UNIVERSITY FOR DEVELOPMENT STUDIES

#### NUTRITIONAL IMPROVEMENT OF FALSE YAM (ICACINA OLIVIFORMIS) SEED MEAL

FOR POULTRY

MOHAMMED ALHASSAN

THESIS SUBMITTED TO THE DEPARTMENT OF ANIMAL SCIENCE, FACULTY OF AGRICULTURE, UNIVERSITY FOR DEVELOPMENT STUDIES IN PARTIAL FULFILMENT OF THE REQUIREMENTS FOR THE AWARD OF DOCTOR OF PHILOSOPHY DEGREE IN ANIMAL NUTRITION.

#### UNIVERSITY FOR DEVELOPMENT STUDIES

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BY

MOHAMMED ALHASSAN (B.Sc. Agriculture Technology, M. Phil. Animal Science, Nutrition

Option)

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JUNE 2019

#### DECLARATION

#### Student

I hereby declare that this thesis is the result of my own original work and that no part of it has been presented for another degree in this University or elsewhere.

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------------------------	-----

Name: Mohammed Alhassan

#### Supervisors

I hereby declare that the preparation and presentation of the thesis was supervised in accordance with the guidelines on supervision of thesis laid down by the University for Development Studies.

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#### ABSTRACT

Five separate experiments were conducted to assess the nutritive value of false yam seed meal (FYSM) when subjected to sequential processing techniques (water treatment, chemical treatment and blanching). Matured false yam (Icacina oliviformis) fruits were harvested and seeds extracted. Five different FYSM samples were prepared. One untreated sample (Un\_T) was prepared by crushing fresh false yam seed and sun-dried to 12% moisture. Four treated samples (T), were crushed and each soaked in water (1:2; w/v) for 12 d with water replaced every 3 d. Afterwards, each sample was re-soaked in 1 M concentration of urea (Urea\_T), sodium chloride (NaCl\_T), sodium hydroxide (NaOH T) or potassium hydroxide (KOH T), for 24 h, washed, blanched and sun-dried to a moisture content of 12%. In experiment 1, the nutrient compositions of the untreated and treated FYSM samples were determined. The range of nutrient contents between untreated and mean of treated FYSM samples were DM (91.3 - 91.2%), CP (13.2 - 4.5%), EE (1.5 - 2.13%), NFE (71.4 - 79.9%), ash (2.5 - 2.1%), Ca (280 - 108 mg/kg), mg (52.8 - 15.1 mg/kg), K (110.6 -272.6 mg/kg), Na (557.7 - 618.2 mg/kg) and P (0.31 - 1.44 mg/kg). Processing of the 4 FYSM samples drastically reduced the concentrations of essential amino acids compared to the untreated samples: Lys. (0.35 - 0.11%), Met. (0.1 - 0.03%), Leu. (0.9 - 0.27%), Ala. (0.6 - 0.18%), Tryp. (0.16 - 0.06%), Val. (0.5 - 0.17%), Threo. (0.42 - 0.12%), Arg. (1.43 - 0.27%), His. (0.36 - 0.14%) and Phenylala. (0.58 - 0.17) respectively. The concentrations of terpenes were reduced by processing; Urea\_T (91.9%), NaCl\_T (88.3%), NaOH\_T (84.1%) and KOH\_T (81.6%). Similarly, the concentrations of saponins were reduced by processing; Urea T (68.0%), NaCl T (76.8%), NaOH\_T (69.6%) and KOH\_T (68.0%). Experiment 2 evaluated feed preference and apparent nutrient digestibility of a maize-based diets containing treated false yam seed meals (TFYSM) at 0, 5, 10, 15, 20, 25, 30, 35, 40, 45 and 50%. Feed preference tests were conducted with 21 day-old



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broiler chicks (Cobb 500) using a RCBD. Apparent nutrient digestibility trial was conducted with broiler chicks in 5x6 factorial design for 15 days using the total collection method. The preference data indicated that feed intake was not compromised when maize was substituted by NaCl T, NaOH T, and KOH\_T FYSM at all levels, while Urea\_T FYSM was tolerated up to 30% and the Un\_T FYSM at 5% dietary level. Dry matter digestibility was generally high (> 75%) in all diets except for Un T (68%). Control diet (74.32%) and NaOH T diets had similar CP digestibility (74.7%), but was higher (P<0.05) than KOH\_T diets (69.2%), while Un\_T diets had reduced CP digestibility (58%). Digestibility of NFE ranged from 77.4% (Un\_T) to 93.8% (KOH\_T). Experiment 3 evaluated apparent metabolizable energy (AME) and nutrient metabolisability (NM) of the maize-based diets containing TFYSM using the same experimental design in experiment 2. AME of the diets indicated increasing trend as TFYSM was increased in the diets. Diets containing TFYSM improved (P<0.001) DM, CP and carbohydrate metabolisability. There was interaction (P<0.017) between method of processing and level of inclusion on gross energy metabolisability. Experiment 4 examined the effect of NaOH\_T FYSM on the growth performance and blood profile of female broiler chickens (Cobb 500). Four dietary treatments containing NaOH T FYSM at 0, 100, 300 and 500 g/kg in broiler finisher diets were tested using a Completely Randomized Design and each treatment had quadruplicate lots (8 birds/replicate). Feed and water were provided ad libitum from 21-56 d of age. Data collected were analysed using GenStat. Growth performance variables of all treatment groups were similar (P>0.05). Birds fed diets containing 0, 100 and 300g/kg NaOH T FYSM had similar (P>0.05) (78.5-79.5%) carcass dressing, but higher (P<0.003) than those fed diets at 500g/kg. All haematological variables were similar (P>0.05) except RBC in birds fed NaOH\_T FYSM diet. Liver function test indicated significant (P<0.048) improvement in only albumin values when NaOH\_T FYSM was included beyond 100g/kg.



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However, all renal function variables increased (P<0.05) with the inclusion of NaOH\_T FYSM in the diets. Economics of feeding NaOH T FYSM revealed a significant (P<0.05) reduction in total feed cost per bird and feed cost per kg gain. Meat quality of the broilers was evaluated and the results indicated that NaOH\_T FYSM inclusion up to 500g/kg improved (P<0.001) protein content of the meat. Furthermore, carcass dress weight, primal cuts and sensory characteristics were not affected (P>0.05). Lipid oxidation in the meat differed (P<0.001) significantly among the treatment groups but were much lower than the maximum permissible level of 25 meq/kg meat. In experiment 5, the effect of varying levels (0, 100, 300 and 500 g/kg) of KOH\_T FYSM on growth and blood profile of male broiler chickens was evaluated as in experiment 4. Growth performance variables as well as percent carcass dressing of all treatment groups were similar (P>0.05). Relative heart weight increased (P<0.05) whereas liver and spleen weights decreased (P<0.05). Among the haematological variables evaluated, Hb, MCV, MCH and MCHC values reduced (P<0.05) significantly with increasing inclusion levels of KOH T FYSM in the diets. Liver function variables remained unchanged except for serum albumin which differed (P<0.05) between the control birds and their counterparts. All renal function variables were similar (P>0.05). Economic variables were similar (P<0.05) for all diets. Similar meat quality evaluation procedures as in experiment 4 was undertaken in experiment 5. The results indicated that KOH\_T FYSM inclusion up to 500g/kg did not affect (P>0.05) carcass characteristics of the birds. Relative weight of breast muscle decreased (P<0.05) at inclusion level of 100 g/kg and subsequently improved at 300 and 500 g/kg. Sensory attributes of the meat were similar (P>0.05) among the treatment groups. The inclusion of KOH\_T FYSM improved (P<0.001) protein content of the meat. Lipid oxidation in the meat differed (P<0.001) significantly among the treatment groups but were much lower than the maximum permissible level of 25 meq/kg meat. It can be concluded that



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sequential use of water-based and chemical treatment methods using NaOH or KOH offers the potential to use FYSM as an alternative feedstuff in broiler chicken diets.



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#### DEDICATION

I dedicate this thesis to my parents Alhaji Alhassan Musah and Madam Abiba Alhassan for the trust they had in me and the investments they committed to my education.



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# LIST OF ABREVIATIONS

GDP	Gross domestic product
USDA	United States Department of Agriculture
IITA	International Institute of Tropical Agriculture
FAO	Food and Agriculture Organization
NRC	National Research Council
NRI	National Research Institute
ANFs	Anti-nutritional factors
NSC	Neem seed cake
TME	True metabolizable energy
TMP	True metabolizable protein
СР	Crude protein
EE	Ether extract
CF	Crude fibre
NFE	Nitrogen free extract
NAS	National academy of Sciences
DM	Dry matter
RFYSM	Raw false yam seed meal
SFYSM	Soaked false yam seed meal
BFYSM	Boiled false yam seed meal
ME	Metabolizable energy
MJ	Mega joule
NSP	Non-starch polysaccharide



TSH	Thyroid stimulating hormone
EDTA	Ethylene diamine tetra-acetic acid
IFCC	International Federation for Clinical Chemistry
CLSI	Clinical and Laboratory Standards Institute
MOLM	Moringa oleifera leaf meal
TOLE	Telfaria occidentalis leaf extract
РКС	Palm kernel cake
FCTW	Fermented cassava tuber waste
WLTS	Waterleaf top supplement
MFCP	Microbial fermented cassava peel
MFCSR	Microbial Fermented Cassava Starch Residue
GGT	Gamma glutamyl transferase
SDFYTM	Sun-dried false yam tuber meal
SBFYTM	Soaked/boiled false yam tuber meal
FFYTM	Fermented false yam tuber meal
SFYTM-SP	Saltpetre-treated soaked false yam tuber meal
FSFYSM	Fermented false yam seed meal
FYS	False yam seed
FYSM	False yam seed meal
AOAC	Association of official analytical chemists
HPLC	High power liquid chromatography
IVGP	In vitro gas production
HGT	Hohenhein gas test



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PEG	Polyethylene glycol
AME	Apparent metabolizable energy
BSI	British standard institute
POV	Peroxide value



#### **CHAPTER ONE**

#### **1.0 INTRODUCTION**

Poultry meat is recognized as the fastest growing component of global meat in terms of production, consumption and trade. Both developed and developing countries including Ghana are playing a significant role in this expansion of poultry meat output. The worldwide poultry sector growth is estimated at 62.2% and a population growth of 40.4% from 2005 to 2050 (http://esa.un.org/unpp/p2k0data.asp). This expansion in the poultry sector is being driven by increasing GDP and rapid population growth.

In the developing countries, where almost all world population increases take place, consumption of meat has been growing at 5 to 6 % p.a. and that of milk and dairy products at 3.4 to 3.8 % p.a. in the last few decades. Aggregate agricultural output is being affected by these trends, not only through the growth of livestock production, but also through the linkages of livestock production to the crop sector which supplies the feeding stuffs (mainly cereals and oilseeds) (de Haan et al., 1998).

Feed development and supply is a major factor promoting the development of the poultry industry. This is because feed is the single largest cost item in poultry production, accounting for 55-64 percent of variable costs and maize (energy) and soybean meal (protein) are the dominant feed ingredients in broiler rations (USDA, 2004).



Maize is the main cereal grain used in poultry feed in sub-Saharan Africa as source of dietary energy. It is also a staple food for an estimated 50% of the population (International Institute of Tropical Agriculture (IITA), 2009).

Therefore, there has been a stiff competition between humans and animals for this commodity, thereby causing shortages and hikes in prices.

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This has led to high feed cost with a consequent negative impact on poultry productivity and profitability. Bell and Weaver (2002) reported that about 85% of the world chicken dietary energy is derived from maize. Also, the production of cereal grains (e.g. maize) in Africa, Asia and Pacific nations has never been adequate for human consumption and industrial use (Reddy and Qudratullah, 2004).

Therefore, it would be preferred if poultry farmers could prepare feed using alternative locally available energy feed resources including non-conventional feedstuffs to reduce feed cost or deal with occasional shortage of maize. The use of non-conventional feedstuffs as substitutes for grains and other feedstuffs have been suggested to be the most active area of animal nutrition research in the tropical world (Ikani and Adesehinwa, 2000) due to abundant availability of non-conventional feedstuffs or insufficient production of grains. These non-conventional feedstuffs include agro-industrial by-products such as cereal brans (Smith, 1990) and oilseed cakes (Nelson, 1998), some food crops like cassava (Smith, 1988), crop residues such as cassava leaf meal (Okai *et al.*, 1984) as well as new plant resources such as false yam (Dei *et al.*, 2011).

The use of false yam (*Icacina oliviformis*) tuber as new feed resource has been shown to have the potential to reduce the demand for maize as a major energy source of feed ingredient for poultry (Dei *et al.*, 2011).



False yam (*Icacina oliviformis*) is a drought-tolerant and fire-adapted shrub that grows in the Savanna regions of West and Central Africa. It produces both seeds and large tubers. Reports by Fay (1993) showed that false yam's tuberous root yield can be as high as 20 mt/ha [compared with ~30 mt/ha for normal yam] and a relatively low seed yield [0.214 mt/ha compared with ~1.3 mt/ha of maize] (Fay, 1991). It is envisaged that with proper agronomic practices, both seed and tuber yields can be substantially increased if cultivated.

The tuber and seed contain toxic compounds called gum resins which are identified as terpenes (Vanhaelen *et al.*, 1986). The gum resins are present in quantities in the tuber ranging from 0.9-2.8% (NRI, 1987), therefore it cannot be eaten directly (Fay, 1991). These anti-nutritional factors (ANFs) reduce the palatability of feed when given to animals (McDonald *et al.*, 2002).

Apart from terpenes (Vanhaelen *et al.*, 1986) as a major ANF in false yam ranging from 0.9-2.8% (NRI, 1987) and as high as 3.75% (Dei et al., 2011), Timothy and Idu (2011) reported the presence of alkaloids, saponins and tannins in the false yam. Rufus (2010) after a phytochemical screening revealed the presence of phenols in the plant. Flavonoids are also present (Nakayoma and Yamada, 1995). Hydrocyanic acid, oxalic acid and phytic acid were found in some species of false yam tuber, especially in *Icacina manii* (Antai and Nkwelang, 1999). These anti-nutritional factors reported in the false yam could have negative impact on the animal's nutrition and production. It has been reported that the objective of processing feedstuffs includes the extraction of non-protenacious component; to remove undesirable taste and odour component; to remove or

inactivate nutritionally undesirable components; and to prepare a protein material that is suitable for application in food products (Iwe, 2003). Processing significantly reduces anti-nutritional factors in foods (Fagbemi et al., 2005).



It is necessary to develop improved methods for detoxifying false yam seeds and tubers so that their potential for use in poultry diets is completely achieved. Processing methods for use in commercial feed production must be simple, economical, feasible and inexpensive. Physical and chemical methods are available for use in processing. In many instances, usage of only one method may not effect the desired removal of anti-nutritional substances and a combination of two or more methods may be required.

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It is known that aqueous solutions can be used in the extraction processes of terpenes (Kamphoff et al., 2007). However, some metabolic activities can take place during soaking which will affect the constituent compounds (Vidal-Valverde et al., 1992). Soaking false yam seed in water for 12 days improved its feed value for poultry over the unprocessed seed (Dei et al., 2012; Mohammed and Dei, 2012). According to Roessler *et al.* (2017), soaking in water removes hydrophilic compounds from the false yams tuber more easily than lipophilic ones.

Chemical treatments that have achieved significant reduction in anti-nutritional factors from plant origin are mostly alkaline in nature (D'Mello and Walker, 1991). None of the chemicals mentioned has been used to treat false yam seeds to remove anti-nutritional factors.

Also, limited research has been conducted to include a sequentially treated false yam seed meals in poultry diets. That is multistage processing involving soaking and chemical treatment using sodium hydroxide and potassium hydroxide. Therefore, this study was conducted to determine chemical compositions and suitability of sequentially treated false yam seeds as feed for poultry.



#### **1.1 RESEARCH QUESTIONS**

- 1. Do false yam seed meals contain adequate amounts of nutrients?
- 2. Do various treated false yam seed meals in the diets of broilers interfere with feed intake and nutrient digestibility?
- 3. How much metabolizable energy can be utilized from these treated false yam seed meals?
- 4. Can dietary supplementation of sodium hydroxide/potassium hydroxide-treated false yam seed meals improve growth performance and blood profile of broiler chickens?
- 5. Can the use of sodium hydroxide/potassium hydroxide-treated false yam seed meals in broiler rations affect their meat quality and sensory characteristics?



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#### **1.2 MAIN OBJECTIVE**

To evaluate the effect of multi-stage processing methods on the nutritive value of false yam seed meal for poultry.

#### **1.3 SPECIFIC OBJECTIVES**

The specific objectives were:

- 1. To determine the proximate composition (CP, EE, CF, NFE and Ash), mineral composition, essential amino acids concentration and some anti-nutrients of processed and unprocessed false yam seed meals.
- 2. To determine the effect of four (4) processing methods (Urea, NaCl, NaOH and KOH) of false yam seed meals on broiler chicken feed preference and apparent nutrient digestibility.
- 3. To determine the effect of four (4) processing methods of false yam seed meals on apparent metabolizable energy and nutrient metabolisability.
- 4. To evaluate growth performance, carcass characteristics, haematology and serum biochemistry of broiler chickens fed diets containing sodium hydroxide/potassium hydroxide-treated false yam seed meals.
- 5. To evaluate meat quality of broiler chickens fed diets containing sodium hydroxide/potassium hydroxide-treated false yam seed meals.



#### **CHAPTER TWO**

# 2.0 LITERATURE REVIEW

#### 2.1 Taxonomy and geographical distribution of false yam plant

False yam is a perennial plant scientifically known as *Icacina oliviformis* and belongs to the family *Icacinaceae*. According to Fay (1987), the plant was first named by J. Poiret as *Hirtella oliviformis* in 1813 but was later changed to *Icacina senegalensis* in 1823 by Andrien de Juss; and again changed to *Icacina oliviformis* by Raynal in 1975. Other species of false yam found are *Icacina trichantha* in Western Nigeria (Burkhill, 1985) and *Icacina manni* in Cross River State of Nigeria (Umoren *et al.*, 2008).

The false yam plant (Plate 2.1) is indigenous to West and Central Africa (Styslinger, 2011) and is found growing wild on light sandy soil in the savanna areas of Senegal, Gambia, Guinea, the northern part of Ghana and parts of Sudan (Fay, 1987; NAS, 2008). It thrives on a variety of soils and in several plant communities (Fay, 1991). It produces bright yellowish or bright-red fruits (Plate 2.2) containing a single seed and a large tuber (Plate 2.3).

Kingdom	Plantae –plants			
Subkingdom	Tracheobionta –vascular plants			
Super division	Permatophyte –seed plants			
Division	Magnoliophyta –flowering plant			
Class	Magnoliopsida-dicotyledons			
Subclass	Rosidae			
Order	Celastrales			
Family	Icacinaceae –icacina family			
Genus	Icacina			
Species	Senegalensis, 'Mumu', Oliviformis			

Sources: Raynal (1975)

According to NAS (2008), the false yam (*Icacina oliviformis*) is a shrubby perennial variable in form, which sends up glabrous or pubescent erect leafy shoots from a large underground fleshy tuber. The aerial stems are light green and may reach about 1m in height.

Raynal (1975) reported that the false yam plant falls under the division Magnoliophyta (flowering plant) (Table 2.1). The flowers are conspicuous, usually white or cream and peduncle, ascending or erect, corymbose cymes, collected into a terminal leafless panicle, or the lower peduncle arising from the axis of reduced leaves (NAS, 2008). The calyx is divided into five sections, the pointed lobes are bright green; the corolla is composed of five narrow, white or creamy-white petals, covered with silky hairs on their outside surface. The fruit is a red ovoid berry, approximately 2.5cm-3cm in length and 2- 2.5 cm in width. It is covered with very short hairs and contains a thin layer of white pulp, approximately 0.26 cm thick, surrounding a single spherical or ovoid seed. Because the false yam plant produces seeds, it falls under the super division permatophyte (seed bearing plants) (Table 2.1).



Plate 2.1: False yam plant



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Plate 2.2: False yam fruit

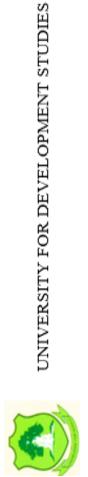
Plate 2.3: False yam tuber

The false yam has different kinds of local names in different parts of African (Fay, 1991) which is known as follows:

- Manankaso in Gambia
- Pane in Sudan
- ➢ Kuraba in Senegal

In Ghana, its local names are as follows:

- > The Ewes call it 'Anyigba fe dzi'
- ➢ The Asantes call it ' Abor ntupe'
- > The Hausas and Dagombas call it 'Tankwara' (Fay, 1991)



#### 2.2.0 Uses of false yam

#### 2.2.1 Food for human and animals

False yam in many countries including Ghana grows in the wild. However, it serves as source of food for humans, particularly during period of food scarcity. The fruit contains a large seed and a thin mesocarp. The flesh can be eaten as snack when the fruit is ripe and mainly by children (Dalziel, 1948). According to NAS (2008), the flour can be used for various food varieties.

The seeds are soaked in water and then ground into flour, which is high in carbohydrates and contains 8 percent protein. It can be a substitute for cassava flour (Styslinger, 2011). Also, the tuber is soaked in water to leach out the toxic component (Fay, 1987), dried and pounded into a white powder, called "gbe-wutu", this can be used in soups, or added to a food known as "igbālò" made from the roasted seeds of water melon (*Citrullus lanatus*) (Burkhill, 1985). The flour can be mixed with millet or beans to make a thick paste known as "enap" in Senegal (NAS, 2008) to prepare porridges (Kay, 1973). The starch can be used for commercial purposes such as tapioca (Fay, 1987). False yam (leaves, seed and tuber) though toxic (Fay, 1987) can be processed into meal for feeding animals. Woot-Tsuen *et al.* (1968) reported that the flour from the tuber contains 74.5% carbohydrate which makes it a right source of energy in the diet of poultry. According to Dei *et al.* (2011), false yam seeds and tubers when processed by boiling and soaking have improved



their nutritive value for poultry. Sun-dried false yam leaves when fed to rabbits have been shown to have no adverse effects on their growth performance (Ansah and Aboagye, 2011).

#### 2.2.2 Ethno medicinal uses of false yam plant

Some members of *Icacina* genus have been reported to exhibit ethno botanical properties. The parts of *Icacina* plants which have been used for ethno medicinal purposes include the leaves and tubers. *Icacina trichantha* is reportedly being used as medicine in rural communities in Nigeria (Asuzu and Abubakar, 1995; Timothy and Idu, 2011). This is supported by the fact that it is regarded as a major handy household medicine for emergency treatment; hence, virtually all households have the macerated tuber in ethanol which is stored in corked bottles. This species has also been cited in other works as to possess analgesic, anti-inflammatory, anti-diabetic activities and antimicrobial activities (Asuzu and Abubakar, 1995).

The tuber of *Icacina trichantha* has also been reported to be used as medicine in treatment of mumps (Rufus, 2010). Antioxidant activities have been reported to be present in some species of *Icacina* (Odukoya *et al.*, 2006). Also, leaves of some species have been known to have antiplasmodial activity. Such species are used in the treatment of malaria (Sarr *et al.*, 2011).

Other species are known to show anesthetic effects in guinea pigs (Asuzu and Abubakar, 1995). Lucindo *et al.* (2008), and Asuzu and Abubakar (1995) have reported anticonvulsant activities in *Icacina trichantha*. Toxicity in certain species however, has been reported (Dalziel, 1947). Tubers of *Icacina trichantha* have been used by herbalists to treat constipation, poisoning, malaria and to induce emesis (Asuzu and Abubakar, 1995). Igbo people in Nigeria consider the species *Icacina trichantha* to be aphrodisiac, and they use it on soft tumours (Burkill, 1985). False yam tubers, leaves and stems are being used for numerous diverse treatments. Leafy twigs in decoction are used for internal hemorrhages and in baths and washes, for cough and all chest infections; and for



feverish states; patients should sleep the night on a bed of newly-cut leaves (Kay, 1973).

Also, extracts from some species have been shown to induce sleepiness and reduce pain in rodents (Asuzu and Abubaker, 1995). Aqueous and methanol extracts of fresh leaves of *Icacina trichantha* have demonstrated a wide range of antimicrobial activities with the methanol extract exhibiting preferable activity than same concentrations of the aqueous extract. Aqueous extract of fresh tuber (Mohammed and Dei, 2017) or seed (Mohammed et al., 2017) of *Icacina oliviformis* replaced favourably with synthetic antibiotic (Oxytetracycline) in cockerel or broiler chicken production without any compromise on health status.

Standard phytochemical screening conducted has revealed the presence of alkaloids, saponins and tannins in both aqueous and methanol extracts of leaves of the plant. Concentrations of these phytochemicals were observed to be higher in the methanol extract than the aqueous extract. The higher concentrations of the phytochemicals in the methanol extract may account for its preferable activity to the aqueous extract. Flavonoids, glycosides, steroids and anthraquinone were however, absent in the aqueous and methanol extracts of *Icacina trichantha* leaves. Both the aqueous and methanol extracts of the plant's leaves have shown activity against gram- positive and gramnegative bacteria, as well as fungi. The antimicrobial potentials demonstrated by these extracts have been dose dependent. *In vitro* studies of these extracts have demonstrated growth inhibition of *Staphylococcus aureus*, *Pseudomonas aeruginosa*, *Escherichia coli*, *Bacillus subtilis*,



Aspergilus niger, and Candida albicans (Timothy and Idu, 2011).

A report by Rufus (2010) on the ethno-botanical uses of *Icacina trichantha*, revealed that the juice from its tubers is used in the treatment of mumps. By the use of both 1, 1-diphenyl-2-picryhydrazyl free radical scavenging and the reducing power (Fe3<sup>+</sup>) methods, methanol extract from *Icacina trichantha* leaves has demonstrated antioxidant activity which is dose specific.

In the same work conducted, phytochemical screening revealed the presence of phenols in the plant's leaves.

The presence of phenols in the leaves of *Icacina trichantha* falls in line with earlier report that there is a strong correlation between phenolic content and antioxidant activity (Velioglu *et al.*, 1998; Odukoya *et al.*, 2005; Odukoya *et al.*, 2006). Methanol extracts of *Icacina trichantha*, folium, *Icacina trichantha*, lignum, *Icacina trichantha*, and radix have been reported to exhibit antioxidant activity and the presence of polyphenols alongside others like alkaloids (Oke and Hamburger, 2002). The antioxidant or antiradical activity exhibited is an indication of the presence of flavonoids (Nakayoma and Yamada 1995).

The treatment of alloxan-induced diabetic mice with the methanolic crude extract of *I. trichantha* leaves demonstrated decreased blood glucose levels (P < 0.01) in a dose-dependent manner (Ezeigbo, 2010). Doses of methanol extract of *Icacina trichantha* tuber had induced sleep in rats and local anaesthetic effects in guinea-pigs. The sleeping time in rats was observed to be dose-dependent. The extract was able to give 80% protection to rats poisoned with pentylenetetrazole but failed to protect rats from strychinine poisoning. It also induced significant dose-dependent analgesia in rats and showed significant muscle relaxant activity in mice (Asuzu and Abubakar,



1995). Chloroform extract from *Icacina trichantha* tubers has proven to significantly inhibit croton oil induced ear edema in mice in a dose-dependent manner than the hexane, ethyl acetate, methanol and water extracts. The chloroform extract has also shown significant reduction in carrageenan-induced paw edema in rats, after oral administration: 50, 100 or 200 mg/kg of the fraction reduced the global edematous response by 15, 20 or 34 %, whereas 10 mg/kg of indomethacin induced 40% inhibition (Asuzu *et al.*, 1999). Other report has it that, the central nervous system active component of *I. trichantha* tuber extract resides almost completely in the chloroform soluble.

This report is buttressed by the fact that chloroform extract of *Icacena trichantha* tuber significantly increased pentobarbitone-induced sleep, reduced sensitivity of mice to pain (analgesia), protected mice from death due to leptazole-induced convulsions and reduced motor co-ordination in treated mice. All these effects are mediated through the central nervous system (Asuzu and Egwu, 1998).

Other results have shown that ethyl acetate extract of *I. trichantha* leaf at the dose of 400 mg/kg was able to reverse paracetamol induced hepatic damage in mature wistar rats better than silymarin (P<0.05). In an *in vivo* study, this same extract has exhibited antioxidant and hepatoprotective activities on paracetamol-induced liver damage in rats. The antioxidant activity did not compare favourably to that of ascorbic acid. No signs of toxicity and death were noticed among the wistar rats. The ethyl acetate extract of *Icacina trichantha* had no adverse effects on rats up to the dose of 2000 mg/kg. This shows that it is relatively safe and corroborates its use in traditional medicine especially as a remedy for hyperglycemia (Ezeigbo, 2010; Udeh and Nwaehujor, 2011).

The report on the hepatoprotective activities of ethyl acetate leaf extract of *Icacina trichantha* on paracetamol-induced liver damage in rats has it that, administration of the ethyl acetate extract of *Icacina trichantha* at the different doses caused a reversal of all the increased levels in the liver enzymes, bilirubin as well as protein (Udeh and Nwaehujor, 2011).



In preliminary *in vitro* tests conducted on the biological activities of *Icacina trichantha* in mice, the ethanol, petroleum ether and aqueous extracts of tubers, roots, stems and leaves did not show contraction of isolated guinea pig ileum up to about concentration of 40.5 mg/ml.

Graded oral doses of the aqueous extract of tubers were reported to produce wet faeces in mice with the number of wet faeces increasing with increasing dose up to 400 mg/kg.

A time-course study of the purgative effect showed the maximum purgative response to be 7-8 h after oral dosing. The aqueous extract of tubers significantly potentiated pentobarbital- induced loss of righting reflex at a dose of 80 mg/kg but did not protect mice from strychnine or leptazole convulsions and death (Asuzu and Ugwueze, 1990).

In an *in vitro* study, conducted by Sarr *et al.* (2011), methanol extract of *Icacina senegalensis* leaves has inhibited growth in *Plasmodium falciparum* chloroquine-sensitive (3D7) and chloroquineresistant (7G8) strains. A preliminary investigation of freeze dried methanolic extract of *I. senegalensis* has revealed no haemolytic effect *in vitro* on red blood cells.

#### 2.2.3 Nutrient composition of false yam

Table 2.3 shows the nutrient composition of false yam tuber and seed as compared to other root/tuber feedstuffs such as cassava, sweet potato, cocoyam and yam. Table 2.4 shows comparative amino acid composition of false yam tuber and seed samples with other root/tuber feedstuffs such as cassava, sweet potato and cocoyam. Table 2.2 shows the comparative nutritional composition of raw, soaked and boiled false yam seed meals.



		Dry matter	Crude protein	Ether extract	Total carbohydrates	Crude fibre	Ash	NFE	Gross energy (kcal/kg)	Sources
Fal	S									
Free	Ĕ	41.0	4.4	1.6	84.5	-	-	-	-	Fay (1991)
Dri	STUDIE	88.3	10.3	0.7	74.5	-	2.8	-	-	NRI (1987)
Sun	5	86.46	5.41	1.6	53.1 (starch)	-	2.2	-	4,067	Dei et al. (2011a)
Sun		87.5	16.45	1.14	-	3.1	1.7	-	3,565	Osei et al. (2013a)
Soa	UNIVERSITY FOR DEVELOPMENT	85.5	5.9	1.5	-	15.7 (NDF)	2.1	74.8	4,067	Dei et al. (2010)
Boi	臣	85.7	6.46	0.98	-	43.12 (NDF)	2.76	-	4,139	Dei et al. (2011a)
Boi	M	86.0	8.84	1.74	-	3.7	2.33	-	3,677	Osei et al. (2013a)
Fal	G									
Rav	Ē.	81.7	0.5	80.7	-	-	-	-	-	Fay (1991)
Rav	T I	87.0	-	72	0.1	0.5	-	-	-	Kay (1973)
Rav	Ë	87.0	0.1	72	-	-	-	-	-	NAS (2008)
Soa	I.	85.9	7.4	0.4	65.5 (starch)	1.7	0.5	-	3,660	Dei et al. (2012c)
Soa	OF	86.1	8.4	0.56	-	9.9 (NDF)	6.3	80.6	4,282	Dei et al. (2012d)
Oth	н									
Fre	ΓY	37.0	1.0	0.3	-	4.4	-	-	1,141	Woolfe (1992)
Free	SIZ	32.0	2.2	0.1	-	5.1	2.80	89.0	-	Jalaludin (1997)
Sun	К.	-	2.9	1.4	-	5.0	2.30	88.4	-	Ravindran et al. (1992)
Boi	ΛE	28.3	0.38	0.04	27.4	0.5	0.46	-	1,112	Favier (1977)
Fre	1Z	30.0	1.5	0.3	-	3.9	-	-	-	Woolfe (1992)
Swe	5	-	4.4	0.6	-	6.9 (NDF)	3.10	-	4,091	Noblet et al. (1990)
Rav		92.69	7.21	4.69	-	1.48	5.13	81.58	1,246	Onu and Madubuike (2006)
Coc		90.70	7.15	4.22	-	1.49	5.12	82.02	1,246	Onu and Madubuike (2006)
Fre		24.0	2.4	0.1	-	1.2	-	-	1,104	Woolfe (1992)
Rav		93.21	10.27	1.15	-	2.31	2.93	76.57	3,756.5	Ezeoch and Ojimelukwe (2012)
Boi		95.26	8.11	0.15	-	1.52	2.48	83.02	3,639.4	Ezeoch and Ojimelukwe (2012)

Table 2.2: Chemical composition of false yam tuber and seed samples compared with other root and tubers (% DM basis)

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Ami	no acid	Sun-dried tuber	Soaked tuber	Sun-dried seed	Soaked (9d) seed	Soaked (15d) seed	Sun-dried cassava root	Fresh sweet potato	Cocoyam	Sun-dried white yam
Ar	DIE	0.792	0.08	0.94	0.57	0.51	0.29	0.34	-	0.660
Gl	STUDIES	0.094	0.08	0.45	0.35	0.32	0.01	-	-	0.525
Hi		0.115	0.16	0.21	0.17	0.16	0.07	0.16	-	0.520
Iso	MEN	0.079	0.08	0.62	0.47	0.43	0.03	-	0.219	-
Le	[dO]	0.124	0.13	0.75	0.56	0.50	0.31	0.57	-	0.750
Ly	DEVELOPMENT	0.192	0.24	0.29	0.22	0.19	0.07	0.41	0.241	0.710
Ph		0.042	0.07	0.44	0.35	0.31	0.03	0.54	0.316	0.530
Μŧ	UNIVERSITY FOR	0.003	0.01	0.05	0.04	0.04	0.03	0.11	0.84	0.548
Th	IY I	0.077	0.07	0.36	0.26	0.24	0.03	0.34	0.257	0.443
Tr	RSI	0.021	0.05	0.08	0.09	0.01	0.29	-	0.88	-
Va	(VE)	0.102	0.13	0.46	0.36	0.34	0.04	0.52	0.382	0.333
Ty	N	-	-	0.28	0.19	0.18	0.01	0.18	0.226	0.410
Су		-	-	-	-	-	-	0.16	0.163	0.527
So		Dei <i>et al.</i> (2011a)	Dei <i>et al.</i> (2012a)	Dei <i>et al.</i> (2012c)	Dei <i>et al.</i> (2012c)	Dei <i>et al.</i> (2012c)	Gil and Buitrago (2002)	Gohl (1975)	FAO (1970)	Ogunlade <i>et</i> <i>al.</i> (2011)

Table 2.3: Comparative amino acid composition (%DM) of false yam tuber and seed samples with other root and tubers

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# Table 2.4: Comparative nutritional composition of false yam seed meal (g/kg, as fed

basis)<sup>1</sup>

		False yam seed products	
Chemical component	RSM	SSM	BSM
Dry matter	884.6	869.0	872.8
GE (kcal/kg)	3,830	3,900	3,850
Crude protein	126.9	97.5	123.8
Ether extract	10.1	4.1	6.7
NDF	124.8	63.7	298.5
Starch	456.1	384.7	385.3
Ash	24.0	4.8	18.7
Calcium	1.0	1.0	1.3
Phosphorus	1.5	0.4	1.6
Essential Amino Acids			
Arginine	11.3	8.8	11.2
Histidine	3.1	2.8	3.3
Isoleucine	7.7	6.9	7.7
Leucine	8.8	7.7	9.3
Lysine	4.1	3.8	4.6
Methionine	1.7	0.9	1.4
Phenylalanine	5.7	4.9	6.0
Threonine	4.8	4.1	5.2
Tryptophan	1.5	1.4	2.1
Valine	5.9	5.2	6.2
Non-essential amino acids			
Alanine	6.2	5.2	6.5
Aspartic acid	13.1	10.6	13.0
Cystine	0.7	0.7	0.7
Glutamic acid	21.6	15.2	18.6
Glycine	5.0	4.3	5.4
Proline	4.9	4.4	5.3
Serine	5.6	5.2	6.4
Tyrosine	4.4	3.4	4.5



<sup>1</sup>Values presented are from 1 replicate analysis of amino acids and means of duplicate analyses for the other chemical components. <sup>2</sup>RSM, SSM and BSM refer to the raw, soaked and boiled seeds meals, respectively. Adapted from Dei *et al.* (2014)

The data show that false yam tuber samples generally contain low protein (4.4-10.4%) and fat (0.7-1.74) contents (Table 2.2). However, Osei *et al.* (2013a) reported a much higher crude protein value of 16.45% which is more than those reported by NRI, (1987) and Dei *et al.* (2011a). Some researchers have reported varying values for NDF content in the false yam tuber, which ranges as low as 15.7% (Dei et al., 2010) and as high as 43.12% (Dei *et al.*, 2011a). The wide range of the values recorded may be due to age and variety of the tuber used as well as analytical method used. These indicate that, the more mature the plant the higher the fibre content. In terms of ash content the values reported for the tuber was 1.7% (Osei *et al.*, 2013a) and 2.8% (NRI, 1987; Dei *et al.*, 2011a). The observed differences in the chemical composition of the false yam tuber samples may be due to differences in the method of processing and analytical procedures. Okai *et al.* (1995) indicated that variation in the proximate values for a feedstuff may be due to the analytical methods and techniques used. The tuber samples (Table 2.2) are quite high in soluble carbohydrates 51.3 to 74.8%. Thus the tuber samples have high gross energy value (Table 2.2). However, the ME values for poultry have not been reported.

The false yam seed samples have wide range of crude protein (7.4-14.0%) and carbohydrate (65.5-80.7%) contents (Table 2.2). The fat content is less variable and low (0.1-0.5%). However, the amount of fibre found in the seed ranges between 0.1% (Kay, 1973) and 9.9% (Dei *et al.*, 2012d). The ash content reported by Kay (1973) was 0.5% and the value reported by Dei *et al.* (2012d) was 6.3%. It can be seen that soaking the seeds for 15 days tended to improve upon the crude protein, fat and energy contents. This suggests that extended soaking duration beyond 9 days had no adverse effect on the nutritional component of the seeds.

Generally, the proximate compositions of false yam tuber and seed appear to be similar. However, one can deduce that the false yam tuber contains more fibre than the seed.



The high tuberous root yield of the tuber (Fay, 1993) can serve as alternative feedstuff all year round, unlike the seed which is seasonally produced. Thus the high soluble carbohydrate contents of the false yam tuber and seed make them a promising energy source for feeding monogastrics (poultry and pigs). Therefore, these products can be a good replacement for maize in poultry diets. Maize is a popular cereal grain for feeding monogastric animals on the basis of its high-energy value in diets (Scott *et al.*, 1947). According to Larbier and Leclercq (1994), maize grain contains 87% dry matter, 2.1% ash, 10.2% crude protein, 4.8% fat, 79.5% nitrogen free extract and 16.4 ME MJ/kg DM.

Comparatively, the nutrient contents of false yam tuber and seeds are generally similar to those of other root and tubers such as sweet potato and water yam, but have higher gross energy than wild cocoyam and cassava (Table 2.2). False yam tuber and seeds also contain higher fibre content, but similar ash content which compare favourably with those of wild cocoyam. The level of essential amino acids (Table 2.3), which are needed by the body of animals, but cannot be synthesized by the animal, namely methionine, threonine, arginine, isoleucine, leucine, valine, phenylalanine and glycine are relatively low in the tuber and the processed seed meal, but slightly higher in raw seed meal. The low concentration of amino acids in the false yam tuber and seeds as compared to root and tubers such as cassava (Table 2.3) is an indication that more dietary supplementation with high protein feeds will be required. The comparative nutritional composition of false yam seed products (Table 2.4) also indicate clearly the need for high protein feed supplementation. According to Swaminathan and Kochar (1989), the low protein content of any feedstuff limits it nutritive value as a source of feed for livestock (particularly poultry).



Even though false yam tuber and seeds are low in protein (Table 2.2) it can be overcome by blending it with high protein sources such as fishmeal and soybean meal in the diets for poultry.

# 2.3.0 Limitations to the use of false yam tuber and seeds and other alternative feedstuffs in poultry diets

Despite the potential of false yam as a food crop, it has a number of limitations for its cultivation; these include drudgery in harvesting, toxicity and presence of anti-nutritional factors.

# 2.3.1 Drudgery in harvesting

According to NRI (1987), the tubers are harvested by hand. They can penetrate to about 25-30 cm below the surface which makes them difficult to dig out. Thus the plant was named "abor ntupe" or 'break hoe' in Northern Ashanti. Further, it is known for destroying shovels and plows. Therefore it is considered to be a troublesome plant in the savannah lands found along roadsides (Fay, 1987).

#### 2.3.2 Toxicity



The main toxic principle which occurs in the false yam plant concerning the seeds and tuber is a chemical compound called gum resin, which has been identified as terpenes (Vanhaelen *et al.*, 1986). The quantity of gum resins in the tuber ranges from 0.9 to 2.8% (NRI, 1987) and 3.75% (Dei *et al.*, 2011a). The concentration may vary greatly between species, processing and also with environmental conditions even though not yet determined. False yam (*Icacina tricachantha*) tuber flour (Umoh, 2013) and (*Icacina manni*) (Umoren *et al.*, 2008) tuber meal have been found to contain hydrocyanic acid, phytic acid, oxalic acid and alkaloids. The fleshly harvested tubers and seeds of *Icacina senegalensis* also contain some toxic complexes-cyanogenic glycosides (Dalziel, 1984).

Okoronkwo *et al.* (2014) reported some types of anti-nutritional factors in false yam seeds as well as the quantitative evaluation of the available anti-nutritional factors as presented in Table 2.5 below;

Sample (%)	Utu (Icacina senegalensis seed)
Oxalate	$2.02\pm0.015$
Tannin	$5.84\pm0.012$
Saponin	$2.59\pm0.012$
Phytate	$2.17\pm0.012$
Alkaloid	$3.92\pm0.025$
HCN	$3.39\pm0.474$
Flavonoid	$2.82\pm0.012$

Table 2.5: Phytochemical composition of Utu (Icacina senegalensis) seeds

Source: Okoronkwo et al. (2014)

## 2.4.0 Anti-nutrients in alternative feedstuffs for monogastric animals

Plants have co-evolved with predator populations of bacteria, insects, fungi and grazing animals, and have developed defense mechanisms for their survival. Many plants also produce chemicals which are not directly involved in the process of plant growth (secondary compounds), but act as deterrents to insects and fungal attack. These compounds also affect animals and the nutritive value of plant materials in which they are found. Anti-nutritional factors may be defined as those substances generated in natural feed stuffs by normal metabolism within plants and by different mechanisms like inactivation of some nutrients, diminution of the digestive process or metabolic utilization of feed which exert effects contrary to optimum nutrition. The ANFs may be regarded as a class of compounds, which are generally not lethal but reduce animal productivity and may cause toxicity during periods of scarcity or confinement when the feed rich in these substances is consumed by animals in large quantities (Cheeke and Shull, 1985).



