PROXIMATE COMPOSITION, FUNCTIONAL PROPERTIES, AND MINERAL COMPOSITION OF WHOLE MEAL, DEFATTED, AND PROTEIN HYDROLYSATES FROM EDIBLE GRASSHOPPER

ABSTRACT

Edible grasshoppers were harvested, sorted, degutted, washed with boiling water for 20 min, oven dried and milled into whole grasshopper protein meal (WGM). The WGM was defatted using acetone to obtain defatted grasshopper protein meal (DGM). Grasshopper protein hydrolysate (GHM) was produced by hydrolyzing DGM with pepsin and pancreatin enzymes to mimic GIT digestion of proteins in humans. The grasshopper samples were evaluated for proximate composition, functional properties, and minerals composition using standard methods. The proximate results revealed, moisture, fat, crude protein, fibre, ash and carbohydrate ranged from 7.33-15.54 %, 0.15-17.55 %, 37.45-87.54 %, 6.34-6.43 %, 0.23-2.76 % and 4.75- 23.27%, respectively. The functional results showed oil absorption capacity ranged between (131.32-212.43 %), water absorption capacity (201.00-237 %), gelation concentration (11.00-23 %), foaming capacity (324.00-367 %), foaming stability (65.00-87.00 %), emulsification capacity (35.43-42.54 m2/g), emulsion stability (67.54-81.43 %) and invitro protein digestibility (67.54-86.54 %). Also, the mineral results revealed, sodium ranged between (43.43-301 %), calcium (1.43-15.43 %), potassium (241.54-2143.53 %) and phosphorus (0.98-7.43 %) showed significant difference ($p<0.05$). The different samples of the studied grasshoppers were high in proteins, essential macromolecules and minerals needed for optimal human health. It is recommended that animal producing farmers should be encouraged to grow grasshoppers to make it easily accessible and available all year round for consumers and researchers.

Keywords: Edible grasshopper protein**,** protein hydrolysate**,** proximate composition**,** functional properties**,** mineral composition.

1.0 INTRODUCTION

The lack of food resources has become a major problem for modern society, and the majority of developing nations struggle to feed their populations. As a result, protein-energy malnutrition is caused by inadequate protein consumption (1). Edible insects are a unique food ingredient with great potential to contribute to global food security, and provide an interesting food alternative. Food security is predicted to be exposed to stress in the coming decades due to rapid global population growth and rising animal protein demand. For thousands of years, edible insects have been used as food to alleviate hunger and improve malnutrition. Some insects have also been used as medicines because of their therapeutic properties. The search for alternative source of food nutrient remains a perpetual event as human population growth is dynamic and ever-increasing under-exploitation and under-utilization of abundant alternative natural resources have now been recognized as one of the militating factors against nutrient glut (1). Over the last decade, the urgency to find alternative and sustainable protein sources has prompted an exponential increase in the interest in insects as a human food source (2). Edible insects contribute suitable amounts of energy and protein, fatty acids, and micronutrients to the human diet. Nutritional values of insects can be manipulated to meet specific

needs. Edible insects in food-insecure countries can contribute to improving diets and preventing undernutrition (2). Edible insects have recently received attention as novel sustainable ingredients owing to their high nutritional value, particularly as an alternative protein sources for animals. Moreover, insects are considered economical and environmental-friendly materials because they require minimal feeding and maintenance as well as emit less greenhouse gases when compared with conventional livestock.The excellent nutritional value of edible insects, especially as an alternative source of animal protein, has drawn attention to them recently as a novel sustainable element. Furthermore, compared to conventional livestock, insects create fewer greenhouse gases and require less upkeep and feeding, making them cost-effective and environmentally friendly resources. According to Capinera (3) on a review of the nutritional composition of 236 edible insects (4), insects are high in energy, with 2-60 % fat on a dry matter basis, which has a high proportion of mono-and polyunsaturated fatty acids, provide satisfactory protein (20-80 %) which meets the human amino acid requirements, are high in minerals such as calcium, copper, iron, phosphorus, magnesium, manganese, and potassium, have an abundance of vitamin A and carotenoids, and though in low amounts, they can contain B vitamins such as riboflavin, pantothenic acid, and sometimes, folic acid. Besides their potential contribution to dietary nutrient intakes, thus improving health, insects are also important for improving and conserving the environment as well as contributing to incomes and livelihoods. Grasshoppers are important sources of proteins, formed by complex biomolecules, found in cells, tissues, and other macromolecules involved in nutrition and protein metabolism. Grasshoppers are good nutritional source, having about 75% protein, 6% fat, 7% crude fibre, 8% carbohydrates depending on the species (5). Bio-active compounds are extra-nutritional constituents that typically occur in small quantities in foods. Besides being a source of valuable nutrients, studies have found bioactive compounds in insects with characteristics that could have the potential to reduce health risks and strengthen the immune system (6). Generally, bioactive peptides are natural compounds of food or part of protein that are inactive in the precursor molecule. However, they may be active after hydrolysis and can be transported to the active site. Biologically active peptides can also be synthesized chemically and characterized. Peptides have many properties, including antihypertensive, antioxidant, antimicrobial, anticoagulant, and chelating effects. These days, food is regarded as a source of physiologically active molecules in addition to dietary compounds that may improve human health and environmental conditions. Customers' selection of raw materials high in vitamins, minerals, and other bioactive substances like polyphenols, peptides, or essential oils reflects their increased understanding of how nutrition affects health (7). With a high fat content and numerous essential minerals and vitamins, grasshoppers are an affordable and excellent source of protein. This study's main goal was to determine whole meal, defatted, and protein hydrolysates from edible grasshopper

2.0 MATERIALS AND METHODS

2.1 Material Procurement

The live edible grasshoppers were obtained from the grasshopper market in Shagari Low Cost, Maiduguri, Borno State and the test diet formulation materials; vitamin premix, casein powder, salt,

vegetable oil were purchased from sudo-pee supermarket in Makurdi, Benue State. For additional processing, these were brought to the Department of Food Science and Technology at Joseph Sarwuan Tarka University in Makurdi, Benue State. Experimental Albino rats were purchased from the National Institute of Trypanosomiasis Research, Vom, Plateau State.

