EFFECT OF PROCESSING ON SOME NUTRIENTS AND ANTINUTRIENTS COMPOSITION OF EGYPTIAN RIVERHEMP (*Sesbania sesban*) SEEDS

BY

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DEPARTMENT OF BIOCHEMISTRY,
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MAY, 2021

DECLARATION

I hereby declare that the work in this dissertation entitled **"Effect of Processing on some Nutrients and Antinutrients Composition of Egyptian riverhemp(*Sesbania sesban)* seeds"** has been performed by me in the Department of Biochemistry under the supervision of Prof. K.M Anigo and Prof. D.A Ameh. The information derived from the literature has been duly acknowledged in the text and a list of references provided. No part of this Dissertation was previously presented for another degree or diploma in another university.

AREKEMASE, Shefiat Olayemi		
Name of Student	Signature	Date

CERTIFICATION

This dissertation entitled **"EFFECT OF PROCESSING ON SOME NUTRIENTS AND ANTINUTRIENTS COMPOSITION OF EGYPTIAN RIVERHEMP(*Sesbania sesban)* SEEDS" by AREKEMASE, SHEFIAT OLAYEMI** meets the regulation governing the award of the degree of Master of Science of Ahmadu Bello University, Zaria and is approved for its contribution to knowledge and literary presentation.

Prof. K.M. Anigo Chairman, Supervisory Committee Department of Biochemistry	Signature	Date
Prof. D.A. Ameh Member, Supervisory Committee Department of Biochemistry	Signature	Date
Prof. A. B. Salau Head of Department	Signature	Date
Prof. S. A. Abdullahi Dean, School of PostgraduateStudies	Signature	Date

DEDICATION

This work is dedicated to ‘Almighty Allah’ the giver of all, for seeing me through from the beginning to the end of this programme.

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ABSTRACT

Egyptian riverhemp (*Sesbania sesban)*is widely distributed throughout Africa including Nigeria. Seeds from the plant contain essential nutrients but levels of antinutritional factors in the seeds present a challenge to its use as alternative feeds for livestock. In this study,the effect of processing on some nutrients and antinutrients composition in the seed of *Sesbania sesban*were assessed using lye. Lye possesses chemical characteristics as slaked lime, which have been used to detoxify or reduce antinutrients in plants. However, slakedlime can not be readily available especially to rural farmers, hence the choice of lye, which can be made readily available for the rural farmers. During processing, the seeds were soaked for (24 hours) and boiled (1 hour) using water, calcium hydroxide and lye. Theprocessed and unprocessed samples were then analyzed using standard methods for proximate, antinutrients, vitamins and minerals composition including *in vitro* protein digestibility. Results obtained for proximate composition revealed significant ($p<0.05$) decrease in moisture content of the processed samples with both samples boiled in calcium hydroxide (BC) and those boiled in lye (BL) which had the highest reduction as compared to raw sample (RS). Crude protein content was significantly ($p<0.05$) higher for sample boiled in water (BW), BC and BL compared to those soaked in water (SW), calcium hydroxide (SC), lye (SL) and RS whereas soluble and insoluble fibre contents were significantly ($p<0.05$) higher in SC and RS. Carbohydrate content obtained showed significant ($p<0.05$) reduction in BC compared to the other processed samples and RS. Analysis for antinutritional factors revealed that boiling with lye resulted insignificant ($p<0.05$) reduction in tannins, saponin, oxalate, cyanogenic glycosides and trypsin inhibitor, while soaking with water revealed more reduction in phytic acid content. Vitamin concentration revealed that processing caused significant ($p<0.05$) reduction in β-carotene (βC) and water-soluble

vitamins, with SC having a higher reduction in βC, B_2 and B_6 as compared to RS while a significant ($p<0.05$) increase in vitamin E was noticed in SL as compared to RS. Mineral contents indicate SC had higher Magnesium, Manganese and Copper concentrations compared to RS whereas Potassium and Iron contents were higher in SW. Boiling with water and boiling with lyehad higher Sodium and Calcium compared to RS. *Iin vitro* protein digestibility showed BL had significantly ($p<0.05$) higher digestibility compared to other processed samples and RS. Based on the findings from this study, samples boiled in lye (BL) had a higher reduction in some of the antinutrients with improved *in vitro* protein digestibility, hence this may enhance the use of the seed of *Sesbania sesban* as a potential alternative for livestock feed.

TABLE OF CONTENTS

CONTENT PAGE

LIST OF TABLES

Table Page

LIST OF PLATES

ABBREVIATIONS

%	Percentage
°C	Degree Centigrade
ANOVA	Analysis of Variance
B	Beta
BGL	Blood Glucose Level
CF	Crude Fibre
CHO	Carbohydrate
CP	Crude Protein
DPPH	2,2 – diphenyl – 1 – picryl hydrazyl
E. coli	*Escherichia coli*
FAO	Food and Agriculture Organization
HDL	High Density Lipoprotein
Inos	Inducible Nitric Oxide Synthase
IVPD	Invitro Protein Digestibility
LC-MS	Liquid Chromatography and Mass Spectroscopy
LDL	Low Density Lipoprotein
mg/dm^3	milligram per decimeter
MT	Metric tones
N	Normal
N_2	Nitrogen
NO	Nitric Oxide
SD	Standard Deviation
STZ	Streptozotocin
TC	Total Cholesterol
TG	Triglycerides

TUI	Trypsin Unit Inhibited
USA	United State of America
USD	United State Dollar
USDA-ARS	United State Department of Agriculture-Agricultural research service
V	Volume
Vit	Vitamin
W	Weight

CHAPTER ONE

1.0 INTRODUCTION

1.1 General Background

Egyptian riverhemp (*Sesbania sesban*) belongs to the family Fabaceae. It is a small perennial tree with woody stems, yellow flowers and linear pods. The origin of *Sesbania sesban* is unclear but widely distributed and cultivated throughout tropical Africa and Asia (Veasey *et al*., 1999). It is a fast growing, leguminous nitrogen (N_2) fixing, multi-purpose tree adapted to subtropical and tropical environments (Makatiani and Odee, 2007).

The centre of diversity is in Africa, and Egyptian riverhemp(*Sesbania sesban)* probably originated there (Evans, 1994). Genetic analysis of African populations by Jamadass *et al*. (2005) found the greatest diversity in East Africa.

The dried leaves ofEgyptian riverhemp(*Sesbania sesban*)are known to be used in indigenous medicines and the preparation in the form of tea is believed to have antibiotic, antihelminthic, antitumor and contraceptives properties. The leaves and flowers are consumed as vegetable by humans, while the immature pods and young branches of *Sesbania sesban* have been reported to be eaten raw by ruminant animals like cattle and goats (Gohl 1981; Verboon, 1966).In India, the mature seeds have been reported to be eaten after cooking by tribal sets of Kharis and Ghondan (Siddhauraju *et al.,* 1995).

Seeds flour has been reported to be used in the treatment of ringworm, skin diseases and wounds (Duke, 1981). The seed endosperm of *Sesbania sesban* are known to produce exudates (Anderson, 1989), which could have potential application in the food industry.

As small-scale farmers cannot afford to use chemicals (Wakjira *et al*., 2011) and improved feeds in their agricultural production system, they resort to the use of natural ways of replenishing soil fertility and feeding livestock through agroforestry.

Sesbania sesban has chemical constituents such as protein, sterol, saponin, flavonoid, fat, glycoside and good source of vitamins (Chartterjee and Pakrash, 1992). It is used in the treatment of ulcer, fever, purgative demulcent, pain reliever and as astringent (Yusuf *et al*., 1994).The plants also exhibit anti-inflammatory, and anti-microbial activities (Tatiya *et al*., 2008; Hossain and Chaudhary 2007: Ibrahim 1992; Sigh 1990;). They are excellent nitrogen fixers and capableof growing rapidly in nitrogen deficient soils, thus possess high utility in agroforestry as inter crop, cover crop, green manure, mulch and folder (Gopalakrishna, 2007).

Agglomeration of chemical, biochemical, clinical, and epidemiological evidences are denoting a positive correlation between the consumption of legume seeds and decreased incidence of several chronic diseases such as cancer, cardiovascular diseases, obesity, and diabetes (Bhathena & Velasquez, 2002). Such conspicuous health benefits of legume grains are ascribed to the existence of certain bioactive compounds, particularly the phenolic constituents (Shahidi & Naczk, 2004).

Flowers of *Sesbania sesban*are known to be added to stews and omelettes in some areas in south India, perhaps mainly as a decorative or festive ingredient in foods (Kathiresh *et al*., 2011; Orwa *et al*., 2009; Pravin *et al*., 2012).Alagesaboopathi (2012) reported that decoction of the leaf is mixed with hot milk and given once a day for seven days for treatment of diarrhea, itches and skin diseases.

1.2. Statementof Research Problem

Legume grains play a pivotal role in the customary diets of human beings throughout the world. They are magnificent sources of protein, starch, dietary fibre, micronutrients, and bioactive compounds with low level of fat. The total per capital consumption of legume grains has increased considerably over the past two decades in USA, due to the aggravated attention given to them as functional foods (Luthria and Pastar-Corrales, 2006).

Underutilized crop species have a potential that needs to be exploited to our advantage. Most of these crop species do not require high inputs, can be grown in marginal and degraded lands and at same time contribute to increased agricultural production, crop diversification and a better environment.

It is since 1970s that different exotic multipurpose fodder trees like *Sesbania sesban* got promoted by different organizations in Africa to alleviate feed shortages (Mekoya *et al*., 2009b). *Sesbania sesban*a N_2-fixing and deep rooting shrub with good-quality foliage and is one of the most promising species for short-duration cover cropping (Desaeger and Rao, 2001) and serve as protein supplement to poor quality roughages or as substitute for commercial protein supplements (Mekoya *et al*.,2009a).

Nigeria is one of the importers of agricultural produce, with a total food import bill 0f USD 4.2 billion annually (Olukunle, 2016).Demand for poultry meat, its consumption in the West Africa region outweighed its production; Nigeria being a highly populated country consumes approximately 1.2 million metric tons of poultry meat and produces approximately 290,000 MT of poultry meat (Ayisi and Adu, 2016).

The price of poultry meat in Nigeria may increase by 70% or more, because of increased prices of some foodstuffs (maize and soyabean) used in the production of

livestock feeds. As such, there is need to replace these conventional foodstuffs with underutilized plant seeds.

In an attempt to address these challenges, efforts are being made to seek and develop novel feed ingredients in order to reduce the high cost of feeding livestock and competition for conventional feed ingredients. *Sesbania sesban* seed has potential as animal feed because of its high crude protein, energy and other essential nutrients needed by livestock (Arekemase *et al.,* 2013). It is against this backdrop that the seed of Egyptian riverhemp (*Sesbania sesban*) was processed so that it can be used for livestock feed.

1.3. Justification of Study

Several plants exist with high nutritive value and yet remain unexploited for human and animal benefit (Oladele and Oshodi, 2007). The reason for underutilization of *Sesbania sesban* seed is because of its high antinutrients and lack of awareness.

Egyptian riverhemp (*Sesbania sesban*) is a good source of protein which can be used to replace soyabean in the formulation of feeds for livestocks, but the major obstacle hindering the utilization is the presence of antinutrients particularly tannin. However, studies have shown that tannin can be reduced to a bearable level using slaked lime (Dakare *et al*., 2012) but the use of slaked lime to detoxify seeds of this plant may not be economically viable, especially to the people in the rural communities. Hence, there is need to explore the use of other alternatives processing methods such as lye which is cheap and easily available to rural communities.

1.4Aim and Objectives

1.4.1Aim

To assess the effect of processing (lye treatment, boiling, soaking and slaked lime) on some nutrients and antinutrients composition of *Sesbania sesban* seeds

1.4.2 Specific Objectives

i. To evaluate the effect of processing (lye treatment, boiling, soaking and slaked lime) on the proximate composition of the seeds of *Sesbania sesban*seed
ii. To determine the effect of processingon antinutritional content of *Sesbania sesban* seed.
iii. To determine the effect of processing on the vitamins and mineral composition of Sesbania sesban seed
iv. To determine the effect of processing on the *in vitro*protein digestibility of *Sesbania sesban* seed

1.4.3Null Hypothsis

Processing has no effect on the nutrients and antinutrients compositions of *Sesbania sesban* seeds

1.4.3.1 Alternate Hypothesis

Processing has no effect on nutrients and antinutrients composition of Egyptian riverhemp (*Sesbania sesban)*

CHAPTER TWO

2.0 LITERATURE REVIEW

2.1 Botany of *Sesbania sesban* Plant

Sesbania sesban is a small perennial tree with woody stem, yellow flowers and linear pod (Arekemase *et al*., 2013). It is a narrow crowned, deep rooting single or multiple stem, short lived shrub or small tree up to 6m tall, and sometimes grow up to 8cm (Mani *et al*., 2011). The wood and the leaves are 7.5 - 15cm long including rachis, paripinnate; stiputes 3-7mm long, linear, acute leaflet 7-28 pairs opposite, 0.6 - 2.5cm long and 0.3 - 0.6cm wide, linear- oblong, glabrous, margins entire, apex obtuse and often faintly apiculate (Sajid *et al*., 2012). *Sesbania sesban* grows very fast (Makatiani and Odee, 2007), and may sometimes grow up to 4.5 to 6.0m high within one year (Niguissie and Alemayehu, 2013).

The leaves of *Sesbania sesban* are about 12- 18cm long, made up of 6-27 pairs of leaflets that are usually long, narrow and in many pairs (Gomase *et al*., 2012). Samajdar and Ghosh (2017) also reported that the leaves of this plant are 10- 20cm long, with 9-20 pairs of leafletsthat are oblong, 2-3cm long.

The pods are subcylindrical somewhat flattened, slightly twisted pendulous, about 20cm long and 3mm wide (Samajdar and Gosh, 2017). Other studies revealed 30cm long and 5mm wide (Arekemase *et al*., 2014). Each pod usually contains about 20-30cm seeds. Gomase *et al*., (2012) reported 40 seeds found in a pod. This implies the content of the seeds found in pods of *Sesbania sesban* varies. The raceme of *Sesbania sesban* has 2-20 flowers, which are yellow with purple or brown streaks (Niguissie and Alemayehu, 2013).

Sesbania sesban is a narrow-crowned, deep-rooting single or multiple stem shrub or short-lived tree, which may grow up to 8m (Hang *et al*., 2011; Mani *et al*., 2011) and up to 20cm stem diameter. The plant is fast growing (Makatiani and Odee, 2007) and it grows 4.5 to 6.0m high in one year (Siddiqi *et al*., 1985) and produces ripe pods within the first year after planting (Heering, 1995). If the trees planted are widely spaced they usually develop many side branches. The many branches give the tree a shrubby appearance (Orwa *et al*., 2009). The plant has up to 20 flowers which are yellow with purple or brown streaks on the corolla. Flowers are attractive, yellow (Pandhare *et al*., 2011; Mani *et al*., 2011), red, purplish, variegated or streaked, seldom white, large or small on slender pedicels, solitary or paired in short axillary racemes.

Sesbania sesban is widely planted, and has a long history of use throughout Africa and Asia including India. While the origin of the species is unclear, Evans (1994) suggests that it may have been spread across southern Asia from northeastern Africa by man.

Plate2.1: *Sesbania sesban*Plant with fresh leaves and pod

Source: www. harbalshop.com

Plate 2.2: *Sesbania sesban*Plant with flowers
Source: www. harbalshop.com

2.2. Ecological Adaptation

The plant (*Sesbania sesban)*can be found in areas with a semi-arid to subhumid climate (Degefu *et al*., 2011; Heering, 1995), with a rainfall between 500and 2000 mm per year (Orwa *et al*., 2009; Heering, 1995) and temperature of 18°C to 23°C (Orwa *et al*., 2009). In the regions with low precipitation however, they occur primarily on poorly drained soils which are subjected to periodic water-logging or flooding. Because of its good tolerance to low temperatures (Heering, 1995), it can be grown at an altitude of 100 to 2300 m (Orwa *et al*., 2009). It has moderate shade tolerance as well, and is adapted to a wide variety of soil types, ranging from loose sandy soils to heavy clays. Furthermore, it has an excellent tolerance to waterlogging and flooding (Heering, 1995) as well as saline, acidic, alkaline soils (Orwa *et al*., 2009) and soils laden with heavy metals (Gupta *et al*., 2011).

2.3 Pests and Diseases

Sesbania sesban is attacked by nematodes, insects, fungi andviruses. *Mesoplatys ochroptera*, the leaf-eating beetle, has been reported as a serious pest of *Sesbania sesban*in Ethiopia, Kenya,Malawi, Mozambique, Tanzania, Zambia and Uganda; while the other leaf eating beetle, *Exosoma and Ootheca spp*., has been reported so far only from Malawi and Zambia (Sileshi and Hailu, 2006). The *M.ochropteracan*can reduce forage yield or completely defoliate leading to mortality, if controlling measures were not taken during establishment (mostly 2 months after planting) (Orwa *et al.*, 2009; FAO, 2007). Insects such as *Anoplocnemis curvipes, Exosoma sp., Formicomus sp., Hilda patruelis*and *Medythia quaterna*, have also been reported to attack *Sesbania sesban*in southern Africa (Sileshi *et al*., 2000). *Alcidodes buho* is a weevil that damages the plant. The larvae of *Azygophelps scalaris*attack the plant boringthe stems. The

bacterium, *Xanthomonas sesbaniae*affects the stems and foliage. The seeds are often destroyed by a number of bruchid and other beetles (Orwa *et al*., 2009; FAO, 2007).

2.4 Distribution and Cultivation.

The exact origin of *Sesbania sesban* is unclear but widely distributed and cultivated throughout tropical Africa and Asia (Arekemase *et al*., 2013; Usman *et al*., 2013). They are found in Chad, Egypt, Kenya, Uganda, Angola, Australia, Benin, Botswana, Burkina Faso, Burundi, Cameroon, Cape Verde, Central African Republic, China, Congo, Cook Islands, Cote d'Ivoire, Democratic Republic of Congo, Djibouti, Equatorial Guinea, Eritrea, Ethiopia, Fiji, French Polynesia, Gabon, Gambia, Ghana, Tonga, United States of America, Vanuatu, Vietnam, Zambia, and Zimbabwe. Itis very common throughout Africa and in Asian countries like India, Malaysia, Indonesia and Philippines (Sandeep *et al*., 2014).

2.5 Chemical Composition.

Sesbania sesban contains numerous chemical compositions including sterol, saponin, flavonoids, cyanogenic glycosides (Arekemase *et al*., 2013), some of which are beneficial to health. Similarly, preliminary phytochemical screening uncovered the presence of triterpenoids, starches, tannins, saponins, glycosides and steroids. Blossoms contain cyanidin and delphinidin glucosides. Leaf contains campesterol, cholesterol, beta-sitosterol, triterpenoids, protein and tannins. Bark and stem contain glucose, fructose, erythritol, arabinitol, and myo-inositol. Different sorts of lignins made out of guaiacyl, syringyl and P-hydroxyphenylpropane building units and furthermore antitumor vital kampferol disaccharide (Samajdar and Ghosh, 2017). The plant parts also contain variable primary metabolites such as carbohydrates, proteins etc. (Kadam *et al*., 2013). Campesterol, β-sitosterol, Cyanidine, Delphinidin glycosides, α-Keto

glutaric, Oxaloacetic and pyruvic acids, Oleanolic acid, saponins, Palmitic acid, Stearic acid, Oleic acid, Linoleic acid and Linolenic acid are reported in Whole plant (Sandeep *et al*., 2014; Dande *et al*., 2010). Most of these plant metabolites are responsible for the broad-spectrum medicinal value of the plant. Medicinal plants play an important role in the development of potent therapeutic agents. There are over 1.5million practitioners of traditional medicinal system using medicinal plants in preventive, promotional and curative applications (Akao *et al*., 2008).

2.6 Nutritional Attributes

Leguminous plants have been used as feed for livestock in many parts of the world (Tolera, 2007), probably because of their high nutrient content. *Sesbania sesban* is one of such plants rich in almost all the essential nutrients needed by livestock (Arekemase *et al*., 2013). Because of it high nutritive value, farmers had been encouraged to feed Sesbania fodder to lactating cows to enhance milk secretion (Kadam *et al*., 2013).

Sesbania sesban foliage is protein-rich forage. Its crude protein content is generally above 22% DM and it can be higher than 30%. Hossain and Becker, (2001) reported 30 to 40% crude protein content in the seed of *Sesbania sesban*, while Arekemase *et al*. (2013) revealed lower crude protein content (20.22 ± 0.03g/100g). The crude lipid, although relatively small in quantity (5-6%) is rich in essential fatty acids, particularly linoleic acid (51.32%), although relatively lower in linoleic acid (2.02-2.64%) (Arekemase *et al*., 2014). Because of its high nutritive value, the plant has been considered as good source of animal feeds, especially in many tropical countries (Manaye *et al*., 2009; Mekoya *et al*., 2009). *Sesbania sesban* can be used as supplementary protein to roughage-based diets or to concentrate mixtures for sheep and goats. Level of inclusion for optimal growth rate or milk yield was about 30% of the

diet when it was used as supplement to teff straw (*Eragrostis tef*), Napier grass or sorghum stover (Mekoya *et al*., 2009c; Manaye *et al*., 2009; Mengistie, 2009; Sampathi *et al*., 1999; Kaitho *et al*., 1998). *Sesbania sesban*has shown to be used as supplementary protein to cattle fed tropical roughages (Tessema and Baars, 2004).

2.7 Potential Constraints

In sheep and goats, deleterious effects on growth and reproduction of male and female animals have been reported (Mekoya *et al*., 2009a; Mekoya *et al*., 2009b), tubular degeneration, changes in scrotal circumference, interstitial fibrosis (Woldemeskel *et al*., 2001), reduced occurrence of estrus, abortion in pregnant ewes, and even death have been reported (Melaku *et al*., 2004). The use of *Sesbania sesban* as a supplementary protein source in roughage-based diets had a positive effect on the rumen degradation of Dry Matter (DM), OM, Crude Protein (CP), Crude Fibre (CF) and NDF (Tessema and Baars, 2004). *In vitro* DM digestibility was found to be about 75% (Gutteridge and Shelton, 1991), while *in vivo* DM digestibility measured in goats ranged between 66 and 71% (Gutteridge *et al*., 1991). Many experiments have also shown that sesban foliage increased the digestibility of DM, OM and CP in cattle, sheep or goats fed on a roughage basal diet (Manaye *et al*., 2009; Melaku *et al*., 2004).

