

**EFFECTS OF BOILED AND FERMENTED AFRICAN LOCUST BEAN (*PARKIA
BIGLOBOSALINN.*)SEEDSDIETARY INCLUSIONS ON GROWTH
PERFORMANCE AND CARCASS QUALITY OF WEANER RABBITS**

BY

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JULY, 2015

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BIGLOBOSA* LINN.) SEEDS DIETARY INCLUSIONS ON GROWTH
PERFORMANCE AND CARCASS QUALITY OF WEANER RABBITS

BY

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JULY, 2015

DECLARATION

I declare that the work in this thesis entitled “**EFFECTS OF BOILED AND FERMENTED AFRICAN LOCUSTBEAN(*PARKIA BIGLOBOSA* LINN.) SEEDSDIETARY INCLUSIONSON GROWTH PERFORMANCE AND CARCASS QUALITY OF WEANER RABBITS**” has been carried out by me in the Department of Biological Sciences,Ahmadu Bello University, Zaria, Nigeria. The information derived from the literature has been duly acknowledged in the text and in a list of references provided. No part of this thesis was previously presented for another degree or diploma at this or any Institution.

ADEOLA ADEBIMPE DANIEL _____
Signature Date

CERTIFICATION

This thesis entitled **EFFECTS OF BOILED AND FERMENTED AFRICAN LOCUSTBEAN (*PARKIA BGLOBOSA* LINN.) SEEDSDIETARY INCLUSIONS ON GROWTH PERFORMANCE AND CARCASS QUALITY OF WEANER RABBITS** by **ADEOLA ADEBIMPE DANIEL** meets the regulations governing the award of the degree of Master of Zoology of the Ahmadu Bello University, and is approved for its contribution to knowledge and literary presentation.

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DEDICATION

I dedicate this thesis to Almighty God for seeing me through my studies, my beloved husband, Barrister F. B. Daniel, my loving children Annabella and Arnold, my late father Pa B. O. Efunkoya, my beloved mother, Mrs. B. O. Efunkoya and my siblings.

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ABSTRACT

A study was conducted to evaluate the effect of boiled and fermented locust bean seeds as dietary inclusions on growth performance and carcass quality of weaner rabbits. Samples of raw, boiled and fermented seeds were taken to the laboratory for proximate composition and anti-nutritional factors evaluation using Association of Official Analytical Chemists (AOAC). The results of proximate composition of the seeds revealed that fermented seeds had significantly ($P<0.05$) high Crude protein, low Dry Matter (DM), low Crude Fibre (CF), Nitrogen Free Extract (NFE) and less than raw and boiled seeds. Processing methods significantly ($P<0.05$) reduced phytic acid, saponins, tannin, cyanide and oxalate contents of seeds. The reduction was more significant ($p<0.05$) in the fermented seeds; phytate from 409.83% to 55.63%; saponins from 207.83% to 44.38%; tannin, 176.50% to 63.74%; cyanide, 87.83% to 68.38%; and oxalate, 181.83% to 68.17%. Four experimental diets were thereafter compounded; treatment 1 was the control while treatments 2, 3, and 4 were diets with raw, boiled and fermented locust bean seeds respectively. Twelve (12) % of the seed was included in the diets and the diets were compounded to be iso-caloric and iso-nitrogenous to meet the nutrient requirement of growing rabbits. A total of 20 weaner rabbits were used, five (5) rabbits per treatment with each serving as a replicate in a completely randomized design and the experimental diets were *fed ad libitum* for eight (8) weeks. The results of the growth performance of the rabbits revealed that rabbits fed fermented locust bean seeds had significantly ($P<0.05$) better average daily weight gain (125.09g/wk), final weight (1312.50g) and feed conversion ratio (5.04) than the raw seeds. However, boiling of the African locust bean seeds significantly ($P<0.05$) improved palatability (protein) of the diet as animals on this diet consumed more feed compared to other treatments. The proximate composition of

rabbit meat was taken to be a reflection of the nutrient composition of the seeds. Since meat from rabbits on the diet with the fermented seeds had a significantly ($P < 0.05$) high crude protein of 41.54, low fibre and low ash. Meat from rabbits on the raw seed diet had significantly ($P < 0.05$) higher ash content in the liver, heart, kidney and lungs. This could be a reflection of the effect of anti-nutritional factors. The results therefore suggest that feeding processed locust bean seeds as protein source to rabbits have no adverse effect on growth performance and meat quality of rabbits and the fermented seed performed better than the raw and boiled.

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CHAPTER ONE

1.0 INTRODUCTION

1.1 Background of the Study

Nigeria and other developing countries are experiencing protein deficiencies. Olugbemi *et al.* (2010) reported that there was protein deficiency gap for which careful attention is necessary to prevent metabolic diseases in Nigerian citizens. Nigeria's population is increasing rapidly and this indicates the need to strategize action and device means of improving the production of animal protein for the populace. Rabbit production is one of the easiest ways of meeting the protein demand of the average Nigerians due to their short generation interval and rapid population growth rate (Oyawoye, 1989; Esenwah and Ikenebomeh, 2008).

Protein–Energy–Malnutrition (PEM), is a serious problem facing most developing nations as a result of inadequate intake of good quality protein source such as meat, fish and poultry product, which are out of reach to many people due to poor economy, increase in population pressure, high cost of feed and other natural calamities such as drought and flood (Ladeji *et al.*, 1995; Nordeide *et al.*, 1996). In these nations such as Africa, about 60% of the population suffer PEM, which results to high rate of mortality, permanent brain damage and decrease in learning capability of children (Abdullahi, 2000).

A lot of effort is therefore required in popularizing the raising of rabbits among the rural populace and other low income groups. Rabbits hold a lot of promise for increasing the supply and intake of animal protein among these classes of people. This is because they multiply and grow rapidly producing an average of 6-8 kittens and kindle 3-4 times yearly as a result of their short gestation period of 28-30 days. Rabbits produce high quality meat and can utilize very cheap feed materials (Aduku, 1992).

The prices of animal products have soared in the last two decades; this is as a result of increases in the prices of protein feedstuffs used in livestock feed formulation (Adeniji, 2000; Esonu *et al.*, 2005). This is also mainly due to competition between human beings, industries and livestock for the available feedstuffs. These crippling realities that are characteristic of third world countries have led to the use of locally available, and cheap industrial by-products, novel crops and animal wastes as feed ingredients. Feed cost and quality determines the growth rate and the population of the animals that can be kept on the farm (Aduku, 1992). There is therefore, the need to intensify research into alternative feed sources that are affordable and available to cut down cost of production.

The use of African locust bean (*Parkia biglobosa*) as feed ingredient was explored in this research. The seeds of the locust bean is reported to contain about 20% of semi-liquid oil of which 54% of the fat is unsaturated with linoleic acid predominating, while 32% consist of palmitic acid. It is low in sulphur amino acid for example, methionine and cystine and similarly low in histidine but high in lysine (Oyenuga, 1978; Ogundun, 2007).

1.2 Statement of the Research Problem

Africa has been recently reported to have high cases of adult mortality as a result of high blood pressure (Abdullahi, 2000). In recent times, the production and population of rabbits is declining and there is a need to encourage the raising of this animal because of its outstanding qualities such as protein value, docile nature and so forth. Feeding rabbits as well as other livestock with quality diets has continued to be the problem of most farmers because of the high price of feed ingredients. In order to arrest this situation, much attention has been focused on the exploitation and utilization of non-conventional feed materials that are affordable and available to be used as

livestock ingredients. Among the leguminous seeds that can be used in feeding livestock particularly in some African countries, is the African Locust bean(*Parkia biglobosa*) seed. Furthermore, *P. biglobosa* is a plant legume with an outstanding protein quality and its protein and amino acid composition have been reported by several authors (Ega *et al.*, 1988; Nordeide *et al.*, 1996; Glew *et al.*, 1997; Cook *et al.*, 2000; Lockett *et al.*, 2000).

1.7 Justification

One attribute of the domestic rabbits above other livestock and pets is that they are attractive animals for handicapped children who enjoy being involved in their care and management and are also preferred to be used as laboratory animals for learning and teaching because of their docile nature. However, inadequate feeds and feeding have constituted a major constraint limiting production and the popularity of rabbits. Also nutritional solution to prevention of high blood pressure has often been linked to the consumption of diets that are low in cholesterol and rabbit meat has been reported to have low cholesterol and high digestible nutrients.

The performance of a good and healthy rabbit can become poor when inadequately fed while low producing ones can be improved on with good feed, because an animal which is not well fed cannot give its best (Sandford, 1979). Therefore, the type of feed provided for rabbits must be given special attention to ensure it meets the requirements for energy, protein, vitamins, minerals and other micro nutrients necessary for optimum growth and development. Hence, the key to abundant animal production is the availability of cheap and balanced feed. Feed cost and quality also determine the growth rate and the population of the animals that can be kept on the farm (Aduku, 1992).

