# Effects of Different Inclusion Rates of Black Soldier Fly Larvae (BSFL) on Physical Properties of *Macrobrachium rosenbergii* juvenile feed

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## DECLARATION

I declare that the work embodied in this thesis is from my own research except for the content that have been cited and summarised and I have made reference for every source.

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**Effects of Different Inclusion Rates of Black Soldier Fly Larvae (BSFL) on Physical Properties of *Macrobrachium rosenbergii* juvenile feed**

## ABSTRACT

In the animal feed industry, aquaculture is the fastest-growing sector. One of the freshwater aquaculture species contributing to the national economy is the giant freshwater prawn (*Macrobrachium rosenbergii*). However, the rising cost of food containing imported protein sources like fish meal has made it difficult for many farmers to keep their farms afloat and stay in the industry. Compared to expensive fish feed, black soldier fly larvae are one of the protein sources that are not yet well-known as a primary source of protein. Its advantages include a high feed conversion rate and high nutritional content and a short life cycle that can be effectively optimized in feed formulation. This study aimed to produce a sustainable physical properties of shrimp feed for juvenile *Macrobrachium rosenbergii* growth. Differences in total percentage consumption of BSFL in 5 different diets which is Diet I, Diet II, Diet III, Diet IV, and Diet V which represent 0% BSFL as a control, 10% BSFL, 20% BSFL, 30% BSFL and 40% BSFL. The results of this study showed the physical properties of feed formulation for BSFL diet 30% better than compared to other diet test. This study proved that the physical properties with the addition of BSFL did not affect the feed formulation for *Macrobrachium rosenbergii* for growth and it paves the way for more research on new protein sources for animal feed, as well as bringing the potential benefits of BSFL to public attention.

Keywords: *Macrobrachium rosenbergii*, Black Soldier Fly Larvae (BSFL), Feed Formulation, juvenile, physical properties

**Kesan Kadar Kemasukan Berbeza Larva Lalat Askar Hitam (BSFL) terhadap Sifat Fizikal makanan juvana *Macrobrachium rosenbergii***

## ABSTRAK

Dalam industri makanan haiwan, akuakultur adalah sektor yang paling pesat berkembang. Salah satu spesies akuakultur air tawar yang menyumbang kepada ekonomi negara ialah udang air tawar gergasi (*Macrobrachium rosenbergii*). Walau bagaimanapun, peningkatan kos makanan yang mengandungi sumber protein yang diimport seperti tepung ikan telah menyukarkan ramai petani untuk mengekalkan ladang mereka dan kekal dalam industri. Berbanding makanan ikan yang mahal, larva lalat askar hitam merupakan salah satu sumber protein yang belum terkenal sebagai sumber protein utama. Kelebihannya termasuk kadar penukaran makanan yang tinggi dan kandungan nutrisi yang tinggi serta kitaran hayat yang singkat yang boleh dioptimumkan dengan berkesan dalam penggubalan makanan. Kajian ini bertujuan untuk menghasilkan sifat fizikal makanan udang yang mampan untuk pertumbuhan *Macrobrachium rosenbergii* juvana. Perbezaan jumlah peratusan penggunaan BSFL dalam 5 diet berbeza iaitu Diet I, Diet II, Diet III, Diet IV dan Diet V yang mewakili 0% BSFL sebagai kawalan, 10% BSFL, 20% BSFL, 30% BSFL dan 40 % BSFL. Hasil kajian ini menunjukkan sifat fizikal formulasi makanan untuk diet BSFL 30% lebih baik berbanding ujian diet lain. Kajian ini membuktikan bahawa sifat fizikal dengan penambahan BSFL tidak menjejaskan formulasi makanan untuk *Macrobrachium rosenbergii* untuk pertumbuhan dan ia membuka jalan untuk lebih banyak penyelidikan tentang sumber protein baharu untuk makanan haiwan, serta membawa potensi manfaat BSFL kepada perhatian umum.

Kata kunci: *Macrobrachium rosenbergii*, Black Soldier Fly Larvae (BSFL), formulasi makanan, juvenil, sifat fizikal

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## LIST OF ABBREVIATIONS AND SYMBOLS

| | |
|---|---|
| BSFL | Black Soldier Fly Larvae |
| FM | Fish Meal |
| *M. rosenbergii* | *Macrobrachium rosenbergii* |
| TPA | Textural Profile Analysis |
| % | Percentage |
| mg | Milligram |
| g | Gram |
| kg | Kilogram |
| mm | Millimetre |
| mL | Millilitre |
| L | Litre |
| per | Or |
| s | Second |
| cm | Centimetre |
| h | Hour |
| °C | Degree Celsius |
| i.e. | Id est / That is |

# CHAPTER 1

# INTRODUCTION

## 1.1 Research background

*Macrobrachium rosenbergii* is the world's largest freshwater prawn and is widely cultivated for food in several countries. In Malaysia, the giant freshwater prawn or scientifically known as *Macrobrachium rosenbergii* (de Man, 1879) or locally known in Malaysia as "*udang galah*" (Jee, 1998). It is the largest genus of Macrobrachium. *M. rosenbergii* is a Southeast Asian prawn native to Thailand, Malaysia, Vietnam, Myanmar, Sri Lanka, Bangladesh, India, Cambodia, and the Philippines. According to Food and Agriculture Organization (FAO), Shao-Wen Ling, the expert who found out that *M. rosenbergii* larvae, can endure in brackish habitat. *M. rosenbergii* is mainly found in the lower reaches of river systems affected by tides (Ling, 1969).

Protein is an important ingredient in the shellfish diet, and for maximal development and reproduction, shellfish require adequate protein levels in their diet (Sundaravadivel et al., 2015). The ideal dietary protein level required by freshwater prawns varies by species, also among different phase of development in the same species with variable protein requirements for distinct prawn species. Furthermore, due to excellent growth performance gained from a low protein diet, the protein requirement of the freshwater prawn *M. rosenbergii* is thought to be lower than that of other prawn species. However, the optimal dietary protein level for prawn changes depending on several factors such as nutrition, protein quality, dietary composition and so on (Teshima et al., 2006).

*Hermetia illucens sp*., or black soldier fly larvae, is a member of the Stratiomyidae family of invasive insects. Although originally from America, this BSFL can be found throughout the world in temperate regions. Since they are more endemic in tropical and hot climates, BSFL can be found on every continent except Antarctica due to less resistance to cold (Wang & Shelomi, 2017). The BSFL is a sleek insect that behaves like a wasp but is not a pest fly. BSFL still has no stings and is harmless to humans. BSFL is commonly associated with outdoor areas and poultry, which can be detected on rotting organic matter such as animal waste or carcasses and decaying crops. BSFL is used to eliminate pollution in agricultural pig and poultry farms (Larry Newton, 2005).

BSFL has the potential to be a protein source to sustain global demand. Where BSFL provides high-quality protein for a range of animals to feed. Furthermore, because BSF is not a pest, its rearing does not necessitate any special precautions, and it minimises the presence of pathogenic bacteria. Because of their high protein and lipid content, even when fed with plant-based waste streams, BSFL meal and oil are already regarded animal-grade alternatives to fish meal and fish oil used to feed carnivorous fish and in other animal diets. Because of its cheaper cost, BSFL has been suggested as an alternative. Finally, the ability of BSFL to efficiently produce protein-rich edible biomass from potentially protein-poor organic waste has led many researchers to believe that BSFL can make a significant contribution to sustainable aquaculture as part or all a replacement feed for aquatic invertebrates such as prawn (Wang & Shelomi, 2017).

## 1.2 Problem statement

In Malaysia's aquaculture sector, reliance on imported raw materials is a major source of concern. Farmers have to bear the high cost of aquaculture feed and the limited availability of protein sources. This is because of the problem in high protein cost and dependent on imported feed materials. Fish meal (FM) was once the primary source of animal protein in aquaculture. Overfishing has escalated, posing a threat to many wild fish populations, and this, together with rising demand, has resulted in higher FM prices and a decrease in aquafeed availability and demand. The use of BSFL as a significant alternative source of protein in giant freshwater prawn culture is still in its early stages of development and has yet to be thoroughly tested in farms as partial protein replacement. The cost of imported raw materials used in prawn feed production is increasing, making it impossible for farmers to procure at a fair price. Because of the high production cost, shrimp farmers have an inadequate source of nutrients, causing the prawn's metabolism to fall short of expectations, implying that the amount of nutrients they need is insufficient. By used BSFL as protein replacement in feed, the larvae of black soldier flies (BSFL) are used to decompose trash or transform it into animal feed. Fly larvae are among the most effective biomass converters.

In juvenile *M. rosenbergii*, a lack of nutrients causes stunted development and a poor survival rate. Fishmeal is the most sustainable and best aquaculture diet despite the costliest macro ingredients. Commercial food with decent quality is the top option for having the highest yield and fast development. However, this price is essentially meaningless for small farmers who spend exclusively on shrimp feed. As a result, the presence of BSLF as an alternative source of protein can make it possible to achieve profitable results in this giant freshwater prawn diet at a lower and more sustainable cost.

## 1.3 Objectives

i. To determine the effect of different inclusion rates of BSFL on the physical characteristics of *Macrobrachium rosenbergii* juvenile feed.

## 1.4 Hypothesis

$H_o$: There is no significant effect of different inclusion rates on the physical characteristic of *M. rosenbergii* feed.

$H_1$: There is a significant effect of different inclusion rates on the physical characteristic of *M. rosenbergii* feed.

## 1.5 Scope of the study

In this study, feed for *M. rosenbergii* was formulated using Winfeed software. This study focuses on the effect of different inclusion rates of BSFL on physical properties and textural profile analysis on the *M. rosenbergii* pellet. The different percentage of BSFL was compared to investigate their effect on the physical properties of *M. rosenbergii* pellet.

## 1.6 Limitation of the study

The formulation of feed Giant freshwater prawn (*Macrobrachium rosenbergii*) in this study uses the manual pelletizing system. The study was conducted from October 2021 until December 2021, which is only 2 months for running the experiment. The result might differ with pelletizing machine size and shape than manual. BSFL is used as the feed formulation material in this experiment. Due to the limited supply of BSFL in stage prepupae, there might be a bit of an obstacle to getting this BSFL in terms of quantity and price. Due to time constraints, the experiment was conducted for nine weeks, including the experiment's preparation in the first week. *M. rosenbergii* needs at least three months to do a feeding trial but only had time to make feed formulation of BSFL pellet.

## 1.7 Significance of the study

The addition of new BSFL formulations to shrimp feed can help farmers lower productivity costs by providing low cost and massively available feed ingredients.

# CHAPTER 2

# LITERATURE REVIEW

## 2.1 General introduction of *Macrobrachium rosenbergii*

The world's largest palaemonid is the enormous freshwater prawn, previously known as *Macrobrachium rosenbergii* (de Man, 1879) (Wowor, 2007). Giant freshwater prawn is a highly valued cultured species due to its widespread consumer acceptance, ease of culture, delicious species, and export potential. *M. rosenbergii* de Man, a giant freshwater prawn, can be found throughout Southeast Asia, South Asia, Northern Australia, and the Western Pacific Islands (New, 2000). High-regard species, for instance, shrimp, prawns are significantly traded, explicitly towards more prosperous countries (FAO, 2018). The *M. rosenbergii* are commercially valuable as a food source. This species has been widely distributed because of its popularity in commercial aquaculture.

According to Food and Agriculture Organization (FAO) the expert, Shao-Wen Ling, a researcher at the Marine Fisheries Research Institute in Penang, Malaysia, made a significant discovery in 1961 that during the larval stages, this species prefers to be in an estuarine environment. *M. rosenbergii* juveniles migrate upstream in search of freshwater. Rivers, lakes, swamps, and irrigation canals are home to large adults (New, 2002). Larvae can survive for a few days in freshwater before moving to salty water. Aquaculture has become increasingly popular with this species. The product's price has increased in every country as market demand continues to rise.

The *M. rosenbergii* contributes significantly to global aquaculture production. Life cycle of *M. rosenbergii* starts from eggs, develop to post larvae, juvenile, and the last stage is adult. *M. rosenbergii* size is about 7 to 10 mm long and 6 to 9 mg for weight in the juvenile stage. At the water's surface, shrimp are very active. When swimming, juvenile shrimp movement travel in a head-forward orientation and with their dorsal (back) uppermost leg.

The characteristic morphology of *M. rosenbergii* has total body weight depending on the male sex has 32 cm long and female sex has 25 cm long (Soesanto, 1980). The colour body of *M. rosenbergii*. *M. rosenbergii* consists of bluish-green and sometimes brown. The male releases spermatophores on the underside of the female's thorax, between her walking legs, during mating. After that, the female extrudes eggs that pass through the spermatophores. The female carries the fertilized eggs until they hatch; the time varies, but it is usually less than three weeks. Up to five times every year, females lay 10,000–50,000 eggs. The first larval stage of crustaceans, zoeae, develops from these eggs. They go through numerous larval stages in brackish water before metamorphosing into post larvae, which are 0.28–0.39cm in (7.1–9.9 mm) long and look like adults. This transformation occurs typically 32 to 35 days after hatching. After then, the post larvae return to freshwater.

