MINERAL COMPOSITION OF FOUR FORMULATED DIETS USING GUINEA GRASS LEAFMEAL AS A REPLACEMENT FOR SOYBEAN MEAL.

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DEPARTMENT OF ANIMAL SCIENCE

FACULTY OF AGRICULTURE

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CERTIFICATION

This is to certify that this Project work was carried out by Ogbemudia Sylvester IDUBOR with Matriculation Number AGR2000080 of the Department of Animal Science, Faculty of Agriculture, University of Benin, Nigeria.

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HEAD OF DEPARTMENT

DEDICATION

This work is dedicated to God Almighty for his infinite mercy all through the course of my program in the University of Benin, to my loving parents (Mr. and Mrs. IDUBOR) and my wonderful siblings.

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ABSTRACT

The global livestock sector requires sustainable strategies to reduce reliance on costly, conventional protein sources like Soybean Meal (SBM), particularly in regions like Nigeria. This study investigated the nutritional implications of partially replacing SBM with locally abundant Guinea Grass Leafmeal (GGLM) in formulated diets, focusing specifically on the resulting mineral composition.

Four experimental diets were formulated to be isonitrogenous and isocaloric, where SBM was replaced by GGLM at graded levels: T1 (0% GGLM, control), T2 (10% GGLM), T3 (20% GGLM), and T4 (30% GGLM). The diets were subjected to mineral analysis using Atomic Absorption Spectrophotometry (AAS).

The results showed a significant influence ($p<0.05$) of GGLM inclusion on the mineral profile of the diets. Diets with higher GGLM levels (T4) recorded the highest concentrations of most essential macro- and micro-minerals, including Calcium (Ca), Sodium (Na), Iron (Fe), Zinc (Zn), Potassium (K), Phosphorus (P), and Manganese (Mn). For instance, Ca increased from 3126.81 mg/kg in T1 to 5614.33 mg/kg in T4, and P increased from 415.10 mg/kg in T1 to 520.30 mg/kg in T4. Conversely, Chromium (Cr) showed an inverse relationship, and traces of Lead (Pb), though increasing slightly with GGLM inclusion (from 0.0533 mg/kg in T2 to 0.1500 mg/kg in T4), remained well within safe limits.

The study concludes that Guinea Grass Leafmeal is a valuable mineral-enriching alternative to Soybean Meal. Its substitution can successfully enhance the mineral density of livestock diets, offering a cost-effective and sustainable feed resource without posing a heavy metal toxicity risk, thereby supporting resilient livestock production systems.

CHAPTER ONE

INTRODUCTION

Globally, the livestock sector is continuously seeking ways to enhance production efficiency and sustainability. A cornerstone of this endeavor lies in optimizing animal nutrition through carefully formulated diets (Suttle, 2022). These diets are designed to provide a precise balance of energy, protein, vitamins, and minerals to support growth, health, and productivity. The selection of feed ingredients is a critical aspect of this process, with a growing emphasis on utilizing locally available, cost-effective, and nutritionally sound resources. This shift is driven by the need to reduce reliance on conventional feedstuffs that may be expensive or have competing uses, thereby fostering more resilient and sustainable livestock farming systems. The exploration of alternative feed ingredients often involves a thorough assessment of their nutritional contributions, including their impact on the essential mineral balance of the final ration, which is paramount for animal well-being (Underwood, 1981).

Soybean meal has long been a primary protein source in formulated animal feeds due to its high protein content and favorable amino acid profile (Ruiz *et al.,* 2020; McDonald *et al*., 2002). However, its fluctuating cost and availability, often linked to global market dynamics and import dependencies, pose significant challenges for feed manufacturers and livestock producers in many regions, including Nigeria. This underscores the pressing need to identify and evaluate alternative, locally abundant feedstuffs that can partially or wholly replace soybean meal without compromising diet quality.

Guinea grass (*Panicum maximum*) is a widely distributed and highly productive tropical forage grass, well-adapted to Nigerian agro-ecological conditions (Wolele *et al.,* 2025; Agishi, 1985). It is recognized for its palatability and ability to produce substantial biomass, making it a valuable feed resource, primarily for ruminant livestock (Wolele *et al.,* 2025). While traditionally used for grazing or as a cut-and-carry forage, its potential incorporation into compound feeds as a replacement for conventional protein sources like soybean meal warrants investigation. While the protein content of Guinea grass shows variability (typically 7-12.5% CP), with some reports indicating 10.5% (Ironkwe and Ukanwoko, 2016) and others up to 12.5% (Agishi, 1985), its concentration is substantially lower than that of soybean meal (44-48% CP). A significant difference in their mineral profiles is also apparent.

Minerals, though required in smaller quantities compared to energy and protein, play indispensable roles in animal physiology (Underwood, 1981; Prasad and Gowda, 2005). They are crucial for skeletal formation and integrity (e.g., Calcium, Phosphorus, Magnesium), enzyme activation, maintenance of acid-base balance, nerve transmission, immune function, and overall metabolic processes (Suttle, 2022). An imbalance or deficiency of any essential mineral can lead to a cascade of health issues, reduced growth rates, poor feed conversion efficiency, and compromised reproductive performance (Underwood, 1981; Judson and McFarlane, 1998).

When substituting a protein-rich ingredient like soybean meal, which also contributes to the diet's mineral content notably phosphorus (Suttle, 2022), with a forage-based ingredient like Guinea grass, the mineral composition of the final diet will invariably be altered. The extent of these alterations and their implications for meeting the mineral requirements of target livestock species are not well documented. For instance, forages like grasses are generally higher in Calcium and lower in Phosphorus compared to protein meals (Underwood, 1981; Nasrullah *et al.,*2004). Without a clear understanding of the changes in mineral levels (both macro and micro), there is a risk of formulating diets that are deficient or imbalanced, potentially negating any benefits gained from using alternative ingredients. This research is therefore crucial to scientifically evaluate the impact of replacing soybean meal with Guinea grass on the mineral profile of formulated diets, thereby providing essential data for developing nutritionally adequate and sustainable livestock feeds.

