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Characterization of Mealworm Powder and its Fortification In all-purpose Flour Bread

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Characterization of Mealworm Powder and its Fortification

In all-Purpose Flour Bread.

A THESIS APPROVED FOR

THE DEPARTMENT OF HUMAN ENVIRONMENT SCIENCES

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To my father, Ghasem Ali Akbaran

And my mother, Mohtaram Ghazanfari

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Abstract

In the face of increasing population globally, food security has become a worldwide concern especially in the developing countries. Protein is mainly derived from animal food, and the demand for access to live stock, land, and water is increasing. Agriculture productivity will diminish as the population rises; therefore, insects as a food source has become more common globally but still is not well known.

The research was conducted to determine in defatted mealworm powder what percentage of valuable protein is present in order to apply it as protein ingredients in food industries. The first objective was to compare the extraction of fat using two solvents and evaluate the function of both solvents. Objective two was to determine the potential use of non-defatted mealworm powder as an alternative high protein source in bread.

The ethanol defatted mealworm powder was found to be comprised of approximately 61.4% protein, 4.2% ash, 397 KCal/100g, 14.1% carbohydrates, 11.9% moisture and 8.24% fat whereas the hexane defatted mealworm powder was found to be 69.1% protein, 5.40% ash, 358 kcal/100g, 15.4% carbohydrates, 4.23% moisture and 5.68%fat. The fat extracted using the rotary evaporator was found to be 20.50% using ethanol and hexane is about 23.13 %

Bread containing mealworm powder showed higher content of protein and the result confirmed the potential application of insect's powder as a protein source. Proximate analysis of bread enriched with 10 % and 20 % compared with all-purpose flour shows the mealworm powder could be added to bread or low nutrient value food to improve the nutritional value. The main goal of this research would allow individuals to consider mealworm as an additional source of protein in their diet.

Keywords: Insects, Protein, defatting, enriching bread with mealworm powder, Dough Rheology

1.1 CHAPTER ONE: INTRODUCTION

The demand for food and especially protein is increasing globally and is expected to increase between 2012 and 2050 by 70-80% (Dennis, 2012). Protein is the main macronutrient that is essential for human bodies. It is mainly found in animal products such as meat, poultry, seafood, and eggs; however, it can also be found in other sources such as nuts, beans, and legumes. Livestock production has a major impact on the environment, so finding a comparable source to replace animal meat and livestock could help to reduce environmental crisis. However, finding a good alternative source is still under study (Elzerman, Hoek, Van Boekel, & Luning, 2011). Scientists reviewed sixteen studies to assess the production of pork, chicken, beef, milk and eggs using life cycle analysis (LCA) (Vries & Boer, 2009). The livestock sector is responsible for about 15% of the total greenhouse emissions and uses about 70% of all agricultural land (Dennis, 2012). According to a study by Hartmann and Siegrist (2017), food consumers may not be aware of how much their daily food choices impact the environment and climate. Researchers found that roughly 20% to 30% of the total environmental crisis caused by humans is related to food production (Vries & Boer, 2009).

In literature by Vries and Boer (2009), they stated that the production of 1kilogram (kg) of beef uses the most land and energy and had the highest global warming potential (GWP) followed by pork and chicken. Moreover, Hartmann and Siegrist (2017) mentioned in their study that for induviduals who eat meat, the greenhouse emissions are two times higher than for vegans. A study by Arnold van Huis (2013) reported that livestock must consume about 6 kg of plant protein in order to produce 1kg of high-quality animal protein (Van Huis, 2013). As the cost of agricultural crops increases, the prices for beef, pork, and poultry will increase 30% by the year of 2050 compared to the year of 2000, and climate change can increase the price by 18-21%

(Van Huis, 2013). The increase in food and food prices encourages the search for alternative protein sources such as cultured meat, seaweed, vegetables and fungi, and small livestock (Van Huis, 2013).

Edible insects are one of the alternative resources that researchers are considering as a potential protein source. They already are consumed in many regions of Asia, Africa, and America (Klunder, Wolkers-Rooijackers, Korpela & Nout, 2012). About 1,800 insect species have been used for human consumption globally (Jongema, 2015). The protein content of insects depends on the species and the stage in the life cycle and varies between 40% and 75% of the dry weight (Jongema, 2015). Researchers suggest that most insects can be collected from natural environments, and this collection depends on the species, the season, and local restrictions (Jongema, 2015). According to Klunder (2012), the insects can be consumed in three ways: as whole insects, as insects processed into powder or paste, and as insect extract such as protein isolate.

In many parts of the world, especially Africa, Latin America, and Asia, eating insects, or entomophagy, is an important dietary behavior (Hartmann, Shi, Gisusto & Siegrist, 2015). Researchers performed an experiment to determine the nutrient content of insects, especially comparing its iron content to that of sirloin. In this experiment, there were significant differences in the iron content of a grasshopper, a cricket, a mealworm, and sirloin (Latunde-Dada, Yang, & Vera Aviles, 2016). A study compared the nutritional value of six edible, cultivated, and wild insect groups that include the cricket (Gryllus sp.), the giant mealworm (Zophobas morio F.), the yellow mealworm (Tenebrio molitor L.), the silkworm (Bombyx mori L.), the Javanese grasshopper (Valanga nigricornis Burm), and the paddy locust (Nomadacris succincta L.).

According to the results, 100-gram dry weight insects have about 3.15-4.1 milligram of iron (Fe) and 24.82-31.22 milligrams of calcium (Kuntadi, Adalina, & Maharani, 2018).

Due to the demand of the rising global population, food industries are trying to find new alternative protein sources to add to human foods (González, Garzón, & Rosell, 2018). Protein is another nutritional consideration which researchers have studied in edible insects. Most commercial food is enriched by proteins that come from rye, soy, oats, and legumes; however, protein in insects is higher than legumes, beans, etc. Researchers have found that beans have about 23.5% protein, lentils have 26.7%, and soybeans have about 41.1% protein. The protein content of insects varies from 45% to 57% (González et al., 2018). However, according to a different study that was conducted by Kuntadi, Adalina, & Maharani, (2018), insects contain about 32.59%-76.69% protein. They also contain about 6.9%-29.47% fat (Kuntadi et al., 2018). In a different study, researchers stated the fat content is between 27% and 36% (González et al., 2018).

These studies prove insects can be used as an alternative source of protein to enhance nutritional value in food application and also can be utilized against malnutrition (Kuntadi et al., 2018). According to a study by González et al. (2018), to increase the nutritional value of food products, some of the insect species can be added in the form of powder. When insects are not visible in food products, consumers are open to eating them. Making them invisible in the form of powder or other food ingredients causes it to be more pleasant for consumers to use insects as a source of food (González et al., 2018).

In the study conducted for this paper, there were three samples with different powder mixes: 100% all-purpose flour, 10% mealworm powder, and 20% mealworm powder incorporated with the all-purpose flour to evaluate the functional properties and nutritional value of the finished product. The focus is mostly on mealworms as a source of non-recognizable protein to enrich the protein content in low-nutrient foods such as all-purpose flour bread.

Research Question:

Can mealworm-fortified, all-purpose flour bread be a good alternative to formulate a nutrient-dense, good-quality bread?

Objective One

To defat mealworm powder by two solvents (ethanol and hexane) to evaluate the percentage of fat and proximate analysis.

Objective Two

To determine the effects of incorporating 10% and 20% of mealworm powder with all-purpose flour on physio-chemical, textural properties of all-purpose bread.

CHAPTER TWO: LITERATURE REVIEW

2.1. Insect Species and Geography:

Edible insects are one of the foods that researchers consider a protein source. They are consumed in many regions such as Asia, Africa, and America (Klunder, Wolkers-Rooijackers, Korpela & Nout, 2012). It has been assessed that about 1800 insect species are used for human consumption worldwide (Jongema, 2015). About 150 different kinds of insects, which are mainly wild-harvested, are consumed in Thailand (Dobermann, Swift, & Field, 2017)

Most of the insect species are known as natural resources (Dossey, Morales-Ramos, & Rojas, 2016). In many parts of the world, insects can be used for commercial food products for humans and animals (Dossey et al., 2016). More than 65 species of insects have been identified as food in Congo, Africa (Van Huris, 2013). According to Gonzalez (2018), in European countries some insect species are used as a substitution source of animal protein for human and animal consumption. The same study explains the most common insect species for food are "*Lepidoptera* (caterpillars of butterflies and moths), *Hymenoptera* (bees, wasps, ants), *Orthoptera* (grasshoppers, locusts, cricket, termites), *Hemiptera* (cicadas, leaf and plant hoppers, true bugs and scale insects), *Odonata*(dragonflies) and *Diptera* (flies) and *mealworms* (larval form of the mealworm beetle)," (González et al., 2018). The same article suggests that insects have an advantage compared to other protein sources due to a high level of food-chain energy retention. In addition, they can be fed with limited products such as cereal grains and secondary products from food manufacturing (González et al., 2018). Insects are also an essential source for many animal species' diet such as trout and bass, as well as frogs, turtles, and snakes (Srivastava, Babu, & Pandey, 2009). The figures 1 and 2 illustrate the edible insects and spiders in the world and by region (Dossey et al., 2016).

