Proximate analysis and amino acid profile in fresh meat, meat meal, and shell meal of bamboo clam *Solen* sp. from Kwanyar Coast, Bangkalan-Madura, Indonesia

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Abstract. This study aims to know the proximate and amino acid content in fresh meat, meat meal, and shell meal of bamboo clam *Solen* sp. collected from Kwanyar Coast, Bangkalan-Madura, Indonesia. The chemical composition was examined using proximate test and amino acid determination used Ultra Performance Liquid Chromatography (UPLC). Results showed that the fresh meat of *Solen* sp. contained 74.52% protein, 3.09% fat, 1.84% carbohydrate, 8.21% water, and 12.28% ash. The meat meal held 76.09% protein, 5.62% fat, 4.81% carbohydrate, 9.49% water, and 3.99% ash. The shell meal contained 2.92% protein, 0.32% fat, 1.17% carbohydrate, 0.57% water, and 95.04% ash. The amino acids consisted of 10 essential amino acids and 8 non-essential amino acids. Arginine is the highest content of the essential amino acid, 1.03% in fresh meat, 6.59% in meat meal, and 1.14% in shell meal. In non essential amino acid, glutamic acid was the highest, 1.24% in fresh meat, 10.52% in meat meal, and 0.12% in shell meal, respectively.

Key Words: proximate, bamboo clam, *Solen* sp., protein, fat, carbohydrate.

Introduction Marine and coastal areas are rich in biodiversity and have been utilized as food source for animal protein. One of these marine animals is mollusks. Recently, the utilization of mollusks as food source taken in fresh form or processed products has highly developed. Several mollusks, particularly bivalves whose nutritional values are studied, are adalah *Anadara antiquata* (Abdullah et al 2013), knife clam *Solen* sp. (Nurjanah et al 2008, 2013), tofu clam *Meretrix meretrix*, and snow clam *Pholas dactylus* (Abdullah et al 2017), *Unio terminalis* and *Potamida littoralis* (Ersoy & Şereflişan 2010), *Solen marginatus* (Prato et al 2019), *Chamelea gallina* (Orban et al 2007), *Ostrea edulis, Mytilus galloprovincialis, Tapes decussatus, Ruditapes philippinarum*, and *Rapana venosa* (Celik et al 2014).

Mollusks contain high quality protein with essential amino acid content for human body maintenance and growth. King et al (1990) stated that mollusks are categorized as low fat and high protein food that can be included in low fat diet. Protein will influence the food texture and taste. Its quality is usually assessed from amino acid composition, so that amino acid composition helps determine the nutritional value of an organism (Suprayitno & Sulistiyati 2017). The biochemical composition of shells has been widely studied by many researchers worldwide (Bongiorno et al 2015).

Studies on bamboo clam (*Solen* sp.) in Indonesia have focused on environmental condition and exploitation (Trisyani & Irawan 2008; Nurjanah et al 2008; Trisyani 2018), population dynamics (Trisyani et al 2016), genetic characteristics (Trisyani & Rahayu 2020), and metal concentration (Trisyani 2020), but information on nutritional content is still limited. Protein content of bamboo clam is potential as animal protein source alternative. Animal protein has higher biological value than plant protein, since animal protein has more complete amino acid composition and content (Suprayitno & Sulistiyati

2017; Nurjanah et al 2013). Bamboo clam can be consumed in fresh form or processed products. The clam shell can also be used as a mixture to make cookies, animal feed, and cement (Agustini et al 2011; Alfred 2015).

The study was carried out to analyze and to determine proximate composition and amino acid profile of bamboo clam (*Solen* sp.) from Kwanyar Coast, Bangkalan, Indonesia, in the forms of fresh meat, meat meal, and shell meal.

Material and Method. Sample collection was conducted in February to March 2020. Bamboo clams were collected by fishermen of Kwanyar Coast, Bangkalan Regency at the geographic position of 7°10'2.00"S and 112°52'12.00"E (Figure 1). Samples were kept in a cool box and taken to the laboratory. Handling of fresh samples was done by cleansing and removing the meat for immediate analysis. To obtain the meat meal, the meat was boiled for 30 sec and dried in an oven at 50^{o}C for 5 h. The samples were then sieved through 80 µm mesh size to get fine particles of meat meal. Shell meal was obtained through shell drying in the oven at 50^{o}C for 3 h, blended, and sieved through 80 µm sieve.

Figure 1. *Solen* sp. sampling location in Kwanyar Coast, Bangkalan, Indonesia.

