NUTRITIONAL VALUE IN FRESH AND SMOKED *CORBICULA FLUMINEA* (ETAK) TISSUE VIA TRADITIONAL SMOKING PROCESS

by

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DECLARATION

I declare that this thesis entitled "Nutritional Value in Fresh and Smoked *Corbicula Fluminea* (Etak) Tissue via Traditional Smoking Process" is the result of my own research except as cited in the references. The thesis has not been accepted for any degree and is not concurrently submitted in candidature of any other degree.

Signature: _____

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Date : _____

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Nutritional Value in Fresh and Smoked *Corbicula fluminea* (Etak) Tissue via Traditional Smoking Process

ABSTRACT

Corbicula fluminea is a clam that is widely consumed by the society and it is a popular snack among Malaysians. In Kelantan, *C. fluminea* is famously known as etak. Currently, there is no study yet conducted to determine the nutritional value of *C. fluminea*. Thus, the purpose of this study is to determine and compare the nutritional value of fresh and smoked *C. fluminea* from six different stalls which include moisture content, ash, fat, protein and carbohydrate. In this study, Association of Official Analytical Chemicals (AOAC) method was used to analyse the nutritional value content. The result shows that the content of moisture was found the highest in fresh *C. fluminea* ($p < 0.05$) from stall 2 and stall 3 ($p < 0.05$) with the value of 81.09 and 80.86% respectively while ash content was found the highest in smoked *C. fluminea* ($p < 0.05$) that came from stall 3 and stall 6 ($p < 0.05$) with the value of 1.96% and 1.81% respectively. Fat content was recorded as 11.66% which means the highest in fresh *C. fluminea* from stall 1 ($p < 0.05$). *C. fluminea* is a type of bivalve that is known to have high protein content. This study recorded that smoked *C. fluminea* has higher protein content than fresh *C. fluminea* and was found the highest in smoked *C. fluminea* from stall 6 with 10.92%. Carbohydrate was found the highest in smoked *C. fluminea* from stall 5 with the value of 1.91%.

Nilai Nutrisi Dalam Tisu *Corbicula fluminea* Segar dan Salai (Etak) Melalui Proses Salai Tradisional

ABSTRAK

Corbicula fluminea merupakan kerang yang dimakan secara meluas oleh masyarakat dan ia adalah makanan ringan popular di kalangan rakyat Malaysia. Di Kelantan, *C fluminea* lebih dikenali sebagai etak. Pada masa ini, tiada kajian yang dijalankan untuk menentukan nilai nutrisi yang terkandung di dalam *C. fluminea.* Oleh itu, tujuan kajian ini dijalankan adalah untuk menentukan dan membandingkan nilai nutrisi di dalam *C. fluminea* yang segar dan juga *C. fluminea* yang disalai yang dibeli dari enam gerai yang berbeza. Nilai nutrisi yang dikaji termasuk kandungan kelembapan, abu, lemak, protein dan karbohidrat. Kaedah AOAC telah digunakan di dalam kajian ini untuk menganalisis kandungan nilai nutrisi. Hasil kajian menunjukkan bahawa kandungan kelembapan didapati paling tinggi dalam *C. fluminea* segar ($p < 0.05$) dari gerai 2 dan gerai 3 ($p < 0.05$) dengan masing-masing bernilai 81.09% dan 80.86% manakala kandungan abu didapati paling tinggi dalam *C. fluminea* yang disalai ($p < 0.05$) dari gerai 3 dan gerai 6 ($p < 0.05$) dengan nilai 1.91% dan 1.86% . Kandungan lemak direkodkan dengan nilai 11.66% yang bermaksud kandungan paling tinggi dalam *C. fluminea* segar dari gerai 1 ($p < 0.05$). *C. fluminea* adalah sejenis bivalvia yang diketahui mempunyai kandungan protein yang tinggi. Kajian ini mencatatkan bahawa *C. fluminea* yang disalai mempunyai kandungan protein yang lebih tinggi daripada *C. fluminea* segar dan didapati paling tinggi kandungannya dalam *C. fluminea* yang disalai dari gerai 6 dengan nilai 10.92%. Karbohidrat juga didapati paling tinggi dalam *C. fluminea* yang disalai dari gerai 5 dengan nilai 1.91%.

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LIST OF ABBREVIATIONS

AOAC	-	Association of Analytical Chemicals
UMK	-	Universiti Malaysia Kelantan
ANOVA	-	Analysis of Variance

LIST OF SYMBOLS

%	-	Percentage
℃	-	Degree celcius
g	-	Gram
mL	-	Millilitre
ppt	-	Part per trillion
cm	-	Centimetre
N	-	Nitrogen
K_2SO_4	-	Potassium sulphate
$CuSO_4$	-	Copper sulphate
TiO_4	-	Titanate
NH_3	-	Ammonia
H_2SO_4	-	Sulphurci acid
NaOH	-	Sodium hydroxide
H_3BO_3	-	Boric acid
HCl	-	Hydrochloric acid

CHAPTER 1

INTRODUCTION

1.1 Background of Study

Corbicula fluminea (Figure 1.1) or Asian clam (Kramer-Wilt, 2008) is one of the filter feeder bivalves. Scientific classification of *C. fluminea* or Asian clam is illustrated in Table 1.1 (Manan, 2014):

Table 1.1: Scientific classification of *Corbicula fluminea*

Kingdom	Animalia
Phylum	Mollusca
Class	Bivalvia
Subclass	Heterodonta
Order	Veneroida
Suborder	Sphaeriacea
Superfamily	Corbiculoidea
Family	Corbiculidea
Genus	Corbicula
Species	*Corbicula fluminea*

Figure 1.1: Live *C. fluminea* or known as Asian clam

Other than known as Asian clam, it is also commonly called as "golden clam". *C. fluminea* is a type of freshwater bivalve mollusc that is also known to be occurring in brackish water. *C. fluminea* is a clam that comes in yellow-brown, light brown to black in colour. It comes in the size that is less than 1.5 inches but can reach 2.25 inches in term of its length. *C. fluminea* is just a small bivalve that has shape of oval triangular (Kramer-Wilt, 2008).

In Kelantan, *C. fluminea* is famously known as 'etak' and it was a popular snack that is widely consumed by the society in Malaysia (Lee et al., 2013). The smoked *C. fluminea* can easily be found and bought from the stalls at the road side. Anyone who visits the markets in the main city of Kelantan will not miss the sight of sellers of 'etak'.

Besides, *C. fluminea* is served as various kind of dishes. They are either eaten as a snack or breakfast with other dishes such as 'nasi kerabu' and so on. They also can be a taste enhancer due to its sweet and salty taste.

C.fluminea is an ecologically and economically important freshwater shellfish for South-East Asian especially in Malaysia and Thailand. C. fluminea also is one of the most harvested freshwater shellfish in certain states like Kelantan, Terengganu and even Pahang for consumption purpose. Although Asian clam is least known or neglected among Malaysian, but the demand for Asian clam is growing among Malaysia markets.