The African locust bean seed if used with other leguminous seeds like groundnut and soybean may reduce the cost of production of livestock to 45 – 55%. The African locust bean seeds are available all year round and affordable. The fat content of the seeds is appreciably lower than that of groundnuts. The seeds are also known to contain 30.36% crude protein, 5.3% ash, 20.3% ether extract and 8.82% crude fiber. The contents of its crude fiber and carbohydrate are also suitable for all classes of livestock (Oyenuga, 1978; Ogundun, 2007).

1.8 Aim

To evaluate the effect of raw, boiled and fermented *Parkia biglobosa* seeds as dietary inclusions on growth performance of weaner rabbits.

1.9 Objectives of the Study

The specific objectives therefore include:

- i. To determine the proximate composition of raw, boiled and fermented African locust bean seeds.
- ii. To determine the effects of boiling and fermentation on the anti-nutritional contents of the African locust bean seed over raw.
- iii. To determine the effects of dietary inclusions of boiled and fermented African locust bean seed on the growth performance of rabbits.
- iv. To determine the effect of the diets on the carcass composition of rabbits.

1.10 Research Hypotheses

- i. There is no significant difference in the proximate content of boiled and fermented African locust bean.

- ii. There is no significant difference in the antinutritional content of raw, boiled and fermented African locust bean meals.
- iii. There is no significant difference on the effect of feeding boiled and fermented African locust bean meals on the growth performance of rabbits.
- iv. The experimental diets have no effects on the carcass composition of the rabbits.

CHAPTER TWO

2.0

LITERATURE REVIEW

2.1 Origin and Distribution of Rabbits

The domestic rabbit originated from the wild rabbit (*Oryctolagus cuniculus*). It is native to south-west Europe. Some 2000 years ago the Romans kept rabbits in enclosure, 'leporaria', for their meat. This method was later used in France and England; it served to meet the desire for hunting. The first wild rabbits came to Germany around 1300 BC, and they were kept on the island of Amrum. Real domestication was completed in the Middle Ages, initially in France, by keeping them in enclosures (Oglesbee, 2006). The first books on this animal species also appeared in France. Domestic rabbits were kept in Germany from the middle of the twelfth century, i.e. after the wild form. They were still kept extensively in enclosures. Soon after the animals became domesticated, the first different colours and forms appeared. At the end of the eighteenth century there were already several breeds, including the Angora rabbit and animals with lop ears. Currently, there are about 80 breeds with approximately 200 colour strains as shown in Table 2.1, which can be divided into five groups which are depending on size and hair length. In addition, there are breeds which do not fit into the normal classification, as well as new breeds (Whitman, 2004).

2.2 Breeds of Rabbit

The concept of breed was first used in animal breeding in the sixteenth century, but its meaning has undergone many changes with time and is still different in the minds of different people.

Table 2.1: The Rabbit breeds of the world

S/N	Breeds	Fur Colours
1	Angora	Grey
2	British giant	Brown grey
3	Cashmere lops	Brown
4	Kalisto	Grey
5	Yokozuna	Brown grey
6	Beige	Various colour
7	Paris	Blue Butterfly
8	Siamese	Smoke Butterfly
9	Maximus	Black Dutch buck
10	Hershey	Chocolate Dutch
11	Tiger	Tri-colour Dutch
12	Fox	Blue
13	Thuringer	Deep yellow ochre colour with blue-black ticking.
14	Hotshot	Opal buck
15	Humbug	Sooty Fawn
16	Agouti	Agouti Butterfly
17	Mindi	Black Butterfly
18	Eve	Blue
19	Fuji	Blue Point
20	Portia	Blue Point
21	Geromino	Brown
22	Possum	Chocolate
23	Possum	Sable (dark)
24	Little foot	Seal Point
25	Peter pan	Seal Point with darker points
26	Bonzai	Seal Point with darker shadings
27	Mia	Siamese Smoke Pearl (lighter)
28	Gidget	Sooty-Fawn
29	Mini rex	Siamese Smoke
30	Nazara	Black Otter
31	Chinchilla	Various colour patter
32	Netherland dwarf	Black
33	Merula	Black otta
34	Cino	Chocolate
35	Winsome	Chocolate otter
36	Cinnamon	Brownish-grey
37	Comet	Lilac
38	Cinnamon	Orange
39	Bisket	Siamese sable

Table 2.1: Continued.....

40	Winston	Silver fox
41	Rex	Chocolate
42	Coco	Chocolate
43	Aurelius	Orange
44	Tan	Black
45	Licorice	Black
46	Cocoa	Chocolate
47	American fuzzy lop	Slate blue
48	American sable	White
49	French Angora	Brown
50	Belgian Hare	Grey
51	Beveren	Dense black
52	Britannia petite	Otter, black, white
53	California	White
54	Chinchilla gigantea	Deep blue slate
55	Dutch	Various colours in the standard Dutch pattern contrasting with white
56	Fox	Black, blue, Lilac or Chocolate
57	French lop	Various colour
58	German angora	Red eyed white
59	Jersey wooly	Various colour
60	Harlequin	One or combination of blue, blue, brown or lilac colours
61	Mini Cashmere Lop	Various colour and pattern
62	Miniature Lop	Various colour and pattern
63	Plush Lop	Various colour and pattern
64	Polish	Various colour and pattern
65	Satin	White and any colour
66	Tan	Black and tan, blue and tan, chocolate and tan and lilac and tan
67	Swiss Fox	White, black and combination of many colours.
68	Mini Cashmere Lop	Various colours

Table 2.1: Continued.....

69	Beige	Slate blue-ticking
70	Blanck de hotot	White
71	Californian	White with red eye
72	English Spot	The unique markings on their body, including eye circles, the butterfly mark, herringbone, coloured ears, cheek spots, and a chain of small spots
73	Flemish Giant	White underside and a dark base colouring
74	Golden Glavcot	Blueish undercolour
75	Havana	Chocolate
77	Lionhead	Brown
78	Perlfee	Mainly greyish blue
79	Silver Marten	black, blue, sable, and chocolate
80	Thrianta	Red rabbits with the fawn colouring on their tail and paws.

Source: Whitman, 2004.

Among others, the following definitions have recently been used. Encarta Encyclopedia (2011) defined breed as a population or a group of population which can be distinguished from other populations of the same species on the basis of different allele frequencies, chromosome changes or genetically determined phenotypic characteristics.

2.2.1 New Zealand White Breed

Sandford (1986) reported that there are about four different strains of the New Zealand breed which include; New Zealand White, New Zealand Blue, New Zealand Red and New Zealand Black, respectively. The New Zealand Red strain was first developed in California with a system of selecting similar breed to that used in France on the Burgundy Fawn Strain (Lebas, 1983). The genetic constitution of the New Zealand Red strain include the gene for the elimination of all black pigment from the Agouti colouring, thus giving it a complete extension of the yellow colour (Sandford, 1986). Thereafter, a number of modifying genes to produce the bright reddish buff colour was introduced. The New Zealand White strain although very similar to the Red, Blue and the Black strains, differs considerably. It is heavier and larger, weighing up to 4.07 Kg (Sandford, 1986; Lebas *et al.*, 1986). Aduku and Olukosi (1990) reported that New Zealand White breed of rabbit can reach adult weight of 4.1 to 5.4 Kg.

2.2.2 Californian Breed

The California is an American breed, which was presented for the first time in 1928 in Californian by its breeder, whose objectives was a meat animal that has a very good fur (Lebas *et al.*, 1986). According to Sandford (1986), the Californian breed was developed by crosses of New Zealand White and Himalayan with further introduction of the Chinchilla gene but the breed, according to the breeder, did not become well known in America for some 15 years

after its original development. Lebas *et al.* (1986) stated that the California breed is today possibly the second most popular meat producing rabbit. The adult Californian weighs between 3.6 - 5.0 Kg (Lebas *et al.*, 1986; Aduku and Olukosi, 1990).

2.2.3 Chinchilla Breed

The Chinchilla rabbit falls under both the medium and small class breeds. The Chinchilla *giganta*, also called Grand Chinchilla, weighs between 4.10 and 5 Kg, while the small breed weighs between 2.5 and 3.4 Kg (Lebas *et al.*, 1986; Sandford, 1986; Aduku and Olukosi, 1990). Sandford (1986) reported that the intention in the production of the Chinchilla *Giganta* in the early 1920s by Chris Wren was to produce a large, fitness rabbit, with good meat qualities and good pelt with the Chinchilla colouring. The advantage of the small breed, according to Lebas *et al.* (1986), is that they usually develop very quickly and make excellent mothers. They eat less than the medium and large breeds and could be crossed or used as purebred in developing countries to produce light, meat carcass of 1 to 1.2 Kg (Bawa *et al.*, 2005).

