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Nutritive value of cultured white leg shrimp *Litopenaeus vannamei*

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Biochemical assays play a major role in recent years. The biochemical composition is the yardstick to measure and assess the nutritional quality of food sources. In the present study, protein, carbohydrate, lipid, moisture and ash in *Litopenaeus vannamei* was 35.69, 3.20, 19, 76.2 and 1.2% respectively. Calcium content was maximum (154.5 mg) followed by sodium (67.7 mg) and potassium (56.7 mg). Manganese was reported to be minimum (0.898 mg). Copper and chromium were observed in trace. Totally 18 amino acids were detected, among these, argentine, histamine, isoleucine ,leucine, methionine, phenylalanine, tryptophan, lysine and valine are essential amino acids and alanine, asparagine, aspartic acid, cysteine, glutamic acid, glycine, proline, serine and tyrosine are non-essential amino acids. In individual essential amino acids, valine (23.72%) was maximum, followed by lysine (13.42%) and methionine (13.06%). Histidine was minimum (1.08%). Glycine (9.8%), cystine (5.56%) and proline (4.26%) contributed as major non-essential amino acids. Ten individual fatty acids were identified, which includes both unsaturated and saturated fatty acids. Three saturated fatty acids (SFA) were recorded (Palmitic acid, Margaric acid and Stearic acid). The polyunsaturated fatty acids (PUFA) were the most dominant common fatty acids (38.5%) with the higher levels of linoleic acid (16.3%) and alpha-linolenic acid (11.2%). Oleic acid is the only monounsaturated fatty acid (MUFA) contributed 12.48% of total fatty acids. At the same time the Omega – 6 and omega - 3 fatty acids accounted for 16.3 and 35.4% of the total PUFA (51.7%). the present study confirming that based on the results, *L.vannamei* species can be considered as a good source of fatty acid as well as protein.

Key words: *Litopenaeus vannamei*, proximate composition, fatty acids, amino acids, minerals, shrimp nutrition.

INTRODUCTION

Shrimp is one of the world's most popular shellfish. It provides high quality rich protein, calcium and various extractable compounds and minerals for human body, while low in calorie and fat (Abdullah et al., 2009). Lipid of shrimp contains mostly polyunsaturated fatty acids (essential fatty acids). These essential fatty acids are available in shrimp provides health benefits for human e.g., eye (retina) and brain development and function (Conner et al., 1992).

There are many inorganic elements in the body of shrimp that support associated vital physiological functions. Although shrimps are capable of extracting some of the elements from water, they do respond to dietary sources (Deshimaru and Yone, 1978; Kanazawa

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et al., 1984; Davis et al., 1992. Since these micronutrients are essential, their absence in the diet may lead to deficiency disease. Some elements, such as copper, zinc, manganese, iron and chromium have useful biological function and are found in shrimp at acceptable levels are very useful for human health (Abdullah et al., 2009). In India, there is no published report on biochemical composition of *Litopenaeus vannamei*. Therefore, the present investigation is the first of its kind on proximate composition of basic biochemical constituents, such as total protein, carbohydrate, lipid, amino acids, fatty acids, moisture and ash in the muscle of *L. vannamei*.

MATERIALS AND METHODS

The shrimps (*L. vannamei*) were collected from the Suriya Marine shrimp farm in Bhimavaram, West Godavari district, Andrapradesh, India. Their lengths ranging from 125 to 130 mm. Samples were washed with deionized water to remove any adhering contamination and drained using filter paper. Samples were brought to the laboratory in ice using insulated containers. The shrimp exoskeletons were pealed out and the meat was homogenized. The grounded samples were oven dried at 70°C and ground into a fine powder.

Analysis of proximate composition

The protein, carbohydrate and lipid contents were estimated by adopting the standard methods of (Lowry et al., 1951; Dubois et al., 1956; Folch et al., 1956) respectively. 1 g of powdered tissue in a porcelain crucible was kept in a muffle furnace at 60°C for 4 h. The residue ash content was weighed and the percentage was calculated. Moisture content was estimated by hot air oven method and minerals were analyzed by following the method of A.O.A.C. (1990). Triplicate reading were taken.