Traditionally, the business of *C. fluminea* has been practiced in a small-scale basis enterprise in Kelantan. Most of *C. fluminea* that are been sold at the stalls in Kelantan were imported from certain states such as Pahang, Terengganu and even Thailand. This is due to the mining project that been conducted along the main river in Kelantan.

According to Manan (2014), fresh *C. fluminea* has been imported by vendors as much as two tonnes with an income of approximately RM 2,400 per vendors per month. *C. fluminea* are been sold at the price of RM 1.20 per kg for the fresh one and RM 2.40 per kg for the smoked one. This means that the vendors that have been involved with importing the Asian clams have great income and get a good profit for being in the business (Manan, 2014).

1.1 **Problem Statement**

C. fluminea has been known as popular snack and is widely consumed among people in Kelantan. It has been served as an appetizer because of its light and delicious taste. However, Kelantan people keep consuming it without actually knows the content of nutritional values of the clams has. Even though the awareness among community has increased regarding this matter, there is no study yet been carried out

to analyse the nutritional value of *C. fluminea*. Thus, it is necessary to carry out this study to determine its nutritional value content.

1.3 Objectives

The objectives of carrying out the study are:

1. To determine nutritional value in fresh *C. fluminea* tissue.
2. To determine nutritional value in smoked *C. fluminea* tissue.
3. To compare nutritional value in fresh and smoked *C. fluminea* tissue via traditional smoking process.

1.4 Scope of Study

This study was performed to determine the nutritional value that contained in fresh and smoked *C. fluminea* tissue. Three components of nutritional value which are fat, protein and carbohydrate were analysed using Association of Official Analytical Chemicals (AOAC) method. The *C. fluminea* samples were collected at Tumpat and Pasir Mas. The samples were collected from this two district areas as Tumpat and Pasir Mas are the main location of collection of *etak* sources from all over the state of Kelantan.

1.5 Significant of Study

The significance of carrying out this study is that the results will contribute to useful and relevant information to the people of Kelantan regarding the content of

nutritional value contained in fresh and smoked *C. fluminea* tissue. Since there is not yet a published study that carry out the nutritional value analysis of *C. fluminea*, this study will help to provide information on how much content of nutritional value the Asian clams has and which method that is suitable to determine the nutritional value of *C. fluminea.*

CHAPTER 2

LITERATURE REVIEW

2.1 Ecology and History of *C. fluminea*

According to Kramer-Wilt (2008), the Asian clam is commonly occur in freshwater but it also can occur in brackish water as well. *C. fluminea* can be easily found in all sizes of estuarine, lake and river habitats. They have the ability to withstand in a multitude of substrates that includes sand, silt and clay. However, in preference, it will choose fine, clean sand, clay and coarse sand substrates (Kramer-Wilt, 2008). *C. fluminea* need well-oxygenated water and temperature ranging between 2℃ to 36℃. Although they can tolerate this temperature range, in order to reproduce it requires temperature above 16℃ (Kramer-Wilt, 2008).

C. fluminea are filter feeders. It has the ability to clear away the particles from the water column. It primarily feed on phytoplankton and particulate organic matter (POM) (Kramer-Wilt, 2008). Previous studies showed that *C. fluminea* is an extremely efficient filter feeder. The study showed that the filtration and assimilation rates of *C. fluminea* are higher than other freshwater clams.

Besides, *C. fluminea* is also be able to deposit feeding where they carry sediment across its labial palps by ciliary tracts on its foot (Kramer-Wilt, 2008). A study figured that *C. fluminea* is a filter feeder and it adopts deposit feeding to

complement its diet when there is a lack of food resources or the size of certain particles limits its efficiency of filter feeding. The existence of *C. fluminea* is usually at the surface with the distance of not more than 10 cm below the surface. These Asian clam or *C. fluminea* can be found in densities as great as 20,000 per meter squared (Kramer-Wilt, 2008)

According to Kramer-Wilt (2008), it has been recorded that the first batch of *C. fluminea* that was found in US found in 1938 in Washington State. It was found along the shores of the Columbia River near the town. The original vector of the invasion is yet unknown however many researcher believe that it has been introduced by Chinese immigrants that use them as a food soure (Kramer-Wilt, 2008).

2.2 Preparation of Traditional Process Smoked Asian Clam, *C. fluminea*

According to the traditional way of smoking process of 'etak', the preparation of the process is very easy. Firstly, the clam were washed in clean water in order to let the the clam dispose out the mud and sand particles that cover its body. To get a clean clam, it is necessary to soak the clams in clean water for at least three hours. However, it will be best to soak the clams overnight in order to get a cleaner clam. After that, the clams were marinated with a paste that was made of salt, lemon grass, shallots, ginger and garlic that has been blended for at least fifteen minutes. After marinating it with the paste, the clams are collected and cooked. The clams are cooked by roasting it over a small fire.

Roasting process takes place on a platform that is raised for about 2.5 feet above the floor. Long split bamboo pieces are used to build the platform. In order to prevent the clams from slipping through the platform, the bamboo pieces are built

closely together with very little space. During the roasting process, the clams are frequently and constantly swayed over a slow fire. A plank that is attached to a long wooden handle is used to turn the clam as shown in Figure 2.1. It is necessary to constantly swaying the clams to make sure that exposure of each clam to the heat below the platform is even. Roasting process usually takes time about ten to fifteen minutes before it is ready to be eaten (Manan, 2014).

Figure 2.1: The traditional process of smoking etok

2.3 Moisture Content

Analysing moisture content of food is one of the most crucial and essential analytical procedures that can be conducted on food products. However, this analytical procedure can also be the hardes analyses to achieve accurate and precise data. The ease to remove water from food products depends on how the water exist in the food product itself. Water in food products exists in three states which are free water, adsorbed water and water of hydration (Bradley, 2010).

The state of free water maintain its physicsal properties and it acts as the dispersing agent for colloids and the solvent for salts. For adsorbed water, the water particles is held tightly or is occluded in cell walls or protoplasm and is held tightly to proteins. Thus, to choose which method to be used in determining moisture content depends on which form the water exists in the food products (Bradley, 2010).

Moisture content can be done by drying in air-circulation oven. According to this method, the sample is dried under certain conditions and the loss of weight of the sample is used to calculate the moisture content. The moisture content of food products can be depends on the type of oven used, conditions of the oven itself and also the time and temeprature recorded for drying process. This is a simple method and many ovens allow for simultaneous analysis of large numbers of samples. The time taken for drying process might be in the range of few minutes to over 24 hours (Bradley, 2010).

2.4 Ash Content

Ash indicates to the excess inorganic that are left after refers to the inorganic residue remaining after either flaming or complete oxidation of organic matter in a foodstuff. There are two type of ashing that are commonly used which are dry ashing and wet ashing. Dry ashing is mainly used for proximate composition and for analysing some types of specific mineral whereas wet ashing is used as a preparation to analyse certain minerals (Marshall, 2010).