2.3 Feeds and Feeding of Rabbits

Rabbits can be maintained on ration consisting of roughages, home grown vegetables and cereal grains. These feeds are generally supplemented with additives such as lysine, methionine, vitamin-premixes and a source of calcium such as limestone or bone meal. Concentrates can be fed in a mash form when it is home produced or in pelleted form as in most commercial operations.

Anyanwu *et al.* (1982) reported that succulent forages can be offered to rabbits. The digestion of these green forages according to the authors is as a result of the nature of their simple stomach that has a large caecum in their digestive tract. They further stated that this large caecum

contains a few microorganisms, which are involved in the conversion of cellulose in the forages to the digestible glucose.

2.3.1 Energy Requirement of Rabbits

Appetite in rabbit is mostly regulated by a chemostatic mechanism; hence, the total quantity of energy ingested daily tends to be constant (Lebas, 1980). The author further stated that rabbits grown in good sanitary conditions, naturally consume sufficient feed to meet their energy requirements. However, reproduction does have high energy requirements for pregnancy, lactation and concurrent pregnancy and lactation that are often not covered by an adequate voluntary energy intake. Voluntary energy intake is proportional to metabolic live weight ($LW^{0.75}$). In growing rabbits, voluntary intake is about 900–1000 kJ DE/ day/ kg $LW^{0.75}$ and chemostatic regulation appears only with a DE concentration greater than 9 MJ/ kg in the diet (Cheeke, 1987; Parigi-Bini, 1988; Santoma *et al.*, 1989). Below this level, a physical type regulation is prevalent, which is linked to the filling of the gut with dietary material (Cheeke, 1987).

An increase in the level of dietary energy intake also affects body gain composition and the partition of energy retained as protein and fat. The body composition changes are not linearly correlated with dietary energy (DE) intake, because some constituents such as fat tend not to increase in proportion to the dietary energy intake. The efficiency of energy utilization for growth is clearly influenced by the levels of growth, because energy retained from protein sources are less efficiently utilized than that of fat (Blaxter, 1989; Close, 1990). Lebas (1983) stated that a growing rabbit is able to adjust its voluntary feed intake according to the energy

concentration of the diet in order to maintain constant daily intakes of 920 -1000/KJ/kgW^{0.75}DE per day.

Energy required by rabbits is within 9.6-9.64 MJ/kg metabolizable energy, depending on the physiological state of the animals (i.e. maintenance, pregnancy, growth, lactation) and this energy can be supplied mainly from carbohydrates and fats (Lebas, 1980; Aduku and Olukosi 1990). Lang (1988) reported that energy requirements in rabbits are greater for lactation than for other productive functions. Feed intake for rabbits is primarily to satisfy their energy requirement (Lebas, 1980). Like any other monogastric animal, the rabbit enzymes cannot digest cellulose, rather it is broken by the bacteria in their caeca (Fielding, 1991).

2.3.2 Protein Requirements

Many attempts have been made to determine the exact protein requirement of rabbits. Reports obtained so far have shown a dietary requirement for ten amino acids (Adamson and Fisher, 1971; NRC, 1995). Aduku and Olukosi (1990) reported that the quantity and quality of these amino acids are not critical in rabbits as in other animals such as poultry because rabbits practice coprophagy (usually soft faeces) and can adapt to low or poor protein situation, except that the production will not be optimal. With high and good protein quality however, optimum production can be achieved.

In growing rabbits, dietary protein (DP) is estimated to be 2.9 g DP/ day/ kg LW^{0.75} (Partridge *et al.*, 1989; Fernandez and Fraga, 1996; Motta-Ferreira *et al.*, 1996; Fraga, 1998). Lower DP has been found in a new strain of laboratory rabbits (2.11–2.14 DP/ day/ kg LW^{0.75}), which was attributed to a lower basic metabolic rate (Lvet *et al.*, 2009). In non-reproducing adult rabbits, there

is no specific information on DP hence, the same figure is assumed as for growing rabbits. Dietary protein requirements vary according to the growth rate.

The efficiency of utilization of dietary protein intake for growth is estimated to be 0.56% (Partridge *et al.*, 1989; Fernandez and Fraga, 1996; Motta Ferreira *et al.*, 1996; Fraga, 1998). Overall DP retention (RP/DPI) decreases linearly from 0.40% to 0.10% with increasing live weight, due to the increase in dietary protein used for maintenance (Xiccato and Cinetto, 1988; Trocino *et al.*, 2000; 2001). Lebas (1980) and NRC (1995) have recommended 12-13% crude protein for maintenance, 15-16 % for growth, 15-18 % for gestation and 17-18 % for lactation.

The most limiting essential amino acids in rabbit diets are methionine, lysine and threonine (Taboada *et al.*, 1996). A minimum level of 5.4 g total sulphur-containing amino acid /kg (4.0g digestible amino acid/ kg) is required to obtain adequate productivity in growing and non-reproducing rabbits. A higher level (6.3 g total amino acid/ kg and 4.9 g digestible sulphur-containing amino acid/ kg) was recommended for reproducing females to increase milk production, reduce the interval between parturitions and improve feed utilization efficiency (Taboada *et al.*, 1996). Recommended levels of lysine (for lactation diets with 10.5–11 MJ DE/ kg) was 6.8 g total lysine/ kg (5.2 g digestible lysine/ kg) for maximum reproductive performance; and 7.6–8.0 g total lysine (6.0–6.4 g/ kg digestible lysine) /kg for maximum milk production and litter growth (Taboada *et al.*, 1996).

Various units are available for expressing protein requirements (de Blas and Mateos, 1998; Fraga, 1998; Carabano *et al.*, 2000; 2008). Crude protein (CP) and apparent dietary protein (DP) are the most commonly used available description for both requirements and raw material composition (Villamide *et al.*, 1998; Maertens *et al.*, 2002).

In reality, rabbits have specific amino acid requirements and apparent faecal and true ileal digestible amino acids would be more reliable units. All the same, even if increasing information is given on the amino acid concentrations of the most common raw materials, digestible amino acid requirements and concentration in feeds are barely known; and even less information exist on ileal digestible amino acid (Carabano *et al.*, 2008). In practice, due to the chemostatic regulation of appetite in rabbits, nitrogen requirements are expressed in relation to dietary energy by the dietary protein (DP) to the dietary energy (DE) ratio, which is directly correlated to body nitrogen retention and excretion (Carabano *et al.*, 2000).

2.4 Taxonomy, Distribution and Ecology of African Locust Bean Seeds

The genus *Parkia* belongs to the family of Leguminosae and subfamily of Mimosoideae. It is a pantropical genus comprising about 30 species of trees. According to Hopkins (1983), only three species deserve to be retained for the entire African continent [*P. biglobosa* (Jacq.) R. Ex G. Br Don, *P. bicolor* Chev. and *P. filicoidea* Welw. Ex Oliv]. The fourth species *P. madagascariensis* R. Viguier is confined to Madagascar. In Hopkins' revision, *P. Africana* R.Br and *P. clappertoniana* Keay became synonymous with *P. biglobosa* (Ouédraogo, 1995). The francophone term "nééré" is a common name in several countries in West Africa, and it is called "African locust bean" in English (Burkill, 1995). The species occurs from the Atlantic coast of West Africa to Uganda in East Africa (Hopkins and White, 1984). The species supports a wide range of climatic conditions consistent with the main dry season from 5 to 7 months per year. It can grow in areas where rainfall is between 500 mm (Sahel) and 2200 mm (Guinea-Bissau) with records of more than 3500 mm in Sierra Leone and Guinea Conakry 4500 mm. Although preferring deep loamy soils, *P. biglobosa* may also occur on shallow lateritic soils, thick lateritic soils, rocky knolls and rocky hills (Sina, 2006). *Parkiabiglobosa* is found at altitudes ranging

from sea level (50 m above sea level in Senegal and Gambia) up to 1350 m in the mountains of the Fouta Djallon in Guinea Conakry (Hopkins and White 1984; Hall *et al.*, 1997). The average ideal annual temperature for *P. biglobosa* is between 26° C and 28°C.

Even though more than 250,000 plant species have been described worldwide as a source of food to man and animals, less than 30 species provide 90% of the world's food requirement and mostly cereals, to which rice, wheat and corn are the major sources and collectively supply nearly 60% of the world's food supply (Oliveira *et al.*, 2000; Parvathin and Kumar, 2002). Ordinarily, Plants provided nearly two-thirds of the world supply of food protein for human and animals in which 10 – 15% comes from legumes (Baudoin and Maquet, 1999; Pirman *et al.*, 2001). Despite the presence of anti-nutritional factors, legumes owing to their high proportion of protein even though unbalanced, are still regarded as potential source of protein (Baudoin and Maquet, 1999; Pirman *et al.*, 2001). Apart from protein, legumes provide a high proportion of complex carbohydrates, starch, edible oil and fibre (Chau *et al.* 1998; Pirman *et al.*, 2001).