2.1.1 Preparation Whole grasshopper protein meal (WGM)

Edible grasshopper were prepared according the method described by (8) as shown in Figure 1. Edible grasshopper were treated with hot water at 100ºC to kill them and further sorted to remove extraneous materials, washed and oven dried at 55ºC for 10hrs followed by the removal of the wings and viscera and milled into whole grasshopper meal.

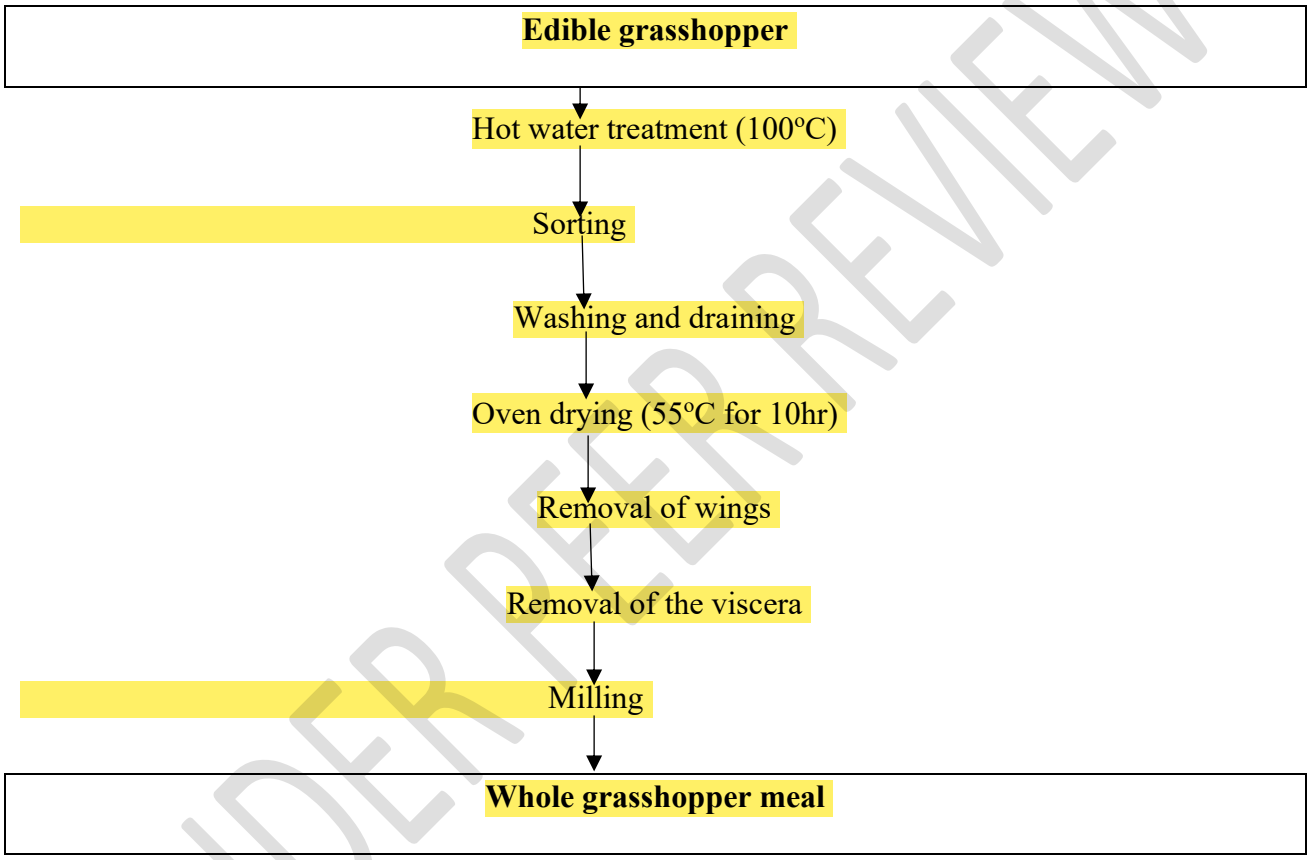


Fig 1. Production of Whole Grasshopper Meal

Source: (8) with modification

2.1.2. Preparation of Defatted Grasshopper Protein Meal (DGM)

Prior to enzymic hydrolysis, the ground whole grasshopper protein meal (WGM) was defatted by mixing 1g with 10ml of acetone (8). After three hours of stirring in a fumehood, the mixture was decanted, and the residue underwent two and three sequential acetone extractions. The resultant

defatted grasshopper meal (DGM) was air-dried in the fume hood for an entire night and kept for use in future research (Figure 2).