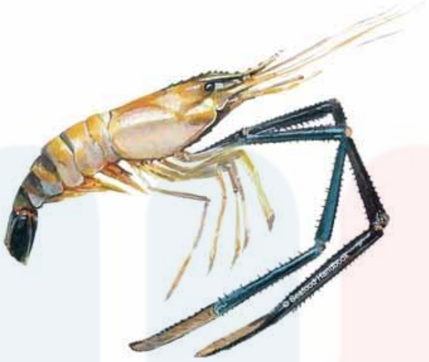


Figure 2.1: *Macrobrachium rosenbergii*

(Source from Seafood Source.com, 2014)

## 2.2 Malaysia production of *Macrobrachium rosenbergii*

According to Food and Agriculture Organization (2008), the first harvest of giant river prawns is recorded in Malaysia in 1970 in the FAO static. Malaysia is one of the 9$^{th}$ largest prawn exporters in the world. In 2020, aquaculture production from the freshwater prawn farming system in Malaysia has recorded a total of 192.69 metric tons according to statistics that released by the Department of Fisheries Malaysia (DOF). Malaysia has natural habitats such as ponds, rivers, estuaries, lakes, and coastal areas, so it has high potential for aquaculture development. However, 2011 is the year the production of this *M. rosenbergii* almost half decreased compared to 2010 due to the supply factor of quality juveniles and other factors contributing over the past decade.

## 2.3 Nutrient requirement in *Macrobrachium rosenbergii*

Freshwater prawns are culture on diets rich in protein and comparatively inexpensive. According to the Food and Agriculture Organization of the United Nations (FAO, 2002), *M. rosenbergii* was not a standardized dietary criterion, but the research starting understanding of a giant freshwater prawn species' nutritional requirements has improved significantly over the last decade. The amount of information available about the species' nutritional needs has increased dramatically improved last decade (New and Valenti, 2000). *M. rosenbergii* are competent in digesting a wide variety of plant and animal-based foods (Mitra, 2005). *M. rosenbergii* has varying nutrient requirements depending on the life cycle.

The dietary requirements of *M. rosenbergii* have to be standardized, but the understanding of the specific nutrient requirements has significantly improved over the last decade. *M. rosenbergii* can digest various feeds, whether plant or animal. *M. rosenbergii* needs 35 - 40% protein in their diet to increase the efficiency and growth rate of shrimp feed, which is better if they are in a pond with natural food (Sarman et al., 2011). Carbohydrates as a source of energy for molting and growth activities. For lipids and fatty acids in *M. rosenbergii*, low dietary cholesterol levels in nutritional foods affect egg quality resulting in low seed production quality. High lipids and cholesterol aid in the maturation and egg quality of *M. rosenbergii*. Shrimp need 60-150 mg of vitamin C for breeding in the brood stock stage. Giving vitamin C to female shrimp can improve carcass quality, accelerate growth at a low level, and tolerance to ammonia pressure. Information on the nutrient requirement of minerals for *M. rosenbergii* is minimal. However, dietary calcium supply appears to improve the growth of *M. rosenbergii*.

### 2.3.1 Protein and amino acid

The growth of *M. rosenbergii* in an apparent water culture system without natural food is ideal for consuming diets containing 35-40% protein (Nesara, 2018). A protein requirement based on the availability of sufficient levels has ten essential amino acids. Only a single protein source or a combination of protein sources in graded levels within dietary treatments has been studied for *M. rosenbergii* protein requirements. Using a single protein source, estimates of protein requirements are usually higher than the combination of sources used, resulting in a deficiency or lack of bioavailability of certain essential amino acids. Many fish and crustacean meals and soybean meals have previously used as primary sources of dietary protein for *M. rosenbergii*. The quantitative nutritional protein requirement of juvenile *M. rosenbergii* is between 30 and 40% (dry weight).

### 2.3.2 Carbohydrate

Diaz-Henera *et al*. (1992) discovered that carbohydrates are the primary energy-producing substrates in larval and post-larval *M. rosenbergii*. The fact that *M.*

*rosenbergii* has a relatively high specific activity of amylase suggests that it efficiently uses carbohydrates as a source of energy. Carbohydrates dominate the prawn's energy metabolism during fasting, followed by lipids and proteins. Simple sugars are not appropriately used as complex polysaccharides like starch and dextrin. Dietary glucosamine (an amino sugar that acts as a link between glucose and chitin) promotes moulting and growth. Diet protein is efficiently utilized when the dietary 8 lipid-carbohydrate ratio is 1:3-1:4, dietary protein is efficiently utilized. Prawns have also known to consume up to 30% dietary fibre.

### 2.3.3 Lipid and fatty acid

Juveniles of both *M. rosenbergii* and penaeid species are capable of efficiently digesting lipids. Both shrimp families appear to tolerate a wide range of dietary triglyceride levels, with low levels (20%) acceptable if essential fatty acid requirements are met and carbohydrates provide sufficient nutritional energy. A wide range of dietary lipid levels (2 to 10%) consists of a 2:1 ratio of cod liver oil and corn oil in diets. It proved to be equally effective when measured by weight gain response (Sheen and D'Abramo, 1991).

### 2.3.4 Vitamin

*M. rosenbergii*'s vitamin needs are likely to be like those of other crustaceans and fish. The prawn needs 60-150 mg of vitamin C per kg of body weight (Nesara, 2018). In prawn brood stock, 60 mg of ascorbic acid and 300 mg of tocopherol per kg of diet are considered adequate for proper reproduction and offspring viability. However, giving female prawns higher doses of both vitamins that around 900 mg/kg each may improve larval quality, including ammonia stress tolerance (Mitra, 2005).

### 2.3.5 Mineral

Data are inadequate on the quantitative mineral requirements of *M.rosenbergii* *M. rosenbergii* growth appears to be aided by calcium supplementation in the diet. On the other hand, Mitra, Chattopadhyay, and Mukhopadhyay (2005) suggested that providing calcium minerals to the prawns could help them grow faster.

## 2.4 Feeding behaviour of *M. rosenbergii*

All aspects of feeding behaviour include searching, detecting, orienting, grasping, and swallowing food. The feeding behaviour is depending on the life stage of *M. rosenbergii.* The creation of formulated feeds necessitates an understanding of feeding behaviour. The research found feeding behaviour of the *M. rosenbergii* juvenile used its first and second pincers to pick up a pellet of standard size, carried it to the mouth with a pair of 3 maxilliped endo pods, shredded only a tiny portion or half of it with a couple of mandibles, spit out the remaining mass, and stopped feeding. The pellets were not crushed by the *M. rosenbergii.* They often kept two or three pellets in their mouth when the pellets were small but masticated them one at a time. The tiny pellets were constantly spitting out by the juveniles, who then stopped feeding (Kawamura, 2018).

## 2.5 Feed formulation of *M. rosenbergii*

Natural feed and formulated feed are the two forms of feed often provided to *M. rosenbergii.* Various forms of live feed are included in the live feed community, depending on the different life cycles of *M. rosenbergii. M. rosenbergii* raised in ponds depend heavily on zooplankton and oligochaete worms for nutrition. Juveniles weighing more than 2 g will benefit directly from zooplankton. Prawn often eats earthworm larvae and insects. In the production of freshwater prawns, increased macroinvertebrate production in ponds is critical, as it significantly improves feed yield. However, when the biomass in the pond rises, the animals must consume high-quality food. Furthermore, using feeds given the results in more consistent *M. rosenbergii* productivity. (Mitra, 2005).

## 2.6 Black soldier fly larvae (BSFL)

Flies are beneficial and essential since they help manage other insect pests, serve as pollinators, recyclers, and scavengers, and are part of the food chain. Insects sustainable reared on organic side streams could offer a suitable alternative animal protein source (Smetana *et al*., 2016). The black soldier fly is a high-value protein component in animal diets with various benefits. The European Union can accept insect proteins in pet feed and aquaculture feed formulations (Mouithys-Mickalad *et al*., 2020). BSFL can be rearing on different substrates, including animal manures, pig liver, fish rendering waste, and fruit waste, human excreta, and food waste (Diener *et al*., 2011; 11 Nguyen *et al*., 2015). BSFL is not a pest, so its rearing requires no specific preventive measures. Compared to other dipteran species like the house fly, *Musca domestica*, it reduces the presence of harmful bacteria (Barragan-Fonseca *et al*., 2017). This is because adult black soldier flies lack a working mouth structure, they are not classified as pests or disease-carrying insects because they are unable to feed or bite. They only spend their adult lives mating and laying eggs (Hawkinson, 2005).

### 2.6.1 The life cycle of BSFL

*Hermetia illucens* or better known as black soldier fly larvae have a life cycle of between 44 days or more depending on their environment and food supply. The life cycle of black soldier fly larvae can be divided by five stages: egg, larva, prepupa, pupa, and adult stage. The female BSFL can produce 500 to 900 more, and the eggs will hatch after four days of being released by the female BSF. Female BSF will lay eggs in clean areas and close to food sources. After hatching, these BSF offspring will continue to begin their feeding mission for up to 13-18 days in the form of larvae, they will eat as much as possible to ensure they have an adequate food supply later in adulthood.

In the process of eating, the black soldier fly (BSF) will inadvertently convert waste products such as waste products from soy and pineapple to a source of protein from its body and compost from its faeces. Pre pupa is the stage where the larval skin will start to turn a little darker and continue the feeding process but in a smaller amount than before. Their movements will be reduced, turning into pupae, and stopping eating in a few days. They will be in a pupa state in 10 days to a month, depending on the environment, before turning into an adult bsfl. Next, the adult BSFL will repeat the task with the same life cycle (Liu *et al*., 2017).

Figure 2.2: The prepupae of BSFL

(Source from mafes, 2015)

### 2.6.2 Nutritional value of BSFL

The larva of BSF is odourless and dry, and it can still dry for storage and friability purposes. People usually feed BSFL to pets, fish, earthworms, and redworms (Veldkamp *et al.*, 2012). Freshly harvested pre-pupae contain 55-65% moisture, a decent amount of crude protein (40-44% dry matter), lipid-rich in omega 3 and omega 6 fatty acids, and crude fiber (7%), among other nutrients, according to nutrient review. Larvae are simpler and less expensive to dehydrate than other fresh by-products because of their high dry matter content (35-45%) (Newton G.L. , 2008). The amount of fat depends on the type of diet and fat content: The recorded values are 15-25% dry matter in poultry manure fed larvae, 28 % in pig manure, 35 % in cow manure, and 42-49% dry matter in waste oil-rich foods fed to larvae. In comparison to housefly maggots, they have less protein and more lipids *(Musca domestica).* The ash content is high but varies, ranging from 11 to 28% dry matter. Calcium and phosphorus are abundant in the larvae (5-8% dry matter) (0.6-1.5% dry matter). New larvae have a high dry matter content, ranging from 35 to 45%, making them more straightforward and less expensive to dehydrate than other fresh by-products. The diet's fatty acid composition affects the larvae's fatty acid composition. Besides, total lipid content rose from 21% to 30% dry matter. Providing a diet of 12 wastes containing beneficial omega-3 fatty acids to BSFL is a way to enrich the final biomass (St-Hilaire, 2007).

### 2.6.3 Demand BSFL in aquaculture

Animal feed is one of the costliest elements of livestock production, and it is also highly harmful to the ecosystem. Based on nutritional value and ability to produce in an environmentally friendly manner, the BSFL is rapidly emerging as a viable fishmeal substitute in aquafeeds. Adult BSFL are can now be found worldwide, and despite their wasp-like nature, they lack any biting or stinging appendages. BSF has the potential to relieve future food and feed insecurity by replacing increasingly expensive protein sources utilized in poultry, aquaculture, and cattle compound diet formulation, such as fish meal and soybean meal (Liu *et al.*, 2017).

BSFL meal has been shown in studies to be able to replace a significant portion of the fish meal used in rainbow trout (*Oncorhynchus mykiss*) diets (Sealey, 2011; St-Hilaire, 2007). In the second decade of the rapid development of the idea of insects being used as food and feed, there has been an increase in a good effect on animals, especially the growth and metabolism of fish black soldiers fly full-fat larval feed (BSFL) on growth performance, physical properties of feed, attractions, and utilisation (Rawski *et al.*, 2021).

### 2.6.4 Potential application of BSFL in animal feed

At this point, BSFL produces good animal feed and is a good protein source. Depending on the food source, BSFL can convert at least 25% of protein and are high in calcium and other nutrients. When defatted, BSFL meal can contain up to 60% protein compared to other insect meal. To substitute the standard commercial feed, feces from BSFL (reared on dried distiller grains) are using in commercial prawn farms. The results were similar to the typical *M. rosenbergii* feed inefficiency, improving the economic returns. The only noticeable difference was that the prawns fed the BSFL feces diet were significantly paler than those fed the standard diet, but the taste was unaffected (Tiu, 2012).

### 2.6.5 Textural Profile Analysis (TPA)

The General Foods Corporation Technical Center first proposed textural profile analysis (TPA) as a tool for characterization and quality control of food products (Trinh, 2012). Texture analysers are primarily used by feed technologists to determine the maximum peak force that causes breakages (hardness) and to estimate tensile stresses. This TPA involves compression of a sample at least twice to simulate the action of jaws (A. F. Haubjerg *et al*., 2015).

# CHAPTER 3

# METHODOLOGY

## 3.1 Defatting process of Black Soldier Fly Larvae

About 2 kg of Black soldier fly larvae (BSFL) powder was purchased from a local producer and seller and posted to University Malaysia Kelantan (UMK), Jeli Campus. Then, the amount from 2 kg of BSFL powder was filled inside a filter and extracted for 5 to 6 h using the Soxhlet method that used Soxhlet apparatus (Soxhlet Extraction Glass 250 with heating mantle) The BSFL powder was extracted using 95% ethanol as an extractor. The defatted BSFL powder was dried following the defatting process. The bsfl powder was dried by using drying oven with 60°C for 24 h for moisture and ethanol removed (Ishak *et al*., 2018).

## 3.2 Feed formulation using Winfeed software

Winfeed 2.8 software was used to calculate the amount of other ingredients to create the feed formulation for this experiment. Firstly, select the animal requirement of *M. rosenbergii* juvenile before starting the formulation. The nutrient requirement and limits for each nutrient were fill in manually the main window to begin the new formulation. The Winfeed software created the excellent nutrient requirement in feed formulation following the specific requirement for targeted species.