2.5 Association of Official Analytical Chemicals (AOAC) Method

Main objectives of AOAC method are to achieve, enhance, establish an exact and detailed method to analyse food, drugs, feeds, fertilizers, pesticides, water or any other substances that can disturb the performance of public health and safety (AOAC, 1990).

2.6 Protein Analysis

Process of analysing protein content in food is done by Kjeldahl method. Kjeldahl method was found by a brewer named Johann Kjeldahl in 1883. In Kjeldahl method, food will be digested with a strong acid and releases nitrogen that can be determined by an appropriate titration technique. The existence of protein can be determined from the concentration of nitrogen in the food products. It is a standard method used to determine protein concentration because the content of protein is not determined directly. Instead, it needs a conversion factor to convert the concentration of nitrogen to a concentration of protein. A conversion factor, which is 6.25 that is equal to 0.16g nitrogen per gram of protein, is required to convert the measured nitrogen concentration to a protein concentration.

CHAPTER 3

MATERIALS AND METHODS

3.1 Materials

3.1.1 Chemicals and reagents

The chemicals used in this study were petroleum ether which was used as a solvent for fat extraction for fat analysis. For protein analysis, sulphuric acid (H_2SO_4), sodium hydroxide (NaOH), boric acid ($H_3BO_{3)}$, and 0.1M hydrochloric acid (HCl) were used.

3.1.2 Apparatus

Aluminium foil, thimble, spatula, crucibles, weighing scale, air-circulation oven, desiccator, measuring cylinder, dropper, beakers, condenser, soxhlet extractor, round bottom flasks, magnetic stirrer, hot plate and conical flasks were used. A blender was used to grind the dried samples.

3.2 Sampling and Sample Preparation

The fresh and smoked Asian Clam, *C. fluminea,* was purchased from three different stalls at Tumpat and three different stalls at Pasir Mas. The clam was kept

in the clean plastic bag and ice box. After that, the clams were brought to the laboratory at Universiti Malaysia Kelantan (UMK) and were kept in a freezer at -40°C until the analysis process of nutritional value. Figure 3.1 shows the location of study area at Pasir Mas and Tumpat and the coordinate of the location of the stall was presented in Table 3.1.

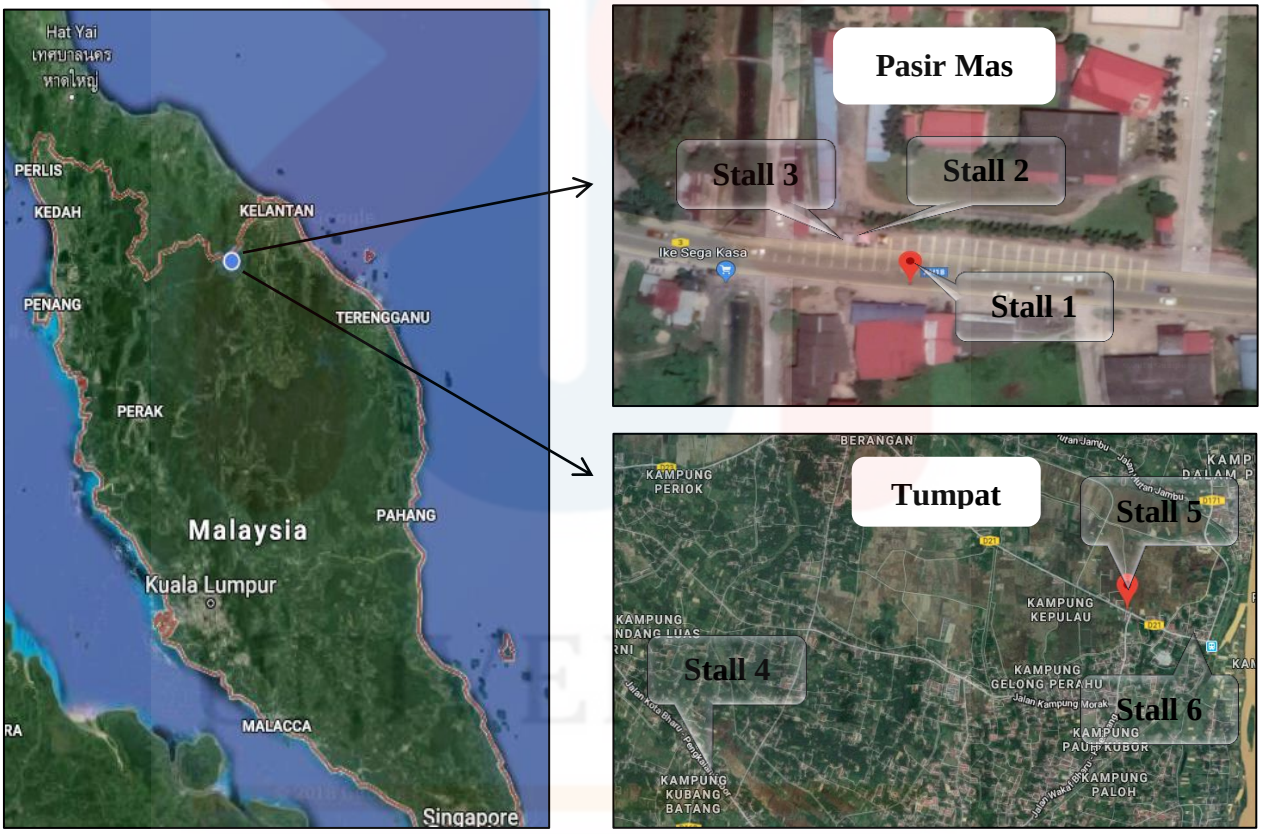

Figure 3.1: Location of study area at Pasir Mas and Tumpat

Table 3.1: The coordinate of the stalls.

District	Stalls	Coordinate
Pasir Mas	Stall 1	6.0495938, 102.1693547
	Stall 2	6.0497306, 102. 1691667
	Stall 3	6.0497333, 102.1691667
Tumpat	Stall 4	6.1379039, 102.170987
	Stall 5	6.1467273, 102.218605
	Stall 6	6.1450348, 102.222301

For sample preparation, the fresh and smoked *C. fluminea* tissue (Figure 3.1) was kept for further analysis. After that, the samples were dried. For drying process, the samples were dried overnight at 65°C for 48 hours in an air-circulation oven to get dried samples.