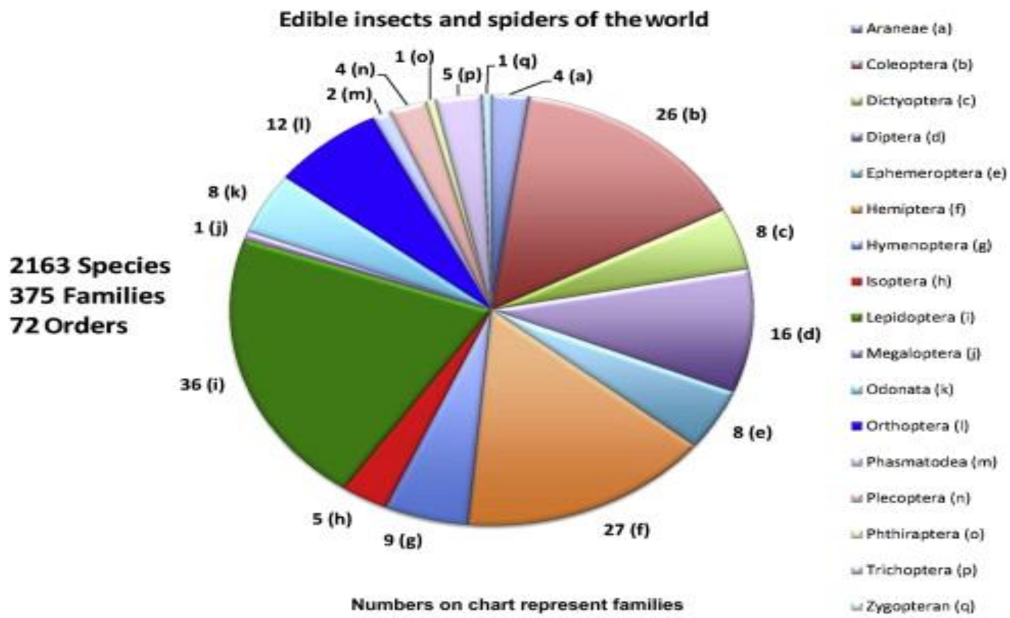

Figure 1: Global list of Edible Insects and Spiders *(Dossey et al., 2016)*

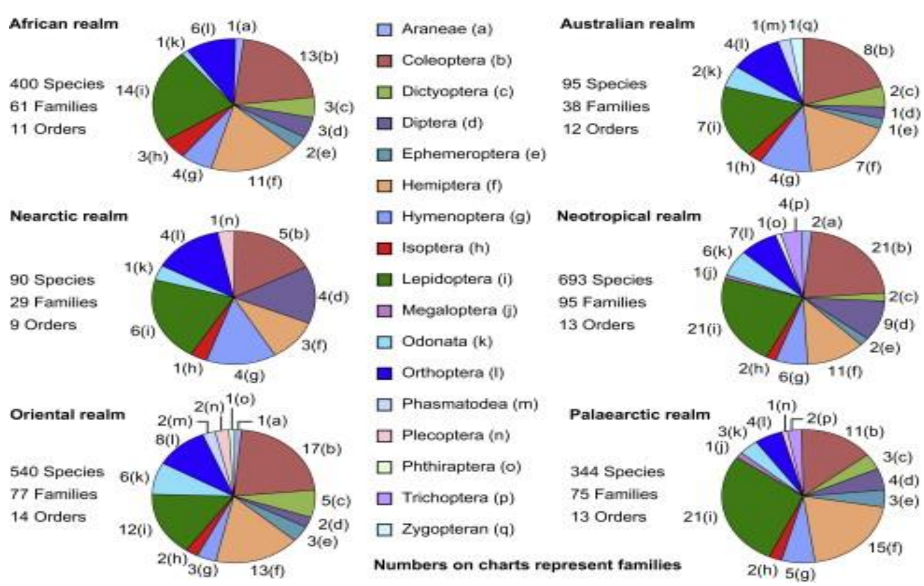

Figure 2: Regional list of Edible Insects and Spiders (Dossey et al., 2016)

Using edible insects in China has a long history, and it is still popular. About 324 insect species from 11 orders have been recognized as edible or related to Entomophagy, but only about 10 to 20 percent of those are used for food and medicine (Feng et al., 2018). Eating insects is very common in China and they are eaten either directly in food or indirectly by consuming livestock which was fed by insects (Feng et al., 2018).

2.2. Insects and Benefits

2.2.1. Environment and Food Production:

Insects are a more environment friendly source of protein compared to other animal sources. Growing population increases the demand for food production. Global warming and changing climates will also impact food production. Availability of land and water concerns meat growers and producers as populations increase (Dobermann et al., 2017). The production of livestock has a significant impact on the environment, and food consumers may not be aware how much their daily food choices affect the environment and climate (Dobermann et al., 2017).To assess the production of pork, chicken, beef, milk, and eggs using life cycle analysis (LCA), scientists reviewed sixteen studies and have found about 20% to 30% of the total environmental disasters initiated by humans is associated to food production (Vries & Boer, 2009).

Usage of beef requires more land and energy leading to a global warming potential. Greenhouse emissions for people who eat meat are two times higher compared to vegans (Hartmann, Shi, Giusto, & Siegrist, 2015). The production of ammonia and environment pollution is also associated with livestock production (Oonincx & De Boer, 2012). Researchers found transporting, slaughtering and storing of meat, also requiring energy, contributes about 17-25 % of GHGs in global warming (Shockley & Dossey, 2014). Insects require less amounts of water, land, and energy resources and produce lower amounts of environmental pollution in comparison to livestock. To produce every kilogram of high-quality animal protein, livestock have to consume approximately 6 Kg of plant protein (Arnold van Huis, 2012). The livestock sector is producing about 15% of the total greenhouse emissions and uses about 70% of all agricultural land (Dennis, 2012). High demand for livestock causes prices to increase by 18-21% leading to further food shortage issues, which might be more significant as agricultural productivity decreases as well (Van Huis, 2013). By 2050, the price of livestock will have increased by 30% leading us to find other alternative sources that are of minimum cost and can be affordable for the world population (Van Huis, 2013).

Figure 3 illustrates the amount of water and land that is required in producing livestock compared to producing insects. Also, Figure 4 shows the amount of carbon dioxide (CO_2) that is produced during production (Dobermann et al., 2017)

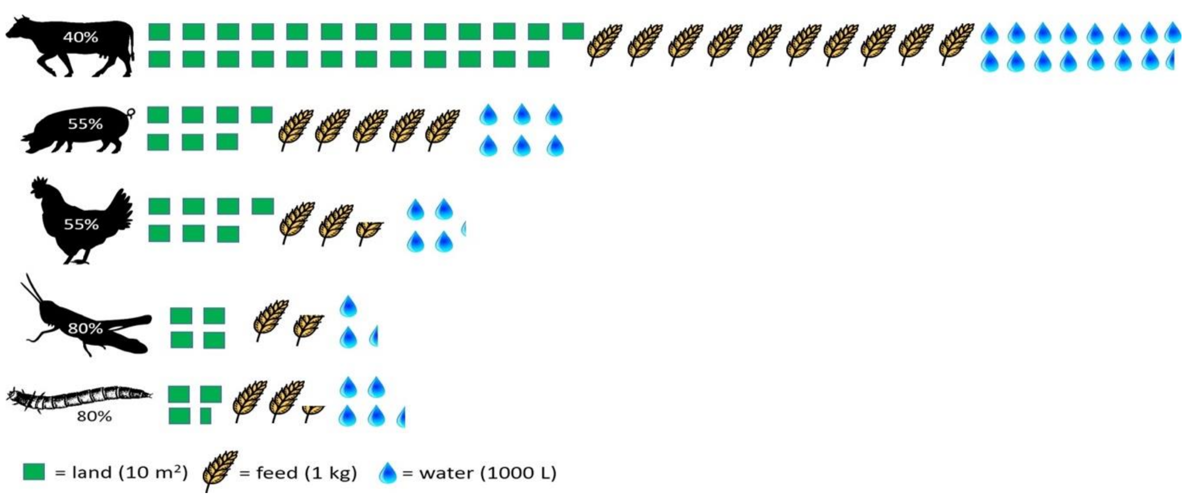

Figure 3: The amount of water and land requires in producing meat (Doberman et al., 2017)

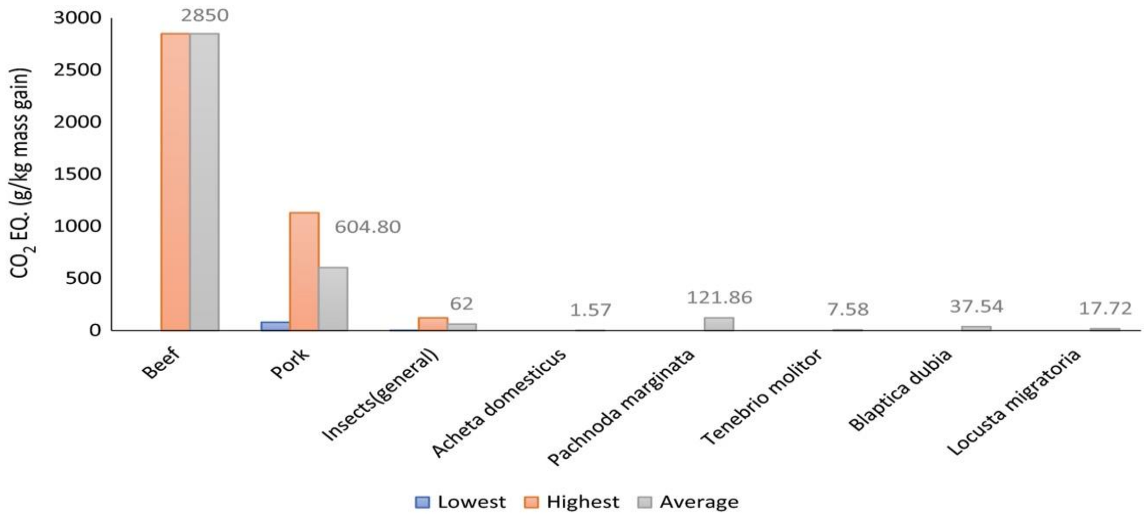

Figure 4: The amount of carbon dioxide (CO2) during production (Doberman et al., 2017).