Table 3.1: Composition of the experiment diet (BSFL) for juvenile *M. rosenbergii.*

| Ingredients | Diet I<br>Control | Diet II<br>10% BSFL | Diet III<br>20% BSFL | Diet IV<br>30% BSFL | Diet V<br>40% BSFL |
|---|---|---|---|---|---|
| BSFL | 0 | 10 | 20 | 30 | 40 |
| Fish meal | 16.75 | 14.25 | 11.75 | 9.25 | 6.75 |
| Corn meal | 16.75 | 14.25 | 11.75 | 9.25 | 6.75 |
| Rice bran | 16.75 | 14.25 | 11.75 | 9.25 | 6.75 |
| Soybean meal | 16.75 | 14.25 | 11.75 | 9.25 | 6.75 |
| Copra meal | 15 | 15 | 15 | 15 | 15 |
| Tapioca flour | 15 | 15 | 15 | 15 | 15 |
| Mineral premix | 1 | 1 | 1 | 1 | 1 |
| Vitamin C | 1 | 1 | 1 | 1 | 1 |
| Palm oil | 1 | 1 | 1 | 1 | 1 |
| Total | 100 | 100 | 100 | 100 | 100 |

Based on the Table 3.1, the formulation for control, the diet I was prepared without BSFL, where fish meal, cornmeal, rice bran, and soybean meal were fixed at 16.75 g The total weight for formulation diet II, 10% of BSFL, is 10 g of BSFL. For fish, corn, rice bran, and soybean meal are 14.45 g for each. The total weight for formulation 20% of BSFL, which is diet III, is 20 g of BSFL. For fish, corn, rice bran, and soybean meal are 11.75 g for each. Next, the total weight for formulation diet IV is 30% of BSFL is 30 g of BSFL. The fish meal, cornmeal, rice bran, and soybean meal are 9.25 g each. The total weight of diet V formulation is 40% of BSFL is 40 g of BSFL. Then, 6.75 g for each ingredient of fish, corn, rice bran, and soybean meals. Copra meal and tapioca flour were 15 g for each and 1 g of each mineral premix, vitamin C, and palm oil for every diet percent of BSFL. Every diet experiment was mixed for pelletizing.

## 3.3 Pellet producing

The various raw ingredients were mixed and appropriately crushed by using blender (Smart Power Machine Blender 2.5L) to become powder to ensure that the powder bonds correctly. Formula for water volume needed:

$$\text{Volume } H_2O = \frac{[(mf)(wi) - (mi)(wi)]}{100 - mf}$$

While;

mf = final moisture (%)

wi = initial weight (g)

mi=initial moisture (g)

Table 3.2: Water volume needed for all sample test diet

| Test diet | Water volume (g) |
|---|---|
| Diet I | 26 |
| Diet II | 25.5 |
| Diet III | 25.8 |
| Diet IV | 26.7 |
| Diet V | 27.3 |

The table shows the total of volume of water was added for each diet. Each raw material was blended for the pelletizing step to obtain a fine texture. Then, all the ingredients are mixed and blended well before forming pellets. The formation of these prawn pellets was done using the hands to create a small circle that fits the prawn mouth. So, since the formation of pellets are done manually, the pellet size should follow the mouth size of juvenile *M. rosenbergii*. The ready-formed pellets are dried in the sun. The sample of the pellet was kept at room temperature for analysis later.

## 3.4 Physical properties in formulated pellet

### 3.4.1 Colour analysis

Colour analysis was done with CR-400 Chroma Meter. The sample from Diet I, Diet II, Diet III, Diet IV, and Diet V was inserted into a small zipper plastic bag size (8 x 12cm) for the test. The chromameter sensor was placed directly on the sample and the measurement button is pressed twice to obtain the result. The outcomes are shown on a chromameter LCD. CIEBLAB colour space used for the identification of colour for each diet. Data were collected, and the procedure was repeated twice for each sample to ensure accuracy.

### 3.4.2 Bulk density

The weight of the 5mL measuring cylinder was obtained by weighing using an analytic balance. Each diet pellet was placed in a measuring cylinder until it reached a volume of 5 mL of the cylinder. After that, measuring the cylinder containing the pellets was weighed again to get the measuring cylinder with the pellet sample. The bottom of the measuring cylinder was then gently tapped on a benchtop more than ten times, from 8-10 cm height or until the constant volume was obtained (Jannathulla *et al*., 2019). Formula on calculating bulk density was given below:

$$Bulk\ density = \frac{W2 - W1}{V}$$

Where, W1 = weight of empty measuring cylinder (g),

W2= weight of measuring cylinder with sample (g),

V= volume of sample after tab (mL)

### 3.4.3 Sinking velocity

About 50 mL of distilled water were poured into a 50 mL measuring cylinder. The height of the distilled water was then measured. A pinch of the sample was dropped into the water, and the timer began as soon as the sample began to sink. When the first two particles reached the bottom of the measuring cylinder, the timer stopped. The amount of time spent was recorded. The step was repeated and replaced with small pieces of pellet control and 10%, 20%, 30% and 40% of BSFL pellet. The sinking velocity was calculated using the formula:

$$Sinking\ velocity = \frac{t}{h}$$

Where, t = time taken for the sample to reach the bottom (s)

h = height of the water (cm)

## 3.5 Textural Profile Analysis (TPA) in formulated pellet

Before withdrawing from the sample, the cylindrical probe moves towards it at a predetermined speed until a predetermined compression is reached. The force applied to the load cell is periodically recorded and analysed graphically. By using Brookfield CT3 texture analyser, a cylinder probe are used as load cell. The pellet was placed at the platform. The test diet I, II, III, IV and V were measured. Data of the hardness, cohesiveness and springiness was recorded.

### 3.6 Data collection and analysis

All the data obtained from the study were analysed using one-way ANOVA in Statistical Package for the Social Sciences (SPSS) version 26 to determine the significant difference between the variation result of percentage of BSFL in physical properties and textural profile analysis *M. rosenbergii* juvenile pellet. All the data was analysed with 3 replicates for each test diet and the significant difference of ($P < 0.05$) were determined.

# CHAPTER 4

# RESULT & DISCUSSION

## 4.1 Physical Properties of Feed

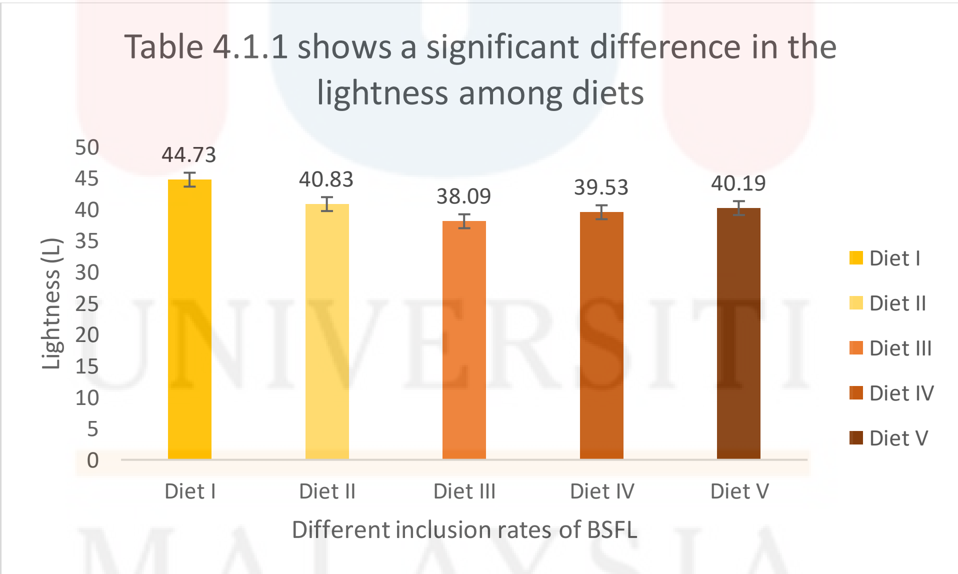
### 4.1.1 Colour analysis

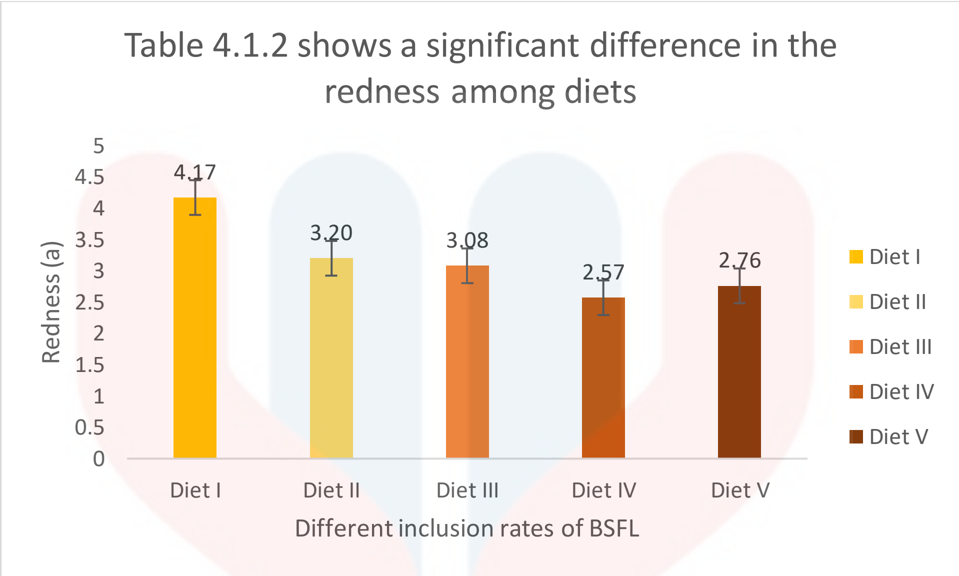
All sample formulations in diet studies were conducted to colour analysis tests. Each pellet sample from Diet I, Diet II, Diet III, Diet IV, and Diet V in a plastic size (8 x 12cm) was determined using a CR-400 Chroma Meter. To achieve the result, the chromameter sensor was placed directly on the sample and the measurement button was pressed twice. The results are displayed on a chromameter LCD. The chromameter LCD provides three data points in screen which is lightness (L), redness (a), and yellowness (Y) (b). Colour analysis test reveals significant differences between the experimental diets ($P<0.05$).

Table 4.1: Colour measurement of samples

| Test diet | Colour measurement of samples (Parameter) | | |
|---|---|---|---|
| | Lightness (L) | Redness (a) | Yellowness (b) |
| Diet I (Control) | $44.73 \pm 0.05^{e}$ | $4.17 \pm 0.03^{e}$ | $7.47 \pm 0.02^{d}$ |
| Diet II (BSFL 10%) | $40.83 \pm 0.02^{d}$ | $3.20 \pm 0.02^{d}$ | $6.60 \pm 0.05^{c}$ |
| Diet III (BSFL 20%) | $38.09 \pm 0.02^{a}$ | $3.08 \pm 0.01^{c}$ | $6.11 \pm 0.05^{b}$ |
| Diet IV (BSFL 30%) | $39.53 \pm 0.02^{b}$ | $2.57 \pm 0.02^{a}$ | $5.17 \pm 0.02^{a}$ |
| Diet V (BSFL 40%) | $40.19 \pm 0.01^{c}$ | $2.76 \pm 0.03^{b}$ | $5.78 \pm 0.15^{b}$ |

Mean within column with different letter(s) indicate significance difference between treatments by Tukey's HSD test at $P \leq 0.05$. Columns represent the mean values ± standard error





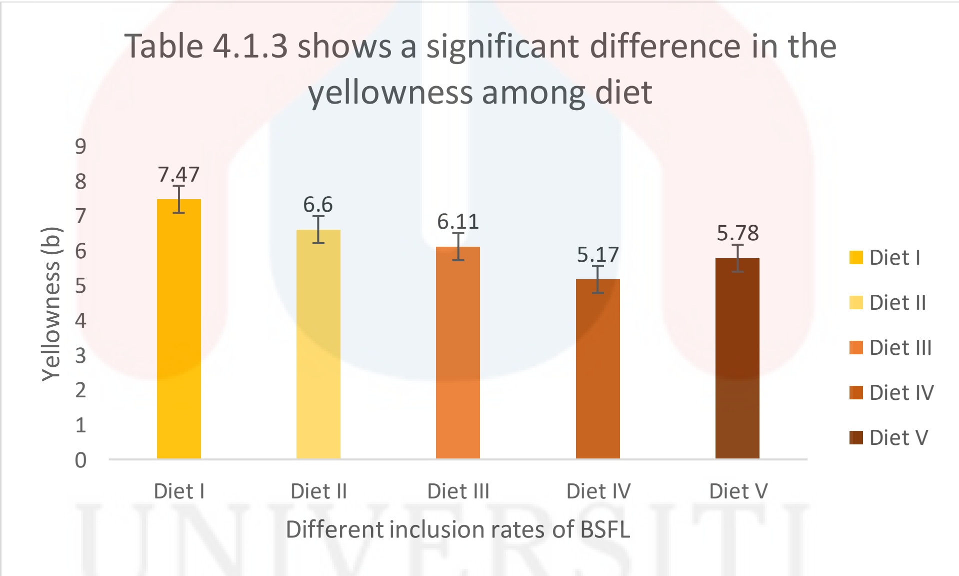




Figure 4.1: Colour Characteristic of BSFL Test Diet with CIE *L*a*b* scale*

Based on Table 4.1, the lightness of the diet I is significant (P <0.005) higher than other test diet formulations. This indicates that the sample has a light or brighter colour and was given a high lightness value. This is because the control does not have BSFL content that may affect the lightness value of the control sample if added. Therefore, the lightness value will be high if the sample is brightly coloured. For the diet test with the addition of BSFL, diet III showed the lowest lightness value. This shows that the BSFL sample of 20% has a darker colour compared to other samples which is a value of 38.09 ± 0.02. However, diet IV which is BSFL 30% shows a lightness value that is not too much different which is i.e., slightly higher with a lightness value of diet III where BSFL 20%. In the sample formulation of diet II which is BSFL 10% showed a slightly higher of lightness value compared to the lightness value of diet V (BSFL 40%) where the possibility of a small percentage content of BSFL on diet II which is BSFL 10%. Probably due to factors when making pellets such as technical errors during mixing or during pellet formation.