Figure 3.2: The tissue of fresh and smoked *C. fluminea* that was kept for further analysis

3.3 Determination of Moisture Content

An empty aluminium foil was weighed and the weight was recorded (W1). Then, approximately 5 g of sample was weighed (W2) and the sample was placed in to the aluminium foil. The sample was dried in air oven at 105°C for 24 hours. After that, the dried sample including the aluminium foil (Figure 3.2) was weighed immediately after drying process (W3). The weighing process was repeated for every hour until a constant reading of weight was recorded. Then, the weight of dry sample was divided by the weight of the wet sample and was multiplied by 100 to get a percentage. The percentage of dried matter was determined by using Equation 3.1.

$$\text{Dried matter (\%)} = \frac{W3 - W1}{W2} \text{ X } 100 \quad (3.1)$$

Then, Equation 3.2 was used to calculate the percentage of moisture content.

$$\text{Moisture content (\%)} = 100 - \text{percentage of dried matter} \quad (3.2)$$

Figure 3.3: Dry tissue that has been kept in an air-circulation oven for 24 hours

3.4 Determination of Ash

An empty crucible was weighed and denoted as W1 and approximately 5 g of sample was weighed that was denoted as W2. The sample was incinerated in furnace at 550˚C for 4 hours and was allowed to cool (Figure 3.3). Next step was that the sample was cooled down in desiccator to room temperature. The weight of crucible and ash were weighed and was denoted as W3. The percentage of ash content was determined by using Equation 3.3.

$$\text{Ash (\%)} = \frac{W3 - W1}{W2} \times 100 \qquad (3.3)$$

Figure 3.4: The tissue that has been furnaced with the temperature of 550˚C

3.5 Determination of Crude Fat

Principle in determination of crude fat is the dried sample will be dissolved with petroleum ether and then the ether will be evaporated in the Soxhlet apparatus (Figure 3.4) and the residues are crude fat.

The round bottom flasks was rinsed with petroleum ether and then dried in an oven at 102℃. After dry, the flask was kept in desiccator to let it cool. The flask was then weighed and denoted as W1. Then, 5 g of grounded and dried sample was weighed which was denoted as W2 and placed in a thimble. The thimble was then placed in the Soxhlet extractor. A clean 150 mL of round bottom flask was filled with 350 mL of petroleum ether. The whole setting of the apparatus was placed on a heating mantle to allow the petroleum ether to boil. The extraction process was continued for several hours. The condensing unit was then removed from the extraction unit and the sample was allowed to cool down. All of the solvent was collected after distillation process. After that, the flask was placed in the oven at 102℃ (Figure 3.5). The sample was dried for 1 to 2 hours until constant weight was recorded and denoted as W3. The flask was cooled in the desiccator and the weight of the flask and sample was recorded. The percentage of crude fat content was determined by using Equation 3.4.

$$\text{Crude fat (\%)} = (W3 - W1) \times \frac{100}{W2} \tag{3.4}$$

Figure 3.5: The set-up of soxhlet apparatus

Figure 3.6: The final product after drying the solvent

3.6 Determination of Crude Protein

3.6.1 Preparation of 40% Sodium Hydroxide (NaOH)

400g of NaOH was diluted in 1L of distilled water. Precaution steps were taken during preparation process as diluted NaOH can be hot.

3.6.2 Preparation of Methyl Red and Bromocresol Green

0.1g of methyl red was diluted in 100 mL of ethanol as well as 0.1g bromocresol green was diluted in 100 mL of ethanol.

3.6.3 Preparation of 4% Boric Acid (H_3BO_3)

6g of boric acid was weighed and diluted with 300 mL of distilled water. Then, it was mixed with 2.1 mL of methyl red and 3 mL of bromocresol green. Then, it was heated until it turned to pink in colour.

3.6.4 Determination of Crude Protein

Principle of determining crude protein was that determination of total nitrogen will be conducted by Kjeldahl method. This method is divided into three parts which are digestion, distillation and titration.

For digestion, 1 g of sample was weighed and was inserted into each digestion tubes and each tube was filled with 10 mL of distilled water and 1 piece of

Kjeldahl tablet. Next, 12 mL of concentrated sulphuric acid, H_2SO_4, was added into each tube inside the fume chamber and were placed inside the digestion rack. The fume manifold was attached tightly on the top of the digestion tube before the H_2SO_4 aspirator was turned completely to prevent the vaporised H_2SO_4 from escaping. The samples were digested in the digestion block (Figure 3.6) at 400°C for 3 hours. After complete total time of 3 hours, the digestion rack was removed into the rack holder inside the fume chamber and let to cool.

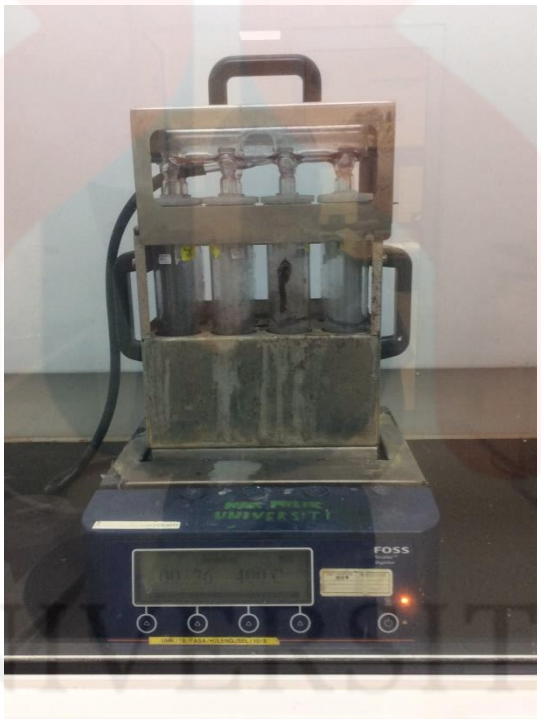

Figure 3.7: The digestion rack and digestion block

As for distillation, the distillation unit (Figure 3.7) was run for 3 times to clean the system. The digested sample was then diluted with 80 mL of distilled water and 50 mL of 40% NaOH. 30 mL of receiver solution was added to the receiver flask and the reaction was allowed to settle. Then, 250 mL Erlenmeyer titration flask was placed on receiving platform and was filled with 4% of boric acid (H_3BO_3) along with indicator and was added into receiver solution tank. The digestion tube that

contain diluted digest was attached to distillation unit and the sample was distilled for 5 minutes. Steam distillated that are green in colour was collected and receiving flask was removed from the unit to continue with titration process.

Figure 3.8: The Gerhardt Vapodest distillation unit

Titration process was started with H_3BO_3 receiving solution titrated with standard 0.1 M HCl in order to reach pink colourisation end point. The volume of HCl used for titration was recorded. Equation 3.5 and Equation 3.6 was used to determine crude protein content.

$$N \quad (\%) = \frac{[V - V\,(blank)] \times N \times 14.007}{W} \tag{3.5}$$

Where,

V = Volume of acid neutralized sample (mL)

N = Concentration of HCl

W = Weight of sample (mg)

$$CP\ (\%) = N\ (\%)\ X\ 6.25 \tag{3.6}$$

3.7 Determination of Carbohydrate

Carbohydrate content can be calculated after analysing all the other components. Carbohydrate content can simply be determined by taking total mass of product and subtract all other components. Carbohydrate content was determined by using Equation 3.7.

$$\text{Carbohydrate (\%)} = 100 - (\text{percentage of moisture + percentage of ash + percentage of fat + percentage of protein}) \tag{3.7}$$

3.8 Data Analysis

Calculation of mean and standard deviation of each element of nutritional value was performed. The comparison of nutritional value content between fresh and smoked *C. fluminea* via traditional smoking process was analysed using SPSS through two-way ANOVA method.