2.2.2. Nutrition:

Increasing world population, expanding economic growth, and urbanization will cause the demand of high-value protein to increase. In 2050, the population is estimated to reach about 9 billion people according to the Food and Agriculture Organization, and food demand will increase by 70% worldwide compared to recent levels (Zielińska, Baraniak, Karaś, Rybczyńska, & Jakubczyk, 2015). The same authors suggest that alternative sources of protein such as insects are required, and recent protein sources are limited and not sufficient due to increasing global population (Zielińska et al., 2015).

Protein is one of the main macronutrients that are essential for human bodies. It contains amino acids, which are the building blocks of protein that help with growth and development of the human body. Protein is mainly found in animal products such as meat, poultry, seafood, and eggs. Arnold van Huis mentioned in his study, that increases in food and food prices tend to encourage the researchers to find sustainable protein sources (Van Huis, 2013). As evidenced by FAO (2012) there are about 870 million undernourished people in the world, therefore, edible insects can help to support the demand of nutritious food in developing countries (Zielińska et al., 2015). According to the World Health Organization about 27% of children five years old and younger are undernourished because they do not have access to good protein sources (Choi, Wong, & Auh, 2017).

Insects are known as a rich source of nutrients and are being considered as an alternative source of proteins and lipids (Kuntadi et al., 2018). According to Zielinska, et. al. (2015) insects are an available source of protein, lipids, carbohydrates, and some vitamins and minerals such as Zinc, iron, and calcium. Researchers performed an experiment to determine the nutrient content of insects, especially iron, by comparing it with sirloin. In this experiment, there were significant differences in the iron content of the grasshopper, cricket, mealworm, and sirloin. The iron level obtained from crickets was (1562 mg/100g dry matter) which is reported to be 180% more than we could obtain from beef. It has been reported the iron compounds in insects are mostly in the form of ferritin, holoferitin, and cytochrome (Latunde-Dada, Yang & Vera Aviles, 2016). This study also reported that the levels of copper, sodium, potassium, iron, zinc, and selenium are comparable in mealworms to beef (Latunde-Dada, Yang & Vera Aviles, 2016).

As evidenced by Rumpold and Schuluter (2013), insects are also a good source of energy, fat, minerals, and vitamins (Dobermann et al., 2017). The energy level of insects is between 400 to 500 per 100 g of dry matter, which is comparable with other protein sources (Dobermann et al., 2017). The protein content of insects depends on species and stage in the life cycle, which can vary from 40% to 75% on dry weight (Jongema, 2015). According to Doberman et al (2017), there are many things which affect the amount of protein, and fat and energy levels such as insect's species, diet, stage of development, sex and natural environments. Most insects can be collected from natural environments, and different species are available depending on the season

(Jongema, 2015). Insects are mainly consumed in three ways: whole insects, non- recognizable insects processed in some powder or paste, and extracts such as protein isolate (Klunder, Wolkers-Rooijackers, Korpela, & Nout, 2012).

2.2.3. Entomophagy:

Entomophagy is dietary consumption of insects and has already become common in many parts of the world, however consuming insects as protein source is still unclear among people. About 113 countries are already using insects traditionally and practicing entomophagy (Rumpold & Schlüter, 2013). In Africa, Latin America, and Asia, eating insects or entomophagy is an important dietary behavior (Htrman, Shi, Gisusto & Siegrist 2015). In many regions, the cultural barriers affect entomophagy and are not accepted in food regardless of their nutritional benefit. Even though insect consumption is high in some countries and is known as a novel food, in many European countries the sale of insects for human consumption is illegal (Hartman & Siegrist, 2016). A study by Gmuer, Nuessli Guth, Hartman, and Siegrist (2016) determined the positive and negative emotional expectations that Western countries have about the consumption of insects. Based on emotional tests, they have shown that the marked products which contain whole insects in comparison to adding them to food products seems to be less preferable (Gmuer, Guth, Hartmann, & Siegrist, 2016).

In a study designed by Verbeke (2015), Entomophagy and eating insects is growing and becoming a favorable way to deal with some nutrient and food challenges. However, according to Verbeke (2015), food neophobia is the most important factor to determine consumer willingness to adopt insects as meat and protein substitutes. A study shows that tasting insects can be a good initial way to reduce food neophobia and encourage people to be familiar with entomophagy (Megido et al., 2016). Changes in cultural tastes can happen and different foods can become popular, for example, sushi, which became popular and caused American people to start eating raw fish (Shelomi, 2015).

The available evidence shows that insects can be an excellent and sustainable source of animal protein. In China, people are more willing to consume food that is made with insects and have higher social acceptance compared with European countries such as Germany (Hartmann, Shi, Giusto & Siegrist, 2015). However, the Germans have a higher willingness to eat processed insect-based foods such as cookies made from cricket flour compared to unprocessed foods (Hartmann, Shi, Giusto & Siegrist, 2015). Health is another essential point that influences their food choices according to Verbeke (2015). The same study also revealed that health has a significant role in people's willingness to adopt insects as a food source (Verbeke, 2015). There is no relationship between age and consuming insects, however, women were less likely to accept insects as a food (Hartmann, Shi, Giusto & Siegrist, 2015).

2.3. Mealworm as a protein source

The demand for food and especially protein is increasing globally and expected to increase between 2012 and 2050 by 70-80% (Dennis, 2012). Demand for novel protein sources is increasing and insects are a potential source of protein (Verkerk, Tramper, Van Trijp, & Martens, 2007).Yellow mealworm (Tenebrio Molitor) is one of the popular edible insects that has been recognized as a protein source for food application worldwide (Zhao, Vázquez-Gutiérrez, Johansson, Landberg, & Langton, 2016). Mealworm contains about 15 % fat and 20 % protein and also includes minerals, vitamins, amino acids, and fatty acids, which can become a potential source of protein in food application (Zhao et al., 2016). A study by Dobermann et al. (2017), reported the amount of minerals and vitamins in Tenebrio Molitor (Mealworm Larve), measured in mg/100g dry matter, are as follows; Calcium 45.77, Potassium 828.28, Magnesium 215.89, Phosphorus 722.74, Sodium 133.16, Iron 5.46, Zinc 12.53, Manganese 1.14, Copper 1.62. Also, mealworm like other insects has a lower impact on the environment due to lower gas emission compared to livestock production. The report shows that production of CO_2 per kg body mass of insect species is higher than pigs and cattle (Oonincx et al., 2010). Figure 5 shows the production of carbon dioxide CO_2 during the raising of livestock and insects, and in comparison, less CO_2 is produced in raising insects (Dobermann et al., 2017).

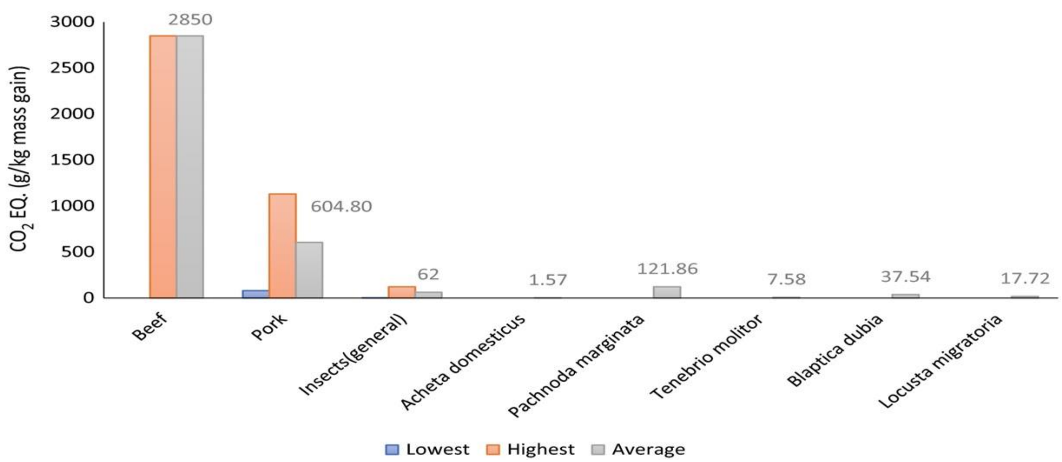

Figure 5: Production of carbon dioxide during raising livestock and insects

Source: (Dobermann et al., 2017)

CHAPTER THREE: METHODLOGY

3.1. Defatting Procedure of Mealworm Powder

According to a study by Zhao (2016), fresh yellow mealworm has about 15% fat and 20% protein (Zhao et al., 2016). This study explained that ethanol is a harmless solvent that has been used for defatting soybeans, ground maize, and *Quercus Suber L*. fruit for food application in human food, and also it can be used as an extraction solvent for defatting food materials for protein extraction (Zhao et al., 2016). Similarly, hexane is less toxic in comparison to other solvents (Hara & Radin, 1978). This procedure was performed in the laboratory of the chemistry department at the University of Central Oklahoma in Edmond, Oklahoma. The aim of this study was to evaluate the effectiveness of two different solvents on fat extraction.

Mealworm powder was obtained from All Things Bugs LLC in Oklahoma City, Oklahoma. In the defatting process, two different extraction solvents were used: ethanol (99.5%) and hexane (100%). The ratio of sample to solvent was 1:2 w/v. In this procedure, 10 g of mealworm powder was weighed and then transferred to 50 ml plastic conical centrifuge tubes (10 g of MWP for each tube). Figure 6 illustrates the mealworm powder in the centrifuge tube. 20 ml of solvent were added to each tube (Figure 7), and then the solution vortexed (Vortex-Genie 2) for 15 minutes (Figure 8). The slurry solution was centrifuged for 10 minutes at 4° C at 2500 G. The solvent and fat were removed via decanting and collection. A second extraction was carried out on the pellet with the same process (Zhao et al., 2016). Analyses were done in triplicate for both solvents to achieve maximum defatting.