Next, the redness colour value showed a slightly significant decrease from the control sample to BSFL 30% for the redness colour. However, the value of redness colour in the BSFL 40% sample showed a slight increase of 2.76 ± 0.03. As for the yellowness value also showed a slightly significant decrease from the control sample to BSFL 30%. In the 40% BSFL sample, the yellowness value increased slightly at 5.78 ± 0.15. Meyers and Hagood (1984) discovered that *M. rosenbergii* larvae preferred light-coloured feed flakes over darker-coloured flakes (Kawamura *et al.*, 2016). There are several possible factors where the values of redness and yellowness at low measurements are due to the presence of dark-coloured BSFL as taken from the prepupae stage. Therefore, the determination for suitable prawn feed requires determination through feeding trial.

### 4.2.1 Bulk Density

Bulk density tests were performed on all sample formulation BSFL test diets, including controls. Using a 5ml measuring cylinder, a few pellets are inserted into it to meet the volume of the measuring cylinder. After that, the measuring cylinder is tapped several times until a compact and constant volume is taken. Sample pellet from Diet I, Diet II, Diet II, Diet IV and Diet V is not significantly different in bulk density except for diet with 40% BSFL. The bulk density of all samples is shown in Table 4.2.

Table 4.2: Bulk density of all samples

| Test diet | Bulk density |
|---|---|
| Diet I (Control) | $0.40 \pm 0.00^{b}$ |
| Diet II (BSFL 10%) | $0.38 \pm 0.01^{b}$ |
| Diet III (BSFL 20%) | $0.37 \pm 0.00^{b}$ |
| Diet IV (BSFL 30%) | $0.40 \pm 0.01^{b}$ |
| Diet V (BSFL 40%) | $0.32 \pm 0.01^{a}$ |

Mean within column with different letter(s) indicate significance difference between treatments by Tukey's HSD test at $P \leq 0.05$. Columns represent the mean values ± standard error

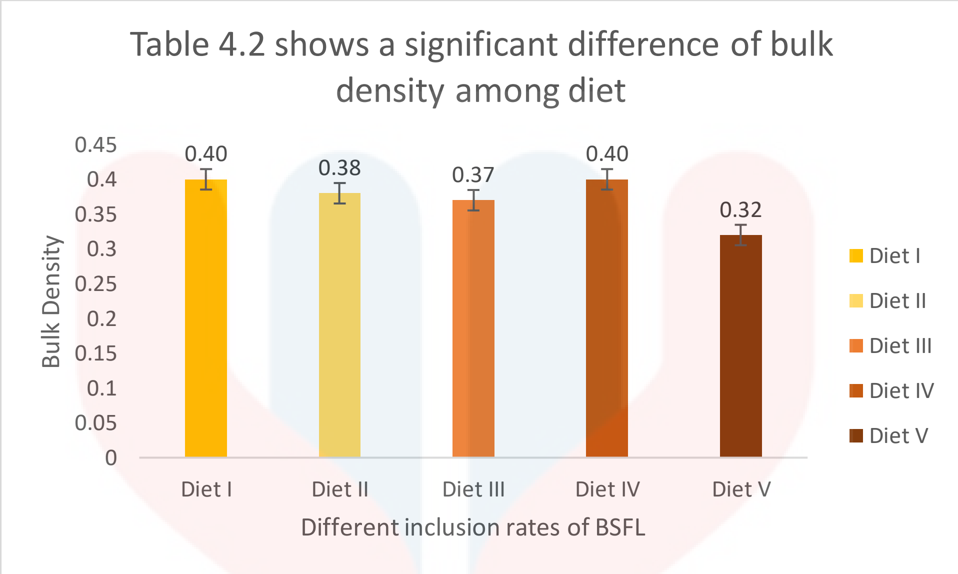



Figure 4.2: Bulk density of BSFL formulation test diet.

The results shown in Table 4.2 highlight that the bulk density of the sample formulation test diet of diet I which is control and diet IV which is BSFL 30% had the same high amount indicating significant ($P < 0.05$) i.e., 0.40 ± 0.00 g/mL. The lowest bulk density value was determined by a diet V that refer 40% BSFL sample of 0.32 ± 0.01 g/mL. Which of the diet II (BSFL 10%) and diet III (BSFL 20%) formulation samples showed a small difference with values of 0.38 ± 0.01 and 0.37 ± 0.00. The possibility of unrestricted size affects different mean bulk density values. Furthermore, research have shown that the bulk density of shrimp feed pellets increases as particle size increases (Tanveer *et al.*, 2018). According to Khater et al., 2002, the results show that the average weight and bulk density of fish feed pellets increases with increasing pellet size and protein ratio. It shows that as the pellet size increases from a slight volume to a larger one, the average weight of the pellets and the bulk density will increase. The increasing dry bulk density causes solid particles to fill water and air voids, leading to a reduction in moisture content (Mouazen *et al.*, 2002). The physical properties of the pellets shall be high to withstand during pneumatic handling, transport, and delivery, without the generation of excessive amounts of dust and fine particles (Khater *et al*., 2014).

### 4.2.3 Sinking Velocity

Sinking velocity tests were performed on all BSFL test diet sample compositions. This test determines how long (times) each sample composition takes to sink and reach the bottom (base). The sinking velocity of all samples is shown in Table 4.8.

Table 4.3: Sinking velocity of all samples

| Test diet | Sinking velocity |
|---|---|
| Diet I (Control) | $0.30 \pm 0.02^{a}$ |
| Diet II (BSFL 10%) | $0.38 \pm 0.02^{b}$ |
| Diet III (BSFL 20%) | $0.29 \pm 0.01^{a}$ |
| Diet IV (BSFL 30%) | $0.29 \pm 0.01^{a}$ |
| Diet V (BSFL 40%) | $0.33 \pm 0.01^{ab}$ |

$^{ab}$ means with different superscripts in a row is significantly different ($P<0.05$)

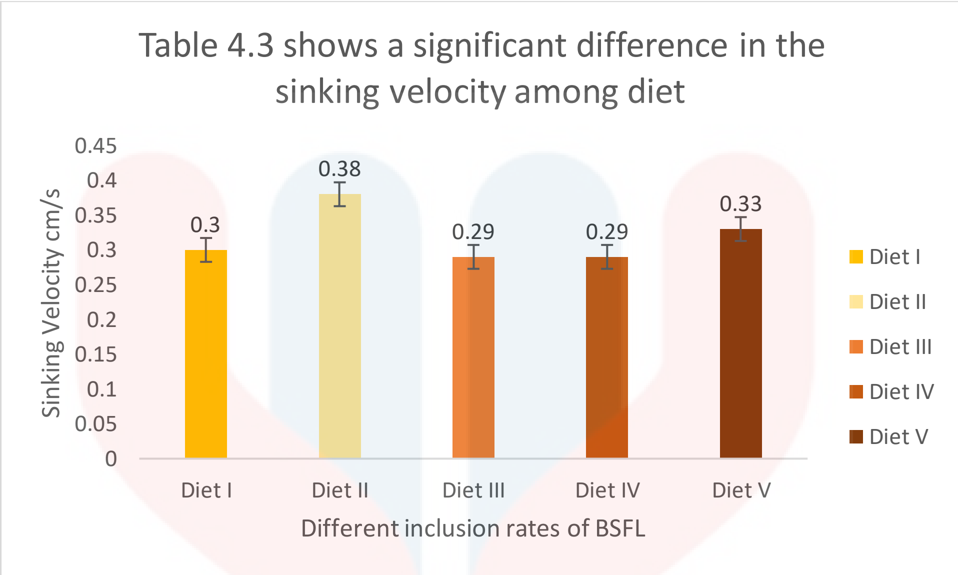



Figure 4.3: Sinking velocity of BSFL formulation test diet

In the Table 4.3 shown that all sample for formulation test diet are significant (P <0.05) including control and BSFL added. Diet III which is BSFL 20% and diet IV which is BSFL 30% addition formulation had the lower sinking velocity rate of 0.29 ± 0.01 s/cm which mean the pellet quickly sinks. Meanwhile, the diet II refers to BSFL 10% formulation yielded the higher sinking velocity rate of 0.38 ± 0.02 s/cm are mean the pellet slowly sinks. However, diet V which is BSFL 40% showed a sinking velocity rate of 0.33 ± 0.01. The higher the sinking velocity, the longer it takes for the sample to sink, and the lower the sinking velocity, the faster it takes There are several possibilities that cause fast velocity shrinkage rates such as pellet compression space and the size of manually made feed pellets giving slightly different sizes. Because prawn are bottom feeders, they require a submersible feed that is water stable. The speed of sinking is an important statistic in prawn nutrition, as it indicates how rapidly feed pellets drop to the pond's bottom (Jannathulla *et al.*, 2019). This is because, there was one study found that after juveniles experienced a random touch of pellets and took them with maxillipeds, and the resulting satiety, they apparently changed their foraging behaviour by bowing their heads, actively swimming forward, sweeping the bottom with maxillipeds, tracking pellets, grabbing them, and swallow it (Kawamura *et al.*, 2017). Therefore, samples with a fast time taken of sinking velocity rate are very suitable for shrimp feed fore juvenile.

## 4.2 Textural Profile Analysis of Feed

The textural profile analysis of treatment BSFL pellet reported in Table 4.2. the TPA (hardness, cohesiveness, and springiness) shown significantly different ($P < 0.05$) in all BSFL test diets.

### 4.2.1 Hardness

The hardness test was conduct to all sample diet formulation including control and bsfl added. Table 4.4 shows the hardness of the test diet.

Table 4.4: Texture profile analysis of hardness of BSFL formulation

| Parameter | Test diet | | | | |
|---|---|---|---|---|---|
| | Diet I (Control) | Diet II (BSFL 10%) | Diet III (BSFL 20%) | Diet IV (BSFL 30%) | Diet V (BSFL 40%) |
| Hardness | 652.33 ±22.26$^{a}$ | 885.67 ±32.37$^{b}$ | 1261.00 ±32.01$^{c}$ | 1826.33 ±16.60$^{d}$ | 1339.33 ±17.46$^{c}$ |

Mean within column with different letter(s) indicate significance difference between treatments by Tukey's HSD test at $P \leq 0.05$. Columns represent the mean values ± standard error

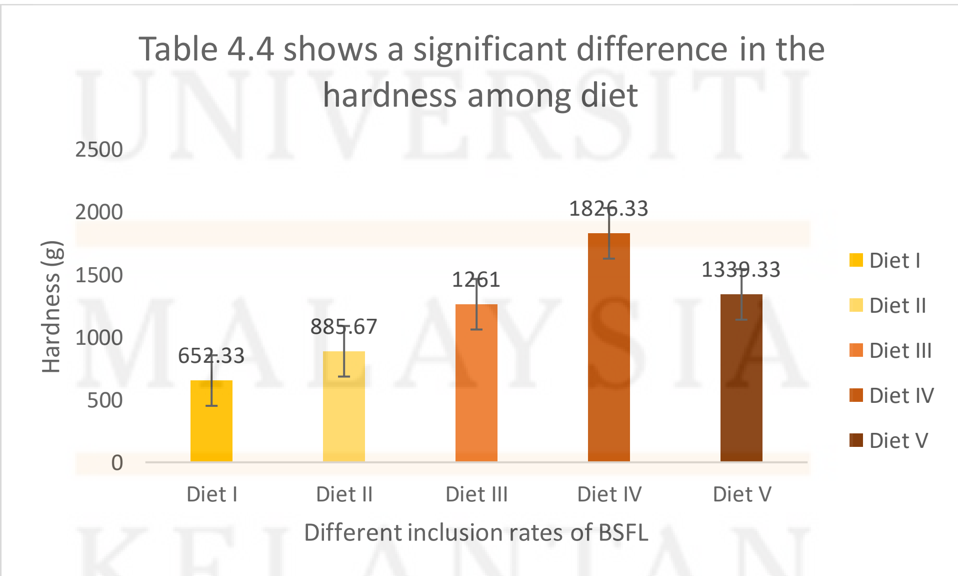



Figure 4.4: Hardness in TPA of BSFL formulation

The textural profile analysis for hardness has been attached at the Table 4.4. The hardness was shown statistically significant (P <0.05) to all sample of test diet formulation included control and different percentage of BSFL. Diet IV which is BSFL 30% shows the most hardness which is 1261.00 ± 32.01. Meanwhile, the diet I that are control without the addition of BSFL had less hardness of 652.33 ± 22.26. However, the hardness rates at diet II that is BSFL 10% and diet III (BSFL 20%) i.e., 885.67 ± 32.37 and 1261.00 ± 32.01 were increasing which means getting harder due to the addition of BSFL in the diet test for shrimp. However, the hardness rate for diet V which is BSFL 40% which is 1339.33 ± 17.46 is slightly decreased but still at a flexible hardness level. During the pressing process on the pellets, it appears that when diet I which is the control pellet is not too crushed from the first compression as well as the crushed BSFL mixture pellets produce few dust particles. Probably due to the presence of tapioca flour as a binding reagent that gives the texture characteristics are not too crushed.