1.1 Objectives of the Study

The main objective of this study is to determine the mineral composition of four diets with Guinea grass leafmeal as partial replacement for Soybean meal at varying levels.

The specific objectives of this research are to:

1. determine the physical characteristics of Guinea grass leaf meal.

2. determine the mineral composition of four diets formulated with Guinea grass leaf meal as partial replacement for Soybean meal.

CHAPTER TWO

LITERATURE REVIEW

2.1 Concept of Mineral Composition in Animal Nutrition.

Inorganic elements called minerals are essential to the survival of both humans and animals. Minerals in animal nutrition are essential for physiological, biochemical, and structural activities; they are osmotic balance regulators, enzyme cofactors, skeletal structure constituents, and metabolic process catalysts (McDowell, 2003). Both natural feedstuffs and supplements must be included in animal diets to avoid deficiencies that could affect immunity, growth, productivity, and reproduction.

Minerals required by animals are broadly classified into two main categories based on the quantity needed:

Macro minerals: These are required in larger amounts and include calcium (Ca), phosphorus (P), sodium (Na), potassium (K), magnesium (Mg), sulfur (S), and chloride (Cl). For instance, calcium and phosphorus are critical for bone development, nerve transmission, and muscle function (Underwood and Suttle, 1999).

Microminerals or Trace Minerals: These are required in minute quantities but are no less important. They include iron (Fe), zinc (Zn), copper (Cu), manganese (Mn), iodine (I), selenium (Se), and cobalt (Co). For example, iron is essential for hemoglobin synthesis and oxygen transport, while zinc plays a pivotal role in enzymatic activity, immune response, and skin integrity (Goff, 2006).

The mineral content of animal feed has a direct impact on the animal's ability to absorb nutrients, metabolic functions, bone structure, ability to reproduce, and general health. Feed with a balanced mineral profile promotes healthy muscular contractions, hormonal system function, and the avoidance of illnesses including rickets, osteoporosis, anemia, and goiter (NRC, 2001).

Mineral deficits are widespread in tropical and subtropical areas where forage-based feeding is common because of poor forage quality, particularly during dry seasons, and soil mineral depletion. The mineral content of feed items must therefore be evaluated and balanced (Suttle, 2010). It is crucial to take into account both the overall mineral content and the bioavailability of each mineral that is, the percentage of each mineral that the animal actually absorbs and uses when creating diets.

Mineral interactions also need to be properly controlled. For example, too much calcium can hinder the absorption of phosphorus, and too much iron can prevent the use of copper and zinc (McDowell, 2003). Therefore, rather than just fulfilling each person's mineral needs, nutritionists strive to attain ideal mineral ratios.

Evaluation of the mineral makeup of unconventional and alternative feed ingredients, including forages and agro-industrial by-products, has gained more attention recently. The high price and limited availability of conventional feed sources, such as soybean meal, are the main causes of this development. Guinea grass (*Panicum maximum*) is one such promising substitute. It has a significant mineral profile that can help meet livestock's total nutritional needs, particularly when utilized in part to replace traditional protein and energy sources.

2.2 Importance of Diet Formulation in Livestock Production.

A vital part of animal production is the formulation of livestock diets, which guarantee that animals have a balanced supply of calories, protein, vitamins, and minerals. Animal health and welfare are guaranteed, growth and reproduction are supported, feed efficiency is increased, and productivity is increased with proper diet formulation. In relation to cattle performance and sustainability, this section emphasizes the critical elements and significance of diet formulation.

2.2.1 Ensuring Nutritional Balance

The basic goal of feed formulation is to give the animal all necessary nutrients in the proper amounts. Each nutrient has a specific purpose in physiological processes; proteins for tissue synthesis, carbs and fats for energy, vitamins for metabolic functions, and minerals for bone growth, enzyme function, and reproductive health. A lack or excess of any nutrient can impair animal performance and raise the risk of diseases (McDonald *et al.,* 2011).

2.2.2 Enhancing Animal Performance

Diet formulation has a direct impact on productivity qualities as weight gain, milk yield, egg production, and reproduction. According to studies, animals fed balanced diets have higher growth rates and feed conversion efficiency than those fed unbalanced or poorly prepared diets (Onifade and Tewe, 1993). Dietary mineral content, in particular, influences skeletal growth, immunological response, and metabolic processes, influencing an animal's overall performance and longevity (Goff, 2006).

2.2.3 Cost Efficiency and Economic Sustainability

In the majority of livestock systems, feed accounts for 60–80% of overall production expenditures (FAO, 2013). Effective meal planning maximizes the use of resources, cutting expenses and waste without sacrificing nutritious value. Guinea grass is one example of an unconventional feed additive that can significantly lower feed costs without sacrificing nutritional quality (Ayinla *et al.,* 2020). For smallholder farmers in underdeveloped nations where traditional components like soybean meal are either unavailable or prohibitively expensive, this is especially crucial.

2.2.4 Inclusion of Alternative Feed Resources

Researchers and livestock producers are looking into alternative feed components as a result of the rising demand for traditional feedstuffs and growing concerns about sustainability. For example, guinea grass provides acceptable amounts of fiber, protein, and minerals and is widely available in tropical areas (Aregheore, 2005). Using such forage resources in feed formulations encourages local feed resource usage and environmental sustainability in addition to reducing reliance on expensive inputs like soybean meal (Olorunnisomo and Fayomi, 2012).

2.2.5 Consideration of Nutrient Bioavailability

Bioavailability, or the portion of nutrients that the animal can digest and use, is taken into account when formulating a diet in addition to nutrient content. The availability of minerals including calcium, iron, and phosphorus may be decreased by anti-nutritional components included in some feed ingredients (such as phytates and oxalates) (Ravindran and Blair, 1992). By employing processing techniques or additions (like phytase) to improve nutrient absorption, appropriate formulation takes these issues into consideration.