However, accessions of sesban rich in condensed tannins were reported to decrease NDF digestibility of the diet (Kaitho *et al*., 1998a). It has a positive effect on N retention, as evident in cattle (Umunna *et al*., 1995), though palatable to sheep and goats (Karbo *et al*., 1996). The effects of sesban feeding on reproductive performance are also debated. Sesban was reported to have deleterious effects (degeneration and necrosis) on the seminiferous tubules of male sheep and goats (Woldemeskel *et al*., 2001). Prolonged and uninterrupted sesban intake may also hinder sexual development (scrotum

circumference changes) and live-weight gains in male sheep and goats (Kaitho *et al*., 1998b).

The effects of feeding sesban foliage to sheep and goats have been studied for a long time and are still much debated. Though sesban foliage was shown to have high *in vitro* and *in vivo* digestibilities as well as positive N balance, its effects on feed intake (DM intake) are not consistent between authors. Some studies report higher feed intake or DM intake when sesban foliage is used as supplementary protein in roughage-based diets even at high levels of inclusion (Mengistie Taye, 2009; Manaye *et al*., 2009; Mekoya *et al*., 2009b).

Study on the effects of *Sesbania sesban* seeds on poultry nutrition, have shown to decrease performance as such, should be used with caution (Gutteridge and Shelton 1991). Supplementing ewes with sesban could compromise oestrus at high levels of inclusion (13.3 g DM/kg LW) and could cause abortion or deaths of pregnant ewes (Melaku *et al*., 2004). However, a series of studies from Mekoya *et al*. in 2009 showed that long-term feeding of *Sesbania sesban* foliage resulted in improved reproductive performance in both male and female sheep. It was also shown that feeding sesban from post-weaning to puberty reduced the age of puberty and improved sexual development (Mekoya *et al*., 2009a; Mekoya *et al*., 2009b). *Sesbania sesban* was introduced in growing rabbit diets up to 15% without any harmful or adverse effects on performance (Ghazalah *et al*., 1998).

2.8 Agricultural Importance

Most countries that have resorted to the use of natural ways of replenishing or improving soil fertility, due to lack of or inability to afford chemicals that can enrich the nutrient composition of the soil (Wakjira *et al*., 2011) have resorted to the use of

Sesbania sesban. It is worthy of green manure (Patra *et al*., 2006) that can immensely improve soil nutrient composition. Apart from it soil enrichment attribute, the plant also controls soil erosion and maintain soil fertility (Degeful *et al*., 2011; Shaheen *et al*., 2004). Also *Sesbania* species are widely used asfertilizer in different agricultural systems because it improves soil fertility, soil organic matter, water infiltration and holding capacity (Goswami *et al*., 2016).

2.9 Nutritional Profile

The seeds of *Sesbania sesban* are reported to contain 30 to 40% crude protein (Hossain and Becker, 2001), 5 to 6% of crude lipid and 2.7 to 3.3% of ash (Hossain *et al.*, 2002). Debela *et al*. (2011) reported that the crude protein contents of *Sesbania sesban*fractions varied from 194 g/kg dry matter in twigs to 297 g/kg dry matter in leaves. In addition, Akkasaeng *et al*.(1989) found that the in vitro dry matter digestibility of *Sesbania sesban*was 75%.

2.10 Phytochemical Properties

Sesbania sesban seed contains various antinutritional factors such as tannins, saponins and trypsin inhibitors. These antinutritional factors are the major problems when the seed is used as animal feed (Hossain and Becker, 2001). Phytochemical investigations in the seeds led to the isolation of oleanolic acid, stigmastane-5.24(28)-diene-3β-O-β-D-galactopyranoside and galactomannan (Das *et al*., 2011). The extracts had a high content of phenols, flavonoids and anthocyanins (Kathiresh *et al*., 2012). The pods and leaves contain campesterol and beta-sitosterol. Flowers contain cyanidin and delphinidin glucosides. Pollen and pollen tubes contain alpha-ketoglutaric, oxaloacetic and pyruvic acids (Pandhare *et al*., 2011).

2.11 Uses of *Sesbania sesban* Seeds

2.11.1. Anti-inflammatory activity

Inflammation is a common clinical condition that manifest to diseases. However, several reports suggested that bark of *Sesbania sesban* is use in diarrhea, spleen enlargement and inflammation. Patil *et al*. (2010) examined the effects of prophylactic administration of extracts of bark of *Sesbania grandiflora* and *Sesbania sesban* on the development of carrageenan induced paw oedema and adjuvant induced arthritis to assess influence of high nitric oxide level in the form of exogenous herbal extracts of bark of *Sesbania grandiflora* and *Sesbania sesban* in the progress of inflammation, which was assessed by measuring paw swelling. Their findings revealed that the petroleum ether extracts of bark of *Sesbania grandiflora* and *Sesbania sesban* suggested to exhibit significant anti-inflammatoryactivity as compared to chloroform and methanol extracts in inflammation. Petroleumether extracts of bark of *Sesbania grandiflora* and *Sesbania sesban* found to exhibitsignificant anti-arthritis activity and play negative feedback regulating role on iNOS andtherefore influence inflammatory process.

Similarly, Dande *et al*. (2010) evaluated the topical anti-inflammatory activity of the crude saponins extract by carrageenan induced rat paw edema method by preparing the gel formulation. The activity was carried on Wistar albino rats, receiving two strengths of crude saponin gel at a concentration of 1% w/w and 2%w/w respectively and Diclofenac sodium gel (1%w/w) was used as reference drug. Their findings showed that the development of carrageenan induced inflammation is a biphasic event; the first phase occurs within an hour of injection and is attributed to the release of histamine, and kinins, while second phase which can be measured around 3-4h is related to release of prostaglandins. The crude saponin extract have been able to control the increase in

paw edema in the early phase; however it has exhibited significant activity at the 3rd hour and further time point, which support the fact that it is related to the inhibition of prostaglandins release which inhibits the second phase of inflammation (Dande *et al*., 2010).

2.11.2 Antidiabetic activity

Because of better cultural acceptability, better compatibility with the human body and lesser sideeffects the herbal medicine is still the mainstay of about 75 - 80% of the world population, mainly in the developing countries, for primary health care (Goswami *et al*., 2016). *Sesbania sesban* amongst other plants has multiple medicinal values including antidiabetic. The aqueous leaves extract of *Sesbania Sesban* was evaluated for its antidiabetic potential on normal and streptozotocin (STZ)-induced diabetic rats at the doses of 250 and 500 mg/kg body weight per day for 30 days. The fasting Blood Glucose Levels (BGL), serum insulin level and biochemical data such as glycosylated hemoglobin, Total Cholesterol (TC), Triglycerides(TG), High Density Lipoproteins (HDL) and Low-Density Lipoproteins (LDL) were evaluated and all were compared to that of the known anti-diabetic drug glibenclamide (0.25 mg/kg b.w.).

Their findings reveal that the aqueous leaves extract of *Sesbania sesban* has beneficial effects in reducing the elevated blood glucose level and lipid profile of STZinduced diabetic rats, but has no effect on normal rats. The potential hypoglycemic effect of aqueous and ethanol extracts of*Sesbania sesban* was evaluated using STZ-induced diabetic mice and compared with metformin (MT) as a reference standard. The extract showed improvement in various body and serum parameters as well as regeneration of β-cells of pancreas and so might be of value in diabetes.

Manjusha *et al*. (2012) also suggests that *Sesbania sesban* may have hypolipidemic potential for type-2-diabetes and support the traditional use of the roots as hypolipidemic therapy.

Vadivel *et al*. (2012) examined the total free phenolics content, antioxidant, and type II diabetes related enzyme inhibition properties of methanolic extract of raw and traditionally processed *Sesbania sesban* seeds with a view to promote them as a dietary ingredient in the supplementary foods to be formulated with therapeutic value. Their findings revealed Methanol extract of the presently analyzed *S. sesban* seed materials was found to contain appreciable total free phenolics levels with promising antioxidant and type II diabetes related enzyme inhibition properties. Among the investigated processing methods, sprouting + oil-frying was identified as more suitable and mild treatment to increase the total free phenolic content as well as antioxidant properties of*Sesbania sesban* seeds. Therefore, such suitably processed underutilized legume grain with favorable antioxidant the formulation of supplementary foods with therapeutic value towards potential management of type II diabetes. Nevertheless, the questions on safe level, frequency of consumption, and serving size of *Sesbania sesban* grains to improve the health status of human beings should warrants further research. Identification of phenolic constituents of *Sesbania sesban* seeds using liquid chromatography and mass spectrophotometer (LC-MS) and *in vivo* evaluation of antioxidant and antidiabetic characteristics are under progress.

2.11.3. Antioxidant activity

Sesbania sesban has been evaluated for antioxidant acitivityThe antioxidant activity has been evaluated by DPPH and nitric oxide scavenging activity of the ethanol extract of the plant. The findings revealed to be dose dependent. For 100µg/ml concentration was

found as most active free radical scavenger. 76.25% and 72.18% have beenreported as percentage scavenging activity for 100µg/ml respectively for 2, 2- diphenyl - 1- picryl hydrazyl (DPPH) and nitric oxide (NO) scavenging (Mani *et al*., 2011). Saponin, Tannin, Phenolic compound, Anthocyanins were extracted with methanol and acidified methanol from the *Sesbania sesban* flower petals and their antioxidant properties were investigated. DPPH scavenging activity or the Hydrogen donating capacity was quantified in presence of stable DPPH radical on the basis of Blois method. Compounds from *Sesbania sesban* flower petals exhibited a dose dependent free-radical scavenging activity against DPPH radical, superoxide anions and hydroxyl radical (Samajdar and Gosh, 2017). Anthocyanins were extracted with methanolic and acidified methanol from the *Sesbania sesban* flower petals and their antioxidant properties were investigated. Anthocyanins from *Sesbania sesban* flower petals exhibited a dose dependent free-radical scavenging activity against DPPH radical, superoxide anions and hydroxyl radical (Kathresh *et al*., 2011).

2.11.4Antimicrobial activities

Throughout human history, most people have relied and still relying on traditional medicine, so as to promote and maintain good health. According to World health organization (WHO), about 80% of the population in the developing countries relies on traditional medicine (Al-Dawah, 2014). *Sesbania sesban* is among the widely used plants for treatment of infectious diseases, including microbial diseases. Broad antimicrobial activities of medicinal plants could be due to the presence of some plant secondary metabolites such as flavonoids, tannins, alkaloids etc. (Usman and Osuji, 2007). For example, Phytochemical screening of the ethanol, ether and chloroform extract of bark of *Sesbania sesban* indicates the presence of carbohydrates, flavonoids, steroids, alkaloids, tannins and saponins (Ahmed *et al*., 2013). These metabolites maybe

responsible for its efficacy against microbial diseases, and it reflect a possibility for the development of more novel chemotherapeutic agents or templates from the plant so that the plant may serve for the production of improved therapeutic plant based drugs (Ahmed *et al*., 2013).

Antimicrobial activity of *Sesbania sesban* has been investigated against five Gram-positive bacteria, nine Gram-negative bacteria and seven fungi by disc diffusion and broth macro-dilution assay (Ahmed *et al*., 2013). Their findings revealed High significant degree of activity against the test bacteria *B. subtilis* followed by *E. coli*. While the carbon tetrachloride partitionate of the methanol leaf extract of *Sesbania sesban* showed the strongest inhibitory activity against *E. coli.* Moreso, A fluctuating trend of inhibition zone was found against some pathogens in the analysis. Similarly, Mythili and Ravindhran (2012) examined the in vitro biological screening effects of the methanol stem extract was tested against ten bacterial species and five fungal species, and

High significant activity was observed against the bacteria *Erwinia amylovora* followed by *E. coli*, while in the case of fungi, *Curvularia lunata* and *Fusarium oxysporum* were inhibited completely.

2.11.5. Green manure

Sesbania sesban is a fast growing nitrogen-fixing leguminous tree species which has the capacity of rapid decomposition when incorporated into soil serving as a green manure (Patra *et al*., 2006) in alley cropping (Heering, 1995) which could bring about substantial increment in crop available nitrogen and soil organic carbon.It has a high level of foliage nitrogen and is an excellent supplement to protein-poor roughage (Sabra *et al*., 2010; Manaye *et al*., 2009; Orwa *et al*., 2009). The leaves and tender branches of

this tree have high levels of protein (with 20 to 25% crude protein), and easily digestible when consumed by ruminants (Pravin*et al*., 2012). It has a long history of use as a source of cut-and-carry forage (Naik *et al*., 2011). In Ethiopia, feeding*Sesbania*leaves and young twigshave become increasingly important as a protein rich supplement to a basal diet of either grass or poor-quality forage for ruminants (Tessema and Baars, 2004). For example, Manaye *et al*. (2009) reported that the sheep fed with diet containing 300 g/kg *Sesbania sesban* foliage showed 103 g/day average daily body weight gain.

2.11.6. Polluted water and soil treatment

The ability of *Sesbania sesban* to grow at different ammonium concentrations soil culture has been studied by different workers (Dan *et al*., 2011; Dan and Brix, 2009., Indieka and Odee, 2005), and it was shown that its seedlings can tolerate ammonium concentrations up to 800 mg/L. This high tolerance suggests that this plant has a potential for use in treatment systems of waste or polluted water (Dan *et al*., 2011; Dan and Brix, 2009; Indieka and Odee, (2005) andremoval of heavy metals from soil, that is, phytoremediation of sites contaminated with heavy metals(Gupta *et al*., 2011; Yang *et al*., 2003).

2.12. Antinutrients

Antinutrients are chemicals which have been evolved by plants for their own defense, among other biological functions and reduce the maximum utilization of nutrients especially proteins, vitamins, and minerals, thus preventing optimal exploitation of the nutrients present in a food and decreasing the nutritive value (Edet *et al*., 2015). These factors can also be defined as those substances generated in natural feedstuffs by the normal metabolism of species and by different mechanisms (for example inactivation of

some nutrients, diminution of the digestive process or metabolic utilization of feed) which exerts effect contrary to optimum nutrition (Akande *et al*., 2010). Antinutrients interfere with metabolic processes such that growth and bioavailability of nutrients are negatively influenced (Agbaire, 2011).

There are many groups of chemical compounds that have health benefits, others can be very toxic and fatal to humans when consumed (Natesh *et al*., 2017). Most of the plant foods contain anti-nutrients. Their presence in foods affects mineral bioavailability and protein absorption in humans and animals, thereby causing mineral deficiencies and may also lead to malnutrition (Ertop and Bektaş, 2018). In countries where plants based seems to be the major source of food, the presence of antinutrients disrupt micronutrients utilization. Infact, antinutrients reduce the maximum utilization of nutrients (especially proteins, vitamins or minerals), and as a consequence they obstruct an optimal bioavailibility of the nutrients present in a food and decrease its nutritive value (Eltayeb *et al.,* 2007). The deficiencies of micronutrients such as minerals and vitamins have caused to be most serious health problems (Jorge *et al.,* 2008). Therefore, the determination of antinutrients and toxic substances in plant based foods is an imperative facet in nutritional studies as it establishes the baseline concentrations index for phytotoxins in the foods (Musa and Ogbadoyi, 2014).Most of these phytotoxins such as trypsin inhibitors, phytates, oxalates, nitrates, alkaloids, tannins and cyanogenic glycosides poses health problem if present in high amount (Proph *et al*., 2006). The major antinutrients mostly found in plant protein sources are toxic amino acids, saponins, cyanogenic glycosides, tannins, phytic acid, gossypol, oxalates, goitrogens, lectins (phytohaemagglutinins), protease inhibitors, chlorogenic acid and amylase inhibitors (Akande *et al*., 2010).

The toxic non-protein amino acids such as djenkolic acids, mimosine and canavanine appear to play a major role in determining the nutritional value of a number of tropical legumes, as these amino acids act antagonistically towards certain nutritionally important amino acids (Akande *et al*., 2010). Phytate has long been recognized as an antinutritional factor affecting the bioavailability of major minerals such as Ca and trace ones such as Fe, Cu, Zn and Mn (Ertop and Bektaş, 2018). However, many antinutrients besides their primary effects on the bioavailability of nutrients may also be toxic beyond a certain dose. For instance, ingestion of high concentration of cyanogenic glycoside leads to respiratory poisoning and inhibition of ATP synthesis in electron transport chain (Musa, 2012; Musa *et al*., 2011). Nitrate on the other hand, is one of the major culprits in cancer and metheamoglobinaemia (Anjana *et al*., 2007). Overall, antinutrients in plant foods are responsible for deleterious effects related to the absorption of nutrients and micronutrients. Phytic acid, tannins, saponins, and protease inhibitors have been shown to reduce the availability of nutrients and cause growth inhibition (Edet *et al*., 2015). Most of the anti nutritive substances become ineffective or their level can be reduced with simple treatments such as heating, soaking, germination or autoclaving (Ertop and Bektaş, 2018).

2.13 Effects of Processing on Nutrients and Antinutrients

Ramadan (2012) study the effect of processing (soaking and germination, ordinary cooking and autoclaving) on the chemical composition, sugars and phytic of two varieties soybean seeds, Giza, 21 and Giza, 35. The processing methods caused increase in both protein and crude fiber contents. Meanwhile, crude oil and carbohydrates contents were decreased of the studied soybean seeds. Generally, the processing and cooking methods resulted in a decrease of raffinose, stachyose, verbascose, maltose and sucrose accompanied by an increase in glucose. These results revealed that the

processing (soaking and germination) and cooking methods (ordinary cooking and autoclaving) was more effective in eliminating the contents of oligosaccharides and phytic acid in both varieties soybean seeds.

Removing antinutrients, the bioavailability of some cation (Ca, Fe and Zn) and the absorption of proteins make to increase and consequently nutrition value of food increase (Ertop and Bektaş, 2018). Idoko *et al*. (2014) investigated the effects of heat treatment (boiling and autoclaving) on the nutrient and anti-nutrient components of melon husk. Variations in the proximate composition of the differently treated husks were significant except for the lipid content. However, no significant difference exists between the proximate compositions of raw and autoclaved melon husks except for ash and carbohydrate contents. The results of mineral analysis showed that boiling caused significant reductions in all the mineral elements analyzed. All the detected antinutrients were significantly reduced by heat treatments (boiling and autoclaving) and autoclaving was found to be more effective in reducing the levels of antinutrients than boiling. Suggesting, that melon husk, if heat treated, could be an alternative source of feed for live stock. One of theadvantage of cooking as a traditional processing method result in destruction of germs or parasites, the removal of toxic substances and the improvement of nutritional quality. However, these phenomenons depend on plant texture, temperature and cooking time (Vodouhe *et al*., 2012).

Irrespective of the processing methods, time or duration of processing plays a key role in the bio-accessibility of nutrients and anti-nutrients of plant based foods. (Suzanne*et al*. 2017) studied the effects of cooking time on some antinutrients contents and on proteins digestibility of Leaves Proteins of *Gnetum* spp. Observation revealed that *Gnetum* vegetables have high contents of some antinutrients which significant amounts are easily remove with long cooking time and improves digestibility of their proteins.

Lewu *et al*. (2009) investigated the effect of cooking on the mineral and antinutrient contents of the leaves of seven accessions of *Colocasia esculenta* (L.). It was revealed that cooking significantly reduced the phosphorus, potassium and zinc contents, while calcium,magnesium, sodium and copper levels were not significantly different in almost all the accessions tested. In contrast, cooking appears to increase the iron levels in all the samples. Suggesting that the consumption of the leaves of cocoyam may increase the blood level and may therefore recommended as a good food for anemic patients.

Furthermore, boiling remarkably reduced the level of the anti-nutritional factors, thereby improving the food quality of all the accessions tested, however, boiling for 5 min led to16-78% drop in oxalate level, 28 - 61% in tannin and 17 - 41% reduction in phytate contents in some of the accessions. Though, boiling can help to reduce the oxalate content in the leaves of this species, it may also reduce the nutritional value of food crops arising from significant losses and changes in major nutrients during cooking (Lewu *et al*., 2009). Similarly, Embaby, (2011) studied the effect of heat treatments (boiling, autoclaving, microwave cooking and roasting) on the levels of certain antinutritional factors (phytic acid, trypsin inhibitor, α-amylase inhibitor, lectin activity and tannins) and *in vitro* protein digestibility (IVPD) of peanut and sesame seeds. All heat treatments significantly reduced the levels of all the investigated antinutrients and improved the IVPDof peanut seeds. Of the attempted treatments, autoclaving, boiling, roasting-salting and oil-roasting werethe most effective in reducing the levels of antinutrients and improving IVPD of peanut.

Also, Fekadu (2014) investigated the raw and boiled Anchote (Coccinia abyssinica (Lam.) Cogn.) tubers and compared for their nutritional composition: moisture, crude protein, total ash, crude fiber, crude fat, utilized carbohydrate and gross energy; minerals: Ca, Fe, Mg, Zn, and P and antinutritional factors: phytate, oxalate, tannin and

cyanide. The study uncovered information on the nutritional composition (crude fiber, crude fat, crude protein, total ash, moisture content, utilized carbohydrate, gross energy, Zinc, Iron, Calcium, Sodium, Magnesium and Phosphorus) and antinutritional factors (Phytate, Oxalate, Tannin and Cyanide) of raw and processed Anchote tubers from western Ethiopia.The study conclusively revealed that raw Anchote contains appreciable quantity of carbohydrate, crude Protein,crude fiber, calcium, magnesium, iron and low levels of antinutrients (Oxalate, tannin, and cynide) exceptphytate, when compared to other reported raw roots and tubers. The traditional processingmethods of Anchote were very important because that increases the crude fibre content in the Anchotetubers.