2.4.1 Description of the African locust bean plant

2.4.1.1 *Trunk and branches*

Parkia biglobosa can measure up to 30 m, with a robust trunk up to 1.5 m of diameter (Bonkougou, 1987). The crown has a form of a ball or an umbrella (Ouédraogo 1995). The trunk is covered with a crust of varying color, striated, giving a scaly appearance as shown on Plate I.



Plate I: Aerial parts of *Parkia biglobosa* tree



Plate II: The fruits of *Parkia biglobosa*

2.4.1.2 Leaves, flowers, pods and seeds

The leaves of *P. biglobosa* are alternating bipinnate with a spine of 20 to 40 cm of length, counting 6 to 18 pairs of pinnae. A leaf comprises from 13 to 60 pairs of leaflets. (Bonkougou, 1987; Ake and Guinko, 1991). The inflorescences are spherical heads from 4.5 to 7 cm long and 3.5 to 6 cm of diameter. They hang at the end of stems of 10 to 35 cm of length. The pods are glabrous, slightly flattened, more or less linear from 12 to 30 cm long and 1.5 cm to 2 cm wide (Ouédraogo 1995). The seeds are ovoid, hard-coated and smooth.

2.4.1.3 Roots

Parkia biglobosa has a taproot system. The length of the main root is generally less than 2 m (Breman *et al.*, 1988). In addition to the main root, *P. biglobosa* produces lateral roots whose length can reach 10 m (Tomlinson *et al.*, 1998).

2.5 Effect of Processing Methods on Nutrient and Anti-nutrients of African Locust Bean

2.5.1 Protein

Crude proteins of un-cooked and cooked Locust bean were reported to be $29.99 \pm 0.01\%$ to $18.43 \pm 0.06\%$, respectively. Previous work reported decrease in protein content of *Canavalia cathartica* from 32.1 to 28.1% after cooking (Seena *et al.*, 2006). In addition, Baiyeri *et al.* (2011), reported reduction in protein content of cooked banana from 3.21 to 2.48%. The Locust bean has the potential of being a source of plant protein. Protein quality is a measure of the usefulness of a food protein for the purpose of growth and maintenance of tissues. Thermal denaturation results in coagulation and decreased solubility (Ihekoronye and Ngoddy, 1985).

2.5.2 Fat

Akinoso and Raji(2011) reported that cooking reduced the fat content of the locust bean from $20.03 \pm 0.06\%$ to $1.23 \pm 0.06\%$. The African locust bean oil ranged from 19.0 to 22.5% (Akinoso and Raji, 2011). A wide margin in fat content of cooked and un-cooked African locust bean can be traced to aqueous extraction of oil from the seeds during cooking. Heating fractionated intact oil bodies and ruptured cellular structure, thus aiding oil extraction.

2.5.3 Carbohydrate

Rahman and Naimat (2013) in Congo reported the carbohydrate content of the raw and cooked African locust bean seeds to be $29.93 \pm 0.13 \%$ and $18.93 \pm 0.06\%$, respectively. According to the author, the remarkable reduction in the carbohydrate content is due to hydrolysis of starch to simple sugars during the cooking period. Hydrophilic groups in carbohydrate molecules caused it to take up moisture in proportion to the relative humidity of the environment (Ihekoronye and Ngoddy, 1985). This characteristic behaviour encouraged moisture uptake and apparent reduction in percentage of the carbohydrate.

2.5.4 Ash

Rahman and Naimat (2013) reported that ash content is high in raw African locust bean seed than the cooked. The values they recorded for the raw and cooked seeds were $5.25 \pm 0.01\%$ and $2.67 \pm 0.15\%$, respectively. The 5.25% obtained for raw in their study was similar to the 5.40% obtained (Omafuvbe *et al.*, 2004) but slightly higher than the 4.24% obtained by Elemo *et al.*(2011). Usually, there is no appreciable loss of ash in legumes during cooking (Table 2.1).

2.5.5 Moisture content

Cooking the locust bean for four hours increased its moisture level from $6.09 \pm 0.04\%$ to $52.23 \pm 0.15\%$ (Rahman and Naimat, 2013). Rise in moisture content of the locust bean from 8.8% to 56.7% after 6 hours of cooking was reported. Moisture uptake of 46.14% was recorded during the cooking process (Omafuvbe *et al.*, 2004). Significant changes in chemical composition of the crop during cooking are attributed to re-distribution due to this high moisture uptake as recorded by Elemo *et al.* (2011) in Table 2.1.

2.5.6 Crude fiber

The crude fiber content in the African locust bean is $8.71 \pm 0.04\%$ and $6.50 \pm 0.15\%$ for raw and cooked samples, respectively (Rahman and Naimat, 2013). However, fiber content has been reported to reduce during treatment such as dehulling, cooking and fermentation. The hull contains a high portion of the fiber present in the seed (Akinoso and Raji, 2011). Reduction in crude fiber of the African locust bean from 11.7% to 4.45% after 6 hours of cooking had also been reported (Omafuvbe *et al.*, 2004) compared to what is recorded in Table 2.2 by Elemo *et al.* (2011) as standard.

2.6 Anti-nutritional Factors Present in African Locust Bean Seeds

The use of the African locust bean seeds and other legumes as protein sources is limited by the presence of anti-nutritional factors which are a diverse range of naturally occurring compounds in many tropical plants. The anti-nutritional factors cause poor protein digestibility in man and animals and are capable of precipitating other deleterious effects. Manifestations of toxicity from the consumption of legumes containing anti-nutritional factors range from severe reduction in food intake and nutrient availability or utilization, to profound neurological effects and even death (Osagie, 1998).

Table 2.2: Proximate composition of African locust bean (*P. biglobosa*).

Chemical Components	Composition (%)
Protein	27.90
Starch	0.39
Dry matter	88.79
Ash	4.24
Fat	15.48
Carbohydrate	41.10
Moisture	11.21
Total protein	1.07
Glucose	0.09

Source: Elemo *et al.* (2011)

Table 2.3:Fibre composition of African locust bean (*P. biglobosa*)

Fibre Component	Composition (%)
Crude fibre	11.23
Natural detergent fibre	28.34
Acid detergent fibre	19.60
Hemicellulose	8.80

Source: Elemo *et al.* (2011)

The African locust bean seed like any other leguminous seed is known to contain some anti-nutritional factors such as tannins, phytic acid, saponins, cyanide and oxalate (Oyenuga, 1978). The anti-nutritional factor of tannins (40.0 mg/g) that has been recorded by Elemo *et al.* (2011) in *P. biglobosa* seed are known to affect protein utilization by binding to lysine and making it unavailable for digestion and absorption. As low as 3-7% levels of tannin can cause death in monogastric animals (Bagepallis *et al.*, 1982). The phytic acid composition of African locust bean seeds of 163.0 mg/g as reported by Esenwah and Ikenebomeh (2008) has the ability to bind and forms indigestible phytates that make mineral unavailable thereby leading to mineral deficiency in monogastrics (Thompson, 1993). Saponins (187.0 mg/g) are described as natural detergents because of their foamy nature and have anti-carcinogenic properties which inhibit the growth of cancer cells and the lower cholesterol activities (Seigler, 1998). Hydrogen cyanide (53.2 mg/g) of African locust bean seeds has been reported to cause vitamin B₁₂ deficiency and retard physical growth of animals (Anhwanghe *et al.*, 2004).

To improve the nutritional quality and organoleptic acceptability of leguminous seeds, processing techniques have been employed to reduce or destroy the anti-nutrients present in them. Some of the commonly used processing techniques include soaking in water, boiling at high temperatures in water, alkaline or acidic solutions, sprouting, autoclaving, roasting, dehulling, microwave treatment, steam blanching and fermentation.

2.7 Socio-Economical Importance of *Parkia biglobosa*

Parkia biglobosa is one of the most important tree species of the West African parklands (Lamien and Bayala, 1994; Ouédraogo 1995; Lamien *et al.*, 1996). It is a multipurpose tree, highly used in West Africa (Ouédraogo, 1995). The leaves, the bark, the roots and the flowers are used for

medicines. The seeds are utilized for “soubala”, a fermented condiment used in sauces (Arbonnier, 2000). The trunk and the branches are appreciated as fire wood. Although the tree has multiple uses and is highly appreciated by the rural population in West Africa (Arbonnier, 2000), the species is characterized by ageing populations with very low regeneration rates (Bouda and Nikiema, 1996; Sina, 2006; Ræbild *et al.*, 2011), leading to fragmentation and smaller stands of the species.