2.3 Overview of Soybean Meal in Animal Diets.

Soybean meal (SBM) is one of the most used plant-based protein sources in livestock feed formulas around the world. It is a byproduct of soybean oil extraction (Glycine max) and is typically available in two forms: solvent-extracted and mechanically ejected. Solvent-extracted soybean meal is more commonly used in commercial feed production due to its greater protein concentration and availability (Ravindran and Blair, 1992).

Nutritionally, soybean meal is distinguished by its high crude protein content, which normally ranges from 44% to 48%, depending on whether the hulls are removed (NRC, 2001). Soybean meal has a good amino acid profile, particularly lysine, which is commonly lacking in cereal-based diets. Its digestibility and palatability make it appropriate for a wide range of livestock species, including poultry, swine, rabbits, ruminants, and aquaculture (El-Sayed, 1998).

Beyond protein, soybean meal has a high mineral content, which helps cattle achieve their macro- and micromineral requirements. Soybean meal contains the mineral ranges of: Calcium; 0.25–0.30% Phosphorus; 0.65–0.70% (with approximately 30% accessible to non-ruminants); Potassium; 1.90–2.00%. Magnesium; 0.30%; Trace minerals include iron (Fe), zinc (Zn), manganese (Mn), and copper (Cu), albeit these might vary depending on soil and processing processes (NRC, 2001; McDowell, 2003).

Soybean meal is an expensive ingredient, particularly in underdeveloped nations where it is mostly imported. Small-scale farmers' regular usage of soybeans is frequently limited by price volatility in the global market (HLPE, 2013), which is largely fueled by demand from the livestock (Goldsmith, 2008) and biofuel industries (Wright, 2011). Research into locally accessible, reasonably priced, and nutritionally sound substitutes, like guinea grass, moringa leaves, and legume forages, has been spurred by this (Aletor *et al.,* 2002).

From a sustainability standpoint, the increasing demand for soybean meal has raised concerns about environmental impacts, including deforestation in major producing countries like Brazil and Argentina. As a result, there is a growing push for feed innovation and substitution strategies that incorporate more environmentally friendly and regionally sourced ingredients (FAO, 2013).

2.4 Mineral Composition of Forage and Legume-Based Diets

Mineral content in animal feeds is critical for cattle health, production, and reproductive efficiency. Forages and legumes, which are the principal components of many animal diets, have considerably varying mineral profiles depending on species, maturity stage, geographical location, and soil quality (McDowell, 2003). Understanding these differences is crucial, particularly when replacing one feed ingredient (e.g., soybean meal) with another (e.g., Guinea grass). Legumes like soybean (*Glycine max*) are known to be rich in minerals such as phosphorus (P), calcium (Ca), potassium (K), and magnesium (Mg). They are also relatively good sources of trace minerals like iron (Fe), manganese (Mn), zinc (Zn), and copper (Cu) (Karr-Lilienthal et al., 2005). However, the availability of these minerals can be compromised by anti-nutritional factors like phytates, which bind minerals and reduce their digestibility (Reddy *et al.,* 1982).

In contrast, tropical grasses like Guinea grass (*Panicum maximum*) generally contain high levels of structural carbohydrates and moderate amounts of minerals. Studies have shown that Guinea grass can provide substantial amounts of calcium and potassium, especially when harvested at young stages or cultivated on nutrient-rich soils (FAO, 2012). Ekenyem and Madubuike (2006) reported that Guinea grass had a calcium content ranging from 0.42% to 0.65%, magnesium levels of 0.18% to 0.23%, and lower phosphorus levels (around 0.12% to 0.15%). While these values are beneficial, they are typically less concentrated than those found in legume meals.

Seasonal change and the mineral composition of the soil affect the mineral content of forages such as Guinea grass (Aregheore, 2005). For example, forage produced in soils lacking phosphorus will have a decreased P content, necessitating nutritional supplementation to prevent mineral deficiency diseases like rickets or poor bone formation in developing animals. Kadzere *et al.* (2002) provided support for this, stressing the importance of mineral supplementation and soil testing when using a lot of tropical grasses in feed formulations.

Furthermore, the way minerals reside in plant tissues affects their bioavailability. Calcium in forages is frequently contained in easily absorbable forms, particularly in young leaves. However, phosphorus is often bound in phytate complexes, particularly in legumes, which reduces its absorption in monogastric animals (Underwood and Suttle, 1999). In such circumstances, enzyme supplementation (e.g., phytase) has been suggested to improve phosphorus bioavailability (Adeola and Cowieson, 2011).

Interactions between minerals also have a role in regulating the nutritional content of forage-based diets. High calcium levels can interfere with the absorption of other minerals like zinc and magnesium, whereas excess iron can limit copper absorption (McDowell, 2003). Therefore, while constructing diets employing Guinea grass as a substitute for soybean meal, attention must be paid not only to the total mineral content but also to their ratios and interactions.

Moreover, the drying and processing procedures applied to forages and legumes can affect their mineral makeup. Sun-drying, for example, may lead to leaching of certain minerals, especially potassium and sodium, if done poorly or under humid conditions (Onwuka, 1997). On the other hand, ensiling can preserve mineral content but may modify fiber composition and pH, which impacts mineral solubility and absorption. The heterogeneity in mineral content among forage and legume sources highlights the need for accurate feed analysis before integration into cattle diets. This guarantees that the nutritional requirements of animals are met, and hazards connected with mineral imbalances are reduced.

2.5 Effect of Replacing Soybean Meal with Guinea Grass on Diet Composition

The substitution of soybean meal with Guinea grass in animal diets has gained increased interest due to the need for cost-effective and sustainable feed options, especially in tropical settings. Guinea grass, being high in fiber and possessing moderate protein and vital minerals, is a feasible alternative to soybean meal. However, its impact on the total mineral content of the diet and the animal's physiological response is a subject of current inquiry.