#### 2.4.1 Tannins

Tannins (Figure 2.1) are phenolic plant compounds that reduce the activities of digestive enzymes mainly due to their binding interactions with proteins (Akande *et al.*, 2010; Sadeghi *et al.*, 2009; Brown, 2001; Miller, 2001). Tannins are astringent, bitter-tasting plant polyphenols that bind to and precipitate proteins (Haslam, 1989). The precipitation of protein by tannins depends on pH, ionic strength, and molecular size of tannins (Akande *et al.*, 2010). Tannins are also known to form complexes with carbohydrates, particularly starch (Mahmood, 2006). They can combine with proteins, including enzymes in the digestive tract thereby negatively affecting the digestibility of proteins, and carbohydrates (Teguia and Beynen, 2005; Van Soest, 1994). They also have covalent bonding capability (Akande *et al.*, 2010). The astringency associated with tannins decreases feed intake and negatively affect digestibility (Brown, 2001; Reed, 1995; Van Soest, 1994). They reduce feed intake by decreasing palatability and by negatively affecting digestion (Hagerman, 2002). They also decrease organic matter, and fibre digestion (Brown, 2001). They depend on the molecular size especially in condensed tannins and on the animal's tolerance, which in turn is dependent on characteristics such as type of digestive tract, feeding behaviour, body size, and detoxification mechanisms (Van Soest, 1994).



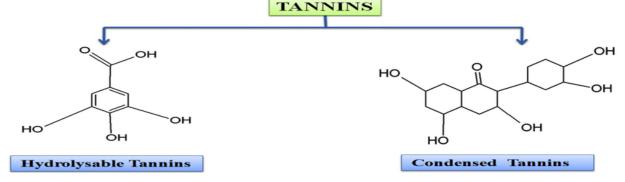


Figure 2.1: Structures of major classes of tannins. Adapted from Debosree, (2015)

Non-ruminants fed diets with tannin level above 5% have been shown to experience depressed growth rates; low protein utilization; damage to the mucosa lining of the digestive tract; alteration in excretion of certain cations; increased excretion of proteins, and essential amino acids (Brown, 2001). In chickens, tannin-protein or tannin-carbohydrate combination affects growth rate, feed efficiency, and availability of metabolisable energy (Teguia and Beynen, 2005; Van Soest, 1994). Levels of 0.5-2.0% tannins in poultry diets cause growth depression, and decreased egg production; levels of 3.0-7.0% tannins cause death (Brown, 2001). However, a study showed that feeding tannin extracts of Quebracho (used in leather industry) reduced the parasite burden from infected sheep (Max *et al.*, 2003). Giving tannin extracts from locally available plant materials to small ruminants had the potential to provide an alternative to using chemically based anthelmintics in controlling parasitic worms (Max *et al.*, 2003).

# 2.4.2 Saponins

Saponins (Figure 2.2) are steroid or triterpenoid glycosides that belong to the class known as terpenoids (Van Soest, 1994). They are low molecular weight secondary plant metabolites containing either a triterpenoid or steroidal aglycone and one or more sugar chains, and are capable of forming stable foam in aqueous solutions (Alexander *et al.*, 2009). Structurally, they possess

one or more hydrophilic glycoside moieties combined with a lipophilic triterpene derivative.



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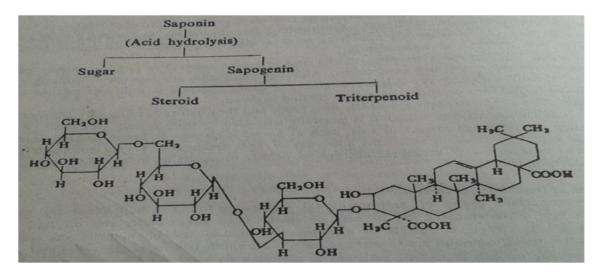


Figure 2.2: Chemical structure of the saponins (Banerjee, 1988)

Saponins are characterized by properties such as foaming, toxicity to fish, insects and fungi (Laitha *et al.*, 1987). They are widely distributed in legumes and can cause haemolysis of red blood cells (Van Soest, 1994; Laitha *et al.*, 1987). Large amounts of saponins are found in rapeseed, and high levels of the seed in diets cause growth depression of chicks (Musharaf, 1989). They are also found in soyseed, quinoa, sugar beet, and beech mast (Alexander *et al.*, 2009). Saponins significantly affect growth, feed intake, and reproduction in animals (Francis *et al.*, 2002). They are antioxidants; impair the digestion of protein, and the uptake of vitamins and minerals in the gut; cause hypoglycaemia, and act as antifungal and antiviral agents (Francis *et al.*, 2002). Although saponins cause anti-nutritional and toxic effects, they also have beneficial attributes. They are used as detergents, pesticides, molluscicides, foaming, and surface active agents (Alexander *et al.*, 2009).

Saponins have the potential as dietary additive with optimum level in the diet favouring higher growth rate, and better feed efficiency (Francis *et al.*, 2002). With dietary additive attributes of saponins, scientists are putting their efforts to make saponins as an important additive rather than being an anti-nutrient for better economic poultry diet formulation (Miah *et al.*, 2004).



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Inclusion of 75 mg/kg of exogenous saponins as feed additive in broiler diets was shown to improve growth, and carcass quality of broilers (Miah *et al.*, 2004).

# 2.4.3 Phytate

Phytate (Figure 2.3), also known as phytic acids or inositol hexaphosphates are phosphorus compounds found primarily in cereal legumes and nuts (Van Soest, 1994). Phytates are the primary storage forms of phosphates and inositols in plant seeds (Kumar *et al.*, 2010). Phytic acid accounts for 50-80% of the total phosphorus in different cereals (Coulibaly *et al.*, 2011). Phytic acid is present in large quantities within many of the major legumes and oilseeds including soyseed, rapeseed, and cotton seed (Coulibaly *et al.*, 2011; Akande *et al.*, 2010). Various polyphosphorylated inositols exist in nature and this has resulted into confusion in terminology about the nomenclature of these compounds with terms such as phytin, phytates, and phytic acid (Maga, 1982). Phytins are mixed Ca and Mg salts of myo-inositol 1,2,3,4,5,6-hexakis (dihydrogen phosphate) and are also known as phytic acid (Lolas and Markakis, 1975). Phytin is a calcium-magnesium salt of phytic acid while phytate would mean the mono to dodeca anion of phytic acid (Maga, 1982).



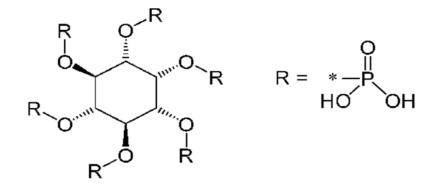


Figure 2.3: Chemical structure of phytic acid (Kumar et al., 2012)

Phytic acid binds trace elements and macro-elements such as iron, calcium, magnesium, and zinc in the gastrointestinal tract to form insoluble complexes thus interfering with their absorption (Akande *et al.*, 2010; Umaru *et al.*, 2007; Ramakrishna, 2006; Van Soest, 1994; Maga, 1982). Phytic acid affects protein and lipid utilization (Kumar *et al.*, 2010) because it inhibits enzymes needed to digest food such as pepsin, amylases, and trypsin (Coulibaly *et al.*, 2011; Ramakrishna, 2006). The major part of the phosphorus contained within phytic acid is largely unavailable to animals due to the absence of the enzyme phytase within the digestive tract of monogastric animals (Akande *et al.*, 2010). Total inositol phosphate contents of dry seeds significantly differ among the varieties and range from 2.9 - 5.0 g/kg (Muzquiz, 1999). Phytate is not known to have any negative effects on mineral absorption in ruminants, but some accounts in literature suggest that it may have the effect in simple gutted animals (Van Soest, 1994). Phytate complexes are poorly available to monogastric animals, and possess several anti-metabolic properties (Makarska *et al.*, 2008).

#### 2.4.4 Non-starch polysaccharides

Non-starch polysaccharides (NSP) (Figure 2.4) cover a large variety of polysaccharide molecules excluding starch (Choct, 1997). Non-starch polysaccharides are categorized into three main groups namely cellulose, non-cellulosic polymers, and pectic polysaccharides (Choct, 1997). The major detrimental effects of NSP are linked with the viscous nature of the polysaccharides, morphological effects on the digestive tract, and interaction with micro flora of the gut (Choct, 1997). The method in which NSP impact their effects includes altering time, modification of intestinal mucosa, and changes in hormonal regulations to a varied rate of nutrient absorption (Irish and Balnave, 1993). However, there is still no direct evidence that the increase in digesta viscosity



is responsible for the inhibition of digesta associated with poor broiler growth depressions, and feed efficiency (Irish and Balnave, 1993). The NSPs may inhibit digestion by directly complexing with digestive enzymes (Annison, 1993).

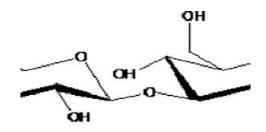


Figure 2.4: Cell wall polysaccharides (Annison, 1993)

Studies on the role of wheat NSPs in broiler nutrition proposed that soluble non-starch polysaccharide cell-wall components of wheat were responsible for the low apparent metabolisable energy (Annison, 1993). The apparent metabolisable energy depression is a result of inhibition of starch, lipid, and protein digestion in the fore gut (Annison, 1993). When NSP are fed to broiler chickens either as dietary supplements or as endogenous components of dietary ingredients, digesta viscosity can be increased thereby depressing bird's performance (Choct, 1997).

Attention has been drawn to studies in which poor growth was observed when broilers were fed

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diets containing soyseed meal as the sole dietary protein concentrate. The growth of broilers was improved by replacing 25% of soyseed meal with sunflower or rapeseed meal or cottonseed meal suggesting that the feeding quality of the soyseed meal may be questionable (Choct, 1997). The poor performance of broilers fed the soyseed diets did not appear to be related to inadequate treatment or to the NSP content or composition of soyseed meal. The measurement of free sugars in the supernatant of the digesta in the ileum indicated that starchyose derived from oligosaccharides of the soyseed meals appeared to exert anti-nutritional effect when soyseed meal was present at high concentrations as the sole protein concentrate in broiler diets (Irish and Balnave, 1993). The anti-nutritive activity of soluble NSP with well-defined chemical structure can be eliminated effectively by supplementation of the feed with xylanases, which cause a partial de-polymerization on NSP to smaller polymers so that their ability to form highly viscous digesta is greatly reduced (Choct, 1997).

### 2.4.5 Oxalates

Oxalate or ethanedioate is the dianion with formula  $C_2O_{42}$ – (Figure 2.5). Many metal ions form insoluble precipitates with oxalate, for instance, calcium oxalate. Oxalates have an effect on metabolism of calcium and magnesium and combine with proteins to form complexes that inhibit peptic digestion (Akande *et al.*, 2010). Cooking feedstuffs containing oxalates reduces oxalates (Akinmutimi, 2006).

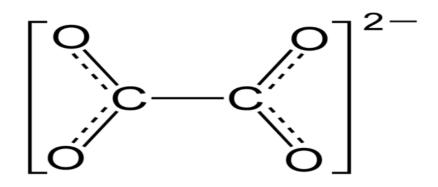


Figure 2.5: Chemical structure of oxalate ion (Wikipedia File)

#### 2.4.6 Protease inhibitors

Protease inhibitors (Figure 2.6) are sulphur-containing enzymes that interfere with normal protein digestion in the intestinal tract by inactivating proteolytic enzymes secreted by the pancreas

(Sadeghi *et al.*, 2009; McDonald *et al.*, 1987). Protease inhibitors are widely found in the seeds of most cultivated legumes, and are the most encountered class of anti-nutritive compounds of plant origin (Akande *et al.*, 2010). Protease inhibitors common in JS are trypsin, chymotrypsin, elastase, trypsin/chymotrypsin, elastase/chymotrypsin, and trypsin/chymotrypsin inhibitors (Sadeghi *et al.*, 2009). Trypsin and chymotrypsin inhibitors are the main protease inhibitors found in raw legume seeds (Akande *et al.*, 2010). Cooking has been reported to completely eliminate trypsin inhibitors from JS (Akinmutimi, 2006).

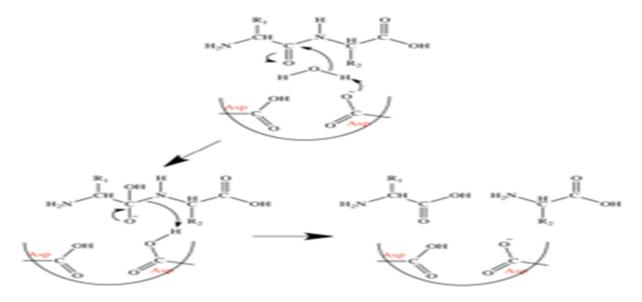


Figure 2.6: structure of protease inhibitors (Wikipedia File)

# 2.4.7 Alkaloids

Alkaloids (Figure 2.7) are heterocyclic nitrogenous compounds responsible for inhibition of digestion and poor feed utilization in livestock (Van Soest, 1994). The high content of alkaloids in diets with traditional seeds of high-alkaloid varieties has been associated to reduced efficiency of many body systems such as nervous, circulatory, digestive, respiratory, immune, and reproductive

systems (Zdunczyk *et al.*, 1997). The study on fodder lupin varieties containing less than 1 g/kg of alkaloids was reported to limit the anti-nutritional effect of alkaloids on palatability, consumption, and feed utilization (Zdunczyk *et al.*, 1997).

Alkaloids

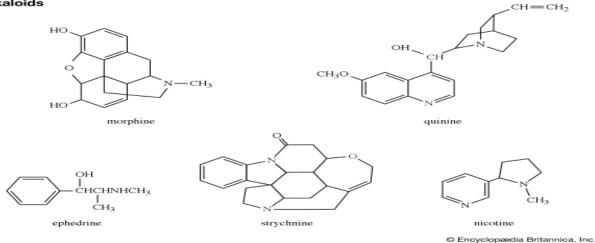


Figure 2.7: Structure of alkaloids (Wikipedia File)

## 2.5.0 Effects of anti-nutritional factors in monogastric animals

The effects of anti-nutritional factors in feedstuffs that can limit maximum utilization by animals include; reduction in feed intake, growth and digestibility as well as reduction in mineral absorption. Some of them can cause reduction in blood cells or death.



# 2.5.1 Reduction in feed intake

The toxic compound, canavanine in jack bean has been found to reduce feed intake when included 300 g/kg in the diet of non-ruminant (Tschiersch, 1962). Canavanine is believed to exert its toxic influence by virtue of its structural similarity with the nutritionally indispensable amino acid known as arginine and this may antagonize arginine and interfere with Ribonucleic Acid (RNA) metabolism (Rosenthal, 1982). Carew *et al.* (2003) found a significant reduction in feed intake

when raw mucuna beans were included in broiler diets. The raw mucuna seed meal at 6% inclusion level reduced feed intake in hen diets, due to the presence of some anti-nutritional factors like tannins and trypsin inhibitors in the diets (Tuleun *et al.*, 2009). High dietary saponin content (9 g/kg) reduced feed intake of chicks (Jenkins and Atwal, 1994) caused by its bitter taste (Cheeke, 1971).

#### 2.5.2 Reduction in growth

According to Tamir and Alumot (1970), tannins found in Carob (*Ceratonia siliqua* L.) were shown to depress growth in rats when added to a commercial diet. Similar observation was also reported in sorghum grains and faba bean containing tannins thereby causing toxicity to chicks (Iji *et al.*, 2004) with the levels from 5 to 20 g/kg in the diet. Lienier (1996) reported that 50% of growth inhibition observed in rats when fed with raw soya bean was attributed to lectin, whilst the rest was due to trypsin inhibitor (40%) and other anti-nutritional factors (10%). The effects of saponins on chicks have been reported to reduce growth and feed efficiency (Jenkins and Atwal, 1994). Goitrogenic substances which cause enlargement of the thyroid gland have been found in legumes such as soybean and groundnut which reduces growth. Because it inhibits the synthesis and secretion of thyroid hormones, since thyroid hormones play an important part in the control of body metabolism, their deficiency results in reduced growth (Olomu, 1995). Monogastric animals such as mice, pigs and poultry fed high levels of cocoa pod husk meal have poor growth (Owusu-Domfeh, 1972; Clarke and Clarke, 1979; Peckham, 1984) and may be due to theobromine (Atuahene *et al.*, 1998).

The ingestion of fresh cassava or processed cassava based diets has been reported to reduce growth rate in rats and pigs (Tewe *et al.*, 1977; Tewe and Maner, 1981; Tewe, 1983). The mechanism of



HCN acting on the animals' growth, according to Tewe *et al.*(1984), is that cyanide inhibits the intra-thyroidal uptake of iodine and thereby causing an increase in the secretion of Thyroid Stimulating Hormone (TSH) and a reduction in thyroxin level which is necessary for growth (Tewe, 1991). Terpenes can also impair the availability of nutrients (McDonalds *et al.*, 2002) and reduce growth in animals.

#### 2.5.3 Reduction in digestibility

Digestibility is the amount of nutrient in a feed that is digested and absorbed. Thus it is available for metabolism by the animal (Cheeke, 2005). The effect of saponins on chicks has been reported to interfere with the absorption of dietary lipids, cholesterol, bile acids and hence reduce digestibility (Jenkins and Atwal, 1994). Animal diets containing legume seeds with lectin constituents, for example, have been shown to impair the absorption of various nutrients such as amino acids and carbohydrates (Pusztai *et al.*, 1981). This has been attributed primarily to the binding of the lectins to the surface of the intestinal epithelial cells causing among other alterations, a non-specific interference with the final digestion and absorption of nutrients (Thompson, 1993). Nutrient digestibility was decreased in animals such as pig, chicken, rat and mouse fed raw *Plukenetia conophora* seed meal and the decrease of protein digestibility was more pronounced (Chunmei *et al.*, 2010; Gilani *et al.*, 2012). Also, Li *et al.* (1996) reported that protein digestibility was decreased by 20 to 40% in animals fed with diets containing high levels of trypsin inhibitor in the raw soybean. The findings of Qin (2003) indicated that anti-nutritional factors such as lectin present in plant-based feed ingredients including soybean (Lajolo and Genovese, 2002) could combine with a specific receptor (polyose) of the epithelial cell surface in the small intestine wall,



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destroying the brush border mucosa structure of the small intestine, interfering with the function of many enzymes in the brush border mucosa.

The phytic content of Mucuna utilis leaf meal could also present possible depression in performance of the chicks offered higher levels of inclusion. Phytic acid can bind with protein to form phytate-protein complexes (Saio et al., 1967) in feedstuff such as cassava (Singh and Krikorian, 1982). This complex can adversely affect the digestibility of protein (Reddy et al., 1982) by inhibiting a number of digestive enzymes in the gastro-intestinal tract such as pepsin (Camus and Laporte, 1976), trypsin (Cadwell, 1992) and chymotrypsin (Singh and Krikorian, 1982) thereby reducing the availability of amino acids. A known anti-nutritive component of poultry feedstuffs, soluble non-starch polysaccharide (NSP), stands out as a major determinant of the available energy and other nutrients for poultry (Hughes et al., 2001). One of the modes of action of soluble NSP is to form a viscous gel in the gut which in turn affects the rates of digestion and absorption of nutrients (Choct, 1999). Tannin in faba beans adversely affects poultry digestibility (Aramananious et al., 1973) due to cross linking between them and proteins and also glucoproteins (Goldstein and Swain, 1965) by disrupting the digestive enzymes (Huisman and Van der Poel, 1989). Saponins in legume have been reported to reduce protein digestibility (Shimoyamada et al., 1998) due to the formation of saponins-protein complexes (Potter et al., 1993). Complexed-tannins found in salseed Shorea robusta meal have been shown to depress nutrient digestibility in poultry (Mahmood et al., 2006). Lectins are present in most legumes (Liener, 1989), they interfere with nutrient digestion and absorption and increase wasteful protein synthesis, resulting in reduced efficiency of nutrient utilization.



The decrease in protein and fat digestibility of diets containing soaked false yam tuber meal in layer chickens have been attributed to anti-nutritional factors such as gum resins in the soaked false yam tuber meal (Mohammed and Dei, 2013).

#### **2.5.4 Reduction in mineral absorption**

Phytate is a cyclic compound that chelates with mineral ions (magnesium, calcium, iron, zinc, and molybdenum). It has been found in cassava, with approximately 624 mg/100 g in the roots (Marfo *et al.*, 1990). This forms compounds such as mineral-phytic acid complexes which lead to reduce mineral bioavailability (Khare, 2000) or not readily absorbed in the intestine (Liener, 1989), thereby reducing animal performance. Barbara (2009) noted that all legumes contain phytate (also known as phytic acid) to some extent, but the soybean is particularly rich in this anti-nutrient. Olomu (1995) reported that pigeon pea contains about 0.38% oxalic acid this binds with calcium and forms calcium oxalate which is insoluble and can adversely affect the absorption and utilization of calcium in the animal body.

# 2.5.5 Reduction in blood cells



Pigeon pea seed is reported to contain haemagglutinin (Amaefule, 2002) which is known to affect the blood formation in animals by reduction of packed cell volume (Akinmutimi, 2004). There have been reductions in haemoglobin and red blood cell counts in birds fed tiger nut diets beyond 50% dietary levels. This was particularly due to phytate present in the feed that chelates divalent metals utilization in monogastric animal metabolism, and may increase the values of white blood cells in diets containing tiger nut (Akinwutimi *et al.*, 2004; Obidinma, 2009). High cyanide levels induce haematological changes of growing pigs fed sun-dried cassava based rations (Tewe, 2006).

#### 2.5.6 Death

According to Kumar and D'Mello (1991), *Napoleona imperialis* seed meal contains oxalate substances which decrease the availability of dietary essential minerals (calcium) at high concentration and causes death in animals due to its corrosive effects. Raw sorghum contains tannins (Douglas *et al.*, 1993) and can cause death to poultry with inclusion level of about 30-70 g/kg in their diets (Salunke and Chavan, 1990; Farrel *et al.*, 1999; Iji *et al.*, 2004). Feedstuffs such as cassava contain hydrocyanic acid which is highly poisonous because it rapidly inactivates cellular respiration thereby causing death to poultry (Salkowski and Penny, 1994).

#### 2.6.0 Methods of reducing or removing adverse effects of anti-nutritional factors

Information on the effect of different processing techniques on the anti-nutritional factors in animal feeds will attract interests by animal nutritionists on the exploitation of these techniques so that the nutritive values of feeds could be efficiently maximized. Although data on the necessary optimal processing techniques, if any, is scanty, this should be established to ensure optimal utilization of animal feeds. Processing techniques that have been used to reduce the negative effects of anti-nutritional factors in plant materials include peeling or dehulling, heat treatment, soaking, boiling, fermentation, plant breeding, autoclaving, urea and alkaline treatments.



# 2.6.1 Peeling or Dehulling

This method is the physical removal of the peels, shells and hulls from feedstuffs. These compounds are mostly confined to the seed coat such as mucuna beans and removal therefore, can be achieved by dehulling (Van der Poel *et al.*, 1991) and has also reduced tannins activity in faba

bean by improving the nutritional value of faba beans. Catalano *et al.* (1977) reported that peeling of sweet potatoes tuber removes most of the toxic components and fibre content.

### 2.6.2 Heat treatment

Heating is normally achieved by dry heating or roasting and sometimes boiling. Anti-nutritional factors in feedstuffs are mostly reduced when subjected to boiling. Boiling or roasting have been effective in removal of cynoglucoside in sweet potato, yellow yam and cocoyam and phytic acid in yellow yam and cocoyam (Omoruyi et al., 2007). Khattab and Arntfield (2009) stated that boiling, roasting, microwave cooking and autoclaving brought a total removal of trypsin inhibitor of cowpea, pea, and kidney bean. The heat applied in processing is identified as the single most important factor that affects soybean meal protein quality. These treatments were primarily applied in order to reduce the contents of anti-nutritional factors and their harmful effects (Bengala-Freire et al., 1991). According to Rasha et al. (2011), the ordinary boiling of soaked seeds for different time periods brought about a significant decrease in trypsin inhibitory activity in soya bean as compared with raw seeds. The most effective methods for inactivation of trypsin inhibitors activities are boiling (90 min) and autoclaving for 10 min (Rasha et al., 2011). Rehman and Salariya (2005), reported that cooking reduced phytic acid contents by 21 and 24% in red and white kidney beans respectively. Cooking velvet beans removed the negative effect of trypsin inhibitor by 100% (Iyayi and Taiwo, 2003). Dry heating treatment can reduce the non-protein factor, L-Dopa (L-3, 4-dihydroxyphenylalanine) in mucuna bean (Siddhuraju et al., 1996).



#### 2.6.3 Soaking

According to Cooke and Maduagwu (1985), there is a significant reduction in total cyanide in cassava when the soaking water is routinely changed over a period of days. Shimelis and Rakshit (2007) found that trypsin inhibitors activity was reduced in kidney bean to 9-18%, by hydration. El-Hady and Habiba (2003) observed a 36% reduction in phytic acid in kidney beans after an overnight soaking in water at room temperature. Some polyphenols and oxalates that inhibit iron and calcium absorption respectively may also be lost by soaking (Erdman and Pneros-Schneier, 1994). Chlorogenic acid is reported to be readily removed from sunflower seeds using aqueous extraction methods (Dominguez *et al.*, 1993).

Soaking prior to cooking has been found to improve the extraction efficiency of protease and alpha-amylase inhibitors (Moneam, 1990). This removes about 20% of free cyanide in fresh cassava root chips. Cracking the seed into two or four pieces before soaking in water and cooking was more effective and improved the performance of broilers (Emenalom *et al.*, 2002). Nergiz and Gokgoz (2007) also reported 57-58% reduction in phytic acid after cooking beans that had been soaked 12 hours prior to cooking. Okai *et al.* (1995) found a significant reduction in tannin content of shea nut meal (40-70%) when processed either in soaked or boiled water. This improved feed intake and protein digestibility in weanling rats. About 97% hydrocyanic acid (HCN) in cassava is removed by prolonged soaking (six days) in water (Bourdoux *et al.*, 1983). Soaking of mucuna dehulled beans for 24 hours reduced the non-protein factor, L-Dopa (L-3,4-dihydroxyphenylalanine) by 30% while in the whole bean by only 6% (Nyirenda *et al.*, 2002).



#### 2.6.4 Fermentation

Fermentation is a process of using microbes (bacteria, mould and yeast) to metabolize raw material forming new products with higher value than the initial material (Frazier and Washoff, 1988). One of the several functions of fermentation according to Steinkraus (1995) is the enrichment of diet and the removal of toxic component. Fermentation process can produce organic acids such as lactic acid (Delaude, 1974) which has been found to break down saponins and tannins (Yosioka *et al.*, 1966) that detoxify these compounds (Reddy and Pierson, 1994). The fermentation of cassava enables softening of the roots which has the combined effect of enabling linamarin and linamarase to mix and also to enable leaching of the cyanogens (Westby and Choo, 1994). Fermentation has been the most effective processing method that drastically reduced phytic acid and trypsin inhibitor activity in oilseeds (Fagbemi *et al.*, 2005). According to Annongu *et al.* (1996), tannin concentration in shea nut meal was reduced when wet incubation was applied.

# 2.6.5 Breeding



Introduction of new nutritionally superior cultivars has enhanced the attractiveness of the faba bean crop as an animal feedstuff by not only removing anti-nutritional properties but also by lowering crude fibre and lignin contents (Bjerg *et al.*, 1984; Garrido *et al.*, 1989). Breeding zero-tannin cultivars (NIAB, 1992) permanently eliminated tannins from the seed of faba bean (*Vicia faba*). The International Institute of Tropical Agriculture (IITA) has developed a high yielding and low cyanide cultivar (T. M. S 4(2) 1425) of cassava. These varieties seemed to be well adapted to different ecological conditions, were high yielding, had low cyanide levels and were poundable after boiling (Silvestre, 1989). The amount of full fat soybeans used has been increasing in the livestock industry due to development of new varieties with limited number or levels of anti-nutritional factors (Gu *et al.*, 2010). Perić *et al.* (2011) indicated that soybean breeding programme

at the Maize Research Institute was aimed at developing the cultivars with reduced trypsin inhibitors content. As a result, two Kunitz-free (KTI free) varieties Lana and Laura were released. Trypsin inhibitors content in new cultivars was about 50% reduced as compared with the conventional cultivars (standard grain type). It has been observed that the effect of raw Kunitzfree soybean in diet of young chicks and pigs was beneficial in terms of better growth performance compared with conventional cultivars (Palacious *et al.*, 2004). Tagliapietra *et al.* (2007) suggested that raw Kunitz-free soybean can be included in the diets of finishing pigs (80 to 170 kg live weight) up to 10% of the complete feed without negative effects on growth performance and health status.

#### 2.6.6 Autoclaving

Autoclaving entails cooking under pressure. The time of cooking is shortened by this method (Akande *et al.*, 2010). Udedibie *et al.* (1988) reported that when jack beans were autoclaved for 30 minutes at 125°C and 151b pressure, thermo-labile inhibitory substances such as cyanogenic glycosides, saponins, terpenoids and alkaloids could not be detected after autoclaving. Kessler *et al.* (1990) stated that there was little nutritional advantage in autoclaving for more than half an hour. They reported that autoclaving of Jack beans was asatisfactory technique for ensuring survival of birds receiving jack beans diets.



Autoclaving of jackbeans for 10 minutes ameliorated the necrotizing effect but did not improve upon its feeding quality. However, combined heat treatment and water washing rendered the neem kernel as good protein supplement compared to groundnut cake (Uko and Kamalu, 2008). Even though the processed neem kernel supported final weight of broilers similar to groundnut cake, growth rate of the experimental birds fluctuated throughout the period of feeding (Uko and Kamalu, 2008).

### 2.6.7 Urea Treatment

Urea is a very strong protein – denaturing agent and can achieve this by competing for hydrogen bonds with the peptide backbone, thereby breaking up the secondary structure of these native protein and disrupting their biologically active structures (Udedibie and Nkwocha, 1990).

Raw jackbean seeds were soaked in 3% solution of urea for 6 days at room temperature in plastic containers. During this period, strong ammonia gas odour was released from the solution. At the end of the period, the beans were rinsed with tap water and then cooked for one hour, dried in oven at 80°C and then ground. Feeding trials with the resultant jackbean meal involving young broiler chicks demonstrated that jackbeans so proccessed could be tolerated by broiler chicks of up to 25% inclusion levels in the diet (Udedibie and Nkwocha, 1990).

#### 2.6.8 Alkaline treatment



The addition of alkaline salts such as sodium bicarbonate has been shown to reduce soaking and cooking time (Singh *et al.*, 1988). Polyphenols were removed by soaking legumes in water and sodium bicarbonate, which makes the process efficient (Laurena *et al.* 1986). Omueti *et al.* (1992) reported that soaking and blanching of soya beans is an effective way of inactivating trypsin inhibitors and removing a significant proportion of polyphenols and oligosaccharies. Nelson *et al.* (1976) stated that removal of trypsin inhibitor by blanching was made more effective by addition of sodium bicarbonate. Ayanwale (1999) also reported that sodium sesquicarbonate (trona) can be used without detrimental effect on broiler performance and carcass quality.

#### 2.7.0 Effect of nutrition on haematological and blood biochemical components of poultry

Generally, both the haematological and biochemical components of the blood are influenced by the quantity and quality of feed and also the level of anti-nutritional elements or factors present in the feed (Akinmutimi, 2004). Biochemical components are sensitive to elements of toxicity in feeds. Esonu *et al.* (2006) noted that haematological parameters are good indicators of the physiological status of the animal and its changes are of value in assessing the response of animals to various physiological situations. It has been observed by many research workers (Khan and Wassilew 1987; Abdel-Hameed and Neat, 1972) that, there is a definite change in the profile of the blood cell throughout life. Available information indicates that haematological values of avian species are also significantly influenced by poultry diseases including fowl typhoid (Kokosharov and Todorova, 1987), mycoplasmolysis (Branton *et al.*, 1997), avian coccidiosis (Koinarski *et al.*, 2001) and Newcastle disease (Galindo-Muniz, 2001).

#### 2.7.1 Avian haematology

Blood can be collected from a variety of sites in avian patients. The choice of a blood collection site is influenced by the species of bird, preference of the collector, physical condition of patient and the volume of blood needed. For best results, venous blood should be collected for haematologic studies. Blood collected from capillaries (e.g. blood from clipped nails) often results in abnormal cell distributions and contains cellular artifacts such as macrophages and material not normally found in peripheral blood. Blood to be used for haematology should be collected into a collection tube containing EDTA (ethylene diamine tetra acetic acid) as anticoagulant. Other anticoagulants, such as heparin, interfere with cell staining and create excessive cell clumping, resulting in erroneous cell counts and evaluations (Campbell, 2013). Valuation of the avian haemogram involves counting the various blood cells per microliter of blood as well as cytologic



evaluation of the cells. The techniques involved in the evaluation of the avian haemogram are easily performed by in-house veterinary laboratory personnel. Because avian blood does not store well (e.g. during transport), haematologic results obtained soon after collection are preferred over those performed several hours after (Campbell, 2013). Blood volume in birds depends on the species and varies from 5ml/100g in the ring-necked pheasant to 16.3 to 20.3ml/100g in the racing pigeon. In general, birds are better able to tolerate severe blood loss than mammals, which is due to greater capacity for extracellular fluid mobilization. However, there is a marked variation among avian species in response to blood loss, which may be a reflection of differences in blood volume or extracellular fluid depots (Campbell, 2013). According to Forbes (2008) the average blood volume of most birds is approximately 10% of body weight. Ten percent of this volume of 1% of bird's body weight may be removed for testing. In comparison to mammals, avian blood cells show unique morphological characteristics e.g. erythrocyte and thrombocyte contain nucleus (Pendi, 2010). According to Forbes (2008) avian red cells are nucleated which is why manual white cell counts are typically not possible. White cells are similar to mammalian lines, except that the mammalian neutrophils are replaced with heterophils and mammalian platelets are replaced with thrombocytes. Important differentials for leukocytosis with profound heterophilia and monocytosis include chlamydophilosis, aspergilosis and tuberculosis. Mitchell and Johns (2008) reported that the interpretation of avian blood cells presents many challenges.



According to Forbes (2008) interpretations that can be made are listed in Table 2.6.

<b>Table 2.6.</b>	CBC	findings
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CBC Results	Interpretation
Leukocytosis	Infection, inflammation, necrosis, neoplasia, heavy metal toxicosis and stress (particularly in macaws, there should be no toxic changes in white cells)
Severe heterophilia	Chlamydophilosis, aspergillosis, tuberculosis (often with toxic changes in white cells)
Moderate heterophilia	Infection, cellular necrosis
Lymphocytosis	Viral infection, certain stages of chlamydophilosis
Monocytosis	Chronic infection with extensive necrosis and phagocyte activity (typically aspergillus, chlamydo philosis and tuberculosis)
Eosinophilia	Of inconsistent and unproven significance
Basophilia	Uncommon result most often associated with respiratory infections, resulting tissue damage, parasitism and some stages of chlamydophilosis
Leucopenia	Overwhelming bacterial or severe viral infection (Particularly circovirus). Leukopenia may also be associated with reduced production of cells or increased use, which is demonstrated by the presence of immature or toxic white cells.

Adapted from: Forbes (2008)

The normal PCV of birds ranges between 35 and 55%. A PCV less than 35% is indicative of anaemia and a PCV greather than 55% is suggestive of dehydration or polycythemia. An increase in red cell polychromasia is indicative of red blood cell regeneration. In normal birds, the number of polychromatic erythrocytes (or reticulocytes) found in the peripheral blood ranges between 1 and 5% of erythrocytes (Campbell, 2013).

An anaemic bird with a 5% or less degree of polychromasia (or reticulocytosis) is responding poorly to the anaemia or there has not been enough time for the bird to demonstrate a significant response. Hypochromasia can be associated with certain nutritional deficiencies in birds, especially iron deficiency. There is a wide variation in the normal leukograms among birds of the same species. In general, total leukocyte count greater than  $10,000/\mu$ l are considered suggestive of

leukocytosis in tame, adult psittacine birds. A normal thrombocyte count ranging between 20,000 and  $30,000/\mu$ l of blood or 10 to 15/1000 erythrocyte can be used as a general reference for most birds (Campbell, 2013).

### 2.7.2 Haematological indices of the blood

Blood which is a vital special circulatory tissue is composed of cells suspended in a fluid intercellular substance (plasma) with the major function of maintaining homeostasis (Isaac *et al.*, 2013). Haematological components, which consist of red blood cells, white blood cells or leucocytes, mean corpuscular volume, mean corpuscular haemoglobin and mean corpuscular haemoglobin concentration are valuable in monitoring feed toxicity especially with feed constituents that affect the blood cells (erythrocytes) serve as a carrier of haemoglobin. It is this haemoglobin that reacts with oxygen carried in the blood to form oxyhaemoglobin during respiration (Johnston and Morris, 1996; Chineke et al., 2006). According to Isaac *et al.* (2013) red blood cell count implies a reduction in the level of oxygen that would be carried to the tissues as well as the level of carbon dioxide returned to the lungs (Ugwuene, 2011; Soetan et al., 2013).



The major functions of the white blood cell and its differentials are to fight infections, defend the body by phagocytosis against invasion by foreign organisms and to produce or at least transport and distribute antibodies in immune response. Thus, animals with low white blood cells are exposed to high risk of disease infection, while those with high counts are capable of generating antibodies in the process of phagocytosis and have high degree of resistance to diseases (Soetan *et* 

*al.*, 2013) and enhance adaptability to local environmental and disease prevalent conditions (Kabir et al., 2011; Okunlola et al., 2012; Iwuji and Herbert, 2012; Isaac *et al.*, 2013).

Blood platelets are implicated in blood clotting. Low platelet concentration suggests that the process of clot-formation (blood clotting) will be prolonged resulting in excessive loss of blood in the case of injury. Packed Cell Volume (PCV) which is also known as haematocrit or erythrocyte volume fraction (EVF), is the percentage (%) of red blood cells in blood (Purves et al., 2003). According to Isaac et al. (2013) packed cell volume is involved in the transport of oxygen and absorbed nutrients. Increased packed cell volume shows a better transportation and thus results in an increased primary and secondary polycythemia. Haemoglobin is the iron-containing oxygentransport metallo-protein in the red blood cells of all vertebrates (Maton et al., 1993) with the exception of the fish family, channichthyldae (Sidell and O' Brien, 2006) as well as tissues of invertebrates. Haemoglobin has the physiological function of transporting oxygen to tissues of the animal for oxidation of ingested food so as to release energy for the other body functions as well as transport carbon dioxide out of the body of animals (Ugwuene, 2011; Omiyale et al., 2012; Soetan et al., 2013; Isaac et al., 2013). According to Peters et al., (2011), previous reports stated that packed cell volume, haemoglobin and mean corpuscular haemoglobin are major indices for evaluating circulatory erythrocytes, and are significant in the diagnosis of anaemia and also serve as useful indices of the bone marrow capacity to produce red blood cells as in mammals (Awodi et al., 2005; Chineke et al., 2006). Furthermore, Chineke et al. (2006) posited that high packed cell volume (PCV) reading indicated either an increase in number of red blood cells (RBCs) or reduction in circulating plasma volume. Mean corpuscular haemoglobin and mean corpuscular haemoglobin concentration indicate blood level conditions. A low level is an indication of anaemia (Aster, 2004).



### 2.7.3 Chicken haematology

The poultry industry in Ghana would have improved more than its present status if biochemical and haematological reference values have been established in Ghana to help in accurate diagnosis and treatment of poultry diseases. These values are needed in Ghana so as to fully understand the mechanisms of some diseases and to identify and prevent other diseases than quoting foreign reference values for interpretation of laboratory results. Published literature has confirmed that many of the reference values obtained from the developed countries differ significantly from what is obtainable in most African localities (Koram et al., 2007) thus making it necessary to establish locally relevant values. The Clinical and Laboratory Standards Institute (CLSI, 2008) and the International Federation for Clinical Chemistry (IFCC) recommend that each laboratory establishes its own reference values (Solberg, 1987).

 Table 2.7: Range of values for haematological parameters of chicken.

Haematologic Type	Units – International Standard (SI)	Normal Ranges
PCV	%	35.9 - 41.0
Hb	g/dl	11.60 - 13.68
RBC	X10 <sup>-6</sup> /ml	4.21 - 4.84
WBC	X10 <sup>-3</sup> /ml	4.07 – 4.32
MCV	Fl	81.60 - 89.10
МСН	Pg	27.20 - 28.90
MCHC	%	32.41 - 33.37



Wikivet (2013)

### 2.7.4 Nutrition and haematology

Haematology refers to the study of the numbers and morphology of the cellular elements of the blood – the red blood cells (erythrocytes), white cells (leukocytes), and platelets (thrombocytes) and the use of these results in the diagnosis and monitoring of disease (Merck Manual, 2012).