The study also indicated that traditional processing methods decreased the crude protein, total ash, calcium, iron, zinc content of the tubers. Also, among the traditional processing methods, boiling before peeling proved to be better in some nutrient and mineral contents considered in the investigation.(Ndidi *et al*. 2014) analyzed the effect of processing (boiling and roasting) on the proximate, antinutrient, and mineral composition of *Vigna subterranea* seeds. Observation from the proximate analysis showed significant difference between the levels of crude lipid, crude fiber, gross energy, carbohydrate, and moisture content in the raw and processed *V. subterranea*. However, no significant difference was revealed in the protein content of processed *V. subterranea* as compared to the raw seeds. Analyses of antinutrient composition show that processing significantly reduced the levels of oxalate, tannins, phytate, trypsin inhibitor, and hydrogen cyanide contents of *V. subterranea*. The magnesium, potassium, and phosphorus were the most abundant macrominerals in *V. subterranea*, zinc was the most abundant micromineral. Suggesting that processing significantly lowered the levels of antinutrients in *V. subterranea*, thereby making it safer for

consumption.Sometimes, the nature and period of cooking or boiling determines it effects on the nutrients and antinutrients of plant based foods. For instance, the loss of ascorbic acid was lesser during pressurecooking as compared to boiling. Moreso, prolonged boiling had detrimental effect on ascorbic acid. Pressure cooking for 3 min or boiling for 15 min improved in vitro protein digestibility, although, longer cooking durations reduced ascorbic acid and beta carotene significantly (Deol and Bains, 2010)

Anuonye *et al*. (2013) investigated the effects of extrusion cooking on the nutrient and anti-nutrient composition of raw and extruded blends of pigeon pea and unripe plantain flours were evaluated. The pigeon pea seeds were cleaned and processed into flour while unripe plantain was peeled, sliced, dried and milled into flour separately and sieved to pass 0.85mm mesh. Blending pigeon pea and unripe plantain flour at 25% of unripe plantain flour substitution resulted in the reduction of anti-nutrients.

Extrusion further lowered these anti-nutrients to safer levels. Rehman and Shah, (2005) evaluated the effect of thermal heat processing on antinutrients, protein and, starch digestibility of black grams, chick peas, lentils, red and white kidney beans. Reduction in the levels of antinutrients, along with an improvement in protein and starch digestibility, was observed after cooking thelegumes foods, whereas, ordinary cooking resulted in improvement of protein and starch digestibility of the food legumes. Reduction of the anti-nutritional factors in legumes is very necessary, as poor nutritive values of the food legumes are due to the presence of some antinutritional substances (Rehman and Shah, 2005).

Nutritional deficiencies of proteins, vitamins and minerals are widespread in most developing countries and hence, imperative to obtain foods with superior nutritive value (Gunashree *et al*., 2014**).** Various traditional processes on different cereals and legumes

are diverse, as (Gunashree *et al*., 2014) investigated the influence of various combinations of processes on nutrients and antinutrients of ragi and wheat. Significant difference in nutrients and antinutrients among raw and processed ragi and wheat was observed, suggesting the need to adopt suitable process for highest nutrient retention and antinutrient reduction for composite food preparations. Similarly, Ihemeje *et al*. (2018) studied different processing methods such as boiling, toasting, sprouting and soaking on dietary Fibre and anti-nutrients in African yam bean and red kidney beans. Some leguminous plant such as *Arachis hypogaea* (Groundnuts) are a good source of nutrients, however the presence of antinutrients, such as oxalate, phytate, tannin and trypsin inhibitor, reduces their bioavailability and utilization (Mada *et al*., 2012).

Processing of groundnuts has shown to reduce the level of antinutrients present. The boiling of groundnut significantly ($P<0.05$) decreased most of the antinutrients component in the samples including saponin, tannin, phytate, oxalate and trypsin inhibitor, but the treatment also significantly ($P < 0.05$) decreased the content of protein from 23.50% to 10.80% (Mada *et al*., 2012). Groundnut is an important oil seed and cash crop accounting for more than one-third of the total oil seed production in India (Sahayaraj and Martin, 2003). It is an important item in several confectionery products, and in supplementary feeding programmes such as in weaning food formulations in combination with cereals and pulses in many developing countries such as Nigeria (Mada *et al*., 2012), as such the need to processing it before consumption is necessary.

CHAPTER THREE

3.0MATERIALS AND METHODS

3.1MATERIALS

3.1.1Chemicals and Reagents

All chemicals and reagents used were of analytical grade.

3.1.2Plant Collection and Identification

The mature pods of *Sesbania sesban* was harvested from Bassawa, Sabon Gari local government area of Zaria. The pods were properly dried, threshed and the seeds obtained were driedat room temperature. The identity of the plant was confirmed and a voucher number 900112 deposited at the herbarium, Department of Botany, Ahmadu Bello University, Zaria, Kaduna State, Nigeria.

3.1.3Ash Production

Ash was produced by burning wood (*Gmelina aborea*) completely and ash obtained was sieved with mesh size of 0.400mm to remove pieces of charcoal and other impurities.

3.1.4Lye preparation

About100g of ash was poured into a perforated container with a sieve cloth at the base of the container and 1000ml of distilled water was poured on the ash. The mixture was allowed to stay overnight to collect the lye as a droplet. Thereafter, 1000ml ofdistilled water was poured into the mixture, followed by lye collection as a droplet. The process was repeated tentimes to achieve the desired quantity of lye enough to be used for the treatment of the plant seed.

3.1.5Sample Preparation

3.1.5.1 Boiling with water

Ground raw100g of*Sesbania sesban* seed sample was boiled in 1000ml of distilled water at 100^{o}C for 1hour and it was allowed to cool overnight. The supernatant was decanted and the residue was washed with distilled water several times, sun-dried and it was labelled as BW.

3.1.5.2 Boiling with $(CaOH)_2$

Dried raw 100g of ground *Sesbania sesban* seed was boiled in a solution of 1000ml of distilled water and 100g of $Ca(OH)_2$at 100^{o}C for 1hour and cooled and kept overnight. The supernatant was decanted and the residue was washed with distilled water several times to be free of $Ca(OH)_2$,(the neutrality was tested using litmus paper),sun-dried and labelled as BC.

3.1.5.3Boiling with lye

About 100g of raw ground *Sesbania sesban* seed was boiled in 1000ml of lye water for 1hour and it was allowed to cool overnight. The supernatant was decanted, the residue was washed with distilled water several times, sun-dried and labelled as BL.

3.1.5.4Soaking with H_2O

Ground raw 100 g *Sesbania sesban*seed was soaked in 1000ml of distilled water for 24hours at room temperature with occasional stirring. The supernatant was decanted and the residue was washed with distilled water several times, sundried and labelled as SW.

3.1.5.5Soaking with $Ca(OH)_2$

Raw ground seeds of *Sesbania sesban* (100g) were soaked in a mixture of 1000ml distilled water and 100g of $Ca(OH)_2$ for 24hours at room temperature. The supernatant

was decanted and the residue was washed with water several times, to be free of Ca $(OH)_2$,(its neutrality was tested using litmus paper),sun-driedand labelled as SC.

3.1.5.6 Soaking with Lye

Ground raw 100 g*Sesbania sesban* seed was soaked in 1000ml of lye water for 24hours at room temperature with occasional stirring. The supernatant wasdecanted and the residue washed with water several times, sun-dried and labeled as SL.

3.1.5.7 Raw seed

Raw ground seed of Egyptian riverhemp (*Sesbania sesban*) 100g was labelled as RS which served as control to all other processed samples.

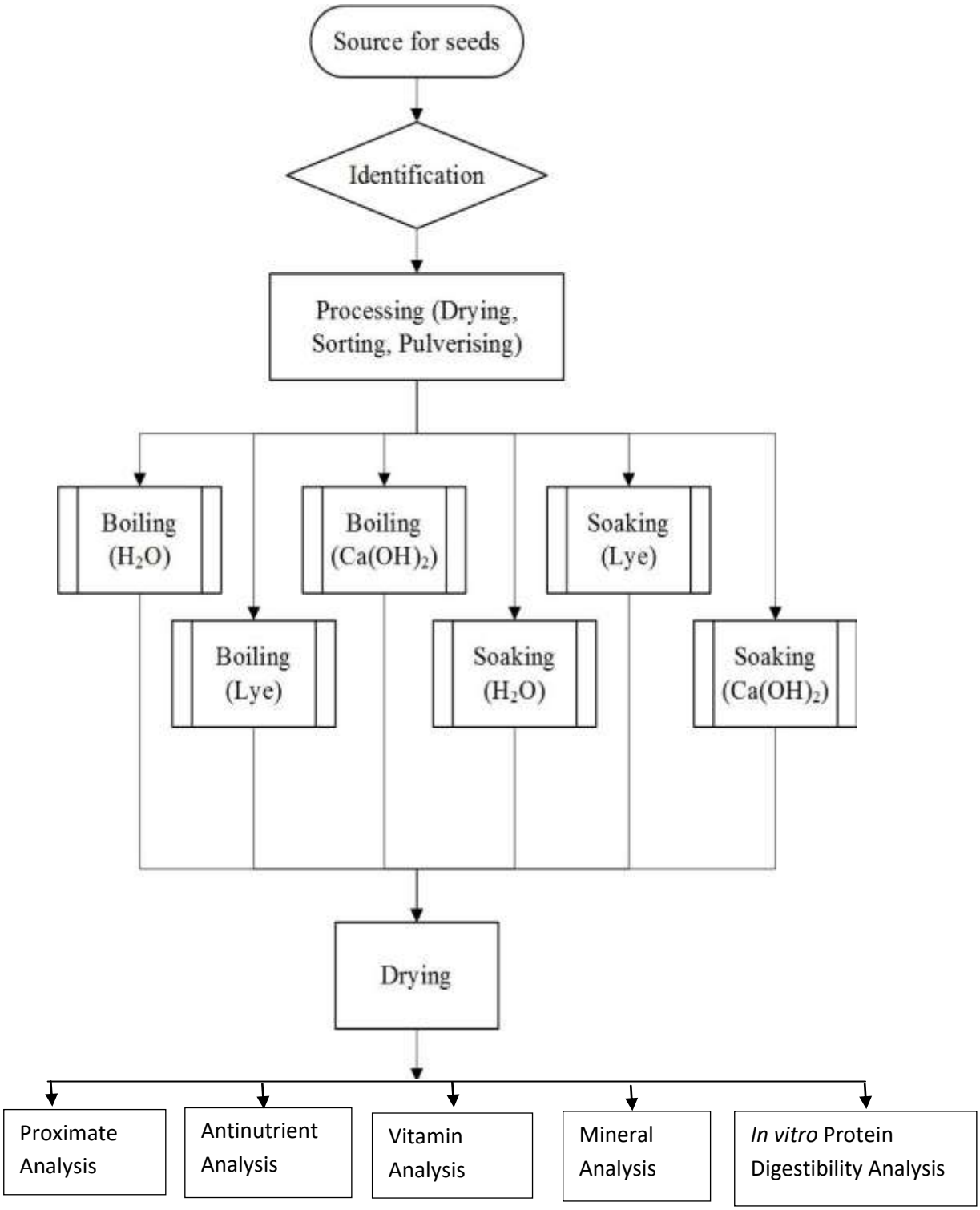

Figure 3.1. Experimental Design

3.2 METHODS

3.2.1Proximate Analysis

3.2.1.1 Determination of moisture content

The method described by Official Method of Analytical Chemist AOAC (2003) was adopted. A clean crucible was dried to constant weight in an air oven at 105^{o}C over a period of thirty minutes, cooled in a desiccator and weighed (W_1). Two grams of sample was weighed into the previously labeled crucible and reweighed (W_2). The crucible was dried in oven to a constant weight (W_3), the moisture content was done in triplicate for all the treated and raw samples. The percentage moisture content was calculated thus:

$$\% \text{ Moisture content} = \frac{W2-W3}{W2-W1} \times 100$$

3.2.1.2. Determination of ash content

The AOAC (2003) method was used for the determination of Ash contents in triplicate for all the treated and raw seeds of *Sesbania sesban.* The porcelain crucible was dried in an oven at 100^{o}C for 10 minutes, cooled in a desiccator and weighed (W_1). Two grams of the sample was placed into the previously weighed porcelain crucible and weighed (W_2). The sample was first ignited and transferred into a furnace, which was then set at 550^{o}C. The sample was left in the furnace for 8 hours to ensure proper ashing. The crucible containing the ash was removed cooled in the desiccator and weighed (W_3). The percentage ash content was calculated as:

$$\% \text{ Ash content} = \frac{w3-w1}{w2-w1} \times 100$$

3.2.1.3. Determination of Crude Fat Content

The crude fat content was determined according to AOAC (2003) method. A clean, dried 500ml round bottom flask, containing few anti-bumping granules was weighed

(W_1) and 300ml of Petroleum ether (40-60^{o}C) for extraction was poured into the flask fitted with soxhlet extraction unit. The extractor thimble containing twenty grams of the sample wasfixed into the soxhlet extraction unit. The round bottom flask and a condenser was connected to the soxhlet extractor and cold-water circulation was put on. The heating mantle was switched on and the heating rate was adjusted until the solvent reflux at a steady rate. Extraction was carried out for six hours. The solvent was recovered and the oil was dried in the oven at 70^{o}C for one hour. The round bottom flask containing the oil was cooled in the desiccator and then weighed (W_2), this was done in triplicate for all the samples. The crude fat content was calculated thus:

$$\% \text{ Crude Lipid Content} = \frac{W2-W1}{Weight\ of\ sample} \times 100$$

3.2.1.4 Determination of crude protein

Protein digestion

The method was based on that reported by Official Method of Analytical Chemists AOAC (2003). About 1.5g of the defatted 15 ml of H_2SO_4 and 3g of digesting mixed catalyst (weighed separately into an ashless filter paper) were dropped into the Kjeldahl flask. The flask was then transferred to the Kjeldahl digestion apparatus and sample digested until a clear green colour was obtained. The digest was cooled and diluted to 100ml with distilled water.

Distillation of the digest

About 20ml of the diluted digest was measured into a 500ml Kjeldahl flask containing anti- bumping chips and 40ml of 40% NaOH was slowly added by the side of the flask. A 250ml conical flask containing a mixture of 50ml of 2% Boric acid and 4 drops of mixed indicator was used to trap the ammonia liberated. The conical flask and the

Kjeldahl flask were then placed on the Kjedahl distillation apparatus, with the tubes inserted into the conical flask. The flask was heated to distill out NH_3 evolved. The distillate was collected into the boric acid solution. From the point when the boric acid turned green, 10 minutes was allowed for complete distillation of the ammonia present in the digest. The distillate was titrated with 0.1M HCl and this was done in triplicate for all the samples.

Calculation:

$$\% \text{ N} = \frac{14 \times M \times Vt \times Tv}{Weight\ of\ sample\ (mg) \times Va} \times 100$$

% Crude protein = % Nitrogen (N_2) x 6.25

Where M = Actual molarity of acid

Tv = Titre volume of HCl used

Vt = Total volume of diluted digest

Va = Aliquot volume distilled

3.2.1.5. Determination of dietary fibre contents(AOAC, 1992)

This method involves sequential enzymatic digestion of the sample by heat-stable α-amylase, protease and amyloglucosidase

About 1g of the sample was weighed inside a 400ml beaker to which 40ml of the buffer solution was added and stirred with a magnetic stirrer to prevent lump formation. The solution was then incubated with 50µl of heat stable α-amylase solution while stirring at low speed and solution was allowed to cool before adding 100ml protease solution and incubated with shaking in water bath at 60°C for 30minutes. The mixture was further

incubated with 200ml amyloglucosidase solution, while stirring in a water bath atthe same temperature of 60°C for 30minutes with constant agitation.

The solution was then filtered, the residue was washed twice with 10ml distilled water, 95% ethanol and acetone, then dried in the crucible for protein / ash determination for insoluble fibre analyses. The filtrate was used for soluble fibre analysis by precipitation 4 volume of 95% extract preheated to 60°C and allowed at room temperature for 60minutes and the precipitate was filtered. The residue was washed successively with two 15ml of 78% ethanol, 95% ethanol and acetone. The residue was later dried, weighed and then ashed and protein content was determined. This was done in triplicate for all the samples

$$\text{Dietary fibre (\%)} = \frac{Weight\ of\ residue\ -protein\ -ash-blank}{Weight\ of\ \ sample} \times 100$$

3.2.1.6. Determination of carbohydrate contents AOAC (2003)

Thecarbohydrate contents of food involve mainly free sugars, oligosaccharides and some polysaccharides were first hydrolyzed into their constituent's monosaccharides by the acids. The monosaccharides are then extracted and reacted with anthrone to form a complex, which absorbs at 620nm

Procedure

To determine the carbohydrate contents of the sample, 1 g of the sample (*Sesbania sesban*seeds) was dispersed in 85 ml distilled water in a 250 ml beaker. An additional 15 ml of 52% perchloric acid was added and stirred thoroughly for 30minutes. The mixture was then filtered using whatman filter paper into conical flask. To the 1ml filtrate, 4ml of anthrone reagent was added and the absorbance at 620nm was measured, using anthrone reagent as a blank.

Standardization

A standard glucose solution of 2.0, 3.0, 5.0, 7.5 and 10mg/$dm^3$from the stock solution was prepared. Then 4 ml of anthrone reagents was added to 1 ml of each of the standard glucose solution check the absorbance measured at 620nm in a Genway model 6000 electronic spectrophotometer using anthrone reagent as blank. A standard curve of absorbance against glucose concentration was prepared.

Calculation

The glucose concentration of the sample was extrapolated from the standard curve. Therefore, carbohydrate content of sample (g/100g or %) =glucose concentration (mg/dm^3) x 10, this was done for all the treated and raw samples in triplicate.

3.2.1.7 Estimation of food energy value of the samples

The energy value was calculated using the factors reported by (Onyeike *et al*, 2015). The value of crude protein content was multiplied by 4, Crude Lipid by 9 and Carbohydrate by 4kCal. The sum of these values areexpressed in kCal/100g sample and this was done in triplicate for all the seven samples

3.2.2.*InVitro*Assay for Protein Digestibility

The *invitro* protein digestibility was carried out according to the method of Mertz *et al*., (1984). To 200 mgof the powdered sample was dispensed in 20 mlof pepsin reagent (1.5mg/ml in 0.1M phosphate buffer of pH. 2.0) and shaken vigorously and then kept in a water bath at 37°C for three hours with constant shaking at 15 minutes interval. After 3 hours, the digestion was stopped by removing the tubes from the water bath and place in ice bath for 30 minutes. The sample was then filtered through whatman No.1 filter paper and the residue washed with buffer and dried at 80°C for 2 hours. The dried residue was placed in a 50 mlmicro-kjedahl flask and analyzed for nitrogen by micro-

kjedahl digestion, this was done in triplicate for all the samples and this was done in triplicate for all the treated and raw samples. The indigestible nitrogen was subtracted from total nitrogen of the sample to obtain digestible nitrogen based on the formula:

Digestible N (mg) = Total N in sample (mg) – N in residue (mg)

Digestible protein = Digestible N (mg)× $Conversion\ factor$

% *invitro* digestibility = $\frac{Digestible\ \ protein}{Total\ \ protein\ \ in\ \ sample} \times 100$

3.2.3Antinutrients Analysis

3.2.3.1 Determination of tannins

The Folin – Denis Spectrophotometric method was usedas described by Pearson (1976) to determine tannin contents in triplicate for the treated and raw seed. A measured weight of each sample (1.0g) was dispersed in 10ml distilled H_20 and agitated. This was left to stand for 30minutes at room temperature (37℃) being shaken every 5 minutes. At the end of 30 minutes, it was centrifuged and extract was gotten. About 2.5ml of the supernatant (extract) was dispersed into a 50ml volumetric flask and 2.5ml of standard tannic acid solution was dispersed into a separate 50ml flask. A 1.0ml folin Denis reagent was measured into each flask followed by 2.5ml of saturated Na_2CO_3 solution. The mixture was diluted to mark in the flask (50ml) and incubated for 90minutes at room temperature. The absorbance was measured at 725nm in a Genway model 6000 electronic spectrophotometer. The tannin content was expressed as follows:

$$\% \text{ Tannin} = \frac{An/As \times C \times 100}{Weig\,ht\ of\ \ sample\ \times five}$$

Where:

An = absorbance of test sample

As = absorbance of standard solution

C = Concentration of standard solution

W = Weight of sample used

V_f = Total volume of extract

V_a = Volume of extract analyzed.

3.2.3.2Determination of saponin:

The gravimetric method of AOAC (1984) employing the use of a soxhlet extractor and two different organic solvents was used. Five gram (5g) of dry ground sample was weighed into a thimble and transferred into the soxhlet extractor chamber fitted with a condenser and a flat bottom flask. Some quantity of acetone, enough to cause a reflux will be poured into the flask. The sample was exhaustively extracted of its lipid and interfering pigments for 3 hours by heating the flask on a hot plate and the solvent distilled off. This was the first extraction. For the second extraction, a pre-weighed round bottom flask was fitted into the soxhlet apparatus (bearing the sample containing thimble) and methanol that was enough to cause a reflux was poured into the flask. The saponin was then exhaustively extracted for 3 hours by heating the flask on a hot plate after which the solvent wasdistilled off. The flask was re-weighed. The difference between the final and initial weights of the flask represent the weight of saponin extracted. This was done in triplicate for all the treated and raw seed of *Sesbania sesban*

$$\% \text{ Saponin} = \frac{Weight\ of\ saponin}{Weight\ of\ sample} \times 100$$

3.2.3.3Determination ofoxalate

The titration method described by (Dey and Underwood 1986) was used to determine the oxalate content. One gram of the sample was weighed into 100ml conical flask where 75ml of 3N H_2SO_4 was added and stirred for 1 hour. It was then filtered using Whatman No.1 filter paper. From the filtrate, 25ml was taken and titrated while hot (80 – 90^oC) against 0.1N $KMnO_4$ solution, until a faint pink colour persisted for at least 30 seconds this determination was done in triplicate for all the seven samples.