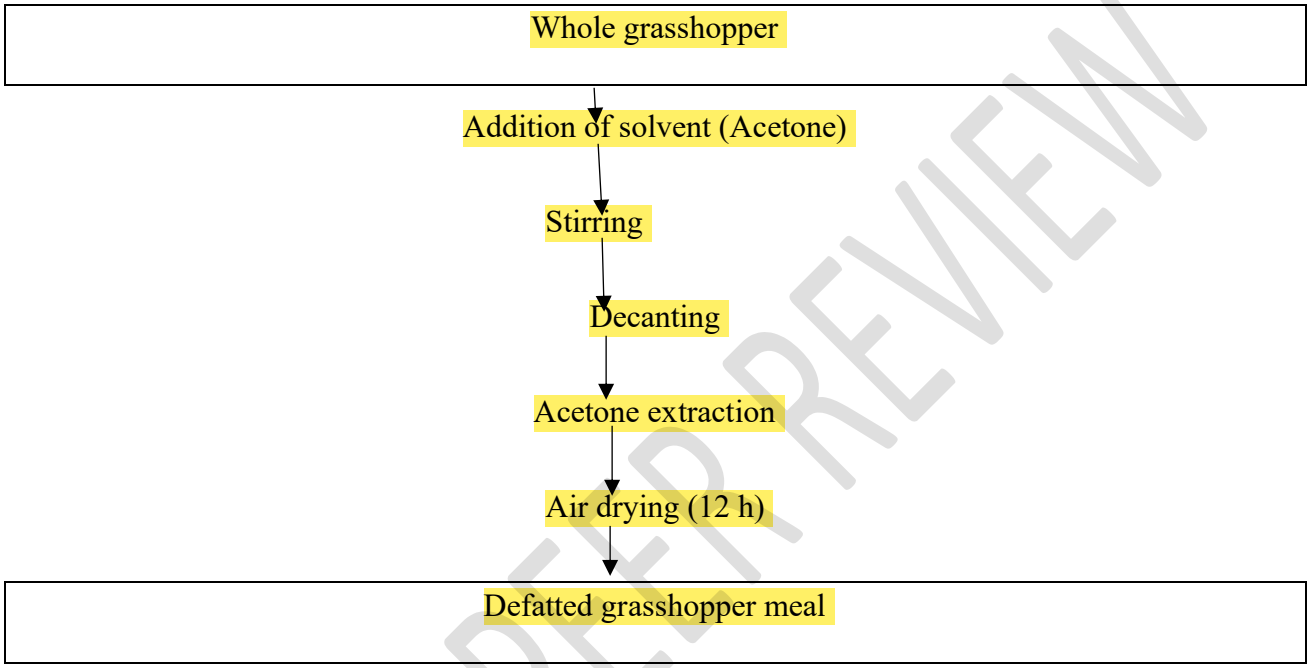
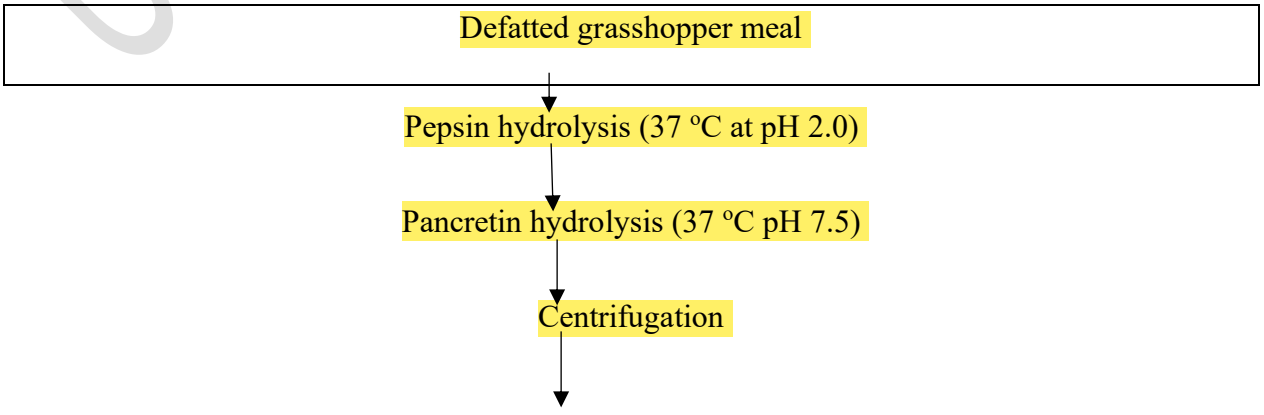


Fig 2. Production of Defatted Protein Meal

Source: Source: (8) with modification

2.1.3 Preparation of grasshopper protein hydrolysate (GHM)

The grasshopper protein hydrolysate was prepared using two enzyme (pepsin and pancreatin) to mimic gastrointestinal digestion of proteins, it was centrifuge at 2500rpm for 15 min and the supernatant was lyophilized



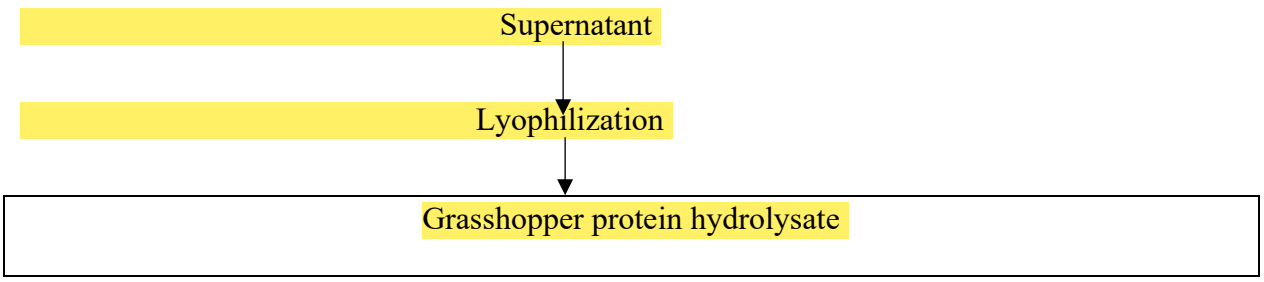


Figure 3. Production of Grasshopper Protein Hydrolysate

Source: (8) with modification

Table 1: Blend Formulation for Feeding Trial

	SAMPLES			
INGREDIENTS	**A** (Control) (g)	**B** (g)	**C** (g)	**D** (g)
WGM	0	15		-
DGM	-	-	15	-
GPH	-	-	-	10
Cornstarch	40	40	40	40
Casein	20	5	5	10
Sucrose + Maltose	23.2	23.2	23.2	23.2
Soy oil	7.00	7.00	7.00	7.00
Rice Husk	5.0	5.0	5.0	5.0
Min-vitamins	4.80	4.80	4.80	4.80

