.62
Eicosapentanoic Acid	17.42±1.14	17.05±1.12	16.74±0.98	16.82±1.12	16.75±1.08	16.96±1.09
Docosahexaenoic Acid	17.33±1.45	17.74±1.38	17.82±1.42	17.38±1.48	17.77±1.54	17.95±1.53
Total	**99.17**	**100.77**	**99.17**	**99.71**	**100.53**	**101.26**
Total (n-3)	78.99	80.02	78.73	79.33	79.48	79.84
Total (n-6)	20.18	20.75	20.44	20.38	21.05	21.42
n-3/n-6 Ratio	3.91	3.86	3.85	3.89	3.78	3.73

All values Mean ± SD of Six individual observations

Table 6: ANOVA Test Result between Saturated, Mono Unsaturated and Poly Unsaturated Fatty Acids

ANOVA					
Score					
	Sum of Squares	**df**	**Mean Square**	**F**	**Sig.**
Between Groups	10455.582	2	5227.791	10461.208	Significant
Within Groups	7.496	15	.500		
Total	10463.078	17			

Prawns are considered to be high-range Protein containing dietary substances. Generally the Biochemical composition of any organism known to reflect its nutritional quality and is being influenced by several biotic and abiotic factors including season, size of the animal, food, temperature and stage in the life cycle etc., Crustacean group of animals play an important role in the food chain cycle as they are being consumed in large numbers by human beings. Due to their delicious nature, high protein and good amino acid content several crustaceans are preferred as food stuffs for human consumption. The penaeid prawns constitute one of the major groups of crustaceans due to possessing of highly nutritious and forms of good sources of proteins and high amounts of amino acids. In addition, due to the presence of relatively lower fibre content in the edible position of penaeid shrimps has got the nutritional advantage, which generally reduces the constipation and other related problems in human beings. The nutritive values of crustaceans depend upon their biochemical composition, such as protein, amino acids, lipids, fatty acids, carbohydrates, vitamins, minerals, related biomolecules etc., In the present study, an attempt was made to probe into the understanding of the biochemical composition of penaeid prawns *Penaeus monodon, Penaeus indicus, Penaeus semisuclatus, Litopenaeus vannamei, Metapenaeus monoceres and Metapenaes dobsoni*. The above selected penaeid prawns constitute one of the most valuable fishery resources of the world generally and particularly with reference to India, and are considered to be most important candidate species for Aquaculture practices. In Aquaculture the length–weight data provide some useful information in market oriented farm management [9, 23, 25]. In the present study, the collected prawns were of marketable size and such sized prawns fetch up to Rs 400-800/-Kg in the local market depending on the availability and demand. The analysis of proximate composition includes the estimation of organic and inorganic constituents of the body. The main constituents are Proteins, Amino acids, Carbohydrates and Lipids. In addition to that prawns also contain a significant proportion of minerals such as Calcium, Phosphorous, Magnesium, Manganese, Chloride etc., and vitamins such as A, D and C [2]. The proximate body composition including moisture, fat, protein, ash etc., are also considered as good indicators of physiological condition of an organism. The higher the protein and lipid contents of the tissues also reveals their involvement in energy production at cellular level [11]. However, the quantities of these constituents may vary considerably within the species and between the species, size, sex, sexual condition, feeding season, athletic activity, Molting stage, Reproductive stage of the life cycle etc., [19, 29]. The results obtained in the present investigation clearly demonstrate that, the proportion of protein content was dominating over carbohydrates and lipid contents in muscle tissue. One of the main objectives of the prawn culture operation is the transformation of dietary protein into tissue protein. Generally, the proteins are essential for normal function, growth, and maintenance of body tissue and hence protein content is considered to be an important tool for the evaluation of biochemical and physiological standards of a

given organism. It has been reported that the protein content of prawn species [1, 30, 38] but in *P. indicus* it was reported as 45% [30]. The amino acids are the building blocks of protein serve as body builders and generally they are being utilized to form different cell structures, of which they are key components and they serve as sources of energy. The edible crustacean muscle contain relatively greater quantities of the amino acids, such as Isoleucine, Histidine, Methionine, Valine, Glycine, Proline, Cysteine, Tyrosine etc.,[6, 10, 21, 26]. The high protein content in the lowest size groups may be attributed to the increased protein synthesis rates during the active growth phase as it has been observed and reported in the case of different shrimp species [3, 4, 22, 32]. In the present investigation the protein content ranged around 30-37%. Carbohydrate content exhibited an inverse relationship with protein content.