Proximate analysis. Proximate analyses were carried out to know the protein, fat, carbohydrate, water, and ash content. The test samples were fresh meat, the meat meal, and the shell meal of bamboo clams. Water content was measured by drying the sample at 105^{o}C for 24 h following the protocol of Association of Officiating Analytical Chemists (AOAC 2005). Ash level was analyzed by drying the sample at 6000^{o}C for 6 h (AOAC 2005). Fat content analysis used Soxhlet method through sample extraction for 4-6 h, then heated in an oven at 60^{o}C for 24 h, whereas protein content was obtained through destruction, distillation, and titration method (AOAC 2005). Total carbohydrate level was tested using by difference method (AOAC 2005).

Amino acid analysis. Amino acid composition in fresh meat, meat meal, and shell meal of *Solen* sp. was analyzed in the Laboratory of Saraswanti Indo Genetech, Bogor, using Ultra Performance Liquid Chromatography (UPLC) Guide, internal method of Laboratory of Saraswanti Indo Genetech, Bogor. Preparation of sample and standard solution was conducted according to literature methods (Waters 2012). Determination of amino acid content was done by using UPLC condition as follow: Column (AccQ Taq Ultra C18 1.7 µm (2.1 x 100 mm)); flow rate (0.7 mL min^{-1}); temperature (49^{o}C); detector (FDA, wave length 260 nm); injection volume (1 µL) and mobile phase (mobile phase A = Eluent A

Concentrate AccQ Taq Ultra from Waters (Part No. 186003838), mobile phase B = 10% mobile phase D, mobile phase C = Aquabidest and mobile phase D = Eluent B AccQ Taq Ultra from Waters (Part No. 1860005859). Sample preparation: about 0.1 g sample was added to 5-10 mL 6N HCl and then hydrolysed for 22 hours at 110°C. The hydrolysed mixture was cooled and transferred into volumetric flask 500 mL diluted with distilled water. The solution was filtered through a 0.45 µm filter. About 500 µL of filtrate was added 40 µL AABA and 460 µL Aquabidest. About 10 µL of solution was added to 70 µL AccQ Fluor Borate and 20 µL reagent fluor A and then injected into UPLC system. Preparation of standard solution was performed as follow: About 40 µL internal standard AABA and 920 µL Aquabidest were added and then homogenized. About 10 µL of standard solution, 70 µL AccQ Fluor Borate and 20 µL of reagent fluor A were added and then vortexed for one min. The solution was incubated for 10 min at 55°C and then injected into UPLC system.

Results and Discussion

Proximate analysis. Protein, fat, and carbohydrate concentrations, water content, and ash in fresh meat, meat meal, and shell meal of *Solen* sp. based on the dry weight are presented in Table 1.

Table 1

Proximate composition of fresh meat, meat meal, shell meal of bamboo clam *Solen* sp. from Kwanyar Coast, Bangkalan, Indonesia

No	*Parameter*	*Fresh meat*	*Meat meal*	*Shell meal*
1	Protein (%)	74.52±2.10	76.09±1.55	2.92±0.03
2	Fat (%)	3.09±0.04	5.62±1.32	0.32±0.00
3	Carbohydrate (%)	1.84±0.06	4.81±0.40	1.17±0.50
4	Water (%)	8.21±0.24	9.49±2.99	0.57±0.01
5	Ash (%)	12.28±1.58	3.99±0.25	95.04±0.46

Notes: mean value ± standard deviation of 4 measurements (n = 4).

Mean protein content in the fresh meat of *Solen* sp. was 74.52% (based on dry weight) or 13.11% (based on wet weight). It is higher than that of *Solen* sp. from Pamekasan, 9.79% (Nurjanah et al 2008) and *Solen marginatus* from Italy, 10.96% (Prato et al 2019), but lower than that of *Solen* sp. from Cirebon, 14.48% (Nurjanah et al 2013). The meat meal of *Solen* sp. from Bangkalan contained mean protein of 76%, while the shell meal held mean protein of 2.9%. The highest fat and water content were recorded in the meat meal of *Solen* sp. and the lowest in the shell meal. In contrast, the highest ash content was found in shell meal and the lowest in the fresh meat (Table 1). The highest carbohydrate was found in the meat meal and the lowest in the shell meal. Drying the fresh meat of *Solen* sp. could increase the nutritional content and the durability. High protein content in the meat meal can be benefitted as alternative processed product using the clam meat meal as basic material. Protein in the shell meal was relatively low so that the clam shell is utilized for biodiesel production (Abdul 2014), mix for concrete material (Alfred 2015), and cookies (Agustini et al 2011).