2.8 Effects of Processing Leguminous Seeds on Growth Performance and Carcass Characteristic of Rabbits

McDonald (1995) reported that the African locust bean seeds contain a number of toxic stimulatory and inhibitory substances including allergenic, goitrogenic and anticoagulant factors. These factors according to the author are responsible for growth retardation in farm animals. The retardation has been attributed to inhibition in protein digestion. The methods of processing the seeds to lower or totally eliminate these anti-nutrients have been a major challenge to most farmers (Okagbare and Akpodiete, 2006). If the seeds are not properly processed, you may end up denaturing the protein in the seeds. A less than 25 % inclusion level of fermented locust bean seed in the diet of rabbits, improved feed intake (Akpet *et al.*, 2012) and declined thereafter. It was believed that beyond this level, the aroma declined while the unpleasant smell increased making the animals reduce their feed intake.

Amaefule *et al.* (2004), Udedibie *et al.* (2005), Bawa *et al.* (2005, 2007) and Taiwo *et al.* (2006) reported a decline in the growth performance and carcass characteristics of rabbits in treatments with raw pigeon pea seed meal, jack bean, neem seed cake, mucuna seed meal and African locust beans. Their results were contrary to the reports of Ani and Ugwuowo (2011) who fed graded levels of processed mucuna to rabbits. The authors reported that there were remarkable differences in

growth performance observed in the study was either due to the fact that cooking reduced the anti-nutritional factors (ANFs) in raw *Mucuna* seeds to such a level that did not cause distortions or disproportionate growth of rabbits, or the levels of cooked Mechanical Separated Meat (MSM) in the diets were not high enough as to depress growth performance of rabbits.

CHAPTER THREE

3.0 MATERIALS AND METHODS

3.1 Experimental Site

This study was carried out in the Department of Biological Sciences, Ahmadu Bello University, Zaria. Zaria is located in the northern Guinea savannah, with an altitude of about 650m above sea level. It lies between latitude 10°N and longitude 7°E, with an average rain fall of 1100mm which starts from late April and early May to mid-October (Meteorological Unit, IAR, 2010).

3.2 Source and Processing of African Locust Bean Seeds

The African locust bean seeds were purchased from retailers in a local market in Zaria, Nigeria. The dry bean seeds were stored at room temperature (25 °C) until ready for use. The raw bean seeds were prepared by depulping, dehulling, dehydrating, and defatting as described by Ikenebomh and Kok (1984). The sample (1.0 kg) was soaked in 5l of tap water for 12 h to soften the adhering pulpy materials. The pulp was removed by rubbing the seeds between the palms and washing them with water. The depulped cleaned seeds were dried during dry season at room temperature of 25°C for 48 h and dehulled to free them from the dark brown testae as follows: Tap water was added to the sample in a flask to give seed: water ratio of 1: 5 (w/v) and boiled on a hot plate for 8 h. The evaporated water was replaced every 2h in order to keep the seeds covered. The content was allowed to cool to 28±2°C and excess water was drained. The testae were removed by rubbing the seeds between the palms and washing with water. The seeds free of the testae were referred to as dehulled bean seeds. They were hydrated further by boiling with water for 30 min, cooled and drained. The dehulled and hydrated bean seeds were referred to as processed substrate. This was allowed to ferment by solid substrate fermentation which involved weighing 25 g of processed substrate into a flat tray, the bottom of which was lined with a sterile

filter paper. The processed substrate was then covered with another sterile filter paper and the Petri dish covered to form the fermentation unit which was incubated at 37°C for 72 h to obtain the fermented bean seeds.

3.3 Experimental Animals

A total of 20, six weeks old Newzealand white cross bred weaner rabbits were used in this study with an average weight of 634.30g. The rabbits were purchased from National Animal Production Research Institute, Shika, Zaria. The rabbits were individually caged and fed the four experimental diets. Each treatment was replicated five times in a completely randomized design. Treatment 1 was the control diet and rabbits on treatments 2, 3 and 4 were fed diets containing raw, boiled and fermented African locust bean seeds. Prior to the commencement of the study, the cages were disinfected, and the animals were dewormed. The animals were also given the respective experimental diets three days to the commencement of the trial to get adjusted to the feed. Feed and water were given *ad libitum*.

3.4 Experimental Design and Diets

Raw, boiled and fermented African locust bean seed was used at 12% dietary level in weaner rabbits' diet. The diets were used in a completely randomized design designated T₁, T₂, T₃ and T₄. They were formulated to meet the rabbit nutrient requirement according to NRC (1995) in Table 3.1.

Table 3.1: Composition of Experimental Diets Containing Locust Bean Seed (%)

Ingredients	T1 (Control)	T2 (Raw)	T3 (Boiled)	T4 (Fermented)
Maize	56.00	50.00	50.00	50.00
African locust bean seed	-	12.00	12.00	12.00
Soybeans meal	10.00	7.00	7.00	7.00
Fish meal	5.00	5.00	5.00	5.00
Groundnut cake	15.50	12.50	12.50	12.50
Rice bran	12.00	12.00	12.00	12.00
Bone meal	3.00	3.00	3.00	3.00
Vit. Premix	0.25	0.25	0.25	0.25
Table Salt	0.50	0.50	0.50	0.50
Methionine	0.10	0.10	0.10	0.10
Lysine	0.15	0.15	0.15	0.15
Total	100.00	100.00	100.00	100.00
<hr/>				
Calculated Analysis				
M. E.	2980.50	2980.50	2980.50	2980.50
Crude Protein	18.02	18.12	18.25	18.36
Crude Fibre	8.95	10.71	8.76	7.94

3.5 Determination of Proximate Composition

The Association of Analytical Organization Chemists(2005) methods were used for determination of protein, fiber content, fat, ash, and moisture content, respectively. The analysis was carried out at the Biochemistry laboratory of the Department of Animal Science, Faculty of Agriculture, Ahmadu Bello University, Zaria.

3.5.1 Determination of crude protein content

Protein was determined by Kjeldahl procedure using a protein factor of 6.25. A sample of about 1.2g was weighed into a digestion tube and conc. tetraoxosulphate (IV) acid (conc. H_2SO_4) was added using a dispenser. The tube was placed in a preheated digester at $420^\circ C$ for 30 minutes until a clear solution was obtained. The tube was removed from the digester, cooled and diluted with water and placed in the distillation unit. A conical flask containing 25ml of boric acid (indicator) was placed under the condenser outlet. About 25ml of 40% Sodium hydroxide (NaOH) was dispensed in the flask and distillation carried out for 5 min. The ammonium borate solution formed was titrated with 0.1M tetraoxosulphate (VI) acid to purplish-grey end. Percentage nitrogen (percentage N_2) was calculated.

$$\text{Nitrogen in sample (\%)} = 100[A \times B / C] \times 0.014$$

$$\text{Crude protein (\%)} = \text{nitrogen in sample} \times 6.25$$

Where:

A = chlorhydric acid used in titration (ml)

B = normality of standard acid

C = weight of sample (g)

3.5.2 Determination of lipid content

Lipid (Fat) analysis was carried out using soxhlet extraction method. Twenty-five(25g) of ground sample was mixed with about 100ml of n-hexane. The mixture was vigorously shaken with the separation flask knob opened at intervals to release the accumulated air pressure, which may burst the flask if left there. The fat in the spirit was evaporated to dryness over a soxhlet extraction, which extracts n-hexane from its solution of fat. The fat left behind in the flask was placed in the oven to dry at 105°C for 1½ hours. The round bottom flask was cooled in desiccators and weighed. Percentage of fat in sample was calculated.

$$\text{Crude lipid content (\%)} = 100 [B-A/C]$$

Where:

A = weight of clean dry flask (g)

B = weight of flask with fat (g)

C = weight of sample (g)

3.5.3 Determination of crude fiber content

Fiber content of the sample was measured using the enzyme-modified, neutral detergent fiber (NDF) method of analysis. Dried samples whose fat content were extracted using soxhlet extraction were treated with standard NDF procedures up to the point that fiber-containing residues were filtered and washed with water. The filtered residues were incubated with a porcine α – amylase solution at 37°C over-night. The residue was filtered after incubation, washed very well, and dried. The NDF was calculated as filtered residual.

$$\text{Crude fibre content (\%)} = 100 [A-B/C]$$

Where:

A = weight of crucible with dry residue (g)

B = weight of crucible with ash (g)

C = weight of sample (g)

3.5.4 Determination of ash content

Ash content of the samples was determined by putting 25g of sample in a dish of known weight (W_4) and dried in an oven for 4 hours at 105°C. It was removed, cooled in desiccators and weighed (W_5). The sample in dish was ash in a muffle furnace at 550°C until white or grey ash resulted. It was cooled and reweighed (W_6). The percentage ash content was calculated.

$$\text{Ash content (\%)} = 100 [A-B/C]$$

Where:

A = weight of crucible with sample (g)

B = weight of crucible with ash (g)

C = weight of sample (g)

3.5.5 Determination of moisture content

Moisture content was determined by weighing 25g of sample into cans of known weights (W_1). The samples in cans (W_2) were placed in an oven for 6 hours at 105°C and then cooled in desiccators and reweighed (W_3). Difference in weight was moisture loss.

$$\text{Moisture content (\%)} = 100 (B-A) - (C-A)/(B-A)$$

Where:

A = weight of clean, dry scale pan(g)

B = weight of scale pan + wet sample (g)

C = weight of scale pan + dry sample (g)

3.6 Determination of Anti-nutritional content of African Locust Bean Seeds

Anti-nutritional factors were determined at the Biochemistry Laboratory of the Department of Animal Science, Faculty of Agriculture, Ahmadu Bello University, Zaria.