3.2. Drying Procedure

The drying procedure was performed in the Chemistry Department at the University of Central Oklahoma (Figure 9). Each sample was attached to nitrogen gas, and nitrogen was passed over the resulting solid material (defatted mealworm powder). The time of the drying process for each sample was dependent on the type of solvent and ranged from 1 hour to 5 hours. The process of drying was faster for samples with hexane compared to ethanol. After drying the samples completely, they were kept under a desiccator to avoid the absorption of moisture. Then the samples were subject to proximate analysis.

3.3. Evaporating Solvents by Rotary Evaporator

The procedure was performed in the Chemistry Department at the University of Central Oklahoma to remove the solvent. The solvent and fat that were collected from the defatting procedure were transferred to 50 ml centrifuge tubes. The fat and solvent mixture was centrifuged to ensure there were no solids left. Then the solution was transferred to a flask and attached to the rotary evaporator (Figure 10). The weights of the empty flask and flask with solution were recorded. For each solvent, the temperature of the evaporation was adjusted separately. The flask rotation speed of the rotary evaporator was adjusted between 0-220 rpm, and the temperature was 1-7 ° C. Due to the lower boiling point of hexane (68° C) compared to ethanol (78° C), this solvent evaporated faster than ethanol. When the solvent was removed completely from fat, the weight of the flask was recorded and the fat percentage was calculated.

Figure 6: Transferring mealworm powder in centrifuge tubes

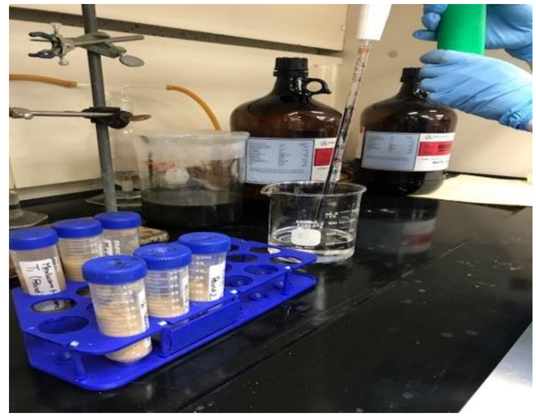

Figure 7: Adding solvent to mealworm powder by pipette

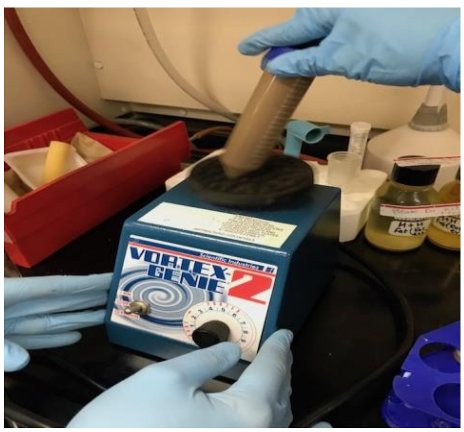

Figure 7: Mixing mealworm powder and solvent using vortex

Figure 9: Passing nitrogen gas to dry sample

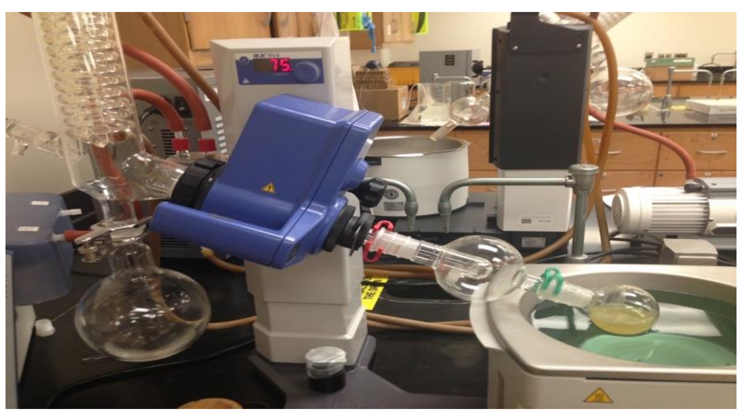

Figure 8: Removing solvent from fat by rotary evaporation

3.4. Bread Preparation

Bread was baked according to AACC-International method 10-10.3 Optimized Straight-Dough method. The ingredients were adjusted due to being incorporated with non-defatted mealworm powder. The formulation of the bread is shown in Table 1. The following ingredients of basic all-purpose flour were used in the straight-dough method: 520 g all-purpose flour (Great Value, Walmart Inc.), yeast (Fleischmann's Bread Machine Yeast 4 oz, Walmart Inc.), vegetable shortening, iodized salt, sugar (Great Value), non-defatted mealworm powder (All Things Bugs, Inc.), and 532.3 g of water.

Table 1: Formulation of non-defatted mealworm powder-fortified all-purpose flour bread

Ingredients	Amount in standard recipe Flour Basic (%)	Optimization	Adjusted with 10 % of non-defatted mealworm powder	Adjusted with 20% non-defatted mealworm powder
Flour	100.0	520.0 g	468 g all-purpose 52 g of MWP	416 g all-purpose 104 g MWP
Sugar	6.0	6.0 g	6 g	6g
Salt	1.5	1.0 g	1.0	1.0
Yeast, active dry	5.3	Followed yeast package instruction	6.3 g	6.3 g
Shortening	3.0	5.0 g	5.0 g	5.0 g
Water	Optimum	591.4 g	591.4 g	591.4 g

3.5. Bread Preparation Procedure

This procedure was performed in the Food Science Lab in the Human Environmental Sciences building at the University of Central Oklahoma. The yeast was prepared according to the manufacturers' instructions (1 ½ teaspoon of yeast was added to 236.59 g of warm water at 100° C and stood for 10 minutes until it doubled in volume). The dry ingredients (all-purpose flour, non-defatted mealworm powder, salt, sugar, and shortening) were placed into a Kitchen Aid, Artisan 5 qt. stand mixer mixing bowl and mixed for 1 minute at speed 1 in Edmond, Oklahoma, USA. With a wooden spatula, a pocket was made in the middle and the yeast solution was added and mixed for another minute at speed 2, and then 591.4 g of water was added and mixed for 1 minute. The sides were scraped down with a spatula and continued mixing at speed 3 for another minute. The dough was then kneaded with the mixer at the same speed for an additional 3 minutes. The total time of mixing and kneading was 5 minutes. The dough was placed in a bowl that was slightly sprayed with cooking oil. Then the dough was placed in a fermentation cabinet for 52 minutes at 27 °C. A first punching was done for 2 minutes. The dough was placed on a lightly floured board and folded and punched by hand. It was then transferred back to the fermentation cabinet for 25 minutes. The second punching was performed for another 2 minutes, and the dough was transferred back again to the fermentation cabinet for 13 minutes. The recipe called for 90 minutes of fermentation. However, it also mentioned that to achieve the maximum amount of proofing, 33 minutes should be added. Then the dough was molded in a 9-inch (9 x 41/2 In) aluminum foil pan and put back in the fermentation cabinet for an additional 33 minutes. It was then baked in a preheated oven for 40 minutes at 450 °F, and 1 liter of water in a beaker was placed in the oven for the entire cooking time. Then the bread was cooled on a cooling rack to dry completely. The same procedure was performed incorporating

10% and 20% non-defatted mealworm powder with all-purpose flour. The preparation steps are illustrated in Figure 11.