The proximate composition in *Solen* genus varied between species. It could result from difference in season, individual size, maturity, food availability, environmental condition (Orban et al 2007; Celik et al 2014; Koral & Süleyman 2016). Biochemical composition of meat quality changes with environmental condition and reproductive period (Bakan & Büyükgüngör 2000). The biochemical composition, such as protein, carbohydrate, and fat, is very important for body maintenance and growth. Celik et al (2014) reported that increased protein, fat, and carbohydrate of mollusks in Turkey occur during gametogenesis and egg development, in spring and summer, as more food is available, and then these decline after spawning. Increase in protein, fat, and carbohydrate levels is highly needed because marine organisms require more energy in reproductive process. In the present study, sample collection was done in February to March, 2020, the first transitional season, as early larval development after the spawning

(Trisyani et al 2019). Mean size of *Solen* sp. in Indonesia also shows high variations. *Solen* sp. has mean shell length of 5.1±0.48 cm in Cirebon, 4.3±0.85 cm in Bangalan, 2.8±0.41 cm in Pamekasan (Trisyani 2018), and variations in protein level are directly proportional to the clam body size, in which the bigger the clam is, the higher the protein level will be.

Carbohydrate is the main energy source in all human food. Carbohydrate level is less than other nutrients, such as protein and fat in animal tissue, particularly aquatic organisms (Babu et al 2010). Carbohydrates in the tissue are stored as glycogen, free sugar, and protein-bound sugars, that function as energy reserves for various metabolism processes. In mollusks, the carbohydrate reserves can generally be used in unfavorable conditions, and high variations in the tissue indicate that the carbohydrate reserves transferred can widely and fast fluctuate as response to the fluctuative conditions (Nagabhushanam & Mane 1978). Present study found 1.84% carbohydrate in the fresh meat of *Solen* sp., 4.8% in the meat meal, and 1.17% in the shell meal, while *Solen* sp. from Cirebon contains 3.68% carbohydrate (Nurjanah et al 2013), and that from Pamekasan holds 4.95% carbohydrate (Nurjanah et al 2008).

Fat is very efficient as energy source and contains twice the energy of carbohydrate and protein (Nelson & Cox 2004). Fat content in the fresh meat of *Solen* sp. from Bangkalan was 3.09%, and it is higher than that from Pamekasan, 0.32% (Nurjanah et al 2008), and from Cirebon, 1.72% (Nurjanah et al 2013). Fat content in the meat meal of *Solen* sp. from Bangkalan was 5.62% and in the shell meal was 0.32%.

Amino acid composition. Table 2 demonstrates percent of essential and non essential amino acids in the fresh meat, the meat meal, and the shell meal of *Solen* sp. Total essential amino acid was 5.15% in fresh meat, 38.49% in meat meal, and 2.11% in shell meal, respectively, whereas total non essential amino acid was 4.97% in fresh meat, 38.9% in meat meal, and 3.98% in shell meal. Total amino acid was 10.12% in fresh meat, 76.98% in meat meal, and 6.09% in shell meal.

Table 2

Amino acid composition (%) in fresh meat, meat meal, and shell meal of *Solen* sp. from Kwanyar Coast, Bangkalan, Indonesia

EAA	*Fresh meat*	*Meat meal*	*Shell meal*
Histidine	0.32±0.004	2.16±0.010	0.06±0.000
Threonine	0.62±0.002	4.65±0.006	0.07±0.002
Arginine	1.03±0.003	6.59±0.008	1.14±0.002
Methionine	0.02±0.000	1.06±0.001	0.08±0.000
Valine	0.48±0.002	3.59±0.002	0.16±0.002
Phenylalanine	0.64±0.001	4.64±0.020	0.34±0.002
Isoleucine	0.45±0.001	3.70±0.003	0.09±0.001
Leucine	0.79±0.003	6.48±0.013	0.07±0.001
Lisyne	0.50±0.002	5.02±0.02	0.08±0.001
Tryptophan	0.09±0.000	0.60±0.003	0.02±0.000
Sub total	5.15	38.49	2.11
NEAA			
Aspartic	0.72±0.002	6.40±0.006	0.12±0.002
Glutamic	1.24±0.005	10.52±0.013	0.12±0.003
Serine	0.65±0.063	4.17±0.015	0.12±0.002
Glycine	0.95±0.002	4.48±0.004	0.79±0.001
Alanine	0.51±0.003	3.94±0.001	0.07±0.001
Tyrosine	0.49±0.001	3.68±0.009	0.19±0.001
Proline	0.37±0.001	2.62±0.100	1.15±0.003
Cysteine	0.04±0.000	2.68±0.002	1.42±0.000
Sub Total	4.97	38.49	3.98
Total	10.12	76.98	6.09

Notes: EAA – essential amino acid; NEAA – non essential amino acid (n = 4).

