EFFECT OF PROCESSING ON THE NUTRITIONAL VALUES OF LOCUST BEAN SEED

BY

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BEING A FINAL YEAR PROJECT SUBMITTED IN PARTIAL FULFILMENT OF THE REQUIREMENTS FOR THE AWARD OF BACHELOR OF ENGINEERING (B. ENG.) DEGREE IN AGRICULTURAL AND BIORESOURCES ENGINEERING FEDERAL UNIVERSITY OF TECHNOLOGY, MINNA

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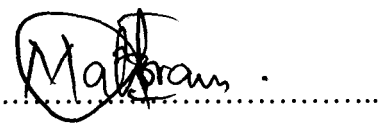
DECLARATION

I hereby declare that this Project is a record of research work that was undertaken and written by me. It has not been presented before for any degree or diploma or certificate at any University or Institution. Information derived from personal communications, published and unpublished works of others were duly referenced in the text.



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CERTIFICATION

This Project entitled "The Effect of Processing on the Nutritional Values of Locust Bean" by Mahmood Babatunde Ibrahim meets the regulations governing the award of the degree of Bachelor of Engineering (B. ENG.) of the Federal University of Technology, Minna, and it is approved for its contribution to scientific knowledge and literary presentation.

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This project is dedicated to Almighty Allah, the most gracious and the most merciful; my beloved brother and sisters and my parents Alh. Ibrahim Mahmood and Alhj. Mahmood Salimat.

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ABSTRACT

Locust bean seed (*Parkia biglobosa*) is a grain legume found growing in wide savannah area of West and Central Africa. It is fermented and consumed as a condiment which is added to soups and stews to enhance the flavour and improve the nutritional values. The seeds were boiled and soaked for 12 hours and 8 hours respectively and re-boiled for 1 hour with the addition of soften agent "*kuru*" and were later subjected to fermentation for 24 hours. The raw and fermented locust bean seeds were taken to laboratories for proximate analysis, bulk density and pH determination. From the results of the nutritional analysis it was found that *dawadawa* or *iru* is richer in crude protein (38.5%) and fat (21.172%) but deficient in crude fibre (2%) and ash (1.8%). This is advantageous over the raw locust bean because the less the crude fibre content the fewer waste products will be passed out. The moisture content (35.21%) is higher which implies short period of storage and easily prone to the attack of microorganisms. The results obtained for pH (6.25%) and some minerals such as: Magnesium (7.67ppm), copper (2.5ppm), iron (1.118ppm) and sodium (19.575ppm) are lower in concentration which needed by body in few quantities. Higher concentration of potassium (63.04ppm) and zinc (6.293ppm) with no lead (0.00ppm) concentration were obtained compared to that of raw locust bean. This study has shown that there was a great influence of processing on the raw locust bean to *dawadawa* when compare the level of difference. This study equally shown that the fermented locust bean "*dawadawa*" is highly nutritious and can be used as a condiment in stew and soup also can be used as substitute for animal protein.

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CHAPTER ONE

1.0 INTRODUCTION

1.1 Background to the Study

The high cost of animal protein has directed interest towards several leguminous seeds. Among the plant species, grain legumes are considered as the major sources of dietary protein. They are consumed world wide, especially in developing and under-developed countries where consumption of animal protein is limited as a result of economic, social and cultural factors (Ikenebomeh and Esenwah, 2008).

Seeds of legumes may account for up to 80% of dietary protein and may be the only source of protein for some groups. Their cooked forms are eaten as meals and commonly used in fermented form as condiments to enhance the flavour of food (Odunfa, 1985a; Aidoo, 1986; Achi, 1992). With high contents of protein, legume condiments can serve as a tasty complement to sauces and soups and can substitute for fish or meat.

Locust bean seed (*Parkia biglobosa*) is a grain legume found growing in wide savannah area of West Africa. It is fermented and consumed as a condiment which is added to soups and stews to enhance the flavour and improve the nutritional values as a result of its high protein content (Odunfa, 1985a). Odunfa (1981a) stated that the fermented locust bean seed is used and consumed in Ghana, Nigeria, Togo and also in savannah of West and Central Africa. The seeds of locust bean are referred to by different local names peculiar to each locality. It is called *"iru" in* Yoruba, *"dawadawa"* in Hausa, *"ogiri-igala"* in Igbo. It is also referred to as *"kinda"* in Sierra-leone and *"kpalugu"* in Ghana.

Fermented food spices have remained popular among Nigerians especially now that the industrial food flavouring products such as curry and thyme are costly beyond the reach of many people. Soups prepared with "*iru*" are preferred in Yoruba region because they produce better taste and flavour, and also it is an important protein supplement generally in Nigerian diet (Ogunbunmi and Bashir, 1980).

1.2 Description of African Locust Bean

African locust bean belongs to the family *leguminosae.* It is a large tree with a very broad stem, supported by a trunk usually short and thickset. There are two botanically related species: *Parkia biglobosa* occurring in the Sahel, and *Parkia clappertoniana* occurring typically in wet savannah regions. The leaves are compound, and the leaflets themselves are composed of many simple, smaller leaflets about 1.5 centimetres long and 0.5 centimetres wide. The leaflets are sub-rectangular with rounded tips. The fruits are long with black seeds embedded in yellow pulp (Dupriez, 1992).

The locust bean is perennial with large pods as fruits. The pods bear both the pulp and the seeds. The dry, yellow powdery pulp is rich in sweet carbohydrate and can be mixed with cereals, meat or soup (Adewakun, 1988).

The seeds are extremely hard and practically inedible, but can be processed and fermented into a palatable product called "*dawadawa*": This is the most important food condiment in the entire savannah of West and Central Africa (Odunfa, 1981a).