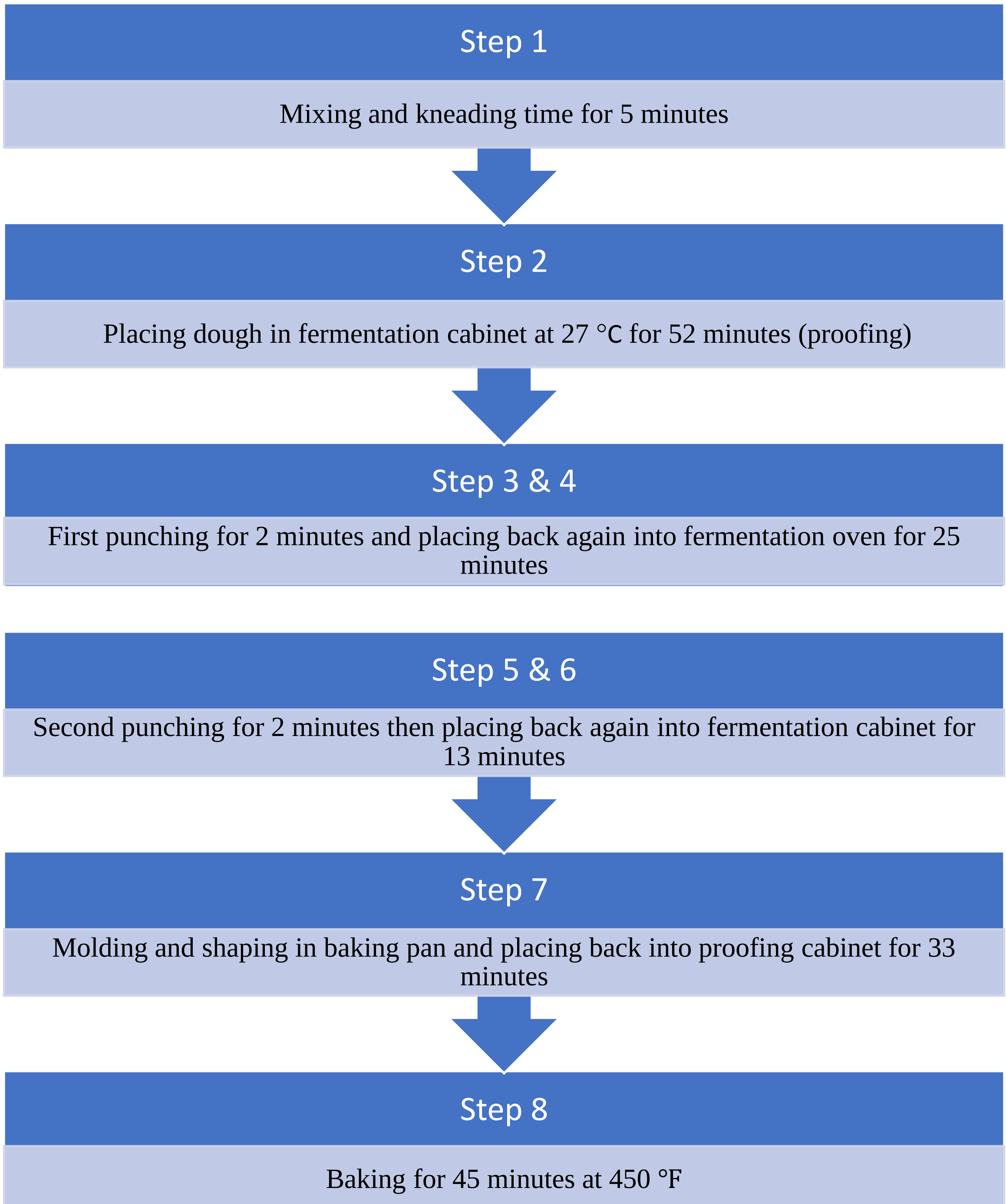

Figure 11: The process of preparation of bread using the straight-dough method.

Figure 12 illustrates the bread sample and the effect of non-defatted mealworm powder at different quantities.

Figure 9: The effect of 10% and 20% mealworm powder compared with control bread

3.6. Bread Analysis

3.6.1. Moisture Content

The moisture content of each bread sample was obtained by an Air-Oven Method using the AACC International method 44-15.02. In this method, 45 g of each sample was weighed and placed into a small aluminum dish and let to sit at room temperature for 60 hours for drying. The mass was recorded and calculated. Then 2 g of the same sample was weighed and placed in the oven at 103 $\pm$ 1 °C for 4-6 hours. Then bread samples were placed under a desiccator to avoid exposure to any moisture during cooling time and were then weighed. The data was placed in the formula below, and the moisture content was calculated.

Calculation

Equation 1 (one-stage and 103° air oven):

$$\% \text{ Moisture} = \frac{A}{B} \times 100$$

in which A = moisture loss in grams, B = original weight of sample.

Equation 2 (two-stage):

$$\% \text{ Total solids} = \frac{Y \times Z}{X}$$

where X = weight of original sample used for air-drying, Y = weight of sample after air-drying, Z = percent total *solids* in prepared ground sample (total solids = 100% – percent moisture at assay).

Equation 3:

$$\% \text{ Total moisture} = A + \frac{(100 - A)B}{100}$$

where A = percent moisture loss on air-drying, B = percent moisture loss as determined by oven-drying.

Figure 10: Moisture content equation

(Ref: AACC international method 44-15.02, Moisture Air-Oven Method)

3.6.2. Color Measurement

This experiment was performed at the Food and Agriculture Products Center at Oklahoma State University in Stillwater, Oklahoma. Each bread sample color was measured with HunterLab MiniScan XE Plus spectrophotometer (Model 45/0 LAV, 2.54-cm diameter aperture, illuminant A, 10° observer) at room temperature. Results were obtained within CIE lab color parameters: "L" was the measurement for lightness and varies from 100 white to zero. "a" indicates redness when (+ a) is positive, grayness when it is zero, and greenness when (- a) is negative. The "b" value indicates yellowness when (+ b) is positive, grayness when it is zero, and blueness when (- b) is negative. Measurements were carried out in triplicate.

3.6.3. Bread Firmness Measurement

The AIB Standard Procedure (White pan bread firmness measurement) was followed by using the TA-XT2 setting at 25 mm probe. Starting with slice 4 from the end of the loaf, two slices of the bread sample were placed in the fixture for each measurement (6 measurements per loaf x 2 loaves per variable = 12 readings per variable per testing day). As the test ran, the probe pushed through the center of the bread, and the maximum peak force value was identified and the results were placed in the file spreadsheet. Also, the mean significant difference of samples was performed by using ANOVA (analysis of variance) and Tukey HSD comparisons using SPSS.

Figure 11: Texture Analysis

3.6.4. Proximate Analysis of Enriched Bread with Mealworm Powder

The experiment was conducted at the Food and Agriculture Products Center at Oklahoma State University in Stillwater, Oklahoma. Three bread samples dried prior to submission and were tested for percent of moisture (18^{th} ed AOAC Official Method 950.46-moisture in Meat, JAOAC 33, 749(1950); 36, 279 (1953), ash percentage (18^{th} ed AOAC Official Method 920.153-ash of meat), fat percentage (18^{th} ed AOAC Official Method 922.06-fat in flour, JAOAC 6, 508

(1922); 9, 41, 429 (1926), and protein percentage (AOCS Official Methods, 5^{th} Edition, 2004: Ba 4e-93-Generic Combustion Method for Determination of Crude Protein). The percent of carbohydrates was calculated by subtraction. Analyses were done in duplicate.

3.6.5. Nutrition Analysis

Nutrition analysis of the control bread and bread enriched with mealworm powder was performed using Genesis R & D Software (ESHA Research, Salem, OR; Version 9.12.1.) at the University of Central Oklahoma.

3.7. Dough Rheological Properties

Rheological behavior analysis was performed using AACCI approved method for Farinograph (54-21.02), constant flour procedure. The apparatus was Farinograph E equipped with a 50 g bowl. (C.W. Brabender Instruments Inc., South Hackensak, NJ). Analyses were done in triplicate. The experiment was performed at the Food and Agriculture Products Center at Oklahoma State University in Stillwater, Oklahoma.

3.8. Flour Properties

The functional properties experiments of 0%, 10%, and 20% non-defatted mealworm powder-fortified all-purpose flour were conducted in collaboration with the Food Science Lab in the Human Environment Sciences and Chemistry Departments at the University of Central Oklahoma. The functional properties experiments were conducted as follows: Water Holding Capacity, Emulsion Stability, Foaming Stability, and Solubility (Alu’datt et al., 2012).

3.8.1. Water Holding Capacity

Aqueous dispersion of the sample was prepared at 16% w/v. 16 g of the flour sample was measured with 100 mL of distilled water. For 10% substitution of mealworm powder, the grams of the sample were adjusted to 14 g all-purpose flour and 1.6 g of mealworm powder; for 20%, 12 g of all-purpose flour and 3.2 g of mealworm powder were used. These dispersions then were heated at 90 °C for 30 minutes in a hot water bath. The now gel-like substance was cooled to 4 °C and maintained this temperature for 24 hours. The gels then were centrifuged at 10,000 x G for 15 minutes, and the separated supernatant layers were measured and recorded.

3.8.2. Emulsion Stability

The emulsion was prepared by using 2 g of each sample and mixing the sample with 20 mL of distilled water and 20 mL of olive oil. This emulsion then was blended for 120 seconds at 1600 rpm and then transferred to 4 calibrated centrifugal tubes. The tubes containing the emulsion were heated at 80 °C for 30 minutes in a water bath. The tubes then were cooled for 15 minutes using tap water. The total height of the emulsion was measured within the tubes, and then the tubes were centrifuged at 1500 G for 15 minutes. The height of the emulsified layer (H1) was measured immediately. Then by following the formula, the emulsion stability was measured (Alu’datt et al., 2012).

$$(\%) = (H1/HT) \times 100\%$$

3.8.3. Foam Stability

2 g of sample control was mixed with 40 mL of distilled water using a blender. The grams of non-defatted mealworm powder and all-purpose flour were adjusted based on 0%, 10% and 20% of non-defatted mealworm powder. Each mixture was mixed for 5 minutes at 1600 rpm in the blender, alternating between blending and shaking to produce a foam. The volume of the foam was recorded immediately following the mixing (VT), and then a second recording was measured after 60 minutes (V1). By using the below formula, the foaming stability was calculated (Alu'datt et al., 2012).

$$\% = (V1/VT) \times 100$$

3.8.4. Solubility Test

2 g of each sample were dispersed in 40 mL of distilled water and then stirred for 30 minutes. The pH of the solution was adjusted to 7 using a phosphate buffer. The buffered dispersion was adjusted to room temperature. The solution was centrifuged at 2000 x G for 15 minutes. Then data was obtained by reading each tube.

3.9. Statistical Analysis

All analyses were conducted in triplicate. Means, standard deviation, and standard error were computed using Microsoft Excel 2017. A T-test was calculated between the two variables of ethanol and hexane for the defatting procedure experiment, and to determine the significance of the mean value. All data from the flour properties were analyzed by the ANOVA table. A P-Value of less than 0.05 was considered statically significant.

CHAPTER FOUR: RESULTS AND DISCUSSION

Table 2 illustrates the proximate analysis of the non-defatted mealworm powder that has been done at Eurofins Scientific Inc. center. This table shows the amount of protein, ash, calories, carbohydrates, moisture, and fat in the mealworm powder.

Table 2: Nutrition Analysis of non-defatted mealworm Powder (%)

Nutrition Analysis of Original Mealworm Powder (%)	
Protein	51.82 %
Ash	5.82 %
Calories	509 Kcal /100 g
Carbohydrates	10.71%
Moisture	2.9 %
Fat	28.75 %

4.1. Fat percentage in mealworm powder defatted by ethanol and hexane

Fat was extracted from mealworm powder by using ethanol and hexane. As shown in Figure 15, the non-defatted mealworm powder has a fat content of 28.75%. The fat extracted using ethanol was 20.5%, and the fat extracted using hexane was 23.1%. Hence, carrying out the defatting procedure using the two solvents, ethanol and hexane, could reduce the fat content to 8.24% and 5.68%.