The overall redox reaction is $MnO_4^- + C_2O_4^{2} + 8H+ Mn^{2+} + 4H_2O + 2CO_2$

Oxalate (mg/100mg) = $\frac{T \times Vme \times D.F}{ME \times Ms(g)} \times 100$

Where T = titre value of $KMnO_4$ (ml)

V_{me} = volume mass equivalent (1 ml of 0.05M $KMnO_4$ solution is

equivalent to 0.00225 anhydrous Oxalic acid.

D.F. = dilution factor (V_t/A = 75/25 = 3) where

Vt = total volume of titrate (filtrate, 75ml) and A is the aliquot used (25ml).

ME = molar equivalent of $KMnO_4$ redox reaction

Ms = mass of sample used.

3.2.3.4 Determination of cyanogenic glycoside

The extraction was according to Wang and filled method as described by Onwuka (2005) was used. A portion (5g) of sample was made and dissolved into 50ml distilled water. The extract was filtered and the filtrate was used for cyanide determination. To 1ml of sample filtrate, 4ml of alkaline picrate was added and absorbance was recorded

at 550nm and cyanide content was extrapolated from a cyanide standard curve. This was done in triplicate for all the samples.

Cyanide (mg/g) = $\frac{Absorbance \times G.F \times D.F}{W}$

Where

G.F. = Gradient factor and

D.F. = Dilution factor

3.2.3.5 Determination of Phytic Acid

Phytic acid was determined using the procedure described by Lucas and Maskakas (1975). A portion (2g) of each sample was weighed into 250ml conical flask; 100ml of 2% concentrated hydrochloric acid was used to soak each sample for 3hrs. The mixture was filtered and 50ml of each filtrate was placed in 250ml beaker and 107 ml of distilled water was added to each solution as indicator and it was titrated with standard iron chloride solution which contained 0.00195 iron per ml. This was done in triplicate for all the seven samples

% Phytic acid = Y x 1.19 x 100

Where Y= titre value x 0.00195

3.2.3.6Determination of Trypsin Inhibitor

Trypsin inhibitor was determined by method described by Onwuka, (2005). Extraction of sample: A known weight (1g) of grounded sample was dispersed into 50 ml of 0.5M NaCl solution.The mixture was stirred for 30minutes at room temperature and centrifuged. The supernatant was filtered through whatman filter paper. The filtrate (extract volume) was used for the assay. Preparation of standard trypsin and substrate: 1mg/ml of trypsin in 0.1M HCl was prepared, 1% casein substrate in 0.1M phosphate

buffer pH7.7 was prepared. To 2ml of trypsin standard solution, 1ml of trpsin inhibitors was added and incubated for 10minutes at 370^{o}C for 10minutes. A blank of 5ml substrate was prepared in a test-tube with no trypsin inhibitor extract added. The contents in the tube were left for 10minutes and the reaction was stopped by adding 3ml 5% TCA. It was filtered and then measured spectrophotometrically at 410nm. The trypsin inhibitor activity was expressed as the number trypsin unit inhibited (TUI) per unit weight of the sample analysed.

Calculation: TUI/MG = (b-a)/0.1 x F. Where b=absorbance of test sample solution, a=absorbance of blank. F=(1xVfxD)/W. Where W=Weight of sample, Vf=total volume of extract used in the assay. D= Dilution factor, Va = Volume of standard. This determination was done in triplicate for the seven samples.

3.2.4. Mineral Analysis

One gram (1g) of the samples was digested using 15ml of Hcl and 5ml of Nitric acid (3:1).Mineral compositions of the digested samples were determined using Shimadzu AAS 6800 Atomic absorption spectrophotometer.Sodium (Na), Potassium (K), Zinc (Zn), Iron (Fe), Calcium (Ca) and Magnesium (Mg) were determined by Atomic Absorption Spectrophotometry (AAS) AOAC (2003).

Wet digestion of sample

For wet digestion of sample, approximately (1.0g) of the powdered sample was taken in digesting glass tube. Twelve millilitres (12ml) of HNO_3 was added to the samples and mixtures were kept overnight at room temperature. Then 4.0 ml perchloric acid ($HClO_4$) was added to the mixture and was kept in the fumes block for digestion. The temperature was increased gradually starting from 50℃ and increasing up to 250-300℃. The digestion was completed in about 70-85 minutes as indicated by the appearance of

white fumes. The mixture was left to cool down and the content of the tube was transferred to 100ml volumetric flasks and the volumes of the contents was made to 100ml with distilled water. The wet digestion solution was tansfered to plastic bottles labeled accurately and stored for mineral determination.

Procedure

Digested samples were analyzed for mineral contents. The absorption measurement of the elements for the samples was read out. Different electrode lamps were used for each mineral. The equipment was run for standard solutions of each of the mineral before and during determination to check that it is working properly. The dilution factor for all minerals except Mg was 100, for determination of Mg, further dilution of the original solution was done using 0.5ml original solution and enough distilled water was added to it to make the volume up to 100ml. Also, the determination of calcium (Ca), 1.0ml lithium oxide solution was added to the original solution to unmask calcium from magnesium. The concentration of minerals was determined using the formula below

Mw = absorbance (ppm) x dry wt. x D/wt of sample x100

Where Mw = conc. of mineral

D = Dilution factor

Determination of sodium and potassium were done by method of flame photometry. The same wet digested sample solution as used in AAS was used for the determination of Na and K. Standard solutions of 20, 40, 60, 80 and 100milli equivalent/L were used for both Na and K. The calculations for the total minerals involve the same procedure as given in AAS. All mineral determined was done in triplicate for all the samples.

3.2.5 Vitamin Determination

Vitamins were determined using Shimadzu UV – 2550 UV-Vis spectrophotometer. Vitamin A (β-carotene), B_1, B_2, B_3, B_6, B_{12}, C, and E were determined using Shimadzu UV-2550 UV spectrophotometer, using the method described by Onwuka (2005). The vitamin was done in triplicate for all the treated and raw samples.

3.2.6 Statistical Analysis

The data obtained were expressed as mean ± standard deviation (SD). The data generated were analyzed using ANOVA (SPSS version 20). Duncan multiple range test was used for multiple mean comparism to determine differences between the means. Significance was accepted at 0.05 of probability ($P<0.05$).

CHAPTER FOUR

4.0 RESULTS

4.1 Effect of Processing on Proximate Compositionof *Sesbania sesban*seeds

The effect of processing on the proximate composition of *Sesbania sesban* seeds is shown in Table 4.1. Crude protein content (25.66±0.52%) revealed that the raw seed (RS) differ significantly (P<0.05) compared to the value revealed in a sample soaked with water(SW) (24.86±0.04%). The crude protein in the RS also differ significantly (P<0.05)compared to the composition (24.75±0.58%) revealed in a sample soaked with calcium hydroxide (SC) and sample soaked with lye (SL) (24.77±0.60%). Also, the percentage crude protein contents revealed in the boiled samples (BC)differ significantly (P<0.05) as compared to the compositions revealed in the soaked samples (Table 4.1).

The percentage carbohydrate content (59.51±0.29%) of the raw seed (RS) differ significantly (P<0.05) as compared to the composition (49.52±0.72) revealed in the sample boiled using water(BW). The Carbohydratecontent in RS also differ significantly compared to the content (52.96±0.61) revealed in SW. Also,significant (P<0.05) differences exists between the carbohydrate content in RS as compared to the compositions revealed in boiled with calcium hydroxide (BC), boiled with lye (BL), soaked with calcium hydroxide (SC) and soaked with lye (SL).

Moisture content indicates RS to be significantly different (p<0.05) from other processed samples. BW, BC and BL moisture showed significant (p<0.05) difference from that of SW, SC and SL.The percentage ash composition of RS (2.66 ± 0.10%) differ significantly (P < 0.05) as compared to the compositions revealed in the BW, BL, SC and SL respectively.

The percentage crude fat contents are indicated in Table 4.1. The percentage crude fatcomposition (6.09 ± 0.04%) of sample soaked with water (SW) differ significantly ($p<0.05$)compared to the compositions (6.23 ± 0.03 and 6.34 ± 0.09) revealed in a sample boiled with water(BW) and sample boiled with lye (BL). Similarly, the percentage crude fat content (6.34 ± 0.09%) of BL differ significantly ($P < 0.05$) as compared to the composition (6.18 ± 0.12%) revealed in raw sample(RS).

Soluble fibre content showedthat RS (3.63 ± 0.02)was significantly ($P < 0.05$) different when compared to other processed samples BW (4.15 ± 0.04),sample boiled with calcium hydroxide 3.18 ± 0.04(BC), BL (3.46 ± 0.02) , SW (4.31 ± 0.01) , soaked with calcium hydroxide 4.72 ± 0.05(SC) and sample soaked with lye 3.83 ± 0.05 (SL). The percentage composition revealed in the BW differ significantly ($P < 0.05$) as compared to BC, BL SW, SC and SL respectively. Similarly, the percentage soluble fibre composition indicated in the BL differs significantly ($P < 0.05$) compared to the compositions revealed in the BW, BC, SW, SC, SL and RS respectively. Also, the composition revealed in the SW differ significantly ($P < 0.05$) as compared to the compositions showed in BW, BC, BL, SC, SL and RS. Similar observation was revealed between SC and BW, BC, BL, SW, SL and RS (Table 4.1).

Observation in this study indicated that the processed seeds and the unprocessed seed contained insoluble fibres. Findings in this study revealed that the percentage insoluble fibre compositions in the RS (11.72 ± 0.08) differ significantly ($P<0.05$) as compared to the composition revealed in the processed samples BW (10.08 ± 0.05), BC (10.50 ± 0.20), BL (9.75 ± 0.12), SW (8.25 ± 0.08), SC (9.84 ± 0.09), SL (8.40 ± 0.02) (Table 4.1). Similarly, the composition showed in BW differ significantly ($P < 0.05$) as

compared to the composition revealed in the BC, BL, SW, SC, SL and RS. Also the composition revealed in the BC differ significantly ($P < 0.05$) as compared to the compositions revealed in the BW, BL, SW, SL, SC and RS (Table 4.1). The RS shows higher food energy value (FEV) as compared to the compositions showed in the BW, BC, BL, SW, SC and SL (Table 4.1).

Table 4.1: Effect of Processing on Proximate Composition (%) of *Sesbania sesban* Seed

Proximate	BW	BC	BL	SW	SC	SL	RS
Moisture	4.22 ± 0.10^{a}	4.11 ± 0.08^{a}	4.11 ± 0.12^{a}	5.11 ± 0.06^{c}	5.12 ± 0.05^{c}	5.10 ± 0.09^{c}	4.64 ± 0.12^{b}
Ash	2.52 ± 0.03^{ab}	2.60 ± 0.05^{bc}	2.47 ± 0.04^{ab}	2.56 ± 0.12^{abc}	2.45 ± 0.04^{a}	2.51 ± 0.07^{ab}	2.66 ± 0.10^{c}
Crude fat	6.23 ± 0.03^{bc}	6.16 ± 0.05^{ab}	6.34 ± 0.09^{c}	6.09 ± 0.04^{a}	6.11 ± 0.03^{ab}	6.10 ± 0.08^{a}	6.18 ± 0.12^{ab}
Crude Protein	26.07 ± 0.05^{b}	26.09 ± 0.03^{b}	26.14 ± 0.06^{b}	24.86 ± 0.04^{a}	24.75 ± 0.58^{a}	24.77 ± 0.60^{a}	25.66 ± 0.52^{b}
Soluble Fiber	4.15 ± 0.04^{d}	3.81 ± 0.04^{c}	3.46 ± 0.02^{a}	4.31 ± 0.01^{e}	4.72 ± 0.05^{f}	3.83 ± 0.05^{c}	3.63 ± 0.02^{b}
Insoluble Fiber	10.08 ± 0.05^{c}	10.50 ± 0.20^{d}	9.75 ± 0.12^{b}	8.25 ± 0.08^{a}	9.84 ± 0.09^{b}	8.40 ± 0.02^{a}	11.72 ± 0.08^{e}
Carbohydrate	49.52 ± 0.72^{a}	48.64 ± 0.84^{a}	52.44 ± 0.40^{bc}	52.96 ± 0.61^{c}	53.33 ± 0.41^{c}	51.54 ± 0.34^{b}	59.51 ± 0.29^{d}
FEV(kCal)	358.43	354.40	371.38	366.04	367.40	360.14	396.30

Values are mean ± SD of triplicate determinations. Values across the rows with different superscripts differ significantly ($P<0.05$). BW – Boil with water, BC – Boil with calcium hydroxide, BL – Boil with lye, SW – Soak with water, SC – Soak with calcium hydroxide, SL–Soak with lye, RS – Raw sample, FEV−Food EnergyValue

4.2 Effect of Processing on Antinutrients Compositions of *Sesbania sesban* seeds

Result in Table 4.2 showedthe tannin contents in raw samples (RS), samples boiled with water(BW),sample boiled with lye(BL), sample boiled with calcium hydroxide(BC), sample soaked with water (SW), sample soaked with calcium hydroxide(SC) and sample soaked with lye(SL). The boiled samples (BW, BC and BL) revealed significant ($P < 0.05$) reduction in tannins content as compared to the composition revealed in the soaked samples (SW 0.76 ± 0.03, SC 0.59 ± 0.03 and SL 0.83 ± 0.04). The BW (0.39 ± 0.03) showed a significant ($P < 0.05$) composition of tannin as compared to the compositions revealed in BC 0.33 ± 0.02 and BL 0.31 ±0.03. The composition of tannins in SW differs significantly as compared to the composition of tannins in SC and SL. Generally, the processed samples (BW, BC, BL, SW, SC and SL) showed a significant ($P < 0.05$) reduction in the tannin composition as compared to the composition revealed in the RS.

The composition (0.43 ± 0.04%) of saponin revealed in the raw sample (RS) differ significantly ($P < 0.05$) as compared to the contents of saponin indicated in sample boiled with water (BW), sample boiled with calcium hydroxide(BC),sample boiled with lye (BL), sample soaked with water(SW), sample soaked with calcium hydroxide(SC) and sample soaked with lye (SL)(Table 4.2). The RS contained more saponin than in the processed samples. The boiled samples (BW 0.25 ± 0.01%, BC0.18 ± 0.02 and BL0.18 ± 0.02%) showed statistically significant ($P < 0.05$) reduction in the saponin composition as compared to the contents in the soaked samples0.38±0.02SW,0.33±0.05SC and 0.32±0.04SL.

The composition (0.25 ± 0.01%) showed in the BW differ significantly ($P < 0.05$) as compared to the compositions (0.18 ± 0.02 and 0.18 ± 0.02%) revealed in the BC and BL.

The oxalate compositions shown in the processed samples and the unprocessed sample are shown in table 4.2. The results showed that the soaked samples revealed significant reduction in oxalate compared to the boiled samples (BW and BC). Generally, the processed samples (BW3.13±0.04, BC3.09±0.02, BL2.10±0.01, SW2.86±0.04, SC 2.81± 0.06and SL2.79 ±0.11) showed significant ($P<0.05$) reduction in the oxalate composition as compared to the RS4.24 ± 0.08.

Table 4.2: Effect of Processing on Antinutrients Composition of *Sesbania sesban* Seed

Test	BW	BC	BL	SW	SC	SL	RS
Tannins(%)	0.39 ± 0.03^{b}	0.33 ± 0.02^{a}	0.31 ± 0.03^{a}	0.76 ± 0.03^{d}	0.59 ± 0.03^{c}	0.83 ± 0.04^{e}	1.32 ± 0.03^{f}
Saponin(%)	0.25 ± 0.01^{b}	0.18 ± 0.02^{a}	0.18 ± 0.02^{a}	0.38 ± 0.02^{e}	0.33 ± 0.05^{d}	0.32 ± 0.04^{c}	0.43 ± 0.04^{e}
Oxalate(mg/100g)	3.13 ± 0.04^{c}	3.09 ± 0.02^{c}	2.10 ± 0.01^{a}	2.86 ± 0.04^{b}	2.81 ± 0.06^{b}	2.79 ± 0.11	4.24 ± 0.08^{d}
Phytate(mg/100g)	0.64 ± 0.05^{ab}	0.70 ± 0.03^{c}	0.75 ± 0.05^{d}	0.62 ± 0.06^{a}	0.63 ± 0.05^{ab}	0.64 ± 0.08^{ab}	1.29 ± 0.09^{e}
Cyanogenic Glycoside(mg/kg)	6.70 ± 0.23^{b}	6.27 ± 0.04^{a}	6.24 ± 0.24^{a}	7.69 ± 0.08^{c}	7.55 ± 0.13^{c}	7.62 ± 0.03^{c}	12.81 ± 0.08^{d}
Trypsin Inhibitor(TIU)	5.84 ± 0.06^{c}	4.72 ± 0.10^{b}	4.32 ± 0.11^{a}	8.22 ± 0.10^{e}	7.29 ± 0.12^{d}	8.88 ± 0.07^{f}	13.50 ± 0.11^{g}

Values are mean ± SD of triplicate determinations. Values across the rows with different superscripts differ significantly ($P<0.05$). BW – Boil with water, BC – Boil with calcium hydroxide, BL – Boil with lye, SW – Soak with water, SC – Soak with calcium hydroxide, SL – Soak with lye, RS – Raw sample

Also, in the case of phytate, the processed samples (BW 0.64 ± 0.05, BC 0.70 ± 0.03, BL 0.75 ± 0.05, SW 0.62 ± 0.06, SC 0.63± 0.05and SL 0.64 ± 0.08) showed a significant ($P < 0.05$) reductionin the phytate composition as compared to the contents (1.29 ± 0.09mg/100g) shows in RS(Table 4.2).

The cyanogenic glycosides compositions revealed in sample boiled with water (BW), sample boiled with lye (BL), sample boiled withcalcium hydroxide(BC), sample soaked with water (SW), sample soaked with calcium hydroxide (SC), sample soaked with lye (SL) and raw sample (RS)are shown in table 4.2. The result showed a significant ($P < 0.05$) increase in cyanogenic glycoside composition (12.81 ± 0.08mg/Kg) of RS as compared to the compositions revealed in BW 6.70 ± 0.23, BC 6.27 ± 0.04, BL 6.24 ± 0.24, SW 7.69 ± 0.08 , SC 7.55 ± 0.13 and SL 7.62 ± 0.03mg/kg. Similarly, the compositions revealed in the soaked samples (SW, SC and SL) differ significantly ($P < 0.05$) compared to the composition revealed in the boiled samples (BW and BC), except the composition (6.27 ± 0.04mg/Kg) revealed in BL.

The trypsin inhibitor is significantly ($P < 0.05$) higher in the RS (13.50 ±0.11TIU) as compared to the levels shows in the processed samples. Generally, the trypsin inhibitors differ significantly ($P < 0.05$) in all the samples (Table 4.2). The levels of trypsin inhibitors revealed in the boiled samples differ significantly ($P < 0.05$) as compared to the levels observed in the soaked samples SW 8.22± 0.10, SC 7.29 ± 0.12 and SL 8.88±0.07. The compositions in the boiled samples (BW 5.84 ± 0.06, BC 4.72 ±0.1 and BL 4.32 ± 0.11) differ significantly ($P < 0.05$) differ from one another. Likewise, the compositions revealed in the soaked samples (SW, SC and SL) also differ significantly ($P < 0.05$) from one another.

Table 4.3: Effect of Processing on Percentage Reduction of Antinutrients

Antinutrients	BW	BC	BL	SW	SC	SL
Tannins	70.45	75.00	76.52	42.42	55.30	37.12
Saponin	41.86	58.14	58.14	11.63	23.25	25.58
Oxalate	26.17	27.12	50.47	32.55	33.73	34.20
Phytate	50.39	45.74	41.86	51.94	51.16	50.39
Cyanogenic Glycoside	47.70	51.05	51.29	39.97	41.06	40.52
Trypsin Inhibitor	56.74	65.04	68.00	39.11	46.10	34.22

BW – Boil with water, BC – Boil with calcium hydroxide, BL – Boil with lye, SW – Soak with water, SC – Soak with calcium hydroxide, SL – Soak with lye

4.4 Effects of Processing on Vitamin Content of*Sesbania sesban*seeds

The effect of processing on the vitamin Acontents of *Sesbania sesban* seed is shown in Table 4.4. The sample boiled with water BW(4.087 ± 0.074), sample boiled with calcium hydroxide BC 5.208± 0.196 and sample boiled with lye (BL 7.535± 0.463) and the sample soaked with water SW 4.438 ± 0.291, sample soaked with calcium hydroxide SC 3.099± 0.930 and sample soaked with lye SL(6.71 ± 0.350) revealed significant ($P < 0.05$) reduction in the βC contents as compared to the composition revealed in the raw sample (RS).

The composition (30.789 ± 0.535) of vitamin B_1 showed in (RS)was significantly ($P < 0.05$) higher than the contents revealed in the processed samples (BW 9.874 ±0.558, BC 4.315 ± 0.273, BL 23.641 ± 0.532, SW 13.560 ±0.466, SC 12.843 ±0.168, and SL 5.075 ±0.122 (Table 4.4). Similarly, the contents (29.233 ± 0.361) of vitamin B_2 shown in the RS differ significantly, as compared to the compositions revealed in other processed samples BW 17.296 ± 0.431, BC 17.191 ± 0.599, BL 19.932 ± 0.425, SW 18.003 ± 0.028, SC 1.393 ± 0.178, and SL 24.283 ± 0.733. The composition (1.399 ± 0.178) of vitB_3 shown in RS, differ significantly as compared to the contents revealed in the boiled samples BW 0.929 ± 0.065, BC 1.017 ± 0.001 and BL 0.921 ± 0.046. Similarly, the contents shown in the soaked sample (SW 1.152 ±0.224, SC 0.962 ± 0.029 and SL 0.936 ± 0.048) also differ significantly ($P < 0.05$) as compared to the content indicated in the RS. Just like vit B_1, B_2 and B_3, the content of vitB_6 11.321 ± 0.304 in the RS differ significantly ($P<0.05$) compared to the contents revealed in the processed samples BW 7.223 ± 0.120, BC 8.362 ± 0.402, BL 10.554 ± 0.205, SW 8.857 ± 0.655, SC 7.043± 0.208, and SL 9.680 ± 0.046. Also, the

contents of vitB_6 revealed in soaked samples (SW and SL) differ significantly ($P < 0.05$) as compared to the composition revealed in the boiled samples (BC and BL).