CHAPTER 4

RESULTS AND DISCUSSION

The research was done to study the nutritional value in both fresh and smoked *C. fluminea* tissue via traditional smoking process. The analysis of moisture content, ash, fat, protein and carbohydrate were done. The data was collected, analysed and were summarized in this chapter.

4.1 Moisture content

The percentage of moisture content and ash content was determined in order to calculate the carbohydrate content which will be calculated at the end of analysis. The mean and standard deviation of moisture content in both stages for each stall are presented in Table 4.1.

Table 4.1: Mean and standard deviation of moisture content in both stages.

Stage / Stall	Stall 1	Stall 2	Stall 3	Stall 4	Stall 5	Stall 6
Fresh *C. fluminea*	80.30±0.14	81.08±0.36	80.86±0.75	80.66±0.27	80.13±0.25	80.23±0.22
Smoked *C. fluminea*	79.68±0.72	79.52±0.26	78.36±0.26	77.60±0.09	77.77±0.55	75.90±0.14

According to Table 4.1, fresh *C. fluminea* from stall 2 and stall 3 has the highest moisture value which is 81.08% and 80.86% respectively. The lowest moisture value is from smoked *C. fluminea* from stall 4 and stall 6 which is 77.60% and 75.90% respectively. The values of moisture content from the two stalls are just slightly different. This can be seen clearly in the graph below as shown in Figure 4.1.

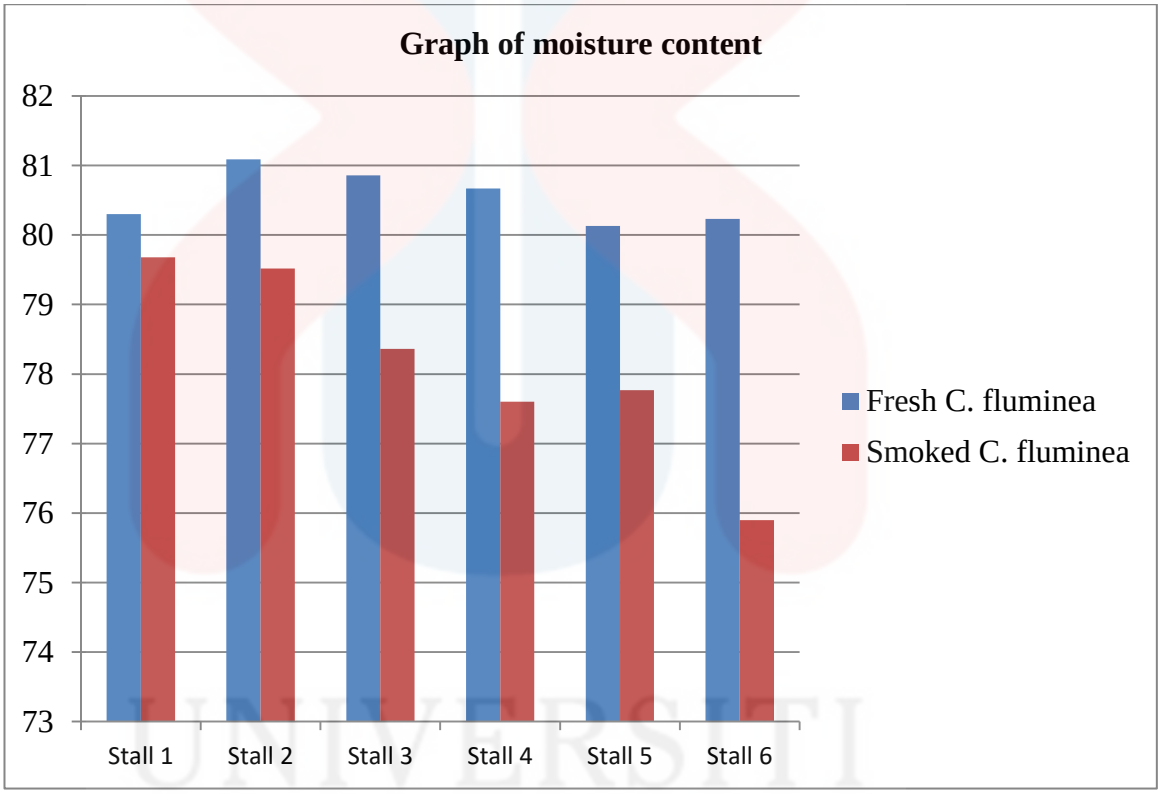

Figure 4.1: The graph of moisture content in fresh and smoked *C. fluminea*

The analytical data reveals showed that there are statistically significant interaction between stages and stalls where $p < 0.05$. The analytical data also showed that there are statistically significant difference in mean stages and stalls where $p < 0.05$.

Water is one of the most important nutrients in any living system. An adult human body consist of 60% water on average amount. Insufficient amount of water in human body can cause dehydration where it can cause imperfect physiological response (Borghi *et al*. 1996). Water can be associated with biochemical mechanisms and it is required to control and retain body temperature.

Based on USDA (2018a), shellfish contain approximately 75% amount of water. In this study, the values of moisture content in both fresh and smoked *C. fluminea* range between 75.90% - 81.08%. The amount of moisture content in this study exceeds the standard approximate amount of moisture content in shellfish.

4.2 Ash content

As mentioned, the analysis of ash content is done for preparation to calculate the carbohydrate content in *C. fluminea* tissue. The tissue of fresh and smoked *C. fluminea* was furnace at the degree of 550℃ for 4 hours.

Table 4.2 presents the mean and standard deviation of ash content in fresh and smoked *C. fluminea* from different stalls. According to Table 4.2, smoked *C. fluminea* from stall 3 and stall 6 has the highest value of ash content which is 1.96% and 1.81% respectively while the lowest value of ash content is fresh *C. fluminea* from stall 0.61% and 0.66% respectively. The range of ash content in both stages which is fresh and smoked *C. fluminea* is in between 0.61% - 1.96%.

Table 4.2: Mean and standard deviation of ash content in both stages.