The blood transports or conveys nutrients and materials to different parts of the body. Therefore, whatever affects the blood, either drugs, pathogenic organism or nutrition will certainly affect the entire body adversely or moderately in terms of health, growth, maintenance and reproduction (Oke et al., 2007). A readily available and fast means of assessing clinical and nutritional health status of animals on feeding trials may be the use of blood analysis, because ingestion of dietary components have measurable effects on blood composition (Church et al., 1984; Maxwell et al., 1990) and may be considered as appropriate measure of long term nutritional status (Olabanji et al., 2007). According to Togun and Oseni (2005), haematological studies have been found useful for disease prognosis and for therapeutic and feed stress monitoring. Adamu et al. (2006) observed that nutrition had significant effect on haematological values like PCV, Hb and RBC. Togun et al. (2007) reported that when the haematological values fall within the normal range, it is an indication that diets did not have any adverse effect on haematological parameters during the experimental period but when the values fall below the normal range, it is an indication of anaemia. Low values for haematological parameters as reported by Bawala et al. (2007) could be due to the harmful effects of high dietary contents. Physiological and nutritional status of animals could cause differences in values observed for PCV and MCV. Immune status is a function of leucocytes, neutrophils and lymphocytes. Lymphocytes are known to play key roles in immune defense system of both man and animals (Ameen et al., 2007). When WBC (leucocytes), neutrophils and lymphocytes fall within the normal range, it indicates the feeding patterns do not affect the immune system, most immunological abnormalities observed in malnutrition are usually corrected after nutritional rehabilitation (Ameen et al., 2007). According to Adenkola et al. (2008) increase in neutophils; lymphocyte ratio is a good indicator of stress (Minka and Ayo, 2007) which could be nutritional stress. Blood cells arise in the bone marrow from stem cells and able to undergo



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processes of proliferation and differentiation in the haematopoietic microenvironment (Bianco, 2011; Mensez-Ferrer et al., 2010). Adequate haematopoisis is dependent on an intact and functional bone marrow microenvironment, which is an environment fully competent to provide the appropriate signals through the production of soluble factors and cell-cell contact interactions regulating by several mechanisms, directly or indirectly, the self-renewable, proliferation, survival, migration and differentiation of haematopoietic cells (Mensez-Ferrer et al., 2010). Feeding birds with protein deficient diets decreases the production of blood cells, leading to bone marrow hypoplasia and inducing structural alterations interfering with both innate and adaptive immunity (Xavier et al., 2007; Borelli et al, 2009; Fock et al., 2009). Furthermore, Cunha et al. (2013) reported that protein malnutrition (PM), as a result of feeding birds with protein deficient diets, results in pathological changes that are associated with leucopenia, bone marrow (BM) hypoplasia and alterations in BM microenvironment leading to haematopoietic failure, however, the mechanisms are poorly understood. The BM mesenchymal stem cells (MSCs) are cells intimately related to the formation of the BM microenvironment, and their differentiation in the adipocytes is important because adipocytes are cells that have the capability to negatively modulate haematopoiesis (Cunha et al., 2013). In a study that subjected experimental animals to proteinenergy malnutrition with a low protein diet containing 2% protein, whereas control animals were fed a diet containing 12% protein, the malnourished animals had anaemia and leucopenia as well as spleen and bone marrow hypoplasia and reduction in the expression of CD45 and CD117 positive cells from BM. The alterations found in the malnourished animals led to the conclusion that malnutrition committed MSC differentiation leading to cytokines production contributing to an impaired haematopoietic microenvironment and inducing the bone marrow failure commonly observed in protein malnutrition states. According to Wanbi et al. (2008) maintenance of



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antioxidant diet was associated with improved recovery of the bone marrow after sublethal or potentially lethal irradiation. Oral supplementation with antioxidants combinations containing Lselenomethionine (SeM), vitamin C, vitamin E succinate, alpha-lipoic acid and NAC appears to be an effective approach for radioprotection of haematopoietic cells and improvement of animal survival, and modulation of apoptosis is implicated as a mechanism for radioprotection of haematopoietic system by antioxidants. Koury *et al.* (1997) posited that folate and vitamin B (Cobalamin) are essential nutrients in diets. Deficiency of either leads to megaloblastic anaemia. In megaloblastic anaemia, the deficiency appears to predispose to other types of malignancy. Decreased incidences of premalignant and malignant matopoietic precursor cells of erythrocytes, granulocytes and platelets are destroyed by programmed cell death, changes in uterine, cervical, bronchial and colonic epithelia were found in susceptible animals who had received oral (apoptosis). This apoptosis can result directly from decreased intracellular folate in folate deficiency or indirectly folate supplementations as compared with similar animals who were not supplemented.

### 2.7.5 Effects of different diets on haematology of different species of poultry



A feeding trial was carried out by Rafiu *et al.* (2013) to evaluate the blood parameters of broiler chickens fed *Moringa oleifera* leaf meal (MOLM). MOLM was incorporated into experimental diets at varying replacement levels of 0, 5, 10 and 15% for soybean meal. The RBC, Hb, MCV, MCH and MCHC counts all showed significant differences. PCV and Hb levels decreased as MOLM inclusion increased. Rafiu *et al.* (2013) reported that MOLM could be included especially when the meat quality is targeted.

Saleh *et al.* (2013) conducted a study on the effects of aqueous extract of tamarind pulp (TP) on blood parameters of broiler chickens under a semi-arid environment. The birds were divided into four treatment groups and received 0g/l tamarind pulp, 20g/l, 30g/l or 40g/l tamarind pulp in drinking water, feed and water were supplied ad libitum and result showed a significant increase in RBC and WBC. No significant effect of TP was seen on Hb, PCV and MCHC and reported that up to 40g/l of tamarind pulp extract can be offered to broiler chicken without any adverse effect on blood constituents.

In a study conducted by Ayoola *et al.* (2010) on physiological response of broiler starter chickens to oral supplementation with *Telfaria occidentalis* leaf extract (TOLE), the birds were allotted into five treatment groups of oral supplementation. PCV, Hb, RBC and WBC counts were significantly affected by the TOLE supplementation. Ayoola *et al.* (2010) stated that the erythropoietic effect of TOLE on blood samples of broiler starter chickens is a good pointer that it can be used to replace synthetic vitamins in broiler production at starter stage.

Alu (2014) carried out a trial on the effect of replacing bone ash with eggshell meal at 0, 25, 50, 75 and 100% on blood parameters of broiler chickens. It was observed that the haematological parameters were not affected by the diet except for corpuscular haemoglobin which was best in birds fed 100% eggshell. Similarly, values obtained for MCV were improved in birds fed 100% eggshell. According to Alu (2014), since there was no deleterious and adverse effect for including eggshell meal in diets on blood parameters, farmers can use eggshell meal as a major source of dietary calcium in broilers diets.

Olerede *et al.* (2009) conducted a study on the effects of the processing methods of *Faidherbia albida* "Gao" on the haematology of broiler chickens fed at 15% inclusion level of the Fapm as partial replacement for groundnut cake meal. The birds were divided into four experimental groups



and fed with 0% Fapm diet (control), 15% raw Fapm, 15% soaked Fapm or 15% boiled Fapm. There were no significant differences in the haematological parameters. However, low values of PCV, RBC count, Hb, MCHC and MCH indicative of anaemic state were recorded in broilers on the experimental diets. Olerede *et al.* (2009) suggested that treatment of Fapm has improvement on haematology of broiler fed 15% inclusion level with boiling showing a better improvement.

Afolabi *et al.* (2010) conducted a research on haematologic parameters of the Nigerian local grower chickens fed varying dietary levels of palm kernel cake (PKC). The birds were randomly allotted to five experimental diets. Five isonitrogenous (17%CP) grower diets containing 2,813 – 3079Kcal/ME/kg diet with varying levels of 10, 15, 20 and 25% PKC inclusion were used to replace maize and soybean in diets 2, 3, 4 and 5, respectively were formulated. Diet without PKC (%PKC) was the control. PCV, Hb, RBC, Platelets, heterophils and eosinophils were similar among birds across diets whereas variations in white blood cell, lymphocytes and monocytes were significant. The values of haematological parameters obtained were within the normal range of values documented for healthy chickens. Inclusion of dietary PKC up to 25% in the diet of the Nigerian local growing chicken elicited no adverse effect on the haematology.

dietary inclusions of fermented cassava tuber wastes. Seven experimental diets were fed to the birds, diet with 0% microbially fermented cassava tuber wastes (MFCTWs), 20% microbially fermented cassava peel (MFCP), 40% MFCP, 60% MFCP, 20% Microbially Fermented Cassava Starch Residue (MFCSR), 40% MFCSR and 60% MFCSR. The results showed that the whole blood viscosities were statistically similar while plasma viscosities were influenced significantly in all the treatments. The whole blood and plasma viscosities, however, showed a gradual decline in value as the level of inclusion of the two types of fermented cassava tuber waste (FCTWs)

In another study carried out by Aro and Ojo (2013) on blood viscosity of finisher cockerel fed

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increased in the diets and concluded that cassava tuber wastes may decrease both plasma and whole blood viscosity and significantly so if plasma viscosity is taken into consideration. Aro and Ojo (2013) further reported that the lower viscosity of birds fed CTW at a higher inclusion is suggestive of a positive influence of the CTW at a higher inclusion on the mechanical and geometric properties of the red blood cells.

Similarly, Aro et al (2013) carried out a trial on the haematology of finisher cockerel fed graded levels of microbially enhanced cassava tuber diets. The birds were fed with seven different cassava tuber waste based (CTW) diets containing 0% CTW (control diet), 20% microbially fermented cassava peel (MFCP), 40% MFCP, 60% MFCP, 20% microbially fermented cassava starch peel (MFCSR), 40% MFCSR and 60% MFCSR. The cassava tuber wastes were inoculated with Lactobacilli (*L. delbruecki* and *L. coryneformis*) and one fungus (*Aspergillus fumigatus*) for their protein enrichment and fibre degradation. The results showed that the MCV was highest in birds fed 40% MFCP and 40% MFCSR diets with significant differences among diets, levels of inclusion and interactions between diets versus levels of inclusion. Birds fed MFCP had higher MCH than those fed MFCSR diets. The MFCR diets were, however, better in PCV, RBC and Hb. Aro et al (2013) reported that the use of CTW in cockerel rations would not compromise the haematological status of cockerel birds.



A feeding trial was also carried out by Nworgu *et al.* (2013) on haematological indices of laying hens fed optimal and sub-optimal rations supplemented with waterleaf (*Talinum triangulare*) tops. The birds in the six treatment groups were fed 117, 121 and 125g of feed/day without Waterleaf Tops Supplement (WLTS) and 117, 121 and 125g of feed/day with 30g of WLTS at 3 days interval respectively. The results depicted that hens fed 117, 121 and 125g of feed/day without WLTS had lower percentages of haemoglobin compared to those on WLTS based diet. The quantity of feed

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served to the hens significantly and progressively increased with Hb, PCV, WBC and RBC. The WLTS supplementation significantly improved the Hb, PCV, RBC and WBC of hens. Nworgu *et al.* (2013) posited that laying hens need to be fed 121 to 125g of feed/day with 30g of WLTS at 3 days interval for better blood formation.

In a trial conducted by Abeke *et al.* (2009) on effects of dietary levels of cooked lablab purpureus beans on haematological parameters of broiler finishers. The beans were processed by boiling in water for 30minutes at 100°C. Seven isonitrogenous diets containing 20.9% crude protein were formulated to contain lablab seed meal at 0.00, 5.0, 10.0, 15.0, 20.0, 25.0 or 30.0 percent level of inclusion. Diet 1 had no lablab and served as the control. The PCV and Hb status of the blood indicated significant decrease as the level of seed meal increased. Abeke *et al.* (2009) stated that lablab can be included up to 10% in broiler diets.

In a study conducted by Nwanbe and Elechi (2009) on haematological indices of broiler finisher fed potash boiled Bambara groundnut (*Voandzein subterranean* (L) Thour) meal as replacement for soybean meal. Potash boiled Bambara groundnut meal partly replaced soybean meal at 0%, 25%, 50%. 75% and 100% graded levels. The results of the haematological parameters indicated significant differences only in the PCV and Hb. According to Nwanbe and Elechi (2009) the results of the study showed that soybean meal can be replaced with potash boiled Bambara groundnut meal up to 100% level without any deleterious effects on blood constituents of the broiler birds.



Moreover, Kehinde *et al.* (2013) carried out a research on haematology of guinea fowl fed cassava peel based diets. The trial comprised three treatments with 0%, 10% and 20% cassava peel inclusion. Haematological parameters investigated include PCV, Hb, RBC, WBC and MCHC. The values for PCV, Hb, RBC and MCHC were significant in the treatments. Kehinde *et al.* (2013)

concluded that the results showed that guinea fowl tolerated 10% dietary inclusion of processed cassava peel based on the haematological indices.

Ugwuene and Onwudike (2010) conducted a trial on the replacement value of dietary palm kernel meal for maize on the haematology of local broiler turkeys. Six treatment diets in which palm kernel meal replaced maize at 0, 20, 40, 60, 80 and 100 percent were formulated. There were no definite trend in the haematological values for the birds with increase in the level of replacement of maize with PKM. However, in most of the haematological indices, turkeys fed diets 3 and 4 (40% and 60 % replacement) performed as well as that of diet 0% replacement. Ugwuene and Onwudike (2010) recommended that PKM can replace maize at 60 percent in the diets of turkey without adverse effects on haematology of the animals.

Washburn and Lowe (1974) conducted a trial on the effect of iron, copper deficient skimmed-milk diets on haematology of Japanese quail. The feeding of iron, copper deficient diet resulted in rapid and severe reductions in PCV, Hb and MCHC. Addition of iron and copper to the skim-milk partially alleviated its adverse effect on haematology.

Coenen (1994) conducted a study on the haematology of Japanese quail fed dietary tri-n-butylin oxide during reproduction and observed the absence of serious effect in blood parameters in both adult and developing chicks.



In a trial carried out by Elaroussi *et al.* (2007) on the effects of dietary vitamin E on haematological indices of Japanese quails. The birds were fed on starter and layer diets containing, 0, 1, 5 or 10 times the NRC recommended supplements for vitamin E. It was observed that as the level of vitamin E increased the percentage of erythrocytes haemolysed.

Sharifi *et al.* (2011) carried out an experiment on the effect of dietary protein levels and symbiotic on blood characteristics of Japanese quails (Coturnix coturnix Japonica). The treatment consisted

of combination of 3 levels of crude protein, sufficient protein diet (24%, high CP) and low protein diet (22.08%; low CP). The results together with that of symbiotic showed that there were no significant differences in the haematological indices among birds.

In an investigation to ascertain the nutritive value of false yam (*Icacina oliviformis*) tuber meal for broiler chickens, inclusion of sun-dried and boiled false yam tuber meals (*Icacina oliviformis*) in the diets of broiler chickens did not affect the health of the birds (Dei *et al.*, 2011). This conclusion was attributable to no adverse changes in values of blood haemoglobin, haematocrit, and white blood cells of broilers fed sun-dried and boiled false yam tuber meals (Dei *et al.*, 2011).

Agyemang (2010) indicated that broilers fed processed (soaked/cooked) false yam tuber meal did not show negative effects on their blood characteristics of the broilers. Okyere (2011) studied the effect of raw and processed false yam seed meals on the haematology of broilers and indicated that raw and processed false yam seed meals can be substituted for maize in the diets of broilers up to 50g/kg and 100g/kg respectively without any significant effect on their haematology.

In the evaluation of false yam (*Icacina oliviformis*) leaf meal as an ingredient in the diet of weaner rabbits (*Oryctolagus cuniculus*) to improve blood profile, it has been reported that, there were no significant differences (P>0.05) in haemoglobin (Hb) concentration, packed cell volume (PCV) and red blood cells. However, all the erythrocytes values increased from the initial low values to higher values which were all within the normal ranges for rabbits (Ansah and Aboagye, 2011).



## **2.8.0 Biochemical indices of the blood**

The globulins are composed of three fractions, designated alpha, beta and gamma globulins. Alpha-globulins are a group of proteins manufactured almost entirely by the liver. Normally, these proteins increase with acute nephritis, severe active hepatitis, active, usually systemic inflammation, malnutrition and in nephrotic syndromes (Margaret, 2001).

Globulin level has been used as indicator of immune responses and source of antibody production (Abdel-fatta *et al.*, 2008). According to Griminger (1986), high globulin level and low A/G (Albumn/ Globulin) ratio signify better disease resistance and immune response.

Albumin serves as the major reservoir of protein and is involved in colloidal osmotic pressure, acid-base balance, and it acts as a transport carrier for small molecules such as vitamins, minerals, hormones and fatty acids (Margaret, 2001).

Lumeij (1997) also submitted that in acute or chronic conditions, a rise in total protein caused by elevated globulin fraction may occur. Often albumin concentrations are decreased in these situations. The combined effect of these changes is a decrease in the albumin/globulin ratio. Often the total protein concentration is within the reference range, while the albumin/globulin ratio is decreased; therefore the albumin/globulin ratio is often of greater clinical significance than the total protein from globulin.

Total serum protein retained has been reported as an indication of the protein retained in animal's body (Akinola and Abiola, 1991; Esonu *et al.*, 2001), while total blood protein and creatinine contents have been shown to depend on the quality and quantity of dietary protein (Esonu *et al.*, 2001).



The effects of essential oils in the clinical chemistry of broilers are still unclear. Serum concentrations of amylase and lipase in broilers can be measured for pancreatic function evaluation. An increase in these serum parameters might be related to a pancreatic (Lumeij, 1997) or renal injury (González and Silva, 2006). Avian renal function can be evaluated by serum urea and uric acid measurements, the latter being a more reliable parameter; the elevation of these parameters in the serum occurs when 30% or less of the kidneys are functional (Lumeij 1997; Campbell 2007). In birds, an increase in serum urea levels occurs after a decrease in glomerular

filtration rate and may indicate a kidney disease or a physiological response to fluid restriction. However, uric acid excretion occurs via tubular secretion, which is slightly influenced by urine flow and hydration state, increasing only when there are very severe prerenal causes and extensive tubular damage (Lumeij 1997; Phalen 2000). Serum parameters can also be elevated after high protein intake, since they are involved in nitrogen metabolism (Campbell, 2007; Schmidt et al., 2007).

Hepatic function of birds can be evaluated by serum concentration of liver enzymes aspartate aminotransferase (AST) and gamma glutamyltransferase (GGT), cholesterol, and albumin, since its synthesis occurs in the liver (González and Silva, 2006). Increase AST serum levels may be caused by hepatocellular disease (Campbell, 2007), while GGT elevation is usually related to hepatobiliary disease (Tennant, 1997). However, AST is not a specific liver injury enzyme and may also be altered by muscle injuries, as indicated by a concurrent increase in creatine kinase (CK) levels (Tennant, 1997; Campbell, 2007; Schmidt et al., 2007).

### 2.8.1 Plasma chemistry



The use of plasma chemical parameters to diagnose renal disorders is limited. Reference values are missing or inadequate for most avian species. Reference ranges have been produced for some species, but small numbers of birds have been used and these may have been kept under different conditions to the birds. A single parameter falling outside the reference range has limited diagnostic use, and several reasons could explain an aberrant value. Consistent and repeatable abnormal results for several parameters produces a clearer picture and may help direct further investigation such as radiology, endoscopy, and biopsy.

Uric acid and urea values can be assessed, but normal physiologic variations have to be considered. Postprandial values rise in healthy raptors with peaks up to 8 hours after feeding (Lumeij and Remple, 1991). Therefore in raptors a 24-hour fasting period prior to blood sampling is recommended for assessment of renal function. In addition, pathologic increases in uric acid can only be detected if 70% or more of kidney function is lost making this parameter useless for early detection (Krautwald-Junghanns, 1999). Urea can be used to detect dehydration but not to confirm renal dysfunction.

The ratio of plasma urea and uric acid can be used to differentiate prerenal and renal causes of azotemia (Lumeij, 2000). Prerenal azotemia with elevated urea levels produces a high urea:uric acid ratio. The ratio is calculated: plasma urea concentration  $[mmol/L] _ 1000$ : plasma uric acid concentration [lmol/L]. In peregrine falcons (Falco peregrinus) the ratio is >6.5 (Lumeij, 2000). Such normal ratios must be established for each species. Renal failure is likely when uric acid concentration is above the species reference range in a fasted individual. Severe tissue damage could lead to an increased uric acid concentration following release of nucleic acids (Hochleithner, 1994).

Creatinine has limited diagnostic value. In birds, creatine is mostly excreted in urine before it is converted to creatinine so levels of plasma creatinine are low (Bell and Freeman, 1971). Creatinine is excreted by glomerular filtration and reabsorbed in the tubules. Both mechanisms keep the plasma concentration constant, and postprandial elevations have not been observed (Lumeij and Remple, 1991). Greater amounts of creatinine are released in cases of severe muscle damage, but excretion appears to remain constant resulting in an elevated plasma concentration (Hochleithner, 1994). Reduction of glomerular filtration can also lead to an increased creatinine concentration

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(Gylstorff and Grimm, 1998). In theory, if the glomerular filtration rate is preserved but tubules are damaged, plasma creatinine concentration will fall.

Hyperkalemia can be seen in acute renal failure, and can cause severe electrocardiographic changes including cardiac arrest. Sample handling can influence the potassium result, and cells should be separated from the plasma within 1 minute of collection (Lumeij et al., 1998). Hypernatremia is seen after an increased sodium intake or during dehydration.

In renal disease sodium loss can be high and uncompensated. Hyponatremia can be an indicator of renal failure (Lumeij, 1994). Diarrhoea can also lead to significant sodium losses (Woerpel and Rosskopf, 1984). Calcium is reabsorbed after glomerular filtration. In renal failure calcium losses are high, resulting in hypocalcemia. Nutritional and alimentary disorders should also be considered. Hypocalcemia can cause tetany. Interpretation of total and ionized blood calcium should only be done in conjunction with albumin concentration. Hypoalbuminemia reduces the quantity of bound calcium and decreases total calcium concentration, although the level of biologically active ionized calcium may be normal (Lumeij, 1990).

Reduced glomerular filtration rates can lead to high plasma concentrations of inorganic phosphorus (Woerpel and Rosskopf, 1984; Amand, 1986). Decreased phosphorus levels may reflect an alimentary problem such as hypovitaminosis D3 or malabsorption.



# 2.9.0 Processing methods of false yam products to improve their nutritive value for monogastric animals

Removal of ANFs in false yam products is essential in order to effectively utilize their full potential as a feedstuff. It has been established in other root/tuber crops that cooking, soaking, fermentation and other processing methods exerted beneficial effect by destroying the anti-nutritional factors

inherent in them when fed to monogastric animals (Feng *et al.*, 2003). There is evidence that, these proposed processes can be used to improve the nutritional value of false yam tuber and seed meals for monogastric animals. Generally, the processing of the tuber involves peeling and chopping it into pieces of about 2cm long with knife. The chopped tuber can be soaked in water, boiled in water or soaked in water using chemical agents (Sodium chloride or saltpetre). In the case of the seed, it involves cracking the fruits using stone to remove the seeds and seeds then soaked in water or boiled in water. In any of these methods, the products are dried and milled before feeding to monogastric animals.

### 2.9.1 Processed false yam tuber meals and their nutritional value for broiler chickens

Dei *et al.* (2011a) evaluated the nutritive value of sun-dried false yam tuber meal on growth performance of broiler chickens. The tubers were peeled, chopped into chips and sun-dried for 5 days on a concrete floor, milled into gritty floor and labelled SDFYTM. SDFYTM replaced maize in a fish meal-soy based diets. The results of the study shows that SDFYTM when substituted for maize at 3% had no adverse effect on growth performance of broilers. Dei *et al.* (2010) again evaluated the effect of soaked false yam tuber meal on the growth performance of broiler chickens. In this experiment, the chopped tuber was soaked in water for 9 days in the ratio of 1 part of fresh tuber chips to 2 parts of water, while changing the water every 3 days, sundried on concrete floor for 7 days and milled into gritty meal and labelled SFYTM. The nutrient composition of SFYTM sample compared with that of maize is shown in Table 2.8. The SFYTM was substituted (w/w) for maize at four dietary levels (0, 30, 60, 90 g/kg) in a broiler grower diet and fed from 21 to 56 days of age. Results are presented in Table 2.9.



Apart from the CP, NDF and EE content of the tuber, the overall nutrient density of the material (Table 2.8) appeared to be similar to that of maize (Larbier and Laclercq, 1994). The nitrogen-free extracts level was high in the tuber indicating a high concentration of soluble carbohydrates such as starch. The gross energy content also was quite high due to high content of carbohydrates in the tuber.

Nutrient Component	Soaked False Yam Tuber Meal (SFYTM) Dei <i>et al.</i> (2010)	Maize Larbier and Leclercq (1994)		
Dry matter	85.5	87		
Crude protein	5.9	10.2		
Ether extract	1.5	4.8		
Neutral detergent fibre	15.7	2.0		
Ash	2.1	2.1		
Nitrogen Free Extract	74.8	79.5		
Gross Energy (MJ/kg DM)	17.0	16.4		

### Table 2.8: Comparison of nutrient composition of SFYTM with maize (% DM basis)

# Table 2.9: Effect of soaked (9 days) false yam tuber meal on mean feed intake, weight gain and gain/feed ratio of broilers (3-8 weeks of age)

	Diets containing SFYTM (g/kg)						
Parameters	Control (0)	30	60	90	±SED		
Feed intake (g/bird/day)	120.0	122.3	123.0	119.0	6.39ns		
Weight gain (g/bird/day)	56.7	51.4	51.2	46.2	4.74ns		
Gain: Feed Ratio	0.473	0.420	0.416	0.388	0.037ns		
GED 1 1 1 1166				1 (201			

SED-standard error difference, ns- not significant, (P>0.05). Source: Dei et al. (2010)

The similarities in feed intake between the control birds and those birds fed diets containing the SFYTM (Table 2.9) indicate that soaking might have removed the bitter compound in the tuber, thereby making it more palatable. Soaking might have also improved the utilization of the product. Therefore, replacing maize with the product at levels tested in the experimental diets tended to give similar growth performance of broilers (Table 2.9).



In a similar experiment, Dei *et al.* (2012a) investigated the effect of SFYTM on growth performance of broiler chickens.

In this experiment, the soaking duration of the tuber was extended to 15 days and the product replaced maize (w/w) in maize-fishmeal based grower diet at 120 and 150 g/kg. The nutrient composition of SFYTM in terms of its dry matter, energy content, crude protein, ether extract, neutral detergent fibre, ash, starch and essential amino acids concentrations is shown in Table 2.10. The results of the nutrient analysis (Table 2.11) corroborate the findings in Table 2.8 that SFYTM is high in energy and carbohydrate as compared to maize. Thus it is capable of replacing maize partially in a broiler and other non-ruminant diets. But the low protein content of SFYTM demands that it should be fed with other ingredients high in protein in order to balance for protein. The level of essential amino acids, which are needed by the animal's body, but cannot be synthesized by the animal's body namely methionine, threonine, arginine, isoleucine, leucine, valine, phenylalanine and glycine are low in SFYTM as compared to maize (Table 2.10) and as such must be provided in their diets.

The study on growth of broilers (Table 2.11) showed that SFYTM when included up to 12 and 15% had adverse effects on growth performance of broiler chickens due to significant reduction in feed intake. This probably may be due to increase in concentration of residual anti-nutritional factor still present in the SFYTM when fed at 120 and 150 g/kg. McDonald *et al.* (2002) indicated that terpenes can actually impair the availability of nutrients and reduce performance in animals as observed in this study.

Table 2.10: Comparison of nutrient composition of SFYTM (15 days) with maize (% DM basis)

Components	SFYTM (Dei <i>et al.</i> , 2012a)	Maize (NRC, 1994)
Dry matter	82.79	89
Crude protein	3.63	7.6
Ether extract	1.10	3.4
Neutral detergent fiber	23.14	2.0
Ash	1.69	2.1
Starch	70.33	79.5
Gross Energy (MJ/kg DM)	14.27	16.40
Essential Amino Acids		
Methionine	0.01	0.16
Lysine	0.24	0.23
Threonine	0.07	0.26
Tryptophan	0.05	0.05
Arginine	0.08	0.33
Isoleucine	0.08	0.26
Leucine	0.13	0.88
Valine	0.13	0.35
Histidine	0.16	0.20
Phenylalanine	0.07	0.33
Glycine	0.08	0.29

Table 2.11: Effect of soaked (15 days) false yam tuber meal on mean feed intake, weight gain and gain/feed of broiler chickens (3-8 weeks of age)

	-		-			
1	Parameters	Control (0)	120	150	±SED	Р
Ì	Feed intake (g/bird/day)	111.1ª	86.1 <sup>b</sup>	72.0 <sup>b</sup>	7.13	0.004
Į	Weight gain (g/bird/day)	43.1 <sup>a</sup>	26.3 <sup>b</sup>	19.9°	2.74	< 0.001
	Gain: Feed Ratio	0.39 <sup>a</sup>	0.30 <sup>b</sup>	0.28 <sup>c</sup>	0.012	< 0.001
		0.07	0.00	0.20	0.012	

SED-standard error of difference, P- probability, Means with the same superscripts in a row are not significantly different (P>0.05). Source: Dei *et al.* (2012a)

In this study, the inclusion level of SFYTM was increased beyond 90 g/kg on the assumption that

prolonged soaking would further improve its nutritional value.



Unfortunately this resulted in adverse effects on growth performance (Table 2.11). This suggests that SFYTM cannot be fed beyond 90 g/kg. Therefore, alternative methods of processing the false yam tuber should be employed to improve the nutritive value of the false yam tuber.

Table 2.13 evaluated the effect of soaking false yam tuber in 0.1% saltpetre solution for 12 days on the growth of broilers. The solution was changed every 3 days, the sample was sun-dried and milled. The experiment was conducted with the aim that saltpetre may react with terpenes found in the tuber. Saltpetre (potassium nitrate) is known to have many uses and can react with many different compounds including terpenes (Pommer, 2003). The processed product replaced maize (w/w basis) in maize-fishmeal based grower diet at 80, 100 and 120 g/kg (Table 2.12). It is concluded that soaking false yam tuber in saltpetre solution improved its nutritive value for broilers up to 120 g/kg. In a similar experiment (Table 2.13), this time replacing saltpetre with common salt at the same concentration during the processing of the tuber, soaking false yam tuber in 0.1% common salt solution had adverse effect on growth performance of broiler chickens.

 Table 2.12: Effect of false yam tuber soaked (12 days) in saltpetre on feed intake, weight gain and gain-to-feed ratio of broiler chickens (23-56 days of age)

	Diets containing saltpetre treated tuber meal (g/kg)					
Parameters	Control (0)	80	100	120	±SED	Р
Feed intake (g/bird/day)	119.4	120.9	117.6	115.5	3.37	0.461
Weight gain (g/bird/day)	40.6	37.4	37.2	37.2	1.31	0.078
Gain: Feed Ratio	0.34	0.31	0.32	0.31	0.016	0.307

SED-standard error difference, P- probability, Means with the same superscripts in a row are not significantly different (P>0.05). Source: Dei *et al.* (2013a)



				•	5		
	Diets containing salt treated tuber meal (g/kg)						
Parameters	Control (0)	80	100	120	±SED	Р	
Feed intake (g/bird/day)	98.7	95.1	97.5	100.6	3.59	0.519	
Weight gain (g/bird/day)	42.4 <sup>a</sup>	33.4 <sup>b</sup>	35.0 <sup>b</sup>	33.0 <sup>b</sup>	2.76	0.030	
Gain: Feed Ratio	0.43 <sup>a</sup>	0.35 <sup>b</sup>	0.36 <sup>b</sup>	0.33 <sup>b</sup>	0.021	0.005	

 Table 2.13: Effect of soaked (12 days) false yam tuber treated with sodium chloride on feed intake, weight gain and gain-to-feed ratio of broiler chickens (23-56 days of age)

SED-standard error difference, P- probability, Means with the same superscripts in a row are not significantly different (P>0.05). Source: Dei *et al.* (2013b)

According to NAS (2008), sliced false yam tuber and seeds can be placed in boiling water for a number of hours to help to remove the toxic compounds thereby rendering the product palatable. Heat treatment such as boiling has been effective in eliminating some anti-nutritive factors in seed meals (Barnes *et al.*, 2009) as well as terpenes in leaves (Yang *et al.*, 2007).

Results in Table 2.14 show that, false yam tuber can also be processed by boiling in water. The chopped tuber was boiled in water for 2 hours at a ratio of 1 part of fresh tuber to 1 part of water. The water was discarded, and the boiled tubers were sun-dried for 8 days and then milled into a gritty meal and labelled BFYTM. The BFYTM was substituted (w/w) for maize at four dietary levels (0, 30, 60, 90 g/kg) in a broiler grower diets and fed from 21 to 56 days of age. The concentration of the total anti-nutritional factors (gum resins) in the sun-dried false yam tuber meal (SFYTM) was 37.5g resin/kg DM (Dei *et al.*, 2011a), boiling has been effective in reducing the resins by 39% (37.5g resins/kg DM to 22.88g resins/kg DM). The growth response of broilers to BFYTM in their diets indicated that the BFYTM could be added in the diets of broiler chickens up to 90g/kg without any adverse effect on their growth performance (Table 2.14).



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Table 2.14: Effect of boiled (2 hours) false yam tuber meal on feed intake, weight gain and
gain-to-feed ratio of broiler chickens (21-56 days of age)

Diets con	g			
Control (0)	30	60	90	SEM
114.8	116.5	119.8	117.3	3.41ns
50.8	52.4	52.9	51.5	3.70ns
0.44	0.45	0.44	0.44	0.020ns
	Control (0) 114.8 50.8	Control (0)         30           114.8         116.5           50.8         52.4	Control (0)         30         60           114.8         116.5         119.8           50.8         52.4         52.9	114.8116.5119.8117.350.852.452.951.5

SEM-Standard error of means, ns-not significant (P>0.05). Source: Dei et al. (2011a)

Osei (2011) conducted similar experiment using boiled false yam tuber meal (BFYTM) but at higher dietary levels, BFYTM replaced maize (w/w) at 0, 120 and 150 g/kg in grower broiler diet and fed from 21-56 day of age. There was clear indication that including BFYTM at 120 and 150 g/kg in the diets of broilers had adverse effect on growth (Table 2.15). Similar results were obtained when the soaked tuber was fed at 120 and 150 g/kg (Table 2.11).

 Table 2.15: Effect of boiled (2 hours) false yam tuber meal on feed intake, weight gain and gain-to-feed ratio of broiler chickens (21-56 days of age)

	Diets contair	Diets containing BFYTM g/kg				
Parameters	Control (0)	120	150	±SED	Р	
Feed intake (g/bird/day)	105.8ª	94.2 <sup>b</sup>	78.8 <sup>b</sup>	4.78	0.004	
Weight gain (g/bird/day)	49.41 <sup>a</sup>	38.56 <sup>b</sup>	31.08 <sup>c</sup>	2.390	< 0.001	
Gain: Feed Ratio	0.47 <sup>a</sup>	0.41 <sup>b</sup>	0.40 <sup>b</sup>	0.009	< 0.001	

SED-Standard error difference, P-Probability, Means with the same superscripts in a row are not significantly different (P>0.05). Source: Osei (2011)



Osei *et al.* (2013a) (Table 2.16) repeated the experiment by testing BFYTM at 50 and 100 g/kg in broiler grower diets. They reported again that BFYTM had a negative effect on growth of broilers. According to the authors, there was no clear explanation to the disparities between their work and that of Dei *et al.* (2011a) (Table 2.14), but factors such as false yam varietal differences and different broiler strains could be important.

	Diets contair	Diets containing BFYTM g/kg					
Parameters	Control (0)	50	100	LSD			
Feed intake (kg/bird/day)	129.1	121.9	118.1	0.011			
Weight gain (g/bird/day)	52.0 <sup>a</sup>	41.3 <sup>b</sup>	40.1 <sup>b</sup>	0.004			
Feed conversion ratio	2.48 <sup>b</sup>	2.95 <sup>a</sup>	2.94 <sup>a</sup>	0.498			

Table 2.16: Effect of boiled (2 hours) false yam tuber meal on feed intake, weight gain and feed conversion ratio of broiler chickens (4-8 weeks of age)

LSD-Least significant difference at 5% level, Means with the same superscripts in a row are not significantly different (P>0.05). Source: Osei *et al.* (2013a)

It is not known how varying the boiling duration would influence the nutritive value of the tuber for broiler chickens. Thus, Tibieb (2011) carried out an experiment to evaluate the effect of varying boiling time (1-3 hours) on the nutritive value of false yam tuber for broiler chickens. In this study, chopped tubers were boiled in water (1 part of fresh tuber to 1 part of water) for 1, 2 or 3 hours and sun-dried for 7 days and milled into gritty meals. Each of the processed products replaced maize (w/w) at 100 g/kg in grower broiler diet and fed from 3 to 8 weeks of age. The result of this experiment is presented in Table 2.17.

The study revealed that, boiling durations of the false yam tuber had similar effect on its feeding value for broiler chickens. There is an indication that, birds can be fed the material boiled for one hour. Nevertheless, feeding the boiled tuber sample at 100 g/kg had adverse effect on broiler performance. This reinforces the fact that the boiled tuber cannot be fed beyond 90 g/kg.



Table 2.17: Effect of boiling duration of false yam tuber on feed intake, weight gain and gain-										
to-feed ratio of broiler chickens (3-8 weeks of age)										

	Boiling duration						
Parameters	Control (0)	1HR	2HR	3HR	±SED	Р	
Feed intake (g/bird/day)	121.3 <sup>a</sup>	102.9 <sup>b</sup>	103.8 <sup>b</sup>	104.1 <sup>b</sup>	5.23	*	
Weight gain (g/bird/day)	55.5 <sup>a</sup>	42.5 <sup>b</sup>	42.1 <sup>b</sup>	45.2 <sup>b</sup>	3.00	**	
Gain: Feed Ratio	0.46	0.41	0.41	0.43	0.026	ns	

SED-standard error difference, Means with the same superscripts in a row are not significantly different (P>0.05), ns-non-significant. Source: Tibieb (2011)

Both soaking and boiling of the tuber appear to be promising in improving the nutritional value for broiler chickens. However, it is not known whether there may be synergy in nutritional improvement if the tuber is boiled after soaking. Therefore, Dei et al. (2013c) (Table 2.18) used soaked/boiled tuber during the finishing phase of broiler chickens to determine the effect of the processed false yam tuber meal on their growth performance. Four treatments comprising a control (no tuber meal) and diets containing the processed false vam tuber meal as substitute for maize at 80, 100 and 120 g/kg were tested and results presented in Table 2.18. Based on the results of the study, it was concluded that combined methods of soaking and boiling false yam tuber had improved its feed value and could be fed up to 120 g/kg without adverse effect on performance of broiler finishers.

chickens (4-8 week of age) Diets containing SBFYTM (g/kg) 80 Parameters Control 100 120 ±SED Р 131<sup>a</sup> 118.9<sup>b</sup> 119.1<sup>b</sup> Feed intake (g/bird/day) 133.5<sup>a</sup> 3.57 0.005 Weight gain (g/bird/day) 62.9 62.6 50.7 5.10 0.134 58.1 Gain: Feed Ratio 0.47 0.48 0.49 0.43 0.033 0.336

Table 2.18: Effect of SBFYTM on feed intake, weight gain and gain-to-feed ratio of broiler

SED-standard error difference, P- probability, Means with the same with superscripts in a row are not significantly different (P>0.05). Source: Dei et al. (2013c)



A previous study by Antai and Nkwelang (1999) has shown that, fermentation of the tuber can improve its nutritional value. The concentration of terpenes and other un-identified anti-nutritional factors in false yam tuber and seeds may be reduced by fermentation. According to Antai and Nkwelang (1999), the fermentation of *Icacina manni* paste for six days resulted in a marked decrease in the level of toxicants. Therefore, a study was conducted by Teog (2010) to evaluate the effect of natural fermentation on the nutritive value of raw false yam tuber meal for broilers. The chopped unprocessed tubers were sun-dried for 7 days and milled into gritty meal.

Fermentation was done by native micro-organisms, where water was added to the raw tuber flour (1:1) to form a thick paste in a plastic basin. The paste was in a container for 3 days after which it was sun-dried for 7 days and labelled FFYTM. The fermented product replaced maize (w/w) at 30, 60 and 90 g/kg in maize-fish meal based grower diets. The nutritive value of the fermented product for poultry was determined based on growth performance of broilers (Table 2.19).

 Table 2.19: Effect of FFYTM on feed intake, weight gain and gain-to-feed ratio of broiler chickens (4-8 week of age)

	Diets containing FFYTM (g/kg)						
Parameters	Control (0)	rol (0) 30 60 90		90	±SEI	) P	
Feed intake (g/bird/day)	93.1ª	84.9 <sup>ab</sup>	74.6 <sup>bc</sup>	65.0 <sup>c</sup>	6.67	0.006	
Weight gain (g/bird/day)	41.5 <sup>a</sup>	35.6 <sup>b</sup>	25.8 <sup>c</sup>	19.9 <sup>d</sup>	2.74	< 0.001	
Gain: Feed Ratio	0.45 <sup>a</sup>	0.42 <sup>a</sup>	0.35 <sup>bc</sup>	0.31 <sup>c</sup>	0.02	< 0.001	

SED-standard error difference, P- probability, Means with the same superscripts in a row are not significantly different (P>0.05). Source: Teog (2010)

The depression in feed consumption as the FFYTM was increased in the diets was an indication that the bitter compound in the tuber was not reduced to a level that makes the feed palatable or acceptable to the birds. The reduction in feed intake resulted in growth depression and consequent poor utilization of the diets containing FFYTM. The poor performance of the birds could be attributed to the presence of the anti-nutritional factors (gum resins) in the product, an indication that natural fermentation was not effective in reducing the anti-nutritional factors to a level that is acceptable to the birds.



### 2.9.2 Processed false yam seed meals and their nutritional value for broiler chickens

Similar studies involving broiler chickens have been carried out to determine the utilization of false yam seed. Dei *et al.* (2011b) reported that processing the seeds by soaking in water improves its nutritional value. The seeds were soaked in water (i.e. 1 part of seeds to 2 parts of water) for 3

days and water changed every 24 hours, sundried for 7 days and milled into gritty meal and labelled SFYSM. The SFYSM replaced maize on (w/w) at four dietary levels (0, 50, 75, 100 g/kg) in a broiler grower diet and fed from 3 to 8 weeks of age. Results are presented in Table 2.20. The study showed that, soaked false yam seed meal can replace maize at 100 g/kg without any adverse effects on growth performance of broilers. This is an indication that soaking of the seed might have improved the nutritive value by reducing concentration of the gum resins and other unidentified ANFs and making it more palatable for the birds.

 Table 2.20: Effects of soaked (3 days) false yam seed meal on mean feed intake, weight gain and gain/feed of broilers (3-8 weeks of age)

	Diets conta	g/kg)				
Parameters	Control (0)	50	75	100	±SED	Р
Feed intake (g/bird/day)	105.8	104.8	96.5	94.7	4.35 0	.075
Weight gain (g/bird/day)	60.6	57.3	55.8	56.2	3.80 0	.598
Gain: Feed Ratio	0.47	0.45	0.46	0.48	0.015 0	.375

SED-standard error difference, P- probability. Source: Dei et al. (2011b)

In another experiment to evaluate the effect of soaking duration on the nutritive value of false yam seeds by Dei *et al.* (2012c), the seeds were soaked in water (i.e. 1 part to 1.5 parts of water) for 9, 12 or 15 days and water changed every 3 days, sundried for 7 days and milled into gritty meals and labelled 9SFYSM, 12SFYSM and 15SFYSM, respectively. Each of the processed products replaced maize (w/w) at 100 g/kg in grower diet. The results of nutrient analysis (Table 2.21) of the processed seeds indicate that SFYSM is rich in carbohydrates as in maize, so can be a substitute in broiler diet. The low level of protein means the SFYSM should be fed with ingredients high in protein. Table 2.21 shows that soaking duration has no appreciable effect on nutrient composition of the seed meal. The results of the feeding trial indicate that soaking the seed for 12 days improved its nutritive value for broilers (Table 2.22).