Defatted mealworm powder was sent for further analysis to an outside laboratory to do a proximate analysis. Figure 16 illustrates the amount of fat that remained in the defatted mealworm powder after the extraction. It shows the reduction of the fat present in the final powder that will be used for further analysis. The results indicate that the difference in the final amounts of fat between the two solvents is 2.56%, which demonstrates that hexane is ideally a better solvent in comparison to ethanol. Figure 15 and 16 illustrate the amount of fat that was extracted from the mealworm powder by using ethanol and hexane and the amount of fat remaining after the defatting procedure.

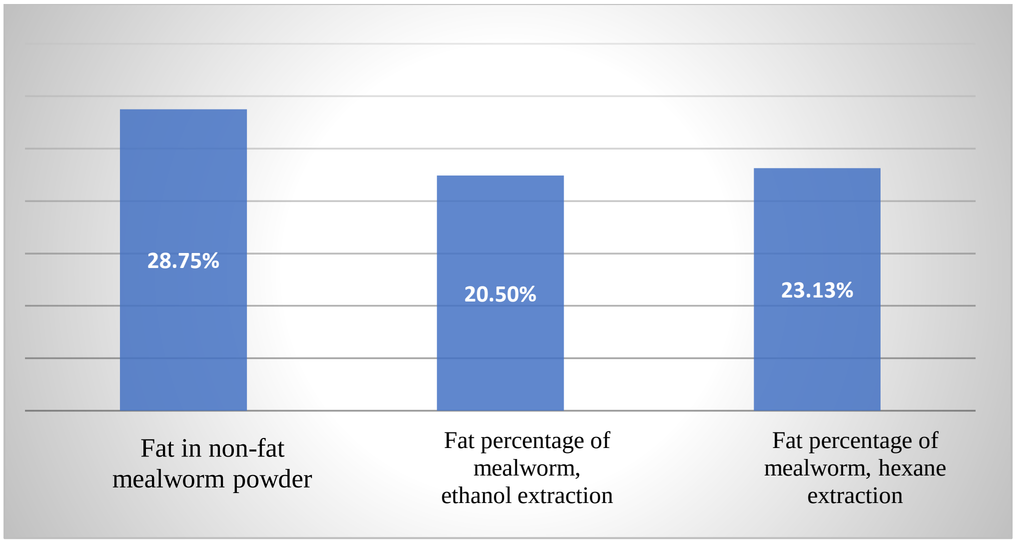

Figure 15: Fat percentage extracted by using ethanol and hexane.

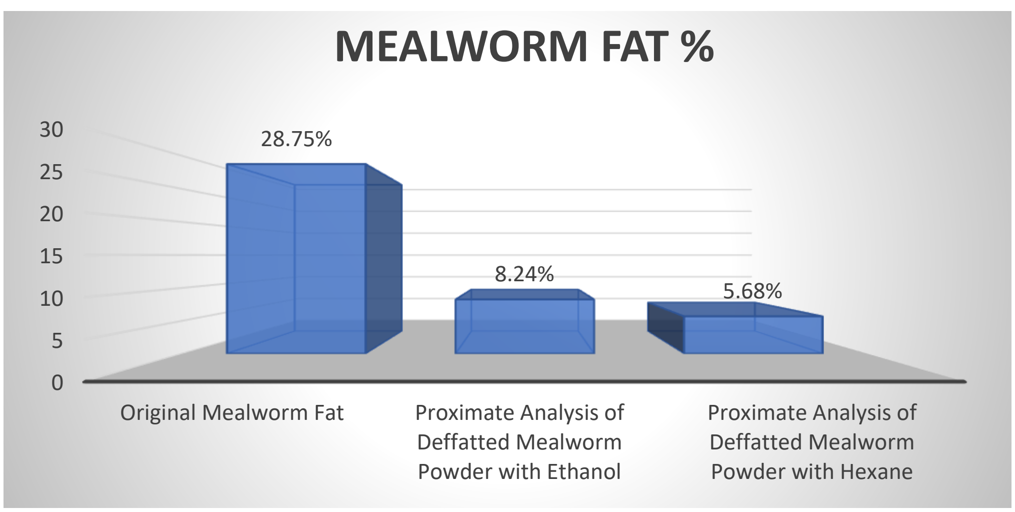

Figure 16: Mealworm fat percentage after defatting procedure.

Table 3 illustrates the proximate analysis of the average amount of protein, ash, calories, carbohydrates, moisture, and fat compared within the defatted mealworm powders of the two solvents. According to researchers, the freeze-dried mealworm contains about 43% protein, 51% crude protein, and 33% fat, and the extracted oil is rich in palmitic and oleic acid (Zhao et al., 2016). Also, mealworm powder contains some amino acids. As evidenced by Yi et al. (2013), all insect species are comparable to the essential amino acid levels in soybean protein. Another study's results interpreted that mealworm contains approximately 70% of protein and about 10% of fat (Kuntadi et al., 2018).

Table 3: Comparison of protein, ash, calories, carbohydrates, moisture and fat in defatted mealworm powder with ethanol and hexane.

	Protein	*Ash*	*Calories*	*Carbohydrates*	*Moisture*	*Fat*
Defatted MW With Ethanol	61.4 ± 5.97	4.2 ± 0.48	373.6 ± 12.6	14.1 ± 3.03	11.9 ± 3.45	8.24 ± 0.90
Defatted MW With Hexane	69.23 ±0.15	5.40 ±0.23	390 ±1	15.4 ± 0.62	4.23 ± 0.28	5.68 ± 0.14
P Value	0.15	0.048	0.21	0.63	0.08	0.05

Means ± standard deviation of 3 measurement.
T-test was obtained between two variances of ethanol and hexane.
The P value of < 0.05 was obtained by using P (T<=t) two-tail.
The P value of fat and ash in the hexane-defatted mealworm shows significant difference.

4.2. Bread Results:

Overall, bread enriched with 10% and 20% of non-defatted mealworm powder showed similar appearance in comparison to the control bread, with the exception of a color difference that appeared darker. Figure 7 shows the differences in all three breads and shows how the color changed as mealworm powder increased.

4.2.1. Nutrition Label

The results in Figure 16 indicate the nutritional content of the control bread, the bread fortified with 10%, and the bread fortified with 20% mealworm powder. According to the obtained data, the bread fortified with insect powder increased in nutritional value, especially in the protein and fat content. Due to the high fat content present in the bread fortified with 20% non-defatted mealworm powder, this process can be modified in future research by using the defatted mealworm powder for further analysis.

Nutrition Facts
Serving Size (74g)
Servings Per Container

Amount Per Serving	
Calories 150	Calories from Fat 5
	% Daily Value*
Total Fat 0g	**0%**
Saturated Fat 0g	**0%**
Trans Fat 0g	
Cholesterol 0mg	**0%**
Sodium 35mg	**1%**
Total Carbohydrate 32g	**11%**
Dietary Fiber 1g	**4%**
Sugars 2g	
Protein 5g	
Vitamin A 0% • Vitamin C 0%	
Calcium 0% • Iron 8%	

*Percent Daily Values are based on a 2,000 calorie diet. Your daily values may be higher or lower depending on your calorie needs:

	Calories:	2,000	2,500
Total Fat	Less than	65g	80g
Saturated Fat	Less than	20g	25g
Cholesterol	Less than	300mg	300mg
Sodium	Less than	2,400mg	2,400mg
Total Carbohydrate		300g	375g
Dietary Fiber		25g	30g

Calories per gram:
Fat 9 • Carbohydrate 4 • Protein 4

Nutrition Facts
Serving Size (74g)
Servings Per Container

Amount Per Serving	
Calories 160	Calories from Fat 15
	% Daily Value*
Total Fat 1.5g	**2%**
Saturated Fat 0g	**0%**
Trans Fat 0g	
Cholesterol 0mg	**0%**
Sodium 35mg	**1%**
Total Carbohydrate 30g	**10%**
Dietary Fiber 1g	**4%**
Sugars 2g	
Protein 6g	
Vitamin A 0% • Vitamin C 0%	
Calcium 0% • Iron 8%	

*Percent Daily Values are based on a 2,000 calorie diet. Your daily values may be higher or lower depending on your calorie needs:

	Calories:	2,000	2,500
Total Fat	Less than	65g	80g
Saturated Fat	Less than	20g	25g
Cholesterol	Less than	300mg	300mg
Sodium	Less than	2,400mg	2,400mg
Total Carbohydrate		300g	375g
Dietary Fiber		25g	30g

Calories per gram:
Fat 9 • Carbohydrate 4 • Protein 4

Nutrition Facts
Serving Size (74g)
Servings Per Container

Amount Per Serving	
Calories 170	Calories from Fat 25
	% Daily Value*
Total Fat 3g	**5%**
Saturated Fat 0g	**0%**
Trans Fat 0g	
Cholesterol 0mg	**0%**
Sodium 35mg	**1%**
Total Carbohydrate 27g	**9%**
Dietary Fiber 1g	**4%**
Sugars 2g	
Protein 8g	
Vitamin A 0% • Vitamin C 0%	
Calcium 0% • Iron 8%	

*Percent Daily Values are based on a 2,000 calorie diet. Your daily values may be higher or lower depending on your calorie needs:

	Calories:	2,000	2,500
Total Fat	Less than	65g	80g
Saturated Fat	Less than	20g	25g
Cholesterol	Less than	300mg	300mg
Sodium	Less than	2,400mg	2,400mg
Total Carbohydrate		300g	375g
Dietary Fiber		25g	30g

Calories per gram:
Fat 9 • Carbohydrate 4 • Protein 4

Control Bread   10 % Mealworm Powder   20% Mealworm Powder

Figure 12: Nutrition facts comparison of bread with 0%, 10% and 20% non-defatted mealworm powder.