In addition, pellets with good hardness can reduce the fraction that will produce powder or dust. It is very unsuitable and will be detrimental to farmers if the resulting pellets do not get the right hardness. In this experiment it was found that the formulation of diet IV which is BSFL 30% diet test has a good value for hardness of 1826.33 ± 16.60. With the research of the acceptance level of *M. rosenbergii* juvenile, a feeding trial is proposed in the future to decide which are suited for feeding *M. rosenbergii* juvenile.

### 4.2.2 Cohesiveness

The cohesiveness test was conduct by using TPA to all sample diet formulation including control and BSFL added. Table 4.5 shows the cohesiveness value of the test diet.

Table 4.5: Texture profile analysis of cohesiveness of BSFL formulation

| Parameter | Test diet | | | | |
|---|---|---|---|---|---|
| | Diet I (Control) | Diet II (BSFL 10%) | Diet III (BSFL 20%) | Diet IV (BSFL 30%) | Diet V (BSFL 40%) |
| Cohesiveness | 0.78 ±0.02[c] | 0.40 ±0.03[b] | 0.44 ±0.01[b] | 0.26 ±0.01[a] | 0.24 ±0.01[a] |

Mean within column with different letter(s) indicate significance difference between treatments by Tukey's HSD test at $P \leq 0.05$. Columns represent the mean values ± standard error

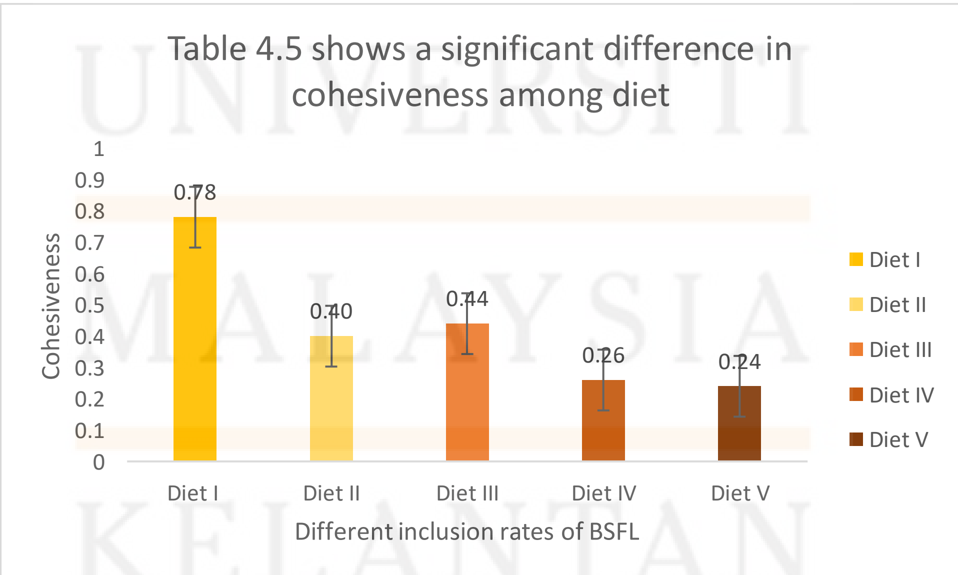



Figure 4.5: Cohesiveness in TPA of BSFL formulation

In the Table 4.5, the result of cohesiveness shown significant ($P < 0.05$) to all sample of test diet formulation. Diet I which is control had the highest of cohesiveness compared to all diet II, III, IV and V from other percentages BSFL formulation which is 0.78 ± 0.02. The lowest cohesiveness is referred to sample from formulation diet V which is BSFL 40% which is 0.24 ± 0.01 and followed by diet IV that is BSFL 30% which is slightly higher than diet V (BSFL 40%) in value i.e., 0.26 ± 0.01. Other formulation such formulation from diet II which is BSFL 10% (0.40 ± 0.03) has a slightly less cohesiveness value with the formulation of diet III which is BSFL 20% i.e., 0.44 ± 0.01. This indicates pellet control shows the sample are more cohesiveness compared to other sample formulations diet. This is because the study found that high-durability pellets can be explained from high cohesion and low elasticity, as measured respectively from landfill tests on fish pellets (Haubjerg *et al.*, 2015). That means the higher the temperature during drying, the higher value cohesiveness. There are possible factors during the pellet drying process that cause the pellet cohesiveness value to differ where the pellet drying process only depends on the sun naturally. In the pelletizing process, steam and heat can cause the starch to gelatinize, that resulting in cohesiveness, which can aid in the binding of different feed ingredients. As a result, the higher the starch content, the better the cohesion. The cohesiveness of extracts from various sources varies. Wheat and barley starch has a higher starch integrity than corn. Soybean meal has good cohesiveness due to its low-fat content.

### 4.2.3 Springiness

The TPA parameter which is springiness were test on all sample diet formulation including control and bsfl added. Table 4.6 shows the springiness of the test diet.

Table 4.6: Texture profile analysis of springiness of BSFL formulation

| Parameter | Test diet | | | | |
|---|---|---|---|---|---|
| | Diet I (Control) | Diet II (BSFL10%) | Diet III (BSFL 20%) | Diet IV (BSFL 30%) | Diet V (BSFL40%) |
| Springiness | 0.15 ±0.02[a] | 0.26 ±0.01[b] | 0.24 ±0.01[b] | 0.23 ±0.01[b] | 0.21 ±0.01[ab] |

[ab] means with different superscripts in a row is significantly different ($P<0.05$)

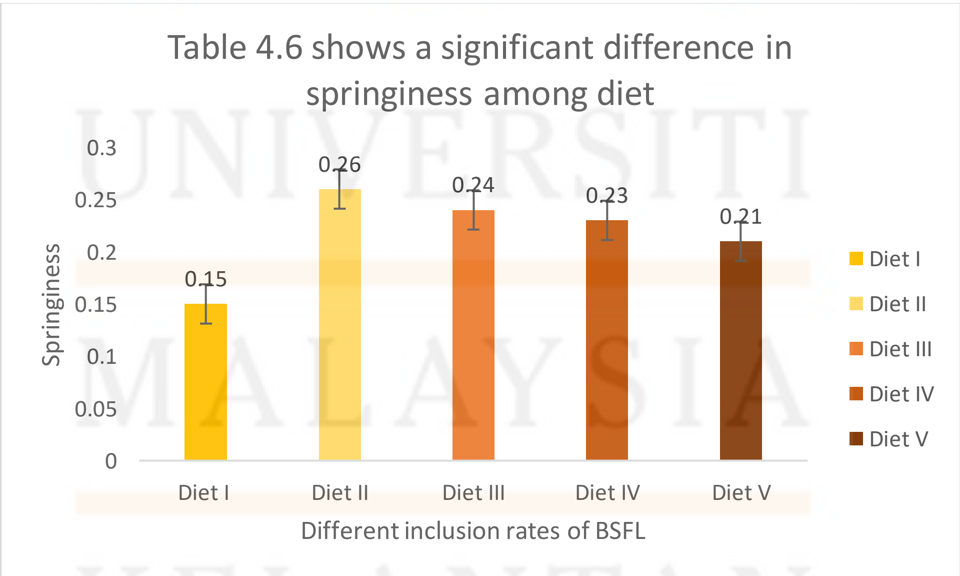



Figure 4.6: Springiness in TPA of BSFL formulation

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Based on the Table 4.6, the result of parameter springiness shown significant ($P < 0.05$) to all sample formulation of test diet for *M. rosenbergii*. Formulation of diet II which is BSFL 10% has the highest springiness which is 0.26 ± 0.01 compared than another sample. Meanwhile the lowest springiness is referred to formulation control which is diet I where there is no addition of BSFL material which is 0.15 ± 0.02. In addition, the formulation of BSFL 20% (diet III) at a value of 0.24 ± 0.01 followed by the formulation of test diet BSFL 30% (diet IV) which is 0.23 ± 0.01 and finally the formulation of BSFL 40% (diet V) with a value of 0.21 ± 0.01. This indicates that at Table 4.6 starting BSFL formulation 20% which is diet III, springiness rate decreases slightly. Springiness is a texture parameter, which is related to the elasticity of the sample. Springiness in the TPA is related to the height of food recovered during the elapsed time during the end of the first bite and the beginning of the second bite. If the redness is high, it requires more chewing energy in the mouth (Shafiur Rahman, 2009). This simplifies the process of shrimp digestion when the shrimp chews pellets in small particles appropriate to the size of its mouth.

# CHAPTER 5

# CONCLUSION

## 5.1 Conclusion

The current study reveals that *Hermetia illucens,* also known as Black Soldier Fly Larvae (BSFL), can be utilised as a partial fishmeal replacement to enhance the growth and productivity of farmed prawns. Where BSFL's ability to produce protein-rich edible biomass from potentially poor protein from organic waste containing BSFL is acceptable for contribution as a partial or entire food substitute for sustainable aquaculture, including invertebrate (Wang, 2017). This is because insect meal has shown to be beneficial in other crustacean diets. In addition, bsfl can be utilised as an alternate protein element in aquaculture diets to replace fishmeal, which is limited.

Physical and textural profile analysis is not very helpful to analyse the experimental test diet on the resulting sample. None of research included a physical properties analysis of the feed or clearly good findings from the use of BSFL. Because there is a lack of information available about the physical and mechanical qualities of feed pellets, which are crucial for understanding the product's behaviour during the processing, transportation, packing, storage, and feeding processes. However, the diet test with BSFL 20% and BSFL 30% addition formulation had the lower sinking velocity rate of 0.29 ± 0.01 s/cm which showed very suitable for juvenile stage *M. rosenbergii* nutrition.

## 5.2 Recommendation

There are some suggestions that can be observed and can be improved where the pellet size should be uniform and follow the different size of the shrimp mouth at each stage. This is because it will affect the feed conversion ratio when the shrimp consume pellets either in small or large quantities depending on the size of the shrimp's mouth. It is highly recommended to form pellets with a suitable pellet machine to get a uniform pellet size and not easily crushed.

Furthermore, a feeding trial for each test diet with BSFL mixture should be conducted to determine the acceptance rate of juvenile *M. rosenbergii* for growth, survivability, and mortality. Furthermore, when the feeding trial is conducted, can also observe the effect of the studied pellets on water quality that may affect the pH, dissolved oxygen, and salinity of juvenile *M. rosenbergii.* Due to the addition of BSFL in pellets as a substitute protein source for the growth of juvenile *M. rosenbergii*.

Prawn feed storage also plays an important role in preventing any damage and preventing the presence of pests. In addition, the storage temperature of *M. rosenbergii* feed should be in good condition with proper ventilation to prevent the growth of microorganisms such as fungi and bacteria.

## REFERENCES


Barragan-Fonseca, K. B., Dicke, M., & van Loon, J. J. (2017). Nutritional value of the black soldier fly (Hermetia illucens L.) and its suitability as animal feed–a review. *Journal of Insects as Food and Feed*, *3*(2), 105-120.

DOF (2020). *Aquaculture Production From Freshwater Culture System By Species*, 2020. Department of Fisheries. https://www.dof.gov.my/sumber/perangkaan-perikanan-i/

FAO. (2018). *The State of World Fisheries and Aquaculture 2018*. Food and Agriculture Organization of the United Nations. https://www.fao.org/documents/card/en/c/I9540EN/

Haubjerg, A. F., Veje, C. T., Jørgensen, B. N., & Simonsen, B. (2015). Experimental investigation of physical properties and mechanical durability of extruded fish feed in the drying process. Proceedings of 5th European Drying Conference (EuroDrying'2015), Budapest, Hungary,

Hawkinson, C. (2005). *Beneficial in the garden: Black Soldier Fly*. Master Gardener Laura Bellmore. Retrieved 15 January 2022 from

Ishak, S., Kamari, A., Yusoff, S., & Halim, A. (2018). Optimisation of biodiesel production of Black Soldier Fly larvae rearing on restaurant kitchen waste. Journal of Physics: Conference Series,

Jannathulla, R., Ambasankar, K., Syama Dayal, J., Rajamohmed, M., Sravanthi, O., & Rajesh, B. (2019). Studies on functional properties and chemical composition of commercial shrimp feeds used in Andhra Pradesh, India.

Kawamura, G., Bagarinao, T., Yong, A. S. K., Jeganathan, I. M. X., & Lim, L.-S. (2016). Colour preference and colour vision of the larvae of the giant freshwater prawn Macrobrachium rosenbergii. *Journal of Experimental Marine Biology and Ecology*, *474*, 67-72.

Kawamura, G., Bagarinao, T. U., & Yong, A. S. K. (2017). Sensory systems and feeding behaviour of the giant freshwater prawn, Macrobrachium rosenbergii, and the marine whiteleg shrimp, Litopenaeus vannamei. *Borneo Journal of Marine Science and Aquaculture (BJoMSA)*.

Kawamura, G., Bagarinao, T. U., Seniman, N. S., Yong, A. S. K., & Lim, L. S. (2018). Comparative morphology and function of feeding appendages in food intake behaviour of the whiteleg shrimp, Litopenaeus vannamei, and the giant freshwater prawn, Macrobrachium rosenbergii. . *Borneo Journal of Marine Science and Aquaculture, 2(1).*

Khater, E.-S. G., Bahnasawy, A. H., & Ali, S. A. (2014). Physical and mechanical properties of fish feed pellets. *Journal of Food Processing & Technology, 5*(10), 1.

Larry Newton, C. S., D Wes Watson, Gary Burtle, Robert Dove. (2005, January 2005). Using the Black Soldier Fly, Hermetia Illucens, as a Value-Added Tool for the Management of Swine Manure. 17.

Ling, S. W. (1969). *THE GENERAL BIOLOGY AND DEVELOPMENT OF Macrobrachium rosenbergii (DE MAN), Vol.3*(FAO Fisheries Report, 57), 589-606.