Nutrient Component	(I	(Dei et al., 2012c)				
	9SFYSM	12SFYSM	15SFYSM	Leclercq,1994)		
				Maize		
Dry matter	85.90	85.52	86.12	86.95		
Crude protein	8.57	8.11	8.40	10.20		
Ether extract	0.48	0.82	0.56	4.80		
Neutral Detergent Fibre	7.41	7.12	9.85	<sup>1</sup> (2)		
Ash	0.56	0.75	0.63	$^{1}(2.1)$		
Starch	76.34	76.65	77.65	<sup>1</sup> (79.5)		
Gross energy (MJ/kg)	15.34	15.25	15.38	-		

Table 2.21: Comparison of nutrient composition of SFYSM samples and with maize (% DM	
basis)	

Source: <sup>1</sup>Value in bracket adapted from NRI (1994)

 Table 2.22: Effect of SFYSM on feed intake, weight gain and gain-to-feed ratio of broiler chickens (3-8 weeks of age)

	Soaking duration of false yam seed							
Parameters	Control	9d	12d	15d	±SED	Р		
Feed intake (g/bird/day)	111.1 <sup>a</sup>	99.4 <sup>b</sup>	107.4 <sup>ab</sup>	108.0 <sup>a</sup>	2.23	0.004		
Weight gain (g/bird/day)	43.1 <sup>a</sup>	35.4 <sup>b</sup>	39.0 <sup>ab</sup>	38.5 <sup>b</sup>	1.53	0.004		
Gain: Feed Ratio	0.39	0.36	0.36	0.36	0.016	0.096		

SED-standard error difference, Means with the same superscripts in a row are not significantly different (P>0.05). Source: Dei *et al.* (2012c)

Dei *et al.* (2011b) carried out an experiment to determine the effect of boiling on the nutritive value of the seeds. The seeds were crushed and boiled in water (1 part of seed to 1 part of water) for 30 minutes and sun-dried for 7 days and milled into gritty meal and labelled BFYSM. The boiled seed meal replaced maize (w/w) at 0, 50, 75 and 100 g/kg in grower broiler diet and fed from 3 to 8 weeks of age. The results are presented in Table 2.23. The study showed that, boiling the seed for 30 minutes and including it in the diet at 50 g/kg or more had no adverse effect on feed intake but adversely affected broiler chicken growth performance. This suggests that, boiling has no effect on the anti-nutritional factors in the seed that affect feed digestibility hence poor digestion and



utilization of feed (Chakam *et al.*, 2010). It appears that boiling duration may be too short to influence the major ANFs in the seeds.

 Table 2.23: Effect of boiled (30 minutes) false yam seed meal on feed intake, weight gain and gain-to-feed ratio of broiler chickens (3-8 weeks of age)

Diets containing BFYSM g/kg										
Parameters	Control (0)	50	75	100	±SED	Р				
Feed intake (g/bird/day)	105.8	97.5	105.5	88.0	6.52	0.08				
Weight gain (g/bird/day)	49.41 <sup>a</sup>	42.65 <sup>b</sup>	40.43 <sup>b</sup>	39.43 <sup>b</sup>	2.35	0.01				
Gain: Feed Ratio	0.47 <sup>a</sup>	0.44 <sup>a</sup>	0.39 <sup>b</sup>	0.45 <sup>a</sup>	0.02	0.03				

SED-standard error difference, P-Probability, Means with the same superscripts in a row are not significantly different (P>0.05). Source: Dei *et al.* (2011b)

In view of the above findings, Dei *et al.* (2013d) evaluated the effect of different boiling durations of the seed on its nutritive value for broilers. The seeds were boiled in water (i.e. 1 part of seeds to 2 parts of water) for 1, 2 and 3 hours, respectively. Sun-dried for 5 days and milled into gritty meal and labelled BFYSM-1h, BFYSM-2h and BFYSM-3h to represent boiling durations and substituted for maize at 100 g/kg (w/w) in broiler finisher diets. The results indicate that feed consumption, weight gain and feed conversion efficiency were adversely affected for birds fed the BFYSM based diets (Table 2.24). The performances of all the birds fed the BFYSM based diets were similar. This suggests that extended duration of boiling the seeds has no nutritional advantage. Boiling may not be appropriate method for detoxifying the seeds for feeding broilers.



	Diets containing BFYSM							
Parameters	Control	1h	2h	3h	±SED	Р		
Feed intake (g/bird/day)	161.5 <sup>a</sup>	133.3 <sup>b</sup>	123.6 <sup>b</sup>	123.4 <sup>b</sup>	5.72	< 0.001		
Weight gain (g/bird/day)	64.9 <sup>a</sup>	43.5 <sup>b</sup>	37.1 <sup>b</sup>	38.7 <sup>b</sup>	2.76	< 0.001		
Gain: Feed Ratio	0.40 <sup>a</sup>	0.33 <sup>b</sup>	0.30 <sup>b</sup>	0.31 <sup>b</sup>	0.016	< 0.001		

Table 2.24: Effect of boiled false yam seed meals on feed intake, weight gain and gain-to-feed ratio of broiler chickens (4-8 week of age)

SED-standard error difference, P- probability, Means with the same superscripts in a row are not significantly different (P>0.05). Source: Dei *et al.* (2013d)

So far, it is not known whether combined processing method such as soaking and boiling would further improve the nutritional value of the seed for birds. Therefore, Mensah (2013) carried out a study in which broiler chickens were fed diets containing false yam seeds which were soaked (12 days) and boiled (2 hours). The processed product (SBFYSM) replaced maize (w/w) at 80, 100 and 120 g/kg in maize-fishmeal based grower diets. The results showed that feeding SBFYSM at levels tested resulted in lower growth performance (Table 2.25). It has been observed that seeds that are boiled are quite hard therefore not easily digested, since particles of the boiled seed meal were visible in the droppings. This is a further indication that boiling as a method is not appropriate for detoxifying the seeds for birds.

A study was undertaken to determine the effect of adding 15 g/kg of charcoal to graded levels of false yam seed meal-based diets on the growth performance of broiler chickens. False yam seeds as a nonconventional feedstuff have the potential to replace maize in poultry production. Moreover, it has been reported that the major limiting factor of false yam seed is its anti-nutritional factor (gum resin) that can be toxic and reduces palatability of feed when given to animals (NRI, 1987).

Researchers (Gerlach and Schmidt, 2012) described the usefulness of biochar (charcoal) as substance that promotes digestion, improves feed efficiency, and thus in particular energy absorption via the feed. Toxins such as dioxin, glyphosate, mycotoxins, pesticides and polycyclic



aromatic hydrocarbons (PAHs) are efficiently bound by the biochar, thereby obviating any adverse effects on the digestive system and intestinal flora. The health, activity and balance of the animals will also be improved, as will meat and egg production.

Preliminary studies involving soaking false yam seeds improved its nutritive value for broilers up to 9% in their diets (Dei et al., 2011). Mohammed (2016) in a preliminary study using charcoal at 10 g/kg diet in soaked false yam seed meal (SFYSM)-based diet in broiler chicken production concluded that charcoal could be a potential substance that can be used in poultry feed containing SFYSMs. It is assumed that charcoal addition to SFYSM can further reduce the negative effect of residual toxins in the false yam seed meal thereby enhancing its nutritive value for broilers beyond 100 g/kg inclusion level. Four treatments comprising a control (no seed meal) and diets containing SFYSM as substitute for whole diet (Commercial broiler finisher diet) at 0 (Control), 100 (T1), 120 (T2), and 140 (T3) g/kg and each treatment was replicated three times. Powdered charcoal was added to the SFYSM-based diets at 15 g/kg as add-on. The experimental diets were fed in mash form from 4 to 8 weeks of age. There were no significant (P>0.05) differences in growth parameters measured in terms of feed intake, daily weight gain and feed conversion efficiency for all the treatment groups (Table 2.26). The similarities in growth performance observed in this study could be as a result of similar feed consumption of the experimental birds.



This occurrence could mean that the addition of the SFYSM with 15 g/kg charcoal did not alter the palatability of the diets. It could also be concluded that the charcoal addition was able to bind effectively with the residual toxins (terpenes) that might be found in the SFYSM, thereby enhancing its utilization by the birds.

	Diets containing SBFYSM (g/kg)						
Parameters	Control 80	100	120	±SED	Р		
Feed intake (g/bird/day)	161.46 <sup>a</sup> 129.72 <sup>a</sup>	127.05 <sup>b</sup>	120.95 <sup>c</sup>	2.140	< 0.001		
Weight gain (g/bird/day)	64.87 <sup>a</sup> 52.81 <sup>b</sup>	52.62 <sup>b</sup>	48.66 <sup>c</sup>	1.848	< 0.001		
Gain: Feed Ratio	0.40 0.41	0.42	0.40	0.009	0.356		

 Table 2.25: Effect of SBFYSM on feed intake, weight gain and gain-to-feed ratio of broiler chickens (4-8 week of age)

SED-standard error difference, P- probability, Means with the same superscripts in a row are not significantly different (P>0.05). Source: Mensah (2013)

Table 2.26: Effect of graded level of SFYSM with 1.5% charcoal addition to the diets of finisher broiler chickens on growth performance (4-8 weeks of age).

	0	1	· · · · · · · · · · · · · · · · · · ·		0 /			
Parameter		Control	T1	T2	T3	SED	Р	
Feed intake (g/b/d)		153.9	152.9	153.9	154.9	2.16	0.836	
Daily weight gain (g/b/d)		53.8	55.7	52.2	50.3	2.01	0.128	
Gain-to-feed ratio		0.35	0.36	0.34	0.33	0.015	0.138	
Final live-weight (kg/bird)		2.63	2.68	2.59	2.54	0.050	0.128	

Mohammed *et al.* (2017)

### 2.9.3 Processed tuber and seed meals and their nutritional value for layer chickens

The effect of processed false yam tuber and seed meals on feed intake, weight gain and gain-tofeed ratio of pullets (9-19 weeks of age) are shown in Table 2.27. The boiled false yam tuber meal can be added to diets of growing pullets at 25 g/kg without any adverse effect on their growth performance (Dei *et al.*, 2012b). However, subsequent egg production of pullets fed diets containing boiled false yam tuber meal up to 100 g/kg during the grower phase was not compromised (Dei *et al.*, 2012b) (Table 2.27)

The feeding of soaked false yam seed meal up to 100 g/kg in growing pullet diets (Table 2.27) showed no adverse effect on their growth (Dei *et al.*, 2012d). In addition, soaked false yam seed meal can be included in the diets of layer chickens up to 100 g/kg without adverse effect on their egg production (Mohammed and Dei, 2012).



Processed	Treatments	Feed intake	Weight gain	Gain/feed ratio	Source
products	(g/kg)	(g/bird/day)	(g/bird/day)		
Diets containing	Control (0)	72.4ª	13.3ª	0.18	
SFYTM	50	72.4 75.7 <sup>a</sup>	13.3 11.7 <sup>ab</sup>	0.18	Niayale,
51 1 1 11	50 75	73.7 64.0 <sup>b</sup>	11.7 11.1 <sup>b</sup>	0.15	(2013)
	100	62.5 <sup>b</sup>	10.2 <sup>b</sup>	0.16	(2013)
	SED	2.01	0.67	0.10	
	SED P-value	<0.001	0.07	0.143	
	r-value	<0.001	0.011	0.145	
Diets containing	Control (0)	64.1ª	11.0 <sup>a</sup>	0.17	
BFYTM	25	61.8 <sup>b</sup>	10.5 <sup>ab</sup>	0.16	
	50	65.4 <sup>a</sup>	10.0 <sup>b</sup>	0.15	Dei et al.
	75	65.1 <sup>a</sup>	9.8 <sup>b</sup>	0.15	(2012b)
	100	65.4 <sup>a</sup>	9.7 <sup>b</sup>	0.14	· · ·
	SED	1.27	0.41	0.01	
	P-value	0.022	0.04	0.063	
Diets containing	Control (0)	73.08ª	13.25ª	0.18 <sup>a</sup>	
SBFYTM	50	74.50 <sup>a</sup>	11.74 <sup>b</sup>	0.16 <sup>b</sup>	Gyawu,
	75	64.68 <sup>b</sup>	11.79 <sup>b</sup>	0.19 <sup>a</sup>	2013
	100	63.10 <sup>b</sup>	11.36 <sup>b</sup>	0.18 <sup>a</sup>	
	SED	2.21	0.45	0.01	
	P-value	0.002	0.014	0.011	
Diets containing	Control (0)	76.07	13.57	0.18	
SFYSM	50	73.39	13.89	0.19	Dei et al.
	75	73.75	14.14	0.19	(2012d)
	100	75.36	13.18	0.18	· · ·
	SED	2.67	1.09	0.02	
	P-value	0.713	0.831	0.730	
Diets containing	Control (0)	72.4	13.25ª	0.18 <sup>a</sup>	
BFYSM	50	70.8	10.13 <sup>b</sup>	0.14 <sup>b</sup>	Owusu,
	75	63.1	10.45 <sup>b</sup>	0.17 <sup>b</sup>	(2013)
	100	69.8	9.93 <sup>b</sup>	0.14 <sup>b</sup>	. /
	SED	3.49	0.55	0.01	
	P-value	0.113	<0.001	0.002	

# Table 2.27: Effects of processed false yam products on feed intake, weight gain and gain-to-feed ratio of pullets (9-19 weeks of age)

SED-Standard error of difference, Means with the same superscript within column are not significantly different (P>0.05), SFYTM-Soaked false yam tuber meal, BFYTM-Boiled false yam tuber meal, SBFYTM- Soaked and boiled false yam tuber meal, SFYSM- Soaked false yam seed meal, BSYSM-Boiled false yam seed meal.

It has been shown that boiled false yam seed meal in the diets of pullets up to 50 g/kg had adverse effect on their growth performance (Table 2.27) and subsequent egg production (Table 2.28).



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However, soaked false yam tuber meal (SFYTM) can be included in the diets of pullets up to 5% without any adverse effect on growth (Table 2.27) and subsequent egg production (Table 2.29). Soaking the tuber prior to boiling was evaluated on growth and subsequent egg production of pullets and it was found that it had adverse effects on growth performance (Table 2.27) and subsequent egg production (Table 2.28) when included at 50 g/kg or more in the diets of pullets.

 Table 2.28: Performance of layer chickens fed processed false yam tuber meal during grower

 phase on subsequent egg production

Processed	Treatments	Feed intake	Hen-day	Egg	Feed	Source
products	(g/kg)	(g/bird/day)	production (%)	weight(g)	conversion efficiency	
Diets	Control (0)	90.94	68.9ª	49.21	0.54	
containing	50	88.54	60.9 <sup>b</sup>	48.93	0.55	
SFYTM	75	86.38	52.1 <sup>b</sup>	49.95	0.58	Effah,
	100	87.55	47.6 <sup>bc</sup>	49.43	0.57	(2013)
	SED	2.79	6.28	0.88	0.02	
	P-value	0.54	0.038	0.709	0.36	
Diets	Control (0)	93.5	79.1	51.3	1.84	
containing	25	95.2	77.0	53.6	1.78	
BFYTM	50	96.2	78.3	53.8	1.79	Dei et al.
	75	94.4	80.3	53.2	1.77	(2012b)
	100	93.5	82.9	52.9	1.77	
	SED	3.24	4.57	1.28	0.67	
	P-value	0.937	0.746	0.381	0.793	
Diets	Control (0)	90.5	68.9ª	49.2	0.54	
containing	50	101.8	60.2 <sup>b</sup>	49.6	0.50	
SBFYTM	75	90.7	52.4 <sup>c</sup>	49.2	0.55	Ofori,
	100	83.2	51.3°	49.4	0.59	(2013)
	SED	8.65	3.36	0.91	0.05	
	P-value	0.273	0.003	0.966	0.310	
Diets	Control (0)	90.5	70.3 <sup>a</sup>	49.25	0.55	
containing	50	91.2	53.5 <sup>b</sup>	50.56	0.56	Opoku
BFYSM	75	81.5	51.9 <sup>b</sup>	50.23	0.62	-Addae (2013
	100	95.3	48.6 <sup>b</sup>	50.20	0.37	
	SED	6.84	4.87	0.82	0.11	
	P-value	0.304	0.009	0.461	0.219	

SED-Standard error of difference, Means with the same superscripts within column are not significantly different (P>0.05), SFYTM-Soaked false yam tuber meal, BFYTM-Boiled false yam tuber meal, SBFYTM- Soaked and boiled false yam tuber meal, BSYSM-Boiled false yam seed meal.

It is not known how soaking the tuber would influence the apparent nutrient digestibility and performance of layers. In this study, chopped tubers were soaked in water (1 part of fresh tuber to



2 part of water) for 12 days, whilst changing the water every 3 days. The soaked tubers were sundried for 7 days and milled into gritty flour and labelled SFYTM. The SFYTM was used to replace maize on (w/w) at 0, 50, 75 and 100 g/kg layer diet and fed from 19 to 35 weeks of age. The result of apparent nutrient digestibility (Table 2.29) of the diets shows that addition of SFYTM to the diets had adverse effects on crude protein and crude fat digestibility. This is an indication that the residual concentration of the resins (terpenes) after soaking might be high enough to have metabolic effect even though resins level was not determined. This could have influenced the digestibility of protein and fat in the diets. Since, terpenes can actually impair the availability of nutrients and reduce performance in animals (McDonald *et al.*, 2002) as observed in this study. The effect on egg laying performance of layers (Table 2.30) showed that feed consumption of hens in all treatments was similar but egg production trend declined as the SFYTM increased in the digestibility (Table 2.29) of hens. Residual concentrations of ANFs might have affected major nutrients (protein and fat) which are components of egg, hence, egg laying performance was

reduced.

Dietary SF11M levels (g/kg)								
Parameter	Control (0)	50	75	100	±SED	P- value		
Dry matter (%)	69.5	72.9	68.7	68.7	2.60	0.324		
Crude protein (%)	67.7 <sup>a</sup>	53.5 <sup>b</sup>	56.2 <sup>b</sup>	53.9 <sup>b</sup>	4.41	0.033		
Crude fat (%)	81.3 <sup>a</sup>	84.6 <sup>a</sup>	72.9 <sup>b</sup>	71.5 <sup>b</sup>	2.63	< 0.001		
Ash (%)	75.5	66.8	66.4	73.2	4.49	0.052		

 Table 2.29: Effect of SFYTM on apparent nutrient digestibility of layers (19-35 weeks of age)

 Dietary SEVTM levels (g/kg)

SED-standard error difference, P- probability, Means with the same superscripts in a row are not significantly different (P>0.05). Source: Mohammed and Dei (2013)

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Dietary SFYTM levels (g/kg)								
Parameter	0	50	75	100	±SED	P-value		
Feed intake (g/bird/day)	89.6	94.2	93.3	92.1	2.33	0.203		
Hen-day egg production (%)	60.3 <sup>a</sup>	51.2 <sup>b</sup>	44.1 <sup>c</sup>	39.9°	3.23	< 0.001		
Mean egg weight (g)	48.5	48.3	49.3	47.9	0.68	0.246		
Mean egg mass (g)	29.5 <sup>a</sup>	24.4 <sup>b</sup>	21.5 <sup>b</sup>	18.9 <sup>b</sup>	1.47	< 0.001		
Feed/egg mass	3.04 <sup>a</sup>	3.88 <sup>b</sup>	4.34 <sup>b</sup>	4.96 <sup>c</sup>	0.298	< 0.001		
Mortality (%)	0.00	0.50	0.75	1.00	0.489	0.259		

 Table 2.30: Effect of SFYTM on performance of layers (19-35 weeks of age)

SED-standard error difference, P- probability, Means with the same superscripts in a row are not significantly different (P>0.05). Source: Mohammed and Dei (2013)

A study was undertaken to determine the effects of boiled false yam tuber meal on feed digestibility as well as egg laying performance of layer chickens. False yam tubers were peeled with a knife and chopped into small chips (~ 2 cm) and boiled in water (1part of fresh tuber to 2 parts of water) for 2 hours. The boiled tubers were rinsed with clean water to remove the sticky substances and sun-dried on a concrete floor for 7 days. The dried product was ground in a grinding mill into a gritty meal and labelled BFYTM. The boiled false yam tuber meal (BFYTM) replaced maize at different levels of 0, 50, 75 and 100 g/kg on weight by weight basis in a layer mash.

The results of feed digestibility indicated depressed digestibility of all nutrients considered except for dry matter digestibility which was not affected by the inclusion of BFYTM in the diets (Table 2.31). The inclusion of BFYTM above 50 g/kg (Table 2.31) in the diets of layer chickens significantly affected crude protein digestibility. This may probably be due to activities of antinutritional factors present in the BFYTM. These might bind with nutrients, thereby rendering them unavailable to the birds. According to McDonald *et al.* (2002), terpenes actually impair availability of nutrients and reduce performance of animals.

The similarity in feed intake of the experimental birds (Table 2.32) suggests that boiling the tuber has reduced the bitterness of the gum resins and making the diets more palatable to enhance feed



consumption. Dei *et al.* (2011) reported that boiling reduces the gum resins in the tuber by 39%, and thereby improving feed intake of birds. The addition of BFYTM to the diets progressively affected hen-day egg production and feed-to-egg mass ratio of birds. This could be due to the residual concentration of anti-nutritive factors (gum resins) still present in the tuber. According to McDonalds *et al.* (2002), terpenes actually interfere in availability of nutrients and reduce performance in animals. This might hinder availability of major nutrient (protein and fat) which are the major components of egg, hence reduction in efficiency of feed availability. In a previous study, Mohammed and Dei (2013) attributed the low egg production of layers fed diets containing soaked false yam tuber meal at similar levels tested to the effect of residual accumulation of anti-nutritive factors (terpenes), which affected protein and fat digestibility which are major components of eggs.

 Table 2.31: Effect of boiling on nutrients digestibility of false yam tuber meal fed to layer chickens

Dietary BFYTM levels (g/kg)								
Parameter	0	50	75	100	±SED	Р		
Dry matter (%)	75.6	74.5	74.2	73.1	1.57	0.509		
Crude protein (%)	58.2ª	57.8 <sup>a</sup>	49.5 <sup>b</sup>	50.0 <sup>b</sup>	2.94	0.015		
Crude fat (%)	79.8 <sup>a</sup>	83.8 <sup>b</sup>	77.1 <sup>a</sup>	77.9 <sup>a</sup>	1.41	0.002		
Ash (%)	48.4 <sup>a</sup>	55.5 <sup>b</sup>	59.5 <sup>b</sup>	56.3 <sup>b</sup>	2.89	0.014		

SED=Standard error of difference, P-probability, Means with the same superscripts in a row are not significantly different (P>0.05). Source: Nani *et al.* (2014)



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<u>kg)</u>		
100		
100	±SED	Р
92.8	1.57	0.078
43.1 <sup>c</sup>	3.54	0.003
47.8	0.45	0.093
21.2 <sup>c</sup>	1.95	0.008
4.48 <sup>c</sup>	0.34	0.008
2/60	-	-
	43.1° 47.8 21.2° 4.48°	92.8         1.57           43.1°         3.54           47.8         0.45           21.2°         1.95           4.48°         0.34

 Table 2.32: Effect of boiled false yam tuber meal (BFYTM) on performance of layer chickens

 (19-35 weeks of age)

SED=Standard error of difference, P-probability, Means with the same superscripts in a row are not significantly different (P>0.05). Source: Nani *et al.* (2014)

A study was conducted to evaluate the effect of soaking false yam seed in 0.1% saltpetre solution (SFYSM-SP) on feed digestibility, egg laying performance and physical egg characteristics of chickens. Freshly harvested false yam fruits were crushed open to remove the seeds. The seeds were then crushed into smaller pieces. The crushed seeds were soaked in 0.1% saltpetre solution (i.e. one part of fresh seed to two parts of saltpetre solution) for 12 days with the solution being changed every three days. The soaked seed was washed with fresh water after 12 days of soaking and sun-dried to a moisture level of about 12% and milled into a gritty meal and labelled SFYSM-SP. SFYSM-SP was substituted (w/w) for maize at four dietary levels (0, 80, 100 and 120 g/kg) in a concentrate-base layer diet.



The results of the study showed that there were no significant (P>0.05) differences in feed intake among the treatments (Table 2.32). There was slight depression (P<0.05) in hen-day egg production of all the hens fed the treated seed meal. However, there were no significant (P>0.05) differences among the treatments in terms of mean egg weight, egg mass and feed conversion efficiency. The similarity in feed consumption of all the birds suggests debittering of the gum resins by the saltpetre. This could be due to modification of the structure of the toxic compounds in the seed by oxidation. According to Pommer (2003), some terpenes are reactive towards oxidizing agents. Processing of the seed using saltpetre solution appears to improve its feed value, since egg laying performance of the hens tended to be similar (Table 2.34).

The outcome of this study is similar to that of Dei *et al.* (2013) who reported improvement in the utilization of false yam tuber up to 90 g/kg in broiler chicken diets when soaked in water and up to 120 g/kg in broiler chicken diets when soaked in 0.1% saltpetre solution.

The nominal reduction (P>0.05) in egg laying performance of hens fed the treated seed meal relative to the control group may suggest residual effect of the anti-nutritional factors. Anti-nutritional factors such as terpenes can actually impair the availability of nutrients and reduce performance in animals (McDonald et al., 2002).

Table 2.33: Effect of soaked false yam (Icacina oliviformis) seeds in 0.1% saltpetre solutionon apparent nutrient digestibility of ISA Brown laying chickens.

	ē .		• •						
Diets containing treated false yam seed meal									
Parameter	Control	80 g/kg	100 g/kg	120 g/kg	SED	Р			
		SFYSM-	SFYSM-	SFYSM-					
		SP	SP	SP					
Dry matter (%)	77.45	80.50	79.79	79.70	1.590	0.317			
Crude protein (%)	55.7	51.6	50.3	50.7	3.02	0.326			
Crude fat (%)	75.77 <sup>ac</sup>	78.06 <sup>a</sup>	83.78 <sup>b</sup>	73.05 <sup>c</sup>	1.733	0.002			
Ash (%)	65.4	65.2	65.5	63.9	3.14	0.952			

SED = Standard error of difference, P = Probability, means with the same superscripts within rows are not significantly different.

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Source: Mohammed et al. (2017a)

	Diets containing treated false yam seed meal						
Parameter	Control	80 g/kg	100 g/kg	120 g/kg	SED	Р	
		SFYSM-	SFYSM-	SFYSM-			
		SP	SP	SP			
Feed intake (g/b/d)	108.4	108.8	108.1	108.0	3.79	0.997	
Hen-day egg production (%)	76.0	75.4	73.7	70.3	3.93	0.506	
Mean egg weight (g)	59.5	60.1	58.8	60.1	1.35	0.743	
Mean egg mass (g)	53.7	53.9	51.6	50.3	2.03	0.300	
Feed-to-egg mass ratio	2.02	2.02	2.01	2.15	0.112	0.607	
Mortality	0.33	0.00	1.67	0.00	0.882	0.260	

Table 2.34: Effect of False Yam Seeds Soaked in 0.1% Saltpetre Solution on Egg Laying
Performance of ISA Brown Chickens (38–49 Weeks of Age).

SED = Standard error of difference, P = Probability. Source: Mohammed *et al.* (2017a)

Another study was undertaken to determine effect of fermented soaked false yam seed meal (FSFYSM) on egg laying performance of chickens. A freshly harvested false yam fruits were crushed open to remove the seeds. The seeds were crushed into smaller pieces.

The crushed seeds were soaked in water (i.e. one part of fresh seed to two parts of water) for 12 days with water being changed every three days. The soaked seeds were washed with fresh water after 12 days of soaking and then sun-dried to a moisture level of 12% and milled into a gritty meal. The soaked false yam seed meal (SFYSM) was fermented by mixing it thoroughly with water in a ratio of one part of SFYSM to one part of water, and placed in a plastic barrel (uncovered) for seven days. A wooden pole was used to stir the mixture on the third day when a mat was formed on the surface of the mixture. The fermented product (FSFYSM) was sun-dried and incorporated in the diets of layer chickens at 80, 100 and 120 g/kg.

Apparent nutrient digestibility of laying hens as affected by inclusion of FSFYSM is presented in Table 2.35. There were no significant (P>0.05) differences between hens fed the control diet and those hens fed diets containing FSFYSM in terms of apparent digestibility of dry matter, crude



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protein and ash. However, hens fed diets containing FSFYSM recorded a significant lower (P<0.001) fat digestibility than those hens fed the control diet.

The nominal reduction in protein digestibility indicates the presence of residual anti-nutritional factors in the FSFYSM-based diets. Anti-nutritional factors such as terpenes can actually impair the availability of nutrients and reduce performance in animals (McDonald et al., 2002). This suggests that soaking and aerobic fermentation only cannot completely eliminate toxic compounds found in the false yam seeds.

Egg laying performance of chickens fed diets containing aerobic FSFYSM is presented in Table 2.36. There were no significant (P>0.05) differences in all the parameters measured. This is an indication that fermentation had a positive impact on the soaked false yam seed meal. Previous studies by Mohammed and Dei (2012) indicated that soaking the false yam seed could only be tolerated by laying hen up to 100 g/kg, replacing maize in layer diets.

However, additional processing by fermentation has allowed replacement up to 120 g/kg. This could be attributed to the reduction in the anti-nutritive factor in the false yam seed which was identified as terpenes (Vanhaelen et al., 1986). Reddy and Pierson (1994) reported that fermentation is a widely practiced form of food processing that is effective in removing anti-nutritional components from cereals and legumes while improving their digestibility and sensory characteristics.



Essers and Nout (1989) stated that covering fresh cassava with leaves for a number of days will encourage mould to grow prior to sun drying thereby producing air fermented cassava products. The microflora of the mould fermentation commonly includes *Neurospora sitophila*, *Geotrichum candidum*, *Rhizopus oryzae*, Aspergillus, fumagatus and *Mucor racemosis* (Essers, 1994). Essers (1994) has shown that the processing involving fermentation is able to reduce initial levels of cyanogen from 231–559 mg CN equivalents/kg dry weight to 8–41 mg CN equivalents/kg dry weight in the final flour. That is about 96% reduction in CN content.

The similarities observed in this study in terms of production performance can be attributed to the positive effect of aerobic fermentation of the SFYSM. The toxins (terpenes) that are reported to contain in the seed could have further reduced due to the activities of the micro flora present during the fermentation process, thereby enhancing its nutritive value for the laying hens in this study.

 Table 2.35: Effect of fermented soaked false yam (Icacina oliviformis) seed meal on apparent nutrient digestibility of ISA Brown layer chicken

Parameter (%)	Control	80 g/kg	100 g/kg	120 g/kg	±SED	Р
		FSFYSM	FSFYM	FSFYSM		
Dry matter	77.98	75.77	77.14	77.62	1.330	0.418
Crude protein	71.84	67.99	68.18	68.59	1.789	0.185
Crude fat	86.61 <sup>a</sup>	74.07°	79.62 <sup>b</sup>	77.43 <sup>b</sup>	1.266	< 0.001
Ash	74.78	74.92	74.71	74.04	1.141	0.867

SED = Standard error of difference, P= Probability, means with the same superscripts are not statistically different. Source: Mohammed *et al.* (2017b)

Table 2.36: Effect of fermented false yam (Icacina oliviformis) seed meal on egg laying
performance ISA Brown layers (29–41 Weeks of Age).

Parameter	Control	80 g/kg	100 g/kg	120 g/kg	±SED	Р
		FSFYSM	FSFYSM	FSFYSM		
Feed intake (g/b/d)	117.4	120.0	118.7	117.9	3.27	0.862
Hen-day egg production (%)	89.9	86.4	85.7	86.5	3.31	0.602
Mean egg weight (g)	60.29	60.82	57.39	58.87	2.014	0.380
Mean egg mass (g)	52.7	51.8	49.0	50.6	3.60	0.750
Feed-to-egg mass ratio	2.23	2.34	2.43	2.36	0.186	0.765
mortality	0.67	0.33	0.33	0.33	0.624	0.931

SED = Standard error of difference, P = Probability. Source: Mohammed *et al.* (2017b)



#### 2.9.4 Processed seed meals and their nutritional value for guinea fowls

So far, only the processed false yam seed meal has been evaluated in diets for guinea fowls. Soaking false yam seed in water for 12 days improved its nutritive value and can be fed up to 150 g/kg in the diet of local guinea fowls without adverse effect on their growth performance (Table 2.37). Also, boiling the seed for 2 hours can be added to the diets of local guinea fowls up to 150 g/kg without any adverse effects on their growth performance (Table 2.38). However, feeding the boiled seed meal beyond 50 g/kg in the diet significantly increased feed cost (Table 2.38).

Table 2.37. Effect of processed false yam seed meals on daily feed intake, weight gain and feed-to-gain ratios in growing local guinea fowls.

Processed	Treatments	Feed intake	Weight gain	Feed/gain	Feed
products	(g/kg)	(g/bird/day)	(g/bird/day)	ratio	cost(GH¢/bird)
Diets	Control (0)	52.1	8.50	0.16	5.59
containing	50	49.5	7.84	0.16	5.64
SFYSM	100	49.0	7.84	0.16	6.50
	150	47.3	6.77	0.14	7.07
	SED	4.73	0.89	0.01	0.76
	P-value	0.604	0.165	0.452	0.151
Diets	Control (0)	65.2	9.0	0.14	6.72 <sup>a</sup>
containing	50	66.4	7.8	0.12	7.67 <sup>a</sup>
BFYSM	100	62.0	6.4	0.11	10.18 <sup>b</sup>
	150	65.1	5.9	0.09	11.40 <sup>b</sup>
	SED	2.79	1.65	0.03	1.18
	P-value	0.489	0.197	0.082	0.017

SED-Standard error of difference, Means with the same superscript within a column are not significantly different (P>0.05), SFYSM-Soaked false yam seed meal (12 days soaking fed from 5-18 weeks of age), BFYSM-Boiled false yam seed meal (2 hours boiling fed from 9-19 weeks of age) (Dei et al., 2015)

#### 2.9.5 Processed tuber meals and their nutritional value for pigs

Feeding of soaked false yam tuber meal up to 100 g/kg in Ashanti Black pig weaner diets had no adverse effects on their growth performance and that it was economical to use as substitute for maize (Table 2.38). Similarly, the boiled false yam tuber meal when fed at 50 g/kg in the diet had no adverse effect on growth performance and economics of feeding (Table 2.38).

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However, the raw false yam tuber meal had adverse effects on both pig growth performance and cost of feeding, therefore should not be used (Table 2.38). Another interesting observation from this study was that processing the false yam tuber by soaking in water appears to be an effective method of processing the tuber for pigs.

Processed	Treatments	Initial	Weight gain	Feed intake	Feed/gain
products	(g/kg)	weight	(g)	(g)	ratio
		(kg)			
Diets	Control (0)	6.19	306 <sup>a</sup>	803 <sup>a</sup>	2.62 <sup>a</sup>
containing	50	5.21	217 <sup>b</sup>	548 <sup>b</sup>	2.53 <sup>a</sup>
SDFYTM	100	5.18	135°	440 <sup>c</sup>	3.43 <sup>b</sup>
	150	5.29	93°	390°	4.49 <sup>b</sup>
	SED	0.78	22.9	41.4	0.61
	P-value	0.538	<0.001	<0.001	0.040
Diets	Control (0)	6.19	306 <sup>a</sup>	803	2.62
containing	50	5.31	261 <sup>a</sup>	689	2.48
SFYTM	100	5.43	263 <sup>a</sup>	721	2.62
	150	4.63	213 <sup>b</sup>	601	2.60
	SED	0.63	23.4	64.4	0.08
	P-value	0.186	0.027	0.075	0.256
Diets	Control (0)	6.19	307 <sup>a</sup>	803 <sup>a</sup>	2.62 <sup>a</sup>
containing	50	5.34	284 <sup>a</sup>	680 <sup>a</sup>	2.38 <sup>b</sup>
BFYTM	100	5.10	203 <sup>b</sup>	553 <sup>b</sup>	2.76 <sup>b</sup>
	150	5.09	83°	440 <sup>b</sup>	5.16 <sup>a</sup>
	SED	0.58	23.1	84.1	0.64
	P-value	0.261	<0.001	0.013	0.008

Table 2.38: Effect of processed false yam tuber meals on daily feed intake, weight gain and feed-to-gain ratios of Ashanti black weaner pigs (12-20 weeks of age)



SED-Standard error of difference, Means with the same superscripts within a column are not significantly different (P>0.05), SDFYTM-Sun dried false yam tuber meal, SFYTM-soaked false yam tuber meal, BFYTM-Boiled false yam tuber meal (Dei et al., 2013).

# 2.9.6 False yam tuber and leaf meals and their nutritional value for rabbits

So far, false yam leaf and tuber meals have been evaluated in diets for rabbits. It has been shown

that false yam leaf as an ingredient in weaner rabbits' diet up to 50 g/kg improved feed intake and

weight gain of rabbits (Table 2.39).

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In another experiment, false yam tuber meal were evaluated on the growth performance of weaner rabbits. The false yam tuber was chopped into pieces and soaked in water for 12 days, water changed every 3 days, sun-dried and milled into gritty floor which replaced maize (w/w) in the diets of rabbits at 100, 150 or 200 g/kg. Table 2.40 shows the results of the effect of soaked false yam tuber meal on the growth performance of weaner rabbits. The results indicated that soaking false yam tuber in water for 12 days improved its nutritive value and could replace maize up to 200 g/kg in the diets of weaner rabbits without adverse effect on growth performance. According to Roessler *et al.* (2017), soaking false yam tuber in water has the potential of leaching out hydrophilic compounds, but not those that are lipophilic in nature. Ologhobo *et al.* (1993) reported that, in chemical treatments of seeds, higher concentration of anti-nutritional factors were found in base-soluble fractions (E.g. NaOH) than others, indicating a greater extractability of anti-nutritional factors by alkali treatment than by acid, ether or alcohol.

In view of these statements, processing of false yam tuber involving water and chemical treatment was evaluated where false yam tuber was chopped, soaked in water for 12 days, and then soaked in 1M concentration of sodium hydroxide (NaOH) solution for 24 hours, washed clean and sundried and milled. The product replaced maize (w/w) in weaner rabbit diets at 100, 150 or 200 g/kg. Growth performance results showed similar performance irrespective of the inclusion level, suggesting that the product was as similar as maize in terms of palatability and nutrition (Table 2.41).

In a similar experiment, processing the tuber with sodium hydroxide was replaced with potassium hydroxide. The new product (potassium hydroxide treated false yam tuber meal) replaced maize at similar levels as in sodium hydroxide treated false yam tuber meal. Growth performance results

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indicated that the new product was as similar as maize and could replace maize up to 200 g/kg in

weaner rabbit diets (Table 2.42).

# Table 2.39: Effect of false yam leaf meal on growth performance of weaner rabbits (8-16 weeks of age)

	Levels of fals	se yam leaf mea	<u>ul (g/kg)</u>		
Parameters	Control (0)	50	100	SED	
Feed intake (g/day)	125.1 <sup>b</sup>	128.7ª	129.3ª	0.72	
Weight gain (g/day)	17.65 <sup>a</sup>	13.33 <sup>ab</sup>	10.83 <sup>b</sup>	2.32	
Gain-to-feed ratio	0.14 <sup>a</sup>	0.10 <sup>ab</sup>	0.08 <sup>b</sup>	0.02	

SED: Standard Error of Difference, Means with the same superscripts in a row are not significantly different (P>0.05). Source: Ayambire (2011)

Table 2.40: Effect of soaked false yam	tuber meal or	n the growth	performance of weaner
rabbits (7-15 weeks of age)			

Inclusion levels of soaked false yam							
		tuber meal (g/kg)					
Parameter	0	100	150	200	SED	Р	
Initial live-weight (g)	729	728	731	730	95.6	1.000	
Final live-weight gain (g)	1264	1116	1241	1139	74.5	0.177	
Average daily feed intake (g)	53.1	50.7	56.5	51.3	2.97	0.251	
Average daily weight gain (g)	9.55	6.94	9.11	7.30	2.158	0.558	
Feed conversion ratio	6.0	8.1	6.5	10.4	3.47	0.596	
		4 4 4 4 4	~ .	• (2)	0.4.0		

SED= Standard Error of Difference, P= Probability. Source: Azupio (2018)

 Table 2.41: Effect of NaOH-treated false yam tuber meal on the growth performance of weaner rabbits (8-16 weeks of age)



	Inclusion	levels of	of NaOl	H-treated		
	false yam tuber meal (g/kg)					
Parameter	0	100	150	200	SED	Р
Initial live-weight (g)	758.8	788.8	772.5	768.8	90.80	0.990
Final live-weight gain (g)	1405	1406	1406	1535	55.8	0.094
Average daily feed intake (g)	54.2	60.0	59.1	60.4	4.73	0.551
Average daily weight gain (g)	11.54	11.03	11.32	13.68	1.766	0.451
Feed conversion ratio	4.77	5.50	5.43	4.80	0.897	0.763

SED= Standard Error of Difference, P= Probability. Source: Eric (2018)

Inclusion levels of KOH-treated						
	false yam tuber meal (g/kg)					
Parameter	0	100	150	200	SED	Р
Initial live-weight (g)	996	990	948	960	103.6	0.957
Final live-weight gain (g)	1411	1382	1241	1392	87.6	0.245
Average daily feed intake (g)	50.6	50.2	50.9	58.7	5.27	0.355
Average daily weight gain (g)	7.41	7.01	5.25	7.72	1.708	0.497
Feed conversion ratio	6.87	8.54	9.67	8.96	2.397	0.694

 Table 2.42: Effect of KOH-treated false yam tuber meal on the growth performance of rabbits (8-16 weeks of age)

SED= Standard Error of Difference, P= Probability. Source: Lester (2018)

#### 2.10.0 Effects of false yam products on haematological profile of animals

According to Anderson and Anderson (2002), haematology serves as an excellent medium for the measurement of potential biomarkers, because its collection is relatively non-invasion and it emphasizes an enormous range of physiological processes in the body at any given time. Haematological analysis are significant in nutritional studies since blood, metabolites and their concentration provide information for indirect assessment (Harper *et al.*, 1979).

# 2.10.1 Chickens



In an investigation to ascertain the nutritive value of false yam (*Icacina oliviformis*) tuber meal for broiler chickens, inclusion of sun-dried and boiled false yam tuber (*Icacina oliviformis*) meals in the diets of broiler chickens did not affect the haematological profile of the birds. No adverse changes in blood haemoglobin, haematocrit, and white blood cells were observed (Table 2.43). Dei *et al.* (2013a) indicated that haematological parameters of broilers did not vary when fed diets containing soaked false yam tuber meal. Agyemang (2010) also indicated that the haematological indices of broilers fed processed (soaking and cooking) false yam flour meal did not show negative effects on the blood characteristics of the broilers (Table 2.43).

Processed product	Treatments (g/kg)	RBC count $(10^6 \text{mm}^3)$	WBC count $(10^6 \text{mm}^3)$	Hb(g/dl)	Haematocrit (%)	Source
diets containing	control (0)	3.97	4.80	9.93	29.67	
sdfytm	30	3.90	4.33	10.10	30.17	
	60	3.70	5.20	10.00	30.08	dei et al.,
	90	3.87	5.23	9.40	28.50	2011a
	sem	0.144	0.377	0.353	1.037	
	р	ns	ns	ns	ns	
Diets containing	Control (0)	4.30	5.13	10.90	32.69	
SFYTM	30	4.10	4.77	10.47	31.17	Dei et al.,
	60	3.90	5.47	10.13	29.83	2013e
	90	3.93	4.83	10.20	30.00	
	SEM	0.262	0.978	0.706	1.425	
	Р	NS	NS	NS	NS	
Diets containing	Control (0)	3.88	4.76	9.97	30.00	
BFYTM	30	3.97	4.80	10.20	30.33	
	60	3.78	5.60	9.77	29.17	Dei et al.,
	90	4.02	4.80	10.23	30.50	2011a
	SEM	0.134	0.330	0.300	0.886	
	Р	NS	NS	NS	NS	
Diets containing	Control (0)	3.77	4.81	9.67	29.00	
SBFYTFM	30	4.13	4.32	10.57	31.67	
	60	3.92	5.20	10.05	30.17	Agyemang,
	90	3.78	5.23	9.72	29.17	2010
	SEM	0.288	0.377	0.712	2.122	
	Р	NS	NS	NS	NS	

# Table 2.43: Blood constituents of broiler chickens fed varying levels (0, 30, 60 and 90 g/kg) of SDFYTM, SFYTM, BFYTM and SBFYTFM in their diets from 21-56 days of age.