The contents of VitB_9 as revealed in the RS (0.029 ± 0.006) differ significantly as compared to the contents shown in BW 0.040 ± 0.003, BC 0.042 ± 0.003 and SC 0.043 ± 0.004. There was a significant ($p<0.05$) reductionin in the vitamin C contents of the processed samples (BW 0.032 ± 0.004, BC 0.005 ± 0.001, BL 0.066 ± 0.004, SW 0.029 ± 0.002, SC 0.031 ±0.002 and SL 0.031 ± 0.009) as compared to the composition (0.124 ± 0.027) revealed in RS. Similarly, the contents of vitamin C indicated in BC and BL also differ significantly ($P < 0.05$) compared to the contents revealed in BW, SW, SC and SL respectively.

The composition of Vitamin E in the processed samples and unprocessed sample (RS) was shown in table 4.4. The content of vitamin E showed in the RS differ significantly ($P < 0.05$) as compared to the contents revealed in the boiled samples BW 10.193 ± 0.078, BC 10.371 ±.474, BL 9.712 ± 0.136, SC 6.477 ± 0.435 and SL 14.449 ± 0.345 respectively. The content of vitamin E as revealed in the SL is significantly 0 ($P < 0.05$) higher than the contents revealed in RS, boiled samples, SW 8.633 ± 0.102 and SC.

Table 4.4 Effect of Processing on Vitamin (mg/100g) Contents of *Sesbania sesban* Seed

Test	BW	BC	BL	SW	SC	SL	RS
βC(IU)	4.087 ± 0.074^{ab}	5.208 ± 0.196^{c}	7.535 ± 0.463^{d}	4.438 ± 0.291^{c}	3.099 ± 0.930^{a}	6.714 ± 0.350^{d}	10.915 ± 0.995^{e}
Vit B_1	9.874 ± 0.558^{c}	4.315 ± 0.273^{a}	23.641 ± 0.532^{e}	13.560 ± 0.466^{d}	12.843 ± 0.168^{d}	5.075 ± 0.122^{b}	30.789 ± 0.535^{f}
VitB_2	17.296 ± 0.431^{b}	17.191 ± 0.599^{b}	19.932 ± 0.425^{c}	18.003 ± 0.028^{b}	1.393 ± 0.178^{a}	24.283 ± 0.733^{d}	$29.223\pm0..361^{e}$
Vit B_3	0.929 ± 0.065^{a}	1.017 ± 0.001^{a}	0.921 ± 0.046^{a}	1.152 ± 0.224^{ab}	0.962 ± 0.029^{a}	0.936 ± 0.048^{a}	1.399 ± 0.399^{b}
Vit B_6	7.223 ± 0.120^{a}	8.362 ± 0.402^{b}	10.554 ± 0.205^{d}	8.857 ± 0.655^{b}	7.043 ± 0.208^{a}	9.680 ± 0.046^{c}	11.321 ± 0.304^{e}
Vit B_9	0.040 ± 0.003^{bcd}	0.042 ± 0.003^{cd}	0.035 ± 0.004^{abcd}	0.032 ± 0.005^{ab}	0.043 ± 0.004^{d}	0.033 ± 0.008^{abc}	0.029 ± 0.006^{a}
Vit C	0.032 ± 0.004^{b}	0.005 ± 0.001^{a}	0.066 ± 0.004^{c}	0.029 ± 0.002^{b}	0.031 ± 0.002^{b}	0.031 ± 0.009^{b}	0.124 ± 0.027^{d}
VitE(IU)	10.193 ± 0.078^{cd}	10.371 ± 0.474^{d}	9.712 ± 0.136^{c}	8.633 ± 0.102^{b}	6.477 ± 0.435^{a}	14.449 ± 0.345^{e}	8.522 ± 0.380^{b}

Values are mean± SD of triplicate determinations. Values across the rows with different superscripts differ significantly ($P<0.05$). BW – Boil with water, BC – Boil with calcium hydroxide, BL – Boil with lye, SW – Soak with water, SC – Soak with calcium hydroxide, SL – Soak with lye, RS – Raw sample. βC – Beta carotene, Vit B1 – Vitamin B1, Vit B2 – Vitamin B2, Vit B3 – Vitamin B3, Vit B6 – Vitamin B6, Vit B9 – Vitamin B9, Vit C – Vitamin C, Vit E – Vitamin E

4.5 Effect of Processing on the Mineral composition of *Sesbania sesban* Seeds

As shown in table 4.5, the mineral indicates highersodium content. The result showed that the composition of sodium in sample boiled with water (BW 289.57 ± 1.08) and sample boiled with lye (BL 288.17 ± 1.95)is significantly ($P < 0.05$) higher than the sodium composition revealed in sample soaked with water(SW 159.43 ± 1.02), (SC 273.58 ± 1.03) and (SL 154.13 ± 1.93). Similarly, the content of sodium in raw sample(RS) is significantly higher than the content revealed in (BC 239.71 ± 0.10), SW and SL.

The content of Magnesium shown in the soaked samples (SW 50.82 ± 0.74), SC 51.13 ± 0.54, and SL 50.35 ± 0.27) were significantly ($P < 0.05$) higher than the compositions revealed in the boiled samples (BC 44.50 ± 0.05 and BL 45.55 ± 0.10), except BW 47.35 ± 0.78 which showed no significant ($P > 0.05$) difference as compared to the content in the soaked samples. Moreso, statistically significant ($P < 0.05$) difference exist between the contents of magnesium in RS as compared to magnesium contents revealed in BW, BC, SW, SC and SL. The composition of potassium shown in this study irrespective of the processing method differ significantly ($P < 0.05$). The contents of potassium in the boiled samples BW 29.08 ± 0.13, BC 46.05 ± 0.13 BL 30.70 ± 0.25 showed significant ($P < 0.05$) reduction in potassium when compared to the contents revealed in the soaked samples SW 162.95 ± 0.15, SC 116 ± 0.54 and SL 137.28 ± 0.24. The potassium composition (110.68 ± 0.87ppm) shown in RS is significantly ($P < 0.05$) higher than the contents revealed in the boiled samples (BW, BC and BL). However, the content revealed in RS is significantly ($P < 0.05$) lower than the content shown in the soaked samples (SW, SC and SL).

The composition of iron revealed in the raw sample RS (33.67 ± 0.76ppm) differ significantly ($P < 0.05$) as compared to the compositions (16.88 ± 0.28, 20.43 ± 0.08 and

25.56 ± 0.11ppm) revealed in sample boiled with water(BW), sample boiled with lye (BL) and sample soaked with calcium hydroxide (SC) respectively.

The calcium compositions (11.75 ± 0.27 and 15.38 ± 0.27ppm) revealed in sample boiled with calcium hydroxide (BC) and sample boiled with lye (BL) differ significantly ($P < 0.05$) as compared to the content (14.50 ± 0.05ppm) shows in raw sample RS (Table 4.5).

The content (1.00 ± 0.50ppm) of manganese as indicated in RS showed no significant ($P > 0.05$) difference as compared to the contents revealed in boiled samples BW 1.00 ± 0.87, BC 0.83 ± 0.29, BL 3.00 ± 0.50, SW 1.33 ± 0.29 and SL 1.83 ± 0.76, although differ significantly ($P < 0.05$) as compared to the content (4.17 ± 0.87ppm) revealed in SC 4.17 ± 0.87 (Table 4.5). Observation in this study showed that the content (2.50 ±0.02ppm) of copper revealed in RS differ significantly ($P < 0.05$) as compared to the contents indicated in BW 1.18 ± 0.06 , BL 1.55 ± 0.09 , SC 3.56 ± 0.06 , and SL 0.87 ± 0.12 respectively.

Table 4.5: Effect of Processing on Mineral Contents of *Sesbania sesban* Seed

Mineral (mg/100g)	BW	BC	BL	SW	SC	SL	RS
Na	289.57±1.08^{f}	239.71±0.10^{c}	288.17±1.95^{f}	159.43±1.02^{b}	273.58±1.03^{e}	154.13±1.93^{a}	269.83±0.76^{d}
Mg	47.35±0.78^{bc}	44.50±0.05^{a}	45.55±0.10^{b}	50.82±0.74^{c}	51.13±0.54^{c}	50.35±0.27^{c}	45.94±0.10^{b}
K	29.08±0.13^{a}	46.05±0.13^{c}	30.70±0.25^{b}	162.95±0.15^{g}	116.33±0.54^{e}	137.28±0.24^{f}	110.68±0.87^{d}
Fe	6.88±0.28^{c}	31.48±2.700^{d}	0.43±0.08^{a}	39.27±0.68^{e}	2.56±0.11^{b}	33.25±0.23^{d}	33.67±0.76^{d}
Ca	14.32±0.20^{bc}	11.75±0.27^{a}	15.38±0.27^{d}	14.22±0.08^{b}	14.67±0.26^{c}	14.65±0.18^{c}	14.50±0.05^{bc}
Zn	1.00±0.87^{a}	1.17±0.29^{a}	1.83±0.29^{ab}	1.50±0.50^{a}	2.67±0.58^{b}	1.67±0.76^{ab}	3.83±0.76^{c}
Mn	1.00±0.50^{ab}	0.83±0.29^{a}	3.00±0.50^{c}	1.33±0.29^{ab}	4.17±0.87^{d}	1.83±0.76^{b}	1.00±0.50^{ab}
Cu	1.18±0.06^{b}	2.46±0.04^{d}	1.55±0.09^{c}	2.41±0.09^{d}	3.56±0.06^{e}	0.87±0.12^{a}	2.50±0.02^{d}

Values are mean ± SD of triplicate determinations. Values across the row with different superscripts differ significantly ($P<0.05$). BW – Boil with water, BC – Boil with calcium hydroxide, BL – Boil with lye, SW – Soak with water, SC – Soak with calcium hydroxide, SL – Soak with lye, RS – Raw sample. Na – Sodium, Mg – Magnesium, K – Potassium, Fe – Iron, Ca – Calcium, Zn – Zinc, Mn – Manganese, Cu – Copper.

4.6 Effect of processing on *Invitro* Protein Digestibility of *Sesbania sesban* seed

Results in table 4.6 showed the *invitro* protein digestibility. The percentage *invitro* protein digestibility in all the samples varies significantly ($P < 0.05$). The raw sample53.720 ± 0.120% (RS) showed significant ($P < 0.05$) reduction in the*Invitro* protein digestibility as compared to the percentage composition revealed in the processed samples. The soaked samples (SW 79.620 ± 0.140, SC 80.660 ± 0.203 and SL 78.237 ± 0.164) showed higher significant ($P < 0.05$) compositions of *invitro* protein digestibility as compared to the percentage composition (53.720 ± 0.120%) revealed in RS. Also, the percentage *invitro* protein digestibility revealed in the boiledsamples (BW 85.767 ± 0.483, BC 84.363± 0.11 and BL 89.233 ± 0.172) differ significantly as compared to the percentage composition (53.720 ± 0.120%) revealed in RS (Table 4.6).

Table 4.6Effect of Processing on *Invitro* Protein Digestibility of *Sesbania sesban* seed

Sample	*In vitro* Protein Digestibility (%)
BW	85.767 ± 0.483^{t}
BC	84.363 ± 0.118^{g}
BL	89.233 ± 0.172^{e}
SW	79.620 ± 0.140^{c}
SC	80.660 ± 0.203^{d}
SL	78.237 ± 0.164^{b}
RS	53.720 ± 0.120^{a}

Values are mean ± SD of triplicate determinations. Value down the column with different superscripts differ significantly ($P<0.05$). BW – Boil with water, BC – Boil with calcium hydroxide, BL – Boil with lye, SW – Soak with water, SC – Soak with calcium hydroxide, SL – Soak with lye, RS – Raw sample

CHAPTER FIVE

5.0 DISCUSSION

The demand for protein rich foods is beyond recent times, and animal protein has been the major source of protein to people all over the world. For quite sometimes and presently the demand and consumption of poultry has declined obvious reason for this, may be due to economic dwindling especially in the developing and underdeveloped countries. As a result, the prices of most of the foodstuffs used in the preparation of animal feed are on the increase. And this inturn also has made it almost impossible for the average income earners to consume meats. So, the need to providing alternative underutilized nutrient rich feedstuff becomes necessary. *Sesbania sesban* is a nutrient rich plant widely grown, especially in Nigeria but has remain untapned or underutilized especially in Nigeria. The nutritional information of this plant seed is scanty. However, Arekemase *et al*. (2013) revealed that the seed from *Sesbania sesban* contained appreciable nutrients contents and that, if properly processed can be used as an alternative livestock feed. In this present study, different processing methods were employed.

In the proximate composition, the low moisture content in the unprocessed and processed sample can be spared from microbial attack. Low moisture is very necessary to retain the desired quality of the samples. Retaining the desired quality of *Sesbania sesban* will make it convenient for consumption by livestock. Sufficient amount of safe feeds is key to sustaining life and promoting good health of the livestock and that of the consumers. The moisture contents observed in this study are comparable to the moisture content previously revealed in study of Arekemase *et al*., (2013). Although the moisture contents in foodstuffs and feeds absolutely depends on the dgree of dryness, a function of time and temperature.

For nutritional evaluation, ash determination is an important component of proximate analysis. It refers to inorganic residue remaining after complete oxidation of the organic matter in food sample (Ismail, 2017). Generally, the ash contents obtained in this study, were lower compared to the value (8-51%) reported for *Sesbania grandiflora* (Kumar *et al*., 2017).

Low crude fat content obtained in this study is an indication that *Sesbania sesban* seed is a poor source of lipid. As such, there is need to complement the plant seed with lipids rich plant seeds for a good nutritional labeling and balancing. The importance of lipids as a component of feedstuff can not be overemphasized. It presence in feedstuff is important in improving the absorption of fat soluble vitamins, and overall, enhancing productive performance in poultry (Cetingul and Yardimci, 2008).

Lipid inclusion in animal feed is very necessary, especially in feeds of which its primary source isdeficient of lipid. Aside carbohydrate, lipids are high energy yield food molecule, as they mostly contain triglycerides, a high energy molecule. Most lipids are primarily composed of triglycerides, but other lipids compound may also be present, and this can affect their physical and chemical properties, as well as their energy value to livestock (Kerr *et al*., 2015).

The percentage crude fat obtained in this study conform to the composition (5-6%) revealed in study by Ali *et al*.(2015). The percentage crude fat composition is also comparable to the percentage crude fat composition (4.7-6.0%) reported for different species of *Sesbania*, including *Sesbania sesban***.**

Carbohydrate had the highest percentage composition irrespective of the processing methods. The percentage compositions obtained in this study were higher than the composition (26.40 ± 0.02 and 24.93 ± 0.00%) reported for the raw and fermented seeds of *Sesbania Spp* (Ishola *et al*.,

2018). The difference may be attributed to the species, although the exact species was not stated in their study. Moreso, the percentage carbohydrate composition (48.64 ± 0.001) revealed in *Sesbania aculeate* (Nayak *et al*.,2018) was lower than the composition revealed in this study implying that the percentage carbohydrate compositions obtained in this study, irrespective of the processing methods, were reasonable to consider seeds of *Sesbania sesban* a good source of carbohydrate.

Generally, carbohydrate is an important nutrient for healthy life, as it delivers about 4kCal of energy / gram (Kokkinidou *et al*., 2018). Although energy does not come from carbohydrate in the form of calories but as nutrient capable of being absorbed and metabolized by animals to yield energy or for anabolic purposes (Hail and Eastridge 2014). The higher percentage carbohydrate composition in the raw (unprocessed) as compared to the composition revealed in the processed sample, suggest that the carbohydrate compositions of *Sesbania sesban* is subject to changes if treated with calcium hydroxide, lye and water (Boiled or Soaked), although boiling has a more negative (reduction) implication on the carbohydratecomposition than Soaking, as observed in this study.

Protein from plant account for the major dietary protein that are for animal (Henchion *et al*., 2017), but of these plants (e.g soybean) used as a source of protein for livestock are becoming almost impossible to acquire especially among the average and poor farmers. This has led to a shift in seeking for alternative source of plant protein for livestock breeding. The percentage crude protein compositions in the unprocessed and processed samples are reasonable enough to consider the seeds of this plant (*Sesbania sesban*) a good source of protein. Since most animal feedstuff are low in protein, thus supplementing with seeds of *Sesbania sesban* may be necessary and may contributes in overall wellbeing of livestocks. The level of crude protein composition

observed in this study were lower than the content (37.69%) reported for soybean (Etiosa *et al*., 2017) as a source of protein ingredient for livestock. This shows that soybean is still one of the leading source of plant protein in poultry feeds. So,the crude protein reported in this study can conveniently replace soybean as a source of protein for livestock, then there is need to upgrade the protein quality of seeds of *Sesbania sesban*. This may be achieved through fermentation using different micro - organism as fermentation has been shown to improve protein quality of plants-based foods (Steve, 2012).

In this study, the protein contentsobserved in this study were lower than the compositions (33.1% and 34.5%) reported for other species of *Sesbania (Sesbania aculeate and Sesbania gradifora* (Hossain *et al*., 2002; Solorio - Sanchez *et al*., 2000). The crude protein reported in this study can conveniently replace soybean as a source of protein for livestock. The marginal increase in the protein composition of the boiled samples as compared to the soaked sample and raw sample could be attributed to extruding of more water in the sample. This may cause further reduction in the water content in the seeds thereby improve the protein quality of the seeds. The loss of water in foods is subject to time and temperature. At a required temperature and time appropriate, removal of water (free water) can be achieved. But at a higher temperature in the case of boiling bound water to protein (absorbed water) may be extruded resulting in a freer protein. This could be the probable reason as to why we observed a marginal increase of protein in our processed (boiled samples).

Contrary to our observation, study has observed a slight decrease in the protein composition of roasted plant sample (Danhassan *et al*., 2018). Further processing of the plant sample after boiling may in part contribute in the difference. In this study, after boiling, the plants were

thoroughly dried, causing a further decrease in the moisture content, which may be the reason why higher increase was observed in the boiled samples

The soluble and insoluble fibers evaluated in this study are both refered to as dietary fiber. It is part of plant-based food which promotes healthy living especially in humans. In a broader definition, dietary fiber is the edible part of plants, analogous to carbohydrates that are persistant to digestion and absorption in the human intestine with complete or partial fermentation in the large intestine (Li and Komarek, 2017) and promote beneficial physiological well being of an individual.

The processed seeds except boiling with lye (BL) of this plant revealed a slight increase in the soluble fiber content as compared to the raw seeds suggesting boiling or soaking of the raw seeds of*Sesbania sesban* with lye, slaked lime and water softens the fiber contents of the plant seed, thereby making it bioavailable within the food matrix. This observation corrobate with observation revealed in the work of Azizah and Zainon (1997) although in different plants seeds that boiling and soaking slightly increased soluble dietary fiber in soybean, mung bean and groundnut, which are also leguminous plant seeds. Contrary observation was revealed in the case of insoluble fiber. The decrease in the level of insoluble fiber of processed seeds of *Sesbania sesban*conformedwith previous observation in the work of Joshua *et al*. (2012). They highlighted that processing methods like boiling affects (reduces) the fiber content in plant-based food, and reduction is a function of time.

As observed in this study, the high nutrients composition in the seeds of *Sesbania sesban* corroborate with previous findings that *Sesbania sesban* contain apple amount of nutrients needed by livestock (Arekemase *et al*., 2013). However, the antinutritional factors present are

major challenges to utilizing the seeds of Egyptian riverhemp(*Sesbania sesban)* as a source of feeds for livestock.

The presence of antinutrients in feeds interferes with mineral bioavailability and functions. Therefore, the need to further process the raw seeds of *Sesbania sesban* has become necessary. All the processing methods employed in this study reduce the levels of the antinutritional factors in the raw seeds, the reduction of the antinutrients in seeds of *Sesbania sesban* varies from one processing methods as soaking had a better reduction of some antinutrients (oxalate and phytate) while boiling also had a better reduction of some antinutrients (Tannins, saponin, cyanogenic glycoside and trypsin inhibitor). Boiling of the raw seeds using slake lime, lye and water revealed a significant reduction in the levels of tannins as compared to the soaking. Informations on effect of processing on raw seeds of *Sesbania sesban* are relatively scarce. However, in comparison with other legumes, Mamiro *et al*. (2017) affirmed that the processing of different varieties of beans (leguminous plant) significantly reduces the level of tannins. The composition of tannin revealed in the raw seeds of *Sesbania sesban* as observed in this study is comparable to the composition revealed for *Sesbania bispinosa* (Pugalenthi *et al*., (2004). Tannin is traditionally classified as an antinutritional factor for animals, as it negatively affects their growth performance. Although Omnes *et al*. (2017) clearly stated that dietary tannin had no negative influence on protein level, but exogenous supplementation above 10g/kg negatively affects (decreases) protein digestibility, why above 20g/kg affect growth performance.

However, there is growing interest on the use of tannin as a growth promoting substance (Barszcz*et al*., 2018). Redondo *et al*. (2014) also highlighted that tannins are successfully being used as additives in poultry feed to control diseases and to improve animal performance. The content of saponin obtained in this study is comparable to the composition previously reported

(Arekemase *et al*., 2013). Just like tannins, the presences of saponin in feed samples especially from underutilize leguminous plant affects protein utilization. Saponin has been considered a deleterious compound, as the nature of the interaction between saponin and protein reduces protein digestibility (Abdulwaliyu *et al*., 2018). Saponin is regarded as antinutrients, as its presence in feeds impairs growth of animals and sometimes toxic to fish and othercold-blooded animals (Gemede and Ratta, 2014). Since saponin is characterized by a bitter taste, so its presence in feeds limit feed intake by domestic animals of interest. It is one of the active ingredients in *Sesbania sesban* plants responsibility for their medicinal efficacies. Study recently revealed that the presence of saponin in *sesbania* seeds may contribute in part to neuroprotection (Semwal *et al*., 2018). However, such medicinal efficacy of *Sesbania* seeds may pose effective use of the plant seeds by livestock, as they have little tolerance for some of the antinutritional factors. Although, ruminant animals have metabolic mechanism for degrading some of the factors (antinutrients). Despite the negative effects of saponin on animal nutrition, their presence (in smaller quantity is necessary, especially for the pressing need to replace antibiotics with natural secondary metabolites, sincemost of the microorganisms impairing the growth performance of livestock have developed resistance to some of the antibiotics presently.