Analysis of fatty acids

The samples were oven dried at 70ºC for 24 h until no more weight reduction was observed. After that the samples were grounded with pestle and mortar. To the 100 to 200 mg of finely ground tissue samples 1:1 ratio of chloroform: methanol (2 ml) was added and kept for 30 s. The residual matter was removed by filtering through Whatman No.1filter paper (125 mm). This was washed with 1 ml of chloroform: methanol (2:1vol) to remove the inorganic substances from the combined extract by partition and treated with chloroform: methanol: water (8:4:3) where the lower phase evaporated to dryness. The dried matter was subjected in a sealed test tube with 3% Methanolic HCL at 80ºC for 18 h. To this 2 ml of hexane was added to extract the fatty acid methyl esters (FAME) obtained from methanol phase in Hexane. Top 1 ml of the hexane phase was collected in a micro vial. The residual fraction was dissolved in 10/µl of ethyl acetate and injected 1/µl aliquot into a gas chromatograph equipped with flame identification detector and column (HP ULTRA – 225 m, 0.2 mm ID) by gas chromatograph (Kashiwagi et al., 1997).

Estimation of amino acids

The samples were dried at 60°C for 24 h in an oven and packed in airtight polyethylene covers and kept in desiccators. The oven-dried samples were finely grounded before estimating amino acid profile. Amino acids were estimated by using HPLC – Lachromemerck in SPD- 10A VP Detector.

Table 1. Proximate composition in the flesh of *L. vannamei.*

Parameter	%
Crude protein	35.69 ± 0.5
Crude carbohydrate	3.20 ± 0.3
Crude lipid	19.00± 0.6
Moisture	76.2 0± 0.5
Ash	1.20 ± 0.6

Table 2. Mineral composition in the flesh of *L. vannamei.*

Minerals	mg/g
Calcium (Ca)	154.5
Magnesium (Mg)	13.41
Sodium (Na)	67.7
Potassium (K)	56.7
Phosphorus (P)	6.98
Manganese (Mn)	0.898
Iron (Fe)	4.54
Copper (Cu)	in trace
Chromium (Cr)	in trace

RESULTS

Proximate composition

The percentage of protein, carbohydrate, lipid, moisture and ash in *L. vannamei* was 35.69, 3.20, 19, 76.2 and 1.2% respectively (Table 1).

Mineral composition (mg/g)

The minerals of the *L. vannamei* flesh are shown in Table 2. Calcium content was maximum (154.5 mg) followed by sodium (67.7 mg) and potassium (56.7 mg). Manganese was reported to be minimum (0.898 mg). Copper and chromium were observed in trace amounts. The mineral composition in *L. vannamei* flesh was in the following order; Ca > Na >K>Mg>P>Fe>Mn>Cu, Cr.

Amino acid profile

The amino acid profiles detected from the flesh of the *L. vannamei* is presented in Table 3. Totally 18 amino acids were detected, among these, arginine, histidine, isoleucine, leucine, methionine, phenylalanine, tryptophan,

Table 3. Essential and non essential amino acids in the flesh of *L. vannamei.*

Essential amino acids (EAA)	EAA (%)
Arginine	1.2
Histidine	1.08
Isoleucine	12.3
Leucine	5.63
Lysine	13.42
Methionine	13.06
Phenylalanine	1.27
Tryptophan	1.3
Valine	23.72
Total	72.98
Non essential amino acids (NEAA)	**NEAA (%)**
Alanine	1
Asparagine	0.056
Aspartic acid	1.46
Cystine	5.56
Glutamic acid	2.51
Glycine	9.8
Proline	4.26
Serine	2.66
Tyrosine	2.51
Total	29.816

lysine and valine are essential amino acids and alanine, asparagine, aspartic acid, cysteine, glutamic acid, glycine, proline, serine and tyrosine are non-essential amino acids. In individual essential amino acids, valine (23.72%) was maximum, followed by lysine (13.42%) and methionine (13.06%). Histidine was minimum (1.08%). Glycine (9.8%), cystine (5.56%) and proline (4.26%) contributed as major non-essential amino acids. Asparagine (0.05%) showed the lowest concentration among the non-essential amino acids.

Fatty acid profile

The fatty acid profile from the flesh of the *L. vannamei* (Table 4) show the presence of ten individual fatty acids, which includes both unsaturated and saturated fatty acids. Three saturated fatty acids (SFA) were recorded (Palmitic acid, margaric acid and stearic acid). Among three saturated fatty acids, stearic acid was maximum (12.88%) followed by palmitic and margaric acid. The polyunsaturated fatty acids (PUFA) were the most dominant common fatty acids (38.5%) with the higher levels of linoleic acid (16.3%) and alpha-linolenic acid (11.2%). Oleic acid is the only monounsaturated fatty acid (MUFA) contributed 12.48% of total fatty acids. At the same time the Omega – 6 and omega - 3 fatty acids accounted for 16.3 and 35.4% of the total PUFA (51.7%).