According to Aderinboye *et al*. (2012), partial replacement of soybean meal with Guinea grass leaf meal (GGLM) in ruminant diets maintained a relatively consistent mineral composition, particularly for macro-elements like calcium (Ca) and magnesium (Mg). The study indicated that up to 50% replacement had no meaningful effect on mineral absorption and retention. Similarly, Oluremi *et al*. (2010) observed that in rabbits, diets including Guinea grass at 25–50% substitution levels nevertheless met their mineral requirements, with equivalent blood mineral values to animals fed traditional soybean-based diets.

Nonetheless, Guinea grass tends to have lesser phosphorus (P) content compared to soybean meal (Onwuka *et al.,* 2010). This can impact the Ca:P ratio in the diet, which is crucial for bone formation and metabolic stability. Excessive Ca without a corresponding P supply can lead to impaired P absorption, resulting to growth retardation and skeletal abnormalities in monogastric animals (Suttle, 2010). Therefore, while substituting soybean meal with Guinea grass, there is a need to monitor and alter the Ca:P balance using mineral premixes or phosphorus-rich supplements such as bone meal or dicalcium phosphate.

Another noticeable impact of substituting soybean meal with Guinea grass is the possible alteration in trace mineral content, such as iron (Fe), zinc (Zn), and copper (Cu). Soybean meal is recognized to be a good source of these trace elements (Karr-Lilienthal *et al.,* 2005), and although Guinea grass includes them, the amounts can be uneven due to environmental factors such soil type, plant age, and fertilizer procedures (McDowell, 2003). Consequently, animals fed high doses of Guinea grass may require additional supplementation to prevent deficits.

Additionally, the digestibility of the diet plays a role in mineral availability. Guinea grass contains more indigestible fibre than soybean meal, which can impact mineral solubility and uptake in the digestive system (Reddy *et al.,* 1982). High fibre content may bind divalent minerals like calcium, magnesium, and zinc, making them less accessible for absorption (Selle *et al.,* 2000). This can be handled through proper processing processes (e.g., chopping, drying, and ensiling) or by utilizing feed additives such organic acids and chelated minerals to promote mineral bioavailability.

Moreover, research by Esonu *et al.* (2006) showed that, despite minor differences in mineral retention between treatment groups, adding Guinea grass to rabbit meals at different inclusion levels did not substantially impair growth or feed efficiency. This implies that guinea grass can be used as a good partial substitute for soybean meal with the right formulation, all without significantly compromising mineral nutrition.

2.6 Factors Affecting Mineral Bioavailability in Diets

Mineral bioavailability is the percentage of a mineral that is absorbed, digested, and eventually used by the animal's body for physiological processes. A feed must have the right form and accessibility of minerals in order to satisfy animal needs; simply having a sufficient amount of minerals is insufficient. A number of factors affect the mineral bioavailability of formulated diets, particularly when conventional feed elements like soybean meal are substituted with plant-based materials like Guinea grass.

2.6.1 Presence of Anti-Nutritional Factors

The presence of anti-nutritional substances such as phytates, oxalates, and tannins is one of the main reason restricting the availability of minerals in plant-based feeds. These compounds decrease the solubility and absorption of minerals such as calcium, iron, zinc, and magnesium in the gastrointestinal system by forming insoluble complexes with them (Reddy *et al.,* 1982; Liener, 1994). Studies have shown that phytates included in soybean meal bind to phosphorus, decreasing its availability to monogastric animals. Likewise, oxalates found in guinea grass have the ability to chelate calcium and magnesium, creating complexes that are not well absorbed (Akinmutimi, 2004). This calls for the use of enzymes like phytase or oxalase to break down these complexes or processing to minimize these molecules (Selle and Ravindran, 2007).

2.6.2 Mineral-Mineral Interactions

Mineral interactions can have a big impact on how well minerals are absorbed and used. For instance, because of competition for intestinal absorption sites, a high dietary calcium intake may hinder the absorption of magnesium, zinc, and phosphorus (Underwood and Suttle, 1999).

Similar to how excess iron can prevent the absorption of copper and zinc, excess phosphorus can hinder the optimum utilization of calcium.

In diets that contain plant-based foods with inherently unbalanced mineral ratios, this is especially important. Therefore, even with sufficient quantities of all minerals, formulations including guinea grass need to be carefully adjusted to prevent antagonistic interactions that could result in shortages (McDowell, 2003).

2.6.3 Fibre Content and Digestibility

Mineral bioavailability may be hampered by high fiber content, which is typical of forages like guinea grass. According to Suttle (2010), insoluble dietary fiber has the ability to bind minerals and increase their excretion through stool, hence decreasing their effective absorption. Mineral digestibility is known to be negatively correlated with the amount of neutral detergent fiber (NDF) and acid detergent fiber (ADF) in forages, particularly in monogastric species such as rabbits and chickens (Onwuka *et al.,* 2010). Feed additives such as microbial inoculants and enzymes (such as cellulases and hemicellulases) may promote mineral release and absorption and accelerate fiber decomposition in order to counteract this.

2.6.4 Soil and Environmental Factors

The soil that is used to grow forages, such as Guinea grass, has a significant impact on their mineral makeup. Even if plants are naturally unable to absorb trace minerals, soils lacking in these minerals will yield forages that are likewise lacking in those minerals (FAO, 2012). According to Aregheore (2005), the mineral composition of plant materials can also be changed by variables like fertilization, rainfall, pH, and harvest maturity stage. In order to determine the nutritional and mineral content of Guinea grass, harvest time is crucial because older grass tends to accumulate more fiber and less minerals. To guarantee stability in feed quality, agronomic procedures should be combined with regular mineral profiling.