The presence of phytic acid in animal feed is also a major concern for achieving optimal animal nutrition. It presence in feeds affects mineral bioavailability, and also reduces the phosphorus availability required for normal or optimal growth of especially monogastric animals (Singh *et al*., 2018). Bulk of thephosphorus is bound to phytic acid, and the monogastric animals lack the enzyme “phytase” capable of detaching the phosphorusthereby resulting to limited use of the phosphorus by the monogastric animals. The use of phytase as feed additive in poultry seems to overcome poor use of phytic acid by monogastric animals (Addel-hacke *et al*., 2018). Aside it

negative effect on mineral bioavailability, protein- phytate interactions are key to the negative effect of phytic acid protein/amino acid availability (Selle *et al*., 2012).

On the other hand, phytic acid is well tolerated by ruminant animals. They (ruminant) have the enzyme (phytase) capable of degrading phytic acid. As such, the bulk of the phosphorus in phytic acid may contribute largely to the dietary requirement of phosphorus by ruminant animals.

The composition of phytic acid revealed in the raw seed of *Sesbania sesban,* as observed in this study is lower than the compositions (2.05, 1.54 and 1.91g/mg) reported for *Sesbania aculeate, Sesbania rostata* and *Sesbania cannabina* (Siddhuraju *et al*., 2002). The difference in the phytic acid composition reported in the study as compared to the present study could be attributed to specie difference. The raw seeds subjected to various treatments (boiling with BL, BC, BW and soaking with SC, SL, SW) significantly reduces the phytic acid composition in the raw seeds of *Sesbania sesban* contrary to our observation, studies revealed that soaking and boiling had no effect (reduction) on phytic acid of *Sesbania* species (Siddhuraju *et al*., 2002; Duodu *et al*., 1999). Also, the phytic acid composition revealed in this study (irrespective of the treatments) were lower than the compositions (2.35 and 2.37g/100g) revealed in the seeds of *Sesbania sesban* (Hossain and Becker, 2001). The differences may be attributed to the stage of the plant seeds harvested and/or geographical zone. In comparism to the phytic acid content of soybean, the composition of phytic acid as revealed in this study were lower than the compositions (2.89 and 2.52g/100g) and 354, 113.90, 178.90 and 97.6192g/100g) revealed for raw, cooked, toasted and roasted seeds of soybean (Kumari *et al*., 2014; Ari *et al*., 2012).

Oxalate also exerts effects against optimal utilization of nutrients in feeds and foods. Observation in this study revealed that the raw seeds of *Sesbania sesban* contain oxalate,

although not in ample amount. Plant seeds high in antinutrients like oxalate are not usually recommended as a source of livestock feeds, except if properly processed. Despite the oxalate composition present in the raw seeds is not too high, processing methods employed in this study further reduces the oxalate composition in the raw seed of this plant. Observation in this study corroborate with previous finding that boiling of oxalate rich plant material promotes the loss of soluble oxalate (Poeydomenge and Savage, 2007). Soaking of plant material has also demonstrated to induce significant reduction of oxalate (Kumoro *et al*., 2014) and this conform withpresent observation as observed in this study. The presence of calcium in slake lime, and also monovalent cations (Na^{+}, K^{+}) and divalent cations (Ca^{2+}, Mg^{2+}) present in lye may be responsible for significant reduction of oxalate. Kumoro *et al*. (2014) revealed that sodium ion promotes decomposition of calcium oxalate into oxalate ion and may also increase the solubility of calcium oxalate. The presence of oxalate in feeds may disrupt optimal nutrition for livestock. However, the ruminant is less sensitive to oxalate toxicity than the monogastric animal as the former possesses oxalate degrading bacteria in the rumen (Rahman *et al*., 2012). Oxalate binds minerals thereby making these (minerals) unavailable. It (oxalate) exists in feeds and foods in two forms as soluble and insoluble oxalate. So, the nature of oxalate present in feeds or foods determines its antinutrients feasibility. Soluble oxalate binds to monovalent cations, while the insoluble oxalate binds to divalent cations like calcium, magnesium, iron etc (Rahman *et al*., 2012).

Many utilized and underutilized plants contain toxic substances that can pose health risk to humans and animals. Cyanogenic glycosides are toxic plant secondary metabolites which may affect normal cellular processes when ingested, especially if consumed in large quantity. It becomes toxic when ingested, as it releases cyanide upon hydrolysis.

Observation in this study revealed the presence of cyanogenic glycoside which is contrary to previous report that *Sesbania leptocarpa* absence of cyanogenic glycoside (Osman *et al*., 2015). The difference may lie on the specie of *Sesbania*. Also, this study revealed that raw seed of *Sesbania sesban* contain reasonable composition of cyanogenic glycosides. Although the composition is lower than the value 29.16mg/100g reported for corn seed (Onojah and Odin, 2015) often used as energy feed source for lovestock. However, the processing methods employed in this study significantly reduce cyanide contents. In the raw seeds of *Sesbania sesban*, although absolute reduction was not achieved. Suggesting that both soaking (SW, SL, SC) and boiling (BW, BL BC) are not very effective for total removal of cyanogenic glycosides.

Montagnac *et al*. (2009) also revealed that boiling was not very effective for the removal of cyanide, and this was attributed to the fact that high temperature denatures heat libile linamirase, an enzyme responsible for hydrolyzing linamarin to cyanohydrin (metabolite of linamarin). As such, linamarin, a type of cyanide cannot be hydrolysed. The target of processing plant containing antunutritional factors is to achieve total removal of the antinutrients present or a significant reduction to tolerable levels that would have no effect on the livestock. Aside aforementioned antinutrients:

The presence of trypsin inhibitor in the raw seed of this plant may also affect overall growth performance of animals. Findings from this study revealed that the trypsin inhibitor present in the raw seed of this plant can be further reduced via different treatment. Among the processing methods employed in this study, boiling (BW, BC, BL) has significant($p<0.05$) effect (reduction) on the trypsin inhibitor contents as compared to soaking (SW, SC, SL). Although soaking also promotes significant ($p<0.05$) reduction in trypsin inhibitor reduction, as evident in the significant ($p<0.05$) differences between the contents observed in the raw seeds and the contents

of the soaked samples. Observation in this study agrees with previous report that soaking of especially leguminous plant seeds promotes significant reduction of trypsin inhibitor (Adeleke *et al*., 2017).

Aside energy and protein, minerals also contribute to optimal growth performance and reproduction of animals. Deficiencies of certain mineral element may induce reproductive disorder, as they are important in health and reproduction of livestock (Balamurugan *et al*., 2017). Some mineral elements are required as co-factors for normal and optimal metabolism of macromolecules (carbohydrate, protein etc). Among the mineral elements analysed in this study, sodium (Na) ranked the highest in composition, this implies that the seeds of *Sesbania sesban*, either processed or not are a good source of Na. Observation in this study suggests that seeds of this plant could be used to augment sodium contents in sodium deficient feeds. Na is the main cation of the extracellular fluid and in alliance with chloride and bicarbonates, sodium regulates the body acid-base balance also involved in the intestinal absorption of glucose, amino acids etc (Abdulwaliyu *et al*., 2018). Just like humans, the mineral requirement of mineral elements by animals depends on certain factors like the age, stage of pregnancy, stage of lactation (Balamurugan *et al*., 2017), as well as the physiological state and disease conditions of the animals. The significant increase of Na in BW and BL is an indication that boiling (BW) improves sodium bioavailability in the seeds of *Sesbania sesban*.

The content of sodium as observed in this study suggest that the use of *Sesbania sesban* may contribute in part to the dietary requirement of Na by livestock, although the amount required may differ from one animal to another.

The potassium (K) composition obtained in the study, soaked samples and RS were higher than the composition (104ppm) revealed in the raw seeds of *Sesbania bispinosa* (Parab and Vaidya, 2016). K is the most abundant intracellular cation, necessary for optimal metabolic functions (Ahmad *et al*., 2008, especially in processes vital to the body homeostasis like osmotic pressure regulation, acid-base equilibrium etc (Oliveira *et al*., 2005). This study also revealed that boiling (BW, BL, BC) has negative effect (reduction) on the k composition, as evident in the significant differences revealed in the soaked and raw samples, as compared to the boiled samples. Implying boiling the seeds of this plant reduces the potassium composition in the seeds. Similar to this observation, study revealed that boiling causes a significant K reduction (Bethke and Jansky, 2008).

Magnesium (Mg) is an energy requiring mineral element that is necessary for many metabolic processes, such as hormone secretion, neuromuscular excitability, protein synthesis, intermediate metabolism etc (Kharb *et al*., 2018). The metabolic requirement of Mg in humans is similar to its requirement by livestock. The purpose of exploiting underutilized plant is to make available plant seeds that could be used to conveniently replace the conventional plant seeds like maize, soybean etc. In comparison to soybean, the Mg composition obtained in this study was lower than the composition revealed soybean (Etiosa *et al*., 2017). Study also revealed higher Mg composition than the composition revealed in this study (Batal *et al*., 2010).

Calcium (Ca) is also necessary for optimal health performance of livestock. Ca is key to muscle contraction, strong bone formation, fluid balance within cells etc. (Pravina *et al*., 2013). This study revealed that boiling (BL, BW, BC) and soaking (SW, SL, SC) have little effect on the Ca level. The composition of calcium observed in this study was low, although the optimal level required for growth and bone mineralization still remains a matter of debate, especially in

poultry industry (Hamdi *et al*., 2015). The low calcium level observed in this study affirms that the Ca level in feedstuffs is usually low (Li *et al*., 2017), and presence of nutrients inhibitors (phytate, oxalate etc) may further deprive its calcium bioavailability.

Some trace elements (Iron (Fe), Zinc (Zn), Manganese (Mn) and copper (Cu) were also analysed in this study. These elements, though required in minute quantity (Bruns, 2017) are also necessary for ensuring optimal health and immunity in humans and animals. They (trace minerals) contributes to growth, production and reproduction in humans and animals (Yatoo *et al*., 2013). Since the trace minerals are required in minute quantity, usually < 100mg/kg dry matter (Yatoo *et al*., 2013), the composition obtained in this study may in part contributes to dietary trace mineral requirement by livestock. The contents of Fe revealed in the raw seeds of this plant is comparable to the contents (32.50mg/100g) previously reported in the raw seeds of this plant (Arekemase *et al*., 2013). The contents of Fe obtained in this study were higher than the composition (78.56µg/g) revealed in soybean (Dan *et al*., 2017). Also, the composition of Fe revealed in the RS, SL, SW were higher than the contents (9.34-13.30mg/100g) revealed in genus Muccung, (Tresina and Mohan, 2013) which hasalso been proposed to be used as an alternative source of livestock feeds.

Iron (Fe) is important to animals, as it is important to humans since ancient times; the importance of Fe has been recognized in health and diseases (Abbaspour *et al*., 2014). Aside its role in heme synthesis (Chung *et al*., 2012). Fe is also an important co-factor in some enzyme catalyzed reaction (Moos *et al*., 2018). Zn is also involved in the metabolic processes that affect (positively) the well being of an organism (Sloup *et al*., 2017). It is essentially involved in many biochemical processes and its deficiency affects the activities of Zn dependent enzyme like superoxide dismutase (SOD) (Sun *et al*., 2011). Observation in this study revealed higher Zn

contents in RS, as compared to the contents observed in the processed samples. This implies that the processing methods (boiling and soaking) does not favour Zn bioavailability, suggesting the need for exogenous addition of Zn to the processed seeds, especially if the content in the seeds failed to meet dietary Zn requirement. Just like the need for Zn,Mn and Cu are also key to optimal growth and reproduction of livestock.

On a general note, the mineral compositions revealed in the study may not satisfy the macro and trace mineral requirement by livestock. Although, it is often difficult to access plant-based food that may provide the entire mineral requirement by humans and animals.

Among all the vitamins analyzed in this study, vitamin B_1 ranked higher than the other vitamins in the seed of *Sesbania sesban,* while vitamin B_9 ranked the least. This implying that this seeds plant is a poor source of vitamin B_9. The plant seed also revealed a lower content of vitamin C, as such the seeds of *Sesbania sesban* as a feed source cannot provide the dietary requirement for Vitamin C. Suggesting the need for exogenous supplement of these vitamins (Vitamin B9 and C). This is necessary especially further reduction revealed in the processed samples. Processing of the raw seeds of *Sesbania sesban* reduces the water-soluble vitamins, as revealed in this study. The target of processing most of the underutilized plant as a source of feeds for livestock is to further reduce the antinutritional factors thereby, enhancing their nutrients composition. As evident in this study, it was revealed that soaking (SL, SW, SC) and boiling (BL, BC, BW) influences (reduces) water soluble vitamins.

Vitamins are complex organic compound, present in food and feedstuffs in traces and also needed in small quantity for normal metabolism, growth, health and reproduction (Magbool *et al*., 2017) in both human and animals. The composition of vitamin B_1, B_2, B_6, C and vit E may

contribute in part to the dietary requirement by livestock. So using the seeds of *Sesbania sesban* as source of livestock feeds requires more or total supplement of Vit B_3, Vit B_9 and Vit C as their contents were lower than the aforementioned vitamins in the seed of *Sesbania sesban*. The ascorbic acid level obtained in this study was lower than the content revealed in soybean (Sharma *et al*., 2013). However, the contents observed in this study is comparable to the contents revealed in matured soybean, as it was clearly revealed that the ascorbic acid content of mature soybean is negligible (Lokuruka *et al*., 2010).

The high level of *invitro* protein digestibility observed in the processed samples suggests that processing of the raw seed of *Sesbania sesban* may in part contribute to the protein quality of the plant seeds. As protein quality is determined by the digestibility of the protein, composition of the amino acids and their bioavailability (Gilani *et al*., 2005). The high levels of antinutrients in the raw sample may be a contributing factor for the lower *in vitro* protein digestibility revealed in raw sample as compared to the processed samples. Study affirmed that the presence of antinutritive factors found in some raw legumes influences protein digestibility (Gilani *et al*., 2012). Possible interaction between protein and non-protein component as observed in this study may also contribute to poor protein utilization (Usman *et al*., 2018). Lower level of *in vitro* protein digestibility revealed in the raw sample seeds may result in poor utilization of protein in *Sesbania sesban* by any targeted animals.

The *in vitro* protein digestibility revealed in the processed samples of *Sesbania sesban* were higher than the value (65.82%) reported for *Sesbania bispinosa* (Pugalenthi *et al*., 2004). The significant increase in the *in vitro* protein digestibility of processed seeds of *Sesbania sesban* would significantly improve protein utilization of the processed seeds. The percentage in vitro protein digestibility revealed for BW and BC are comparable to the composition (85.89%)

revealed for soybean meal fermented with Aspergillus, while the seeds processed with lye (BL) revealed higher percentage of *invitro* protein digestibility, as compare to the percentage *in vitro* protein digestibility reported for soybean meal (87.50%), soybean meal fermented with Aspergillus (85.89) and soybean protein concentrate (87.63) (Chen *et al*., 2010) Suggesting that the seeds of *Sesbania sesban* if properly processed may be employed as an alternative source of feed protein to soybean meal, a widely and most acceptable source of protein for livestock.

CHAPTER SIX

6.0 CONCLUSION AND RECOMMENDATIONS

6.1 Conclusion

Based on the findings from this study, it is concluded that:

i. Proximate compositions showed sample boiled with lye (BL) had higher levels of crude protein (26.14 ± 0.06), carbohydrate (52.44 ± 0.40) and food energy value (371.38) compared to other processed samples.

ii. All the processing methods employed significantly ($p<0.05$) reduced the antinutrient contents of *Sesbania sesban* present in the seed with boiling with lye(BL) having the highest percentage (76.52%) reduction in tannins, 58.14% in saponin, 50.47% in oxalate, 51.29% in cyanogenic glycosides and 68.00% in trypsin inhibitor.

iii. Processing methods caused significant ($p<0.05$) reduction in beta-carotene and water-soluble vitamins except for vitamin B_9, while vitamin E was significantly improved in s a sample soaked with lye (SL).

iv. Processing as demonstrated in this study improved*invitro* protein digestibility of *Sesbania sesban* seeds in which that of sample boiled with lye (BL) had the highest digestibility therefore, the seed of *Sesbania sesban* processed with boiling with lye(BL) maybe used for animal feed.

6.2 Recommendations

i. Based on these findings samples boiled with lye(BL) significantly reduced the tannins, saponin, oxalate, cyanogenic glycosides and trypsin inhibitor present in the seeds than other processing methods. To adequately reduce the antinutritional factors in the seed of

Sesbania sesban, it is therefore, recommended that the seed may be processed with boiling with lye(BL).

ii. Based on these findings, boiling with lye (BL) of the seeds of *Sesbania sesban* significantly increased the protein contents and improves the *in vitro* protein digestibility. Therefore, it is recommended for processing of *Sesbaania sesban*for livestock feeds.

iii. Further studies should focus on the effects of other processing methods such as fermentation, roasting, sprouting on the levels of nutrients, antinutrients and *in vitro* protein digestibility.

6.3 Contribution to Knowledge

i. Boiling with lye (BL), boiling with calcium hydroxide(BC), boiling with water (BW) and soaking with water (SW), soaking with lye(SL), soaking with calcium hydroxide(SC) of *Sesbania sesban* seeds can effectively reduce some of the antinutritional factor present.

ii. This study revealed for the first time that BL, BC, SL, SC can be employed to favorably reduce the antinutrients present in the seeds of the plant.

REFERENCES

Abbaspour N, Hurrell R and Kelishade R (2014). Review on iron and its importance for human health. *J. Res. Med. Sci, 19 (2): 164 – 174.*

Abdel- hack M, Alagawany M, Arif M, Emam M, Saeed M, Arain MA (2018). The uses of microbial phytase as a feed additive in poultry nutrition- A Review. *Ann. Anim. Sci, 18(3):639-658.*

Abdulwaliyu I, Idowu OO, Arekemase S.O, Batari ML, Nkeonye OL, Odjobo BO (2018). The nutritional potential of senna alata seed. *Int. Fd. Res. J, 25 (6): 2628-2633.*

Adeleke OR, Adiamo OQ, Fawale OS, Olamiti G (2017). Effect of soaking and boiling an antinutritional factors, oligosaccharide contents and protein digestibility of newly developed Bambara groundnut cultivars. *Turkish. J. Agri. Fd. Sci. Tech, 5(9): 1006 – 1014.*

Agbaire, P.O (2011). Nutritional and Anti-nutritional Levels of Some Local Vegetables(Vernomia anydalira, Manihot esculenta, Teiferia occidentalis, Talinum triangulare, Amaranthus spinosus) from Delta State, *Nigeria. J. Appl. Sci. Environ. Manage, 15 (4) 625 – 628.*

Ahmad T, Khalid T, Mushtaq T, Mizra MA, Nadeem A, Babar ME and Ahmad G (2008). Effect of potassium chloride supplementation in drinking water on broiler performance under heat stress conditions. Poultry Sci, 87: 1276-1280. Doi: 10. 3382/ps. 2007-00299.

Ahmed A, Howlader SI., Dey SK., Arpona Hira A., and Hossain H (2013).Phytochemical screening, antimicrobial and cytotoxic activity of different fractions of *Sesbania sesban* bark.*Internat. J. Med Sci. Pharm.3(1):6 – 12.*

Akande K.E., Doma U.D., Agu H.O., and Adamu H.M (2010). Major Antinutrients Found in Plant Protein Sources: Their Effect on Nutrition. *Pakistan J. Nutri, 9 (8): 827-832.*

Akao Y, Yoshihito N, Munekazu L, Yshinori N (2008). Anticancer Effects of Xanthones from Pericarps of Mangosteen.*Int J Mol Sci,* , 9, 355-370.

Akkasaeng R, Gutteridge RC and Wanapat M (1989) Evaluation of trees and shrubs for forage and fuelwood in Northeast Thailand. *Int. Tree Crops J*. 5:209-220.

Alagesaboopathi C (2012). Ethnobotanical studies on useful plants of Sirumalai Hills of Eastern Ghats, Dindigul District of Tamilnadu, Southern India. *Int. J. Biosci. 2(2):77-84.*

Al-Dawah NKJ., Al-safi SM., Aboktifa MA., AL-Zeiny SSM., AL- Shimmary BAA., Albayati MTN and Muhammad NA (2014). Comparative of Phytochemical and Antimicrobial of*Sesbania grandiflora* Leaves Extract. *Med J. Babylon, 11(3).*

Ali Z., Ashraf M., Al – qurainy F., Khan Sand Akram NA (2015). Appraising Drought Tolerance in local Accession of Sesbania (Sesban sesban (L.) Merril) using Biomass production relative membrane permeability and photosynthetic capacity as selection Criteria. *Pak. J. Bot,47(3):845-850.*

Anderson DMW. (1989). *Sesbania* species as sources of gum exudates and seed galactomannan gums. In: Macklin B, Evans DO, editors. *Perennial J species in agroforestry systems.* Proceedings of an International Workshop at ICRAF.Nairobi, Kenya, NFTA Special Publication 90-01. p99-194.

Anjana, S.U., Muhammed, I. and Abrol.Y.P. (2007). Are nitrate concentrations in leafy vegetables within safe limits? *Current Science, 92(3): 355-360.*

Anuonye J. C., Jigam A. A and Ndaceko G. M (2012). Effects of Extrusion-Cooking on the Nutrient and Anti-Nutrient Composition of Pigeon Pea and Unripe Plantain Blends. *Journal of Applied Pharmaceutical Science 02 (05); 158-162.*

AOAC (1984). Official Methods of Analysis. Association of Official Analytical Chemists. 14th Edition, AOAC, Arlington.