SDFYTM-sun-dried false yam tuber meal, SFYTM-soaked false yam tuber meal, BFYTM-boiled false yam tuber meal, SBFYTFM-soaked and boiled false yam tuber flour meal, RBC-Red blood cell, WBC- White blood cell, Hb- haemoglobin, NS-Not significant (P>0.05), P-probability, SEM-standard error of means.

Studies on the effect of raw and processed false yam seeds on the haematology of broilers indicated that raw and processed false yam seed meals can be substituted for maize in the diets of broilers up to 50 g/kg and 100 g/kg, respectively without any significant negative effects on their haematology (Table 2.44).



Processed product	Treatments (g/kg)	RBC count (10 <sup>6</sup> mm <sup>3</sup> )	WBC count (10 <sup>6</sup> mm <sup>3</sup> )	Hb (g/dl)	Haematocrit (%)
Diet containing	Control (0)	4.67	-	11.87	35.5
SDFYSM	50	4.47	-	11.37	34.0
	75	4.60	-	11.70	35.0
	100	4.50	-	11.47	34.3
	SEM	0.240	-	0.606	1.840
	Р	NS	-	NS	NS
Diet containing	Control (0)	4.67	-	11.87	35.5
SFYSM	50	4.37	-	10.73	32.17
	75	4.87	-	12.43	36.83
	100	4.80	-	11.77	35.17
	SEM	0.392	-	1.034	3.266
	Р	NS	-	NS	NS
Diet containing	Control (0)	4.67	-	11.87	35.50
BFYSM	50	4.30	-	10.97	32.83
	75	4.20	-	10.70	36.00
	100	4.67	-	11.90	35.67
	SEM	0.438	-	1.092	3.282
	Р	NS	-	NS	NS

Table 2.44: Blood constituents of broiler chickens fed varying levels (0, 50, 75 and 100 g/kg)
of SDFYSM, SFYSM, and BFYSM in their diets from 21-56 days of age

SDFYM-sun-dried false yam seed meal, SFYSM-soaked false yam seed meal, BFYSM-boiled false yam seed meal, RBC-Red blood cell, WBC- White blood cell, Hb- haemoglobin, NS-Not significant (P>0.05), P-probability, SEM-standard error of means (Okyere, 2011).

# 2.10.2 Rabbits

In the evaluation of false yam (Icacina oliviformis) leaf meal as an ingredient in the diet of weaner

rabbits (Oryctolagus cuniculus), it has been reported that, there were no significant differences

(P>0.05) in haemoglobin (Hb) concentration, packed cell volume (PCV) and red blood cells.



Erythrocytes values were all within the normal ranges for rabbits (Table 2.45).

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False yam product	Treatments	RBC count	WBC count	Hb	Haematocrit (%)
	(g/kg)	(106/mm <sup>3</sup> )	(106/mm <sup>3</sup> )	(g/dl)	
Diets containing	Control (0)	4.99	8.93	12.80	38.40
FYLM	50	4.56	7.44	11.62	34.80
	100	4.28	9.84	10.92	32.80
	SEM	0.82	3.46	2.04	6.12
	Р	NS	NS	NS	NS

Table 2.45: Blood constituents of rabbits fed varying levels (0, 50 and 100 g/kg) of false yam leaf meal (8-16 weeks of age)

RBC-Red blood cell, WBC- White blood cell, Hb- haemoglobin, FYLM-False yam leaf meal, NS -Not significant (P>0.05), P-probability, SEM-standard error of means. Source: Ansah and Aboagye (2011)

Haematological indices are an index and a reflection of the effects of dietary treatments on the animal in terms of the type, quality and amounts of the feed ingested, and were available for the animal to meet its physiological, biochemical and metabolic necessities (Ewuola et al., 2004). Reports by Aletor (1989) indicate that blood variables most consistently affected by dietary influence include red blood cell (RBC), pack cell volume (PCV), plasma protein and glucose. Form the results of feeding weaner rabbits with diets containing soaked false yam tuber meal (SFYTM) presented in Table 2.46, the PCV values (37.3-41.9%) were slightly above the range of values (31-38%) reported by Shah *et al.* (2007). This suggests that soaking in water as a method of processing was good enough for detoxification of false yam tuber for rabbits. Low PCV is an index of toxicity reduction in the blood and usually suggests presence of a toxic factor which has adverse effect on blood formation (Oyawoye and Ogunkunle, 1998). Values of the Red blood cells was significantly increasing as the SFYTM was increasing in the diet of the rabbits contrary to the findings of Ewuola and Egbunike (2008). The total white blood cell size is an indication of animals' ability to fight against diseases. The rabbits fed diets containing 200g/kg had high count



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of total white blood cells, indicating that they were more resistance to disease than other rabbits

especially the control rabbits.

# Table 2.46: Effect of soaked false yam tuber meal (SFYTM) on the haematological parameters of weaner rabbits (7-15 week of age)

Inclusion levels of soaked false						
		yam tube	r meal (g/	'kg)		
Parameter	0	100	150	200	SED	Р
Red blood cells $(10^{12}/l)$	5.3 <sup>b</sup>	5.6 <sup>ab</sup>	5.5 <sup>b</sup>	6.1 <sup>a</sup>	0.19	0.013
Haemoglobin (g/dl)	13.6	11.2	11.3	11.7	1.16	0.206
Packed cell volume (%)	37.3	39.7	37.7	41.9	2.40	0.272
Mean corpuscular volume (f/l)	70.3	70.7	69.3	68.6	2.54	0.837
Mean corpuscular haemoglobin (pg)	25.5ª	19.8 <sup>b</sup>	18.9 <sup>b</sup>	19.1 <sup>b</sup>	1.74	0.016
Mean corpuscular haemoglobin conc. (g/dl)	36 <sup>a</sup>	28 <sup>b</sup>	27 <sup>b</sup>	28 <sup>b</sup>	1.5	0.001
Total white blood cells $(10^9/l)$	3.92 <sup>b</sup>	5.17 <sup>b</sup>	5.60 <sup>ab</sup>	7.24 <sup>a</sup>	0.591	0.003
White blood cell differentials (%)						
Lymphocytes	13.8	17.8	11.1	15.1	2.85	0.210
Monocytes	11.5 <sup>b</sup>	11.5 <sup>b</sup>	13.4 <sup>ab</sup>	17.9 <sup>a</sup>	1.76	0.02
Neutrophils	73.8	70.3	73.7	70.9	2.95	0.528
Eosinophils	0.53	0.38	0.39	0.39	0.061	0.106
Basophils	0.34	0.67	0.68	0.74	0.169	0.15

SED= Standard Error of Difference, P= Probability, Means with the same superscripts in a row are not significantly different (P>0.05). Source: Sam (2018)



Haematological parameters of weaner rabbits fed diets containing sodium hydroxide-treated false yam tuber (NaOH-FYTM) were evaluated and results presented in Table 2.47. The results of the study indicated that, the red blood cells (RBC) and haemoglobin (Hb) values were within the normal ranges for rabbit as reported by Benson and Paul-Murphy (1999), suggesting that dietary protein quality was not altered. This was explained in the works of Gilani *et al.* (2012), who demonstrated that some plant ANFs interfere with the quality of dietary protein and consequently affect red blood cells formation.

The relatively low park cell volume (PCV) values compared to those reported by Brewer, (2006) and Melillo (2007)(31-50%) suggest some levels of residual toxins in the processed false yam tuber. This statement is in agreement with Oyawoye and Ogunkunle (1998) that PCV depicts an index of toxicity and that low PCV concentrations in the blood usually would suggest presence of a toxic factor (e.g. haemagglutinins). Another researcher confirmed this assertion; that most plant species contain haemagglutinins (Sharon and Lis, 2004). It is possible that traces of haemagglutinins were still present in the processed tuber meal that may be responsible for the reduced PCV values.

The values obtained for the white blood cells (WBC) in this study were within the normal ranges  $(5.2-12.5 \times 10^6 \text{m}^3)$  as reported by Benson and Paul-Murphy (1999). This suggests that the processed false yam tuber meal had no negative influence. Hrapkiewicz and Medina (2007), revealed that high concentration of WBC in the blood indicates microbial infection or the presence of foreign body or antigen in the circulating system.



The values of mean corpuscular haemoglobin (MCH) and mean corpuscular volume (MCV) in this study were found to be within the stipulated ranges (17.1–23.9pg, and 28.2–37g/dl respectively), this shows that the experimental rabbits were not anaemic. This agrees with Washington and Van Hoosier (2012) who established that low values of MCH, MCV and MCHC are markers of anaemia. The relatively lower values of eosinophils, basophils, neutrophils and monocytes in this study compared to the reference ranges (0-5%, 0–10%, 20–75% and 1–10%) reported by Hrapkiewicz and Medina (2007) and Wilson *et al.* (2012) respectively and signify a good health.

Inclusion levels of NaOH-							
treated false yam tuber meal							
		(g/	kg)				
Parameter	0	100	150	200	SED	Р	
Red blood cells $(10^{12}/l)$	6.9	7.1	6.9	6.9	0.09	0.177	
Haemoglobin (g/dl)	13.6	13.7	13.5	13.6	0.29	0.917	
Packed cell volume (%)	29.7	30.4	29.9	29.8	0.67	0.748	
Mean corpuscular volume (f/l)	43.4	43.2	42.1	43.5	0.87	0.409	
Mean corpuscular haemoglobin (pg)	19.7	20.0	19.5	19.8	0.33	0.584	
Mean corpuscular haemoglobin conc.	46.0	46.0	46.5	46.0	0.46	0.633	
(g/dl)							
Total white blood cells $(10^{9}/l)$	6.7	6.3	6.8	10.2	2.61	0.312	
White blood cell differentials (%)							
Lymphocytes	24.7 <sup>ab</sup>	22.1 <sup>b</sup>	28.9 <sup>ab</sup>	39.2 <sup>a</sup>	4.64	0.027	
Monocytes	6.5	4.9	6.2	3.5	1.67	0.314	
Neutrophils	65.7	70.6	63.2	63.1	4.39	0.339	
Eosinophils	1.01	0.80	0.83	0.72	0.185	0.492	
Basophils	2.1	1.7	0.9	1.65	0.80	0.531	

Table 2.47: Effect of sodium hydroxide-treated false yam tuber meal on the haematological
parameters of weaner rabbits (8-16 week of age)

SED= Standard Error of Difference, P= Probability, Means with the same superscripts in a row are not significantly different (P>0.05). Source: Eric (2018)

In another experiment, weaner rabbits were fed diets containing potassium hydroxide-treated false



yam tuber meals (KOH-FYTM) and the results presented in Table 2.48. Form the results, all the haematological parameters measured were within the normal physiological ranges reported for rabbits; haemoglobin, packed cell volume, red blood cell, white blood cell, neutrophils, lymphocytes and eosinophils (Jenkins, 1993; Hillyer, 1994). Madubuike and Ekenyem (2006) indicated that it is evident that haematological characteristics of livestock suggest their physiological disposition to the impact of nutrition. It may then be suggested that, the different diets imposed on the rabbits were balanced in their formulation to support relatively high performance and maintain the normal haematological profile of the rabbits. The values of haemoglobin, packed cell volume, red blood cell and total white blood cell reported in this study

were generally higher when compared to values reported by other researchers (Ahamefule et al., 2006; Yakubu et al., 2008; Ahamefule et al., 2008). Even though there was a significant difference in packed cell volume, haemoglobin and red blood cells levels, this difference was positive for rabbits fed potassium hydroxide-treated false yam tuber meal-based diets as these parameters increased marginally on rabbits fed the potassium hydroxide-treated false yam tuber meal-based diets. High values may apparently mean an increase in the circulation of red blood cells or an increase in plasma volume (Frandson 1986). Njidda and Hambagda (2006) indicated that PCV, RBC and Hb are the most dependable blood features for assessing the health status of animals. The findings in this study therefore agrees with Ansah and Aboagye (2011) who concluded that, there were no negative effects of processed false yam leaf meal on the weaner rabbit's haematology since all parameters fell within the normal ranges reported for rabbits.

 Table 2.48: Effect of potassium hydroxide-treated false yam tuber meal on the haematological parameters of rabbits (8-16 week of age)

	Inclusion	Inclusion levels of KOH-treated false				
	yam tuber meal (g/kg)					
Parameter	0	100	150	200	SED	Р
Red blood cells $(10^{12}/l)$	5.53 <sup>b</sup>	5.50 <sup>b</sup>	6.05 <sup>a</sup>	6.10 <sup>a</sup>	0.206	0.031
Haemoglobin (g/dl)	11.25 <sup>b</sup>	11.20 <sup>b</sup>	12.60 <sup>a</sup>	12.75 <sup>a</sup>	0366	0.004
Packed cell volume (%)	43.60 <sup>c</sup>	42.50 <sup>c</sup>	48.95 <sup>b</sup>	54.20 <sup>a</sup>	1.161	< 0.001
Mean corpuscular volume (f/l)	79.0	81.0	81.2	82.3	6.87	0.854
Mean corpuscular haemoglobin (pg)	20.40	21.60	20.85	21.40	0.850	0.519
Mean corpuscular haemoglobin conc.	25.85	25.65	25.70	25.80	0.307	0.909
(g/dl)						
Total white blood cells $(10^9/l)$	10.43	10.33	11.03	10.56	1.731	0.977
White blood cell differentials (%)						
Lymphocytes	70.3	70.0	66.1	65.1	3.64	0.411
Monocytes	7.40	7.40	6.75	6.75	0.843	0.759
Neutrophils	17.95	19.40	21.60	21.60	2.404	0.401
Eosinophils	0.10	0.05	0.10	0.05	0.050	0.596
Basophils	2.20	2.25	1.95	1.95	0.399	0.810

SED= Standard Error of Difference, P= Probability, Means with the same superscripts in a row are not significantly different (P>0.05). Source: Lester (2018).



# 2.11.0 INFERENCES FROM LITERATURE REVIEW

- The false yam plant is indigenous to West and Central Africa and is found growing wild on light sandy soil in the savanna areas of Senegal, Gambia, Guinea, the northern part of Ghana and parts of Sudan.
- The false yam can be used as food for humans and animals and has medicinal properties such as analgesic, anti-inflammatory and anti-diabetic activities and antimicrobial activities.
- Nutrient composition and amino acid profiles of false yam tuber and seeds are generally lower than that of maize.
- False yam tuber and seeds as alternative feed ingredients contain anti-nutritional factors that limit their utilization in poultry diets.
- Processing the tuber and seeds of false yam to feed poultry has achieved a varying degree of success depending on the method and nature of additives used.
- Acceptable inclusion levels of these alternative feed ingredients in poultry diets vary depending on the method of processing given the nature of anti-nutritional factors present.
- Anti-nutritional factors have been found to have significant negative influence on nutrient digestibility, animal productive performance, as well as blood chemistry of animals, particularly poultry.
  - There is room for improving the nutritive value of false yam seeds using chemical treatment methods.



#### **CHAPTER THREE**

#### **3.0 GENERAL MATERIALS AND METHODS**

#### 3.1 Sample collection

Matured fruits of false yam plants growing in the wild around Nyankpala Campus of the University for Development Studies, Tamale, were harvested by hand picking. The fruits were partially sundried (5 days) to facilitate cracking to obtain the seeds. After cracking the false yam seeds (FYS) were partially sun-dried (7 days) to reduce their moisture content. Afterwards, the FYS were crushed to reduce size and increase surface area to facilitate processing.

#### 3.2 Sample processing

Five samples of false yam seed meal (FYSM) were prepared as follows:

Freshly crushed FYS were sun-dried to approximately 12% moisture content on a cement floor. The remaining four seed samples were subjected to multiple-stage processing where each seed sample was first soaked in ordinary water (i.e., addition of fresh seeds in ordinary water at a ratio of 1:2, wt./vol.) for 12 days, with water changed every 3 days. After the 12 days of soaking, the seed samples were washed with clean ordinary water. In the second stage of processing, each soaked FYS sample was soaked in a solution of 1M concentration (i.e., addition of soaked seeds into 1M concentration at a ratio of 1:2 wt./vol.) of an industrial chemical additive (either urea, sodium chloride, sodium hydroxide or potassium hydroxide) for 24 hours, after which all the samples were washed thoroughly with clean ordinary water. The last stage of processing involved blanching of all samples that had been soaked in the chemicals. In this process, seed samples were immersed in hot water (90°C) for 20 minutes and then transferred into cold water (4°C) for 40 minutes.



The samples were then washed with clean ordinary water, sun-dried on a cemented floor to a moisture content of about 12%. Maize was bought from Tamale maize market in a single batch.

## 3.3 Sample milling

The processed false yam seed samples were ground into gritty flour using a hammer mill (2 mm screen size). The processed false yam seed meals were labeled as follows: sun-dried=Un\_T; urea treated=Urea\_T; sodium chloride treated=NaCl\_T; sodium hydroxide treated=NaOH\_T and potassium hydroxide treated=KOH\_T. Maize served as control.

#### 3.4.0 Proximate analysis

Proximate components were determined for maize, untreated FYSM and the chemically treated FYSM.

#### 3.4.1 Moisture content determination

The basic method of AOAC (2000) was used. Cleaned crucibles were dried in the oven at  $100^{\circ}$ C to obtain a constant weight and then cooled in the desiccator. Two grams each of the samples was weighed into the crucible and dried in an Ov150SF oven at 100°C until a fairly constant weight was obtained. Percentage moituren was calculated as follows:

% Moisture content = 
$$\begin{bmatrix} W & 2 - W \\ W & 2 - W \end{bmatrix}$$
 x 100 = Equation 3.1

- W1 = Initial weight of empty crucible
- W2 = Weight of dish + sample before drying



W3 = Weight of dish + sample after drying.

#### 3.4.2 Ash determination

The AOAC (2000) method was used.

Two grams (2g) of finely ground sample was weighed into a preheated cooled crucible. This sample was charred on a Bunsen flame inside a fume cupboard.

Sample was transferred into a pre-heated OH85TR muffle furnace at 550<sup>o</sup>C for 3hrs until a white or light grey ash was obtained. It was cooled in a desiccator and then weighed.

=Equation 3.2

The ash content was calculated as follows:

% ash = 
$$\begin{bmatrix} W3-W1 \\ W2-W1 \end{bmatrix} \times 100$$

Where

W1 = weight of empty crucible

W2= Weight of crucible +sample before ashing

W3 = Weight crucible +ash (sample after ashing)



# 3.4.3 Crude protein determination

The protein content of the samples was determined according to the standard methods of AOAC (2000) using the Kjeldahl method which involved digestion, distillation and titration.

# **Digestion of Sample**

Two grams (2g) of sample was weighed into NAAFCCO kjeldahl flask. Five grams of anhydrous sodium sulphate as catalyst was added. Twenty five milliliters (25ml) conc. H<sub>2</sub>SO<sub>4</sub> was added with

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few boiling chips. It was heated in the fume chamber until solution became clear. The solution was cooled to room temperature after which it was transferred into a 250ml volumetric flask and made up to the level with distilled water.

### Distillation

The distillation unit was cleaned and the apparatus set up. A 100ml conical flask, (receiving flask) containing 5ml of 2% boric acid was placed under the condenser with the addition of few drops of methyl red indicator. Five milliliters (5ml) of the digest was poured into the apparatus through the small funnel, washed down with distilled water followed by addition of

5ml of 60% NaOH solution. The digestion flask was heated until 100ml of distillate was collected in the receiving flask.

#### Titration

The solution in the receiving flask was titrated with 0.049M H<sub>2</sub>SO<sub>4</sub> to get pink colour. The same procedure was carried out on the blank (with filter paper only).

Calculation

% Nitrogen of sample (% N) = Titre x Normality of acid x  $14g \times 100$  =Equation 3.3

Weight of sample x mole

Titre = titre of sample - titre of blank



14g = atomic weight of nitrogen

Mole = mole  $H_2SO_4$ =mole  $NH_3$ =mole N in the sample

% Crude protein = % N x 6.25

#### 3.4.4 Fat determination

The method of AOAC (2000) was used. A Soxhlet extractor with a reflux condenser and a 250ml round between flask was fixed. Two grams (2g) sample was weighed into a thimble. Petroleum ether (300ml) was filled into the round bottom flask. The extractor thimble was sealed with cotton wool. The Soxhlet apparatus was allowed to reflux for about 6 hours. The thimble was removed with care and petroleum ether collected in the top and drained into a container for re-use. When the flask was free of ether, it was removed and dried at 105°C for 1h in an oven, then cooled in a desiccator and weighed.

## 3.4.5 Crude Fibre determination

The method of AOAC (2000) was used. Petroleum ether was used to defat 2g of sample. This was put in boiling 200ml of 1.25% H<sub>2</sub>SO<sub>4</sub> and boiled for 30minutes. The solution was filtered through muslin cloth on a fluted funnel. It was washed with boiling water until it was free of acid. The residue was returned into 200ml boiling NaOH and allowed to boil for 30 minutes. It was further washed with 1% HCl and boiling water, to free it of acid. The final residue was drained and transferred to silica ash crucible (porcelain crucible) dried in OV150SF oven to a constant weight, cooled and weighed.

% crude fibre = loss in weight after ignition (drying) x 100 = Equation 3.5

weight of sample

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#### 3.4.6 **Determination of carbohydrate**

The standard method of AOAC (2000) was used and the carbohydrate content of the samples was determined by difference as follows.

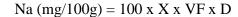
% carbohydrate = 100 - (% moisture + % ash + % protein + % fat + % crude fibre) = Equation 3.6

#### 3.5.0 Determination of minerals

Mineral content of the samples were determined by dry ashing and extraction method. Two grams of sample was ashed in a muffle furnance at 550°C for 3 hours. The resulting ash was dissolved in 10ml of IM HCl and then diluted to 100ml in volumetric flask using distilled water. The digest obtained was used for the various analysis. Mineral concentrations were determined for maize, untreated FYSM and the chemically treated FYSM.

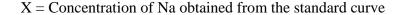
#### 3.5.1 **Determination of sodium**

Weight of 0.2542g of NaCl was dissolved in 1 litre of distilled water to give 100ppm Na. This working standard solution was diluted to produce a range containing 0 – 10ppm sodium and made up to 100ml mark and 2ml sample aliquot (sample stock solution) was read using a JENWAY PFP7 flame photometer. The concentration of the test mineral in the sample was calculated with reference to the graph (standard curve) and obtained as follows:



=Equation 3.7

W = Weight of the sample analyzed



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- VF = Total volume of digest/extract (100ml)
- Va = Volume of extract used
- D = Dilution factor

## 3.5.2 Determination of iron

Standard solution containing 100mg/ml of Fe3<sup>+</sup> ions was prepared from 1g pure iron wire. The wire was dissolved in 20ml concentrated HNO<sub>3</sub>, boiled in water bath and diluted to 1000ml with distilled water. Standard solutions with concentrations 0, 0.5, 1.0, 2.0 and 4.0ppm was prepared. Two milliliters of sample aliquot was diluted to 100ml and was used to determine the absorbance of the sample using an AGILENT (Model 5805, Agilent Spec England) atomic absorption spectrophotometer at 510nm. The standard and samples absorbance were noted and concentration of iron in the sample was determined from the standard curve.

### 3.5.3 Determination of calcium

Calcium was determined using the atomic absorption spectrophotometer. Calcium carbonate (2.495g) was dissolved and diluted to 100ml with de-ionized water. This solution contains 1000mg Ca2<sup>+</sup> ions and from this stock solution, calcium standard of the following concentration levels 0.0, 3.0, 6.0, 9.0 were prepared. The absorbance of both the sample and the standard working aliquot was determined in the AGILENT (Model 5805, Agilent Spec England) atomic absorption spectrophotometer at 239.9nm.

The concentration of the test mineral in the sample was calculated with reference to the graph (standard curve) and obtained as follows:

$$Ca (mg/100g) = 100 \text{ x X x Vf x D}$$

W = Weight of the sample analyzed

X = Concentration of Ca obtained from the standard curve

Vf = Total volume of extract

Va = Volume of extract used

D = Dilution factor

#### 3.5.4 **Determination of phosphorus**

Phosphorus was determined using spectrophotometer. Phosphorus in the sample was determined by the molybdate method using hydroquinone as a reducing agent. Sodium sulphate (1.0ml), ammonium molybdate (1.0 ml) and hydroquinone (1 ml) were added to 1ml of the sample digest. The mixture was agitated and allowed to stand for 30 minutes for the blue colour to develop. The absorbance of the sample was determined using the spectrophotometer at 600nm. The phosphorus standard was prepared by dissolving 1.1g of monobasic potassium phosphorus (KA<sub>2</sub> PO<sub>4</sub>) into a 500ml volumetric flask containing 500 ml of distilled water. Five drops of toluene was added to diminish microbial activity. Twenty millilitres of the standard stock was collected and made up to 100ml. This contained 100ppm. Standard stock (0.1ml) = 0.2ppm. Zero to one millilitre of the 100ppm phosphorus stock solution was poured into 100ml volumetric flask separately and treated the same way as the sample. The reading of the standard was taken at 600 nm in an AGILENT spectrophotometer (Model 5805, Agilent Spec England) and a standard curve was plotted.



=Equation 3.8

$$P (mg/100g) = 100 x Au x C x Vf$$

Where

W = Weight of sample analyzed

- AU = Absorbance of test sample
- AS = Absorbance of standard phosphorus solution
- C = Concentration (in mg/ml) of sample
- VF = Total volume of extract
- Va = Volume of extract analyzed

#### 3.6.0 Analysis of amino acid concentrations

To determine essential amino acids (except tryptophan), ion chromatographic methods were used that conformed to the German Food and Feed Code (§64 LFGB L 49.07-2). Tryptophan was quantified in accordance with procedures specified by the Association of German Agricultural Analytic and Research Institutes using HPLC methods (VDLUFA, 2006). Amino acid concentrations were determined for maize, untreated FYSM and the chemically treated FYSM.



# 3.7.0 Total steroidal saponins determination (Baccou et al., 1977).

# Reagent.

Reagent A: Add 0.5mL of anisaldehyde with 99.5 mL of ethyl acetate and mix thoroughly Reagent B: Concentrated sulphuric acid (95-98 %).

Reagent C: 50 mL of concentrated sulphuric acid plus 50 mL of ethyl acetate.

=Equation 3.9

Standard saponin solution: 10 mg of diosgenin was weighed and dissolve in 25 mL of ethyl acetate (0.4 mg/mL).

#### Preparation of calibration curve

Zero to fourty (0-40)  $\mu$ g of steroid sapogenin was dissolved in 2 mL of ethyl acetate in the test tube and 1 mL of reagent A and 1mL of reagent C were added. After stirring, the test tubes were placed in a water-bath maintained at 60°C for 20 min, then allowed them to cool for 10 min in a water-bath maintained at room temperature. The absorbance was measured at 430 nm against the reagent blank. It was possible to effect this determination without heating the tubes at 60°C water bath if reagent B was used instead of reagent C.

## Determination of saponin

Known amounts of extracted residues were dissolved in 80% methanol and placed in test tubes (corresponding to a sapogenin content of between 0 and  $40\mu g$ ), and the tubes place in a boiling water bath in order to remove alcohol and, after cooling, 2 mL of ethyl acetate was added and the determination carried out as for sapogenin. Total steroidal saponin concentrations were determined for maize, untreated FYSM and the chemically treated FYSM.



#### 3.8 Quantitative determination of total saponins (Hiai et al., 1976).

#### Reagents.

Vanillin reagent (8%, w/v): Dissolve 800 mg of vanillin in 10 mL of 99.5% ethanol (analytical grade) 72% (v/v) sulphuric acid: To 28 mL of distilled water, add 72 mL of sulphuric acid (analytical grade, 95%, w/w).

#### Standard saponin solutions

Ten (10) mg of Diosgenin was weighed and dissolved in 16 mL of Methanol and 4 mL of  $H_2O$  was added. The final concentration of diosgenin in the solution was 0.5mg/mL of 80% methanol. It was mixed thoroughly and pipetting started immediately.

# Preparation of calibration curve

Zero to one hundred and twenty five  $(0-125) \mu g$  of diosgenin was dissolved in 0.25 mL of 80% methanol in the test tube and 0.5 mL of vanillin solution added. The tube was then transfered into ice-cold water bath and 2.5 mL of sulphuric acid added slowly on the inner side of the wall. The solution was mixed well and left as such for 2-3 min. It was warmed in a water-bath settled at 60°C for 10 min., and then cooled in ice-cold water for 3-4 min. The absorbance was measured at 544 nm against the reagent blank and a standard curve prepared.

## Determination of sample.

Known amounts of extracted saponin residues were weighed in 80% methanol. An aliquot of 0.25 mL was taken and carried out the determination as for the standard saponin. Saponin concentrations were determined for maize, untreated FYSM and the chemically treated FYSM.

#### **3.9.0** Gas production test (Hohenheim Method)

#### **Principle**

The gas production test is based on the association between rumen fermentation and gas production. The *in vitro* gas production method can be used to measure the metabolizable energy of feeds and to quantify utilization of nutrients.

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# Procedure

- Weigh 200 mg of feed sample (1 mm ground) and insert carefully in 100 ml calibrated glass syringe
- Include blank syringes without feed
- Include syringes with standard feed (concentrate or/and hay)
- Keep syringes overnight in circulating water bath at 39 °C
- Prepare fermentation buffer solution except rumen fluid and reducing solution in 21 Woulff bottle
- Keep bottle in water-bath at 39 °C with stirring overnight.

# Next morning

- Keep under CO<sub>2</sub>
- Add reducing solution and wait 20 minutes until colour changes from blue to purple to colourless
- Add rumen fluid
- Keep stirring under CO<sub>2</sub> for 10 minutes
- Fill 30 ml into glass syringes
- Incubate syringes in 39 °C
- Shake syringes every hour during first four hours, then twice every hour
- Record gas production at 8 hours and push back piston to 30 ml if gas production exceeds 70 ml
- Record gas production at 24 hours (V24) and terminate experiment.

## **Reagents / Solutions**

# Rumen fluid

• Collect rumen fluid before morning feeding from 2 rams

- Filter through two-layer cheese cloth into thermos container
- Keep at 39 °C and under carbon dioxide (CO<sub>2</sub>).

# Fermentation buffer solution

- 630 ml of bicarbonate buffer
- 315 ml of macro mineral solution
- 0.16 ml of micro mineral solution
- 1.6 ml of resazurine solution
- 945 ml distilled water
- 60 ml of freshly prepared reducing solution
- 660 ml rumen fluid.

# Bicarbonate buffer

- 35 g sodium bicarbonate (NaHCO<sub>3</sub>)
- 4 g ammonium carbonate
- Dissolve in 500 ml distilled water and then make up to 11itre.

# Macro mineral solution

- 6.2 g potassium dihydrogen phosphate (KH<sub>2</sub>PO<sub>4</sub>)
- 5.7 g disodium hydrogen phosphate (Na<sub>2</sub>HPO<sub>4</sub>)
- 0.6 g magnesium sulphate (MgSO<sub>4</sub>.7H<sub>2</sub>O)
- Dissolve in 500 ml distilled water and then make up to 1 liter.

# Micro mineral solution

- 10 g manganese chloride (MnCl<sub>2</sub>.4H<sub>2</sub>O)
- 13.2 g calcium chloride (CaCl<sub>2</sub>.2H<sub>2</sub>O)
- 1 g cobalt chloride (CoCl<sub>2</sub>.6H<sub>2</sub>O)

• Dissolve in 50 ml distilled water and then make up to 100 ml.

#### Resazurine

• 0.1 g resazurine in 100 ml distilled water.

#### **Reducing** solution

- 996 mg sodium sulphide (Na<sub>2</sub>S.9H<sub>2</sub>O)
- Dissolve in 94 ml distilled water
- 6 ml 1 *N* sodium hydroxide solution (NaOH)
- 1 N NaOH = 4 g NaOH in 100 ml distilled water.

#### Calculations

## Calculation for gas production

 $G24 = [(V24 - V0 - G0) \times FSt \times 200]/Ws$ 

G24 = gas production value (ml/200 mg) at 24 hours

- G0 = gas production of blank syringes (ml)
- V0 = volume in ml at begin
- V24 = volume in ml at 24 hours
- Ws = weight of dried sample in mg



FSt = GSt/GSt measured

Reference value (FSt) is gas production GSt of the standard sample (e.g. hay and/or concentrate) as per supplier (e.g. Landesarbeitskreis. Fütterung, Baden Württemberg e.V. (LAF), Hohenheim, 70578 Stuttgart, PF 7200220) compared to the measured gas production in the test (GStmeasured).

=Equation 3.10

#### 3.10.0 Haematological analysis

The Principles of Impedance Haematological Analysis

Blood cells were diluted in a buffered electrolyte solution. A measured volume of sample was passed through an aperture tube (example 100um in diameter) between two electrodes. Interpretation of the current by the non- conducting blood cells altered the electrical charge and a pulse was produced. The amplitude of each pulse is proportioned to the volume of the cell which causes it. A threshold circuit ensures only those pulses that exceeded the present threshold level are counted. The cell count was determined from the total number of pulses obtained from a measured volume of blood.

Note; Analysis of the pulse heights enables mean cell volume (MCV) to be measured and the haematocrit to be calculated from the MCV value and red cell count. In sysmex impedance analyzers, the haematocrit was determined from voltage pulse data and the MCV was calculated from the haematocrit values.

The red blood cell (RBC) counts, total white blood cell (WBC) counts, haemoglobin (Hb) concentration and packed cell volume (PCV) parameters were determined following standard procedures described by Davice and Lewis (1991).



#### 3.11.0 Serum biochemical analysis

The blood samples were centrifuged at 500 rpm (revolution per minute) for 3 minutes in a microcentrifuge to obtain serum that was free from cell debris for the biochemical analysis using a spectrophotometer at a wavelength of 500 nm. The serum obtained was analysed colorimetrically for total protein by the Biuret method with kits (PLASMATEC) supplied by Plasmatec Laboratory Products Ltd., U.K. Colorimetric determination of total protein was based on the principle of Biuret reaction (copper salts in alkaline medium) in which cupric ions form a blue complex, in alkaline solution, with NH<sub>2</sub> of two or more peptide bonds. The intensity of the blue colour formed was proportional to the protein concentration in the plasma or serum. Albumin concentration was determined by the Bromocresol Green (BCG) method (Peters et al., 1982); albumins bind with BCG to form a green compound. The concentration of albumins is directly proportional to the intensity of the green colour formed. Globulin concentration was computed as the difference between total protein and albumin concentrations.

#### 3.12.0 Peroxide value (POV) determination

#### Protocol for meat samples

- Weigh 3 grams of meat sample into 25 ml conical flask
- Heat on a water-bath @ 60°C to melt the fat
- Thoroughly agitate for 3 minutes with 30 ml acetic acid-chloroform mixture (3:2 v/v) to dissolve fat
- Filter solution through Whatman number 1 filter paper to remove meat particles.
- Add saturated KI (0.5 ml) to filtrate followed by starch solution (0.5 ml) as indicator.
- Titrate against Sodium Thiosulphate (0.01NNa<sub>2</sub>S<sub>2</sub>O<sub>3</sub>)

POV  $(meq/kg) = (T \times N/W) \times 100$ 

=Equation 3.11

T= titre value

N=Normality of thiosulphate

W= sample weight

#### **CHAPTER 4**

# 4.0 EXPERIMENT 1: PROXIMATE COMPONENTS, MINERALS, AMINO ACIDS AND SOME ANTI-NUTRIENTS IN PROCESSED FALSE YAM SEED MEALS: POTENTIAL BENEFITS FOR POULTRY NUTRITION

#### **4.1.0 INTRODUCTION**

This experiment was conducted to investigate the proximate and mineral compositions, essential amino acid profiles as well as the concentrations of some anti-nutritional factors and in vitro gas productions of unprocessed and processed false yam seed meals for poultry. A number of authors have reported the presence of some anti-nutritional factors in the false yam seeds that are bitter and toxic and affect its utilization as feed for livestock especially poultry (Vanhaelen et al., 1986; NRI, 1987; Kumar, 1992; Dei et al., 2011; Frohne and Pfander, 2005; Gershenzon and Dudareva, 2007). The negative influence of anti-nutritional factors comes as a result of the anti-nutritional factors reacting with protein, enzymes, or essential amino acids and forming various complexes, thus affecting digestibility and nutrient utilization in poultry (Pekel *et al.*, 2015).

Various processing methods have been employed in an attempt to improve the nutritional value of the false yam seed with varying degrees of success in poultry trials (Dei et al., 2014). Ologhobo *et al.* (1993) reported that, after chemical treatment of false yam seeds, a higher concentration of antinutritional factors was found in base-soluble fractions, indicating a greater extractability of antinutritional factors by alkali treatment than by acid solutes, ether or alcohol.



However, in many instances, usage of only one detoxification method may not achieve the desired removal of anti-nutritional substances and a combination of two or more methods may be required for significant nutritional improvement.

In this experiment, water extraction, various approaches (urea, NaCl, NaOH and KOH) of chemical extraction and blanching were considered in a sequential approach so as to determine www.udsspace.uds.edu.gh

how a combination of processing methods influences the concentration of nutrients and selected anti-nutritive components in false yam seed meals.

# **4.1.1 OBJECTIVES**

The objectives of this experiment were to determine the

- Proximate composition (CP, EE, CF, ASH and NFE) of unprocessed and processed false yam seed meals
- Mineral composition of unprocessed and processed false yam seed meals
- Essential amino acid profiles of unprocessed and processed false yam seed meals
- Terpenes and saponin concentrations of unprocessed and processed false yam seed meals
- *In vitro* gas productions of unprocessed and processed false yam seed meals

# **4.1.2 HYPOTHESIS**

The concentration of nutrients and selected anti-nutritive components in false yam seed meals will not differ when processed by the use of water extraction, various approaches of chemical (urea, NaCl, NaOH and KOH) extraction and blanching



#### 4.2.0 MATERIALS AND METHODS

#### 4.2.1 Procurement and processing of false yam seed meals

Matured fruits of false yam plants growing in the wild around Nyankpala Campus of the University for Development Studies, Tamale, were harvested by hand picking. The fruits were cracked and the false yam seeds (FYS) were partially sun-dried (7 days) to reduce their moisture content. Afterwards, the FYS were crushed to reduce size and increase surface area to facilitate processing. Five samples of false yam seed meal (FYSM) were prepared as described under sections *3.1, 3.2 and 3.3*.

#### 4.2.2 Chemical analysis

#### **4.2.2.1 Proximate composition**

Samples of all FYSM and maize (as positive control, bought in bulk from Tamale maize market) were analyzed using standard methods (AOAC International, 2000) as outlined under section **3.4.0**.

#### 4.2.2.2 Mineral determination

Mineral element analysis followed the official method of the Association of Official Analytical Chemists (AOAC, 2000). One gram (1.0 g) of each sample was suspended in 20 ml HNO<sub>3</sub> in a 100 ml beaker. The mixture was placed on a hot plate and the temperature maintained at 130°C for four hours until the solution became clear. After cooling, the solution was filtered through Whatman filter paper (11 µm pore space) to remove the insoluble particles and made up to a final volume of 50 ml with distilled water in a standard flask. Appropriate dilutions were made for each sample before analysis. Potassium and sodium were determined using a Jenway Digital Flame Photometer (Cole-Parmer, Beacon Road, Stones Staffordshire, ST15, OSA, UK), while other mineral elements were determined using a Buck Scientific Atomic Absorption Spectrophotometer



(Unicam Model 929, Unicam Cambridge, England). The resulting solutions were analyzed using the concentrations of the metals extrapolated from the calibration graphs generated using standard mineral solutions. The procedures employed in these determinations followed the protocols in the manufacturer's manual for the equipment (*see section 3.5.0*).

#### 4.2.2.3 Amino acid determination

To determine essential amino acids (except tryptophan), ion chromatographic methods were used that conformed to the German Food and Feed Code (§64 LFGB L 49.07-2). Tryptophan was quantified in accordance with procedures specified by the Association of German Agricultural Analytic and Research Institutes using HPLC methods (VDLUFA, 2006). Amino acid concentrations were determined for maize, untreated FYSM and the chemically treated FYSM.

# 4.2.2.4 Total steroidal saponins and total saponin determination

Quantitative estimation of total steroidal saponins followed the method of Baccou *et al.* (1977) while quantitative determination of total saponins was done by the method described by Hiai *et al.* (1976) as outlined under sections **3.7.0** and **3.8.0** respectively.

#### 4.2.2.5 In vitro gas production

In vitro gas production (IVGP) of samples was determined by using the Hohenheim Gas Test (HGT) method (Menke et al., 1979) as outlined under section **3.9.0**.



# 4.3.0 RESULTS

# 4.3.1 Proximate, mineral and amino acid components

The proximate composition of maize, sun-dried and treated FYSM is presented in Table 4.1. The results revealed that maize and FYSM treated with potassium hydroxide had highest dry matter contents (92.1% and 92.8%, respectively). The lowest dry matter content was observed in FYSM

treated with sodium hydroxide (89.0%). Protein content was highest in sun-dried FYSM (13.2%) and lowest in KOH\_T (2.2%). The crude fiber content of maize (1.7%) compared favourably to that of Urea\_T (1.3%) and NaCl\_T (1.7%). Ether extract (EE) content of maize (4.3%) was higher than those of FYSM. Among the processed FYSM, Un\_T (1.5%) and KOH\_T (1.5%) were similar in concentration of EE but lower than those of Urea\_T, NaCl\_T and NaOH\_T. Ash content varied widely from 3.8% for KOH\_T to 0.5% for NaCl\_T. On the other hand, NaCl\_T contained the highest share (83.9%) of nitrogen free extractives (NFE); yet NFE concentration of FYSM was generally high, indicating that treated seed meals may be a good source of energy. At 3,464 kcal/kg DM, the calculated ME content of maize was higher than those of the FYSM, where ME contents (per kg DM) ranged from 3,073 kcal for NaOH\_T to 3,284 kcal for NaCl\_T.

The concentrations (per kg DM) of macro-elements (Table 4.2) indicated higher levels of calcium (800 mg Ca) and magnesium (78.4 mg Mg) in maize than in the processed FYSM. However, Un\_T recorded highest calcium (280 mg Ca) and magnesium (52.8 mg Mg) levels within the FYSM. Potassium concentration, on the other hand, was higher in processed FYSM than in maize (87.5 mg K), ranging from 111 mg K (Un\_T) to 368.8 mg K (KOH\_T). With the exception of KOH\_T (281 mg Na), all processed FYSM had higher sodium concentrations than maize (440 mg Na). Total phosphorus concentration was lower (0.31 mg P) in Un\_T and higher (2.37 mg P) in Urea\_T than in maize (4.3 mg P). Among the trace elements (per kg DM) considered in this study, iron concentration was as low as 0.01 mg in Un\_T and as high as 24.6 mg in Urea\_T, whereas the concentration in maize was 5.24 mg. Manganese, copper and zinc concentrations (per kg DM) ranged from <0.002 to 1.16 mg, 0.003 to 29.0 mg and 0.76 - 1.84 mg respectively.