Liu, X., Chen, X., Wang, H., Yang, Q., ur Rehman, K., Li, W., Cai, M., Li, Q., Mazza, L., & Zhang, J. (2017). Dynamic changes of nutrient composition throughout the entire life cycle of black soldier fly. *PLoS One*, *12*(8), e0182601.

Mitra, G., Mukhopadhyay, P. K., & Chattopadhyay, D. N. (2005). Nutrition and feeding in freshwater prawn (Macrobrachium rosenbergii) farming. *Aqua feeds: formulation and Beyond, 2(1)*, 17-19.

Mouazen, A. M., Ramon, H., & De Baerdemaeker, J. (2002). SW—Soil and water: Effects of bulk density and moisture content on selected mechanical properties of sandy loam soil. *Biosystems Engineering*, *83*(2), 217-224.

Mouithys-Mickalad, A., Schmitt, E., Dalim, M., Franck, T., Tome, N. M., van Spankeren, M., Serteyn, D., & Paul, A. (2020). Black soldier fly (Hermetia illucens) larvae protein derivatives: potential to promote animal health. *Animals, 10*(6), 941.

Nesara, K. M., & Paturi, A. P. (2018). Nutritional requirement of fresh water prawn and shrimps: A review". *J. Entom. and Zoo. Stud 6*, .( no. 4 ), : 1526-1532.

New, M. B. (2002). *FARMING FRESHWATER PRAWNS: A manual for the culture of the giant river prawn(Macrobrachium rosenbergii)*. FAO Fisheries Technical Paper,.

New, M. B. V., Wagner Cotroni. (2000). In: New MB, Valenti WC, editors. Freshwater Prawn Culture: The Farming of Macrobrachium rosenbergii., pp. 1–10. (Blackwell Science; London:)

Newton G.L. , S. D. C., and Burtle G.J. . (2008). Research Briefs: Black soldier fly prepupae: A compelling alternative to fish meal and fish oil. . *Aquaculture, 4*, 103-109.

Rawski, M., Mazurkiewicz, J., Kierończyk, B., & Józefiak, D. (2021). Black Soldier Fly Full-Fat Larvae Meal Is More Profitable Than Fish Meal and Fish Oil in Siberian Sturgeon Farming: The Effects on Aquaculture Sustainability, Economy and Fish GIT Development. *Animals, 11*(3), 604.

Sarman, V., Vishal, R., Mahavadiya, D., & Sapra, D. (2011). Nutritional aspect for freshwater prawn (Macrobrachium rosenbergii) farming. *culture (Ha), 12*, 2012-2013.

Sealey, W. M., Gaylord, T. G., Barrows, F. T., Tomberlin, J. K., McGuire, M. A., Ross, C., & St-Hilaire, S. . (2011). Sensory analysis of rainbow trout, Oncorhynchus mykiss, fed enriched black soldier fly prepupae, Hermetia illucens. *Journal of the world aquaculture society, 42(1)*, 34-45.

Shafiur Rahman, M., & Al-Mahrouqi, A. I. (2009). Instrumental texture profile analysis of gelatin gel extracted from grouper skin and commercial (bovine and porcine) gelatin gels. *International journal of food sciences and nutrition, 60*(sup7), 229-242.

Smetana, S., Palanisamy, M., Mathys, A., & Heinz, V. (2016). Sustainability of insect use for feed and food: Life Cycle Assessment perspective. *Journal of Cleaner Production, 137*, 741-751.

Soesanto, V. (1980). Report of the training course on prawn farming for Asia and the Pacific. 47-49.

St-Hilaire, S., Sheppard, C., Tomberlin, J. K., Irving, S., Newton, L., McGuire, M. A., ... & Sealey, W. . (2007). Fly prepupae as a feedstuff for rainbow trout, Oncorhynchus mykiss. *Journal of the world aquaculture society, 38(1)*, 59-67.

Sundaravadivel, C., Sethuramalingam, T., & Juliet, K. (2015). Studies on the Optimum Dietary Protein Requirement of Three Size Groups of Fresh Water Prawn Macrobrachium idae (Heller). *Journal of Environmental Science, Toxicology and Food Technology, 9*, 38-43.

Tanveer, J. M., Sivakumar, M., Balasubramanian, S., Vikneswaran, M., & Sabanayagam, S. (2018). Analysis of engineering properties of shrimp feed pellets.

Teshima, S. I., Koshio, S., Ishikawa, M., Alam, M. S., & Hernandez Hernandez, L. H. (2006). Protein requirements of the freshwater prawn Macrobrachium rosenbergii evaluated by the factorial method. *Journal of the world aquaculture society, 37*(2), 145-153.

Tiu, L. G. (2012). *Enhancing sustainability of freshwater prawn production in Ohio*. South Cent. Newsl. Fall.

Trinh, K. T., & Glasgow, S. (2012). On the texture profile analysis test. Proceedings of the Chemeca,

Veldkamp T. Van Duinkerken G., v. H. A., C.M.M. Lakemond, C.M.M., Ottevanger E., Bosch G., & Van Boekel (2012). *Insects as a sustainable feed ingredient in pig and poultry diets: a feasibility study*. Wageningen UR Livestock Research.

Wang, Y.-S., & Shelomi, M. (2017). Review of black soldier fly (Hermetia illucens) as animal feed and human food. *Foods*, *6*(10), 91.

Wowor, D., & Ng, P. K. . (2007). The giant freshwater prawns of the Macrobrachium rosenbergii species group (Crustacea: Decapoda: Caridea: Palaemonidae). *The Raffles Bulletin of Zoology*, *55(2)*, 321-336.

# APPENDICES

## APPENDIX A

Figure A.1: Prepupae of Black soldier fly larvae.

Figure A.2: Manual pelletizing.

Figure A.3: Sample of test diet

Figure A.4: Colour characteristics of sample were tested.

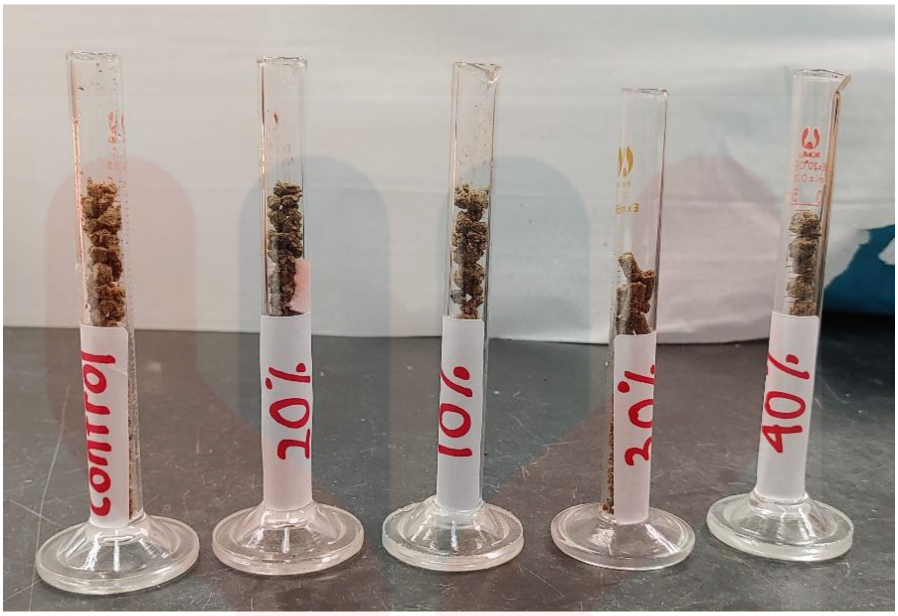


Figure A.5: Bulk density test.

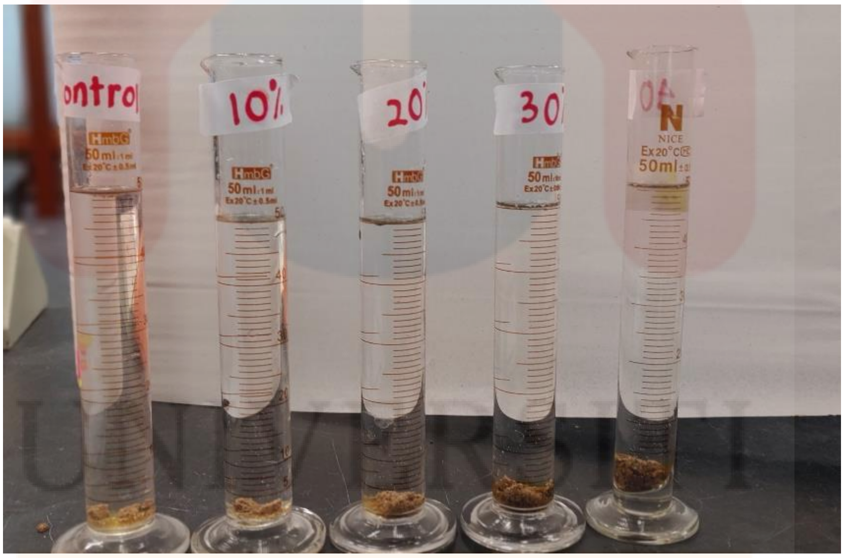


Figure A.6: Sinking velocity test.


Figure A.7: Textural Profile Analysis (TPA) for hardness, cohesiveness, and springiness by using Brookfield CT3 Texture Analyser with cylinder probe.

**APPENDIX B**

Table A.1: Descriptive for one way ANOVA (Colour Analysis)

**Descriptives**

| Color Analysis | | N | Mean | Std. Deviation | Std. Error | 95% Confidence Interval for Mean | | Minimum | Maximum |
|---|---|---|---|---|---|---|---|---|---|
| | | | | | | Lower Bound | Upper Bound | | |
| Color_Lightness | Control | 3 | 44.7267 | .08737 | .05044 | 44.5096 | 44.9437 | 44.63 | 44.80 |
| | BSFL 10% | 3 | 40.8267 | .03215 | .01856 | 40.7468 | 40.9065 | 40.79 | 40.85 |
| | BSFL 20% | 3 | 38.0900 | .04000 | .02309 | 37.9906 | 38.1894 | 38.05 | 38.13 |
| | BSFL 30% | 3 | 39.5267 | .03512 | .02028 | 39.4394 | 39.6139 | 39.49 | 39.56 |
| | BSFL 40% | 3 | 40.1933 | .02517 | .01453 | 40.1308 | 40.2558 | 40.17 | 40.22 |
| | Total | 15 | 40.6727 | 2.29969 | .59378 | 39.3991 | 41.9462 | 38.05 | 44.80 |
| Color_a | Control | 3 | 4.1667 | .04509 | .02603 | 4.0547 | 4.2787 | 4.12 | 4.21 |
| | BSFL 10% | 3 | 3.2000 | .03000 | .01732 | 3.1255 | 3.2745 | 3.17 | 3.23 |
| | BSFL 20% | 3 | 3.0833 | .02082 | .01202 | 3.0316 | 3.1350 | 3.06 | 3.10 |
| | BSFL 30% | 3 | 2.5667 | .03512 | .02028 | 2.4794 | 2.6539 | 2.53 | 2.60 |
| | BSFL 40% | 3 | 2.7567 | .05132 | .02963 | 2.6292 | 2.8841 | 2.70 | 2.80 |
| | Total | 15 | 3.1547 | .57450 | .14834 | 2.8365 | 3.4728 | 2.53 | 4.21 |
| Color_b | Control | 3 | 7.4700 | .03606 | .02082 | 7.3804 | 7.5596 | 7.43 | 7.50 |

| | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|
| | BSFL 10% | 3 | 6.6033 | .09074 | .05239 | 6.3779 | 6.8287 | 6.50 | 6.67 |
| | BSFL 20% | 3 | 6.1067 | .08505 | .04910 | 5.8954 | 6.3179 | 6.01 | 6.17 |
| | BSFL 30% | 3 | 5.1700 | .03000 | .01732 | 5.0955 | 5.2445 | 5.14 | 5.20 |
| | BSFL 40% | 3 | 5.7767 | .26633 | .15377 | 5.1151 | 6.4383 | 5.47 | 5.95 |
| | Total | 15 | 6.2253 | .81244 | .20977 | 5.7754 | 6.6752 | 5.14 | 7.50 |



Table A.2: Descriptive for one way ANOVA (Bulk Density)

**Descriptives**

BulkD

| | N | Mean | Std. Deviation | Std. Error | 95% Confidence Interval for Mean Lower Bound | 95% Confidence Interval for Mean Upper Bound | Minimum | Maximum |
|---|---|---|---|---|---|---|---|---|
| Control | 3 | .3967 | .00577 | .00333 | .3823 | .4110 | .39 | .40 |
| BSFL 10% | 3 | .3800 | .01000 | .00577 | .3552 | .4048 | .37 | .39 |
| BSFL 20% | 3 | .3733 | .00577 | .00333 | .3590 | .3877 | .37 | .38 |
| BSFL 30% | 3 | .3967 | .01528 | .00882 | .3587 | .4346 | .38 | .41 |
| BSFL 40% | 3 | .3233 | .01155 | .00667 | .2946 | .3520 | .31 | .33 |
| Total | 15 | .3740 | .02923 | .00755 | .3578 | .3902 | .31 | .41 |

Table A.3: Descriptive for one way ANOVA (Sinking Velocity)

**Descriptives**

SinkingV

| | N | Mean | Std. Deviation | Std. Error | 95% Confidence Interval for Mean | | Minimum | Maximum |
|---|---|---|---|---|---|---|---|---|
| | | | | | Lower Bound | Upper Bound | | |
| Control | 3 | .3033 | .03055 | .01764 | .2274 | .3792 | .27 | .33 |
| BSFL 10% | 3 | .3800 | .03000 | .01732 | .3055 | .4545 | .35 | .41 |
| BSFL 20% | 3 | .2900 | .01000 | .00577 | .2652 | .3148 | .28 | .30 |
| BSFL 30% | 3 | .2933 | .01528 | .00882 | .2554 | .3313 | .28 | .31 |
| BSFL 40% | 3 | .3267 | .02517 | .01453 | .2642 | .3892 | .30 | .35 |
| Total | 15 | .3187 | .03980 | .01028 | .2966 | .3407 | .27 | .41 |