Key: A (Control) = Sample without test ingredients; B (WGM-) Whole grasshopper Meal, C (DGM) - Defatted grasshopper Meal ; D (GPH) – grasshopper Protein Hydrolysate

2.2 Determination of Proximate Composition of Edible Grasshopper Meal

2.2.1 Moisture content determination using oven method

The air oven drying method was used to determine the moisture content, as given by (9). An oven (GENLAB, England B6S, serial no. 85K054) was used to dry a clean dish with a lid for 30 minutes at 100°C. It was weighed after cooling in a desiccator. Exactly, 2 g of the material was weighed and added to the plate.After that, the dish and its contents were dried to a relatively consistent weight at 105°C in the oven. The weight loss from the initial sample (before to heating) was expressed as a percentage of moisture.

$$\%\ Moisture = \frac{weight\ loss\ (W2-W3)}{Weight of Sample\ (W2-W1)}\ x\ 100 \quad (1)$$

Where

$W1 = weight\ of\ dish,$

$W2 = weight\ of\ dish + sample\ before\ drying,$

$W3 = weight\ of\ dish + sample\ after\ drying.$

2.2.2 Determination of crude protein

Crude protein was determined using the Kjeldahl technique of (9). A digital weighing balance was used to weigh two (2 g) samples into a Kjeldahl digestion flask (3000 g x 0.01g 6.6LB). A 0.88 g catalyst combination containing 0.5% selenium dioxide, 3.5% copper sulphate, and 96% anhydrous sodium sulphate was added. After adding 7 ml of concentrated sulphuric acid, the flask's contents were mixed by swirling. In the fume chamber, the Kjeldahl flask was gradually heated in an inclined position until no sample particles remained on the flask's side. The flask was periodically shaken as the solution was heated more vigorously to bring the liquid to a boil until a clear solution was achieved. After letting the mixture cool, it was diluted with 25 millilitres of distilled water in a volumetric flask. The diluted digest was placed in a steam distillation apparatus in ten millilitres. 8 ml of 40% NaOH was added to the digest to make it alkaline. To the receiving flask, 5 ml of 2 % boric acid solution was added and 3 drops of mixed indicator was dropped. The delivery tube was dipped into the 100 ml conical flask and titrated with 0.01 M HCl before the distillation apparatus was attached to the receiving flask. There was a blank titration. The following formula was used to determine the proportion of nitrogen:

$$\%\ Nitrogen = \frac{(S-B)\times 0.0014\times 100\times D}{sample\ weight} \quad (2)$$

Where, S = sample titre, B = Blank titre, S - B = Corrected titre, D = Dilution factor

% Crude Protein = % Nitrogen x 6.25 (correction factor).

2.2.3 Determination of crude fat

The solvent extraction method described by (9) was used to determine crude fat. A 5 g sample was weighed into a thimble, and its contents were placed into the Soxhlet apparatus's bottom extractor after loose plug fat-free cotton wool was put into the top of the thimble.The extractor was equipped with a 250 ml, flat-bottom flask of known weight that held 150–200 ml of 40–60 0C hexane. For eight hours, the device was heated to extract fat. After recovering the solvent, the flask holding the oil and solvent mixture was placed in a hot air oven (GENLAB, England B6S, serial no. 85K054) and heated to 105 0C for one hour in order to evaporate

the solvent and remove any remaining moisture. After that, it was placed in a desiccator to chill for fifteen minutes before being weighed. The percentage of fat was calculated as

$$\%\ CrudeFat = \frac{Weight\ of\ extracted\ fat}{Weight\ of\ Sample}\ x\ 100 \quad (3)$$

2.2.4 Determination of ash content

According to (9), two grams (2 g) of the sample were weighed into an ashing plate that had been preheated, cooled in a desiccator, and then weighed shortly after at room temperature. After that, the crucible and its contents were heated for six to seven hours at 550°C in a muffle furnace (Shanghai box type resistance furnace, No.: SX2-4-10N). At room temperature, the dish was chilled in a desiccator and weighed. The proportion of the initial sample weight was used to calculate the total ash.

$$\%\ Ash = \frac{(W3-W1)}{(W2-W1)}\ x\ 100 \quad (4)$$

$Where$:

$W1\ =\ Weight\ of\ empty\ crucible,$

$W2\ =\ Weight\ of\ crucible\ +\ sample\ before\ ashing,$

$W3\ =\ Weight\ of\ crucible\ +\ content\ after\ ashing.$

2.2.5 Determination of crude fiber

Diethyl ether was used to extract two grams (2 g) of the sample in accordance with (9). The California Buchner system was used to digest and filter this. After two hours of drying at 130 ± 2 0C, the residue was chilled in a desiccator and weighed. After that, the residue was placed in a muffle furnace (Shanghai box type resistance furnace, No.: SX2-4-10N), ignited for 30 minutes at 550 0C, cooled, and weighed. The crude fibre content percentage was calculated as :

$$\%\ Crude\ fibre\ = \frac{Loss\ in\ weight\ after\ incineration}{Weight\ of\ original\ food} \times 100 \quad (5)$$

2.2.6 Determination of carbohydrate content

According to Onwuka (10)'s description, the carbohydrate content was determined by difference as follows:

$$\%\ Carbohydrate = 100 - (\%\ moisture + \%\ Protein + \%\ Fat + \%\ Ash + \%\ Fibre) \quad (6)$$