Comparing the concentration of essential amino acids (Table 4.3) of Un\_T versus maize, seven out of ten determined amino acids were higher in Un\_T than in maize, and only leucine, alanine



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and methionine concentrations were higher in maize than in Un\_T. However, there was a decline in the concentrations of all amino acids determined within the treated FYSM as follows: Urea\_T > NaCl\_T > NaOH\_T > KOH\_T.



Table 4.1: Proximate components of maize and differently treated false yam (Icacina oliviformis) seed meals (FYSM)

TUDIES		Crude protein	Crude fibre	Ether extra	ot Nitroo	en free	Ash **	*ME (kcal/kg)
н	Dry matter*	Crude protein	Clude lible	Eulei extra	-	ctives	A511	· MIE (KCal/Kg
	<i>∋</i> 2.1 ± 1.67	9.7 ± 1.31	$1.7\pm0.55$	$4.3 \pm 0.21$	1 77.8	± 0.72 1.4	±0.11	3464 ± 64.74
ŝ	$91.3 \pm 0.25$	$13.2\pm0.09$	$2.7 \pm 0.56$	$1.5 \pm 0.50$	) 71.4 :	± 0.73 2.5	$\pm 0.00$ 3	$3132 \pm 12.11$
E	$91.3 \pm 0.75$	$9.1 \pm 0.23$	$1.3 \pm 0.15$	$2.5 \pm 0.50$	) 74.9	± 1.17 1.0	$\pm 0.50$	$3190 \pm 8.74$
Ē	$92.0 \pm 0.00$	$4.0 \pm 0.12$	$1.7 \pm 0.56$	$2.0 \pm 0.00$	) 83.9	$\pm 0.68$ 0.50	$0 \pm 0.00$	$3284 \pm 19.65$
A.	$39.0 \pm 0.00$	$2.7 \pm 0.32$	$2.7 \pm 0.58$	$2.5 \pm 0.50$	) 78.1	± 1.39 3.0		$3073 \pm 2.90$
-	$92.8 \pm 1.25$	$2.2 \pm 0.06$	$2.7 \pm 0.54$			± 1.02 3.8		$3149 \pm 79.11$
ЕĹ					1 1		••••	
2					hydroxide treated, K	OH_T= potassium hydr	oxide treated;	
<u> </u>								
н								
Ř	···· · • • • • • • • • • • • • • • • •	J	- <b>1</b>	J J:6641 41				
FOR I	on of macro- an	d micro-miner	als in maize an	d differently t	reated false ya	m ( <i>Icacina olivifo</i>	rmis) seed r	neals (FYSM
SITY FOR		ad micro-miner ents Mean ± sd		d differently t		m ( <i>Icacina olivifo</i> e elements Mean		
SITY FOR				d differently to Phosphorus		· · · ·		
SITY FOR	Macro elem	ents Mean $\pm$ sd	(mg/kg DM)		Trace	e elements Mean	± sd (mg/kg	DM) Zinc
Y FOR	Macro elem Potassium	ents Mean±sd Magnesium	(mg/kg DM) Sodium	Phosphorus	Trace	e elements Mean Manganese	± sd (mg/kg Copper	DM) Zinc
SITY FOR	Macro elem Potassium 87.5 ± 1.01	ents Mean $\pm$ sd Magnesium $78.4 \pm 0.20$	(mg/kg DM) Sodium 440.2 ± 2.15	Phosphorus 0.4 ± 0.00	Trace Iron 5.2 ± 0.03	e elements Mean Manganese $1.2 \pm 0.10$	$\pm \text{ sd (mg/kg)}$ Copper $0.01 \pm 0.00$	DM) Zinc D 1.2 ±0.02 1.8 ± 0.02
SITY FOR	Macro elem Potassium 87.5 ± 1.01 110.6 ±1.53	ents Mean $\pm$ sd Magnesium 78.4 $\pm$ 0.20 52.8 $\pm$ 1.00 13.3 $\pm$ 0.00	(mg/kg DM) Sodium 440.2 ± 2.15 558.0 ± 1.00	Phosphorus 0.4 ± 0.00 0.3 ± 0.00	Trace Iron 5.2 ± 0.03 0.01 ± 0.002	e elements Mean Manganese $1.2 \pm 0.10$ $< 0.002 \pm 0.001$	$\pm$ sd (mg/kg Copper 0.01 $\pm$ 0.00 2.1 $\pm$ 0.10	(DM) Zinc D $1.2 \pm 0.02$ $1.8 \pm 0.02$ $1.2 \pm 0.02$
SITY FOR	Macro elem Potassium 87.5 ± 1.01 110.6 ±1.53 236.1 ± 1.05	ents Mean $\pm$ sd Magnesium 78.4 $\pm$ 0.20 52.8 $\pm$ 1.00 13.3 $\pm$ 0.00 14.1 $\pm$ 0.01	$(mg/kg DM)$ Sodium $440.2 \pm 2.15$ $558.0 \pm 1.00$ $602.0 \pm 0.20$	Phosphorus 0.4 ± 0.00 0.3 ± 0.00 2.4 ±0.01	Trace Iron $5.2 \pm 0.03$ $0.01 \pm 0.002$ $24.6 \pm 0.02$	e elements Mean Manganese $1.2 \pm 0.10$ $<0.002 \pm 0.001$ $<0.002\pm0.001$	$\pm$ sd (mg/kg Copper 0.01 $\pm$ 0.00 2.1 $\pm$ 0.10 1.7 $\pm$ 0.02	DM) Zinc D 1.2 ±0.02 1.8 ± 0.02
SITY FOR	Macro elem Potassium $87.5 \pm 1.01$ $110.6 \pm 1.53$ $236.1 \pm 1.05$ $210.3 \pm 1.00$	ents Mean $\pm$ sd Magnesium 78.4 $\pm$ 0.20 52.8 $\pm$ 1.00 13.3 $\pm$ 0.00 14.1 $\pm$ 0.01 15.5 $\pm$ 0.10	$(mg/kg DM)$ Sodium $440.2 \pm 2.15$ $558.0 \pm 1.00$ $602.0 \pm 0.20$ $989.0 \pm 1.00$	Phosphorus $0.4 \pm 0.00$ $0.3 \pm 0.00$ $2.4 \pm 0.01$ $1.2 \pm 0.00$	Trace Iron $5.2 \pm 0.03$ $0.01 \pm 0.002$ $24.6 \pm 0.02$ $19.7 \pm 0.02$	e elements Mean Manganese $1.2 \pm 0.10$ $<0.002 \pm 0.001$ $<0.002 \pm 0.001$ $<0.002 \pm 0.001$	$\pm$ sd (mg/kg Copper 0.01 $\pm$ 0.00 2.1 $\pm$ 0.10 1.7 $\pm$ 0.02 2.0 $\pm$ 0.01	DM) Zinc $1.2 \pm 0.02$ $1.8 \pm 0.02$ $1.2 \pm 0.02$ $0.8 \pm 0.02$
	VELOPMENT	$\begin{array}{c} 91.3 \pm 0.25 \\ 91.3 \pm 0.75 \\ 92.0 \pm 0.00 \\ 39.0 \pm 0.00 \\ \underline{92.8 \pm 1.25} \\ \text{f fresh matter; each n} \\ \text{ated, Urea_T= urea tr} \end{array}$	$\begin{array}{c} 91.3 \pm 0.25 & 13.2 \pm 0.09 \\ 91.3 \pm 0.75 & 9.1 \pm 0.23 \\ 92.0 \pm 0.00 & 4.0 \pm 0.12 \\ 39.0 \pm 0.00 & 2.7 \pm 0.32 \\ \hline 2.8 \pm 1.25 & 2.2 \pm 0.06 \\ f  fresh matter; each mean represents triplicated, Urea_T= urea treated, NaCl_T= sodium text and the text of tex of tex of tex of tex of tex of text of text of $	$\begin{array}{c} 91.3 \pm 0.25 & 13.2 \pm 0.09 & 2.7 \pm 0.56 \\ 91.3 \pm 0.75 & 9.1 \pm 0.23 & 1.3 \pm 0.15 \\ 92.0 \pm 0.00 & 4.0 \pm 0.12 & 1.7 \pm 0.56 \\ 39.0 \pm 0.00 & 2.7 \pm 0.32 & 2.7 \pm 0.58 \\ \underline{92.8 \pm 1.25} & 2.2 \pm 0.06 & 2.7 \pm 0.54 \\ \hline \text{f fresh matter; each mean represents triplicate determinations;} \\ \text{ated, Urea_T= urea treated, NaCl_T= sodium chloride treated,} \end{array}$	$\begin{array}{c} \textbf{71.3} \pm 0.25 & 13.2 \pm 0.09 & 2.7 \pm 0.56 & 1.5 \pm 0.50 \\ \hline \textbf{91.3} \pm 0.75 & \textbf{9.1} \pm 0.23 & 1.3 \pm 0.15 & 2.5 \pm 0.50 \\ \hline \textbf{92.0} \pm 0.00 & 4.0 \pm 0.12 & 1.7 \pm 0.56 & 2.0 \pm 0.00 \\ \hline \textbf{39.0} \pm 0.00 & 2.7 \pm 0.32 & 2.7 \pm 0.58 & 2.5 \pm 0.50 \\ \hline \textbf{92.8} \pm 1.25 & 2.2 \pm 0.06 & 2.7 \pm 0.54 & 1.5 \pm 0.50 \\ \hline \textbf{13.5} \pm 0.50 & \textbf{13.5} & \textbf{13.5} & \textbf{13.5} \\ \hline \textbf{13.5} \pm 0.50 & \textbf{13.5} & \textbf{13.5} & \textbf{13.5} & \textbf{13.5} & \textbf{13.5} \\ \hline \textbf{92.6} \pm 0.00 & 4.0 \pm 0.12 & 1.7 \pm 0.56 & 2.0 \pm 0.00 \\ \hline \textbf{92.8} \pm 1.25 & 2.2 \pm 0.06 & 2.7 \pm 0.54 & 1.5 \pm 0.50 \\ \hline \textbf{13.5} \pm 0.50 & \textbf{13.5} & \textbf{13.5} & \textbf{13.5} & \textbf{13.5} \\ \hline \textbf{13.5} \pm 0.50 & \textbf{13.5} & \textbf{13.5} & \textbf{13.5} & \textbf{13.5} & \textbf{13.5} & \textbf{13.5} \\ \hline \textbf{92.6} \pm 0.00 & \textbf{13.5} \\ \hline \textbf{92.6} \pm 0.00 & \textbf{13.5} & \textbf{13.5}$	$\begin{array}{c} 91.3 \pm 0.25 & 13.2 \pm 0.09 & 2.7 \pm 0.56 & 1.5 \pm 0.50 & 71.4 \pm 0.12 \\ 91.3 \pm 0.75 & 9.1 \pm 0.23 & 1.3 \pm 0.15 & 2.5 \pm 0.50 & 74.9 \pm 0.20 \pm 0.00 & 4.0 \pm 0.12 & 1.7 \pm 0.56 & 2.0 \pm 0.00 & 83.9 \pm 0.00 & 2.7 \pm 0.32 & 2.7 \pm 0.58 & 2.5 \pm 0.50 & 78.1 \pm 0.20 \pm 0.00 & 2.7 \pm 0.32 & 2.7 \pm 0.54 & 1.5 \pm 0.50 & 82.6 \pm 0.50 & 78.1 \pm 0.20 \pm 0.00 & 82.6 \pm 0.50 & 82$	$\begin{array}{cccccccccccccccccccccccccccccccccccc$	$\begin{array}{cccccccccccccccccccccccccccccccccccc$

FYSM treatments. On\_1 = untreated, Urea\_T = urea treated, NaCl\_T = sodium chloride treated, NaOH\_T = sodium hydroxide treated, KOH\_T = potassium hydroxide treated, sd = standard deviation.

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Amino acid	Maize	SFYSM*	Un_T	Urea_T	NaCl_T	NaOH_T	KOH_T
Phenylalanine	0.47	0.45	0.58	0.24	0.19	0.15	0.11
Leucine	1.18	0.72	0.9	0.37	0.31	0.23	0.17
Alanine	0.63	ND	0.6	0.26	0.2	0.15	0.11
Valine	0.45	0.49	0.5	0.22	0.19	0.14	0.11
Threonine	0.33	0.35	0.42	0.18	0.14	0.1	0.07
Methionine	0.23	0.06	0.1	0.04	0.03	0.03	< 0.03
Lysine	0.27	0.28	0.35	0.17	0.14	0.07	0.06
Arginine	0.53	0.74	1.45	0.45	0.34	0.17	0.1
Histidine	0.3	0.23	0.36	0.17	0.13	0.15	0.12
Tryptophan	0.07	0.15	0.16	0.08	0.06	0.05	0.04

 Table 4.3: Amino acid concentrations (% DM) of maize and differently treated false yam

 (*Icacina oliviformis*) seed meals (FYSM)

FYSM treatments: Un\_T= untreated, Urea\_T= urea treated, NaCl\_T= sodium chloride treated, NaOH\_T= sodium hydroxide treated, KOH\_T= potassium hydroxide treated; ND= Not determined \*SFYSM= water-soaked false yam seed meal, values according to Dei *et al.* (2012).

# 4.3.2 Total terpenes, saponin and tannin concentrations

Figure 4.1 shows the concentrations of total terpenes in maize, untreated and treated false yam

seed meals.

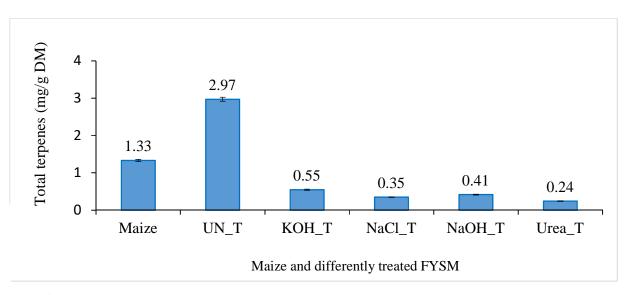
Untreated FYSM (Un\_T) had the highest concentration of total terpenes (2.97 mg/g DM) and

Urea\_T recorded the lowest value. The reduction in total terpenes (Figure 4.1) due to processing

relative to Un\_T was 91.9% (Urea\_T), 88.3% (NaCl\_T), 86.1% (NaOH\_T) and 81.6% (KOH\_T).



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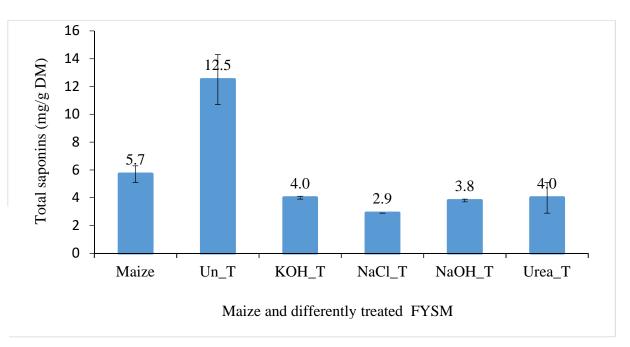


**Figure 4.1:** Concentration of total terpenes in maize, untreated and differently treated false yam seed meals (FYSM) FYSM treatments: Un\_T= untreated, KOH\_T= potassium hydroxide treated, NaCl\_T= sodium chloride treated, NaOH\_T= sodium hydroxide treated, Urea\_T= urea treated. Where visible, error bars represents standard error of means of 3 replicates.

Figure 4.2 shows the concentrations of total saponin in maize, untreated and treated false yam seed meals. Total saponin concentration (mg/kg DM) was highest in Un\_T (12.5) and lowest in NaCl\_T (2.9). The reduction in total saponin concentration (Figure 4.2) relative to Un\_T was 76.8% (NaCl\_T), 69.6% (NaOH\_T), and 68.0% (KOH\_T, Urea\_T).



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**Figure 4.2:** Concentration of saponins in maize, untreated and differently treated false yam seed meals (FYSM). FYSM treatments: Un\_T= untreated, KOH\_T= potassium hydroxide treated, NaCl\_T= sodium chloride treated, NaOH\_T= sodium hydroxide treated, Urea\_T= urea treated. Where visible, error bars represents standard error of means.

Table 4.4 shows the results of *in vitro* gas production of maize and FYSM samples incubated for 24 hours in rumen fluid, with or without adding polyethylene glycol (PEG). Inclusion of PEG resulted in a significant reduction of gas production in maize, KOH\_T, NaCl\_T and NaOH\_T FYSM samples (Table 4.4). Urea\_T FYSM sample also showed a non-significant (P>0.214) reduction of about 4.2 ml gas production when PEG was added to the sample. However, the untreated (Un\_T) FYSM sample had a significant (P<0.012) improvement in gas production when PEG was added to the sample.



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Table 4.4: *In vitro* gas production (IVGP) of maize, untreated and differently treated false yam seed meals when incubated for 24 hours with rumen fluid without (-PEG) or with (+PEG) addition of polyethylene glycol (PEG).

	IVGP (ml/	IVGP (ml/380 mg DM)					
Feedstuff	- PEG at 0.00 mg	+ PEG at 0.75 mg	*Probability				
	$(Mean \pm SD)$	$(Mean \pm SD)$					
Maize	132.4 (3.59)	121.2 (5.87)	0.048				
Un_T FYSM	138.3 (1.19)	143.5 (1.66)	0.012				
KOH_T FYSM	134.7 (2.46)	121.7 (2.32)	0.003				
NaCl_T FYSM	162.7 (1.52)	157.1 (1.29)	0.009				
NaOH_T FYSM	131.8 (2.65)	118.7 (1.41)	0.002				
Urea_T FYSM	163.9 (4.76)	158.9 (0.81)	0.214				

FYSM treatments: Un\_T= untreated, KOH\_T= potassium hydroxide treated, NaCl\_T= sodium chloride treated, NaOH\_T= sodium hydroxide treated, Urea\_T= urea treated; -PEG= without polyethylene glycol, +PEG=with polyethylene glycol, SD= standard deviation, P= probability. \*t- test.



#### 4.4.0 DISCUSSION

The information on proximate composition, mineral concentrations, essential amino acids and antinutritional components of differently treated false yam seed meals provides substantial information on the nutritional value of FYSM and can serve as a guide in diet formulation that considers FYSM as a partial replacement of, for example, maize in diets of monogastric animals. Generally, the dry matter (DM) contents of all false yam seed meals compared favourably with that of maize. The latter was within the range of 88 - 98% reported by Enyisi *et al.* (2014) whereas the DM content of FYSM was similar to values reported by Golly and Amadotor (2013), Salifu *et al.* (2015) and Sunday *et al.* (2016). The similarity in DM content of maize and FYSM indicates that on dry matter basis both feedstuffs can complement each other in monogastric animals, particularly poultry. The crude protein concentration (9.7%) of maize in the present study was within the range of 9 – 10% CP in maize DM reported by Sverker *et al.* (2005) but was higher than values (7.86-8.41%) reported by Deka and Sarkar (1990) and Edema *et al.* (2005) and lower than the value (11.2-12.65%) given by Aminogo and Ogutunde, (2000) and Gupta (2001). Such variations might be associated with varietal differences.

Processing of FYSM resulted in a reduction of the CP content by 31% with Urea\_T and up to 83.5% with KOH\_T as compared to Un\_T. The remarkable decrease in CP concentration in the treated FYSM may be attributed to the leaching out of soluble nitrogenous seed components during the soaking procedure as reported by Longland *et al.* (2011), who indicated that soaking allows greater extratability of total nitrogen, leading to relatively lower CP content. The CP content of soaked false yam seed meal reported by Dei *et al.* (2013) was higher than the values observed in this study. However, Valantine and Sulemana (2012) reported a CP concentration of 14% in the untreated seed, comparable to the value determined in this study. Golly and Amadotor (2013)

observed a 15% reduction in CP content of false yam seeds after soaking them for 17 days in ordinary water. The present drastic reduction in CP content of treated FYSM suggests that the sequential treatment involving the use of chemical solutions aggravated nutrient loss. Yet, a variability in the complexity of the processing methods used could result in wide variation of nutrient concentrations.

Ether extract content of maize (4.3%) was comparable to the values of 4.4% and 4.1% reported by Aminogo and Ogutunde (2000) and Edema (2005), respectively, but was higher than the values determined by Deka and Sarkar (1990). However, ether extract of FYSM observed in the present study was generally lower than that of maize (Table 4.1). Untreated FYSM had an ether extract content similar to that reported for untreated false yam tuber meal by Dei *et al.* (2011).

The nitrogen free extractive content of maize (78%) obtained in our study was higher than the 66 – 70% reported by Ujabadenyi and Adebolu (2005) and the 72 – 73% reported by Wilson *et al.* (1999); as a general rule, the carbohydrate content of maize may be environmentally or genetically controlled. The high carbohydrate content of FYSM treated with NaCl, KOH and NaOH as compared to Un\_T and Urea\_T samples might be explained by differences in the employed chemicals and their reaction with seed proximate constituents. Soaking in NaOH and KOH resulted in a gelatinization of the seed which might enhance the carbohydrate content, as also observed in high-starch tubers (Prathibha et al., 1998).

The ash content of our samples of maize and FYSM falls within the range (1.4 - 3.3%) for maize flour reported by Enyisi *et al.* (2014) and Mlay *et al.* (2005), as well as in the range (1.2 - 1.5%)reported for maize flour by Yadav and Yadav (2002) – with the exception of Urea\_T and NaCl\_T. The ash content of untreated false yam seeds was reported as 2.6% (Golly and Amadotor, 2013), which compares well with the ash content (2.5%) of Un\_T in our study.



Generally, Ca and Mg concentrations in the sun-dried false yam seed meals were lower than in maize, and processing the FYSM further reduced Ca and Mg concentrations. This could be due to leaching during the soaking process. However, K and Na were lower in maize than in FYSM. Golly and Amadotor (2013) reported a concentration of 56.3 mg Ca/kg DM in untreated false yam seeds and 52.1 mg Ca/kg DM in water treated FYSM. The Mg value obtained for maize was higher than the 29 – 47 mg Mg/kg DM reported for maize by Enyisi *et al.* (2014). FYSM concentrations of micro-minerals such as Fe, Cu and Zn were enhanced and Mn reduced due to processing. Differences in mineral composition between differently treated FYSM are most probably due to differences in the chemical agents used for treatment and their reactivity (Karr-Lilienthal et al., 2004).

The concentrations of essential amino acids determined in Un\_T were superior to those reported for untreated false yam tuber meal (Dei et al., 2011) and for soaked false yam seed meal (Dei et al., 2012; see Table 4.3); they were also higher than those of maize determined in this study, except for leucine, alanine and methionine. Soaking has been reported to wash toxic substances out of false yam tubers (Dei et al., 2015) but alongside it removes soluble nutrients from the treated feedstuffs, as observed in this study. In consequence, treated FYSM showed a reduction in total CP as well as individual amino acid concentrations as compared to Un\_T, indicating that the negative effect of soaking on the concentrations of valuable nutrients was reinforced by chemical treatments and blanching. The variation of the amino acid concentrations in the treated FYSM might again be due to differences in the properties of the chemical agents used for treatment. Therefore, to balance the nutrient composition of a diet containing treated FYSM, additional feeds and/or feed additives might be needed to compensate for the low protein content of false yam seed meals as suggested by Dei *et al.* (2015) for false yam tuber meal.



Anti-nutritional factors such as terpenes in false yam (Vanhaelen et al., 1986) are plant biosynthetic substances which have profound effects on animal metabolism (Frohne and Pfander, 2005) and some terpenic components can act as toxins, growth inhibitors, or are deterrent to animals (Gershenzon and Dudareva, 2007). The chemical analysis of the untreated false yam seed sample confirmed the presence of resins identified to be terpenes (Vanhaelen et al., 1986), determined at a concentration of 2.97 mg total terpenes per gram DM. No information is available in literature on the concentration of total terpenes in untreated false yam seeds. However, Dei et al. (2011) reported a total resin concentration of 37.5 g/kg DM in untreated false yam tuber meal which was higher than the maximum amount of 28 g total resin per kg of DM reported by NRI (1987). Chemical treatments used in the present study proved effective in reducing total terpenes in FYSM by 82 - 92% and saponing by 68 - 77%; this was a greater removal than the 39% reduction of total terpenes reported by Dei et al. (2011) for boiled false yam tuber meal, although it is known that aqueous solutions can also be used for the extraction of terpenes (Kamphoff et al., 2007). The significant reduction observed can be attributed to the characteristics of the chemical agents used, which were all basic in nature. A higher concentration of anti-nutritional factors was found in base-soluble than in other fractions (Ologhobo et al., 1993), suggesting a greater extractability of anti-nutritional factors by basic reagents. Limonoids, the triterpenoids in neem seed kernel cake, were reduced by soaking in water (1:5 wt./vol.) containing either NaOH (2% wt./wt.) for 24 h or by ensiling with 2.5% urea (wt./wt.) for 5-6 d (Nagalakshmi et al., 1996, 1999). Alkali treated and urea-ammoniated neem seed kernel cake was found suitable for feeding broiler poultry (Nagalakshmi et al., 1996, 1999) without affecting their growth, nutrient utilization, blood profile and gross and histopathology of vital organs. The observed reduction of anti-nutritional



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factors may thus enhance the feeding value of treated FYSM for monogastric animals.

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Tannins, another major group of plant secondary compounds, cause decreased feed consumption in animals, bind to dietary protein to form complexes that are not readily digestible and inactivate digestive enzymes (Aletor, 1993). They also cause decreased palatability and reduced growth rate (Roeder, 1995). The biological test for tannins conducted in this study, namely the *in vitro* gas production with or without polyethylene glycol addition, yielded reduced 24 h gas production values for maize and all FYSM samples, but improved 24 h gas production in the untreated FYSM sample; this indicates that tannins are a major anti-nutritional factor in Untreated FYSM sample.



#### 4.5.0 CONCLUSION AND RECOMMENDATION

The sequential use of water-based and chemical treatment methods was effective in reducing some anti-nutritional factors in false yam seed meal, which can enhance its usefulness for diet formulation in monogastric animals, particularly poultry. However, sequential treatments also induced losses of crude protein, essential amino acids and macro as well as micro-minerals. Therefore, other ingredients rich in the lost nutrients need to complement the use of treated false yam seed meals in such diets.

The processing methods employed in this study induced nutrient losses and still contained residual anti-nutrients, therefore a further study (experiment) is recommended to investigate the extent of feed preference and apparent nutrient digestibility of the treated false yam seed meals by poultry.



#### **CHAPTER 5**

# 5.0 EXPERIMENT 2: PROCESSED FALSE YAM SEED MEALS IN BROILER CHICKEN DIETS: EFFECTS ON FEED PREFERENCE AND APPARENT NUTRIENT DIGESTIBILITY

#### **5.1.0 INTRODUCTION**

It was confirmed from Experiment 1 that the false yam seed meals contained some anti-nutrients. The sequential use of water-based and chemical treatment methods was effective in reducing some of the anti-nutrients but also induced nutrient losses in the samples evaluated. Hence the need for Experiment 2 to determine the effect of the treated false yam seed meals on feed preference and apparent nutrient digestibility by poultry.

The presence of anti-nutritional factors in feedstuffs has been reported to affect feed digestibility and utilization by animals (Lange *et al.*, 2000). Food processing methods can help control potential adverse effects of plant compounds that are toxic to humans or animals when such foods are consumed. The processing methods employed in Experiment 1 did not completely remove the anti-nutrients studied, then they are capable of reacting with protein, enzymes, or essential amino acids and form various complexes, thus affecting digestibility and nutrient utilization in poultry (Pekel *et al.*, 2015).



# **5.1.1 OBJECTIVES**

- To determine the effect of four (4) treated false yam seed meals on feed preference at ten different inclusion levels (5, 10, 15, 20, 25, 30, 35, 40, 45 and 50 %)
- To determine the effect of four (4) treated false yam seed meals on apparent nutrient digestibility at five different inclusion levels (10, 20, 30, 40 and 50%).

# 5.1.2. HYPOTHESIS

Feed preference and apparent nutrient digestibility by broiler chicken will not differ among the four (4) treated false yam seed meals when included at different inclusion levels in maize-fish meal based diets.



#### **5.2.0 MATERIALS AND METHODS**

#### **5.2.1 Experimental samples**

The 4 treated false yam seed meals were prepared as outlined in Experiment 1 (*Section 4.2.1*). A basal diet was prepared that was based on maize, dehulled soybean meal, wheat bran and fish meal, but not supplemented with any of the four (4) treated false yam seed meals. Ten dietary inclusion levels (5, 10, 15, 20, 25, 30, 35, 40, 45 and 50 %) differently treated false yam seed meals were added to the basal diet that was calculated to contain adequate levels of required nutrients for broiler chickens (*Table 5.2*) for both feed preference test and apparent nutrient digestibility trial. The determined nutrient composition of the four (4) treated false yam seed meals is presented in Table 5.1.

			Fals	se yam seed :	meal <sup>2</sup>	
nemical component	Maize	Un_T	Urea_T	NaCl_T	NaOH_T	KOH_T
y matter	92.1	91.3	91.3	92.0	89.0	92.8
ude protein	9.7	13.2	9.1	4.0	2.7	2.2
ude fiber	1.7	2.7	3.8	1.7	2.7	2.7
her extract	4.3	1.5	2.5	2.0	2.5	1.5
trogen free extract	77.8	71.4	74.9	83.9	78.1	82.6
sh	1.4	2.5	1.0	0.5	3.0	3.8
$E (kcal/kg)^3$	3464	3132	3190	3284	3073	3149
sential amino acids						
enylalanine	0.5	0.6	0.1	0.2	0.2	0.2
ucine	1.2	0.9	0.2	0.3	0.2	0.4
anine	0.6	0.6	0.1	0.2	0.2	0.3
line	0.5	0.5	0.1	0.2	0.1	0.2
Ireonine	0.3	0.4	0.1	0.1	0.1	0.2
Methionine	0.2	0.1	< 0.03	0.03	0.03	0.04
Lysine	0.3	0.4	0.1	0.1	0.1	0.2
Arginine	0.5	1.4	0.1	0.3	0.2	0.5
Histidine	0.3	0.4	0.1	0.1	0.2	0.2
Tryptophan	0.1	0.2	0.04	0.1	0.1	0.1
Anti-nutrients (mg/g DM)						
Total terpenes	1.33	2.97	0.24	0.35	0.41	0.55
Total saponins	5.7	12.5	4.0	2.9	3.8	4.0

Table 5.1 Determined nutrient composition of false yam seed meals (% DM)<sup>1</sup>

DM: dry matter; <sup>1</sup>Values presented are from one replicate analysis of amino acids and means of triplicate analyses for the other chemical components; <sup>2</sup>Un\_T, Urea\_T, NaCl\_T, NaOH\_T and KOH\_T refer to the untreated, urea treated, sodium chloride treated, sodium hydroxide treated and potassium hydroxide treated false yam seed meals, respectively; <sup>3</sup>ME was calculated using the formula of Pauzenga (1985) and used in calculating ME content of experimental diets.

		Inclusion levels (%) of TFYSM										
ngredient	С	5	10	15	20	25	30	35	40	45	50	
Iaize	620	589	558	527	496	465	434	403	372	341	310	
$YSM^1$	0	31	62	93	124	155	186	217	248	279	310	
ish meal	115	107	107	106	106	106	105	105	105	104	104	
Vheat bran	98	93	93	94	94	94	95	95	95	96	96	
oybean meal	137	150	150	150	150	150	150	150	150	150	150	
'it./Min remix <sup>2</sup>	2.5	2.5	2.5	2.5	2.5	2.5	2.5	2.5	2.5	2.5	2.5	
)i-calcium	10	10	10	10	10	10	10	10	10	10	10	
hosphate												
lyster shell	15	15	15	15	15	15	15	15	15	15	15	
ommon salt	2.5	2.5	2.5	2.5	2.5	2.5	2.5	2.5	2.5	2.5	2.5	
<i>utrient</i> <sup>3</sup> (g/kg	DM)											
'Р (8 г	200	200	200	200	200	200	200	200	200	200	200	
la	16.2	15.9	16.0	16.1	16.2	16.3	16.3	16.5	16.6	16.6	16.7	
	8.8	8.5	8.4	8.3	8.3	8.2	8.1	8.0	8.0	7.8	7.8	
ys	10.9	10.9	10.9	10.9	10.9	10.9	10.7	10.7	10.7	10.6	10.6	
let	4.3	4.2	4.2	4.2	4.2	4.1	4.1	4.0	4.0	3.9	3.8	
1E (kcal/kg )M)	2905	2898	2891	2884	2877	2871	2863	2857	2850	2843	2836	

Table 5.2 Ingredients and calculated nutrient composition of the control (C) and each of the treated false vam seed meal (TFYSM) diets

<sup>I</sup>Urea\_T, NaOH\_T, NaCl\_T, KOH\_T and Un\_T



<sup>2</sup>Composition of vitamin/trace mineral premix per kg diet (Arosol Chemicals Ltd, India): Vitamin A, 6250 IU; Vitamin D3, 1250 IU; Vitamin E, 25 mg; Vitamin K3, 25 mg; Vitamin B1, 25 mg; Vitamin B2, 60 mg; Vitamin B6, 40 mg; Vitamin B12, 2 mg; Folic acid, 10 mg; Niacin, 40 mg; D-Biotin, 5 mg; Elemental calcium, 25 g; Elemental phosphorus, 9 g; Elemental magnesium, 300 g; Choline chloride, 500 mg; Sodium (as sodium chloride), 1.5 mg; Copper (as penta-hydrate sulphate copper), 60 mg; Cobalt (as hepta-hydrate sulphate cobalt), 10 mg; Zinc (as zinc oxide), 150 mg; Manganese (as manganous oxide), 100 mg; Iron (as ferrous carbonate); Iodine (as potassium iodine), 20 mg; and Selenium (as sodium selenium), 1.0 mg. Lime lactobacillus spore, 0.2 million cfu. <sup>3</sup>Calculated nutrient concentration and ME

#### 5.3.0 Broiler bioassay

The feed preference test was conducted using twenty four Cobb broiler chicks (12 male, 12 female) aged 21 days and of similar body weights (806 g/bird). They were selected, divided into 6 groups of 4 chicks (individual replicates) and randomly assigned to 24 wire-mesh floor cages (0.4 m x 0.3 m =  $0.12 \text{ m}^2$ /chick). FYSM (1 untreated and 4 treated with either urea, sodium chloride, potassium hydroxide or sodium hydroxide) were used to formulate diets with inclusion levels of 0, 5, 10, 15, 20, 25, 30, 35, 40, 45 and 50% in replacement of maize on weight by weight basis. All diets were formulated to be isonitrogenous with similar caloric values (Table 5.2). Each FYSM was thus used in a total of 10 different diets and each dietary level of each FYSM was fed for 24 hours to 4 chicks to determine the inclusion level at which feed intake of the respective FYSM-based diet will be compromised (feed preference). The control diet (maize-based) was fed for 11 days to 4 chicks and feed intakes of the control and all test diets were determined quantitatively by subtracting feed refusal from the feed supplied for every 24 hours. The difference in feed intake of the control and each of the FYSM-based diets was used to determine the preference/aversion of the supplemented diets.



In the apparent nutrient digestibility trial involving FYSM-based diets, a total of 108 chicks (54 males, 54 females) aged 4 weeks and with similar body weights ( $825g \pm 1.23$ ) were selected and randomly divided into 5 groups of 20 birds per group. Each group was subdivided into 5 treatment groups of 4 birds (individual replicates) housed in individual wire-mesh floor cages (0.4 m x 0.3 m = 0.12 m<sup>2</sup>/chick) in a Completely Randomized Block Design. The control group had 8 birds. The five types of FYSM were tested at inclusion levels of 10, 20, 30, 40 and 50%, respectively, replacing maize (wt. / wt.) in a maize-fish meal-based diet (Table 5.2). Each bird received one of the 26 dietary treatments for the period of 15 days. The first 10 days constituted the preliminary

stage of the trial, where birds were allowed to adapt to their new environment as well as new diets. The last 5 days were used for data collection. During this period, feed and water were provided *ad libitum* and light was provided 24 hours with 10 lux of light intensity.

During the 5 days of data collection, weighed quantities of the diets were supplied daily and eventual refusals reweighed after 24 h. Faeces were collected on plastic sheets placed under the wire-mesh floor of the cages (total collection method) and were removed every 24 hours, weighed and stored at 4°C in a refrigerator. At the end of the trial, the daily samples of feed refusal and faeces collected from chickens in each replicate cage were pooled into one sample per treatment, oven dried (60°C for 24h), weighed, ground (2 mm) and stored in airtight plastic containers at room temperature for chemical analysis.

Triplicate samples of treatment diets and dried faeces were analysed for proximate components in accordance with standard methods described by AOAC (2000) (see section *3.4.0*) and the values were used to compute apparent nutrient digestibility (Equation 5.1).

Apparent nutrient digestibility (%) =Nutrient consumed – Nutrient excreted in faeces x 100 Equation 5.1 Nutrient consumed



#### 5.4.0 STATISTICAL ANALYSIS

Feed preference data were analysed using descriptive statistics (Mean  $\pm$  SD), while digestibility data were subjected to two-way analysis of variance (ANOVA) and post-hoc Tukey's honest significant difference (HSD) test with 95% family-wise confidence level.

# 5.5.0 RESULTS

The results of the feed preference trial of broiler chickens fed diets containing FYSM are shown in Table 5.3 and Figure 5.1. The feed intake data indicated that broiler chickens' feed intake was not compromised when maize was substituted with NaCl\_T, NaOH\_T or KOH\_T up to 50% in their diets. The levels of feed intake of these diets were comparable to those of birds in the control group. Broiler chickens preferred diets containing up to 30% Urea\_T and up to 5% Un\_T, whereas higher inclusion levels (>50%) of the respective FYSM compromised the birds' feed intake (Figure 5.1).

Table 5.3 Effect of false yam seed meal (FYSM)<sup>1</sup> on feed preference of broiler chickens (data

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ш	g/I	)Iru/	uay	per	level)

			l	Number o	of days of	f feeding	basal die	et				
	1	2	3	4	5	6	7	8	9	10	11	
	104.8	103.3	119.3 <sup>ab</sup>	125.3 <sup>a</sup>	133.3ª	138.3ª	138.5ª	139.3ª	142.5ª	144.5 <sup>a</sup>	148.5ª	
	Inclusion level (%) of differently treated FYSM (n=4)											
I	0	5	10	15	20	25	30	35	40	45	50	
	107.8	86.0	95.5 <sup>b</sup>	93.3 <sup>b</sup>	87.5 <sup>b</sup>	82.8 <sup>b</sup>	80.3 <sup>b</sup>	79.0 <sup>b</sup>	78.8 <sup>b</sup>	78.0 <sup>b</sup>	78.0 <sup>b</sup>	
<u>\_</u> Т	108.0	102.5	118.8 <sup>ab</sup>	120.8 <sup>ab</sup>	123.5 <sup>ab</sup>	134.0 <sup>a</sup>	134.0 <sup>a</sup>	128.3ª	124.5 <sup>a</sup>	125.0 <sup>a</sup>	126.3ª	
T	109.3	105.0	124.3 <sup>a</sup>	124.5 <sup>a</sup>	128.3 <sup>a</sup>	137.0 <sup>a</sup>	140.0 <sup>a</sup>	141.3 <sup>a</sup>	142.3 <sup>a</sup>	150.3 <sup>a</sup>	149.5 <sup>a</sup>	
[_T	105.8	103.3	118.5 <sup>ab</sup>	121.5 <sup>ab</sup>	132.8 <sup>a</sup>	133.0ª	138.5ª	139.0ª	141.5ª	142.5 <sup>a</sup>	143.5 <sup>a</sup>	
T	112.3	102.3	126.3 <sup>a</sup>	127.3 <sup>a</sup>	127.5 <sup>a</sup>	134.3 <sup>a</sup>	137.0 <sup>a</sup>	139.3ª	141.5 <sup>a</sup>	143.0 <sup>a</sup>	145.0 <sup>a</sup>	
	20.69	17.36	18.31	19.03	24.22	23.91	18.92	17.21	21.58	26.05	26.39	
ıe	0.979	0.245	0.030	0.014	0.008	< 0.001	< 0.001	< 0.001	< 0.001	< 0.001	< 0.001	

<sup>1</sup>Sun-dried=Un\_T, Urea=Urea\_T, Sodium chloride=NaCl\_T, Sodium hydroxide=NaOH\_T and Potassium hydroxide=KOH\_T.

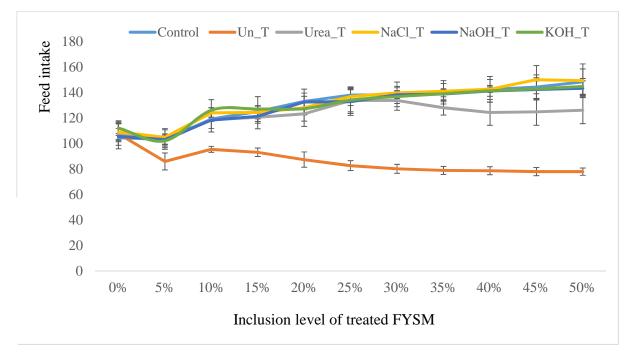


Figure 5.1 Effect of false yam seed meal (FYSM) inclusion into a maize-based control diet at varying levels on feed intake of broiler chickens. Error bars represent standard error of mean feed intake (n=4 per data point).

The results of the effect of partially replacing maize with the FYSM on apparent nutrient digestibility are presented in Table 5.4. There was no significant (P>0.05) difference in DM digestibility between birds fed the control diet and those birds fed diets containing Urea\_T, NaOH\_T and KOH\_T at any of the inclusion levels tested; the same observation was valid for up to 40% inclusion of NaCl\_T. However, there was a significant (P<0.001) reduction in DM digestibility in birds fed various levels of Un\_T.



Birds fed the control diet had a CP digestibility similar (P>0.05) to birds fed diets containing different levels of NaOH\_T, but a higher (P<0.001) CP digestibility than those fed diets containing KOH\_T. However, amongst birds on KOH\_T diets the CP digestibility was similar (P>0.05). CP digestibility was compromised beyond 20% inclusion of Urea\_T, and there was a significant (P<0.001) reduction in CP digestibility with the inclusion of Un\_T at all tested levels.

Crude fiber digestibility of the control diet was similar (P>0.05) to those diets containing various levels of Un\_T. However, all diets containing Urea\_T, NaCl\_T, NaOH\_T and KOH\_T at the various levels had a CF digestibility superior (P<0.001) to diets containing various levels of Un\_T and the control diet.

A higher (P<0.001) fat digestibility than the control diet was observed in diets containing Urea\_T, NaCl\_T, NaOH\_T and KOH\_T beyond the 10% inclusion level. However, fat digestibility of the control diet was comparable to the diets containing 10% NaOH\_T and Urea\_T, but higher than diets containing Un\_T. The latter ingredient decreased fat digestibility by trend as inclusion levels increased.

Generally, the digestibility of NFE was quite high among the treatment diets, with the minimum average digestibility of 77.4% observed for the 50% Un\_T diet and the maximum average digestibility of 93.8% obtained for the 20% KOH\_T diet. Furthermore, NFE digestibility of KOH\_T diets was superior to NaCl\_T and NaOH\_T diets which had similar (P<0.05) mean values. Urea\_T diets were of higher NFE digestibility than Un\_T diets, even though a quite high average of 84.3% was calculated for the latter.