1.3 Statement of the Problem

Since it has been noticed that there is a rapid reduction in the demand for animal protein in under-developed and developing countries as a result of economic, social and cultural factors (*i.e.* the rate of consumption of animal protein is low), and since protein

and other dietary nutrients are highly needed by the human body, there is need for a substitute for animal protein such as *"dawadawa"*. This condiment is added to soups and stews to enhance flavour and improve their nutritional contents.

1.4 Objectives of the Study

The objectives of this study are:

1. To determine the nutritional contents of unfermented locust bean *i.e.* the raw locust bean.
2. To determine the nutritional contents of fermented locust bean *i.e.* *"dawadawa"* or *"iru"*.
3. To determine the physicochemical changes during fermentation process.

1.5 Justification of the Study

The justification of this study lies on the need to have knowledge of nutritional contents, organoleptic (flavour) and physicochemical changes of fermented locust bean during fermentation process. Information provided in this study will help the processors and consumers of locust bean to know the economic importance, palatability and benefits one can derive from fermented locust bean.

1.6 Scope of the Study

The scope of this study is limited to the effect of processing on the nutritional contents of locust bean. The nutritional contents to be assessed are: Moisture content, crude fibre content, lipid content, crude protein content, ash content, carbohydrate content, minerals content, bulk density and pH value of locust bean. These are based on the availability of laboratory equipment and reagents.

CHAPTER TWO

2.0 LITERATURE REVIEW

2.1 History of Locust Bean

The history of fermented locust bean can be traced back to several centuries when locust bean trees were traditionally planted around communities. A Scottish surgeon, Mongo Park encountered locust bean tree when he explored the Niger Basin from 1795-1799. He described this tree in his book, "Travels in the interior district of Nigeria" (Eka,1980). The locust bean tree was named *parkia biglobosa* after him by Robert Brown in 1826 (Anon., 1981).

Odunfa (1986) stated that, *dawadawa,* a West African fermented meat substitute and flavour prepared from the seeds of African locust bean, also has its name originated from the Arabic language. Basically, the word *'dawa'* means medicine or cure in Arabic language, but in Sudan, it means a spice. *Dawa-mulah* is a mixture of a particular assortment of spices used to flavour sources and relishes for sorghum porridge or bread (Hamid, 1994).

A large traffic of *dawadawa* reportedly entered Sudan from northern Nigeria via the border town of Geneina of Darfur region, traveling all the way across the country to Sudan port on the red Sea. *Dawadawa* enters Sudan under the name of its next of kin, *"kawa"* in order to avoid misunderstanding at the custom checking points. This instance provides a strong connection between the people of Sudan and the people of West Africa. As a corollary of this, it is suggested here that, a survey should be carried out from the Nile to West Africa to find out the extent of these meat substitutes in the region and to assess their role in nutrition (Hamid, 1994).

2.2 Characteristics and Taxonomy of Locust Bean

Throughout Nigeria, many names are applied to the multitude of fermented food condiments. Table 2.1 shows the variety of names by which the products are known in different parts of the country. The exact origin of such names could be attributed to the region or area of manufacture, type of legume or oil seed used and spelling according to region or area.

The Yorubas of the southwestern Nigeria locally call fermented condiments "*iru*" while the Hausas who inhabit most of the northern part of Nigeria call it "*dawadawa*", "ogiri" is the name used by the Ibos of the southeastern Nigeria. "*Owoh*" on the other hand, is its popular name among the Urhobos and Itsekiris in the Niger Delta region. Similarly, "*okpiye*" is the popular name among the Igala and Idoma people of the Middle Belt region (Achi, 2005).

The conventional substrates for condiment production are diverse and each can be produced from more than one raw material. Almost any edible plant material can be subjected to fermentation. Judging by the available literature, over nine different fermented products are condiments. A list of these substrates is given in Table 2.1

Table 2.1: Traditional Substrates used for Food Condiments

Raw material	**Local name**	**Botanical name**	**Reference**
Soya bean	*Dawadawa*	*Glycine max*	Popoola and Akueshi, 1984
Melon seed	*Ogiri*	*Citrullus vulgaris*	Odunfa, 1981b
Castor oil seed	*Ogiri-igbo*	*Ricinus communis*	Odunfa, 1985b
Fluted pumpkin seeds	*Ogiri-ugu*	*Telferia ocidentalis*	Barber and Achinewhu, 1992
African locust beans	*Dawadawa (iru)*	*Parkia biglobosa*	Odunfa, 1981b
African oil beans	*Ugba/Ukpaka*	*Pentaclethra macrophylla*	Obeta, 1983
African yam beans	*Owoh*	*Stenophylis stenocarpa*	Ogbonna *et al.*, 2001
Cotton seeds	*Owoh*	*Gosspium hirsitium*	Sanni and Ogbonna, 1991
Bambara nut	*Dawadawa*	*Vigna subterraanea*	Barimalaa *et al.*, 1989

Source: Achi, 2005

2.3 Planting of Locust Bean

The foliage of the African locust bean does not give much shade. The locust bean tree is frequently planted with seeds in the main cereal fields or in orchards where it forms the uppermost canopy. The seeds needed for planting should be collected from freshly falling pods from strong and healthy trees. Viability is short, so it is best to plant the seeds as soon as possible (Dupriez, 1992).

2.4 Harvesting of Locust Bean

The first commercial fruits can be harvested after about 5-7 years of planting. The pods take about 6-8 months to mature, turning from green to chocolate brown in dry season. The matured pods containing a yellow, dry and powdery pulp can be harvested by knocking them off with long poles, preferably aimed at the pods (Ikenebomeh and Kok, 1999). The pods can also be picked from the ground after they have been blown down by heavy winds. The African locust bean tree is commonly found in savannah region and it is usually harvested around the month of March (Odunfa, 1981b).

2.5 Relationship between Locust Bean and other kinds of Grains

Fermented locust bean *"dawadawa"* is produced in large quantities throughout West Africa, extending eastwards to neighbouring Chad. In some areas in West Africa, such as parts of northern Nigeria, the product is the single largest source of protein after sorghum and millet in average diet of the inhabitants (Odunfa, 1986).