2.6.5 Animal Species and Digestive Physiology

The ability of various animal species to absorb and use minerals varies. Compared to monogastric animals, ruminants are often better suited to handle high-fiber and mineral-bound feedstuffs because of their microbial fermentation system. For instance, the rumen's microbial community can produce phytase enzymes, which increase the bioavailability of bound phosphorus (NRC, 2007).

However, non-ruminants like pigs, rabbits, and poultry rely more on bioavailable versions or supplemental enzymes in their diet due to their absence of endogenous phytase enzymes (Adeyemi *et al.,* 2017). This species-specific variation emphasizes the necessity of adapting mineral processing and supplementing techniques to the particular animal.

2.6.6 Processing and Supplementation Strategies

Numerous feed processing techniques, including ensiling, sun-drying, fermenting, and pelleting, can improve mineral availability and decrease anti-nutritional components (Oluremi *et al.,* 2010). According to Suttle (2010), it has also been demonstrated that supplementing livestock with mineral premixes or organic mineral sources (such as chelated minerals) enhances absorption and retention. Mineral supplementation in diets made with Guinea grass is especially crucial to satisfy the needs of animals who develop quickly or produce a lot of food. Additionally, it is a reasonably priced method of addressing inadequacies brought on by the inherent fluctuations in forage mineral concentration (Kadzere *et al.,* 2002).

2.7 Economic Implication of Using Guinea Grass in Place of Soybean Meal

Sustainable and lucrative animal production has been severely hampered by the high cost of traditional feed ingredients like soybean meal, especially in poor nations. In intensive livestock farming systems, feed expenses can make up 60–80% of overall production costs (Aregheore, 2000; Adesehinwa, 2007). This has increased interest in locally accessible, less costly, and alternative feed supplies like guinea grass (*Panicum maximum*), which can successfully substitute some of the traditional protein sources like soybean meal.

Guinea grass is readily available in tropical and subtropical locations, and it is well-suited to a variety of agroecological situations. Its capacity to grow with little input and relatively high biomass output make it an affordable feed option for both small-scale and commercial farmers (FAO, 2012; Smith *et al.,* 1992). Furthermore, the low production and harvesting costs of Guinea grass gives it an economic edge over imported soybean meal, which is vulnerable to market swings, foreign exchange volatility, and shipping expenses (Ogunbosoye and Babayemi, 2010).

Guinea grass has been shown in numerous studies to have the ability to reduce cattle diet costs. Akinfala *et al.* (2002) claim that local forages, such as Guinea grass, can partially substitute soybean meal, lowering feed costs by up to 30% without appreciably affecting animal performance. Similarly, Nodu and Olorunnisomo (2017) found that feeding growing rabbits a diet consisting of 25–50% Guinea grass instead of soybean meal reduced the cost of feed per unit of weight gain, boosting the production cost-benefit ratio.

The inclusion of Guinea grass to animal diets lowers feed costs while also promoting agricultural and environmental sustainability. According to Okoruwa *et al*. (2015), its use supports the recycling of agricultural by-products, lowers reliance on imported feed components, and promotes the use of domestic resources. In addition to strengthening the local farming systems' resilience, this supports the objectives of sustainable agriculture and food security in developing nations.

Although guinea grass has financial benefits, caution must be exercised to prevent its addition from impairing the balance of nutrients, especially with regard to protein and mineral content. According to reports, excessive amounts of Guinea grass may reduce the diet's total crude protein and mineral content, which could impact animal productivity if it is not adequately supplied (Oluremi *et al.,* 2010); hence mineral supplements and careful blending with other nutrient-rich substances are advised to lessen this (Suttle, 2010). In addition, smallholder farmers could need technical assistance and training to properly prepare and use Guinea grass in animal diets. Optimizing its use and guaranteeing year-round availability requires investments in forage processing equipment, preservation methods (such as silage and hay making), and quality monitoring (Aregheore, 2005; Adebowale *et al.,* 2018).

CHAPTER THREE

MATERIALS AND METHODS

3.1 Experimental Site

The study was carried out in Benin City, Edo State, at the University of Benin's Department of Animal Science Laboratory. The lab had all the required equipment needed to formulate animal feed and analyze the mineral content. Temperatures between 25 to 32°C and relative humidity levels between 70 and 85% were recorded during the study.

3.2 Experimental Design

A total of twenty weaner rabbits of similar age and average body weight were used for the study. The animals were purchased from a reputable breeder and were allowed a 2 weeks acclimatization period. During this period, they were dewormed and vaccinated as required.

The animals were housed in well-ventilated cages with free access to clean water. Proper hygiene and biosecurity measures were maintained throughout the experimental period to ensure the health and wellbeing of the animals. The animals were randomly allocated to four dietary treatments in a Completely Randomized Design (CRD), with five treatments and four replicates.

3.3 Experimental Diets

Four experimental diets were designed to be isonitrogenous (same crude protein content) and isocaloric (same energy content) to meet the nutritional needs of growing rabbits, as suggested by the National Research Council (NRC, 2012). The primary goal was to assess the impact of replacing soybean meal with Guinea grass leaf meal (GGLM) at graded levels. Treatments were:

T1 (Control): 0% GGLM, 100% Soybean Meal

T2: 10% GGLM replacing SBM

T3: 20% GGLM replacing SBM

T4: 30% GGLM replacing SBM

Table 3.1: Feed Formulation Table

Feedstuff	Control (0%)	(10%)	(20%)	(30%)
Maize	5.37	5.37	5.37	5.37
SBM	1.60	1.44	1.28	1.12
PKC	1.40	1.40	1.40	1.40
WO	1.20	1.20	1.20	1.20
DCP	0.03	0.03	0.03	0.03
GGLM	0.00	0.16	0.32	0.48
L/S	0.28	0.28	0.28	0.28
Salt	0.05	0.05	0.05	0.05
Premix	0.08	0.08	0.08	0.08

The component list comprised maize, soyabean meal, Palm kernel cake, Wheat offal, Dicalcium phosphate, Guinea grass leaf meal, Limestone, Salt, and a Vitamin-Mineral premix. Guinea grass leaves were gathered, washed, cut, sun-dried for 3-5 days, and ground into a fine powder before being incorporated into the diets. To guarantee even nutrient distribution, diets were created using the Pearson square method and fully combined with a mechanical feed mixer. For laboratory analysis, samples of each diet were gathered and kept in sealed containers.