3.6.1 Determination of trypsin inhibitor activity

Trypsin inhibitor activity of sample was determined by the method of Kakade *et al.* (1974). The digest contained 1.0 g of the sample, 40 µg of trypsin and 2 mg of benzoyl- DL-arginine-P-nitroanilide (BAPA) in Tris buffer. The absorbance of sample was read at 410 nm.

3.6.2 Determination of tannin content

The method of estimation of tannin content in extract by Joslyn (1970) was used for the determination of tannin content in samples. Finely ground sample (0.5 g) was defatted with 5% ethyl ether for 15 min. The tannin in the defatted sample was then extracted with methanol and the absorbance at 760 nm was measured.

3.6.3 Determination of phytic acid

An indirect colorimetric method of Wheeler and Ferrel (1971) was used for phytate determination. This method depends on an iron to phosphorus ratio of 4: 6. Five grams of the test sample was extracted with 3% tri-chloro acetic acid. The phytate was precipitated as ferric phytate and converted to sodium hydroxide. The precipitate was dissolved in hot 3.2 N HNO₃ and the colour read immediately at 480 nm. The standard solution was prepared from Fe (NO₃)₂ and the iron content was extrapolated from a Fe (NO) standard curve. The phytate concentration was calculated from the iron results assuming a 4: 6 iron: phosphorus molecular ratio.

3.7 Growth Parameters Monitored

Parameters monitored and measured on weekly basis were weight gain and feed intake. Feed conversion ratio was calculated as secondary data.

Weight gain was gotten by subtracting the newly measured weight from the initial weight, feed intake was gotten by subtracting feed left in the feeding trough from the known quantity that was served to the rabbits and feed conversion ratio was gotten from dividing feed intake by weight gain using the following formula:

Feed Conversion Ratio = Feed intake/Weight gain

3.7.1 Weight gain

Weight gain is an increase in [body weight](#). This can be either an increase in [muscle mass](#), [fat deposits](#), or excess fluids such as [water](#). Weight gain without increased calorie rich food intake might be a symptom of a serious medical condition. Weight gain may be as a result of imbalance of thyroid hormones which may cause weight gain or weight loss (Nicholaset *al.*, 2008).

3.7.2 Feed intake

This is the food available to animal in its diet. The amount of feed on offer must be sufficient to satisfy appetite and meet production targets. Provide equal access to feed for all cows or rabbit, particularly at troughs or on a feedpad. Feed quality and the palatability of a particular feed are affected by;freshness, mould, spoilage, taste, moisture and temperature (Snowder and Van Vleck, 2003).

3.7.3 Feed conversion ratio

In [animal husbandry](#), feed conversion ratio (FCR), feed conversion rate, or feed conversion efficiency (FCE), is defined as a measure of an [animal](#)'s efficiency in converting [feedmass](#) into

increases of the desired output. For dairy cows, for example, the output is milk, whereas animals raised for meat – such as beef cows, pigs, chickens, rabbits and fish– the output is the mass gained by the animal. Specifically FCR is the mass of the food eaten divided by the output, all over a specified period of time (NRC, 2000).

3.7 Carcass Quality Evaluation

Three rabbits from each treatment were selected based on the average weight per treatment on the eighth (8) weeks of the experiment, fasted overnight and thereafter slaughtered. The heart, liver, kidney, flesh (muscle) and skin were subjected to proximate analysis according to AOAC (2005) methods as stated in section 3.5.

3.8 Statistical Analysis:

Data generated from proximate composition, anti-nutritional factors determination, growth performance and carcass composition of rabbit carcass were analyzed using one-way Analysis of Variance (ANOVA) with model of SAS software (SAS, 2002). Significant differences in means were separated using the Duncan's multiple range test at $p < 0.05$.

CHAPTER FOUR

4.0 RESULTS

4.1 Proximate Composition of African Locust Bean Seeds

The proximate composition is presented in Table 4.1. The Dry Matter (DM) was significantly ($P<0.05$) reduced from the raw and fermented seeds. Fermented seeds also had a significantly ($P<0.05$) higher crude protein (CP) than to the raw and boiled seeds respectively. Crude fibre (CF) was significantly ($P<0.05$) reduced from 11.40 in the raw seeds to 4.50 than in fermented seeds. The boiled seeds had a CF of 9.25. Fermenting locust bean seed also significantly ($P<0.05$) reduced the oil content of the seeds from 10.60 in the raw to 3.78. The boiled had 13.49. Ash and NFE content of the seeds was also reduced by fermentation. Ash was significantly ($p<0.05$) high in the raw seeds than in the boiled and fermented seeds.

4.2 Anti-nutritional Factors Content of African Locust Bean Seeds

The effect of processing on anti-nutritional factor content of African locust bean seed is presented in Table 4.2. It was observed that processing methods used (boiling and fermenting) significantly reduced anti-nutritional factors in locust bean seed. Fermenting was able to significantly ($p<0.05$) reduced tannin level from 409.83 in the raw seeds to 181.82; while the value of tannin after boiling the seeds is 321.50. Saponins was significantly ($p<0.05$) reduced from 207.83 in the raw seeds to 67.83. The value recorded in the boiled seeds is 129.50. Cyanide content of the seeds was significantly ($p<0.05$) reduced from 87.83 in the raw to 27.79 in fermented seeds while boiled seeds recorded 64.50 of cyanide content. Processing locust bean seed reduced oxalate from 181.83 in the raw seeds to 57.87 in fermented seeds, while boiling reduced oxalate to 146.50.

Table 4.1: Proximate Composition of African Locust Bean Seeds

Parameters (%)	Raw	Boiled	Fermented	SEM
Dry Matter	88.11 ^a	81.15 ^b	44.34 ^c	0.05
Crude Protein	26.50 ^c	29.58 ^b	41.54 ^a	0.16
Crude Fibre	11.40 ^a	9.25 ^b	4.50 ^c	0.04
Oil	10.60 ^c	13.49 ^b	15.76 ^a	0.06
Ash	6.44 ^a	4.65 ^b	3.78 ^c	0.17
NFE	44.60 ^a	43.18 ^b	34.29 ^c	0.12

Means with the same superscript along rows were not significantly different ($p \geq 0.05$)

SEM = Standard Error of means

NFE = Nitrogen Free Extract

Table 4.2 Anti-nutritional Factors Content of African Locust Bean Seed

Parameters (mg/100g)	Raw	Boiled (%) Reduction	Fermented	(% Reduction)	SEM
Phytate	409.83 ^a	321.50 ^b (21.55)	181.82 ^c	(55.63)	0.31
Saponins	207.83 ^a	185.50 ^b (10.74)	115.60 ^c	(44.38)	0.38
Tannin	176.50 ^a	129.50 ^b (26.63)	67.83 ^c	(63.74)	0.48
Cyanide	87.83 ^a	64.50 ^b (26.56)	27.77 ^c	(68.38)	0.34
Oxalate	181.83 ^a	146.50 ^b (19.43)	57.87 ^c	(68.17)	0.31

Means with the same superscript along rows were not significantly different ($p \geq 0.05$)

SEM = Standard Error of means

4.3 Growth Performance of Rabbits

The effects of boiled and fermented locust bean seed on the growth performance of weaner rabbits is presented in Table 4.3. The results show that boiling and fermenting locust bean seeds significantly ($P < 0.05$) improved growth performance parameters of the rabbits. Boiling improved feed intake in the rabbits compared to fermented and raw seeds. However, there was improvement in average daily weight gain of the rabbits fed diets with fermented seeds which was similar to that of boiled seeds compared to the control and raw seeds. Rabbits fed diets with the fermented seeds also had a better final weight and feed conversion ratio which was similar to boiled seeds. Diets with raw African locust bean seeds and the control diet had a poor final weight and feed conversion ratio.