Groundnut, cowpea, soya bean, pigeon pea, cotton seeds and others belong to the "papilionaceae" family but the African locust bean *"Parkia biglobosa"* belongs to the *"mimosaceae"* family, but the two families belong to the group of *"leguminosae"* (Vickery and Vickery, 1979).

Cowpea: The cowpea plant is known botanically as *Vigna unguiculata* and it belongs to the family *leguminosae*. It probably originated in Central Africa, but it is today widely grown in many tropical parts of the world. The plant is an annual, which requires 12-15 weeks from planting to harvesting period (Onwuemeh, 1979). Cowpeas are savannah crops; they thrive where temperature is above 20^0C and where annual rainfall is low, about 750mm. These legumes are of major importance in African agriculture; they are propagated from seeds by putting three or four seeds in holes 3cm deep and 30cm apart (Mayhew and Penny, 1988).

Groundnut: The groundnut is a leguminous plant. It originated in Brazil, from where it was introduced into West Africa by Portuguese traders. The plant is an annual, and the seeds are germinated in about 5 days after planting; flowering occurs in 1 month, and the crop is ready for harvest 4-5 months (Ónwuemeh, 1979).

2.6 Processing of Locust Bean

The matured pod contains a yellow, dry and powdery pulp in which dark brown or black seeds are embedded. The seeds need to be processed so as to improve its flavour, texture, colour and removing anti-nutritional contents such as: *Trypsin* Inhibitor Activity (TIA), tannin, phytic acid (Sarkar *et al.*, 1993).

The seeds are removed by pounding the pods gently in a mortar and sieving to reduce the powdery pulp. The seeds are boiled for about 8-hours to soften the *testa* or seed coat. The boiled seeds are repounded in the mortar to remove the softened *testa.* Sand or wood ash may be added to aid the removal of the *testa* (Odunfa, 1981a). The seeds could be rubbed against the wall of a basket or between the palms (Campbell-platt, 1980).

The cotyledons are then washed thoroughly with clean water and the *testa* separated by using a basket, special earthen pot or calabash as sieves. The washed bean cotyledons are further boiled for 1-2 hours and a softening agent *"kuru"* in Yoruba, *"kanwa"* in Hausa and *"akawa"* in Ibo may be added during second boiling to aid in softening of the bean cotyledons, if a softer variety is desired (Campbell-platt, 1980).

The softened hot bean cotyledons are spread in clean calabash trays up to 4cm in depth, the seeds can also be placed in earthen ware placed in a shallow heap or in a hole in the ground and lined with leaves. The calabash trays containing the cooked bean cotyledons are covered immediately and wrapped in 2-3 layers of jute sacks or cotton cloths to provide warmth and humid conditions. The fermenting locust beans may be covered with leaves (Campbell-platt, 1980). The flow chart for processing locust bean into *dawadawa* is shown in Figure 2.1