The digestibility of ash in the control diet was similar to treatment diets, with the exception of a high (P<0.001) ash digestibility in the 30% Urea\_T diet.



	Level			FY	'SM <sup>2</sup>				ANOVA	3
Compo-	(%)	Un_T	Urea_	NaCl_	NaOH	KOH_	Mean	Facto	LSD	P-
nent <sup>1</sup>			Т	Т	_T	Т		r		value
	0						78.48	Т	2.894	< 0.001
	10	74.7	77.3	76.5	75.8	75.6	76.08	L	2.894	0.170
	20	71.4	77.9	76.2	79.4	76.2	76.34	ΤxL	5.202	0.005
DM	30	69.5	76.4	75.1	80.2	78.0	75.97			
	40	65.7	74.8	75.6	78.7	78.6	74.79			
	50	60.1	75.8	74.2	79.6	77.4	73.55			
	Mean	68.26	76.44	75.51	78.75	77.14	75.55			
	0						74.32	Т	3.722	< 0.001
	10	70.5	75.9	67.5	69.7	70.3	71.08	L	3.722	< 0.001
	20	61.4	72.9	65.2	76.6	68.9	69.31	ΤxL	6.691	< 0.001
СР	30	55.3	57.2	67.4	77.5	69.5	65.68			
	40	55.9	54.7	64.8	74.8	68.7	64.10			
	50	46.5	56.6	62.0	77.5	68.5	62.52			
	Mean	57.89	63.47	65.37	75.23	69.19	67.04			
	0						50.29	Т	2.940	< 0.001
	10	50.70	59.69	63.47	71.35	68.69	62.24	$\mathbf{L}$	2.940	0.409
	20	48.98	64.59	64.96	71.42	69.65	63.86	ΤxL	5.284	0.361
CF	30	48.72	65.78	64.63	70.11	72.43	63.80			
	40	48.38	68.96	64.91	71.32	71.84	64.55			
	50	48.17	69.08	65.54	71.53	70.91	64.51			
	Mean	48.99	65.62	64.70	71.15	70.70	62.89			
	0						50.88	Т	4.063	< 0.001
	10	44.5	52.2	42.7	48.0	43.1	45.86	$\mathbf{L}$	4.063	< 0.001
	20	40.6	62.6	65.5	62.8	67.9	59.64	ΤxL	7.304	< 0.001
EE	30	36.0	67.0	65.4	63.9	70.3	60.28			
	40	34.9	66.6	64.8	64.5	68.6	59.64			
	50	32.9	66.4	53.5	70.8	72.5	58.98			
	Mean	37.76	62.96	58.38	62.00	64.50	56.50			
	0						91.12	Т	1.217	< 0.001
	10	86.6	86.3	88.5	88.1	87.4	87.49	L	1.217	< 0.001
	20	88.3	88.1	89.8	89.4	93.8	89.97	ΤxL	2.188	< 0.001
NFE	30	86.1	87.8	88.7	89.8	91.8	88.95			
	40	83.3	88.1	89.5	89.9	91.5	88.56			
	50	77.4	87.4	88.0	90.9	91.2	87.06			
	Mean	84.32	87.52	88.90	89.63	91.13	88.58			
	0						32.90	Т	5.307	< 0.001
	10	28.7	43.0	28.3	26.9	34.1	32.19	$\mathbf{L}$	5.307	0.808
	20	26.9	38.4	28.4	27.1	37.3	31.61	ΤxL	9.539	0.954
Ash	30	29.4	42.4	30.8	34.6	36.1	34.65			
	40	28.0	37.4	29.3	33.8	35.7	32.81			
	50	30.4	37.2	29.4	32.9	34.7	32.92			
	Mean	28.64	39.67	29.24	31.05	35.57	32.84			

Table 5.4 Effect of the 5 false yam seed meal (FYSM) at varying levels on apparent nutrient digestibility (%) of broiler chickens (n= 4 per treatment)

<sup>1</sup>DM= Dry matter, CP= Crude protein, CF= Crude fiber, EE= Ether extract, NFE=nitrogen free extractives.

 $^{2}$ Un\_T= Untreated, Urea\_T= Urea treated, NaCl\_T= Sodium chloride treated, NaOH\_T= Sodium hydroxide treated, KOH\_T= Potassium hydroxide treated.

<sup>3</sup>Two-way ANOVA and post-hoc Tukey HSD test; T= treatment, L=Level, LSD= Least significant difference, P= Probability.

#### **5.6.0 DISCUSSION**

The very similar feed intake observed in the preference test between birds fed the control diet and those consuming diets containing NaCl\_T, NaOH\_T and KOH\_T FYSM, demonstrates the effectiveness of the sequential use of water-based and chemical treatment methods in the removal of the bitter anti-nutritional factors in the false yam seeds as opposed to Un\_T FYSM. Bitter triterpenoids are known to reduce feed palatability (Musalia et al., 2000; Elangovan et al., 2000), whereas alkali treatment and urea ammoniation have been found to improve feed intake of broiler chickens (Nagalakshmi et al., 1996, 1999) and without affecting their growth and nutrient utilization. Since birds have taste sensors for salt and bitterness (Fairchid et al., 2005), it seems evident that the processing methods in this study had the potential of debittering the toxic compounds and possibly also modified the chemical structures of some anti-nutritional factors in the false yam seed meal due to chemical reactivity. The feed intake of birds on diets containing Un\_T and Urea\_T suggest that these FYSM still contained high residual bitter compounds that compromised feed intake beyond inclusion levels of 5% (Un\_T) and 30% (Urea\_T), respectively. The presence of anti-nutritional factors in feedstuffs has been reported to affect feed digestibility and utilization by animals (Lange *et al.*, 2000). Food processing methods can help control potential adverse effects of plant compounds that are toxic to humans or animals when such foods are consumed. Previous studies involving the processing of false yam seeds by soaking in water before drying improved its nutritional value for broiler chickens from 3% up to 9% in a grower diet (Dei et al., 2013). Soaking the seeds in saltpetre solution further improved its nutritive value by up to 12% in broiler grower diets (Dei et al., 2015). This indicates that nutritional improvement of false



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yam products should go beyond water treatment to include the use of additives as seen in this study. The improvement in DM and CP digestibility of FYSM containing diets when the unconventional ingredient was sequentially treated with water-based and chemical treatment methods suggests significant reduction in some anti-nutritional factors as compared to the only sun-dried but further untreated (Un\_T) ingredient.

A previous experiement in this study (Experiment 1, Figure 4.1 and 4.2) involving the sequential use of water-based and chemical treatment methods was effective in reducing total terpenes in FYSM from 2.97 mg/g DM to 0.24 mg/g DM and saponins from 12.5 mg/g DM to 2.9 mg/g DM. This could be due to a washing out of these compounds or a modification of their structures through chemical reactivity. According to Pommer (2003), carbon-carbon double bonds in the structure of some terpenes make the molecules reactive towards oxidizing agents. The significant reduction of total terpenes (92%) and saponins (77%) as observed in this study, could enhance the digestibility of nutrients as seen in this study. D'Mello and Walker (1991) achieved considerable success by using the alkali potassium bicarbonate to detoxify jack beans.

Anti-nutritional factors react with protein, enzymes, or essential amino acids and form various complexes, thus affecting digestibility and nutrient utilization in poultry (Pekel *et al.*, 2015). Addition of Un\_T FYSM to the broiler diets had adverse effects on crude protein digestibility, which showed a decreasing trend with increasing inclusion levels. This could be attributed to the presence of anti-nutrients in the false yam seed (Fay, 1991). According to Payne (1990), the digestibility of a feed is influenced by its chemical components, and since the major contributor to the differences in the diets was the false yam seed meal, it suggests that anti-nutritional factors in the Un\_T FYSM have influenced the utilization of protein in those diets as against diets containing treated false yam seed meals. According to McDonalds *et al.* (1995) terpenes can actually impair



the availability of nutrients and reduce feed digestibility in animals as observed in this study. Saponins are a heterogeneous group of naturally occurring foam-producing triterpenes that occur in a wide range of plants (Jenkins and Atwal, 1994) including false yam seeds. They affect animal performance and metabolism, leading to enzyme inhibition and reduction in nutrient absorption (Cheeke, 1971). Saponins have been shown to bind to the cells of the small intestine, thereby affecting the absorption of nutrients across the intestinal wall (Johnson et al., 1986). The enhanced nutrient digestibility of diets containing the treated false yam seed meals could be attributed to the reduction or modifications of anti-nutrients in these seed batches. Generally, digestibility of nutrients was highest in false yam seed samples treated with NaOH and KOH.

# 5.7.0 CONCLUSIONS AND RECOMMENDATION

The sequential use of water-based and chemical treatment methods was effective in improving nutrient digestibility of false yam seed meal-based diets. The preferred treatment methods identified in this study are sodium hydroxide and potassium hydroxide treatments. These improved diet digestibility up to a 50% replacement of maize by treated false yam seed meals.

Even though this is an enhanced inclusion level of treated false yam seed meals in poultry diets, the time needed to soak the seed, coupled with costs of chemicals, might limit the benefits of false yam utilization in poultry production. Since an improved nutrient digestibility does not necessarily translate completely into a better nutrient utilization, there is a need to assess the nutrient metabolisability by testing the inclusion of the treated false yam seed meals in rations of growing broiler chickens.

# **CHAPTER 6**

# 6.0 EXPERIMENT 3: NUTRIENT METABOLISABILITY OF TREATED FALSE YAM (ICACINA OLIVIFORMIS) SEED MEALS FOR BROILER CHICKENS

# **6.1.0 INTRODUCTION**

There is a relative lack of information on the availability of the nutrients contained within the 4 treated false yam seed meals for poultry. There is therefore a need to evaluate the variability in nutrient availability between the 4 treated false yam seed meal samples. In the previous experiment (**see Experiment 2**) of the study the apparent nutrient digestibilities of the 4 treated false yam seed meals were varied. However, it is not known how the 4 treated false yam seed meal samples with varying residual anti-nutrients would influence dietary nutrient availability for productive functions of broiler chickens.

#### **6.1.1 OBJECTIVES**

- To determine dietary apparent metabolizable energy (AME) of the diets containing the 4 treated false yam seed meals (Urea\_T, NaCl\_T, NaOH\_T and KOH\_T).
- To determine apparent nutrient digestibility coefficient and gross energy metabolisability in the 4 treated false yam seed meals.

# 6.1.2 HYPOTHESIS

• Apparent metabolizable energy (AME) of diets and nutrient digestibility and gross energy metabolisability of the 4 treated false yam seed meals will not differ when included in broiler chicken diets.

#### 6.2.0 MATERIALS AND METHODS

#### **6.2.1 Experimental samples**

The 4 treated false yam seed meals were prepared as described in Experiment 1 (Section 4.2.1).

#### **6.2.2** Broiler bioassay

Cobb 500 broiler chicks were reared in a deep-litter-floored pen and fed broiler starter diet (CP=220.0 g/kg, ME=12.2 MJ/kg) for 21 days. The experiment was a factorial design (5 false yam seed meal samples and 5 dietary levels) with one additional control (no treated false yam seed meals). A total of 108 chicks (54 males, 54 females) aged 4 weeks and with similar body weights  $(825g \pm 1.23)$  were selected and randomly divided into 5 groups of 20 birds per group and each group was subdivided into 5 treatment groups of 4 birds housed in individual wire-mesh floor cages (0.4 m x 0.3 m = 0.12 m<sup>2</sup>/chick) in a Completely Randomized Block Design. The control group had 8 birds (8 replications) to enhance the reliability of the control data, since metabolisability values would be derived from these data. The five types of FYSM were tested at inclusion levels of 10, 20, 30, 40 and 50%, respectively, replacing maize (wt. / wt.) in a maize-fish meal-based diet (Table 5.2). Each bird received one of the 26 dietary treatments for the period of 15 days. The first 10 days constituted the preliminary stage of the trial, where birds were allowed to adapt to their new environment as well as new diets. The last 5 days were used for data collection. During this period, feed and water were provided *ad libitum* and light was provided 24 hours with 10 lux of light intensity. Light is usually provided throughout the night in the northern Guinea Savanna zone to stimulate feed intake, which is depressed by the prevalent high daytime temperatures.

Weighed quantities of the diets were supplied and faeces collected in plastic sheet placed under the wire-mesh floor of the cages using total collection method. Faeces were collected every 24



hours, weighed and stored under cool temperature (4°C in refrigerator). At the end of the trial, the daily samples collected from chickens in each replicate cage were pooled into one sample per treatment, oven dried (60°C), weighed, ground and stored in airtight plastic containers. Triplicate samples of treatment diets and dried faeces were analysed for proximate components in accordance with standard methods described by AOAC (2000). The gross energy of experimental feed and excreta was calculated using the formula of Anderson *et al.* (2011).

# Calculations

The apparent metabolizable energy (AME) values of the diets were calculated from the gross energy (GE) values of the diets and excreta using the following:

 $AME_{diet} = [(feed intake x GE_{diet}) - (excreta output x GE_{excreta})]/Feed intake Equation 6.1$ An apparent nutrient digestibility coefficient estimate of each sample was derived according to calculation of digestibility of a single feed of a mixed ration (Lloyed et al., 1978).

$$S = A + [100 (T - A)]$$
Equation 6.2

 ${f S}$  is the coefficient of apparent digestibility of the test feed ingredient



A is the coefficient of digestibility of the basal diet

 $\mathbf{T}$  is the coefficient of digestibility of the combination of the basal feed plus test ingredient

 ${\bf s}$  is the proportion of test feed ingredient in the mixed diet (T)

#### 6.3.0 RESULTS

Apparent metabolizable energy (AME) values of the experimental diets (Table 6.1) indicated an increasing trend of AME values as the level of FYSM samples were increased in the diets (Figure 6.1). Generally, all the treatment diets showed a higher AME content. However, among the treatment diets, urea, potassium hydroxide and sodium hydroxide-treated FYSM-based diets had the highest (P<0.001) AME values than the untreated FYSM.

The nutrient metabolisability coefficients of the treated FYSM samples are presented in Table 6.2. Varying inclusion levels of each treated FYSM sample did not vary (P>0.05) the coefficients of metabolisability of dry matter (DM) and crude protein (CP). However, in terms of methods of processing, DM metabolisability was significantly higher (P<0.001) in the FYSM samples that were sequentially treated with water-based and chemical treatment methods than in the untreated sample (Table 6.2). The sodium hydroxide-treated FYSM sample had higher (P<0.001) CP metabolisability than its counterparts. Urea and potassium hydroxide-treated samples had similar (P>0.05) CP metabolisability, with the untreated sample recording the lowest (P<0.001) CP metabolisability.



Generally, sequentially treated FYSM samples had increasing values of carbohydrate metabolisability as their levels were increased in the diets. The untreated sample had a significant (P<0.001) reduction in carbohydrate metabolisability beyond 30% inclusion level (Table 6.4). Carbohydrate metabolisability was higher (P<0.001) in potassium hydroxide-treated FYSM sample and lower (P<0.001) in the untreated sample. Improvement (P<0.001) in gross energy metabolisability was observed in sodium chloride, sodium hydroxide and potassium hydroxide-treated samples as their levels were increased in the diets. However, the untreated sample had a declining (P<0.001) gross energy metabolisability as its level was increased in the diets. Among the sequentially treated FYSM samples, sodium hydroxide-treated samples had the highest

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(P<0.001) gross energy metabolisability, while the untreated sample recorded the lowest (P<0.001) gross energy metabolisability. The interaction between level of inclusion and treatment methods (P < 0.017) in the present study showed that treatment methods had different effects on gross energy metabolisability value of FYSM samples depending on the level of inclusion.

Table 6.1: Apparent metabolizable energy of experimental diets

	Level			FYSM <sup>1</sup>		ANOVA				
Component	(%)	Un_T	Urea_T	NaCl_T	NaOH_T	KOH_T	Mean	Factor	LSD	Р
	0						14.8	Т	0.543	<.001
AME	10	16.1	16.9	16.3	16.4	16.3	16.4	L	0.543	<.001
(MJ/Kg,	20	17.3	18.9	18.3	19.2	18.2	18.4	ΤxL	0.976	<.001
DM)	30	19.1	20.1	20.3	21.4	20.7	20.3			
	40	20.4	23.1	22.3	23.1	22.9	22.4			
	50	21.2	24.2	23.9	25.3	24.8	23.9			
	Mean	18.8	20.6	20.2	20.6	21.1	<i>19.8</i>			

<sup>1</sup>Un\_T= Untreated, Urea\_T= Urea treated, NaCl\_T= Sodium chloride treated, NaOH\_T= Sodium hydroxide treated, KOH\_T= Potassium hydroxide treated, LSD=least significant difference, P= probability

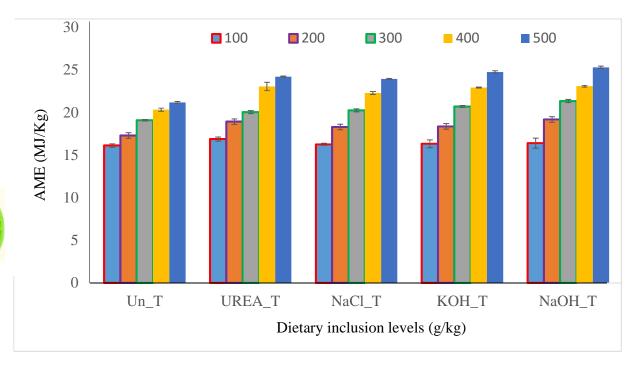


Figure 6.1: Trend of AME values of experimental diets. Where visible, error bars represent standard error of mean feed intake (n=4 per data point).



Variable	Level	Un_T FYSM	UREA_T FYSM	NaCl_T FYSM	NaOH_T FYSM	KOH_T FYSM	Mean	Factor	LSD	Р
	10	0.51	0.78	0.69	0.62	0.60	0.64	S	0.148	< 0.001
	20	0.48	0.81	0.72	0.88	0.72	0.72	L	0.148	0.798
DM	30	0.51	0.74	0.70	0.87	0.80	0.72	S x L	0.330	0.997
	40	0.48	0.71	0.73	0.81	0.81	0.71			
	50	0.43	0.74	0.71	0.82	0.78	0.7			
	Mean	0.48	0.76	0.71	0.80	0.74	0.698			
	10	0.56	1.10	0.26	0.48	0.54	0.59	S	0.193	< 0.001
	20	0.18	0.76	0.37	0.95	0.56	0.56	L	0.193	0.869
СР	30	0.16	0.23	0.56	0.90	0.63	0.5	S x L	0.431	0.011
	40	0.32	0.29	0.54	0.79	0.64	0.52			
	50	0.20	0.41	0.52	0.83	0.65	0.52			
	Mean	0.29	0.56	0.45	0.79	0.61	0.539			
	10	0.51	0.47	0.70	0.65	0.58	0.58	S	0.064	< 0.001
	20	0.79	0.78	0.87	0.84	0.96	0.85	L	0.064	< 0.001
СНО	30	0.76	0.81	0.84	0.88	0.95	0.85	S x L	0.144	0.202
	40	0.72	0.84	0.88	0.89	0.93	0.85			
	50	0.64	0.84	0.85	0.91	0.92	0.83			
	Mean	0.68	0.75	0.83	0.84	0.89	0.795			
	10	0.76	0.79	0.77	0.77	0.76	0.77	S	0.018	< 0.001
	20	0.74	0.81	0.79	0.82	0.79	0.79	L	0.018	< 0.001
Gross	30	0.75	0.81	0.80	0.84	0.82	0.81	S x L	0.040	0.017
energy (AME/GE)	40	0.75	0.87	0.82	0.84	0.84	0.83			
(AME/GE)	50	0.73	0.83	0.82	0.86	0.84	0.82			
	Mean	0.75	0.82	0.80	0.83	0.81	0.803			

Table 6.2: Nutrient digestibility and gross energy metabolisability coefficients in false yam
seed meal samples

Un\_T= Untreated, Urea\_T= Urea-treated, NaCl\_T= Sodium chloride-treated, NaOH\_T= Sodium hydroxide-treated, KOH\_T= Potassium hydroxide-treated, FYSM= False yam seed meal, LSD= Least significant difference, P= Probability. S=Sample, L=Level.



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#### 6.4.0 DISCUSSION

The lower apparent metabolizable energy values in the untreated seed sample could be attributed to the high anti-nutrient contents (Table 6.1). High anti-nutrient contents of a meal could have a pronounced negative effect on its metabolizable energy (Smulikowska et al., 2001). The results of this study suggest that anti-nutrients have an energy effect in poultry diets. These results are consistent with the findings of Farrell *et al.* (1993), who reported significant increases in the AME of sorghum-soya bean meal-based diets with phytase addition. The fact that these negative effects on AME were overcome by sequential use of water-based and chemical treatment methods as seen in this study, anti-nutrients may be partly responsible for the observed depressions in AME values in the untreated FYSM (Figure 6.1).

This may explain the lower CP metabolisability in the untreated than the treated FYSM samples. The low nutrient metabolisability values of the untreated false yam seed meal recorded in this experiment could be due to the high amount of anti-nutrients. Although, the terpenes and saponin contents of the meal could be implicated, Dei *et al.* (2011) reported that high concentration of terpenes in soaked false yam tuber meal fed to experimental birds at increasing level may have been responsible for the poorer growth performance of the birds due to poor nutrient utilisation. According to MacDonald *et al.* (2002), terpenes actually impair the availability of nutrients, particularly protein and reduce performance when fed to animals. Terpenes, reported to be constituents in false yam (Vanhaelen et al., 1986), are plant biosynthetic substances, many of which have pronounced effects on animal metabolism (Frohne and Pfander, 2005). The interactions of anti-nutrients (terpenes) and proteins to form protein complexes have been described (Anderson, 1985) and the nutritional significance of these complexes are insoluble



and less subject to attack by proteolytic enzymes than the uncomplexed protein (Cheryan, 1980). All the false yam seed meal samples had residual terpenes and saponin contents, and in particular, the untreated false yam seed samples had very high terpenes and saponin levels (Figures 4.1 and 4.2). This indicates that the residual anti-nutrients in the false yam seed meals may have contributed significantly to its ME content. The false yam seed meal samples treated with sequential use of water-based and chemical treatment methods had high metabolisability (AME/GE; mean of 0.82) compared with the untreated false yam seed meals may have a deleterious effect on ME, although the sequential treatment methods employed in this study reduced the terpenes concentration by 55.2 to 91.9% and saponin concentration by 54.4 to 76.8% in the false yam seed meal samples. The processing conditions, the presence of anti-nutritional factors and dietary fibre are some factors that influence digestibility and nutrient metabolisability of feed (Leeson and Summers, 2002).

#### 6.5.0 CONCLUSION AND RECOMMENDATION

In conclusion, sodium hydroxide and potassium hydroxide-treated false yam seed meal showed higher AME and better nutrient and gross energy metabolisability values. The content and nature of the anti-nutrients in the false yam seed meal samples as well as the method of processing probably accounted for most of the variability observed in available nutrient concentrations. Therefore, the residual anti-nutrient content is important quality variable of false yam seed meal that affects its nutrients and energy metabolisability values for poultry. It is recommended that varying inclusion levels of nutritionally improved false yam seed meals in diets of broiler chickens be fed to ascertain their effects on performance.



#### **CHAPTER 7**

## 7.0 EXPERIMENT 4: EFFECT OF VARYING LEVELS OF SODIUM HYDROXIDE-TREATED FALSE YAM SEED MEAL ON GROWTH PERFORMANCE, HAEMATOLOGY AND SERUM BIOCHEMISTRY OF FEMALE BROILER CHICKENS

## 7.1.0 INTRODUCTION

The previous experiment (Experiment 3) of the study have demonstrated high availability of nutrients in the sequentially treated false yam seed meals, especially the sodium hydroxide-treated (NaOH\_T) false yam seed meal for broiler chickens. So far, there is no available published report that has shown the relationship between dietary inclusion levels of the sodium hydroxide-treated (NaOH\_T) false yam seed meal and growth performance variables. However, there is a published report (Mohammed et al., 2017) that has shown the relationship between dietary inclusion levels of saltpetre-treated false yam seed meal and laying performance variables in Lohman brown layers. Therefore, there is a need for evaluation of the growth response of broiler chickens fed NaOH\_T FYSM. This experiment was carried out using ingredient (maize) substitution method to establish a relationship between the dietary level of NaOH\_T FYSM and growth performance variables.

## 7.1.1 OBJECTIVES

The specific objectives were to determine the effects of increasing dietary levels (0, 10, 20, 30, 40 and 50%) of NaOH\_T FYSM on the:

- Growth performance of broiler chickens
- Blood biochemistry of broiler chickens
- Carcass characteristics of broiler chickens
- Sensory characteristics of broiler chicken meat
- Proximate and peroxide value of broiler chicken meat.

## 7.1.2 HYPOTHESIS

• The growth performance, blood biochemistry and carcass characteristics of broiler chickens will not differ when sodium hydroxide-treated false yam seed meal in maize-based diet is fed to them.



#### 7.2.0 MATERIALS AND METHODS

#### 7.2.1 False yam seed meal sample

This was prepared as described in Experiment 1 (Section 4.2.1).

#### 7.2.2 Broiler bioassay

Mixed sex day-old broiler chicks (Cobb 500) were obtained at day-old from a local hatchery in Dormaa and reared in a deep-litter floored brooder house and fed a starter broiler diet for 21 days. At 21 d of age, 128 female birds were individually weighed and then randomly assigned to one of four dietary treatments in quadruplicate lots. Each replicate had 8 female broilers. The mean liveweight of birds in each replicate was 824 g ( $\pm$ 0.01). The four treatments (Table 7.1) included the control without NaOH\_T FYSM; Treatments 2, 3 and 4 contained 10%, 30% and 50% sodium hydroxide treated false yam seed meal (NaOH\_T FYSM) respectively replacing maize (wt. /wt. basis) (Table 7.1). The diets were formulated to contain adequate levels of required nutrients for growing broiler chickens. Feed and water were given *ad libitum*. The experiment lasted 35 days. Each replicate lot of experimental birds was kept in a pen (0.16m<sup>2</sup>/bird) within an open-sided conventional house in a completely randomized design. Artificial light was provided in the night as a strategy to encourage feed intake at night to check the anorexic effects of the high day time temperatures (as high as 40°C) in the Savannah regions of Ghana during the experimental period.





	Tr	Treated false yam seed meal levels in diets					
Ingredient (g/kg)	Control	10%	30%	50%			
Maize	620	558	434	310			
NaOH_T FYSM	0	62	186	310			
Fish meal	115	109	125	141			
Wheat bran	98	81	65	49			
Soybean meal	137	160	160	160			
Vitamin /Min. Premix*	2.5	2.5	2.5	2.5			
Di-calcium Phosphate	10	10	10	10			
Oyster shell	15	15	15	15			
Common salt	2.5	2.5	2.5	2.5			
Calculated nutrient analysis (g	v/Kg)						
Crude protein	200	200	200	200			
Calcium	16.2	16.8	17.4	18.1			
Phosphorus	8.8	8.3	8.3	8.3			
Lysine	10.9	11.1	11.3	11.4			
Methionine	4.4	4.3	4.3	4.4			
M.E (kcal/kg)	2905.2	2882.1	2871.6	2850.5			

#### Table 7.1: Composition and nutrient contents of experimental diets

\*Composition of vitamin/trace mineral premix per kg diet: vitamin A (retinyl acetate), 5.2 mg; vitamin D3 (cholecalciferol), 0.125 mg; vitamin E (DL-alpha-tocopherol), 100 mg; vitamin K3 (menadione), 5 mg; vitamin B1(thiamine), 2 mg; vitamin B2 (riboflavin), 9 mg; vitamin B3 (Niacin), 50 mg; vitamin B5 (Calcium pantothenate), 25 mg; vitamin B6 (pyridoxine), 7 mg; vitamin B8 (biotin), 0.3 mg; vitamin B9 (folic acid), 3 mg; Vitamin B12 (cyanocobalamin), 0.24 mg; Fe (FeSO<sub>4</sub>), 90 mg; Cu (CuSO<sub>4</sub>), 5 mg; Mn (MnO), 120 mg; Co (CoSO<sub>4</sub>), 1 mg; Zn (ZnSO<sub>4</sub>), 100 mg; I (Ca(IO3)<sub>2</sub>), 2 mg; and Se (Na<sub>2</sub>SeO<sub>3</sub>), 0.4 mg. NaOH\_T FYSM= sodium hydroxide treated false yam seed meal

#### 7.3.0 DATA COLLECTION

#### 7.3.1 Growth parameters

#### 7.3.1.1 Feed intake

Feed intake was obtained by subtracting the left-over feed in the feed trough at the end of the week from the total feed supplied for the week. This was measured weekly in kilograms by using digital

scale (**JADEVER JPS-1050**) to weigh the feed. Mean feed intake per bird per day was calculated by dividing the feed consumed by the number of birds in the replicate and the number of days in a

week. The answer was then multiplied by 1000 to get feed intake per bird per day in grams.



#### 7.3.1.2 Weight gain

Live-weights of birds in each replicate was measured weekly in kilograms by weighing them in batches using a digital electronic scale (Jadever, JPS-1050), and weekly live weight gains calculated by dividing total weekly live-weight gain by the number of birds in the replicate and by the number of days in a week. The answer was then multiplied by 1000 to get live-weight gain per bird per day in grams.

#### 7.3.1.3 Gain-to-feed ratio

Feed conversion efficiency was defined as live weight gain per unit feed consumed. This parameter was calculated by dividing daily live-weight gain by the amount of feed consumed per day by each replicate bird.

## 7.3.1.4 Mortality

Mortality was recorded as and when they occurred. All dead broilers were autopsied by a Veterinary officer of the Department of Veterinary Science, UDS, Tamale.

### 7.3.2 Carcass parameters



Carcass parameters included carcass dress weight, carcass dressing percentage and relative weights of gizzard, spleen, heart and liver. At the end of the feeding trial, birds were starved for 8 hours and two birds per replicate were randomly selected and slaughtered by jugular venipuncture. Carcasses were then scalded in hot water (about 60°C), de-feathered and eviscerated to get carcass dress weight. Carcass dressing percentage was calculated by dividing carcass dress weight by the bird's live weight, multiplied by 100.

After evisceration, the internal organs were separated and weighed individually. The internal organs weighed included empty gizzard, heart, liver and spleen, and expressed as a percentage of dress weight to set a relative organ weight.

#### 7.3.3 Carcass yield

The dressed carcass was chilled for 24 hours and weight taken. Primal cuts were made from the chilled carcass, and were weighed. The breast and thigh muscles were packed separately in transparent polythene bags and vacuum-sealed, then frozen (-18°C) for sensory and laboratory analyses.

#### 7.3.4 Sensory analysis

A total of fifteen (15) panellists, comprising staff members and students of the University, were randomly selected and trained according to the British Standard Institution guidelines (BSI, 1993) to evaluate the products. The breast muscles were thawed and grilled to a core temperature of 70°C in an electric oven (Turbofan, Blue seal, UK). The products were sliced into uniform sizes (about 2cm<sup>3</sup>) wrapped with coded aluminium foils and presented to the panellists. Each panellist was provided with water and pieces of bread to serve as neutralizers between the products. A five-point category scale was used to evaluate the sensory characteristics of the chicken as follows:



 Table 7.2: Five-point category scale for sensory analysis (Lim, 2011)

		Scale						
Attributes	1	2	3	4	5			
Colour	very pale red	pale red	intermediate	dark red	very dark red			
Juiciness	very juicy	juicy	intermediate	dry	very dry			
Tenderness	very tender	tender	intermediate	tough	very tough			
Chicken flavour	very weak	weak	moderate	strong	very strong			
Flavour-liking	like very much	like	intermediate	dislike	dislike very much			
Overall-liking	like very much	like	intermediate	dislike	dislike very much			

#### 7.3.5 Proximate compositions and lipid per-oxidation (Peroxide values) of the meats

The proximate composition according to AOAC (2000) and lipid per-oxidation (Salam et al., 2004) of the products were conducted and the procedures are outlined in sections *3.4.0 and 3.12.0*.

#### 7.3.6 Blood biochemistry

At the end of the feeding trial, two birds from each replicate treatment were randomly selected for blood sampling. The selected birds were restrained and 2 mL of blood were drawn from their wing veins with a syringe and needle. Blood samples for haematological evaluation were collected into EDTA-containing tubes, while blood samples for serum chemistry evaluation were collected without anticoagulant and span in a centrifuge to generate serum. Samples were kept in cooled condition prior to analysis.

The following haematological parameters were assessed; packed cell volume (PCV, %) following Mukherjee (2005), red blood cell count (RBC × 106/µL) following Dacie and Lewis (2000), white blood cell count (WBC × 103/µL) following Holfbrand and Petit (2000), haemoglobin (Hb, g/dL) following Cheesbrough (2001), white blood cell differentials (heterophils, lymphocytes, eosinophils, monocytes, basophils; all %), mean corpuscular haemoglobin concentration (MCHC, g/dL), mean corpuscular haemoglobin (MCH, pg), and mean corpuscular volume (MCV, µm<sup>3</sup>) using a haemo-analyser (Sysmex Hematology Analyser, XS-500i, Sysmex Europe GmbH, Norderstedt, Germany). The serum biochemical assay was carried out using spectrophotometry. The liver function parameters included total serum proteins (g/L), albumin (g/L), globulins (g/L), alkaline phosphate (units (U)/L), aspartate transferase (U/L) and alanine transferase (U/L), and the renal function parameters were chloride (µmol/L), creatinine (mmol/L), sodium (mmol/L), potassium (mmo/L) and urea (mmol/L).



#### **7.3.7 Economics of feeding**

The cost of feed per kg and the cost of feed consumed per bird to gain a kg body weight were the bases for the economics of production. The cost was calculated based on the prevailing market prices for the feed ingredients at the time of the experiment (Table 7.3). The cost of feed required to produce a kg weight gain was determined by multiplying the feed conversion ratio by the cost per kg weight of feed for the various treatments. Selling price was determined by the prevailing market price (GH¢10.45) per kg dressed weight of the birds at the time of the experiment multiplied by the number of dressed birds in each treatment.

Table 7.3: Feed ingredients and their	prices	per	kg
---------------------------------------	--------	-----	----

Ingredients	Price per les (CH/d/les)	
	Price per kg (GH¢/kg)	
Maize	1.50	
*NaOH_T FYSM	0.86	
*KOH_T FYSM	0.86	
Fish meal	5.60	
Wheat bran	0.8	
Soybean meal	3.2	
Vitamin /Min. Premix	10.00	
Di-calcium Phosphate	2.00	
Oyster shell	0.48	
Common salt	1.00	

\*The feed cost/kg NaOH\_T FYSM and KOH\_T FYSM was computed from the labour and processing cost at the time of the experiment as the false yam seeds were not bought.



## 7.4.0 STATISTICAL ANALYSIS

All variables measured were assessed for normality by the Shapiro-Wilk test. Normally distributed data were subjected to one-way Analysis of Variance (ANOVA) and *post-hoc* Tukey's honest significant difference (HSD) test with 95% family-wise confidence level. All other data were subjected to the Kruskal-Wallis rank sum test, followed by the Wilcoxon rank sum test for pairwise comparison of treatments.

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## 7.5.0 RESULTS

The growth response of female broiler chickens fed diets supplemented with varying levels of NaOH\_T FYSM as shown in Table 7.4 revealed no significant (P>0.05) difference in all growth parameters measured. Figure 7.1 shows the trend of growth of birds from 21 to 56 days of age.

Table 7.4: Growth performance of broiler chickens fed diets containing varying levels of sodium hydroxide-treated (NaOH\_T) false yam seed meal (21-56 days).

	Inclusion					
Variable	Control	100	300	500	SEM	Р
Initial weight (Kg)	0.825	0.825	0.825	0.824	0.0016	0.948
Feed intake (g)	111.2	111.1	117.0	111.5	3.05	0.475
Weight gain (g)	59.0	58.8	60.9	63.1	2.13	0.482
Gain-to-feed ratio	0.53	0.53	0.50	0.53	0.019	0.634
Final live-weight(Kg)	2.89	2.88	2.96	3.03	0.075	0.490
Mortality (%)	0.50	0.25	0.50	0.50	0.280	0.894

SEM: standard error of mean, P: Probability.

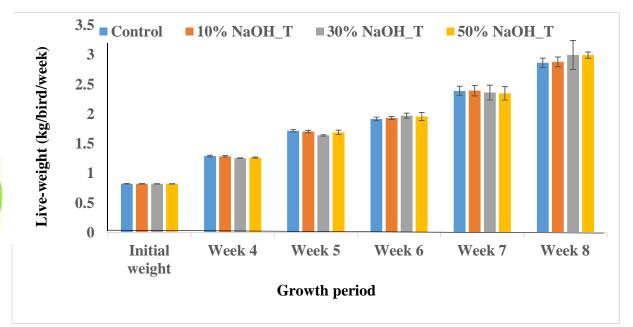


Figure 7.1: Growth performance of broiler chickens fed control diet and diets containing NaOH\_T FYSM. Error bars represent standard error of mean live-weight (n= 8 per data point).



Haematological parameters of experimental birds were similar (P>0.05), except for red blood cell count which revealed an increasing trend (P<0.045) of the cells as the treated false yam seed meal was increased in the diets (Table 7.5).

Table 7.5: Haematology of broiler chickens fed diets containing varying levels of sodium
hydroxide-treated (NaOH_T) false yam seed meal (21-56 days)

	Inclusion levels of NaOH_T FYSM (g/kg)							
Variable	Control	100	300	500	SEM	Р		
RBC (x10 <sup>12</sup> /L)	2.07 <sup>b</sup>	2.18 <sup>ab</sup>	2.25 <sup>ab</sup>	2.29 <sup>a</sup>	0.057	0.045		
PCV (%)	28.43	29.12	29.18	29.77	0.751	0.659		
Hb (g/dL)	6.55	6.58	6.63	6.75	0.168	0.840		
MCV (fL)	131.6	130.9	130.9	130.3	1.12	0.991		
MCH (Pg)	29.27	29.09	20.41	29.45	0.567	0.968		
MCHC (g/dL)	21.75	21.88	22.75	22.50	0.534	0.672		
WBC $(x10^{9}/L)$	238.2	246.1	245.6	244.4	15.89	0.719		
Lymphocytes (%)	47.1	48.0	47.8	47.6	3.44	0.570		
Neutrophils (%)	44.19	44.03	44.44	45.13	1.931	0.897		
Basophils (%)	0.13	0.11	0.11	0.13	0.019	0.875		
Eosinophils (%)	0.065	0.068	0.070	0.070	0.0063	0.952		
Monocytes (%)	9.30	9.36	9.06	9.12	0.359	0.913		

RBC: red blood cell count; PCV: packed cell volume; Hb: haemoglobin; MCHC: mean corpuscular haemoglobin concentration; MCH: mean corpuscular haemoglobin; MCV: mean corpuscular volume; WBC: white blood cell count. SEM: standard error of mean; P: probability. Means with the same superscripts within a row are not statistically different.

The results of the liver function test of birds fed diets supplemented with varying levels of treated false yam seed meal as shown in Table 7.6 revealed no significant (P>0.05) difference in all parameters determined except for blood albumin which increased (P<0.048) as the test material was increased in the diets. All the renal function parameters evaluated showed a significant

(P<0.05) increase in their values as the test material was increased in the diets (Table 7.7).



	Inclusion					
Variable	Control	100	300	500	SEM	Р
Albumin (g/L)	15.85 <sup>c</sup>	16.12 <sup>c</sup>	17.07 <sup>b</sup>	18.01 <sup>a</sup>	0.569	0.048
Globulin (g/L)	16.26	16.86	15.79	17.20	1.086	0.801
Total protein (g/L)	32.08	32.73	32.00	32.92	1.567	0.999
ALP (U/L)	151.2	151.5	148.2	150.1	2.65	0.819
ALT (U/L)	11.21	10.20	9.96	10.06	0.652	0.990
AST (U/L)	142.75	138.25	140.38	141.12	2.255	0.272

Table 7.6: Liver function test of broiler chickens fed diets containing varying levels of sodium hydroxide-treated (NaOH\_T) false yam seed meal (21-56 days)

ALP: alkaline phosphate; AST: aspartate transferase; ALT: alanine transferase. P: probability; SEM: standard error of the mean; Means with the same superscripts within a row are not statistically different.

 Table 7.7: Renal function test of broiler chickens fed diets containing varying levels of sodium hydroxide-treated (NaOH\_T) false yam seed meal (21-56 days)

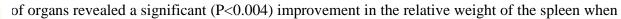
	Inclusion le	Inclusion levels of NaOH_T FYSM (g/kg)							
Variable	Control	100	300	500	SEM	Р			
Chloride (µmol/L)	65.2 <sup>b</sup>	81.6 <sup>a</sup>	84.0 <sup>a</sup>	83.5 <sup>a</sup>	3.17	< 0.001			
Creatinine (mmol/L)	14.32 <sup>b</sup>	16.51 <sup>ab</sup>	18.35 <sup>a</sup>	18.45 <sup>a</sup>	0.685	0.004			
Sodium (mmol/L)	78.9 <sup>b</sup>	107.8 <sup>a</sup>	110.1 <sup>a</sup>	112.8 <sup>a</sup>	4.80	< 0.001			
Potassium (mmol/L)	1.57 <sup>c</sup>	1.86 <sup>bc</sup>	2.39 <sup>ab</sup>	2.58 <sup>a</sup>	0.144	< 0.001			
Urea (mmol/L)	0.73 <sup>b</sup>	0.82 <sup>ab</sup>	0.95 <sup>ab</sup>	0.99 <sup>a</sup>	0.063	0.031			

SEM: standard error of the mean; P: probability; Means with the same superscripts within a row are not statistically different.

Table 7.8 showed the carcass and relative organ weights of female broiler chickens fed NaOH\_T

FYSM supplemented diets. Carcass dressing showed a significant (P<0.003) decrease when maize

was supplemented with the treated false yam seed meal at 500g/kg. However, the relative weight



the maize was supplemented with treated false yam seed meal beyond 100g/kg (Table 7.8). All

other carcass and organ characteristics were similar (P>0.05). The primal cuts as a percentage of

dress weight (Table 7.8) as well as the sensory attributes of the chicken meat evaluated (Table 7.9)

were not also affected (P>0.05) negatively.



	Inclusio	on levels of	f NaOH_T i	FYSM		
Variable	Control	100	300	500	SEM	Р
Dress weight (Kg)	1.87	1.95	1.84	1.86	0.071	0.705
Carcass dressing (%)	79.18a	79.50a	78.54ab	77.01b	0.439	0.003
Organs (% live-weight)						
Gizzard	1.43	1.44	1.41	1.37	0.040	0.630
Heart	0.39	0.39	0.42	0.39	0.017	0.469
Liver	1.60	1.95	1.77	2.09	0.150	0.060
Spleen	$0.078^{b}$	0.081 <sup>b</sup>	0.090 <sup>ab</sup>	0.107 <sup>a</sup>	0.0055	0.004
Cut up parts (% dress weight)						
Breast	9.45	9.20	9.34	9.64	0.450	0.913
Thigh	5.99	6.28	5.83	5.58	0.198	0.111
Drum stick	5.05	4.93	4.86	4.74	0.132	0.428
Wing	4.24	4.18	4.06	3.97	0.091	0.171

# Table 7.8: Carcass and relative organ characteristics of broiler chickens fed diets containing varying levels of sodium hydroxide-treated (NaOH\_T) false yam seed meal (21-56 days)

P: probability; SEM: standard error of the mean; Means with the same superscripts within a row are not statistically different.