4.4 Effect of Boiled and Fermented African Locust Bean Seed on the Proximate Composition of the Rabbit Meat

Table 4.4 shows the effects of boiled and fermented locust bean seed on the proximate composition of rabbit meat (Carcass). Processing methods had significant ($P < 0.05$) effect in all the parameters analyzed for except for crude fibre of flesh, heart, kidney, intestine, liver and lungs which did not show any significant difference. Fermentation was able to improve the dry matter (DM) of the skin, heart, kidney, intestine and the values were similar to boiled seeds. The control diet only improved the DM of the flesh muscle. The raw seeds gave poor dry matter of meat analyzed. Fermenting locust bean seed also improved the crude protein (CP) of the skin, flesh, heart, kidney, intestine, liver and lungs compared to other treatments.

Table 4.3: Effects of Boiled and Fermented African Locust Bean Seeds on the Growth Performance of Rabbits

Parameters	Control	Raw	Boiled	Fermented	SEM
Initial weight (g/wk)	634.30 ^a	635.30 ^a	635.21 ^a	634.20 ^a	0.29
Final weight (g)	1075.00 ^b	1175.00 ^{ab}	1237.50 ^{ab}	1312.50 ^c	54.73
Weight gain (g/wk)	84.91 ^d	110.88 ^c	118.16 ^b	125.09 ^a	1.68
Feed intake (g/wk)	427.63 ^a	442.89 ^{bc}	462.84 ^a	453.60 ^{ab}	5.81
Feed Conversion Ratio (g/wk)	3.60 ^c	3.91 ^c	4.00 ^b	5.04 ^a	0.11

Means with the same superscript along the row were not significantly different ($p \geq 0.05$)

SEM = Standard Error of mean

The results also revealed that the liver and heart had a higher CP (58.94 and 57.90 respectively); this was followed by the kidney and flesh (muscle) which had a CP of 46.57 and 46.92 respectively. The skin recorded a poor CP in all the diets. A value of 14.21 was recorded for the control, while 18.00, 17.11 and 20.11 were values for CP of the skin from animals fed raw, boiled and fermented African locust bean seeds respectively. Crude fibre, was observed to be only present in the skin. The control and raw seeds diets recorded higher values for CF compared to the boiled and fermented seeds. Rabbits that ate diets with raw seeds recorded higher oil content for the skin, flesh, heart, kidney, intestine, liver and lungs. The values were similar to diets with boiled seeds. The control and fermented diets performed poorly with respect to oil being incorporated into the organs. The ash content of the organs was inconsistent. Fermented seeds had high ash content for the skin only while diet with boiled seeds recorded a high ash of the lungs, and intestine. The raw seeds recorded high ash content for the heart, kidney, intestine, and liver. The control diet recorded high ash for the flesh and the liver. The values of nitrogen free extract (NFE) were observed to be higher in the control diets than the rest of the diets containing locust bean seed; the values of the African locust bean seed diets were similar to each other.

Table 4.4: Carcass Composition of the Weaner Rabbit Meat

Parameters (%)	Control	Raw	Boiled	Fermented	SEM
Dry Matter					
Skin	39.33 ^c	40.29 ^b	39.44 ^c	46.76 ^a	0.18
Flesh	25.04 ^a	22.88 ^c	24.26 ^b	23.88 ^b	0.12
Heart	19.12 ^c	20.17 ^b	23.10 ^a	23.95 ^a	0.28
Kidney	24.26 ^b	21.32 ^c	30.93 ^a	31.05 ^a	0.12
Intestine	19.32 ^c	17.96 ^d	20.31 ^b	26.32 ^a	0.18
Liver	28.25 ^b	23.23 ^c	31.44 ^a	28.75 ^b	0.34
Lungs	20.76 ^c	18.33 ^d	32.26 ^a	26.38 ^b	0.20
Crude Protein					
Skin	14.21 ^c	18.00 ^b	17.11 ^b	20.11 ^a	0.18
Flesh	39.96 ^d	43.50 ^c	44.88 ^b	46.92 ^a	0.09
Heart	41.09 ^c	54.99 ^b	55.02 ^b	57.90 ^a	0.18
Kidney	42.5 ^d	45.22 ^c	46.19 ^b	48.57 ^a	0.18
Intestine	21.07 ^d	27.32 ^c	29.85 ^b	32.56 ^a	0.06
Liver	50.02 ^d	55.21 ^c	56.19 ^b	58.94 ^a	0.16
Lungs	33.56 ^c	34.03 ^c	35.36 ^b	36.13 ^a	0.17
Crude Fibre					
Skin	0.74 ^a	0.60 ^{ab}	0.48 ^b	0.31 ^c	0.05
Flesh	0.00	0.00	0.00	0.00	0.00
Heart	0.00	0.00	0.00	0.00	0.00
Kidney	0.00	0.00	0.00	0.00	0.00
Intestine	0.00	0.00	0.00	0.00	0.00
Liver	0.00	0.00	0.00	0.00	0.00
Lungs	0.00	0.00	0.00	0.00	0.00

Means with the same superscript along rows were not significantly different ($p \geq 0.05$)

SEM = Standard Error of mean

Table 4.4 Continues...

Parameters (%)	Control	Raw	Boiled	Fermented	SEM
Lipid					
Skin	1.56 ^c	2.09 ^a	1.82 ^b	1.56 ^c	0.05
Flesh	5.46 ^a	4.19 ^b	5.09 ^a	3.35 ^c	0.14
Heart	5.16 ^a	3.14 ^c	4.62 ^b	3.25 ^c	0.17
Kidney	4.12 ^c	6.39 ^a	5.67 ^b	4.21 ^c	0.14
Intestine	4.31 ^b	5.09 ^a	4.31 ^b	3.51 ^c	0.11
Liver	3.33 ^b	4.17 ^a	4.14 ^a	3.54 ^b	0.06
Lungs	2.29 ^b	2.64 ^{ab}	3.01 ^a	2.60 ^{ab}	0.13
Ash					
Skin	3.40 ^d	4.46 ^c	5.48 ^b	7.29 ^a	0.12
Flesh	12.99 ^a	11.31 ^c	11.52 ^c	12.16 ^b	0.08
Heart	3.17 ^c	14.44 ^a	13.31 ^b	13.56 ^b	0.20
Kidney	10.33 ^c	14.18 ^a	12.48 ^b	10.47 ^c	0.16
Intestine	11.58 ^b	13.18 ^a	13.31 ^a	11.39 ^b	0.08
Liver	15.23 ^a	15.16 ^a	12.39 ^b	12.38 ^b	0.08
Lungs	6.69 ^c	7.47 ^b	8.74 ^a	7.69 ^b	0.18
NFE					
Skin	79.58 ^a	74.46 ^b	74.24 ^b	70.03 ^c	0.17
Flesh	41.08 ^a	38.42 ^b	38.25 ^b	37.27 ^c	0.17
Heart	40.59 ^a	26.38 ^b	26.25 ^b	25.52 ^c	0.16
Kidney	43.07 ^a	34.35 ^c	34.59 ^c	36.54 ^b	0.16
Intestine	62.58 ^a	53.11 ^b	53.03	51.79 ^b	0.42
Liver	31.21 ^a	24.19 ^c	24.54 ^c	25.30 ^b	0.17
Lungs	56.53 ^a	52.32 ^c	52.70 ^c	53.51 ^b	0.23

Means with the same superscript along rows were not significantly different ($p \geq 0.05$)

SEM = Standard Error of mean

NFE = Nitrogen Free Extract

CHAPTER FIVE

5.0 DISCUSSION

5.1 Effect of Boiling and Fermentation on the Proximate Composition of African Locust Bean Seed

The increase in protein content obtained in this study during fermentation agreed with other reports on African locust bean seeds carried out in southern part of Nigeria (Ikenebomeh *et al.*, 1986; Omafuvbe *et al.*, 2004). The values were similar to the values that were reported by Omafuvbe *et al.* (2004). The difference in the levels of protein content obtained in both studies may be attributed to differences in cultivars of *P. biglobosa* studied and experimental procedures used. Though crude fiber contents of the sample was observed to reduce by boiling and fermentation, this could be that during boiling the bonds binding the cellulose, hemicelluloses and lignin together, may have been weakened (Ugwuowo and Ani, 2011). Reduction in fibre was very obvious after fermentation; this may be attributed to the degradation done by microbes during fermentation by converting the fiber to volatile fatty acids for their nutrition (Akpet *et al.*, 2012). The value (11.40%) for the raw seeds was similar to the value (11.2%) reported by Aniet *al.* (2005).