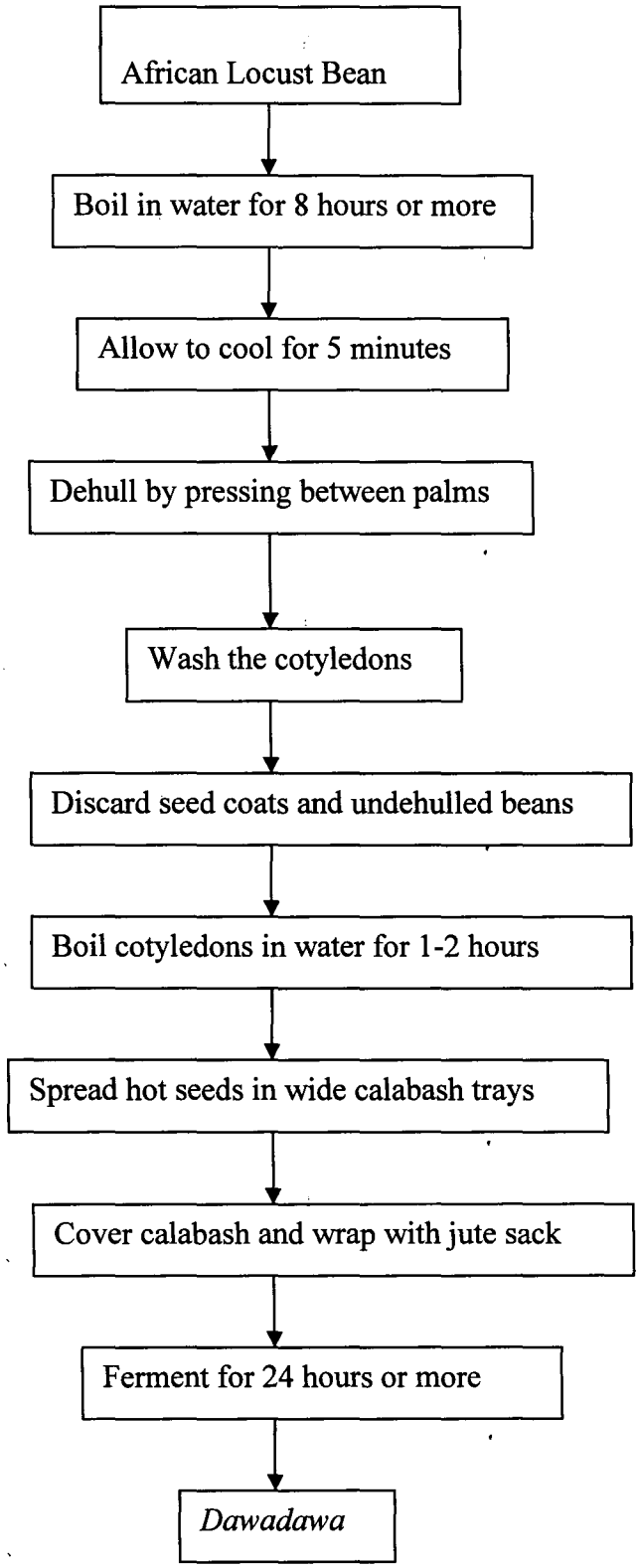


Fig. 2 .1: Flow Chart for Processing Locust Bean into *Dawadawa*

Plate 1: Removing the yellow powdery pulp

Plate 2: Boiling in water for 12 hours

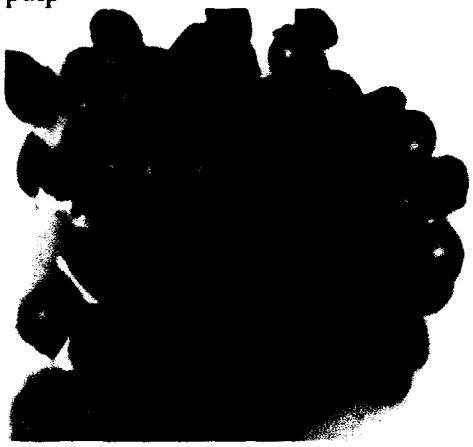


Plate 3: Soaking in water for 8 hours

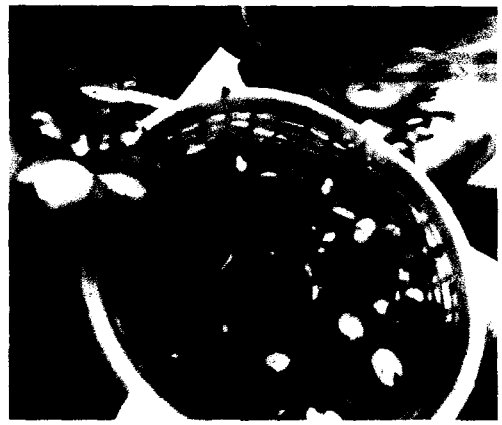


Plate 4: Confirming whether the testa can be removed easily or not

Plate 5: gently repounding the seeds to remove the testa

Plate 6: Addition of River-bed sand for easy removal

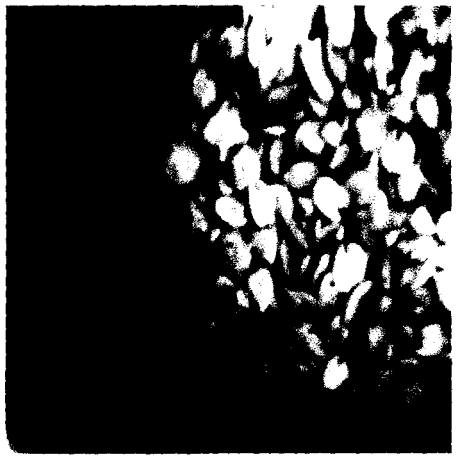


Plate 7: Separating the testa from the cotyledons

Plate 8: Washed and cleaned bean cotyledons

Plate9: Washed bean cotyledons were further boiled

Plate 10: The hot and soften bean cotyledons were spread in a clean calabash

Plate 11: The calabash was covered with several cloths for fermentation

Plate 12: The fermented locus beans *"Dawadawa"*

2.7 Fermentation of Locust Bean into *Dawadawa*

Fermentation is one of the oldest methods of food preservation known to man. In Africa, the art of fermentation is widespread including the processing of fruits and other carbohydrate sources to alcoholic and non-alcoholic beverages, the production of sour t.aste of fermented cereal products *e.g.* "Ogi" in Yoruba, which provides instant energy in breakfast and oil seeds such as African locust bean, melon seed and castor oil seed (Adewusi *et al.,* 1992).

The preparation and preservation of foods by fermentation processes are dependent upon the production by certain micro-organisms, some chemical and physical changes which alter the appearance, body and flavour of the original material. These changes may improve the inhibition to the growth of undesirable micro-organisms.

The micro-organisms that ferment foods to produce desirable changes can be distinguished from those that are responsible for spoilage and food-borne disease. (Ihekoronye and Patrick, 1985).

Fermentation occurs when microorganisms are allowed to break down the carbohydrate to form acids in a limited oxygen environment. It is usually carried out in a moist solid-state, the hot seeds provide the moisture that increases the initial humidity for fermentation. The cotyledons of locust bean are left to ferment for 24-hours or longer depending on the locality. In southwestern Nigeria, the Yorubas allow up to 2 days fermentation whereas in Ghana, it may be up to 4 days. Fermentation is terminated by removing the jute cover from the tray and exposing the fermented beans to air (Campbell-platt, 1980).

During fermentation period, the bean cotyledons change colour from light brown to dark brown and become softer, thus edible.

2.8 Nutritional Contents of Locust Bean

The nutritional contents of locust bean are:

2.8.1 Moisture Content

Moisture content is the ratio of the mass of water present in a sample to the mass of sample after it has been dried to a constant weight or is the volume of water present in a unit volume of sample. Moisture content is usually a dimensionless ratio of two volumes or masses expressed as percentage.

The state of water activity in food is described by the relationship between moisture of the product and the relative humidity of the air surrounding it. The ratio of these two parameters is called "water activity" (a_w). The relative humidity corresponding to each specific moisture content of the product is called "equilibrium relative humidity" (Ihekoronye and Patrick, 1985).

2.8.2 Crude Fibre Content

Crude fibre is that portion of the plant material which is not ash or which dissolves in boiling solution of 1.25% H_2SO_4 or 1.25% NaOH. Crude fibre was originally thought to be indigestible portion of any main food. It is known however that fibre consists of cellulose which can be digested to a considerable extent by both ruminants and non-ruminants. The interest in fibre in food and feed has increased, based on the noticed number of serious illness associated with diet low in fibre (Ibitoye, 2005).