2.3 Determination of Functional Property Indices

2.3.1 Determination of water absorption capacity

For five minutes, a magnetic stirrer set at 1000 rpm was used to combine one gram (1 g) of the sample with either 10 ml of distilled water or 10 ml of vegetable oil with a specified density (0.99 mg/ml). The mixture was centrifuged (Model: SM 800B Uniscope Surgifriends Medicals,England) at 3500 rpm for 30 min and the volume of the supernatant noted. WAC or OAC was calculated and expressed as g of water or oil absorbed or retained per g of sample as described by AOAC (9).

$$Water\ Absorption\ Capacity\ (\%) = \frac{Amount\ of\ water\ added - Freewater}{Weight\ of\ sample} \times density\ of\ water\ \times 100 \quad (7)$$

2.3.2 Determination of oil absorption capacity

The approach of AOAC (10) was used. One gram of each flour sample was weighed independently before being added to sterile centrifuge tubes with known weights. In each tube, flour and groundnut oil were

combined to create a 10 ml dispersion. For fifteen minutes, the tubes were centrifuged at 3500 rpm. The tubes were reweighed after the supernatant was disposed of. The capacity to absorb oil is the mass gain.

$$Oil\ Absorption\ Capacity\ (\%) = \frac{Amount\ of\ oil\ added - Free\ oil}{Weight\ of\ sample} \times density\ of\ oil \times 100 \quad (8)$$

2.3.3 Determination of the gelation concentration

The procedure outlined by (10), was used to investigate the lowest gelation concentration. Test tubes containing suspensions of 2, 4, 6, and 8 up to 20% (w/v) flour in 5 millilitres of distilled water were heated in boiling water for one hour, cooled in ice, and then cooled at 4 oC for two more hours. When the test tube was turned upside down, the sample did not fall or slip at the lowest gelation concentration.

2.2.4 Foaming capacity and stability

The method described in (10) was modified to calculate foam stability and foam capacity. One hundred millilitres of distilled water were used to dissolve a 500 mg protein sample. The mixture was put into a 250 ml measuring cylinder after being homogenised for two minutes on high speed 5max in a blender (O'Qlink, China). The percentage difference between the volume increase and the original volume of protein solution in the measuring cylinder was used to calculate foam capacity, commonly referred to as whippability. Foam stability was expressed as the proportion of foam that stayed in the measurement cylinder after a 30-minute quiescent period as opposed to the initial volume.

$$Foaming\ capacity\ (\%) = \frac{V_2 - V_1}{V_1} \times 100 \quad (9)$$

Where;

$V1$ = $Volume\ before\ whipping\ (ml)$

$V2$ = $Volume\ after\ whipping\ (ml)$

$V3$ = $Volume\ after\ standing\ (ml)$

2.3.5 Determination of emulsion capacity

was ascertained by applying the Onwuka technique (10). In a warring blender set at 1600 rpm, two grams (2 g) of each flour sample were combined with 25 millilitres of distilled water at room temperature. After adding 25 ml of vegetable oil gradually, the mixture was blended continuously for an additional 30 seconds. After that, each mixture was put into a centrifuge tube and spun for five minutes at 1600 rpm. The volume of oil that was separated from the samples was then measured from the tube.

$$Emulsification\ capacity = X/Y \times 100 \quad (10)$$

$Where$

$X =\ height\ of\ emulsified\ layer$

$Y =\ height\ of\ whole\ solution\ in\ centrifuge\ tube$

2.4 Determination of Mineral Composition

2.4.1 Determination of sodium

AOAC's description of flame photometry was used to determine the sodium (9) contents. One litre of distilled water was used to dissolve 0.2542 grams of NaCl, yielding 100 parts per million Na. A 2 mL sample aliquot (sample stock solution) was measured using a flame photometer after this working standard solution was diluted

to provide a range with 0–10 ppm sodium and built up to a 100 mL mark. Using the graph (standard curve) as a guide, the test mineral concentration in the sample was determined as follows:

$$\text{Sodium (mg/100g)} = \frac{100XYV_f \times D}{W \times 100 \times Va} \quad (11)$$

Where:
W=Weight of the sample analyzed
Y=Concntration of Na obtained from the standard curve
Vf=Total volume of digest/extract (100 mL)
Va=$Volume\ of\ extract\ used$
D=$Dilution\ factor$

2.4.2 Zinc and Magnesium

To determine the levels of zinc and magnesium, 1.0 g of zinc or magnesium ribbon was dissolved in 10 ml of concentrated HCl using the method of (9). This created a stock solution that had 1000 mg/ml of either ion. A water bath was then used to boil and evaporate the solution until it was nearly dry. The solution was poured into a 100 ml volumetric flask and marked with de-ionized water after de-ionized water was added. With 1000 mg/ml of Zn2+ or Mg2+ ions, this stock solution was used to create standard solutions with concentrations of 0.0, 0.5, 1.0, and 1.5 ppm. Zinc and magnesium solutions were mixed with a strontium chloride solution until the final solution contained 1500/mg of strontium ions. The concentration of zinc and magnesium were determined using respective calibration curves.