4.2.2. Nutrition Analysis of Bread Enriched with Mealworm Powder

The bread with 0%, 10%, and 20% was subjected to a proximate analysis. Table 4 shows the nutrition analysis of bread enriched with non-defatted mealworm powder.

Table 4: Nutritional composition of bread enriched by 0%, 10%, and 20% non-defatted mealworm powder.

Sample ID/FAPC	% Ash	% Fat	% Protein	% Carbohydrates
Control/143-18	0.89	2.43	9.82	70.63
10% MW/144-18	1.04	4.69	12.65	60.91
20% MW/145-18	1.34	7.29	15.78	55.77

According to Table 5, the proximate nutritional composition of the bread shows higher values when compared to the control bread. The protein content levels in the bread made with 10% and 20% mealworm powder was higher in comparison to the control bread. The amount of protein increased to 15.78% by increasing the level of mealworm powder. Hence, adding the mealworm powder can increase the nutritional value of bread (González et al., 2018). The average ash content percentage in cereals, breads, and pasta varies from 1.5% to 2.5%. In this experiment, the percentage of ash of the control bread was lower than 10% and 20% fortified bread. The data indicates less carbohydrates in the 20% mealworm powder bread when compared to the control bread. On the other hand, the 20% mealworm powder shows a higher protein and mineral content (González et al., 2018). The fat content of the bread made with 10% and 20 % MWP resulted in a higher fat content, which could be corrected by using the defatted powder in future analysis of this project.

4.2.3. Moisture Content of Bread

In this experiment there was no significant difference in moisture content of the control bread after air-drying method compared with 10% and 20% mealworm fortified bread. The percent moisture content (wet weight basis) in enriched white bread is approximately 13.4 % (Nielsen, 1998). Bread made with mealworm powder has high a moisture rate, which could be the result of high fat content. The fat content in bread prevents water evaporation during the baking process, which leads to a high moisture content (González et al., 2018). According to Table 5, the average mean in control bread made with all-purpose flour, 10 %, and 20% enriched with MWP after air-oven drying are 34.72 ± 1.86 , 34.44 ± 1.74 , and 33.66 ± 2.14, which this data indicates that after drying there is not significant difference.

Table 5: The moisture content of bread by air-oven drying method.

Sample	*Mean ± SD*
Control	34.72 ± 1.86a
10% MW	34.44 ± 1.74b
20% MW	33.66 ± 2.14c

Means ± standard deviations of 3 measurements.
The variance analyzed according to Tukey test P value < 0.05.
The P value is greater than 0.05 in this experiment and the result is not significantly difference.

4.2.4. Color Measurement of bread enriched with mealworm powder

Results obtained from this study show the average L, a, and b values which were significantly different in all three samples. Bread made with all-purpose flour was lighter in color in comparison with fortified bread with 10 % and 20 % MWP. All bread formulation with insect powders shows lower luminosity (L), increased redness (a), and yellow tonality (b) (González et al., 2018). As it was observed in Figure 12, bread enriched with mealworm powder showed similar appearance to all-purpose control bread, however; as the level of mealworm increased the crust color became darker and showed that the presence of 20% mealworm powder gave it a brownish crumb. A similar study has previously shown the value of a and b increase by increasing the amount of chickpea flour in wheat bread and producing a darker color. This was attributed to increasing the Maillard reaction during baking due to high protein content (Mohammed, Ahmed, & Senge, 2012). The Maillard reaction usually happens if food with high protein, carbohydrates, and moisture are at the temperature above 50 C (Ramirez-Jimenez, Guerra-Hernández, & García-Villanova, 2000)

Table 6: Comparison of color results of all-purpose flour bread fortified with non-defatted mealworm powder.

Treatment*	Color parameter		
	L*	a*	b*
Mealworm 0 %	72.0 a	0.9 c	25.0 c
Mealworm 10 %	42.9 b	5.4 b	26.0 b
Mealworm 20 %	34.4 c	7.5a	28.0 a

Different superscripts within columns indicate significant differences among treatment ($p<0.05$)
L* = lightness; a* = redness (+) and blueness (–); b* = yellowness.
The results were processed by one-way analysis of variance (ANOVA).
The significance of differences was determined by Tukey HSD using SPSS.

4.2.5. Effect of mealworm powder on bread firmness

Bread containing mealworm powder has similar appearance to the control with the exception of crumb color and volume. The Figure 18 shows the firmness of bread is significantly different in enriched bread with 10% and 20% compared to control bread. The firmness of bread shows higher on day one and three compared to the control bread. On day seven the firmness of enriched bread with 10% and 20% of mealworm powder is higher than control, which could be the result of high fat content in mealworm powder.

Enriched bread with insect powder has harder crumbs, a higher density, and lower elasticity than non-enriched bread (González et al., 2018). Presence of a higher protein content causes the crumb to become thicker because it surrounds the air cell, which strengthens the structure (Mohammed et al., 2012). Bread containing mealworm powder shows similar characteristics to regular bread. Although, the bread containing 20 % of mealworm powder showed lower volume and darker color with thicker crust, which is attributed to higher fat content of mealworm powder (González et al., 2018).

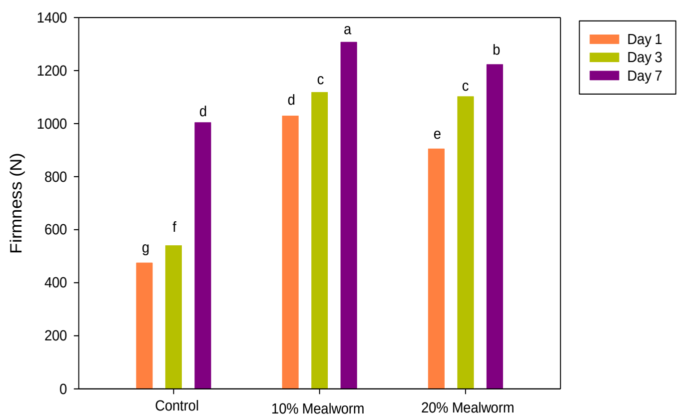

Figure 13: Effect of 10% and 20% of non-defatted mealworm powder on bread firmness.

4.3. Dough Rheological Properties

The presence of non-defatted mealworm powder affected the rheological behavior of the dough during mixing and carried significant changes on the dough mixing behavior. Figure 18 illustrates the changes in composition of mixing (all-purpose flour and mealworm powder in 10% and 20%), protein increased by 40 and 80% with MWP substitutions of 10 and 20% respectively; fat also increased by 229 and 458%, and moisture decreased by 7.8 and 15.7% at 10 and 20% mealworm powder substitution, respectively.

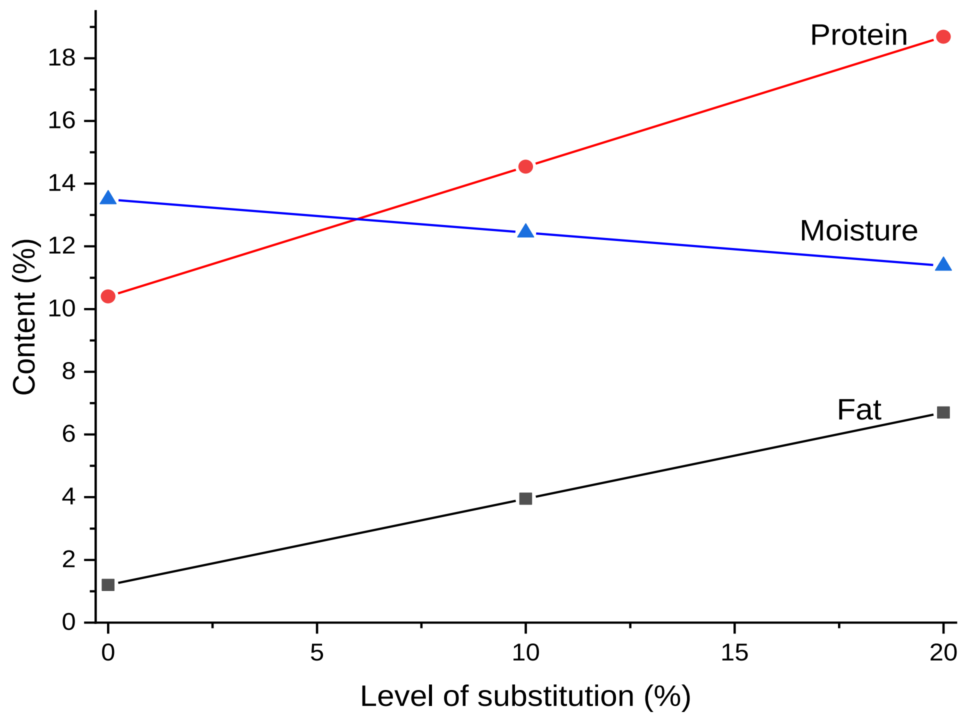

Figure 14: Effect of mealworm powder substitution on protein, moisture and fat content by mass balance.