DISCUSSION

Shrimp is considered as a high-range protein containing nutrient like fish, which contain 8 to 20% protein. It has been reported that protein content of shrimp ranged between 17 and 21% depending on shrimp species (Sriket et al., 2007; Yanar and Celik, 2006). According to Sambhu and Jayaprakash (1994), the protein level in *Penaeus indicus* was varied from 44.62 to 80.87%. The high protein content in the lowest size groups may be attributed to increased protein synthesis during the active growth phase as it has been observed elsewhere in shrimps and mantis shrimps (Achuthan Kutty and Parulekar, 1984; Ajith kumar, 1990; Tanuja, 1996; Pedrazzoli et al., 1998).

According to the study of Sriraman (1978), the protein content of crustaceans and mollusks were around 20%. In the present investigation, the protein content of *L. vannamei* showed 35.69%. Carbohydrate content exhibited an inverse relationship with protein content. Similar findings were recorded by Silva and Chamul (2000), Sriraman (1978), Nair and Prabhu (1990), Reddy and Shanbhogue (1994), Ravichandran (2000). In general, lipid act as major food reserves along with protein and subjected to periodic fluctuations influenced by environmental variables like temperature (Johnstene, 1917; Pillay and Nair, 1973). But this does not affect the lipid composition of muscle tissue to any great extent. In this investigation, the proportion of protein was greater followed by lipid and carbohydrate in the muscle of *L. vannamei*. Similar difference has been already reported by (Nargis, 2006).

Moisture of fresh shrimp is generally reported as 75 to 80% (Yanar and Celik, 2006; Sambhu and Jayaprakash, 1994). In the present study, 76.2% moisture was recorded. The ash content of the *L. vannamei* was calculated as 1.2% in the present study. Ash content of shrimp is generally 1 to 1.5%. Gokoglu et al. (2008) and Yanar and Celik (2006) calculated the amount of ash in black tiger and white shrimps were 0.95 and 1.47%, respectively. These values are very close to the findings of the present study

Determination of mineral composition of shrimp is important for both checking raw material quality and labeling requirement in nutritional point of view hence aiding health. The main functions of essential minerals are to maintain colloidal systems and a-acid-base equilibrium. Fish and shellfish contain considerable amounts of minerals such as, calcium, magnesium, phosphorus, potassium and sodium (Attar et al., 1992; Abdullah et al., 2009).

The mineral content, including Ca, Mg, K, P, Mn, Na and Fe in the edible part of *L. vannamei* were investigated in the present study. Calcium levels of

Table 4. Saturated fatty acid and unsaturated fatty acids in the flesh of *L. vannamei.*

Saturated fatty acids	**Position of the carbon atom**	**%**
Palmitic acid	C 16	7.06
Margaric acid	C 17	1.42
Stearic acid	C 18	12.88
	Total	23.36
Monounsaturated fatty acids		
Oleic acid	18:1 (n-9)	12.48
	Total	12.48
Polyunsaturated fatty acids		
Omega-6 fatty acids		
Linoleic Acid	18:2 (n-6)	16.3
Omega-3 fatty acids		
Alpha-linolenic acid (ALA)	18:3 (n-3)	11.2
Stearidonic acid (SDA)	18:4 (n−3)	in trace
Eicosatrienoic acid (ETE)	20:3 (n−3)	4
Eicosapentaenoic acid (EPA)	20:5 (n−3)	9
Docosahexaenoic acid (DHA)	22:6 (n−3)	11.2
	Total	51.7

L. vannamei were found to be 154.5 mg, which is higher (59.5 mg) than green tiger shrimp (Sambhu and Jayaprakash, 1994), sea bass (63.6 mg) and sea bream (19.2 mg) (Erkan and Ozden, 2007). Calcium is essential for hard tissue structure, blood clotting, muscle contraction, nerve transmission and osmoregulation as a cofactor for enzymatic procession. Calcium recommendation is (RDA) 800 mg/day. In addition, many Ca supplements contain lead, which impairs health in numerous ways. Fortunately, Ca interferes with the absorption and action of lead in the body system (Whithney and Rolfes, 2008). Sodium is the principal caution of the extra cellular fluid, aids acid-base balance and is essential for nervous system (Whithney and Rolfes, 2008). The level of Na in flesh of *L. vannamei* was found as 67.7 mg. Potassium assists in maintaining fluid, electrolyte balance and cell integrity. During nerve transmission and muscle contraction, potassium and calcium briefly exchange places across the cell membrane. Potassium requirement for human is about 2 g day^{-1}. The average K contents of *L. vannamei* were found to be 56.7 mg, which is lesser compared to that reported by Sambhu and Jayaprakash (1994) for green tiger shrimp and Erkan and Ozden (2007) for sea bass and sea bream.