2.4.3 Calcium

The precipitate was transformed into CaO by ignition and reported as calcium weight per sample weight using the precipitation procedure as outlined by AOAC (9). Ten grams of the food sample were weighed into a crucible and then ashed for twenty-four hours at 500 to 550 degrees Celsius in a muffle furnace. SiO2 was dehydrated by dissolving the ash in 10 millilitres of 2 N HCl, then adding 5 millilitres of concentrated HCl and letting it evaporate completely in a steam bath. The residue was moistened with 5 ml of 2 M HCl, about 50 ml of H2O was added and mixture heated for a few minutes in a steam bath, after which it was transferred to a 100 ml volumetric flask, cooled rapidly to room temperature and diluted to volume. A 10 ml aliquot was pipetted into a conical tip centrifuge tube and to each sample in the centrifuge tubes 2 ml of saturated ammonium oxalate solution was added. Two (2) drops of methyl red indicator (0.5 % methyl red in ethanol) was added to each tube and the pH adjusted to 4.5 by mixing. This was allowed to stand for at least 4 h, then centrifuged at 1500 rpm for 15 min.The supernatant was carefully removed by decantation, taking care not to disturb the precipitate. The precipitate was thoroughly washed by sharply shaking of the tube. Centrifuging and washing were repeated three times. After removing the last supernatant, the calcium oxalate precipitate was re-suspended in 2 ml of the water and quantitatively transferred to a carefully dried and weighed crucible. Rinsing was done three times adding the rinse to the crucible which was carefully dried on a hot plate avoiding splattering. Complete ashing was done at 600 0C in a muffle furnace for 12 h, after which the ash was cooled in a desiccator and weighed. The result is expressed as

$$Weight\ of\ sample\ (in\ 10\ ml\ aliquot\ of\ sample) = mg\ calcium\ oxide\ x\ 0.7147 \quad (12)$$

2.4.4 Potassium

Potassium determination was by Flame Photometry described by AOAC (9). One gram (1 g) of sample was dissolved in 20 ml of acid mixture (650 ml of concentrated HNO3; 80 ml Perchloric acid; 20 ml concentrated H2SO4) and aliquots of the diluted clear digest were taken for photometry using the Flame analyzer.

2.4.5 Phosphorus

Potassium was determined by flame photometry, as outlined by AOAC (9). One gram (1 g) of the sample was dissolved in 20 millilitres of an acid mixture made up of 650 millilitres of concentrated HNO3, 80 millilitres of perchloric acid, and 20 millilitres of concentrated H2SO4 before aliquots of the diluted clear digest were obtained for photometry using the Flame analyser.

2.4.6 Determination of iron

Iron was measured using the procedure outlined by AOAC (9). A standard solution was made from 1 g of pure iron wire that contained 100 mg/ml of Fe3+ ions. After being dissolved in 20 millilitres of pure HNO3, the wire was heated in a water bath and then diluted with 1000 millilitres of distilled water. Standard solutions were made with concentrations of 0, 0.5, 1.0, 2.0, and 4.0 ppm. An AGILENT (Model 5805, Agilent Spec England) atomic absorption spectrophotometer was used to measure the sample's absorbance at 510 nm after two millilitres of the sample were diluted to 100 millilitres. The absorbance of the standard and the sample was recorded, and the standard curve was used to calculate the sample's iron content.

3.0 RESULTS AND DISCUSSSION

3.1 Proximate Composition of Grasshopper Protein Meal and Protein Hydrolysate

Results of the proximate composition of grasshopper protein meal and protein hydrolysate is presented in Table 2. It is well recognised that insects are a good source of proteins, fats, carbs, vitamins, and minerals (11). According to Payne et al. (12), because insects are abundant in nature and can be economically bred, they may make a great source of protein. The great diversity of edible insect species contributes to the great variation in their nutritional content. Depending on the insect's stage of metamorphosis, values can vary even among the same group of edible bug species.

The stability of the food's shelf life is indicated by its moisture content; a higher moisture content increases the risk of microbial infection and chemical reactions that could lower the food's quality and stability. The moisture contents of the hydrolyzed, defatted and whole meal grasshopper are below 14 % recommended for long period of storage, hence a good potential during storage (13).

The grasshopper meal recorded highest content of fat which was significantly ($p<0.05$) different from the defatted and hydrolyzed grasshopper. Fat are essential in daily human diets as they increase the palatability of foods by absorbing and retaining their flavour (14). They also play a crucial role in the biological and structural processes of cells and aid in the transportation of fat-soluble vitamins that are necessary for proper nutrition (15). Whole grasshopper meal was a rich source of oil due to its fat content.

When compared to the defatted and whole meal grasshoppers, the hydrolysed grasshopper's protein level was greater. Body fluids including milk and blood, as well as enzymes, antibodies, and other hormones, are largely composed of proteins. They generate protective, supportive, and structural tissues like muscles, cartilages, skin, hairs, and nails, and are vital to all life (16). Food proteins serve as nutrients as well as having physiochemical functions that support well health. Whole meal grasshopper showed highest content of fibre. The least was recorded in the defatted grasshopper but showed no significant difference. No fibre was found in the grasshopper that had been hydrolysed. Crude fibre has a physiological function in the body by preserving internal distension, which facilitates appropriate intestinal tract peristaltic movement (17). "A very low-fiber diet may cause constipation, which may cause the body to experience discomfort from loose stool." Because they provide a feeling of fullness even when little portions are consumed, diets high in fibre have been used to reduce fat and control weight. Crude fibre content differed significantly between insect species, according to literature surveys. This could be because various species have distinct exoskeletons and structures. Banjo et al.'s (18) findings for Orthoptera and the grasshoppers' crude fibre content compare nicely.

Ash is an inorganic residue that indicates the mineral content of any dietary item (19). It is the byproduct of burning biological stuff. As a result, ash is a good source of minerals, including both organic (such as oxalate acetate and pectat) and inorganic (such as phosphates, carbonates, and sulphates) salts. The ash content of the complete defatted grasshopper was higher than that of the whole grasshopper, but there was no discernible difference. As the grasshopper was hydrolysed, the value dropped.

All bodily functions rely on carbohydrates for heat and energy, so when they are insufficient, the body may turn to proteins and fat for the energy it needs, which could result in the depletion of bodily tissues (20). It was found that the complete grasshopper meal had the highest amount of carbohydrates. The defatted grasshopper came next, and the hydrolysed grasshopper came last.