Stage / Stall	Stall 1	Stall 2	Stall 3	Stall 4	Stall 5	Stall 6
Fresh *C. fluminea*	1.01±0.003	0.94±0.02	0.61±0.01	0.75±0.05	0.66±0.02	1.11±0.01
Smoked *C. fluminea*	1.80±0.01	1.60±0.01	1.96±0.03	1.76±0.03	1.73±0.02	1.81±0.02

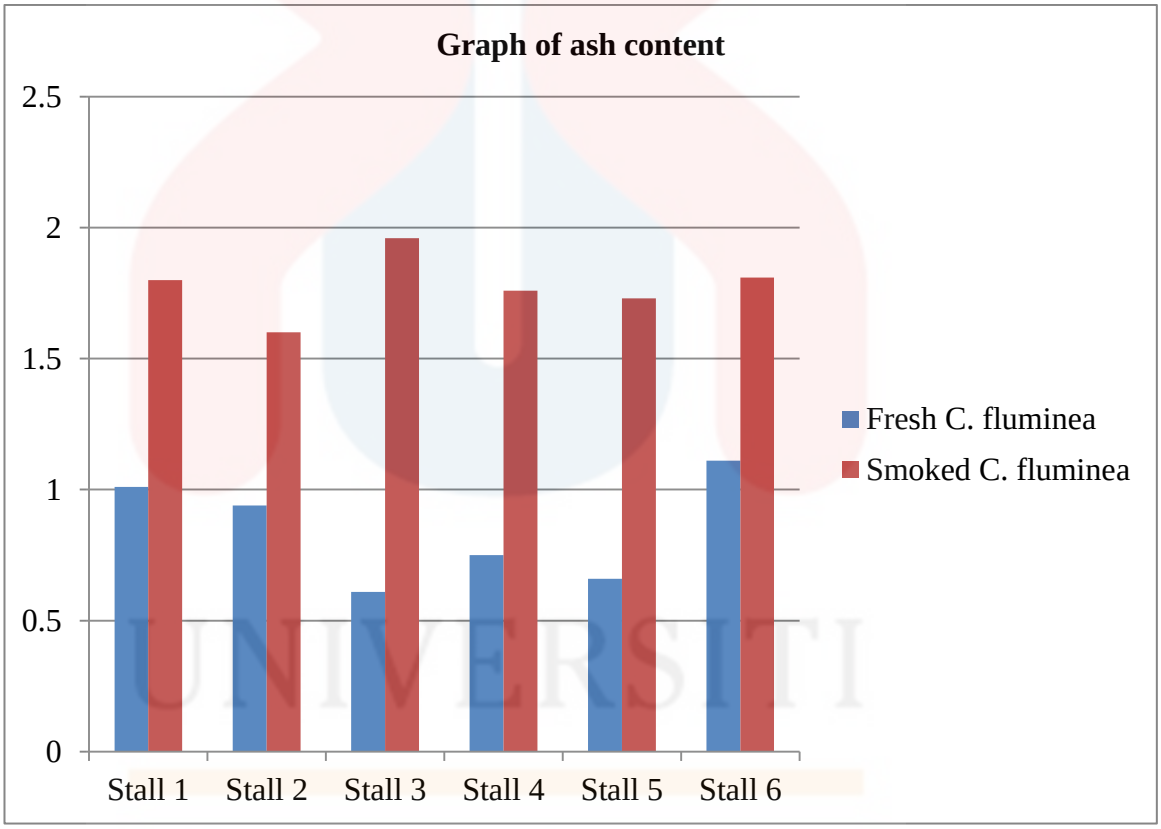

Figure 4.2: The graph of ash content in fresh and smoked *C. fluminea*

According to analytical data, there are statistically significant interaction between stages and stalls where the value of $p < 0.05$. The significance value of stages and stalls also $p < 0.05$ which means there are statistically significance

difference between two stages of *C. fluminea* and there are statistically significant difference between the stalls.

4.3 Fat content

Fat content analysis was done through soxhlet extraction method where the dried sample of fresh and smoked *C. fluminea* was dissolved with petroleum ether and was evaporated in the soxhlet apparatus. The residue of the extracted sample was count as crude fat.

Through this analysis, fat content was found the highest in fresh *C. fluminea* from stall 1 which is 11.66% while the lowest in smoked *C. fluminea* from stall 1 which is 8.75% as shown in Table 4.3 and Figure 4.3.

Table 4.3: Mean and standard deviation of fat content in both stages.

Stage / Stall	Stall 1	Stall 2	Stall 3	Stall 4	Stall 5	Stall 6
Fresh *C. fluminea*	11.66±0.32	10.49±0.36	11.24±0.16	10.23±0.11	10.44±0.12	10.67±0.26
Smoked *C. fluminea*	8.75±0.10	10.02±0.02	10.05±0.05	11.14±0.38	9.74±0.20	10.54±0.32

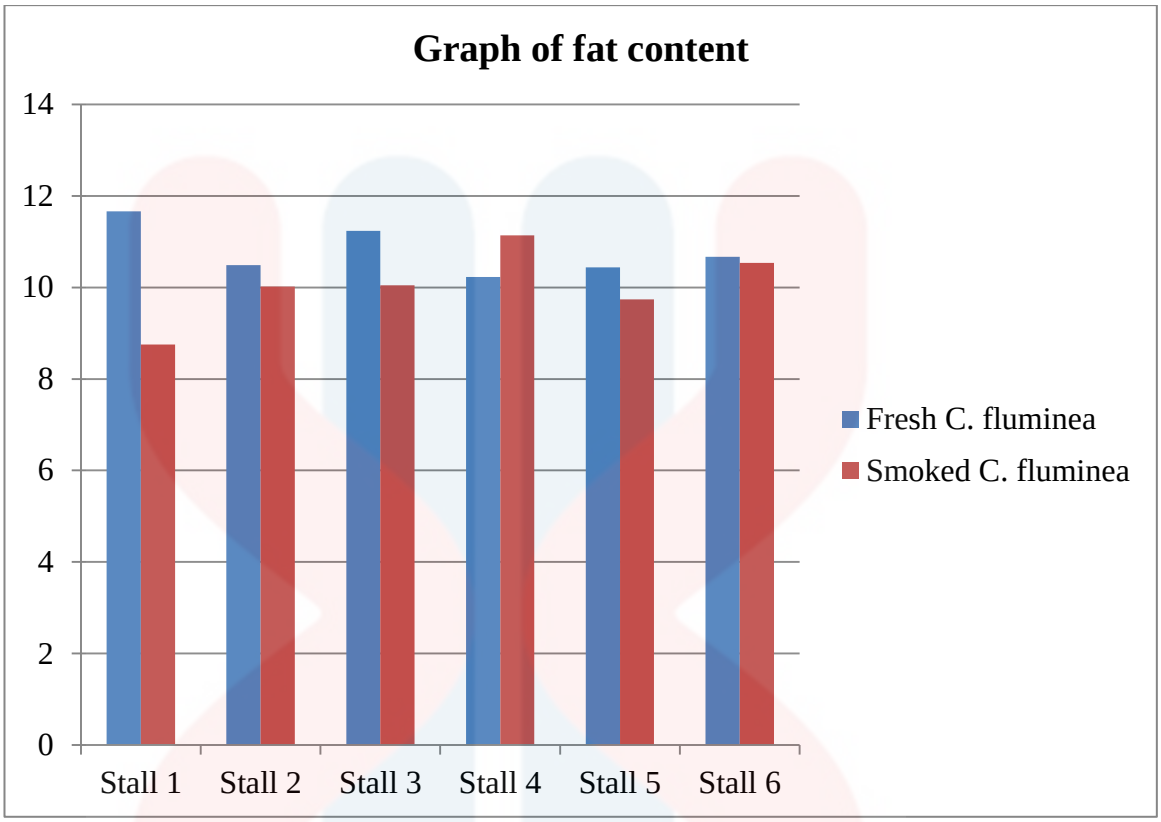

Figure 4.3: The graph of fat content in fresh and smoked *C. fluminea*

For fat content, the analytical data showed that there is statistically significant interaction of stages and the stalls where $p < 0.05$. For mean stages and stalls, there is statistically significant difference for both dependent variables where $p < 0.05$.