Fibres swell and form gelatinous mass with high water retention capacity with the digestive system. Findings show that fibre product can absorb cholesterol, toxic agents and raise the excretion of bile acids and sterols (AOAC, 1980).

2.8.3 Lipid Content

Lipid (fat and oil) is the next predominant nutrient to water and carbohydrate in diet. Some of the dietary sources of these nutrients are readily identified as visible fats and oils. Lipids may be classified as fat and oil in solid and liquid state respectively at room temperature. The primary one being a *triglyceride*, a *trigly* contains 3 fatty acids that are esterified to the 3 hydroxyl alcoholic-glycerol.

Fatty acid has a general formula: R-COOH. R-group contains carbon and hydrogen in it, if the carbon atoms are bonded together with a single bond **C-C,** the compound is saturated, if the carbon atoms are bonded together with double bonds **C=C,** the compound is unsaturated. The fatty acids that are unsaturated can react with Oxygen to reproduce undesirable flavor (Ihekoronye and Patrick, 1985).

From the available literature on the nutritional composition of *dawadawa* and locust bean, it shows that *dawadawa* has high level of lipid contents. Therefore, the lipid has been found to be of high nutritional values because of its high content of polyunsaturated fatty acids including the essential fatty acids. The high content of poly unsaturated fatty acids of *dawadawa* and locust bean including *linoleic* acid is also of value in the regulation of serum cholesterol and triglycerides and hence can help in reducing or preventing coronary heart diseases, *hypelipemia* and hence atherosclerosis (Ikenebomeh and Esenwah, 2008).

2.8.4 Protein Content

The word protein is derived from Greek meaning "holding the first place". Protein literally holds the first place in the architecture of all living things, without protein no life can exist, no plant can grow or trap sunlight. The protein content of locust bean is about 30-40% which is relatively high and can therefore serve as good source of protein.

The higher protein value of the fermented beans which is about 38-50% may be due to nitrogen contributed by microorganisms involved in fermentation (Eka, 1980).

Proteins are polymers of amino-acids linked together through a peptide bond, the shape and function of protein is determined by the sequence of amino-acids, some of which are essential to the nutritional well being of humans. Amino-acids contain amino group ($-NH_2$) and acid group (-COOH).There are twenty amino-acids that are found in protein. When amino-acids combine to form protein, they do this through the NH_2 group of one amino-acid reacting with the (OH)of another amino-acid, splitting of water into H and OH in the process. The nine essential amino-acids are*: histidine, isoleucine, leucine, lysine, methionine, phenylalanine, theoline, tryptophan and valine. Histidine* is only needed in children (Mottram, 1979).

2.8.5 Ash Content

These are inorganic compounds which appear in food analysis *i.e.* they are substances left behind when the carbon, hydrogen, nitrogen and organic compounds have all burnt off. An adult may have over 1kg of calcium in his body, whereas of chromium, he has only 5-10mg and of copper 150mg (National Research Council, 1996).

2.8.6 Carbohydrate Content

The carbohydrate in the food is a mixture of carbon, hydrogen and oxygen. It can be classified as:

1. Simple carbohydrates or monosaccharide
2 Complex carbohydrates or polysaccharides

2.8.6.1 Simple Carbohydrates

Simple carbohydrates are water soluble and contribute to the sweetness of food. There are two general types of carbohydrates: Reducing and non-reducing sugars. A reducing sugar, *e.g.* glucose (a monosaccharide) contains a reactive *aldehyde* (CHO) group that is absent in non-reducing sugars. Thermal processing can cause reaction between reducing sugar and amino group of protein causing browning and altering flavours. This reaction is called or termed Millard reaction. When two monosaccharides link together they form a disaccharide.

Sucrose is the most common disaccharide and is made up of molecules of glucose and fructose. Sucrose is commonly referred to as sugar, while lactose is the major sugar in milk and is made up of one molecule of glucose and one molecule of *galactose* (Onwuemeh, 1979).

2.8.6.2 Complex Carbohydrates

These consist of celluloses which are the major component of plant cell. Cellulose is a polymer of glucose molecules linked together by beta 1-4 linkages and can not be digested by human. Starch is also a polymer of glucose, but the glucose is joined together by alpha 1-4 linkages that can be digested by humans. Polysaccharides are

commonly used as thickening agent in food and they include: agar, *algin*, locust bean starch, and pectin.

There is a progressive reduction in the carbohydrate content during processing of locust bean due to the removal of pulp and *testa.* The carbohydrate content partly contributed by the pulp, is higher than the protein content and its considerable amount is made up of reducing and non-reducing sugars with absolutely no starch, therefore, the sweetness of the pulp can be attributed to the presence of the reducing sugars particularly the fructose (Onwuemeh, 1979).

2.8.7 Mineral Content

Various minerals have been shown to be essential to Man. These can be divided into major nutrients and trace elements, with the later being present in the body tissue. The major mineral nutrients are calcium, potassium, sodium and magnesium. Trace elements include iron, manganese, copper, chromium, tin, zinc, nickel, fluorine and cobalt. The mineral elements are ingested in both organic and inorganic forms (Ihekoronye and Patrick, 1985).

2.9 Bulk Density

The bulk density of a sample is defined as the ratio of the weight of a given quantity of dry sample, including pore spaces, to that of an equal volume of water. It is influenced by the structure, texture and compactness of the sample (Michael and Ojha, 2006).