Table 2: Proximate Composition of Grasshopper Protein Meal and Protein Hydrolysate

Proximate composition (%)	WGM	DGM	GHM
Moisture	12.54±0.34^{a}	8.65±0.48^{b}	7.33±0.19^{c}
Fat	17.55±0.65^{a}	5.43±0.72^{b}	0.15±0.04^{c}
Crude protein	37.45±1.14^{c}	64.04±1.25^{b}	87.54±0.96^{a}
Fibre	6.43±1.12^{a}	6.34±0.65^{a}	Not detected
Ash	2.76±0.62^{a}	2.05±0.54^{a}	0.23±0.05^{b}
Carbohydrate	23.27±1.03^{a}	13.49±0.96^{b}	4.75±0.43^{c}

Values are means ± Standard deviation of triplicate determination. Means in the same row with different superscripts are significantly different ($p \leq 0.05$)

Key: WGM: Whole Grasshopper meal, DGM: Defatted Grasshopper Meal, GHM: Hydrolyzed Grasshopper Meal

3.2 Functional Properties of Flours from Grasshopper Protein Meal and Protein Hydrolysate

Table 3 displays the findings of the functional characteristics of flours made from grasshopper protein meal and protein hydrolysate. The criteria that dictate a food material's use and ultimate use are known as its functional properties (21). The physical characteristics of food proteins known as functional properties dictate how they behave in the food system throughout processing, storage, and consumption. It typically indicates that the food ingredients being studied will either directly or indirectly interact with other food ingredients to impact processing uses, food quality, and final acceptance (22).

The quantity of fat that the proteins are able to retain is known as the oil absorption capacity. It is also the total of the fat that is physically trapped in the protein matrix and the lipids that are bound by hydrophobic interactions between the protein and the fat itself (23). The maximum capacity to absorb oil was demonstrated by whole grasshopper flour. The flour made from grasshopper hydrolysate came next. Protein levels, protein kinds, and the amino acid makeup of proteins particularly hydrophobic residues that interact with the hydrocarbon chains in fat molecules all affect an oil's ability to absorb it.

The high concentration of non-polar amino acids in whole grasshopper flour may be the cause of its high oil absorption capacity, as these amino acids are crucial to the mechanism of oil absorption (24). This is significant since oil improves food palatability and preserves taste (25). In food technology, understanding fat absorption capacity is crucial since it gives products features like improved palatability, flavour retention, and longer shelf life by lowering humidity and fat loss. The meat and pastry sectors are among the many food applications that primarily employ this feature (25). Additionally, the meals' high oil absorption capability indicated that the grasshopper meals' ingredients were lipophilic (26).

The quantity of water that proteins can bind or hold onto is known as their water absorption capacity (WAC). Additionally, compared to the defatted and whole meal grasshopper flours, the hydrolysed grasshopper flour had a greater potential to absorb water. The high concentration of hydrophilic amino acids in the hydrolysed grasshopper flour may be the cause of its greatest water absorption capacity values.

Continuous gas dispersions, usually liquids, are called foams. To form a cohesive protein layer around the gas/air droplets, proteins must be soluble in the aqueous phase and quickly unfolded during synthesis. Compared to the defatted and hydrolysed grasshopper meal samples, the whole grasshopper meal may have a lower foaming capacity due to the physicochemical properties of its proteins. In order for flour to foam, as shown by its foaming capacity, it must have flexible protein molecules that reduce the surface tension of water (27).

The hydrolyzed grasshopper flour showed highest contents of both the foaming capacity and foaming stability as compared to the whole meal and defatted grasshopper flour counterparts while the foaming capacity of flours observed here were higher than those observed in grasshopper by Niphattha et al. (28). The result of the foaming capacities of both the whole, defatted and hydrolyzed grasshopper flours were greatly higher that those reported for whole giant cricket (*Gryllidae sp.*) powder and Omotoso (15), who reported the FC from *Cirina forda* larvae powder as low as 7.1 %. When compared to its foaming capability, the low foaming stability indicates an exceptional ability to stabilise foam against collapse. Foam collapse typically occurs through one of these three mechanisms: (i) bubble disproportionation; (ii) bubble coalescence caused by thin film instability between bubbles; and (iii) water dripping from bubble surfaces to the liquid layer, which removes protein from the bubble-encircling film (29).

Because WGM contains lipids and intact protein structures throughout the meal, it appears to form a gel more readily than DGm and GHM, based on the differences in gelation concentration. It's possible that the protein structure changes during defatting and hydrolysis, decreasing the protein's capacity to form gels at lower concentrations, which explains the rise in gelation concentration for DGM and GHM.

This pattern shows that fat removal and protein hydrolysis affect grasshopper meal's gelation qualities, necessitating larger concentrations to produce gelation. Furthermore, research on the gelation of legume protein isolates by Chandra and Samsher (30) shows that whole protein meals that have not been hydrolysed or defatted have a tendency to form gels at lower concentrations because their intact protein structures interact more easily to form a gel matrix. This is consistent with the current study's lower WGM gelation concentration.

The highest levels of emulsion stability and capacity were found in the hydrolysed grasshopper flour. The emulsion capabilities of the hydrolysed, defatted, and whole meal grasshopper flours were found to differ significantly.

The amino acid profile and the capacity of digestive enzymes to liberate the amino acid dictate the quality of protein in diet. The hydrolysed grasshopper showed significant levels of protein digestibility. The defatted flour comes next, and the whole grasshopper meal had the lowest recorded amount. The results obtained here are comparable to those published for Gryllus assimilis (80.82%), Cirina forda (81.71%), and Ruspolia diferens (82.34%) (31).