4.4 Protein content

The result of protein content in fresh and smoked *C. fluminea* tissue is shown in Table 4.4. According to the table, the highest value of protein content was found in smoked *C. fluminea* from stall 6 which is 10.92%. The lowest value of protein content was found in fresh *C. fluminea* from stall 1 and stall 3 which is 6.24% and 6.21% where the difference of the value is just slightly different.

Table 4.4: Mean and standard deviation of protein content in both stages.

Stage / Stall	Stall 1	Stall 2	Stall 3	Stall 4	Stall 5	Stall 6
Fresh *C. fluminea*	6.24±0.11	6.34±0.08	6.21±0.02	7.89±0.11	8.03±0.06	7.61±0.09
Smoked *C. fluminea*	8.97±0.17	8.33±0.08	9.42±0.03	9.05±0.17	8.86±0.13	10.92±0.11

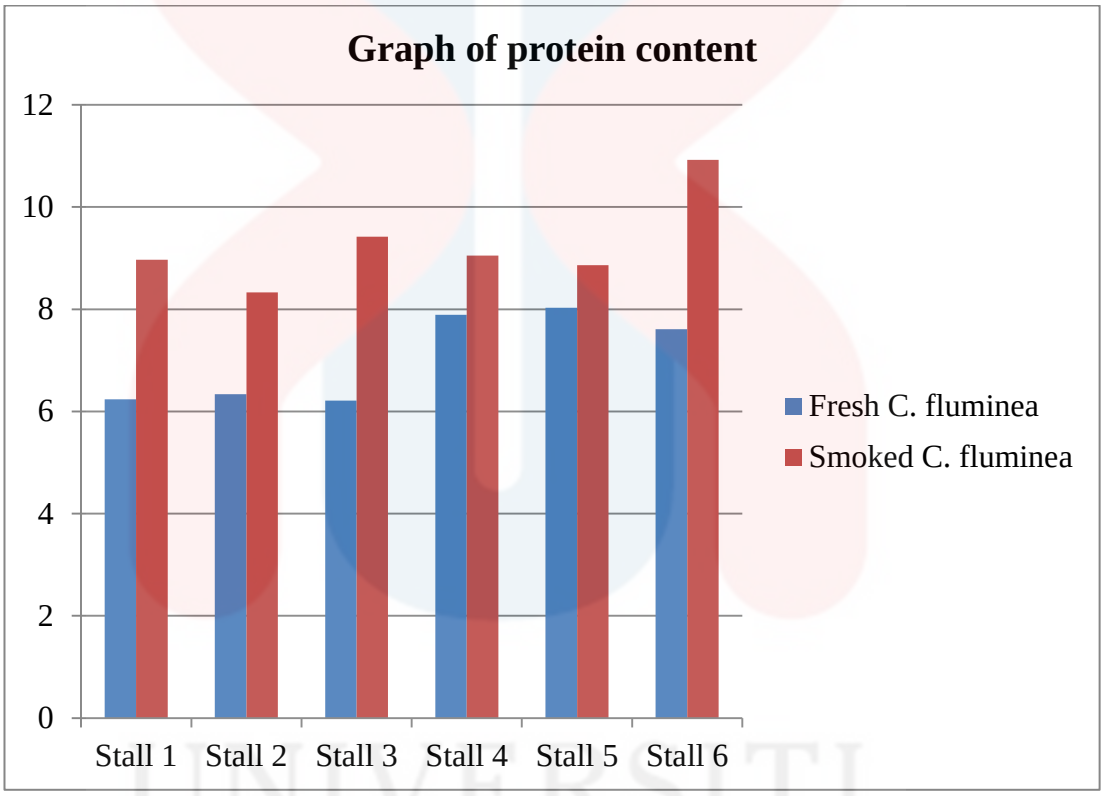

Figure 4.4: The graph of protein content in fresh and smoked *C. fluminea*

The significance value of interaction between stages and stalls is below 0.05 as in $p < 0.05$ which means there is statistically significant interaction. For stages, the significant value is $p < 0.05$ which means it is statistically significant different between the two stages of *C. fluminea*. As for stalls, the significant value is $p < 0.05$ which also means there is statistically significant difference between the stalls.

A normal adult male should eat up 50 g of protein per day based on USDA (2018b). Protein is a dominant component for lean muscle mass and it is an important component for cellular processes. If the human body is lack of carbohydrate to regulate energy, protein can take place to contribute the energy to the human body (Wright, Fan, & Baker, 2018)

Dong (2001) stated that shellfish are rich in protein content. Average approximate composition for protein in shellfish is 9.0% to 13.0%. Shellfish is count as high-quality protein due to its constitutive amino acid profile. Thus, it is classified as a source of food that contains highly digestible protein content.

The amount of protein content in both stages, fresh and smoked *C. fluminea* is in range 6.24% to 10.92% which equal to the range stated by Dong (2001).

4.5 Carbohydrate content

Carbohydrate content was analysed after analysis of all other components has completed. Table 4.5 and Figure 4.5 shows the result of protein content in both fresh and smoked *C. fluminea* tissue from different stalls. According to Table 4.5, it shows that smoked *C. fluminea* from stall 5 has the highest value of carbohydrate content which is 1.91% while smoked *C. fluminea* from stall 3 has the lowest value of carbohydrate content which is 0.22%.

Table 4.5: Mean and standard deviation of carbohydrate content in both stages.