2.10 pH Value

pH is a measurement of $\log_{10}$ of a reciprocal of the hydrogen ion concentration ($moldm^{-3}$)and is represented as $pH = \log_{10} [H^+]$. The pH for optimal growth of most microorganisms is near neutrality (7.0). Yeast can grow in an acid environment and thrive best in an intermediate acid (4.0-4.5). Moulds tolerate a wider range (2.0-8.0), although their growth is generally greater with an acid pH (Norman and Robert, 2006)

2.11 Traditional Open Sun Drying Techniques

Drying is a process of food preservation; it is an adequate method under most conditions in developing economies. The facilities are capable of being locally manufactured and maintained from materials that are locally available and within the economic reach of the people. Sun drying is the simplest method of drying. Dehydration denotes drying effected by artificial means; it is usually reserved for artificial drying methods employing a forced draft of conditioned air by means of fans. All of the drying methods however rely on the removal of water by evaporation in the original sense (Ihekoronye and Patrick, 1985).

One of the oldest methods of food preservation is drying, which reduces water activity sufficiently to delay or prevent bacteria growth. Drying of *dawadawa* has long been practised traditionally by local farmers, mostly in sub-Sahara northern part of Nigeria, where there is high intensity of sunshine and low relative humidity for most part of the year. In particular, traditional open sun drying of *dawadawa* is carried out on an open flat container exposed to the intensity of sunlight for a certain period of time (Hamid, 1994).

2.12 Uses of Locust Bean

Locust bean contributes significantly to the intake of protein, essential amino-acids, fatty-acids and vitamin-B particularly riboflavin (Fetuga *et al.*, 1973). The tree of locust bean is used as timber for making pestles, mortars, bowls, and hoe handles. They are valuable and reliable source of food especially the seeds which serve as source of useful ingredients for consumption (Campbell-platt, 1980).

The tree also plays a vital ecological role in cycling of nutrient from deep soil by holding the soil particles to prevent soil erosion with the aid of its root, it also provides shades for the farmers (Campbell-platt, 1980). The bark and the fruit with or without seeds are used to prepare dyes, tannins, and narcotics for fishing. The leaves and the bark are used in remedies for treating guinea worm, filiariasis, skin infections and burns (Dupriez, 1992).

2.13 Preservation of *Dawadawa*

The post fermentation treatment of *dawadawa* varies with each locality. Yorubas of southwestern Nigeria add salt to fermented locust bean, while the Hausas of the northern part prefer drying as a means of preservation. Refrigeration is another method of preserving the fermented locust bean.

Drying: The most common post fermentation treatment involves drying the *dawadawa* is the sun which yields a stable dark brown or black coloured product. This colour is derived from *polyphenol* oxidation. It is dried in the Sun for a day or two and pounded into flat cakes. Dried *dawadawa* is usually stored in earthenware pots for up to a year (Popoola, 1980).

Salting: *Dawadawa* can be preserved by adding some salts to the fermented products. The salted fermented beans are then molded into small balls. The balls of *dawadawa* are then arranged in calabash trays covered with raffia trays to keep away flies and other insects. The use of salt as a preservative could be more effective if the right concentration is added. A salt concentration of 5% (w/w) inhibits both the growth of *Bacillus subtilis* and its proteolytic activity (Popoola, 1980).

2.14 Factors Influencing the Quality of Stored Products

The major problem in storage operation is the maintenance of products quality during the storage period. The factors responsible for quality deterioration are divided into three major groups. These could be physical, chemical and biological in nature; each of these groups may be from either internal or external sources. The internal factors include products composition and internal activities which could be chemical and/or biological in nature. The external factor could be physical (temperature, humidity and sorption phenomena). Chemical factors are gaseous exchange or biological activities of insect, bacteria, fungi and activities of man (Ajisegiri, 2001).

CHAPTER THREE

3.0 MATERIALS AND METHODS

3.1 Materials

The locust beans sample (*Parkia biglobosa*) used in this study to determine the effect of processing method on the nutritional contents of locust beans were purchased from Minna Central market, Niger State (Latitude 9°32/ and Longitude6° 31/) of Nigeria. The sample was collected in a clean polythene bag stored under room temperature 27±2°C, where particles of dirt, stones and pebbles were removed and cleaned from the pack. The processing of locust bean into *dawadawa* was locally prepared by the project student while the tests and analyses were carried out at Animal Production Laboratory and Crop Production Laboratory, Federal University of Technology, Minna, and Science Laboratory Technology, Bida under the supervision of Mr. Audu, Mr. Zegi, Mr. Bidemi and Mr. Jerry respectively.

3.2 Reagents and instruments/Apparatuses

In the course of the practical work carried out in the project, the reagents and instruments/apparatuses used are listed below:

3.2.1 Reagents:

Sodium Hydroxide (NaOH)

Tetraoxosulphate (IV) acid (H_2SO_4)

Methyl Orange Indicator

Petroleum Ether

Hydrochloric Acid (HCl)

Boric Acid (H_3BO_3)

3.2.2 Instruments/Apparatuses

Crucibles

Petri dishes

Desiccators

Weighing balance (±0.001g)

Filter paper

Oven

Soxhlet extractor flat bottom silica dishes

Bunsen burner

Beaker

Pipette

Thimbles

Conical flasks

Muffle furnace

3.3 Preparation of *Dawadawa* from Locust Bean

350grammes of locust bean seeds were poured in the mortar, little water and ash was added and was gently pounded so as to remove the remaining powdery pulp. The seeds were boiled for 12 hours and were allowed to be soaked in water for 8 hours in order to soften the *testa*. The boiled seeds mixed with sand were gently re-pounded in mortar or rubbed with either hands or legs in order to remove the softened *testa*. The cotyledons were then washed thoroughly in clean water and the *testa* was separated with the aid of sieve. The washed bean cotyledons were further boiled for 1 hour by adding a softening agent "*kuru*" in it. The hot and softened bean cotyledons were spread in a clean

calabash covered with several cloths which provided warmth and fermentation was allowed for 24 hours.

3.4 Experimental Procedures

Ibitoye (2005) guidelines for determining nutritional parameters were followed (Appendix A).

3.4.1 Determination of Moisture Content of Locust Bean

Moisture content is the amount of moisture (water) present per given weight of sample. It is checked in order to increase the shelf-life of *dawadawa* by preventing microbial growth.