AOAC (1990). Official Methods of Analysis (15th ed.). Association of Official Analytical Chemists. Washington D.C.

AOAC (1992). Total, Soluble, and Insoluble dietary fibre in foods. Enzymatic gravimetric method 15thedition. Official Method of Analysis of the Association of Official Analytical Chemists, Arlington, Virginia.

AOAC (2003). Official Methods of Analysis of Analytical of chemists, 17th edn. Association of Official analytical chemists, Arlington, Virginia.

Arekemase S.O., Abdulwaliyu I and Musa M (2014). Some organic contents of Sesbania sesban seed oil. *Int. J. Fd Nutr, saf, 5(3): 115 – 122.*

Arekemase S.O., Abdulwaliyu I., Dakare M.A., Bala S., Ibraheem A.S and Nkeonye O.L (2013). Quantitative evaluation of the nutritional constituents of Sesbania sesban Seeds and Pods.*International Journal of Modern Plant and Animal Sciences* 1 (1) : 16-27,

Ari MM, Ayawale BA, Adama TZ, Olatunji EA (2012). Evaluation of the chemical composition and antinutritional factors (ANFs) levels of different thermally processed soybeans. *Asian J. Agric. Res, 6(2): 91 – 98.*

Ayisi ND, Adu KJ (2016). Challenges and future Prospect for Broiler Meat Consumption in Ghana. *Imperial Journal of Interdisciplinary Research, 2(8):648-654.*

Aziza AH and Zainan H (1997). Effect ofprocessing on dietary fiber contents of selected legumes and cereals. *Mal. J. Nutr, 3: 131 – 136*

Bairszcz M, Taciak M, Tusino A, Skomial J (2018). Effect of dietary level of tannic acid and protein as Internal organ weights and biochemical blood paremeters of rats. PLoSONE 13(1):E0190769. https://doi.org/10.1371/ Journal pone. 0190769

Balamurugan B, Ramamoorthy M, Mandal RSK, Rerthana J, Gopalakrishnan G, Karya KM (2017). Mineral an important nutrient for efficient reproductive health in diary cattle. *Int. J. Sci. Envi, 6 (1): 694-701.*

Batal AB, Dale NM, and Saha UK (2010). Mineral composition of corn and soyabean meal. *J. Appl poultry. Res, 19: 361-364.*

Bethke PC and Jansky SH (2008). The effect of boiling and leaching on the content of potassium and other minerals in potatoes.*J. Fd. Sci, 75 (5) : 80-85.*

Bhathena, S.J. and Velasquez, M.T.(2002). Beneficial role of dietary phytoestrogens in obesity and diabetes. *American Journal of Clinical Nutrition*, 76: 1191–1201.

Bruns HA (2017).Soybean micronutrient content in irrigated plants grown in the mid south. Communication in soil Sci. Plt. Anal, 48 (7) : 808-817.

Cetingul I.S and Yardimci M (2008).The importance of fats in farm animal nutrition. *Kocatepe Vet J, 1: 77-81.*

Chatterjee A. and Pakrashi S., (1992). The treatise of Indian medicinal plants. Deep publication New Delhi, 2:121.

Chen CC, Shih YC, Chiou PWS, Yu B (2010).Evaluating nutritional quality of single stage and two stage fermentedsoybean meal. *Asian – Aust. J. Anim. Sci, 23(5):598-*

Cheung P.C.K, Leung A.Y.H, Ang P.O., (1998). Comparison of supercritical carbon dioxide and Soxhlet extraction of lipids from a brown seaweed, Sargassum hemiphyllum (Turn.) C. Ag. *J. Agric. Food Chem., 1998, 46, 4228-4232.*

Choudhary M., Aggarwal N., Choudhary N., Gupta P and Budhwaar V (2014). Effect of aqueous and alcoholic extract of *Sesbania sesban* (Linn) Merr.root on glycemic control in streptozotocin-induced diabetic mice. *Drug Develop. Therap.*5(2): 115-122.

Chung J, Chen C, Paw BH (2012). Heme metabolic and Erythropoiesis.*Curr.Opin.Hematol:* 19 (3) :156 – 162.

Dagefu T., Wolde-meskel E and Frostegard A (2011). Multilocus sequence analysis reveal several unnamed mesorhi zobium genospecies modulating Acacia species and Sesbania sesban trees in southern regions of Ethopia. *Sys. Appl. Microbial,* 34: 216-226.

Dakare MA., DanladiAA., Abel SA and SundayEA (2012). Effects of processing techniques on the nutritional and antinutritional contents of mango (*Mangifera indica*) seed kernel. *World Journal of Young Researchers,*2 (3): 55-59.

Dan SK, Banerjee G, Nandi A, Ray AK (2017). Nutritional evaluation of soybean meal after fermentation with two dish gut bacterial strains, Bacillus cereus LRF5 and Staphylococcus caprae ccf2 in formulated diets for Labeo rohita fingerlings. *J. Fisheries, 5 (1): 445 – 454.*

Dan TH and Brix H (2009).Growth responses of the perennial legume *Sesbania sesban* to NH4 and NO3 nutrition and effects on root nodulation.Aquat. Bot. 91:238-244. doi:10.1016/j.aquabot.2009.07.004

Dan TH, Quang LN, Chiem NH, Hans B (2011). Treatment of high-strength wastewater in tropical constructed wetlands planted with *Sesbania sesban:* Horizontal subsurface flow versus vertical downflow. Ecol. Eng. 37:711-720.doi:10.1016/j.ecoleng.2010.07.030

Dande P.R., Talekar V.S and Chakraborthy G.S (2010).Evaluation of Crude Saponins Extract from Leaves of *Sesbania sesban* (L.)Merr.for Topical Anti-inflammatory Activity.*Int. J. Res. Pharm. Sci. Vol-1, Issue*-3, 296-299.

Danhassan MS, Salihu A, Inuwa HM (2018).Effect of boiling on protein, mineral, dietary fibre and antinutrient compositions of Nymphaea lotus (Linn) seeds. *Journal of food composition and analysis 67: 184 – 190.*

Das N., Chandran P and Chakraborty S (2011). Potent spermicidal effect of oleanolic acid 3-beta-D-glucuronide, an active principle isolated from the plant *Sesbania sesban* Merrill. *Contracept*. 83:167-175.

Day RA and Underwood AL (1986). Quantitative analysis. 5th ed. Prentice – Hall Publication, New Jersey, USA, p701.

Debela E., Tolera A., Eik LO and Salte R (2011). Nutritive Value of Morphological Fractions of *Sesbania sesban* and *Desmodium intortum.Tropical and Subtropical Agroecosystems*, 14: 793-805793.

Degefu T., Wolde-meskel E and Frostegard A (2011). Multilocus sequence analyses reveal several unnamed Mesorhizobium genospecies nodulating Acacia species and *Sesbania sesban* trees in Southern regions of Ethiopia. Syst. *Appl. Microbiol*. 34:216-226. doi:10.1016/j.syapm.2010.09.006.

Deol J.K and Bains K (2010). Effect of household cooking methods on nutritional and anti nutritional factors in green cowpea (Vigna unguiculata) pods. *J Food Sci Technol, 47(5):579–581.*

Desaeger J and Rao MR (2001). Effect of field establishment methods on root-knot nematode (Meloidogyne spp.) infection and growth of *Sesbania sesban*in western Kenya. *Crop Protect*. 20:31-41.

Dey R.A. and Underwood A.L. (1986).Quantitative analysis. 5th ed. Prentice – Hall Publication, New Jersey, USA, p701.

DinendraNS,Azad-ud-doulaProdhanAKM(2001).Anatomy of *Sesbania sesban.Indian J. Agric. Res.35(4):211-218.*

Duke(1981). Handbook of legumes of world economic importance.Plenum Press, New York, p170-84.

Duodu KG, Minnaqr A, Taylor JRN (1999).Effect of cooking and irradiation as the labile vitamins and antinutrients content of a traditional Africa Sorghum porridge and spinach relish.Food Chemistry, 66; 21 – 27.nd their morphological factors. *J. Fd. Chem, 73 : 421 – 431*.

Edet A., Eseyin O and Aniebiet E (2015).Anti-nutrients composition and mineral analysis of allium cepa (onion) bulbs.*Afri. J. Pharm. Pharmacol*,9(13), pp. 456-459. DOI: 10.5897/AJPP2015.4300.

Eltayeb M.M., Hassn, A.B., Sulieman, M.A., Babiker, E.E. (2007). Effect of processing followed by fermentation on antinutritional factors content of pearl millet (Pennisetum glaucum L.) cultivars. *Pakistan Journal of Nutrition*, 6 (5), 463-467.

Embaby HE (2011).Effect of Heat Treatments on Certain Antinutrients and in vitro Protein Digestibility of Peanut and Sesame Seeds.*Food Sci. Technol*. Res., 17 (1), 31 – 38.

Ertop H.M and Bektaş, M (2018).Enhancement of Bioavailable Micronutrients and Reduction of Antinutrients in Foods with some processes. *Food and Health*,4(3),159-165. DOI: 10.3153/FH18016.

Etiosa OR, Chika NB and Benedicta A (2017).Mineral and proximate composition of soybean. *Asian J. Phys. Chem. Sci*, 4 (3): 1 - 6.

Etiosa OR, Chika NB, Benedicta A (2007).Mineral and proximate composition of soybean. *Asian Journal of physical and chemical sciences*, 4 (3): 1- 6.

Evans D.O, (1994). Sesbania sesban: widely distributed multipurpose NFT. NFT Highlights, No. 94-06.

FAO (2007). Factsheet Sesbania sesban. Http://www.tropicalforage.info/key/forages/media/html.

Fekadu H (2014). Nutritional composition, antinutritional factors and effect of boiling on nutritional composition of Anchote (*Cocciniaabyssinica)*tubers.*Food Science and Quality Management,* 26, 25-38.

Gemede HF and Ratta N (2014). Antinutritional factors in plant foods: potential health benefits and adverse effects. *Int. J. Nutr. Fd. Sci*, 3(4): 284-289.

Ghazalah, A. A.; El-Shahat, A. A.; El-Yamny, A. T., (1998). Evaluation of some tropical forages for nutrition and meat production of rabbits. *Egyptian J. Rabbit Sci.,* 8 (2): 127-139.

Gilani GS, Cockell KA, Sepehr E (2005).Effect of antinutritional factors on protein digestibility and amino acids availability in foods. *J. AOAC International*, 88 (3): 967 - 987

Gilani GS, Xiao CW, Cockell KA (2012).Impact of antinutritional factors in food proteins on the digestibility of protein and the bioavailability of amino acid and protein quality. *British J. Nutr*, 108, 315-332

Gohl, B (1981). Tropical Feeds. FAO Animal Production and Health Series No. 12. FAO Rome, pp. 198-199.

Gomase P., Anjum S., Shakil S and Shahnavaj K.M (2012). *Sesbania sesban* Linn: A review on its Ethnobotany, phytochemical and pharmacological profile. *Asian J. Biomed. Pharmaceut Sci* 2(12): 11 – 14.

Gopalakrishna, T. and Joshi-Saha, A. (2007).Agromorphological and molecular variability in the genus Sesbania. *Genetic Resources and Crop Evolution* 54: 1727-1736.

Goswami S., Mishra K.N., Singh R.P., Singh P and Singh P (2016).*Sesbania sesban*, A Plant with Diverse Therapeutic Benefits: An Overview. *J. Pharmaceut Res & Edu.* 1(1), 2016, 111-121.

Gunashree B.S., Kumar R.S., Roobini R., and Venkateswaran R (2014). Nutrients and antinutrients of ragi and wheat as influenced by traditional processes.*Int.J. Curr.Microbiol.App.Sci, 3(7) 720-736.*

Gupta AM, Su SW, Chen ZS (2011). Heavy-Metal Bioavailability and Chelate Mobilization Efficiency in an Assisted Phytoextraction Process by *Sesbania sesban*(L.)Merr., Commun. *Soil Sci. Plant Anal. 42(2)*:231-245. doi:10.1080/00103624.2011.535073

Gutteridge RC and Shelton HM (1991). Evaluation of *Sesbania sesban* - a new forage shrub species for tropical and subtropical Australia. Final Technical Report, *Meat Research Corporation*, Canberra. 12 pp.

Hail M.B, and Eastridge ML (2014). Invited review: Carbohydrate and fat: Considerations for energy and more. *The professional animal scientist*, 30:140-149.

Hamdi M, Sola- Oriol D, Davin R, Perez J.F (2015). Calcium sources and their interaction with the different levels of non-phytate phosphorus affects performance and bone mineralization in broiler chickens. *Poult. Sci,* 94:2136-2143.

Hang B.P.T, Phuong T.T.B, and Preston T.R (2011). Water hyacinth (Eichornia crassipes) : an invasive weed or a potential feed for goat?. *Livestock Research for Rural Development 23(7), 152*.

Harbone J.B (1973). Phytochemical methods Chapman and Hall, New York.

Heering J.H (1995). Botanical and agronomic evaluation of a collectionof Sesbania sesban and related perennial species. Doctoral thesis,Wageningen Agricultural University, Wageningen, The Netherlands, p.127.

Henchion M, Hayes M, Mullen AM, Fenelon M, Tiwari B (2017). Future protein supply and demand: Strategies and factors influencing a sustainable equilibrium. *Foods* 6, 53.

Hossain A., and Chaudhary S., (2007). Antimicrobial activity of methanolic extract of *Sesbania sesban*. *Journal of Pharmaceutical science, 6(1): 61-63.*

Hossain MA, and Becker K (2001). Nutritive value and antinutritional factors in different varieties of Sesbania seedoybeans and their morphological factors. *J. Fd. Chem, 73: 421 – 431.*

Hossain MA, Focken U, and Becker (2002). Nutritional evaluation of dhaincha (Sesbania aculeate) seeds as dietary protein source for tilapia Oreochromis niloticus. *Aquaculture Res, 33: 653-662.*

Ibrahim A.M., (1992). Antihelmintic activity of some Sudanese plants. *Phytotherapy Research, 6(3):155-157.*

Idoko A.S., Oladiji A.T., Yakubu M.T., and Aska A.S (2014). Effect of Heat Treatment on Nutrient and Anti-nutrient Components of Melon (*Citrullus colocynthis*) Husks.*Res. J. Chem. Sci, Vol. 4(4), 28-32.*

Ihemeje, A., Nwanekezi, E.C., Odimegwu, E.N. & Ekwe, C.C (2018).Effect of Processing Methods of Toasting, Soaking, Boiling, Sprouting on Dietary Fibre and Antinutrients Contents of African Yam Bean and Red Kidney Bean Flour.*European Journal of Food Science and Technology Vol.6, No.1, pp.40-48.*

Indieka SA and Odee DW (2005). Nodulation and growth response of *Sesbaniasesban*(L.) Merr. to increasing nitrogen (ammonium) supply under glasshouse conditions. *Afr. J. Biotechnol*. 4:57-60.

Ishola DT, Olabiran TE, Olajide M.B, Ishola OT, Alejo AO, Awonyemi IO, Ajayi OB (2018). Comparative evaluation of the proximate composition of raw and fermented seeds of Zarmarkee, Sesbania Spp. *Journal of Agriculture and veterinary Science, 11 (6): 20 – 25.*

Ismail B.P (2017). Ash content Determination in: Food Analysis Laboratory Manual. Food Science Text Series. Cham-DOI (https: 11 doi.org/10.1007/978-3-319-44127-6-11

Jamnadass R., Hanson J., Poole J., Hanotte O., Simons TJ and Dawson IK (2005). High differentiation among populations of the woody legume *Sesbania sesban* in sub-Saharan Africa: implications for conservation a nd cultivation during germplasm introduction into agroforestry systems. *Forest Ecology and Management,* 210(1/3):225-238. http://www.sciencedirect.com/science/journal/03781127.

Jorge E.M., Wolfgang, H.P., and Peter, B. (2008). Biofortified crops to alleviate micronutrient malnutrition. *Current Opin-ion Plant Biology,* 11, 166-170.

Joshua ZP, Timothy AG, Suleiman MM (2012).Effect of cooking time on the vitamin C, dietary fiber and mineral composition of some local vegetables. Sci. *World J, 7(1):29-30*

Kadam V.B., Mali M.V., Medhane V.J., Gaikwad V.B (2013). Biochemical evaluation of three medicinal Taxa of genus Sesbania in maharashtra. *J. Drug Deli. Therape, 3(5): 41-43.*

Kaitho, R. J.; Tegegne, A.; Umunna, N. N.; Nsahlai, I. V.; Tamminga, S.; Bruchem, J. van; Arts, J. M., (1998). Effect of *Leucaena* and *Sesbania* supplementation on body growth and scrotal circumference of Ethiopian highland sheep and goats fed teff straw basal diet. *Livest. Prod. Sci., 54 (2): 173-181.*

*K*aitho, R. J.; Umunna, N. N.; Nsahlai, I. V.; Tamminga, S.; Bruchem, J. van, (1998b). Nitrogen in browse species: ruminal degradability and post-ruminal digestibility measured by mobile nylon bag and *in vitro* techniques. *J. Sci. Food Agric., 76 (4): 488-498.*

Kaitho, R. J.; Umunna, N. N.; Nsahlai, I. V.; Tamminga, S.; Bruchem, J. van, (1998a). Effect of feeding graded levels of *Leucaena leucocephala*, *Leucaena pallida*, *Sesbania sesban* and *Chamaecytisus palmensis* supplements to teff straw given to Ethiopian highland sheep. *Anim. Feed Sci. Technol., 72 (3-4): 355-366*

Karbo, N.; Barnes, P.; Rudat, H., (1996). Evaluation of browse forage preferability by sheep and goats in the Northern Guinea Savannah Zone of Ghana. *Bulletin of Animal Health and Production in Africa, 44 (4): 225-230.*

Kathiresh M., Suganya PD and Saravanakumar M (2012). Bioactive compounds in Sesbania sesbanflower and its Antioxidant and Antimicrobial activity. *J. Phar. Res.*5(1):390-293.

Kathresh M., Suganya P., Saravanakumar M. Antioxidant effect of Sesbania sesban flower extracts (2011). *International Journal of pharmaceutical Sciences,3(2), 1307-1312.*

Kerr BJ., Kellner TA and Shurson GC (2015). Characteristics of lipids and their feeding value in some diets. *J. Ani.Sci. Biotech, 6: 30.*

Kharb S, Bhardwaj J, Goel K and Nanda S (2018). Nutritional Aspects of Magnesium in Fetal growth. *Acta scientific nutritional health*, 2 (12): 03 – 07.

Kokkinidou S, Peterson D, Bloch T, Bronston A (2018). The important role of carbohydrates in the flower, function and formulation of oral Nutritional supplement.*Nutrients,* 10, 742:doi:10.3390/nu/0060742.

Krishnaveni S, Balasubramanian T and Sadasivam S., (1984) Phenol Sulphuric acid Method., *Food Chemistry, 1984, 15, 229.*

Kudu Y.S., Alabi J.O., Egene SSA, Umaru M.A., (2008).Effect of four different commercial feeds on cockerel production produc.33rd Ann. Conf. NSAP.Ayetoro, Ogun state, 18th - 20th March.Pp.443-445.

Kumar U., Murithy N., Singh C., Gouri M.D., Rajeshiwari Y.B., Siddeshwara NC, Mateen A and Guruprasad R (2017). Biomass Yield and Chemical Composition of *Sesbania gradiflora and Moringa oleifera. Int. J. Sci. Environ.Tech. 6 (6):3264-3269.*

Kumari S, Krishnan V, Jolly M, Sachdev A (2014). Invivo bioavailability of essential minerals and phytase activity during soaking and germination in soybean (Glycine max L.). *Aust. J. Crop Sci, 8 (8): 1168-1174.*

Kumoro AC, Budiyati CS, Retnowati DS (2014). Calcium oxalate reduction during soaking of giant taro (Alocasia acrorrhiza (L.) Schott corn chips in sodium bicarbonate solution. *Int. Fd. Res. J., 21 (4): 1583 – 1588.*

Lewu M.N., Adebola P.O., and Afolayan A.J. (2009). Effect of cooking on the mineral and antinutrient contents of the leaves of seven accessions of Colocasia esculenta (L.)*Schott growing in South Africa.Journal of Food, Agriculture & Environment Vol.7 (3&4): 3 5 9 - 3 6 3.*

Li OY and Komarek AR (2017). Dietary fiber basics: Health, nutrition, analysis and application. *Fd. Quality and Safety,* 1, 47 – 59

Li X, Zhang D and Bryden WL (2017). Calcium and phosphorus metabolism and nutrition of poultry: are current diets formulated in excess. *Ani. Prod. Sci, 57 :2304 -2310.*

Lokuruka M (2010). Soybean nutritional properties: The good and the bad about soy foods consumption – A review. *Afr. J. Fd. Agric. Nutr. Dev, 10 (4): 2439 – 2459.*

Lucas G.M. and Markakas P., (1975). Phytic acid and other phosphorus compounds of bean (phase Olus Vulgaris). *J. Agric. Educ. Chem., 23: 13 -15.*

Luthria, D.L. and Pastor-Corrales, M.A. (2006).Phenolic acids content of fifteen dry edible bean (*Phaseolus vulgaris* L.) varieties. *Journal of Food Composition and Analysis, 19: 205–211.*

Mada S.B., A. Garba, A. Mohammed, A. Muhammad, A. Olagunju, H. A. Mohammed (2012). Effects of Boiling and Roasting on Antinutrients and Proximate Composition of Local and Some Selected Improved Varieties of Arachis hypogaea L (Groundnut). International *Journal of Food Nutrition and Safety, 1(1): 45-53.*

Makatiani E.T and Odee D.W (2007).Response of Sesbania sesban (L) Merr. To rhizobial inoculation in an N- deficient soil containing low numbers of effective indigenous rhizobial. *Agroforsyst,* 70: 211-216. Doi: 10.1007/10457 – 007-9054-9.