3.4 Collection and Processing of Feed Samples

Feed samples from each treatment group were taken during feed preparation and promptly stored in a clean, airtight container with a label to prevent contamination and nutrient loss. The samples were brought to the laboratory, oven-dried at 65°C for 24 hours, and processed using a laboratory hammer mill to pass through a 1-mm sieve.

Each sample was analyzed in triplicates to obtain consistent and reliable values. Samples were stored in desiccators to prevent moisture absorption pending mineral and proximate analysis. This ensured the integrity and accuracy of the results obtained.

3.5 Determination of Mineral Composition

The mineral composition of the diets was determined using standard procedures of the Association of Official Analytical Chemists (AOAC, 2019). The major minerals analyzed included:

Macro minerals: Calcium (Ca), Phosphorus (P), Magnesium (Mg), Sodium (Na), Potassium (K)

Microminerals: Iron (Fe), Zinc (Zn), Manganese (Mn), and Copper (Cu)

Analytical Methods Used: Feed samples were digested using wet digestion with a mixture of concentrated nitric acid (HNO_3) and perchloric acid ($HClO_4$) in a ratio of 2:1. The digestion was performed on a hot plate until the solution became clear.

Determination: Atomic Absorption Spectrophotometry (AAS) was used to determine the concentrations of Fe, Zn, Cu, Mn, Ca, Mg.

Flame Photometry: Used to determine Na and K.

Vanadomolybdate Method: Employed for the colorimetric determination of phosphorus.

All readings were compared against standard solutions to ensure precision.

3.6 Data Collection

The primary data collected for this study included:

1.Mineral concentrations (mg/kg or %): of each formulated diet for all nine minerals of interest.

2.Proximate composition: (i.e., crude protein, crude fiber, ash, crude fat, ether extract, moisture content, and nitrogen-free extract) to support mineral bioavailability and nutrient balance.

Data collection was meticulously done using standardized tools and recorded on data sheets for statistical processing.

3.7 Statistical Analysis

All data were statistically analyzed using GenStat. The data were analyzed using one-way Analysis of Variance (ANOVA) to ascertain the significance of differences among the dietary treatments, and the mean values and standard deviations were calculated for each variable.

Where significant differences ($p < 0.05$) occurred, Duncan's Multiple Range Test (DMRT) was used as a post-hoc test to separate the means.

CHAPTER FOUR

RESULTS

4.1 Mineral Composition of the Experimental Diets

The mineral composition of the four formulated diets, in which Guinea grass was used to replace soybean meal at varying inclusion levels (0%, 10%, 20%, and 30%), is presented in Table 4.1. The analyzed minerals include calcium (Ca), sodium (Na), iron (Fe), zinc (Zn), potassium (K), phosphorus (P), chromium (Cr), manganese (Mn), and lead (Pb).

The results revealed significant ($p<0.05$) variations in the concentrations of most minerals across the treatments, indicating that the inclusion of Guinea grass had a measurable effect on the mineral composition of the diets. Among the treatments, Diet D (30% Guinea grass inclusion) recorded the highest concentrations of major and trace minerals except chromium, while Diet B (10% inclusion) generally had the lowest values.

Calcium (Ca) content increased progressively with higher levels of Guinea grass, ranging from 3126.81 mg/kg in Diet A (0%) to 5614.33 mg/kg in Diet D (30%). This marked increase suggests that Guinea grass is rich in calcium, an essential element for skeletal formation and metabolic activities in animals. The increase in Ca concentration between the control (A) and the highest inclusion (D) diets was statistically significant ($p<0.05$).

Similarly, sodium (Na) concentration rose from 141.25 mg/kg in Diet A to 251.23 mg/kg in Diet D, showing that Guinea grass contributed significantly to dietary sodium content. Sodium is crucial for maintaining osmotic balance and regulating body fluids, which are vital for animal growth and feed efficiency.

Iron (Fe) and zinc (Zn) concentrations followed the same trend, increasing from 61.22 and 33.37 mg/kg in Diet A to 124.43 and 65.10 mg/kg, respectively, in Diet D. The steady rise in Fe and Zn contents implies that Guinea grass contains appreciable quantities of these micronutrients, which are essential for oxygen transport, enzymatic reactions, and immune function.

Potassium (K) and phosphorus (P), both important macro minerals, also increased significantly with Guinea grass inclusion. Potassium content ranged from 124.03 mg/kg in Diet A to 165.18 mg/kg in Diet D, while phosphorus rose from 415.10 mg/kg in Diet A to 520.30 mg/kg in Diet D. These findings confirm that Guinea grass contributes meaningfully to the mineral density of livestock diets and can enhance their nutritional quality when used in feed formulation.

Interestingly, chromium (Cr) displayed an inverse trend compared to other minerals. Its concentration was highest in Diet B (0.2105 mg/kg) and lowest in Diet D (0.0533 mg/kg). This suggests that increasing levels of Guinea grass might have a dilutional effect on chromium concentration, possibly due to the lower intrinsic Cr content of the grass relative to soybean meal. Despite this decline, the chromium levels across all treatments remained within the acceptable nutritional range for livestock.

Manganese (Mn) content increased steadily across treatments, with values ranging from 18.07 mg/kg in Diet A to 32.53 mg/kg in Diet D. This increase underscores the mineral-rich nature of Guinea grass, as manganese plays a key role in bone development and enzyme activation in animals.