Table 7.9: Effect of sodium hydroxide-treated false yam seed meal on sensory characteristics
of broiler chickens

	Inclusion levels	Inclusion levels of NaOH_T FYSM (g/kg DM)						
Variable	Control	100	300	500	SEM	Р		
Colour	2.27	2.20	2.00	1.87	0.221	0.562		
Tenderness	2.00	2.00	2.07	2.40	0.245	0.612		
Juiciness	2.53	2.53	2.53	2.53	0.278	1.000		
Chicken flavour	3.20	3.20	2.93	2.87	0.288	0.774		
Flavour liking	2.00	2.20	2.13	2.07	0.265	0.957		
Overall liking	2.00	1.87	2.00	1.80	0.183	0.826		

SEM: standard error of the mean; P: probability.

The proximate components and lipid peroxidation values of the chicken breast muscles are presented in Table 7.10 and Figure 7.2 below.

The proximate composition of the chicken breast muscle analysed indicated comparable (P>0.05)

dry matter and fat contents among the treatment groups. However, crude protein content increased

(P<0.001) when NaOH\_T FYSM was increased beyond 100g/kg. Crude protein content of the



control group and the group fed diet containing 100g/kg NaOH\_T FYSM were similar (P>0.05) (Table 7.10).

Lipid peroxidation values (POV) varied significantly (P<0.001) among the treatment group with the control group recording the highest value, while the group fed diet containing 300g/kg had the least POV. Notwithstanding the significant variations, POV recorded in this experiment were below 1 meq/kg meat (Figure 7.2).

 Table 7.10: Effect of sodium hydroxide-treated false yam seed meal on proximate composition of breast muscle of broiler chickens

	Inclusion leve	Inclusion levels of NaOH_T FYSM (g/kg DM)					
Variable (%)	Control	100	300	500	SEM	Р	
Dry matter	37.6	37.6	38.0	37.0	2.43	0.992	
Crude protein	19.4 <sup>c</sup>	19.6 <sup>c</sup>	20.2 <sup>b</sup>	21.8 <sup>a</sup>	0.06	< 0.001	
Fat	3.81	3.75	4.11	4.02	0.918	0.991	

SEM: standard error of the mean; P: probability; Means with the same superscripts within a row are not statistically different.

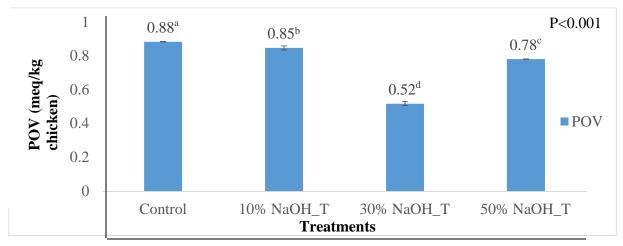


Figure 7.2: Lipid per-oxidation (Peroxide value) of broiler breast muscles fed diets containing NaOH\_T FYSM. Error bars represent standard error of mean Peroxide value (n= 4 per data point).



The economics of adding sodium hydroxide-treated false yam seed meal in the maize-based diets of female broilers are shown in Table 7.11; it was revealed that price per kg feed reduced by 16.3% when NaOH\_T FYSM was included at 500g/kg. Total feed cost per bird decreased (15.9%) (P<0.004) significantly at 500g/kg inclusion, whilst a significant (P<0.001) gain in the feed cost per kg live-weight gain was observed (21.9%) when the product was included at 500g/kg. Again, feed cost per kg dress-weight reduced by 14.7% at 500g/kg inclusion of NaOH\_T FYSM.

 Table 7.11: Economics of feeding varying levels of NaOH\_T FYSM to broiler chickens (21-56 days of age)

	Inclusion lev	Inclusion levels of NaOH_T FYS					
		(g/kg)			_		
Variable	Control	100	300	500	SEM	Р	
Price/kg feed(GHS)	2.15	2.08	1.94	1.80	-	-	
Total feed intake/bird(Kg)	3.89	3.89	4.10	3.90	0.107	0.475	
Total feed cost/bird(GHS)	8.35 <sup>a</sup>	$8.08^{a}$	7.95 <sup>a</sup>	7.02 <sup>b</sup>	0.206	0.004	
Feed cost/Kg live-weight(GHS)	4.07 <sup>a</sup>	3.93 <sup>a</sup>	3.73 <sup>a</sup>	3.18 <sup>b</sup>	0.107	< 0.001	
Feed cost/Kg dress-weight(GHS)	3.47	3.59	3.43	2.96	0.168	0.088	
Price/dressed bird(GHS)	25.41	23.72	24.23	24.84	1.480	0.696	
Gross profit/dressed bird(GHS)	17.05	15.64	16.28	17.83	1.069	0.527	

SEM: standard error of mean, P: Probability. Means with the same superscripts within a row are not statistically different.

## 7.6.0 DISCUSSION



The comparable feed intake values between the control diet and diets containing sodium hydroxide-treated false yam seed meal up to 500g/kg suggest that all the diets did not contain antinutritive compounds at leves that could avert adequate consumption of such diets. This clearly suggests that the NaOH\_T FYSM at the present dietary levels was as palatable as maize and had no adverse effect on consumption. It further indicates that, sequential use of water, sodium hydroxide and blanching treatment might have reduced the level of bitter compound (resins) in the seed. Thus anti-nutrients in diets with tolerable levels could improve feed consumption and efficiency and consequently, growth performance of birds that consume such diets.

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Feeding broiler chickens with diets containing NaOH\_T FYSM up to 500g/kg had no negative effects on their haematological and liver function values; except that of RBC and serum albumin which had seen an improvement in their values as the level of treated false yam seed meal was increased in the diets. The red blood cell levels observed for the birds on the test diets were generally higher than those on the control diet. This is an indication that the treated false yam seed meal improved the oxygen carrying capacity of the birds and did not in any way render the birds anaemic. This finding agreed with that of Ogbuewu *et al.* (2010) but differed from what was reported by Esonu *et al.* (2006).

Examination of blood sample provides the opportunity to clinically investigate the presence of several metabolites and other constituents in the body and it plays a vital role in the physiological, nutritional and pathological status of the animal (Aderemi, 2004). It also helps to distinguish normal state from state of stress which can be nutritional (Aderemi, 2004). According to Togun and Oseni (2005), haematological studies have been found useful for disease prognosis and for the therapeutic and feed stress monitoring. Adamu (2006), observed that nutrition had significant effect on haematological values like PCV, Hb and RBC. Dei *et al.* (2016) reported no adverse effects of feeding laying hens diets containing soaked false yam seed meal on their haematology and serum biochemistry. Although a measurement of enzyme activities in serum is very important diagnostic tool of bird diseases, the wide range of activity make it difficult to interpret (Harr, 2002). AST is a very sensitive, nonspecific biomarker of liver disease in birds. Conversely, ALT is of poor diagnostic value in birds due to its existence in many tissues (Harr, 2002). In this study, the mean plasma AST, ALT and ALP concentrations of the experimental birds were higher than those reported by Dei *et al.* (2016) when such birds were fed diets containing soaked false yam seed meal.

Serum chloride values in this experiment are lower than the range (108.84 to 119.1 mmol/l) reported by Siller and Wight (1997). Sodium is present mainly in the extracellular fluid and is primarily responsible for determining the volume of the extracellular fluid and its osmotic pressure and its levels are maintained within narrow limits, despite wide fluctuation in dietary intake (Ritchie et al., 1994). The normal range of serum sodium in mature birds is 130-150 mmol/l (Sturkie, 1965), which is higher than the values recorded in this experiment. The values of urea and potassium recorded are lower than those reported by Nworgu *et al.* (2007) when broiler chickens were served pumpkin leaf extract as supplement.

Kidneys are the major excretory organs and about 20–25% of the total amount of circulating toxins reach them (Harriet, 2003). The demands of kidneys for nutrients and oxygen are high because of their extensive functional load. Approximately one-third of the total blood volume is filtered through kidneys and at the same time, 98–99% of systemic water and sodium chloride are reabsorbed.

In birds exposed to the toxic effects of ANFs, haemorrhagic and fatty kidney syndrome, thickening of glomerular membranes, degenerative changes in renal epithelial cells and congestion of renal parenchyma are reported (Hussain *et al.*, 2008). At the same time, the toxic effects of ANFs on glomerular membranes and renal tubules in rats consisted in reduced glomerular filtration rate, glucose reabsorption, tubular electrolyte and organic ion transportation rate (Manseld Grunert and Kautna, 1989).

In this study, histopathology of the kidney was not evaluated, it is however believed that residual anti-nutritional factors in the treated false yam seed meal could account for the increasing values of the kidney function parameters measured.

The significant decrease in percent carcass dressing at inclusion level of 500g/kg of NaOH\_T SFYSM suggests that, nutrients in such diets could be skewed towards internal organ development whose extraction from the bird's body cavity could lead to lower carcass dressing. The higher significant values observed in the relative organ weight of the spleen could probably be due to higher physiological activities by this organ triggered by the presence of anti-nutritional factors and their concomitant effects leading to compensatory growth of the organ to counteract the negative effect of residual anti-nutrients in the false yam seed meal.

Considering the plentiful supply of food products in the world today, the concept of quality is of particular importance. Being a complex concept, poultry meat quality is understood in various ways and thus it is difficult to define conclusively. Poultry meat quality is made up of its safety, nutritive value and sensory characteristics. The nutritional quality of poultry meat depends on the content of high-value protein, unsaturated fatty acids, vitamins, macro- and micronutrients, cholesterol and other biologically active compounds. Meat colour, aroma and flavour are essential sensory traits. Simply put, it can be stated that poultry meat is of good quality if it fully meets consumer expectations (Sokolowicz et al., 2016). Teguia *et al.* (2003) reported a reduction in carcass parameters when diets containing anti-nutritional factors are fed to birds.



The inclusion of NaOH\_T FYSM in the diets of broiler chickens did not influence the primal cuts of chickens. Sensory characteristics of broiler breast meat from the various treatments were similar and overall liked by panelists. Meat purchasing decisions are influenced more by product appearance than any other quality factor (Lawrie and Ledward, 2006); colour and flavour represent perceived freshness and are of vital importance to the meat industry and meat science research (Mancini and Hunt, 2005).



The significant increase in crude protein of chicken meat as the test ingredient was increase in the diets suggests that consumers will require relatively low amounts to meet their protein needs. Meat is the major source of protein required for growth and repair of worn-out tissues in man (Lawrie and Ledward, 2006).

Lipid oxidation is a major problem encountered in the storage of chicken, mainly due to the higher levels of unsaturated fatty acids in the fat of chicken. Lipid oxidation results in quality deterioration, which is perceived in the emission of off-flavours leading to a stale, rancid flavour in foods (Kerler and Grosch, 1996). Generally, peroxide values (POV) of the products are less than 1 meq/kg meat, but significantly varied from each product. The POV obtained in this study indicated that the products were safe and could meet consumer preference and were also similar to those values reported by Hugo *et al.* (2009).

False yam seeds at present has no economic value, labour involved in harvesting and processing remained a major concern. In this experiment, cost of harvesting and processing per kg NaOH\_T FYSM was estimated. The decline in price per kg feed as NaOH\_T FYSM was increased in the feed was due to the cost of NaOH\_T FYSM which was lower than that of maize (i.e. GHS 0.86 vr.s GHS 1.50). Total feed cost per bird and feed cost per kg live-weight gain was lowest for birds fed diets containing 500 g/kg NaOH\_T FYSM. Additionally, all economic variables examined in this experiment showed a declining trend of cost per variable, suggesting that the processed FYSM have economic potential for use in poultry diets. Extensive research on non-conventional feed resources for poultry has shown that in most cases only limited amounts (up to 20%) can be used without compromising productivity of birds (Devendra, 1992).



Sequential water-based and chemical (NaOH) treatment method deployed in this study, could render false yam seeds a good replacement for maize up to 500 g/kg in broiler chicken diets.

## 7.7.0 CONCLUSION AND RECOMMENDATION

It is obvious from this experiment that, inclusion of sodium hydroxide-treated false yam seed meal up to 500g/kg in the diets of female broilers had no negative effect on their growth performance. The inclusion improved red blood cell and blood albumin values suggesting improved health. The addition of NaOH\_T FYSM in maize-based diets for broilers had no adverse effects on primal cuts, sensory attributes, proximate and peroxide values of broiler breast meat. Economics of feeding the NaOH\_T FYSM indicate a significant reduction in feed cost per kg live weight gain. There is an economic value for using this product for broiler chickens meant for live-weight sales. It is recommended that NaOH\_T FYSM can be used up to 500g/kg by farmers in broiler diet formulations.





#### **CHAPTER 8**

## 8.0 EXPERIMENT 5: EFFECT OF VARYING LEVELS OF POTASSIUM HYDROXIDE-TREATED FALSE YAM SEED MEAL ON GROWTH PERFORMANCE, HAEMATOLOGY AND SERUM BIOCHEMISTRY OF BROILER CHICKENS

## 8.1.0 INTRODUCTION

The results of the previous Experiment (*section 7.1*) have shown the potential of false yam seed meal treated in a sequential use of water treatment, chemical (NaOH) treatment and blanching as a potential method for improving the nutritive value of false yam seed meal for broiler chickens. The improvement in the nutritive value was thought to be as a result of reduction in some anti-nutritional factors, particularly terpenes and saponins concentrations. Ologhobo *et al.* (1993) reported that, in chemical treatment of seeds, higher concentration of anti-nutritional factors was found in base-soluble fractions than in other chemical fractions, indicating a greater extractability of anti-nutritional factors by alkali treatment than by acid, ether or alcohol.

In this section, another alkali (KOH), an industrial product that has the potential of reducing some anti-nutritional factors in false yam seed meal as observed in Experiment 1 (*section 4.1*) was evaluated. It is not known how false yam seed meal treated with sequential use of water treatment, chemical (KOH) and blanching would influence growth performance of broiler chickens. Also, it is not known whether the growth performance obtained with NaOH would be consistent with that of KOH. That is, if there will not be any variability in bird growth performance in the utilisation of the two nutritionally improved (NaOH\_T and KOH\_T) false yam seed meals.



## 8.1.1 Objectives

The specific objectives were to determine the effects of increasing levels (0, 10, 20, 30, 40 and

50%) of KOH\_T FYSM on the

- Growth performance of broiler chickens
- Blood biochemistry of broiler chickens
- Carcass characteristics of broiler chickens
- Sensory characteristics of broiler chicken meat
- Proximate components and peroxide value of broiler chicken meat

## 8.1.2 Hypothesis

• The growth performance, blood biochemistry and meat quality of broiler chicken will not differ when potassium hydroxide-treated false yam seed meal is used in their diets

#### 8.2.0 Materials and methods

## 8.2.1 False yam seed meal sample



The KOH\_T FYSM was used for this experiment. This was one of the samples that had been shown to have higher nutrient availability among the treated false yam seed meal samples. Also, it is not known whether the growth performance obtained, when NaOH was used as a chemical treatment agent would be consistent with the use of KOH as an agent in chemical treatment since both chemicals are alkaline in nature and also proved to enhance digestibility and metabolisability of nutrients as observed in previous studies (*section 5.1.0 and 6.1.0 respectively*).

#### **8.2.2 Broiler bioassay**

Mixed sex day-old broiler chicks (Cobb 500) were obtained at day-old from a local hatchery in Dormaa and reared in a deep-litter floored brooder house and fed a starter broiler diet for 21 days. At 21 d of age, 128 male birds were individually weighed and then randomly assigned to one of four dietary treatments in quadruplicate lots. Each replicate had 8 male broilers. The mean liveweight of birds in each replicate was 883 g ( $\pm$ 0.05). The four treatments included the control without KOH\_T FYSM; Treatments 2, 3 and 4 contained 10%, 30% and 50% potassium hydroxide treated false yam seed meal (KOH\_T FYSM), respectively replacing maize (wt. /wt. basis) (Table 7.1). The diets were formulated to contain adequate levels of required nutrients for growing broiler chickens. Feed and water were given *ad libitum*. The experiment lasted 35 days. Each replicate lot of experimental birds was kept in a pen (0.16m<sup>2</sup>/bird) within an open-sided conventional house in a completely randomized design. Artificial light was provided in the night as a strategy to encourage feed intake at night to check the anorexic effects of the high day time temperatures (as high as 40°C) in the Savannah regions of Ghana during the experimental period.

#### 8.3.0 DATA COLLECTION

Similar procedures as in Experiment 4 were used for data collection on growth variables, carcass evaluation, blood biochemistry, economics of feeding as well as quality of meat produced (*see sections 7.3.1; 7.3.2, 7.3.3, 7.3.4, 7.3.5, 7.3.6 and 7.3.7*)

#### 8.4.0 STATISTICAL ANALYSIS

All variables measured were assessed for normality by the Shapiro-Wilk test. Normally distributed data were subjected to one-way Analysis of Variance (ANOVA) and *post-hoc* Tukey's honest significant difference (HSD) test with 95% family-wise confidence level. All other data were

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subjected to the Kruskal-Wallis rank sum test, followed by the Wilcoxon rank sum test for pairwise comparison of treatments.

## 8.5.0 RESULTS

The growth response of male broiler chickens fed diets containing varying levels of KOH\_T

FYSM as shown in Table 8.1 revealed no significant (P>0.05) difference in all growth parameters

measured. Figure 8.1 shows the trend of growth of birds from 21 to 56 days of age.

 Table 8.1: Growth performance of broiler chickens fed diets containing potassium

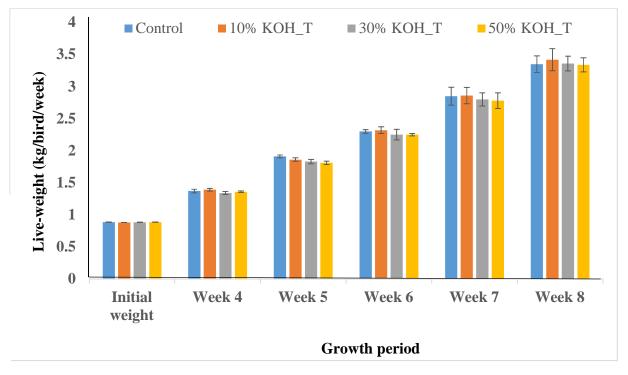
 hydroxide-treated (KOH\_T) false yam seed meal (21-56 days of age)

	Inclusion le					
Variable	Control	100	300	500	SEM	Р
Initial weight (Kg)	0.885	0.881	0.883	0.885	0.012	0.994
Feed intake (g)	121.1	125.3	133.3	133.1	4.73	0.235
Weight gain (g)	70.6	72.4	70.7	70.3	3.87	0.978
Gain-to-feed ratio	0.61	0.59	0.54	0.53	0.031	0.280
Final live-weight (Kg)	3.35	3.42	3.36	3.34	0.134	0.980
Mortality (%)	0.75	0.75	1.00	1.00	0.498	0.996

SEM: standard error of mean, P: Probability.



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**Figure 8.1:** Growth performance of broiler chickens fed control diet and diets containing varying levels of KOH\_T FYSM. Error bars represent standard error of mean live-weight (n= 8 per data point).

Table 8.2 shows the effect of KOH\_T FYSM on the haematology of broiler chickens. Haematological parameters of experimental birds evaluated revealed a significant (P<0.05) decrease in the haemoglobin, mean corpuscular volume, mean corpuscular haemoglobin and mean corpuscular haemoglobin concentration as the treated false yam seed meal was increased in the diets (Table 8.2).



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	Inclusio	Inclusion levels of KOH_T FYSM (g/kg)					
	Control	100	300	500	SEM	Р	
RBC (x10 <sup>12</sup> /L)	2.39	2.36	2.33	2.40	0.035	0.429	
PCV (%)	31.59	31.75	30.75	30.71	0.392	0.146	
Hb(g/dL)	7.74 <sup>a</sup>	7.10 <sup>ab</sup>	6.99 <sup>ab</sup>	6.75 <sup>b</sup>	0.201	0.010	
MCV (fL)	132.22 <sup>ab</sup>	135.44 <sup>a</sup>	132.34 <sup>ab</sup>	129.07 <sup>b</sup>	1.059	0.003	
MCH (Pg)	32.06 <sup>a</sup>	30.21 <sup>ab</sup>	30.23 <sup>ab</sup>	29.06 <sup>b</sup>	0.573	0.006	
MCHC (g/dL)	24.25 <sup>a</sup>	23.12 <sup>ab</sup>	22.38 <sup>b</sup>	22.12 <sup>b</sup>	0.418	0.013	
WBC ( $x10^{9}/L$ )	295.9	297.6	296.1	296.9	9.64	0.999	
Lymphocytes (%)	59.4	59.8	57.6	57.0	4.13	0.952	
Neutrophils (%)	28.5	28.4	28.1	30.7	2.04	0.792	
Basophils (%)	0.080	0.083	0.078	0.078	0.015	0.412	
Eosinophils (%)	1.16	1.19	1.12	1.14	0.079	0.942	
Monocytes (%)	7.37	7.29	7.31	7.46	0.319	0.987	

Table 8.2: Haematology of broiler chickens fed diets containing potassium hydroxide-
treated (KOH_T) false yam seed meal (21-56 days of age)

RBC: red blood cell count; PCV: packed cell volume; Hb: haemoglobin; MCHC: mean corpuscular haemoglobin concentration; MCH: mean corpuscular haemoglobin; MCV: mean corpuscular volume; WBC: white blood cell count. SEM: standard error of mean; P: probability. Means with the same superscripts within a row are not statistically different.

The results of the liver function test of birds fed diets containing varying levels of treated false yam seed meal as shown in Table 8.3 revealed no significant (P>0.05) difference in all parameters determined; except for blood albumin which decreased (P<0.047) as the test material was increased in the diets. All the renal function parameters evaluated showed no significant (P>0.05) difference in their values as the test material was increased in the diets (Table 8.4).



Table 8.3: Liver function test of broiler chickens fed diets containing potassium hydroxide-
treated (KOH_T) false yam seed meal (21-56 days of age)

	Control	100	300	500	SEM	Р
Albumin (g/L)	16.80 <sup>a</sup>	15.45 <sup>b</sup>	16.00 <sup>ab</sup>	16.10 <sup>ab</sup>	0.320	0.047
Globulin (g/L)	18.05	17.96	18.06	17.91	0.550	0.954
Total protein (g/L)	34.88	33.94	34.26	33.77	0.823	0.791
ALP (U/L)	149.8	149.9	148.2	148.4	3.30	0.973
ALT (U/L)	8.80	8.81	8.91	8.91	0.283	0.986
AST (U/L)	128.12	126.12	129.12	131.38	1.830	0.257

ALP: alkaline phosphate; AST: aspartate transferase; ALT: alanine transferase. P: probability; SEM: standard error of the mean; Means with the same superscripts within a row are not statistically different.

	Inclusion	levels of KC				
	Control	100	300	500	SEM	Р
Chloride (µmol/L)	77.1	77.3	77.6	78.9	3.63	0.985
Creatinine (mmol/L)	17.80	18.16	18.21	18.07	0.781	0.982
Sodium (mmol/L)	98.7	100.6	100.3	99.6	4.54	0.991
Potassium (mmol/L)	2.13	2.12	2.18	2.17	0.128	0.495
Urea (mmol/L)	0.71	0.69	0.71	0.71	0.031	0.995

Table 8.4: Renal function test of broiler chickens fed diets containing potassium hydroxide-
treated (KOH_T) false yam seed meal (21-56 days of age)

SEM: standard error of the mean; P: probability.

Table 8.5 showed the carcass and relative organ weights of male broiler chickens fed KOH\_T FYSM supplemented diets. Carcass dress weight and percent carcass dressing showed no significant (P>0.05) differences when the treated false yam seed meal incorporated at 500g/kg. However, the relative weight of organs revealed a significant (P<0.041) increase in the heart weight and a reduction in the weights of the liver and the spleen when the treated false yam seed meal was added beyond 100g/kg (Table 8.5). The primal cuts as a percentage of dress weight were also not affected; except for the relative weight of breast muscles. Breast muscle of birds fed diets containing 100g/kg KOH\_T was significantly depressed (P<0.015), whereas those birds fed diets containing 300 and 500g/kg had increased (P<0.015) relative weight of breast muscle. The sensory attributes of the chicken meat evaluated (Table 8.6) were not affected (P>0.05) negatively.





Table 8.5: Carcass cut up parts and relative organ characteristics of broiler chickens fed diets containing potassium hydroxide-treated (KOH\_T) false yam seed meal (21-56 days of age)

	Inclusion 1	evels of K	M (g/kg)			
Variable	Control	100	300	500	SEM	Р
Dress weight (Kg)	2.44	2.27	2.31	2.39	0.063	0.284
Carcass dressing (%)	77.65	78.17	77.25	76.72	0.498	0.248
Organs (% live-weight)						
Gizzard	1.34	1.28	1.40	1.28	0.045	0.152
Heart	0.39 <sup>c</sup>	0.44 <sup>b</sup>	0.40 <sup>c</sup>	$0.46^{a}$	0.018	0.041
Liver	2.02 <sup>a</sup>	1.62 <sup>b</sup>	1.71 <sup>ab</sup>	$1.78^{ab}$	0.100	0.026
Spleen	0.11 <sup>a</sup>	$0.07^{b}$	0.09 <sup>ab</sup>	0.07 <sup>b</sup>	0.008	0.038
Cut up parts (% dress weight)						
Breast	8.25 <sup>ab</sup>	7.60 <sup>b</sup>	8.36 <sup>a</sup>	8.43 <sup>a</sup>	0.188	0.015
Thigh	5.88	5.62	6.09	6.19	0.228	0.322
Drum stick	5.10	5.21	5.15	5.08	0.117	0.868
Wing	3.80	3.98	3.82	3.80	0.073	0.266

P: probability; SEM: standard error of the mean; Means with the same superscripts within a row are not statistically different.

Table	8.6:	Effect	of	potassium	hydroxide-treated	false	yam	seed	meal	on	sensory
charac	eterist	ics of m	ale	broiler chio	ekens						

	Inclusion le	Inclusion levels of KOH_T FYSM (g/kg DM)					
Variable	Control	100	300	500	SEM	Р	
Colour	2.53	2.33	1.67	2.00	0.259	0.103	
Tenderness	2.53	2.53	2.13	2.47	0.278	0.701	
Juiciness	2.53	2.73	2.67	2.40	0.228	0.739	
Chicken flavour	3.07	2.20	2.40	2.33	0.289	0.160	
Flavour liking	2.33	2.73	2.67	2.67	0.236	0.628	
Overall liking	2.13	2.47	2.67	2.60	0.264	0.494	

SEM: standard error of the mean; P: probability.

The results of the proximate components and lipid peroxidation values of chicken breast muscles are presented in Table 8.7 and Figure 8.2.

The inclusion of KOH\_T FYSM in the diets of male broiler chickens up to 500g/kg did not vary (P>0.05) dry matter and fat contents of the meat. However, crude protein content of the meat increased (P<0.001) significantly as KOH\_T FYSM was increased in the diets (Table 8.7). Figure 8.2 shows the effect of KOH\_T FYSM in the diets of male broiler chickens on the peroxide values



of their meat. There was an increasing trend (P<0.001) of peroxide values as the product was increased in the diets. However, peroxide values obtained in this study were far lower than the maximum permissible level of 25 meq/kg meat.

 Table 8.7: Effect of potassium hydroxide-treated false yam seed meal on proximate composition of breast muscle of male broiler chickens (21-56 days of age)

	Inclusion levels of KOH_T FYSM (g/kg DM)					
Parameter	Control	100	300	500	SEM	Р
Dry matter	34.4	35.9	33.4	39.8	2.50	0.351
Crude protein	17.63 <sup>d</sup>	18.03 <sup>c</sup>	19.64 <sup>b</sup>	20.41 <sup>a</sup>	0.006	< 0.001
Fat	3.3	3.5	3.8	3.8	1.16	0.986

SEM: standard error of the mean; P: probability.

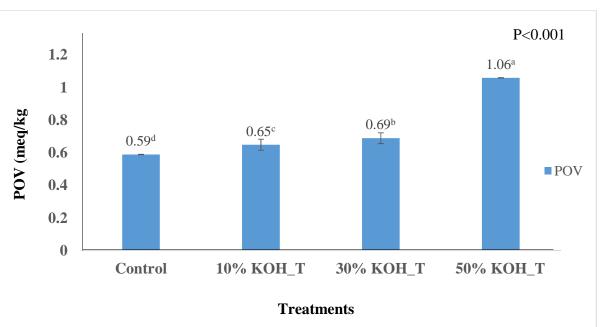




Figure 8.2: Lipid per-oxidation (Peroxide value) of breast muscles of male broiler chickens fed diets containing KOH\_T FYSM. Error bars represent standard error of mean. Peroxide value (n= 4 per data point).

The economics of supplementing maize with KOH\_T FYSM in the diets of male broilers as shown in Table 8.8 revealed that price per kg of feed reduced by 16.3% as the test material was increased to 500g/kg in the diets. However, the rest of the economic variables evaluated showed no

significant (P>0.05) difference. The non-significant reductions in the economic variables include; total feed cost per bird (7.8%) feed cost per kg live-weight gain (7.8%) feed cost per kg dress weight (5.7%) and gross profit per dress bird (5.9%).

	Inclusion le					
	Control	100	300	500	SEM	Р
Price/kg feed (GHS)	2.15	2.08	1.94	1.80	-	-
Total feed intake/bird (Kg)	4.24	4.39	4.67	4.66	0.165	0.235
Total feed cost/bird (GHS)	9.09	9.11	9.05	8.38	0.330	0.373
Feed cost/Kg live-weight (GHS)	3.71	3.64	3.66	3.42	0.189	0.712
Feed cost/Kg dress-weight (GHS)	4.88	4.74	4.95	4.60	0.345	0.892
Price/dressed bird (GHS)	19.55	20.35	19.21	19.46	1.139	0.904
Gross profit/dressed bird (GHS)	10.46	11.24	10.16	11.08	1.262	0.919

Table 8.8: Economics of feeding KOH\_T FYSM to broiler chickens (21-56 days of age)

SEM: standard error of the mean; P: probability.

#### 8.6.0 DISCUSSION



experiment was similar to those birds fed diets containing NaOH\_T FYSM in experiment 7. The similarity in growth performance of the birds could be due to similarity in the nutritional values of false yam seeds (FYS) treated with KOH or NaOH. This suggest that both products provide an opportunity as an alternative maize supplement in broiler chicken maize-based diets. KOH like NaOH is an alkaline substance capable of extracting anti-nutrients in solution. According to Ologhobo *et al.* (1993), higher concentrations of extractable anti-nutrients were found in alkaline-soluble fractions than in acid solutes, ether or alcohol. The major negative influence of anti-nutrients comes as a results of ANFs forming complexes with biologically important compounds like protein, enzymes or essential amino acids, thus affecting digestibility and nutrient utilization by poultry (Lange *et al.*, 2000; Pekel et al., 2015). The potential of KOH in reducing ANFs in

Growth performance of broiler chickens fed diets containing up to 500 g/kg KOH T FYSM in this

solution is clearly demonstrated in Experiment 1 of this study, hence improved nutritional value of the FYS and consequently good performance of birds that consume diets containing KOH\_T FYSM up to 500 g/kg inclusion.

The decrease in the values of some haematological parameters in this study could be attributed to the effect of residual accumulation of anti-nutritional factors found in the KOH\_T FYSM. According to Harvey *et al.* (1991), haematocrit and Hb concentrations have been shown to decrease when chicks were fed a diet naturally contaminated with 18 mg of deoxynivalenol (DON) /kg for 9 weeks.

The liver and renal function parameters were comparable in all treatment groups. However, only serum albumin concentrations were similar within the KOH\_T FYSM treatment groups, but lower compared to the control group. These results are in accordance with Campbell *et al.* (1983), who observed a decrease only in albumin of broiler chickens fed 2,500g of AFB<sub>1</sub>/kg from 1-21 days of age.

The carcass characteristics of the broiler chickens fed diets containing varying levels of KOH\_T FYSM were not adversely effected, except the breast muscle of birds fed diets containing 100 g/kg KOH\_T FYSM. This phenomenon could be attributed to dress weight of the birds fed such diets, because primal cuts of birds' increase with increasing dress weight (Rose, 1997). The reduction in weight of the spleen and liver of birds fed diets containing KOH\_T FYSM suggests that the residual anti-nutrients found in the seed meal could be implicated. Because, the main target of any toxin in the diets of poultry is the liver which is responsible for detoxifications of toxins (Smith, 1992).



The use of KOH\_T FYSM in broiler chicken diets did not negatively influence the sensory characteristics of their meat. When the sensory attributes of meat products vary significantly from the known, such products will be rejected by consumers (Warris, 2010).

Visual appraisal of products is one of the most important characteristics of food, and determines whether a consumer chooses or rejects products. According to Van Oeckel *et al.* (1999) and Bell and Weaver (2002), colour is a major indicator of quality of meat, as the appearance influences consumer acceptance. The comparable colours of the products observed in this study suggests that the KOH\_T FYSM at different inclusion levels have no potential to alter meat colour. Juiciness is directly related to tenderness, thus more tender meat readily liberate juices during chewing, compared with tougher meat. Juiciness in meat arises from moisture released by meat during chewing, and moisture from saliva (Christensen *et al.*, 2000).

Protein and fat are important constituents of meat. Meat serves as a major source of protein in human diets, hence meat is graded according to the level and quality of protein it possesses (Warris, 2010). The improvement in protein levels of meat in this experiment is an indication that the use of KOH\_T FYSM in broiler rations will not reduce the protein content of the meat. Fat improves appearance, juiciness and other sensory qualities of meat (Lawrie and Ledward, 2006). The similarity in fat levels in the meat is an indication that the sensory qualities of the meat will not be adversely affected, hence comparable sensory qualities observed in this experiment.



The lipid per oxidation values (POVs) of the products ranged from 0.59 to 1.06 millequivalent of active O<sub>2</sub> /kg of meat. Even though POVs significantly varied across the products in this experiment, POVs were far lower than the permissible limits of 25 millequivalent of active O<sub>2</sub>/kg of meat reported by Narasimhan *et al.* (1986).

The cost of producing broilers declined with higher levels of KOH\_T FYSM inclusion such that it was approximately 8.0% cheaper to produce live broiler chicken and approximately 6% cheaper to produce dressed broiler chicken on the 500 g/kg KOH\_T FYSM than on only maize-based diet (Control). Supplementing maize with KOH\_T FYSM at 500 g/kg gave the best financial returns of about 5.9% higher than birds on only maize-based diet. The higher profitability is due to the price differences between maize and KOH\_T FYSM. At the time of the trial, KOH\_T FYSM cost 42.7% of maize.

#### 8.7.0 CONCLUSION AND RECOMMENDATION

From this experiment, inclusion of potassium hydroxide-treated false yam seed meal up to 500g/kg in the diets of male broilers had no adverse effect on their growth performance. However, its inclusion influenced internal organ weights such as the liver, spleen and heart. Sensory and peroxide values in the meat were acceptable and protein content of the meat improved.

Economics of feeding KOH\_T FYSM to broilers did decrease cost of feeding, suggesting that, there is an economic value for using this product for broiler chickens as an alternative to maize during periods of scarcity and can be recommended for use by farmers where this plant is available.



#### **CHAPTER 9**

#### 9.0 General discussion

The observed differences in nutrient composition (Tables 4.1, 4.2 and 4.3) and some anti-nutrients (Figure 4.1 and 4.2) between raw and processed false yam seed meals showed the effect of processing on these components in false yam seeds for feeding animals. The raw false yam seeds showed a superior nutrient components but higher anti-nutrient contents, suggesting that, the seed cannot be used as alternative energy source in poultry diets in the raw or untreated form. This assertion is in line with that of Dei *et al.* (2011) who indicated that false yam seeds should be processed before it is fed to poultry.

It has been shown that feed ingredient processing technologies such as soaking (Dominguez *et al.*, 1993; Dei *et al.*, 2013), heat treatment (Omoruyi *et al.*, 2007; Khattab and Arntfield 2009), fermentation (Annongu *et al.*, 1996; Fagbemi *et al.*, 2005), autoclaving (Kessler *et al.*, 1990; Akande *et al.*, 2010), urea treatment (Udedibie and Nkwocha, 1990) and alkaline treatment (Ayanwale, 1999) can be used to improve the nutritive value of non-conventional feed ingredients, and therefore, increase nutrient availability and digestibility.

In terms of nutrient contents of the untreated false yam seeds (Table 4.1), they were similar to what have been reported by Dei *et al.* (2014). The slight variations observed in the nutrient composition could be attributed to the growing conditions of the false yam plant and the analytical method used. The drastic decline in some of the nutrients (CP, Ca, Mg and amino acids) of processed false yam seed meals in this study could be as a result of nutrient solubility during the water treatment. The use of water treatment to reduce or eliminate undesirable components in the feed ingredient could also remove useful soluble nutrients from the treated feed ingredient. The decline in crude protein and consequently in amino acid composition of the processed false yam seed is of a major nutritional concern in livestock feeding, particularly poultry. This is because of the need to use



high protein concentrates or the use of synthetic amino acids as supplements to make up for deficient amino acids, particularly lysine and methionine. This may increase feed cost. However, the use of processed false yam seed meals in this study did not incur additional costs in diet formulations when the diets were supplemented with high protein concentrate and macro-nutrients (Table 7.11 and Table 8.8). This is because the material at the moment is not sold and the only cost item is the processing cost. In fact, feed cost per kg diet was actually reduced by 16.3% when each of NaOH/KOH-treated false yam seed meals was included in the diets up to 500g/kg. The favourable economics of using the processed false yam seed meals in poultry diets (Table 7.11 and Table 8.8) in this study was attestation to the fact that processing has no adverse economic effects on the use of false yam seed as a feed ingredient for poultry. Therefore it can be useful as a feedstuff.

The chemical analysis of the untreated false yam seed sample confirmed the presence of antinutritional factors including terpenes (Figure 4.1)., The processing approaches in this study (i.e. sequential use of water-base, chemical treatment and blanching) resulted in a drastic reduction in anti-nutritional factors (Figure 4.1 and 4.2). For example, total terpenes in the processed false yam seed meals was reduced by 82-92% compared with a 39% reduction in total terpenes in false yam tuber meal (Dei *et al.* 2011). The greater reduction in the total terpenes could be attributed to the multiple stage processing methods used in this study as against a single processing method (soaking in water) used by Dei *et al.* (2011). Thus, in many instances, usage of only one detoxification method may not achieve the desired removal of anti-nutritional substances and a combination of two or more methods may be required for significant nutritional improvement (Akande et al., 2010).



Nevertheless, residual concentrations of anti-nutritional factors in the false yam seed (Figure 4.1 and 4.2) could still pose a nutritional challenge to the use of this material in poultry diets. Antinutritional factors react with protein, enzymes, or essential amino acids and form various complexes, thus affecting digestibility and nutrient utilization in poultry (Pekel *et al.*, 2015). This suggests that the materials in their present processed form could not be wholly substituted for maize in poultry diets even though the apparent metabolizable energy contents of the processed false yam seed meals were similar to that of maize (Table 4.1).

In this study, the experimental birds showed no pathological changes in the blood parameters measured when they were fed the processed false yam seed meals. Though significant variations were observed in blood parameters, they were within the ranges reported by some authors (Merck, 1979; Health and Olusanya, 1985). However, the narrow variations in the values recorded in the present study compared to those in literature could be due to difference in diets of the birds (Sahin et al., 2002), genetic makeup of the flock and age of the birds (Gendi et al., 2000) time of sampling (Adenkola and Ayo 2009), and environmental temperature (Olayemi and Ojo 2007). This suggests that the residual concentrations of the anti-nutritional factors in the processed false yam seed meals might have no health consequences for poultry.



Previous works involving false yam tuber (Dei *et al.*, 2011; Dei *et al.*, 2013; Mohammed and Dei, 2013) and seed (Okyere, 2011; Dei et al., 2016) meals in poultry diets did not show any deleterious effects on their health status. It has been reported that some animals are capable of tolerating low residual concentration of ANFs in their diets and the level of tolerance depends on the age of the bird (Asa, 2004).

The type of feed given to animals is reported to have significant effect on the carcass and sensory characteristics of the meat (Teye et al., 2006). In this study, the inclusion of sodium hydroxide or

potassium hydroxide-treated false yam seed meals up to 50% in broiler chicken diets as replacement for maize did not show any adverse effects on carcass, sensory characteristics and peroxide values of the broiler meat. The favourable carcass characteristics of the broilers in this study is an indication of improved utilisation of the processed false yam seed meals (Table 8.5). Also, the peroxide values for broiler chicken fed either false yam seed product in this study were lower than the minimum permissible level of 25 meq/kg meat, similar to those reported by Teye *et al.* (2011a; 2011b) for broilers fed either raw or soaked false yam seed meal. Besides, the inclusion of either false yam seed products in the diets resulted in improved crude protein (CP) content in the meats (Tables 7.10 and 8.7). The improvement in the CP of the meats was similar to the findings of Teye *et al.* (2011b) who reported improvement in CP content of broiler meat when fed diet containing soaked false yam seed meal.

In fact, the use of the alkali treated false yam seed meals in this study did show reduction in poultry feed cost by 3.3-16.3% (Table 7.11). The objective of every poultry farmer is maximization of profit, therefore, the use of these processed materials would be of economic benefit to farmers and they are encouraged to use them in diet formulations.



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## 9.1.0 General conclusion and recommendation

## 9.2 Conclusion

- Untreated false yam seeds contained adequate amount of carbohydrate and metabolizable energy to serve as dietary energy source for broiler chickens.
- Untreated false yam seeds contained low crude protein which was further depressed by processing.
- Untreated false yam seeds had high amount of total terpenes and saponins that could hinder its use as dietary energy source for broiler chickens.
- Sequential use of water-based and chemical (alkali) treatment methods were effective in reducing total terpenes and saponins in the false yam seed meals, which can enhance its usefulness for diet formulations in poultry. Processing methods caused drastic losses of crude protein, essential amino acids and macro as well as micro minerals.
- Processing the false yam seeds involving NaCl, NaOH and KOH had similar feed preference to the maize-based diets (Control).
- The preferred processing techniques for improved digestibility, AME and gross energy metabolisability were those of NaOH and KOH.



• The inclusion of NaOH-treated FYSM and KOH-treated FYSM up to 500g/kg as substitute for maize in the diets of broiler chickens had no adverse effects on growth performance, blood profile, carcass characteristics and eating quality.

## 9.3 Recommendation

- Processed FYSM would require dietary balance with protein-rich feedstuufs.
- Locally available and cheap sources of alkali substances (e.g. wood ash extract) should be tested for their efficacy in reducing anti-nutrients in the false yam seeds.
- The use of NaOH\_T/KOH\_T at 500g/kg in maize-based broiler chicken diets is recommended for broiler chicken farmers.



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