Stage / Stall	Stall 1	Stall 2	Stall 3	Stall 4	Stall 5	Stall 6
Fresh *C. fluminea*	0.80±0.55	1.14±0.71	1.08±0.59	0.46±0.39	0.74±0.31	0.37±0.30
Smoked *C. fluminea*	0.81±0.69	0.53±0.18	0.22±0.25	0.44±0.26	1.91±0.24	0.83±0.31

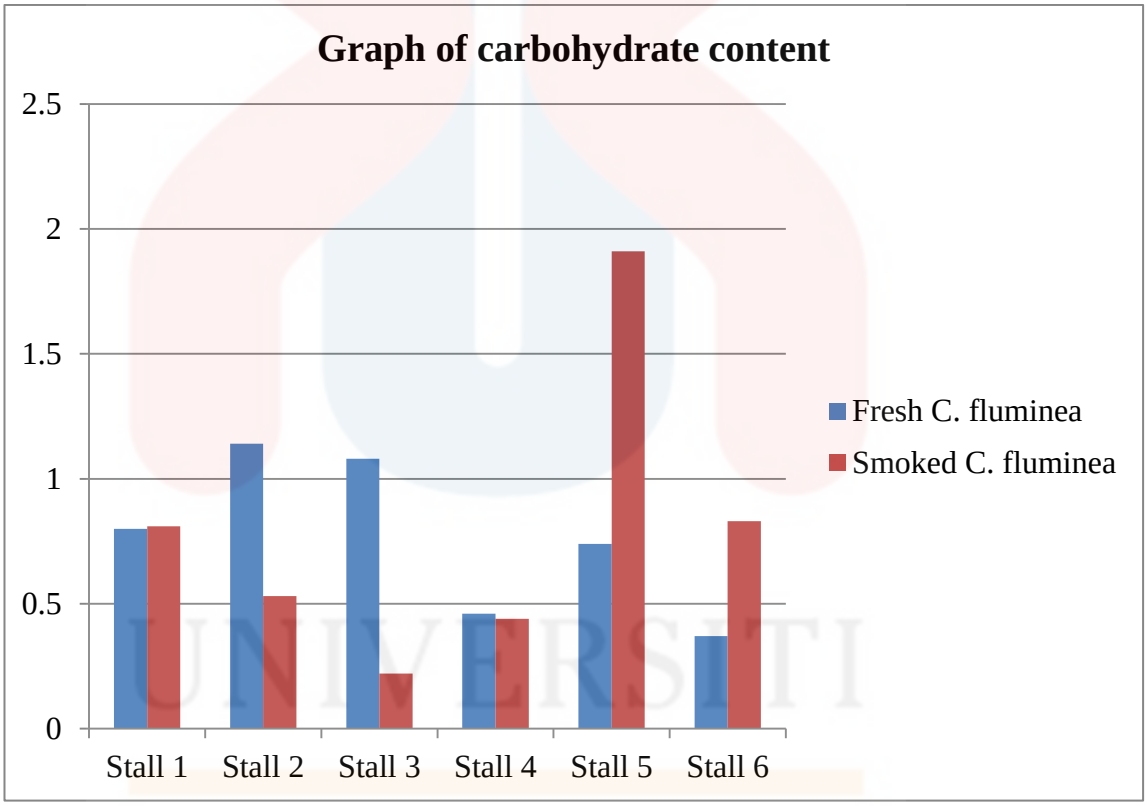

Figure 4.5: The graph of carbohydrate content in fresh and smoked *C. fluminea*

The analytical data showed that there is statistically significant interaction between stages and stalls with $p < 0.05$. For stalls, there is statistically significant difference between the stalls with $p < 0.05$, but there is no statistically significant difference between the stages of *C. fluminea* where $p > 0.05$.

Sugar, fibre and starch are included in types of food carbohydrate which will be changed into glucose. This will provide the energy to the human body. Human body need carbohydrates in only certain quantities. Oversupply of carbohydrates may cause chronic health issues like obesity and Type 2 diabetes (Wylie-Rosett *et al*. 2004).

Carbohydrate content in shellfish tissue is in moderate levels. This may be because of consumption pattern where they are consumed lived or shortly after death. The amount of protein content in shellfish is generally low (Wright, Fan, & Baker, 2018). In this study, it shows that the range of protein content in *C. fluminea* is in the range of 0.37% to 1.91%. According to Venugopal & Gopakumar (2017), the maximum amount of carbohydrate content in Asian clam is about 7.9%

CHAPTER 5

CONCLUSION AND RECOMMENDATIONS

5.1 Conclusion

In conclusion, the objective of determining the nutritional value of both fresh and smoked *C. fluminea* via traditional smoking process is achieved. Fresh *C. fluminea* is found to be high in moisture content and fat where the highest value is 81.09% and 11.66% respectively. For smoked *C. fluminea,* it is recorded that it is high in ash, protein and carbohydrate. The highest value for these three components are 1.96%, 10.92% and 1.91% respectively. The analytical data showed that there are significant differences between the two stages except for carbohydrate where the significance value is $p > 0.05$.

5.2 Recommendations

This research can be studied further in order to investigate the following:

1. The nutritional value in smoked *C. fluminea* tissue that is cooked via modified smoking process.
2. The comparison between nutritional values in smoked *C. fluminea tissue* via traditional and modified smoking process.

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APPENDIX A

Table A.1: Descriptive analysis of moisture content

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	83.662[a]	11	7.606	48.591	.000
Intercept	226629.696	1	226629.696	1447903.909	.000
stage	52.126	1	52.126	333.028	.000
stall	19.472	5	3.894	24.881	.000
stage * stall	12.063	5	2.413	15.414	.000
Error	3.757	24	.157		
Total	226717.115	36			
Corrected Total	87.419	35			

a. R Squared = .957 (Adjusted R Squared = .937)

Table A.2: Descriptive analysis of ash content

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	8.573[a]	11	.779	1412.511	.000
Intercept	61.933	1	61.933	112240.021	.000
stage	7.764	1	7.764	14070.665	.000
stall	.305	5	.061	110.635	.000
stage * stall	.504	5	.101	182.756	.000
Error	.013	24	.001		
Total	70.519	36			
Corrected Total	8.587	35			

a. R Squared = .998 (Adjusted R Squared = .998)

Table A.3: Descriptive analysis of fat content

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	19.221[a]	11	1.747	32.202	.000
Intercept	3903.863	1	3903.863	71941.961	.000
stage	5.063	1	5.063	93.297	.000
stall	2.049	5	.410	7.550	.000
stage * stall	12.110	5	2.422	44.634	.000
Error	1.302	24	.054		
Total	3924.386	36			
Corrected Total	20.524	35			

a. R Squared = .937 (Adjusted R Squared = .907)

Table A.4: Descriptive analysis of protein content

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	67.210[a]	11	6.110	532.686	.000
Intercept	2394.512	1	2394.512	208761.245	.000
stage	43.739	1	43.739	3813.349	.000
stall	15.130	5	3.026	263.824	.000
stage * stall	8.340	5	1.668	145.415	.000
Error	.275	24	.011		
Total	2461.997	36			
Corrected Total	67.485	35			

a. R Squared = .996 (Adjusted R Squared = .994)

Table A.5: Descriptive analysis of carbohydrate content

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	6.785[a]	11	.617	3.244	.008
Intercept	21.717	1	21.717	114.237	.000
stage	.005	1	.005	.025	.876
stall	2.758	5	.552	2.902	.035
stage * stall	4.022	5	.804	4.231	.007
Error	4.562	24	.190		
Total	33.064	36			
Corrected Total	11.347	35			

a. R Squared = .598 (Adjusted R Squared = .414)