The value of ether extract (lipid) obtained in this study is in agreement with the values reported by Esenwah and Ikenebomeh (2008). They reported that soaking and boiling, led to increase in ether extract in the locust bean seed which was further increased by fermentation at 72 h. The increase in ether extract obtained in this study is in agreement with the findings of Omafuvbe *et al.* (2004), Addy *et al.* (1995) and Ikenebomeh (1986) for locust bean seed. Soaking and boiling of the sample might have led to the cleavage of the protein-lipid or carbohydrate-lipid linkages thereby, facilitating the easy extraction of the oil by the extracting solvent. Processing technique

may also contribute to the disparity in values obtained by various researchers. Leaching of oil in processing water can also lead to a reduction in the overall oil content of the seed after processing. Loss in ash may be due to leaching of soluble inorganic salts into the processing water during the boiling of the seeds. A similar result was reported in Ibadan, Nigeria for *Dolichos lablab* bean seeds (Osman, 2007). The nitrogen free extract (NFE) gives a rough estimate of the carbohydrate content present in a material. In this study, it was observed that NFE reduced after boiling the seeds and further reduced on fermentation. This result is in agreement with results of earlier workers (Addy *et al.*, 1995; Omafuvbe *et al.*, 2004; Osman, 2007). Loss in carbohydrate during soaking and boiling may be due to leaching of soluble carbohydrates like sugars into the soaking and cooking water; while loss in carbohydrate during fermentation may be as a result of the utilization of some of the sugars by fermenting organisms for growth and metabolic activities.

5.2 Effect of Boiling and Fermentation on the Anti-nutritional Factors Content of African Locust Bean Seed

In all the anti-nutritional factors examined, boiling and fermentation resulted in decreased levels of anti-nutritional factors. The reduction of tannin in this study by boiling of the locust bean seeds may be due to its solubility in water and its sensitivity to heat during boiling (Bagepalli *et al.*, 1992). Similar reduction in tannin content was recorded during processing of legumes by other workers. Igboeli *et al.* (1997) reported 35.9 to 43.6% reduction in tannin content during the processing of baobab seeds (*Adansonia digitata*) by dehulling, cold water, hot water, hot alkali and acid treatments. El-Adawy (2002) reported 48.0% to 50.1% reduction during processing of chick pea (*Cicer arietinum* L.) by different cooking methods. Mbajunwa (1995) observed that during the processing of African oil bean seeds, *Pentaclethra macrophylla* (Benth.), cooking of

the seed led to 51.6% reduction in the tannin content while fermentation further reduced the tannin content to 56.8% total reduction. Mbajunwa (1995) in a study on the effect of processing on some anti-nutritional and toxic components and on the nutritional composition of African oil bean seed *Pentaclethra macrophylla* Benth reported 11.2 g kg⁻¹ of phytic acid in the raw sample and 2.7 g kg⁻¹ in the fermented sample; which represented 75.9% decrease in phytic acid in the fermented product. The reduction in phytic acid content in this study was similar to 41.27% reported by Adegbulu (2004) for phytic acid after boiling African locust bean seeds, and lower than the 79.29% recorded by Bawa *et al.* (2003) for lablab seeds. The reduction of the cyanide content obtained for boiled and fermented locust bean seeds was similar to the reports of Okolie and Ugochukwu (1989). They observed that the processing of some Nigerian legume seeds caused a drastic reduction in HCN. The study showed that boiling and fermentation was effective in reducing the anti-nutritional factors in African locust bean seeds.

5.3 Effect of Boiled and Fermented African Locust Bean Seed on the Growth Performance of Rabbits

The increase in feed intake observed in this study could be attributed to the aroma and improved palatability of the diets after boiling the locust bean seeds. The pungent smell of the fermented locust bean seeds which might have masked the smell of other ingredients could have been responsible for the slight reduction in feed intake noticed in animal fed with fermented locust bean seed diet. Hence if fermented locust bean seeds are in excess in a rabbit diet, they can make the diet unattractive and unpalatable to the rabbits (Akpet *et al.*, 2012). Abubakar and Yusuf (1991) reported a similar observation when they fed rumen content to broilers. This finding is similar to the earlier report of Odunsi (2003). He reported that feed intake increases in animals if the aroma of their diet is pleasant. This could be because some animals detect their feed through

smell (Akpet *et al.*, 2012). The improved average daily weight gain observed in rabbits fed with fermented and boiled locustbean seeds could also be due to reduction in anti-nutritional contents that would have inhibited the availability of nutrients. Fermented locust bean seeds had given the best weight gain maybe because of the improvement in nutrient quality of the seeds by microbes thereby making the nutrients to be readily digestible (Anhwange *et al.*, 2004). The values of weight gain obtained in this study are similar with the finding of Akpet *et al.* (2012). The final weight and feed conversion ratio in this study are reflections of their average daily weight gain.

5.4 Effect of Boiled and Fermented African Locust Bean Seed on the Proximate Composition of the Rabbit Meat

The trend in proximate composition of the various organs of rabbits in this study is a reflection of the proximate composition of the fed material (locust bean seed). It should be recalled that the results of the proximate composition of raw, boiled and fermented locust bean seed revealed that fermented locust bean seeds had high crude protein, low crude fiber and ash. This trend was also reflected in the carcass of the rabbits fed the diets. In general, internal organs had a higher crude protein than the flesh (Muscle). The skin had a poor CP which may be due to inability of the skin to store it which is similar to that reported by Odunsi (2003) of feeding poultry using lablab leaf. Raw seeds heavily affected the heart, liver, lungs and kidney. This was observed by the high ash content these organs recorded. Ash tends to estimate the mineral content present in a material. The diets with this raw seeds may have had high anti-nutritional factors which may have been reflected in the form of ash. The reasons for the general decline in protein and ash in the flesh (muscle) of the rabbit carcass could be due to the relative high moisture content present in flesh compare to the moisture content of internal organs (liver, heart, kidney etc). Protein concentration in meat increases as moisture content decreases (Aduku and Olukosi, 2000, Apata

et al. 2009). High NFE in meat of rabbits noticed in the control group, may be attributed to the delay in the onset of glycolysis in the rabbits carcasses which could have started earlier in rabbits carcasses on diets with locust bean seeds. Thus, depleting the glycogen reserves more quickly than the rabbits in the control group whose carcasses might have attained lower pH within a short period of time that led to pre-rigor observed in this study. This result agrees with the reports of Apata *et al.* (2009) in which they recorded similar results in their study.

CHAPTER SIX

6.0 SUMMARY, CONCLUSIONS AND RECOMMENDATIONS

6.1 Summary

Fermentation improved the nutrient composition of the seeds as was observed in the proximate composition of the seeds. Anti-nutritional factor analysis revealed that raw locust bean seeds have high tannin, saponins, cyanide, oxalate and phytic contents. These elements are capable of retarding growth in rabbits by impairing digestion, and subsequently lead to mortality. Processing African locust bean seeds was found to reduce these anti-nutritional factors. Fermentation as a processing method showed to be better than boiling because the anti-nutritional factors were more reduced in the fermented seeds. On the feeding trials, rabbits that consumed diets with fermented African locust bean seeds performed better in terms of average daily weight gain, final weight and feed conversion ratio. However, boiling African locust bean seed improved palatability of the diet as animals on the diet consumed more feed compared to other treatments. Results of proximate composition revealed that locust bean seed can be used as a protein source in rabbit diets. The proximate composition of the meat was a reflection of the nutrient composition of the seeds. Meat from rabbits on the diet with the fermented seeds had high crude protein, low or no fibre and low ash. Meat from rabbits on the raw seed diet had high ash content in the liver, heart, kidney and lungs. This could be a reflection of the effect of anti-nutritional factors which could have reduced the synthesis of some food nutrients present in the diet.

6.2 Conclusions

It was therefore concluded that locust bean seeds can be fed to rabbits preferably when processed. Processing African locust bean seeds significantly ($p < 0.05$) reduced the anti-nutritional

factors and also improves the nutrients in the seeds. Fermenting seeds of the African locust bean seeds increased more crude protein for utilization than that obtainable in the raw seeds. Feeding processed African locust bean seeds does not adversely affect the rabbits as there was improved feed intake, weight gain, final weight and feed conversion ratio of rabbits on these diets. Processed African locust bean seeds also improve carcass quality of rabbits.

6.3 Recommendations

From the above results, the following are recommended:

- Fermentation of the African locust bean seeds is recommended over boiling and raw in the formulation of diets for rabbits
- Further research on the effect of raw and processed African locust bean seed on histopathological effect on rabbits internal organ is recommended as it will help in ascertaining the effect the raw and processed seeds have on these internal organs.

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