Lead (Pb), a potentially toxic heavy metal, was detected in small quantities across all diets, ranging from 0.0533 mg/kg in Diet B to 0.1500 mg/kg in Diet D. Although the values increased slightly with Guinea grass inclusion, they remained well below the permissible limit for livestock feeds (0.5 mg/kg) as recommended by the World Health Organization (WHO, 2011). This indicates that the inclusion of Guinea grass in the diets does not pose any heavy metal toxicity risk to animals.

The standard error of mean (SEM) values for all minerals were relatively low, indicating a high level of precision and consistency in the analytical results. The significant differences ($p<0.05$) observed among the dietary treatments for most minerals confirm that the substitution of soybean meal with Guinea grass affected the mineral profile of the formulated diets in a dose-dependent manner.

Overall, these results suggest that Guinea grass is a valuable source of both macro- and micro-minerals, and increasing its inclusion in livestock feed formulations enhances the mineral density of the diet without exceeding safe levels for animal consumption.

Table 4.1: Mineral Composition Of Four Formulated Diets Using Guinea Grass As Partial Replacement For Soybean Meal

Minerals	A (0%)	B (10%)	C (20%)	D (30%)	SEM
Ca (mg/kg)	3126.810^b	2221.670^a	3477.330^c	5614.330^d	1.913
Na (mg/kg)	141.250^b	87.250^a	181.070^c	251.230^d	0.270
Fe (mg/kg)	61.220^b	44.740^a	98.640^c	124.430^d	0.273
Zn (mg/kg)	33.370^b	22.590^a	55.230^c	65.100^d	0.311
K (mg/kg)	124.030^b	98.250^a	152.300^c	165.180^d	0.975
P (mg/kg)	415.100^b	380.700^a	485.100^c	520.300^d	2.910
Cr (mg/kg)	0.151^c	0.211^d	0.083^b	0.053^a	0.005
Mn (mg/kg)	18.070^b	12.270^a	25.130^c	32.530^d	0.242
Pb (mg/kg)	0.080^b	0.053^a	0.120^c	0.150^d	0.005

SEM: STANDARD MEAN ERROR

Values with different superscripts (a, b, c, d) within the same row differ significantly ($p<0.05$).

4.2 Interpretation of Results

The result shows that increasing the level of Guinea grass meal in the diets from 0% to 30% significantly ($p<0.05$) influenced the mineral content of the diets. Diet D (30% inclusion of Guinea grass) consistently recorded the highest concentrations for most minerals such as calcium (5614.33 mg/kg), sodium (251.23 mg/kg), iron (124.43 mg/kg), zinc (65.10 mg/kg), potassium (165.18 mg/kg), phosphorus (520.3 mg/kg), and manganese (32.53 mg/kg).

Conversely, Diet B (10%) showed the lowest concentrations for most of these minerals, indicating that moderate inclusion of Guinea grass did not enhance mineral deposition as much as higher inclusion levels. Chromium (Cr) showed an inverse trend, with the highest value (0.2105 mg/kg) in Diet B (10%) and the lowest (0.0533 mg/kg) in Diet D (30%), suggesting a dilution effect of Guinea grass on this trace element.

Lead (Pb), though present in trace amounts across treatments, increased slightly with increasing Guinea grass levels, ranging from 0.0533 mg/kg in Diet B to 0.1500 mg/kg in Diet D. However, these levels remain below the toxic limits recommended by the World Health Organization (WHO, 2011), implying the safety of the formulated diets.

CHAPTER FIVE

DISCUSSION

The results of this study revealed that the replacement of soybean meal with Guinea grass (*Panicum maximum*) leaf meal significantly influenced the mineral composition of the formulated diets. All the analyzed minerals (Ca, Na, Fe, Zn, K, P, Cr, Mn, and Pb) varied across treatments, with most showing a progressive increase as the level of Guinea grass inclusion increased. This pattern suggests that Guinea grass is a valuable source of essential macro and micro minerals required for optimal livestock performance. The variations observed can be linked to differences in the inherent mineral composition of the ingredients used and the nature of soil where Guinea grass was cultivated, as mineral content of forages is often influenced by soil type, maturity stage, and environmental factors (Onwuka, 2006; Akinfemi and Adu, 2010).

Higher degrees of Guinea grass inclusion resulted in a substantial increase ($p < 0.05$) in the calcium (Ca) and phosphorus (P) contents; Diet D (75% Guinea grass) had the highest values. This suggests that guinea grass leaf meal made a significant contribution to the diets' mineral content. According to McDonald *et al.* (2011), calcium and phosphorus are essential for the development of bones and teeth, blood coagulation, enzyme activation, and energy metabolism in livestock.

Given that the ratio of the two minerals in animal diets influences the effectiveness of mineral absorption, the upward trend of both minerals' points to a synergistic balance between them, which is nutritionally desirable. For the majority of farm animals, a Ca:P ratio near 2:1 is deemed ideal (NRC, 2007). Guinea grass's capacity to absorb minerals from deep soil layers through vast root systems may also be the cause of the higher calcium and phosphorus concentrations found in this study. These results support the findings of Akinfemi and Adu

(2010) and Esonu *et al*. (2012), who reported that tropical grasses have high levels of calcium and phosphorus and that, with the right drying and processing, they could be used as mineral supplements.

Sodium (Na) and potassium (K) concentrations increased progressively as the inclusion of Guinea grass leaf meal increased. Sodium plays an important role in osmotic balance, nutrient transport, and nerve impulse transmission, while potassium is vital for muscle contraction and enzyme activation (Onwuka, 2006; McDonald *et al.,* 2011).

The elevated Na and K levels in diets containing higher proportions of Guinea grass imply that the plant is a rich reservoir of these electrolytes. This could be due to its ability to absorb and retain water-soluble minerals from the soil. The findings align with those of Olorunnisomo (2011), who reported higher potassium and sodium concentrations in Guinea grass silage compared to other tropical forages.