Magnesium content of *L. vannamei* was about 13.41 mg. Magnesium has some useful roles in the body. Magnesium is required for the body's enzyme system, bone health; it is a major part of protein synthesis in soft tissues and energy metabolism (Whithney and Rolfes, 2008). Iron is an essential trace element since. It serves as a carrier of oxygen to tissues from the lungs by red blood cell. Exceeding level of 10 mg kg^{-1} for iron is not permitted (ITS, 2000). Iron content of shrimps used for this study was 4.54 mg. However, the value stated for *L. vannamei* was higher than those stated by ITS (2000). Compared to green tiger shrimp (1.48 mg), speckled shrimp (1.55 mg) (Gokoglu et al., 2008, Sambhu and Jayaprakash 1994) and *Aristeus antennatus* (0.9 mg) (Karakoltsidis et al., 1995). Amino acids are the building blocks of proteins (Babsky et al., 1989). Crustacean muscles contain high concentration of free amino acids, such as arginine, glycine, proline, glutamine and alanine (Cobb et al., 1975), and support osmoregulatory functions (Fang et al., 1992). The amino acid, tryptophan plays an important role in the brain as a precursor of the neurotransmitter: Serotonin, which has a major effect on the feeding behavior of animals (Mullen and Martin, 1992). Valine is involved in many metabolic pathways, protein synthesis and optimal growth (Wilson, 2002). Histidine is also involved in many metabolic functions including the production of histamines, which take part in allergic and inflammatory reactions. It aids osmoregulation and metabolic pathways during certain emergencies/ harsh conditions (Abe and Ohmama, 1987).

In the present study among saturated fatty acids stearic acid was dominant that followed by palmitic and margaric acids. The amount of palmitic acid in *L. vannamei* was 7.06%. However, Yanar and Celik (2006) reported 22.2% in black tiger shrimp and 21.8% in white tiger shrimp respectively. Sargent et al. (1999) showed reports of 17.3% in golden shrimp, 18.0% in pink shrimp and 17.6% in Norway shrimp. It has been reported that incorporation of essential fatty acids in the diet produced better growth rate and survival in aquaculture. Bell and Sargent (2003), Osborn and Akoh (2002) and Watanabe et al. (1989) mentioned in their review article that n-9 fatty acids, found as oleic acids (C18:1 n-9) plays a moderate role in the body. Moreover, n-6 fatty acids cannot be synthesized by humans and are therefore considered as essential fatty acids.

In the present study, the presence of n-3 PUFA, particularly, linoleic, EPA and DHA indicates better growth and survival of *L. vannamei* in the culture pond. The higher levels of EPA and DHA would increase stress tolerance and membrane permeability (Watanbe, 1993; Lin et al., 2003). Findings of C18:1 n-9 of *L. vannamei* species in this study were similar to previously reported findings of white and black tiger shrimps (Yanar and Celik, 2006). In addition, DHA and EPA, belonging to n-3 fatty acids family, are considered as essential (Feliz et al., 2002). DHA and EPA, two of the major PUFA were found as 11.2 and 9% in *L. vannamei*. Results of this present study also showed that the value of EPA was lower than that of DHA in *L.vannamei* although Ackman (1989) reported that, the shellfish tend to have EPA greater than DHA. This previous findings disagrees with the present findings of *L. vannamei*. Furthermore, n-3 fatty acids are essential in growth and development throughout the human life cycle and should be included in the diet.

The n-3 fatty acids have anti-inflammatory and anti-coagulant properties as well as many other important health .The ω-9 fatty acids help to reduce the risk of arteriosclerosis, cardiovascular disease and stroke (Since *L. vannamei* contains considerable amounts of PUFA). Based on the results, *L. vannamei* species can be considered as a good source of fatty acid as well as protein.

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