Procedure

A cleaned, well labeled dish was weighed which has been oven dried (W_1). Enough samples were added into the dish and weighed (W_2). The dish with the content was transferred to the thermosetting oven at about 105°C for about 24 hours, which was later transferred from oven to desiccator and allowed to cool for about 1 hour and weighed (W_3). Percentage moisture content was calculated with the expression below.

$$\%\ Moisture = \frac{W_2 - W_3}{W_2 - W_1} \times 100$$

3.4.2 Determination of Crude Fibre Content of Locust Bean

Crude fibre is that portion of the plant material which is not ash or dissolves in boiling solution of 1.25% H_2SO_4 or 1.25 NaOH. The following diseases are associated with diet low in fibre: Constipation, or if fibre is low the intestine absorbs water and volume of the feaces will decrease.

Procedure

5g sample was transferred into conical flask and 200ml of boiling 1.25% H_2SO_4 was added and brought to boiling which was allowed for 30 minutes. 200ml of boiling 1.25% NaOH was added and few drops of anti-foaming agent brought to boiling within 1 minute and boiled gently for 30 minutes using cooling finger (KOH can be used in place of NaOH). Poplin cloth was used to filter and washed with hot distilled water; it was rinsed four times with hot distilled water and once with 10% HCl, also four times with hot water and twice with methylated spirit and three times with petroleum ether. The residue was savaged into crucible after drained and dried in the oven at 105^0C, cooled in desiccator and weighed (W_2); it was removed into desiccator and was allowed to cool to room temperature and weighed (W_3).

$$\% \, Crude \, Fibre = \frac{W_2 - W_3}{W_1} \times 100$$

3.4.3 Determination of Lipid Content of Locust Bean

Soxhlet Extraction Method

Extraction is carried out with soxhlet apparatus with either ether or petroleum ether. The usual procedure is to continuously extract the fat content with 40-60^{O}C petroleum ether in a convenient extractor.

Procedure

A dried thimble free from fat was weighed (W_1), 50g of sample was added into the thimble and weighed (W_2). A 500ml round bottom flask was weighed (W_3) and was filled with petroleum ether up to 2/3 of the 500ml flask. A soxhlet extractor with a reflux condenser was fitted together, heat source was adjusted so that the solvent boiled gently and was left to siphon over several hours (5-6 hours), condenser was detached and

thimble was removed. The flask containing fat residue was dried in an air oven at 100^{O}C for 5 minutes and was allowed to cooled in a desiccator weighed (W_4), the thimble was placed in a beaker and put in an oven at 50^{O}C which dried to constant weight with the sample. It was cooled in a desiccator and weighed (W_5).

1. Weight of the lipid in the flask after extraction

$$\% \, Lipid = \frac{W_4 - W_3}{W_2 - W_1} \times 100$$

2. Weight loss in the sample

$$\% \, Lipid = \frac{W_2 - W_5}{W_2 - W_1} \times 100$$

3.4.4 Determination of Crude Protein of Locust Bean

The amount of crude protein contains in the seeds, roots, tubers and other stuff can be obtained by multiplying the nitrogen content of the food by 6.25. The factor 6.25 owes its origin to the assumption that all food protein contains 16% nitrogen and that all the nitrogen in a feed is present as protein. It is necessary to digest the sample for certain period until a cleared solution is obtained to ensured accurate results.

Procedure

Digestion (stage 1)

2g of wet sample was weighed into 50ml kjeldahl flask and 20ml of concentrated H_2SO_4 was added with one kjeldahl catalyst tablet, another 0.5g of dryed sample was weighed into 50ml kjeldahl and 5ml concentrated H_2SO_4 with half kjeldahl catalyst was added and weighed (W_1). Heat on the heater was started with a low heat for about 15 minutes, increased to medium heat for about 30 minutes and finally at high heating until digested. The sample residue was allowed to cool and filtered to get the appropriate digest (V_1).

Distillation (Stage 2)

5ml of 2% boric acid (H_3BO_3) was placed into100mlconical flask. H_3BO_3 as an acid was able to trap down the ammonia vapour from the digest and 3-drops of mixed indicator was added (H_3BO_3 and indicator can be prepared together). An indicator of 0.198g *bromocresol* green was mixed with 0.132g methyl red in 200ml alcohol, while the receiving flask was placed so that the tip of the condenser tube was below the surface of the boric acid. 10ml of 40% NaOH was added and distilled about 50ml into the receiving flask (V_2).

Titration and Calculation (Stage 3)

The distillate was titrated with standard mineral acid (0.01 HCl) and a blank was also titrated with the acid.

Sample titre: T_1

Blank titre: T_2

Control titre: $T_1 - T_2 = T$

Molarity of acid: M

Digestion

$H_2SO_4 + 2NH_3 \longrightarrow (NH_4)_2SO_4$

Distillation

$(NH_4)_2SO_4 + 2NaOH \rightarrow 2Na_2SO_4 + NH_3 + 2H_2O$

$NH_3 + H_3BO_3 \rightarrow NH_4 + H_2BO_3$

Titration

$NH_4 + H_2BO_3 + HCl \rightarrow NH_4Cl + H_3BO_3$

1 mole of HCl = 1mole of NH_3

$$Molarity\ of\ HCl = \frac{M \times T}{1000}$$

$$Molarity\ of\ NH_3 = \frac{M \times T}{1000}$$

$$Mass\ of\ NH_3 = \frac{M \times T}{1000} \times 17 \times \frac{14}{17} = M \times T \times 0.014g$$

$$\%\ N = \frac{M \times T}{W} \times 0.014 \times \frac{V_1}{V_2} \times 100$$

% of Crude protein = 6.25 × % of N

3.4.5 Determination of Ash Content Locust Bean

The ash of the biological materials is analytical termed for the inorganic residue that remains after the organic matter has burnt off. The ash is not the same as the inorganic matter present in the original material since there may be losses due to volatilization or chemical interaction between the constituents. The importance of ash content is that it gives an idea of the amount of mineral elements present and contents of the organic matter in the sample.