Table A.4: Descriptive for one way ANOVA (TPA)

**Descriptives**

| | | N | Mean | Std. Deviation | Std. Error | 95% Confidence Interval for Mean Lower Bound | 95% Confidence Interval for Mean Upper Bound | Minimum | Maximum |
|---|---|---|---|---|---|---|---|---|---|
| HARDNESS | Control | 3 | 652.3333 | 38.55299 | 22.25858 | 556.5624 | 748.1043 | 615.00 | 692.00 |
| | BSFL 10% | 3 | 885.6667 | 56.92393 | 32.86504 | 744.2598 | 1027.0735 | 820.00 | 921.00 |
| | BSFL 20% | 3 | 1261.0000 | 55.43465 | 32.00521 | 1123.2927 | 1398.7073 | 1228.00 | 1325.00 |
| | BSFL 30% | 3 | 1826.3333 | 28.74601 | 16.59652 | 1754.9243 | 1897.7424 | 1800.00 | 1857.00 |
| | BSFL 40% | 3 | 1339.3333 | 30.23795 | 17.45789 | 1264.2181 | 1414.4486 | 1305.00 | 1362.00 |
| | Total | 15 | 1192.9333 | 419.25520 | 108.25123 | 960.7575 | 1425.1091 | 615.00 | 1857.00 |
| COHESIVENESS | Control | 3 | .7833 | .04163 | .02404 | .6799 | .8868 | .75 | .83 |
| | BSFL 10% | 3 | .3967 | .05033 | .02906 | .2716 | .5217 | .35 | .45 |
| | BSFL 20% | 3 | .4433 | .02082 | .01202 | .3916 | .4950 | .42 | .46 |
| | BSFL 30% | 3 | .2600 | .01000 | .00577 | .2352 | .2848 | .25 | .27 |
| | BSFL 40% | 3 | .2433 | .01528 | .00882 | .2054 | .2813 | .23 | .26 |
| | Total | 15 | .4253 | .20343 | .05253 | .3127 | .5380 | .23 | .83 |
| SPRINGNESS | Control | 3 | .1567 | .03786 | .02186 | .0626 | .2507 | .13 | .20 |
| | BSFL 10% | 3 | .2633 | .02082 | .01202 | .2116 | .3150 | .24 | .28 |

| | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|
| | BSFL 20% | 3 | .2367 | .01528 | .00882 | .1987 | .2746 | .22 | .25 |
| | BSFL 30% | 3 | .2267 | .02517 | .01453 | .1642 | .2892 | .20 | .25 |
| | BSFL 40% | 3 | .2133 | .01528 | .00882 | .1754 | .2513 | .20 | .23 |
| | Total | 15 | .2193 | .04200 | .01084 | .1961 | .2426 | .13 | .28 |

Table A.5: ANOVA (Colour Analysis)

**ANOVA**

| | | Sum of Squares | df | Mean Square | F | Sig. |
|---|---|---|---|---|---|---|
| Color_Lightness | Between Groups | 74.016 | 4 | 18.504 | 7625.236 | .000 |
| | Within Groups | .024 | 10 | .002 | | |
| | Total | 74.040 | 14 | | | |
| Color_a | Between Groups | 4.606 | 4 | 1.152 | 796.021 | .000 |
| | Within Groups | .014 | 10 | .001 | | |
| | Total | 4.621 | 14 | | | |
| Color_b | Between Groups | 9.064 | 4 | 2.266 | 127.872 | .000 |
| | Within Groups | .177 | 10 | .018 | | |
| | Total | 9.241 | 14 | | | |

Table A.6: ANOVA (Bulk Density)

**ANOVA**

BulkD

| | Sum of Squares | df | Mean Square | F | Sig. |
|---|---|---|---|---|---|
| Between Groups | .011 | 4 | .003 | 25.531 | .000 |
| Within Groups | .001 | 10 | .000 | | |
| Total | .012 | 14 | | | |

Table A.7: ANOVA (Sinking Velocity)

**ANOVA**

SinkingV

| | Sum of Squares | df | Mean Square | F | Sig. |
|---|---|---|---|---|---|
| Between Groups | .017 | 4 | .004 | 7.399 | .005 |
| Within Groups | .006 | 10 | .001 | | |
| Total | .022 | 14 | | | |

Table A.8: ANOVA (Textural Profile Analysis)

**ANOVA**

| | | Sum of Squares | df | Mean Square | F | Sig. |
|---|---|---|---|---|---|---|
| HARDNESS | Between Groups | 2441768.267 | 4 | 610442.067 | 319.927 | .000 |
| | Within Groups | 19080.667 | 10 | 1908.067 | | |
| | Total | 2460848.933 | 14 | | | |
| COHESIVENESS | Between Groups | .569 | 4 | .142 | 141.384 | .000 |
| | Within Groups | .010 | 10 | .001 | | |
| | Total | .579 | 14 | | | |
| SPRINGNESS | Between Groups | .019 | 4 | .005 | 7.904 | .004 |
| | Within Groups | .006 | 10 | .001 | | |
| | Total | .025 | 14 | | | |

Table A.9: Post Hoc Test (Colour Analysis)

**Multiple Comparisons**

Tukey HSD

| Dependent Variable | (I) Group | (J) Group | Mean Difference (I-J) | Std. Error | Sig. | 95% Confidence Interval Lower Bound | 95% Confidence Interval Upper Bound |
|---|---|---|---|---|---|---|---|
| Color_Lightness | Control | BSFL 10% | 3.90000* | .04022 | .000 | 3.7676 | 4.0324 |
| | | BSFL 20% | 6.63667* | .04022 | .000 | 6.5043 | 6.7690 |
| | | BSFL 30% | 5.20000* | .04022 | .000 | 5.0676 | 5.3324 |
| | | BSFL 40% | 4.53333* | .04022 | .000 | 4.4010 | 4.6657 |
| | BSFL 10% | Control | -3.90000* | .04022 | .000 | -4.0324 | -3.7676 |
| | | BSFL 20% | 2.73667* | .04022 | .000 | 2.6043 | 2.8690 |
| | | BSFL 30% | 1.30000* | .04022 | .000 | 1.1676 | 1.4324 |
| | | BSFL 40% | .63333* | .04022 | .000 | .5010 | .7657 |
| | BSFL 20% | Control | -6.63667* | .04022 | .000 | -6.7690 | -6.5043 |
| | | BSFL 10% | -2.73667* | .04022 | .000 | -2.8690 | -2.6043 |
| | | BSFL 30% | -1.43667* | .04022 | .000 | -1.5690 | -1.3043 |
| | | BSFL 40% | -2.10333* | .04022 | .000 | -2.2357 | -1.9710 |
| | BSFL 30% | Control | -5.20000* | .04022 | .000 | -5.3324 | -5.0676 |
| | | BSFL 10% | -1.30000* | .04022 | .000 | -1.4324 | -1.1676 |

| | | | | | | | |
|---|---|---|---|---|---|---|---|
| | | BSFL 20% | 1.43667* | .04022 | .000 | 1.3043 | 1.5690 |
| | | BSFL 40% | -.66667* | .04022 | .000 | -.7990 | -.5343 |
| | BSFL 40% | Control | -4.53333* | .04022 | .000 | -4.6657 | -4.4010 |
| | | BSFL 10% | -.63333* | .04022 | .000 | -.7657 | -.5010 |
| | | BSFL 20% | 2.10333* | .04022 | .000 | 1.9710 | 2.2357 |
| | | BSFL 30% | .66667* | .04022 | .000 | .5343 | .7990 |
| Color_a | Control | BSFL 10% | .96667* | .03106 | .000 | .8645 | 1.0689 |
| | | BSFL 20% | 1.08333* | .03106 | .000 | .9811 | 1.1855 |
| | | BSFL 30% | 1.60000* | .03106 | .000 | 1.4978 | 1.7022 |
| | | BSFL 40% | 1.41000* | .03106 | .000 | 1.3078 | 1.5122 |
| | BSFL 10% | Control | -.96667* | .03106 | .000 | -1.0689 | -.8645 |
| | | BSFL 20% | .11667* | .03106 | .024 | .0145 | .2189 |
| | | BSFL 30% | .63333* | .03106 | .000 | .5311 | .7355 |
| | | BSFL 40% | .44333* | .03106 | .000 | .3411 | .5455 |
| | BSFL 20% | Control | -1.08333* | .03106 | .000 | -1.1855 | -.9811 |
| | | BSFL 10% | -.11667* | .03106 | .024 | -.2189 | -.0145 |
| | | BSFL 30% | .51667* | .03106 | .000 | .4145 | .6189 |

| | | | | | | | |
|---|---|---|---|---|---|---|---|
| | | BSFL 40% | .32667* | .03106 | .000 | .2245 | .4289 |
| | BSFL 30% | Control | -1.60000* | .03106 | .000 | -1.7022 | -1.4978 |
| | | BSFL 10% | -.63333* | .03106 | .000 | -.7355 | -.5311 |
| | | BSFL 20% | -.51667* | .03106 | .000 | -.6189 | -.4145 |
| | | BSFL 40% | -.19000* | .03106 | .001 | -.2922 | -.0878 |
| | BSFL 40% | Control | -1.41000* | .03106 | .000 | -1.5122 | -1.3078 |
| | | BSFL 10% | -.44333* | .03106 | .000 | -.5455 | -.3411 |
| | | BSFL 20% | -.32667* | .03106 | .000 | -.4289 | -.2245 |
| | | BSFL 30% | .19000* | .03106 | .001 | .0878 | .2922 |
| Color_b | Control | BSFL 10% | .86667* | .10869 | .000 | .5090 | 1.2244 |
| | | BSFL 20% | 1.36333* | .10869 | .000 | 1.0056 | 1.7210 |
| | | BSFL 30% | 2.30000* | .10869 | .000 | 1.9423 | 2.6577 |
| | | BSFL 40% | 1.69333* | .10869 | .000 | 1.3356 | 2.0510 |
| | BSFL 10% | Control | -.86667* | .10869 | .000 | -1.2244 | -.5090 |
| | | BSFL 20% | .49667* | .10869 | .007 | .1390 | .8544 |
| | | BSFL 30% | 1.43333* | .10869 | .000 | 1.0756 | 1.7910 |
| | | BSFL 40% | .82667* | .10869 | .000 | .4690 | 1.1844 |
| | | Control | -1.36333* | .10869 | .000 | -1.7210 | -1.0056 |

| | | | | | | |
|---|---|---|---|---|---|---|
| BSFL 20% | BSFL 10% | -.49667* | .10869 | .007 | -.8544 | -.1390 |
| | BSFL 30% | .93667* | .10869 | .000 | .5790 | 1.2944 |
| | BSFL 40% | .33000 | .10869 | .074 | -.0277 | .6877 |
| BSFL 30% | Control | -2.30000* | .10869 | .000 | -2.6577 | -1.9423 |
| | BSFL 10% | -1.43333* | .10869 | .000 | -1.7910 | -1.0756 |
| | BSFL 20% | -.93667* | .10869 | .000 | -1.2944 | -.5790 |
| | BSFL 40% | -.60667* | .10869 | .002 | -.9644 | -.2490 |
| BSFL 40% | Control | -1.69333* | .10869 | .000 | -2.0510 | -1.3356 |
| | BSFL 10% | -.82667* | .10869 | .000 | -1.1844 | -.4690 |
| | BSFL 20% | -.33000 | .10869 | .074 | -.6877 | .0277 |
| | BSFL 30% | .60667* | .10869 | .002 | .2490 | .9644 |

*. The mean difference is significant at the 0.05 level.

Table A.10: Post Hoc Test (Bulk Density)

**Multiple Comparisons**

BulkD

| Dependent Variable: | (I) Group | (J) Group | Mean Difference (I-J) | Std. Error | Sig. | 95% Confidence Interval Lower Bound | 95% Confidence Interval Upper Bound |
|---|---|---|---|---|---|---|---|
| Tukey HSD | Control | BSFL 10% | .01667 | .00843 | .342 | -.0111 | .0444 |
| | | BSFL 20% | .02333 | .00843 | .112 | -.0044 | .0511 |
| | | BSFL 30% | .00000 | .00843 | 1.000 | -.0278 | .0278 |
| | | BSFL 40% | .07333* | .00843 | .000 | .0456 | .1011 |
| | BSFL 10% | Control | -.01667 | .00843 | .342 | -.0444 | .0111 |
| | | BSFL 20% | .00667 | .00843 | .928 | -.0211 | .0344 |
| | | BSFL 30% | -.01667 | .00843 | .342 | -.0444 | .0111 |
| | | BSFL 40% | .05667* | .00843 | .000 | .0289 | .0844 |
| | BSFL 20% | Control | -.02333 | .00843 | .112 | -.0511 | .0044 |
| | | BSFL 10% | -.00667 | .00843 | .928 | -.0344 | .0211 |
| | | BSFL 30% | -.02333 | .00843 | .112 | -.0511 | .0044 |
| | | BSFL 40% | .05000* | .00843 | .001 | .0222 | .0778 |
| | BSFL 30% | Control | .00000 | .00843 | 1.000 | -.0278 | .0278 |
| | | BSFL 10% | .01667 | .00843 | .342 | -.0111 | .0444 |
| | | BSFL 20% | .02333 | .00843 | .112 | -.0044 | .0511 |
| | | BSFL 40% | .07333* | .00843 | .000 | .0456 | .1011 |
| | BSFL 40% | Control | -.07333* | .00843 | .000 | -.1011 | -.0456 |
| | | BSFL 10% | -.05667* | .00843 | .000 | -.0844 | -.0289 |
| | | BSFL 20% | -.05000* | .00843 | .001 | -.0778 | -.0222 |
| | | BSFL 30% | -.07333* | .00843 | .000 | -.1011 | -.0456 |

*. The mean difference is significant at the 0.05 level.