The highest concentration of essential amino acid in all *Solen* sp. sample from Bangkalan occurred in arginine, 1.03% in fresh meat, 6.59% in meat meal, and 1.14% in shell meal, respectively. The lowest was recorded in methionine, 0.02% in fresh meat, and tryptophan, 0.6% in meat meal and 0.02% in shell meal. In non essential amino acid, glutamatic acid had the highest content, 1.24% in fresh meat and 10.52% in meat meal, respectively. Nevertheless, all amino acid concentrations found in this study are higher than those recorded by Nurjanah et al (2008) in samples from Pamekasan.

Shells contribute high quality protein and essential amino acids for human body maintenance and growth (Ersoy & Sereflisan 2010). Essential amino acids are highly needed for nutrition, normal growth maintenance of nitrogen equilibrium, while non essential amino acids are physiologically important and take part in general metabolismic reactions (Jayaprabha 2016). Essential amino acids are not synthetized in the body and have to be obtained from food intake. Without these amino acids, the tissues will be inhibited to grow, recover, or maintain (Hoffman & Falvo 2004). Nevertheless, amino acid content in marine organisms depends on species, size, season, and geographic condition (Wesselinova 2000).

Amino acids are protein building components. Each amino acid binds through covalent bond. Protein can be broken down to simpler units, amino acids, through hydrolisis (Nelson & Cox 2004). Protein in *Solen* sp. from Bangkalan had complete amino acids, 10 essential and 10 non essential amino acids. The present study indicated that there were different types and levels of amino acids in fresh meat, meat meal, and shell meal. Processing could affect the chemical components of the product by increasing or reducing the chemical composition. Akintola et al (2013) stated that drying can increase arginine and histidine levels.

Glutamic acid in fisheries product is the mostly found amino acid and can add the flavor to the food. The hydrogen group of glutamic acid can be substituted with sodium to form monosodium glutamate that has high savory taste intensity so that it is commonly used as flavor enhancer. According to Oladapo et al (1984), glutamic acid and aspartic acid are important to typical aroma and taste of food. Glutamic acid can be produced by human body. Glutamic acid has glutamate ion that can stimulate several nerves in human tongue. This characteristic could be used by the flavoring industry. Glutamic acid is amino acid found in several fisheries products, such as peptone of fish intestines (Nurhayati et al 2013), and the highest non essential amino acid in *Solen* sp. from Pamekasan (Nurjanah et al 2008).

Arginine is an amino acid formed in the liver and some of it in the kidney. According to Emmanuel et al (2008), arginine is very important for children. It functions also to increase the release of growth hormone and raise the fertility of man. Glutamic acid is beneficial for withholding excessive alcohol consumption, accelerating intestinal wound healing, increasing mental health, and holding depression. Aspartic acid functions in urea biosynthesis and as gluconic and pyrimidin precursor. It is also good for chronic fatique handling and energy development. Proline is amino acid whose R group is nonpolar and hydrophobic. Proline has free amino group and forms an aromatic structure. This amino acid can be obtained from casein hydrolysis (Suprayitno & Sulistiyati 2017). According to Erkan et al (2010), histidine functions in body tissue growth and repairs and produces erythrocytes.

Amino acid is also needed by microorganisms to meet the need for nitrogen. This need is very high in the exponential phase so that reduction of several amino acid components in the media will be significant. According to Selvarasu et al (2008), serine, aspartate, and glutamate are amino acids highly required by bacteria. Amino acid in the bacterial growth during the lag phase did not change, but when it enters exponential phase, amino acid levels reduced drastically. It indicates that bacteria use amino acid in their growth.

Conclusions. Bamboo clam (*Solen* sp.) from Kwanyar Coast, Bangkalan, had the highest protein content in the meat meal, 76.09%, followed by the fresh meat, 74.52 %, the lowest in the shell meal, 2.92%. *Solen* sp. was found containing 18 types of amino acid in which six of them were essential amino acid while the other eight were non-essential amino acid. The highest content was arginine in the essential amino acid category and glutamic acid in the non-essential category.

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