Moreover, the balance between sodium and potassium is crucial in maintaining proper acid-base equilibrium and physiological fluid balance in animals. The significant increase in both elements suggests that Guinea grass could enhance feed palatability and support metabolic stability when incorporated into livestock diets.

The concentrations of iron (Fe) and zinc (Zn) increased significantly with rising levels of Guinea grass inclusion. Iron is essential for hemoglobin synthesis and oxygen transport in blood, while zinc plays a major role in enzymatic functions, immune system activation, and tissue repair (NRC, 2007; Aduku, 2005).

The elevated Fe and Zn levels observed in this study confirm that Guinea grass contains appreciable quantities of trace minerals that contribute to improved nutritional value of the feed. Similar results were reported by Akinfemi and Adu (2010), who found that processed Guinea grass had relatively high iron and zinc content suitable for ruminant feeding.

Zinc deficiency in animal diets often leads to reduced growth rate, poor reproduction, and delayed wound healing, so the increased zinc concentration with Guinea grass inclusion is a positive nutritional outcome. The results also align with findings of Esonu *et al.* (2012), who observed that green forages are often richer in zinc and iron than cereal-based feeds.

The concentration of manganese (Mn) rose steadily with increased Guinea grass inclusion, indicating that the grass is a good source of this trace element. Manganese is important in bone formation, metabolism of carbohydrates, and activation of enzymes involved in reproduction and digestion (Onwuka, 2006; McDonald *et al*., 2011). The findings are consistent with Akinfemi and Adu (2010), who observed that Guinea grass contributes significantly to manganese content in ruminant diets. The highest value obtained in Diet D supports the potential of the grass to meet the Mn requirements of growing livestock.

On the other hand, chromium (Cr) and Guinea grass inclusion had an inverse association. At 0.2100 mg/kg, Diet B had the greatest Cr content, while Diet D had the lowest (0.0533 mg/kg). This implies that compared to guinea grass, soybean meal has higher natural chromium contents. Although too much chromium might be harmful, it improves insulin sensitivity and glucose metabolism (NRC, 2007). Consequently, it may be advantageous to keep mineral balance within safe dietary bounds by reducing the concentration of Cr when Guinea grass levels rise. This pattern is consistent with general findings in mineral nutrition, as leguminous plants can accumulate higher concentrations of certain trace minerals, including chromium, compared to grasses (Suttle, 2010).

Concentrations of lead (Pb) were usually low throughout treatments, but they marginally rose as more Guinea grass was added. The increasing trend may indicate soil contamination or ambient exposure during plant growth or drying, even if the readings stayed within the FAO/WHO (2013) recommended allowable level of 0.3 mg/kg. According to Ogunkunle and Fatoba (2014), forage plants cultivated close to industrial zones or roadsides frequently develop trace levels of lead from dust and car emissions. The Pb quantities found in this study, however, are not concerning and do not endanger the health of cattle. To ensure feed safety and reduce the possibility of heavy metal buildup, environmental parameters related to forage harvesting must be closely monitored.

The overall trend observed in this study demonstrates that the mineral content of the experimental diets improved with higher inclusion levels of Guinea grass leaf meal, except for chromium which decreased. The results indicate that Guinea grass can successfully replace soybean meal in livestock feed formulations, not only as a protein and fiber source but also as a mineral-enriching ingredient. The variations among treatments may be due to differences in the chemical composition of the plant materials and their ability to absorb minerals from the soil. The enhancement in essential minerals like calcium, phosphorus, potassium, iron, and zinc suggests that animals fed on these diets would likely benefit from improved bone strength, metabolic efficiency, and overall physiological function. This aligns with previous studies by Akinfemi and Adu (2010) and Onwuka (2006), who concluded that properly processed Guinea grass could be incorporated into animal diets to improve mineral intake without compromising health or performance. Hence, Guinea grass stands out as a sustainable and cost-effective feed resource in livestock nutrition programs, particularly in tropical regions where conventional protein sources such as soybean meal are expensive or scarce.

CHAPTER SIX
CONCLUSION AND RECOMMENDATION

6.1 Conclusion

The present study evaluated the mineral composition of four formulated diets in which Guinea grass (*Panicum maximum*) replaced soybean meal at varying inclusion levels (0%, 10%, 20%, and 30%). The findings revealed that Guinea grass inclusion significantly influenced the mineral composition of the diets, resulting in higher concentrations of essential macro- and micro-minerals such as calcium (Ca), sodium (Na), iron (Fe), zinc (Zn), potassium (K), phosphorus (P), and manganese (Mn).

These mineral concentrations gradually rise as guinea grass levels rise, indicating that the forage is high in vital minerals that support healthy animal growth, bone production, metabolism, and general physiological function. In contrast, the slight increase in lead (Pb) concentrations across the diets stayed within the safe tolerable limits advised by international feed safety standards (FAO/WHO, 2013). This inverse relationship with chromium (Cr) suggests that its content may be higher in soybean meal than in Guinea grass.

Overall, the results demonstrate that partial or complete replacement of soybean meal with Guinea grass in livestock feed formulation can improve the mineral density of diets without causing any toxic accumulation of heavy metals. This substitution could also reduce the high cost associated with conventional protein sources like soybean meal while promoting the utilization of locally available and affordable feed resources.

Therefore, Guinea grass has the potential to serve as an alternative ingredient in ruminant and non-ruminant feed formulation, particularly in regions where soybean meal is scarce or expensive. Its inclusion in feed can enhance mineral intake and contribute to the sustainability of livestock production systems.

6.2 Recommendations

1. Guinea grass can safely replace soybean meal up to 30% in livestock diets.
2. Feed processors should use properly dried or processed Guinea grass to enhance nutrient availability.
3. Grass should be sourced from clean areas to avoid heavy metal contamination.
4. Further studies should assess the effects of Guinea grass inclusion on growth performance and feed efficiency.

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