Table 3: Functional Properties of Grasshopper Protein Meal and Protein Hydrolysate

Samples	WGM	DGM	GHM
Oil absorption capacity (%)	212.43±1.32[a]	193.43±1.04[b]	131.32±2.32[c]
Water absorption capacity (%)	201.00±0.43[c]	231±0.32[b]	237±0.95[a]
Gelation concentration (%)	11.00±0.00[c]	17.00±0.00[b]	23.00±0.00[a]
Foaming capacity (%)	345.00±0.43[b]	324.00±1.92[c]	367.00±0.85[a]
Foaming stability (%)	65.00±1.50[c]	70.00±1.00[b]	87.00±1.24[a]
Emulsification capacity (ml/g)	35.43±0.65[c]	38.64±0.96[b]	42.54±1.04[a]
Emulsion stability (%)	67.54±0.97[c]	75.67±1.03[b]	81.43±1.22[a]

Values are means ± Standard deviation of triplicate determination. Means in the same row with different superscripts are significantly different ($p \leq 0.05$)
Key: WGM: Whole Grasshopper meal, DG: Defatted Grasshopper Meal, GH: Hydrolyzed Grasshopper Meal

3.3 Mineral Composition of Grasshopper Protein Meal Protein Hydrolysate

Table 4 presents the findings of the mineral composition of grasshopper protein hydrolysate and meal. The body uses micronutrients, which are substances needed in trace amounts to maintain healthy growth and development, to make hormones, enzymes, and other compounds necessary for maintaining regular bodily processes. The inorganic components, known as minerals, play a crucial and specialised function in metabolism. In addition to proteins, carbs, lipids, and vitamins, they are needed in lower amounts. They are inorganic, or "ash constituents," of food that cannot be eliminated by heating. Despite the fact that they don't produce energy, they are vital to numerous bodily functions (32).

The mineral composition result indicated that the potassium content of the grasshopper hydrolysate and the entire meal did not differ significantly. Despite this, all of the samples' potassium contents showed high values. The outcome demonstrated that the potassium content of grasshopper meal is considerably decreased by defatting and hydrolysing it. The levels of sodium, magnesium, calcium, phosphorus, iron, zinc, copper, and manganese showed a similar pattern. In general, defatting and hydrolysing had a major impact on the mineral content of grasshopper meal. This is comparable to the findings of a previous study by (33). Calcium is essential for the development and maintenance of bones, teeth, muscles, nerve impulse transmission, blood coagulation, and a regular heartbeat. The recommended daily allowance (RDA) for calcium for humans between the ages of 18 and 50 is 1,000 mg per day. For the development of bones and teeth, those under the age of 18 require a higher concentration of calcium (1,300 mg) (34). One of the six major macrominerals, magnesium is necessary for over 300 metabolic processes and plays a part in healthy bones, proper muscle contraction, ideal blood pressure, and a healthy heart rate. According to Sales and Pedrosa (35) magnesium is necessary for DNA synthesis and stability. After calcium, phosphorus is an essential mineral for human health and plays a key part in several metabolic processes, such as energy metabolism, bone mineralisation, and the structure of DNA and RNA (36). Together with Na

and Ca, potassium controls fluid balance, keeps the heart beat regular, and is responsible for nerve and muscle signals (37).

Table 4: Mineral Composition of Grasshopper Protein Meal Protein Hydrolysate

Mineral composition (mg/100g)	WGM	DGM	GHM
Sodium	301.44±0.65[a]	276.54±0.75[b]	43.43±0.57[c]
Magnesium	8.05±0.06[a]	8.01±0.03[b]	3.24±0.05[c]
Calcium	15.43±0.31[a]	10.43±0.65[b]	1.43±0.07[c]
Potassium	2143.53±0.95[b]	1245.32±0.95[a]	241.54±0.85[b]
Phosphorus	7.43±0.61[a]	5.35±0.55[b]	0.98±0.03[c]
Iron	6.43±0.08[a]	3.21±0.21[b]	0.26±0.05[c]
Zinc	4.83±0.08[a]	2.32±0.04[b]	1.22±0.83[c]
Copper	1.32±0.83[a]	1.03±0.03[a]	0.73±0.07[b]
Manganese	0.78±0.08[a]	0.21±0.09[b]	0.07±0.01[c]

WGM: Whole grasshopper DGM: Defatted grasshopper, GHM: Grasshopper hydrolysate

4.0 CONCLUSION AND RECOMMENDATIONS

4.1 Conclusion

The result of the proximate composition revealed that the WGM DGM and GH had high protein contents, with WGM recording high fat content and substantial amounts of sodium, calcium and iron minerals. Protein percent was found to appreciate significantly ($p<0.05$) with defatting and enzymatic hydrolysis of the grasshopper meal resulting in higher protein contents in DGM (64.04%) and GH (87.54%).

This study also showed that functional properties of the grasshopper meals such as oil absorption capacity, decreased with increase in the protein content of the samples. While the reverse was the case for water absorption capacity, gelation concentration, forming stability and emulsion stability increased with increased in protein content of samples. WGM exhibited

superior functional properties than the DGM and GH samples indicating their suitability in product development of bioactive products. The results of the mineral composition of this study showed significant different.

4.2 Recommendations

Based on the findings of this study, the following recommendations were made:

1. Grasshopper protein meals and hydrolysates were found to possess substantial nutrients with varying potent bioactivities suggesting they could be used in food fortification to produce functional foods and nutraceuticals to help in the management of chronic malnutrition

2. Grasshoppers farming should be encouraged to increase their production and utilization for both food and medicinal purposes. Animal producing farmers should be encouraged to grow grasshoppers to make it easily accessible and available all year round by consumers and researchers.

3. Further studies should be carried out through bioassay guided HPLC purification to elucidate the structural composition and sequences of the active components of grasshopper proteins showing the observed bioactivities.

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