In the present investigation both essential-amino acids and non-essential amino acids in the Muscle tissue were quantified. Under Essential-amino acids category, Arginine, Histidine, Isoleucine and threonine and in Non-essential amino acid banner, Alanine, Asparagine, Aspartic acid, Cysteine, Glutamic acid, Glycine, Proline, Serine and Tyrosine were quantified then the muscle tissue of selected Penaeid prawns and presented in Table. 3. The free amino acids are known to play an important role in different functions as the body of crustaceans such as Osmoregulation [12], neurotransmitter [17], metabolic pathways of growth including protein synthesis [6, 21, 37], allergic and inflammatory reactions. The essential amino acid content of muscle tissue dominate over non-essential amino acids, clearly reflects the need and requirement of essential-amino acids for the substances of physiological and biochemical activity in the penaeid prawns. Generally, the essential amino acids are being procured through diet to meet the body requirements of prawns i.e., to show the proportionate muscle growth. In the present investigation the prawns either collected from the culture environment or from the wild environment, they are showing the relatively higher the total quantities of essential amino acid quantities compared to Non-essential amino acid quantities of muscle tissues in all the Penaeid prawns selected in the present investigation.

Among the body organic nutrients, Carbohydrates are considered to be the first substances to be utilised for the synthesis of energy required for physiological activities. Carbohydrates serve as precursors for the synthesis of dispensable amino acids and certain nutrients, which in free and bound state along with proteins as protein-bound sugars and glycogen. In the present investigation the carbohydrate content in all the penaeid prawns ranges in an around 3% of the body weight. Several factors are known to influence carbohydrate content, in different tissues of crustaceans including, gonad developmental stage, Starvation, feeding rates, physical activity and other physiological conditions of the animal. From the results obtained, it is very clear that carbohydrate content of muscle tissue registered relatively very low compared to proteins and lipids, the other important organic molecules of the cellular environment.

The lipids are extremely important organic molecules available at the cellular level, and play an important role in maintaining structural and physiological integrity of the cellular membranes. Generally lipids act as major food reserves and known to play an important role in the production of energy at cellular level and also play vital role in the production of cells by maintaining structural integrity of the cells. The lipid also acts as vehicles for the transport of lipid soluble vitamins A, D, E and K. In the present study, the total lipid content appears to be ranging around 15 to 17% of the total body weight in all the penaeid prawns selected in the present investigation. Three different types of fatty acids were quantified in the muscle tissue of selected penaeid prawns namely Saturated Fatty acids (SF), Mono unsaturated Fatty acids (MUFA), and Poly unsaturated Fatty acids (PUFA) were presented in the Table. 5. Among the Three groups of Fatty acid profiles, PUFA are more dominating over SF and MUFA in the Muscle tissue of all the penaeid prawns selected in the present investigation. In the case of crustaceans, the hepatopancreas is considered to the main seat of metabolism and is considered to be the depot for lipids, including Triglycerides, Phospholipids etc., but it has been already reported that the muscle tissue contains only Phospholipids [8, 18]. The lipid quantity is relatively lower in the muscle tissue of Crustaceans and also in the case of penaeid prawns. Therefore, prawns and other sea foods are being preferred for consumption by human beings. But lipid storage and its subsequent utilization is also recorded to be majority from hepatopancreas of crustaceans. The higher quantity of total lipids and fatty acids recorded in the hepatopancreas of prawns may be attributed for performing certain specific physiological activity related that the incorporation of fatty acids in the diet produced better growth rates and survival in several candidate species of aquaculture. In the present study the quantities of PUFA, particularly Linoleic acid, EP and DHA are higher compared to other PUFA, whereas MUFA and SFA except margaric acid are showing almost similar range of values. The higher levels of EPA and DHA would also increase the stress tolerance and membrane permeability [35, 36]. The Arachidonic acid is considered to be the precursor for Prostaglandins, which in terms play an important role in the gonad growth and maturation phenomenon. Moreover interaction and intricacies between omega-3, omega-6 and omega-9 fatty acids are more crucial for the maintenance as good health. The omega-3 fatty acids have Anti-inflammatory and Anti-coagulant properties as well as play an important role in health benefits. The omega-6 fatty acids have their role in female reproduction, whereas omega-9 fatty acids help to reduce the rise of Atherosclerosis, cardiovascular disease and stroke. From the results obtained it is very clear that the muscle tissue of penaeid prawns possessing considerable amounts of PUFA hence it can be a good and healthier choice of daily diet. Ash content is one of the least studied biochemical constituents of crustaceans. In the present study the Ash content of muscle tissue of selected penaeid prawns was appears to be around 1.5% body weight. Similar kind of results was also supported in several prawns including penaeids and palemonids [6, 21, 26]. The moisture content of the muscle tissue of penaeid prawns was almost in the range around 75%. The results obtained in the present study are in accordance with previous for different species of prawns.

In the conclusion, the penaeid prawns selected in the present investigation are showing reasonably good proximate composition and sufficient quantities of organic molecules in terms of nutrition. From the data of the present results obtained it clearly indicates that all the penaeid prawns are possessing good protein source with low fat and higher amounts of essential amino acids and PUFA, all these prawns species can be recommended as a healthy and good nutritious food choice for consumption by humans.

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