Table A.11: Post Hoc Test (Sinking Velocity)

**Multiple Comparisons**

SinkingV

| Dependent Variable | (I) Group | (J) Group | Mean Difference (I-J) | Std. Error | Sig. | 95% Confidence Interval Lower Bound | 95% Confidence Interval Upper Bound |
|---|---|---|---|---|---|---|---|
| Tukey HSD | Control | BSFL 10% | -.07667* | .01932 | .018 | -.1403 | -.0131 |
| | | BSFL 20% | .01333 | .01932 | .954 | -.0503 | .0769 |
| | | BSFL 30% | .01000 | .01932 | .984 | -.0536 | .0736 |
| | | BSFL 40% | -.02333 | .01932 | .748 | -.0869 | .0403 |
| | BSFL 10% | Control | .07667* | .01932 | .018 | .0131 | .1403 |
| | | BSFL 20% | .09000* | .01932 | .006 | .0264 | .1536 |
| | | BSFL 30% | .08667* | .01932 | .008 | .0231 | .1503 |
| | | BSFL 40% | .05333 | .01932 | .113 | -.0103 | .1169 |
| | BSFL 20% | Control | -.01333 | .01932 | .954 | -.0769 | .0503 |
| | | BSFL 10% | -.09000* | .01932 | .006 | -.1536 | -.0264 |
| | | BSFL 30% | -.00333 | .01932 | 1.000 | -.0669 | .0603 |
| | | BSFL 40% | -.03667 | .01932 | .377 | -.1003 | .0269 |
| | BSFL 30% | Control | -.01000 | .01932 | .984 | -.0736 | .0536 |
| | | BSFL 10% | -.08667* | .01932 | .008 | -.1503 | -.0231 |
| | | BSFL 20% | .00333 | .01932 | 1.000 | -.0603 | .0669 |
| | | BSFL 40% | -.03333 | .01932 | .462 | -.0969 | .0303 |
| | BSFL 40% | Control | .02333 | .01932 | .748 | -.0403 | .0869 |
| | | BSFL 10% | -.05333 | .01932 | .113 | -.1169 | .0103 |
| | | BSFL 20% | .03667 | .01932 | .377 | -.0269 | .1003 |
| | | BSFL 30% | .03333 | .01932 | .462 | -.0303 | .0969 |

*. The mean difference is significant at the 0.05 level.

Table A.12: Post Hoc Test (Textural Profile Analysis)

**Multiple Comparisons**

Tukey HSD

| Dependent Variable | (I) GROUP | (J) GROUP | Mean Difference (I-J) | Std. Error | Sig. | 95% Confidence Interval Lower Bound | 95% Confidence Interval Upper Bound |
|---|---|---|---|---|---|---|---|
| HARDNESS | Control | BSFL 10% | -233.33333* | 35.66573 | .000 | -350.7122 | -115.9545 |
| | | BSFL 20% | -608.66667* | 35.66573 | .000 | -726.0455 | -491.2878 |
| | | BSFL 30% | -1174.00000* | 35.66573 | .000 | -1291.3789 | -1056.6211 |
| | | BSFL 40% | -687.00000* | 35.66573 | .000 | -804.3789 | -569.6211 |
| | BSFL 10% | Control | 233.33333* | 35.66573 | .000 | 115.9545 | 350.7122 |
| | | BSFL 20% | -375.33333* | 35.66573 | .000 | -492.7122 | -257.9545 |
| | | BSFL 30% | -940.66667* | 35.66573 | .000 | -1058.0455 | -823.2878 |
| | | BSFL 40% | -453.66667* | 35.66573 | .000 | -571.0455 | -336.2878 |
| | BSFL 20% | Control | 608.66667* | 35.66573 | .000 | 491.2878 | 726.0455 |
| | | BSFL 10% | 375.33333* | 35.66573 | .000 | 257.9545 | 492.7122 |
| | | BSFL 30% | -565.33333* | 35.66573 | .000 | -682.7122 | -447.9545 |
| | | BSFL 40% | -78.33333 | 35.66573 | .256 | -195.7122 | 39.0455 |
| | BSFL 30% | Control | 1174.00000* | 35.66573 | .000 | 1056.6211 | 1291.3789 |
| | | BSFL 10% | 940.66667* | 35.66573 | .000 | 823.2878 | 1058.0455 |
| | | BSFL 20% | 565.33333* | 35.66573 | .000 | 447.9545 | 682.7122 |
| | | BSFL 40% | 487.00000* | 35.66573 | .000 | 369.6211 | 604.3789 |
| | BSFL 40% | Control | 687.00000* | 35.66573 | .000 | 569.6211 | 804.3789 |
| | | BSFL 10% | 453.66667* | 35.66573 | .000 | 336.2878 | 571.0455 |
| | | BSFL 20% | 78.33333 | 35.66573 | .256 | -39.0455 | 195.7122 |
| | | BSFL 30% | -487.00000* | 35.66573 | .000 | -604.3789 | -369.6211 |
| COHENSIVENESS | Control | BSFL 10% | .38667* | .02591 | .000 | .3014 | .4719 |
| | | BSFL 20% | .34000* | .02591 | .000 | .2547 | .4253 |
| | | BSFL 30% | .52333* | .02591 | .000 | .4381 | .6086 |
| | | BSFL 40% | .54000* | .02591 | .000 | .4547 | .6253 |

| | | | | | | | |
|---|---|---|---|---|---|---|---|
| | BSFL 10% | Control | -.38667* | .02591 | .000 | -.4719 | -.3014 |
| | | BSFL 20% | -.04667 | .02591 | .423 | -.1319 | .0386 |
| | | BSFL 30% | .13667* | .02591 | .003 | .0514 | .2219 |
| | | BSFL 40% | .15333* | .02591 | .001 | .0681 | .2386 |
| | BSFL 20% | Control | -.34000* | .02591 | .000 | -.4253 | -.2547 |
| | | BSFL 10% | .04667 | .02591 | .423 | -.0386 | .1319 |
| | | BSFL 30% | .18333* | .02591 | .000 | .0981 | .2686 |
| | | BSFL 40% | .20000* | .02591 | .000 | .1147 | .2853 |
| | BSFL 30% | Control | -.52333* | .02591 | .000 | -.6086 | -.4381 |
| | | BSFL 10% | -.13667* | .02591 | .003 | -.2219 | -.0514 |
| | | BSFL 20% | -.18333* | .02591 | .000 | -.2686 | -.0981 |
| | | BSFL 40% | .01667 | .02591 | .964 | -.0686 | .1019 |
| | BSFL 40% | Control | -.54000* | .02591 | .000 | -.6253 | -.4547 |
| | | BSFL 10% | -.15333* | .02591 | .001 | -.2386 | -.0681 |
| | | BSFL 20% | -.20000* | .02591 | .000 | -.2853 | -.1147 |
| | | BSFL 30% | -.01667 | .02591 | .964 | -.1019 | .0686 |
| SPRINGNESS | Control | BSFL 10% | -.10667* | .01989 | .002 | -.1721 | -.0412 |
| | | BSFL 20% | -.08000* | .01989 | .016 | -.1455 | -.0145 |
| | | BSFL 30% | -.07000* | .01989 | .035 | -.1355 | -.0045 |
| | | BSFL 40% | -.05667 | .01989 | .099 | -.1221 | .0088 |
| | BSFL 10% | Control | .10667* | .01989 | .002 | .0412 | .1721 |
| | | BSFL 20% | .02667 | .01989 | .675 | -.0388 | .0921 |
| | | BSFL 30% | .03667 | .01989 | .403 | -.0288 | .1021 |
| | | BSFL 40% | .05000 | .01989 | .163 | -.0155 | .1155 |
| | BSFL 20% | Control | .08000* | .01989 | .016 | .0145 | .1455 |
| | | BSFL 10% | -.02667 | .01989 | .675 | -.0921 | .0388 |
| | | BSFL 30% | .01000 | .01989 | .985 | -.0555 | .0755 |
| | | BSFL 40% | .02333 | .01989 | .766 | -.0421 | .0888 |
| | BSFL 30% | Control | .07000* | .01989 | .035 | .0045 | .1355 |
| | | BSFL 10% | -.03667 | .01989 | .403 | -.1021 | .0288 |
| | | BSFL 20% | -.01000 | .01989 | .985 | -.0755 | .0555 |
| | | BSFL 40% | .01333 | .01989 | .959 | -.0521 | .0788 |

| | | | | | | |
|---|---|---|---|---|---|---|
| BSFL 40% | Control | .05667 | .01989 | .099 | -.0088 | .1221 |
| | BSFL 10% | -.05000 | .01989 | .163 | -.1155 | .0155 |
| | BSFL 20% | -.02333 | .01989 | .766 | -.0888 | .0421 |
| | BSFL 30% | -.01333 | .01989 | .959 | -.0788 | .0521 |

*. The mean difference is significant at the 0.05 level.

Table A.13: Homogenous Subset (Colour Analysis)

**Color_Lightness**

Tukey HSD[a]

| Group | N | Subset for alpha = 0.05 | | | | |
|---|---|---|---|---|---|---|
| | | 1 | 2 | 3 | 4 | 5 |
| BSFL 20% | 3 | 38.0900 | | | | |
| BSFL 30% | 3 | | 39.5267 | | | |
| BSFL 40% | 3 | | | 40.1933 | | |
| BSFL 10% | 3 | | | | 40.8267 | |
| Control | 3 | | | | | 44.7267 |
| Sig. | | 1.000 | 1.000 | 1.000 | 1.000 | 1.000 |

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 3.000.

**Color_a**

Tukey HSD[a]

| Group | N | Subset for alpha = 0.05 | | | | |
|---|---|---|---|---|---|---|
| | | 1 | 2 | 3 | 4 | 5 |
| BSFL 30% | 3 | 2.5667 | | | | |
| BSFL 40% | 3 | | 2.7567 | | | |
| BSFL 20% | 3 | | | 3.0833 | | |
| BSFL 10% | 3 | | | | 3.2000 | |
| Control | 3 | | | | | 4.1667 |
| Sig. | | 1.000 | 1.000 | 1.000 | 1.000 | 1.000 |

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 3.000.

**Color_b**

Tukey HSD[a]

| Group | N | Subset for alpha = 0.05 | | | |
|---|---|---|---|---|---|
| | | 1 | 2 | 3 | 4 |
| BSFL 30% | 3 | 5.1700 | | | |
| BSFL 40% | 3 | | 5.7767 | | |
| BSFL 20% | 3 | | 6.1067 | | |
| BSFL 10% | 3 | | | 6.6033 | |
| Control | 3 | | | | 7.4700 |
| Sig. | | 1.000 | .074 | 1.000 | 1.000 |

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 3.000.

Table A.14: Homogenous Subset (Bulk Density)

**BulkD**

Tukey HSD[a]

| Group | N | Subset for alpha = 0.05 | |
|---|---|---|---|
| | | 1 | 2 |
| BSFL 40% | 3 | .3233 | |
| BSFL 20% | 3 | | .3733 |
| BSFL 10% | 3 | | .3800 |
| Control | 3 | | .3967 |
| BSFL 30% | 3 | | .3967 |
| Sig. | | 1.000 | .112 |

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 3.000.

Table A.15: Homogenous Subset (Sinking Velocity)

**SinkingV**

Tukey HSD[a]

| Group | N | Subset for alpha = 0.05 | |
|---|---|---|---|
| | | 1 | 2 |
| BSFL 20% | 3 | .2900 | |
| BSFL 30% | 3 | .2933 | |
| Control | 3 | .3033 | |
| BSFL 40% | 3 | .3267 | .3267 |
| BSFL 10% | 3 | | .3800 |
| Sig. | | .377 | .113 |

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 3.000.

Table A.16: Homogenous Subset (Textural Profile Analysis)

**HARDNESS**

Tukey HSD[a]

| GROUP | N | Subset for alpha = 0.05 | | | |
|---|---|---|---|---|---|
| | | 1 | 2 | 3 | 4 |
| Control | 3 | 652.3333 | | | |
| BSFL 10% | 3 | | 885.6667 | | |
| BSFL 20% | 3 | | | 1261.0000 | |
| BSFL 40% | 3 | | | 1339.3333 | |
| BSFL 30% | 3 | | | | 1826.3333 |
| Sig. | | 1.000 | 1.000 | .256 | 1.000 |

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 3.000.

## COHESIVENESS

Tukey HSD[a]

| GROUP | N | Subset for alpha = 0.05 | | |
|---|---|---|---|---|
| | | 1 | 2 | 3 |
| BSFL 40% | 3 | .2433 | | |
| BSFL 30% | 3 | .2600 | | |
| BSFL 10% | 3 | | .3967 | |
| BSFL 20% | 3 | | .4433 | |
| Control | 3 | | | .7833 |
| Sig. | | .964 | .423 | 1.000 |

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 3.000.

## SPRINGINESS

Tukey HSD[a]

| GROUP | N | Subset for alpha = 0.05 | |
|---|---|---|---|
| | | 1 | 2 |
| Control | 3 | .1567 | |
| BSFL 40% | 3 | .2133 | .2133 |
| BSFL 30% | 3 | | .2267 |
| BSFL 20% | 3 | | .2367 |
| BSFL 10% | 3 | | .2633 |
| Sig. | | .099 | .163 |

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 3.000.