Mamiro P.S, Mwanri H.W, Mongi R.J., Chiviaghula T.J, Nyagaya M. and Ntwenya (2017).Effect of cooking on tannin and phytate content in different bean (Phaseolus vulgaris) varieties grown in Tanzania.*African Journal of Biotechnology 16 (20). Pp 1186-1191.*

Manaye T., Tolera A and Zewdu T (2009). Feed intake digestibility and body weight gain of sheep fed Napier grass mixed with different levels of Sesbania sesban. *Livestock Sci*, 122:24-29.

Mani R.P., Pandey A., Goswani S., Tripathi P., Kumudhavalli V., and Singh A.P (2011). Phytochemical screening and invitro evaluation of Sesbania sesban (L) Merr. *Free ridic. Antioxidant*, 3(1):66 – 69- doi: 10.530/ax. 2011.3.9.

Manjusha1, Neha Aggarwal1, Nitesh, and Pankaj Gupta (2012). Effect of petroleum ether extract of Sesbania sesban (Merr.) roots in streptozotocin (STZ) induced diabetes in mice.*Asian Pacific Journal of Tropical Biomedicine S1254-S1260.*

Maqbool MA., Aslam M., Akbar W and Iqbal Z (2017). Biological importance of vitamins for human health: A review. *J. Agric. Basic Sci, 2 (3): 50 – 58.*

Mekoya A., Oosting S.J., Fernandez-Rivera S., Tamminga S and Vander-Zijpp A.J (2009c).Effect of supplementation of Sesbania sesban to lactating ewes on milk yield and growth rate of lambs. *Livestock Sci, 121:126-131.*

Mekoya, A.; Oosting, S. J.; Fernandez-Rivera, S.; Tamminga, S.; Tegegne, A.; Zijpp, A. J. van der, (2009a). Effect of supplementation of *Sesbania sesban* on post-weaning growth performance and sexual development of Menz sheep (Ethiopia).*Livest. Sci., 121 (1): 108-116.*

Mekoya, A.; Oosting, S. J.; Fernandez-Rivera, S.; Tamminga, S.; Tegegne, A.; Zijpp, A. J. van der,(2009b). Effect of supplementation of *Sesbania sesban* on reproductive performance of sheep.*Livest. Sci., 121 (1): 117-125.*

Melaku, S.; Peters, K. J.; and Tegegne, A., 2004. Feed intake, live weight gain and reproductive performance of Menz ewes supplemented with *Lablab purpureus*, graded levels of *Leucaena pallida* 14203 and *Sesbania sesban* 1198. *Livest. Prod. Sci., 87 (2-3): 131-142.*

Mengistie Taye, (2009). Growth of Washera ram lambs fed on Napier (*Pennisetum purpureum*) and Sesbania (*Sesbania sesban*) mixture at different levels of combination. *Livest. Res. Rural Dev., 21 (12).*

Mertz E.T, Hassen, M.M., Cairs-Whittern, C., Kirleis, A.W., Tu, L., Axtell, J.D. (1984).Pepsin digestibility of proteins in sorghum and other major cereals. *Proceedings of National Academy of Sciences,* 81(1): 1-2.

Montagnac JA, Davis CR, Tanumihardj S.A (2009).Processing techniques to reduce toxicity and antinutrients of cassava for use as a staple food.Compre. Rev. *Fd. Sci. Fd safety, 8: 17 - 27*

Moos T, Skjorring T and Thompson LL (2018). Iron deficiency and iron treatment in the fetal developing brain – a pilot study introducing an experimental rat model, 15 (Suppl 1); 93: Pp 118 – 126.

Musa A and Ogbadoyi E.O (2014). Determination of Anti-nutrients and Toxic Substances of Selected Fresh Leafy Vegetables Obtained from Minna Town, Nigeria. Nigerian *J. Bas. Appl. Sci*, 22(3&4): 79-83. DOI: http://dx.doi.org/10.4314/njbas.v22i3.5.

Musa, A. (2012). Influence of plant leaf locations on the bioaccumulations of phytotoxins and nutrients in Corchorus olitorius at market maturity. *International Journal of Biology.4(3): 130-139.*

Musa, A., Oladiran, J.A., Ezenwa, M.I.S., Akanya, H.O. and Ogbadoyi, E.O. (2011).Effect of heading on some micronutrients, anti-nutrients and toxic substances in Amaranthus cruentus grown in Minna, Niger State, Nigeria.*American Journal of Food and Nutrition.1(2): 147-154.*

Mythili T and Ravindhran R (2012).Phytochemical Screening and antimicrobial activity of *Sesbania sesban.Asian J Pharm Clin Res, Vol 5, Issue 4, 2012, 179-182*

Naik NN., Tare H.L, Sherikar AK., Deore SR and Dama GY (2011). Central nervous system stimulant effect of extracts obtained from the barks of Sesbania sesban. *Int. J. Inst. Pharm. Life Sci*. 1(1):77-92.

Natesh H.N., Abbey L and Asiedu S.K (2017).An overview of nutritional and anti nutritional factors in green leafy vegetables.*Horticult Int J. 2017;1(2):58–65*.

Nayak KC, Rath SC, Giri SS, Mohanta K.N (2018). Evaluation of Dhaincha seed (Sesbania aculeate) as a non-conventional feed ingredient for Labeo rohita (Ham) fry). *International Journal of Fisheries and Aquatic studies, 6(2):272-279.*

Ndidi U.S., Ndidi C.U., Aimola I.A., Bassa O.Y., Mankilik M. and Adamu Z (2014). Effects of Processing (Boiling and Roasting) on the Nutritional and Antinutritional Properties of Bambara Groundnuts (Vigna subterranea [L.]Verdc.)from Southern Kaduna, Nigeria. *Journal of Food Processing, 9 pages, http://dx.doi.org/10.1155/2014/472129.*

Niguissie Z and Alemayehu G (2013). *Sesbania sesban (L)* Meroill: potential uses of an underutilized multipurpose tree in Ethiopia- A Review. *Afr. J. Plant Sci, 7 (10) : 468 – 475.*

Obadoni B.O., Ochuko P.O., (2011). Phytochemical studies and comparative efficacy of the crude extract of some homeostatic plants in Edo and Delta states of Nigeria. Global. *J. Pure Applied sci., 86: 203 – 208.*

Oladele P.E. and Oshodi A.A., (2007). Nutritional potential of Berlandier nettle spurge (Jatropha Catharical) seed. *Parkistan Journal of nutrition, 6(4): 345-348.*

Oliveira J.E, Albino L.F.T, Rostagno H.S, Paez LE and Carvalho DCO (2005).Dietary levels of potassium for broiler chickens.*Brazilian J. Poultr. Sci, 7 (1) : 33-37.*

Olukunle O.T (2016). Economic analysis of profitability and competitiveness of sugarcane enterprise in Nigeria. *Journal of Development and Agricultural Economics, 8(6):160-171.*

Omnes M, Goasduff JL, Delliou HL, Bayon NL, Quazuguel P, Robin JH (2017). Effects of dietary tannin on growth, feed utilization and digestibility, and Carcass composition in juvenile European seabass (Dicentrarchus labrax L.). *Aquaculture Reports,* 6 : 21-27.

Onojah PK and Odin EM (2015). Cyanogenic glycoside in food plants. *Int. J. Innov. Sci. Math, 3 (4) : 197 – 200.*

Onwuka G.I (2005). Food analysis and instrument (theory and practice).Department of food science and Technology, Micheal Opara University of Agriculture, Umudike, Abia state, Nigerialic. Naphthalic prints, Surulere, Lagos, Nigeria, 219-230.

Onyeike EO., Anyalogbu EA and Monanu MO (2015). Effect of Heat Processing On the Proximate Composition and Energy Values Of African Walnut (Plukenetiaconophora) And African Elemi (Canariumschweinfurthii) Consumed As Masticatories In Nigeria. *International journal of scientific & technology research* volume 4, issue 08.

Orwa C, A Mutua, Kindt R , Jamnadass R, and S Anthony. (2009) Agroforestree Database:a tree reference and selection guide version 4.0 (http://www.worldagroforestry.org/sites/treedbs/treedatabases.asp)

Osman ME, Ahmed EM, Barda HS, Eltohami MS (2015).Antioxidant and phytochemical properties of the seeds of Surib (Sesbania leptocarpa).*World. J. Pharm. Pharmaceu. Sci, 4 (1): 1598 – 1604.*

Pacheco WJ, Stark CR, Ferket PR, Brake J (2014). Effects of trypsin inhibitor and particle size of expeller extracted soybean mean on broiler live performance and weight of gizzard and pancreas. *J. Poultry Sci,93: 2245-2252*.

Pandhare RB, Sangameswaran B, Mohite PB, and Khanage SG (2011). Antidiabetic Activity of Aqueous Leaves Extract of Sesbaniasesban (L.)Merr.in Streptozotocin Induced Diabetic Rats. *Avicenna J. Med. Biotech. 3(1):37-43*.

Parab N and Validya S (2016).Determination of some trace elements and macro minerals of Sesbania bispinosa (Jacq) W.F. Wight. *Int J. Pharm. Pharmaceu. Res, 6 (2): 383-401*.

Patil R.B., Nanjwade B.K., and Manvi F.V (2010).Effect of *Sesbania Graniflora* and *Sesbania sesban* Bark on Carrageenan Induced Acute Inflammationand Adjuvan Induced Arthritis in Rats. *Inter. J. Pharm. Sci, 1(1): 75-89*.

Patra A.K., Chhonker P.K and Khan M.A (2006). Effect of green manure Sesbania sesban and nitrification inhibitor encapsulated calcium carbite (ECC) on soil mineral, enzyme activity and nitrifying organism in rice-wheat cropping system. *Eur. J. Soil Biol, 42: 173-108.doi:10.1016/J.ejsobi. 12.007.*

Pearson D.A (1976). Chemical analysis of foods (7^{th} edition).Churchill living tone, Eduinburgh.

Poeydomenge GY and Savage GP (2007). Oxalate content of raw and cooked purslane. *J. Fd. Agri. Envi., 5(1): 124-128.*

Pravin G., Priti G., Shaikh S., Khan M.S. (2012). *Sesbania sesban Linn*: A review on its ethnobotany,phytochemical and pharmacological profile. *Asian Journal of Biomedical and pharmaceutical science 2(12): 11-14.*

Pravina P, Sayaji D and Avinash M (2013).Calcium and its role in human body. *Int. J. Res. Pharmaceu. Biomed, Sci, 4(2): 659-668.*

Prohp T.P., Ihimire I.G., Madusha A.O., Okpala H.O, Erebor J.O. and Oyinbo.C.A. (2006). Some Antinutritional and mineral contents of Extra – cotyledonous deposit of pride of Barbados (Caesalpina pulcherrima). Parkistan *Journal of Nutrition,* 5(2); 114-116.

Pugalenthi M, Vadivel V, Gurumoorth P, Janardhanan K. (2004). Comparative nutritional evaluation of little-known legumes (*Tamarindus indica*, *Erythrina Indica* and*Sesbania bispinosa*. *Irop.Subtrop. Agrosyst,* 4: 107-123

Rahman MM, Abdullah RB, Wan Khadijah WE (2012). A review of oxalate poisoning in domestic animals: tolerance and performance aspects. *J. Anim. Physiol. Anim. Nutr, 97 (4): 605 – 14*.

Ramadan E. A. (2012). Effect of Processing and Cooking Methods on the Chemical Composition, Sugars and Phytic Acid of Soybeans.*Food and Public Health,* 2(1): 11-15 DOI: 10.5923/j.fph.20120201.03.

Redondo LM, Chacana PA, Domingueze JE, Miyakawa MEF (2014). Perspectives in the use of tanning as alternatives to antimicrobial growth promoter factors in poultry.Frontier in Mionuol, 5, 118/2.Reduviidae) as augmented control in groundnut pests.*J. Central Eur. Agric., 4: 103-110.*

Rehman Z., Shah W.H. (2005). Thermal heat processing effects on antinutrients, protein and starch digestibility of food legumes. *Food Chemistry 91: 327–331.*

Sabra HA., Hassan SG and Mohamed MI (2010). Effect of *Sesbania sesban*(Sesbania egyptiaca) Supplementation on the Reproductive Performances of Baladi Sheep as Compared to Bereseem (Egyptian Clover). *J. Reprod. Infertil*. 1(3):66-70.

Sahayaraj, K., and Martin, P. (2003).Assessment of Rhynocoris marginatus (Fab.) (Hemiptera: Reduviidae) as augmented control in groundnut pests. *Journal of central Europeans agriculture*, 4(2).

Sajid S., Vijay P.T and Usman R (2012).Antiinflammatory activity of Sesbania sesban (L) Merr. *Int. Res.J. Pharm, 3(1): 176 – 180.*

Samajdar S and Ghosh A.K (2017). Pharmacological effects of Sesbania sesban Linn: An overview. *J. Pharma. Tutor 5(7): 16-21.*

Sampathi, L.; Reddy, K. J.; Naidu, M. M., (1999). Nutritional evaluation of *Sesbania sesban* hay in the ration of sheep. *Indian J. Anim. Nutr., 16 (1): 53-55.*

Sandeep K., Singh B.B and Narinder K (2014). Evaluation of Anti-bacterial Activity of Plant *Sesbania sesban.Int J Pharm; 4(1): 385-396.*

Selle PH, Cowieson AJ, Cowieson NP, Ravindran V (2012). Protein – phytate interactions in pig and poultry nutrition: a reappraisal. *J. Nutr. Res. Rev, 25: 1-17*

Semwal BC, Verma M, Murti Y, Yadav HN (2018). Neuroprotective activity of *Sesbania ugradifolara* seeds Extract against Celecoxib induced amnesia in mice. *Pharmacogen J., 10(4):747-752.*

Shaheen N., Hussain M.M., Yousaf F., Qureshi M.S and Idrees S (2004).Effect of Rhizobium strains on growth of two Sesbania species. *Pak. J. Life Soc. Sci, 2(1):79-81.*

Shahidi, F. and Naczk, M. (2004).*Phenolics in food and nutraceuticals*, Washington, , USA: CRC Press.

Sharma D., Gupta R and Joshi I (2013). Nutrients analysis of raw and processed soybean and development of value-added soybean noodles. *Inventi Rapid: Life style,* 1: 1-5.

Siddhuraju P, Osoniyi O, Makkar HPS, Becker K (2002).Effect of soaking and ionizing radiation on various antinutritional factors of seeds from different species of an unconventional legume, Sesbania and a camman legume, green gram (Vigna radiate). *J. Fd. Chem 79: 273 – 281*

Siddhuraju P., Vijayakumari K., Janardhanan K., (1995). Studies on the underexploited legume, *Indigofera linofolia* and *Sesbania bispinosa*: nutrient composition and antinutritional factors. *Int J Food Sci Nutri.*,**46**: 195-203.

Siddiqui MA, Adam M, Hayat MY and Sandhu GR (1985). Nodulation studies in *Sesbania sesban* (L) Merr.2. Green manuring for wheat. *Pakistan J. Sci. Indus Res. 28:(407-411).*

Sileshi G and Hailu G (2006). Modelling the spatial patterns and interspecific interactions between three chrysomelid beetles defoliating the multipurpose agroforestry tree *Sesbania sesban* in Africa. *Afr. Entomol.* 14(2):337-348.

Sileshi G., Maghembe JA., Rao MR., Ogol CKPO and Sithanantham S (2000). Insects feeding on Sesbania species in natural stands andagroforestry systems in southern Malawi. *Agrofor. Syst.*49:41-52.

Singh S.P., (1990). Fertility control of female through Sesbania seeds. *Journal of research and Education in Indian medicine, 9(4): 27-32.*

Singh V, Mehra R, Bisht S, Shekhar M, Kuman A (2018). Phytin: A nutritional inhibitor in food and feed- Review of strategies and challenges to overcome the menace in maize. *Int. Curr. Microbiol. Appl.Sci, 7(6):3264-3279.*

*Slou*p V, Jankovska I, Nechybova S, Perinkova P and Langrova I (2017). Zinc in the animal organism: Areview. *Sci. Agri. Biochem, 48 (1) : 13 – 21*

Solorio-Sanchez FJ, Yanez IA, KU-Vera (2000). Chemical composition and in vitro dry matter digestibility of some fodder trees from south – east Mexico. *Livestock Res for Rural Dev, 12(4): 35.*

Steve I.O (2012). Influence of germination and fermentation on chemical composition, protein quality and physical properties of wheat flour (Triticum aestivum). *Journal of cereals and oil seeds, 3(3):35-47.*

Sun JY, Wang JF, Zi NT, Jing MY and Weng XY (2011).Effect of zinc supplementation and deficiency on bone metabolism and related gene expression in rat.*Bio. Trace Elem. Res*, 143, 394-402. doi: 10.1007/011- 010 – 8869-9.

Suzanne B.B.S., Mathieu N., Bertin S.E.,Calvin M.J., Annie N.N.R and Clergé T (2017). Effects of Cooking Time on Some Antinutrients Contents and in vitro Digestibility of Leaves Proteins of Gnetum spp. *International Journal of Nutrition and Food Sciences, 6(2): 99-104*.

Tatiya A.U., Patil U.K., Nikam DD., Surana S.J., (2008). Evaluation of antifungal activity of *Sesbania sesban* leaves. *Indian Drugs, 45(1): 44-47.*

*T*essema, Z. and Baars, R. M. T., (2004). Chemical composition, *in vitro* dry matter digestibility and ruminal degradation of Napier grass (*Pennisetum purpureum* (L.) Schumach.)mixed with different levels of *Sesbania sesban* (L.) Merr. *Anim. Feed Sci. Technol., 117 (1-2): 29-41*.

Tolera A (2007).Feed resources for producing export quality meat and livestock in Ethiopia (Examples from selected Weredas in Oromia and SNNP regional states), Addis Abab, Ethiopia.

Tolera A, (2017). The role of forage supplements in smallholder mixed farming system. In: Hare, M.D., Wongpichet, K., (Eds). Forages: A pathway to prosperity for smallholeder farmers. Proceedings of an international forage symposium. Ubon *ratchanthsni university, Thailand.165-186.*

Tresia PS and Mohan VR (2013).Assessment of nutritional and antinutritional potential of underutilized legumes of the genus Muccuna. *Trop. Subtrop. Agroecosys, 16: 155-169.*

Umunna, N. N.; Osuji, P. O.; Nsahlai, I. V.; Khalili, H.; Mohamed-Saleem, M. A., (1995). Effect of supplementing oat hay with lablab, sesbania, tagasaste or wheat middlings on voluntary intake, N utilization and weight gain of Ethiopian Menz sheep.*Small Rumin. Res., 18 (2): 113-120.*

Usman H and Osuji JC, (2007). Phytochemical and in vitro antimicrobial assay of the leaf extract of Newbouldia leavis. *Afr. J. Trad. CAM*. 4(4): 476-480.

Usman MA., Bolade MK, Hussein JB (2018). Selected antinutritional factors and in vitro protein digestibility of some sorghum types as influenced by germinating time during malting. *Int. J. fd. Sci. Biotech*, 3(2): 40-45.

Usman MRM., Patil SB., Patil SS and Patil RS (2013). *Sesbania sesban* Linn.: an overview. *International Journal of Pharmacy and Life Sciences* (IJPLS), 4(5):2644-2648. http://www.ijplsjournal.com/issues%20PDF%20files/may-2013/4.pdf.

Vadivel V., Patel A and Biesalsiu H.K (2012).Effect of traditional processing methods on the antioxidant, α-amylase and α-glucosidase enzyme inhibition properties of *Sesbania sesban* Merrill seeds. *CyTA J. Fd*,10 (2): 128-136.

Veasey, E.A., Schammass, E.A., Vencovsky, R., Martins, P.S. and Bandel, G. (1999).Morphological and agronomic characterization and estimates of genetic parameters of Sesbania Scop.(Leguminosae) accessions. *Genetics and Molecular Biology* 22: 81−93.

Verboom W.C., (1966). The grassland communities of Barotseland.*Trop Agric* (Trinidad), **43**:107-16.

Vodouhe A.; Dovoedo V. B.; Anihouvi R. C.; and Tossou, M. M. (2012). Influence du mode de cuisson sur la valeur nutritionnelle de Solanum macrocarpum, Amaranthus hybridus et Ocimum gratissimum, trois légumes feuilles traditionnels acclimatés au Bénin Sènan.*Int. J. Biol. Chem. Sci.* 6 (5): 1926-1937.

Wakjira M, Berecha G, and Bulti B (2011). Phytotoxic effects of multi-purpose tree species on germination and growth of Parthenium hysterophorus L. *Int. J. Agric. Res.*, 6(2):149-162. doi: 10.3923/ijar.2011.149.162.

Wakjira M., Berecha G and Bulti B (2014). Phytotoxic effects of multipurpose tree species on germination and growth of Parthenium hysterophorous L. *Int. J. Agric. Res*, 6(2): 149-162. Doi:10.3923/ijar.149.

Woldemeskel, M.; Tegegne, A.; Umunna, N. N.; Kaitho, R. J.; Tamminga, S., (2001). Effects of *Leucaena pallida* and *Sesbania sesban* supplementation on testicular histology of tropical sheep and goats. *Anim. Repr. Sci.*, 67 (3-4): 253-265.

Yang B, Shu WS, Ye ZH, Lan CY, and WongMH (2003). Growth and metal accumulation in vetiver and two Sesbania species on lead/zinc mine tailings. *Chemosphere*52:1593-1600. doi:10.1016/S0045-6535(03)00499-5.

Yatoo MI, Saxena A, Deepa PM, Habeeb BP, Devi S, Jatar RS and Dimri U (2013). Role of trace elements in animals: a review. *Veterinary world*, 6 (12): 963-967.

Yusuf M, Chowdhury J, Wahab M, Begum J., (1994). Medicinal plant of Bangladesh council of scientific trial research (BCSIR).Dhaka, Bangladesh, P223.

Zhishen J, Mengecheng T, Jianming W, (1999). The determination of flavonoid content Mulberry and their scavenging effects on superoxide radical. *Food Chem.*, 64, 555-559.