4.3.1. Water Absorption, Developing Time, and Stability

Water absorption significantly decreased by 8.6 % and 15.6 % when 10% and 20% of flour was substituted by mealworm powder, respectively. The study was approved by González et al., (2018) that the presence of five percent of insect powder decreases the water absorption in comparison with control flour. Absence of starch and high content of protein can reduce water absorption, which mostly is the result of amino acids in insect powders (González et al., 2018). Studies show, protein extract from mealworm contains essential amino acids such as lysin, leucine, etc. (Zhao et al., 2016). Figure 19 shows the relationship between water content behavior, mealworm powder substitution, and fat. The effect of fat and mealworm powder substitution in water absorption of the dough was linear (Fig. 20), but with different slopes. While 1% of change on level of substitution causes -0.47% of changes on water absorption, 1% on the fat content causes -1.72% changes on water absorption. These two parameters, fat content and level of substitution, are related, and they have similar statistical parameters like R^2 (0.9914) and residual (1.1606). MWP substitutions (10 and 20%) in all-purpose flour resulted in increased protein by 40 and 80% and fat by 229 and 458%, respectively.

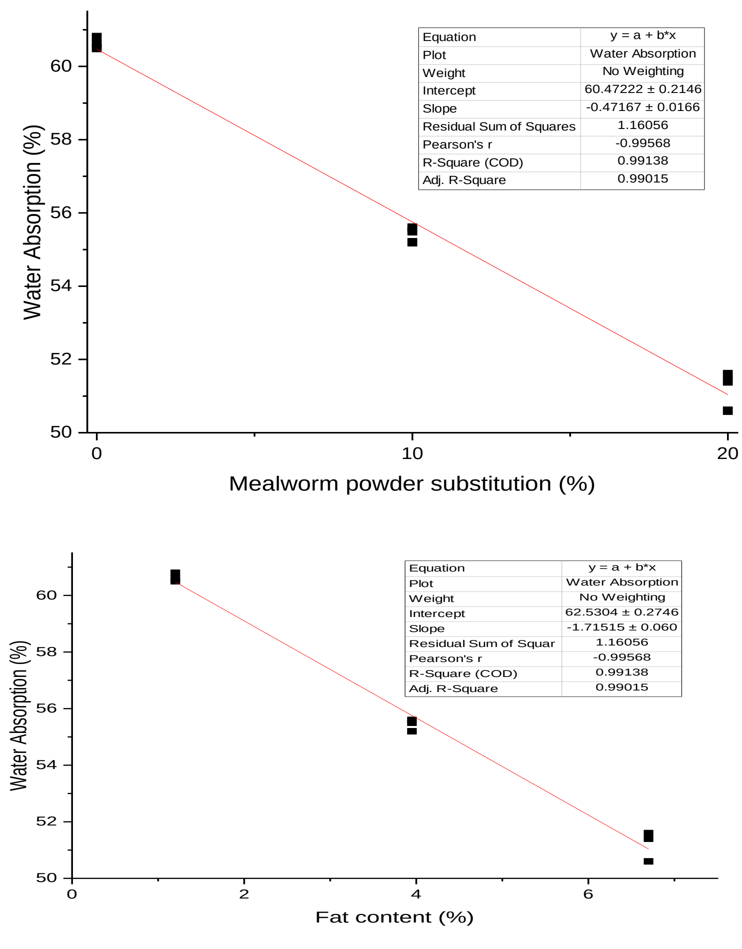

Figure 20: Relationship between water content vs a) MWP substitution and b) fat content

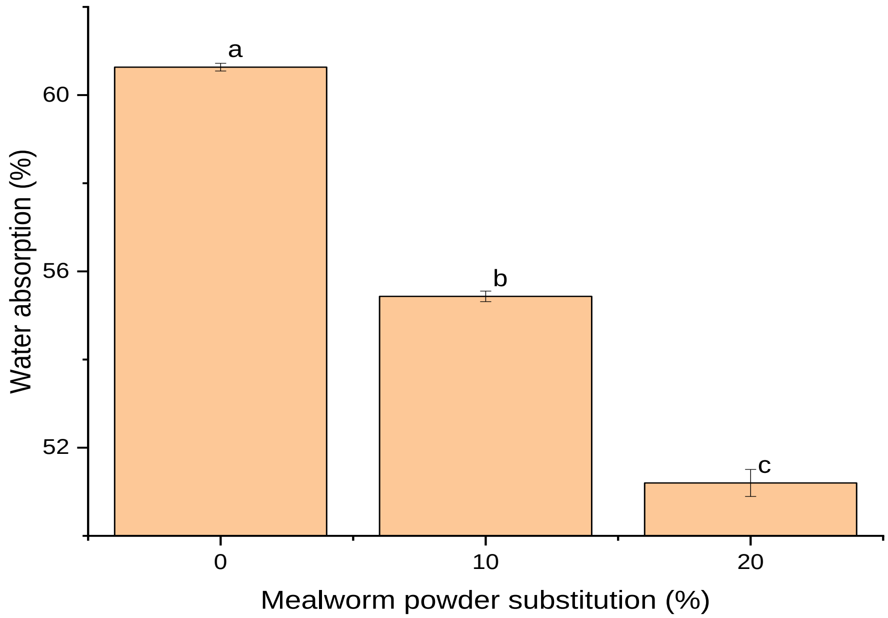

Figure 21: Water absorption behavior vs. mealworm powder substitution

Development time was significantly reduced by 27% when comparing control versus the treatments, but not between treatments. According to González et al., (2018), developing time increases when comparing *A. domestica or H. illucens* (insect powder) to control. In a different study, development time was significantly higher ($p < 0.05$) when wheat-chickpea flour was compared with control (Mohammed et al., 2012).

A similar trend was observed for dough stability. Stability significantly decreased by adding 10 % and 20 % mealworm. According to González et al., (2018), water stability and development time are showing the strength of the flour and stronger dough. In the same study, the presence of *H.illucens* powder (insect powder) shows higher stability due to higher fat content (González et al., 2018); however, in our experiment we observed lower stability and dough development time, which can be the result of composition of mealworm powder versus other insects.

Table 7 indicates how the level of non-defatted mealworm substitution affected water absorption, development time, and stability. The non-defatted mealworm powder substitution significantly decreased water absorption. The effect on the rheological properties is related to changes in fat content.

Table 7: Water absorption and mixing characteristics of flour and non-defatted mealworm powder substitution, by farinograph [a].

Level of substitution	**Water absorption 14 % m.b (%)**	**Developmnt Time (min)**	**Stability (min)**
Control (0%)	60.6 ± 0.1 a	2.0 ± 0.11 a	6.3 ± 0.90 a
10%	55.4 ± 0.1 b	1.4 ± 0.06 b	1.3 ± 0.10 b
20%	51.2 ± 0.3 c	1.3 ± 0.06 b	1.3 ± 0.10 b

[a] Means (n=3) ± standard error followed by different letter within same column are significantly different (Tukey, $p < 0.05$).

[b] Water absorption to reach 500 UF, and reported at 14% moisture basis.

4.4. Flour properties

Table 9 shows the effect of 10 % and 20 % of the non-defatted mealworm powder being added to all-purpose flour as well as the different effects on flour properties. According to obtained data, the solubility is decreasing as the amount of mealworm powder increases, and this could be related to higher fat and protein content of mealworm powder or could be the result of the pH (pH=7) of the solution (Suliman, El Tinay, Elkhalifa, Babiker, & Elkhalil, 2006). The foaming stability decreased gradually as mealworm powder increased, which is attributed to the presence of fat in flour enriched with 20% mealworm powder (Heywood, Myers, Bailey, & Johnson, 2002); however a different study showed that higher foam stability was achieved by making foam with non-hydrolyzed cricket protein (Hall, Jones, O'Haire, & Liceaga, 2017). Water holding capacity also increased in 20% mealworm powder, which can be the result of high fat (hydrophobic) and a lower hydrophilic binding substance (Heywood et al., 2002). The presence of hydrophobic material such as fat could lower hydrophilic binding for protein to hold water (Heywood et al., 2002). A study reported the highest emulsion stability for fortified wheat flour was with barley flour, however, there were significant differences in the decrease of emulsion stability, which could be the result of the composition of mealworm powder and fat content. Therefore, there was a difference between 10 % mealworm powder, control, and 20 % mealworm powder in comparing the standard deviation. Hence, the P value is lower than 0.05, so the results are significantly different (Alu'datt et al., 2012).

Table 8: Comparison of functional properties in all-purpose flour and non-defatted mealworm powder

Groups	*Solubility*	*Foaming Stability*	*Emulsion Stability*	*Water Holding Capacity*
Controls	41 ± 1.47 a	0.81 ± 0.10 a	0.36 ± 0.03 a	40.0 ± 0.50
10% MW	39.53 ± 1.84 *b*	0.78 ± 0.14 b	0.36 ± 0.02 a	40.2 ± 0.17
20% MW	38.14 ± 0.96 c	0.72 ± 0.09 c	0.35 ± 0.03 a	40.8 ± 0.75

Means ± standard deviations of 3 different measurement shows significantly different. ANOVA single factor were obtained for each experiment.

CHAPTER FIVE: CONCLUSION

In this study, the data obtained from defatting mealworm powder indicated that ethanol and hexane both are good solvents for defatting. The fat percentage, the most crucial factor, seemed to show a high reduction from the original mealworm powder, which contained about 28.5% to 5.68% after the defatting process proving both solvents to be effectively efficient.

Proximate analysis of the mealworm powder shows a higher protein content of 69.1% after defatting, which proves that the mealworm powder can be a good alternative source of protein in the food industries.

Moisture content did not appear to be greatly affected by the defatting or protein extraction process. The results indicate that there is a noticeable difference in the nutrient compositions of mealworm powder. Further research is required to do amino acid profiling for the defatted mealworm powder.

Data from the bread indicates that mealworm powder or edible insects in the form of powder could be incorporated into baked products to enhance the nutritional value, especially protein (González et al., 2018). The result from the bread confirmed that by increasing the amount of mealworm powder the nutrient value of the bread increased.

Sensory testing needs to be performed to determine the effect of fortification on food products and to determine the shelf-life of the baked goods enriched with mealworm powder that include a high protein source.

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