Procedure

A crucible was placed in muffle furnace for about 15 minutes at 350^{O}C which was later removed and cooled in desiccator for 1 hour and weighed to be (W_1). Enough sample was added into the crucible and weighed (W_2), the crucible was replaced inside the muffle furnace and slowly increased temperature from 200^{O}C to 400^{O}C in order to avoid incomplete ashing (if ashing is not complete, evidence of black particles will be there). The crucible was removed from furnace and returned to desiccator which was allowed to cool to room temperature. The crucible with the content was reweighed (W_3).

Calculation

$$\% Ash = \frac{w_3 - w_1}{w_2 - w_1} \times 100$$

3.4.6 Determination of Carbohydrate Content Locust bean

This is the subtraction of total protein content and lipid content from the organic matter.

% Carbohydrate = 100% – (% protein + % lipid + % ash + % crude fibre).

3.4.7 Determination of Mineral Content of Locust Bean

The mineral contents such as: magnesium, potassium, iron and sodium, zinc, copper and lead in the residue remaining after the destruction of organic matter of plant material (ashing), were dissolved in HCl. The concentration of the elements in the solution was determined by a machine called Atomic Absorption Spectrophotometer (AAS) and Flame Photometer Machine. These machines use a method of chemical analysis based on the absorption or attenuation and emission by matter of electromagnetic radiation of specified wavelength or frequency.

3.4.8 Determination of Bulk Density of Locust Bean

A 50g locust bean sample was put inside a 100ml graduated cylinder. The cylinder was tapped 40 times and the bulk density was calculated as weight per unit volume:

$$\text{Bulk density} = \frac{\text{loose volume}}{\text{packed volume}} \times 100$$

3.4.9 Determination of pH Value of Locust Bean

The pH is defined as the negative value of the logarithm to the base 10 of the hydrogen ion concentration.

$pH = -\log_{10}[pH]$

The pH of locust bean was determined using an electronic machine called Electronic pH meter (Kent 7640). The grinded locust bean poured inside a distil water and shook vigorously. The meter was calibrated using buffer solution of pH7, the machine's protrude was deepen inside the solution of locust bean and the sample pH was recorded.

CHAPTER FOUR

4.0 RESULTS AND DISCUSSION

4.1 Results

The results of the nutritional analysis (proximate composition), pH value and bulk density of unfermented (raw) locust bean (*parkia bigglobosa*) and fermented locust bean "dawadawa" are presented in Table 4.1.

Table 4.1: Results of the Nutritional Content of Unfermented and Fermented Locust Bean

Parameter	Nutritional value	
	Unfermented	Fermented
Moisture content (%)	7.01	37.1
Crude fibre (%)	4.7	2.0
Lipid (%)	9.569	21.172
Crude protein (%)	28.0	38.5
Ash (%)	2.0	1.8
Carbohydrate (%)	55.731	36.528
Copper (ppm)	9.5	2.5
Iron (ppm)	2.647	1.118
Lead (ppm)	0.00	0.00
Magnesium (ppm)	7.87	7.67
Potassium (ppm)	41.86	63.04
Sodium (ppm)	81.3	19.575
Zinc (ppm)	3.467	6.293
Bulk density (gcm^{-3})	6.25	15.5
pH	8.3	6.25

The data obtained in this study were subjected to the statistical analysis using statistical programming in Microsoft Office Excel. The statistical analysis carried out was

Analysis of Variance (ANOVA) using single factor (Appendix B).This was used to determine the level of significance when the two samples were compared. The analysis showed that there was a significant difference between the fermented and unfermented locust beans ($p<0.05$) as a result of processing.

4.2 Discussion of Results

The results of the nutritional analysis performed showed that: The moisture contents of unfermented (raw) and fermented locust bean were 7.01% and 35.21% respectively. The increase in moisture content may probably due to boiling in water and followed by soaking in water. It may also be as a result of metabolic activities of microorganisms which give moisture as one of their end products. This agrees with Omafubve *et al* (2004). The high moisture however suggests that the fermented locust bean will not keep well in storage *i.e.* it is subject to rapid deterioration by mould growth.

The crude fibre content was determined to be 4.7% for unfermented which was very high compared to 2% obtained for the fermented locust bean. The boiling and de-hulling of the locust bean during processing might have led to the reduction in crude fibre which indicates that the product "*dawadawa*" is highly nutritive than unfermented (raw) locust bean.

The crude lipid contents of unfermented locust bean and fermented "*dawadawa*" were 9.569% and 21.172% respectively. The lipid content obtained in this work is in agreement with the findings of Omafuvbe *et al* (2004), Addy *et al* (1995) and Ikenebomeh (1986). The increment recorded in the crude protein and lipid content may be due to the reduction in the carbohydrate, ash and crude fibre. The crude lipid content

present in *dawadawa* could serve as a ready source of energy when incorporated into diet.

The values obtained for crude protein determination were 28% obtained for unfermented locust bean and 38.5% for fermented "*dawadawa*", this is considerably higher *i.e.* there was a significant difference ($p<0.05$) between the two samples. The increase in the protein content obtained in this study agrees with other reports on African locust bean seeds (Ikenebomeh, 1986; Omafuvbe *et al.*, 2004). The microorganisms involved in the fermentation of the locust bean which showed proteolytic activity may contribute to high protein content in "*dawadawa*". *Bacillus substilis* have been known to cause hydrolysis of protein to amino-acids and peptides releasing ammonia which increases the protein content of the fermented product (Odunfa, 1981a). This result also agrees with Odunfa (1986) who also observed a high percentage of protein in "*dawadawa*"

The ash contents of unfermented locust bean and fermented were 2% and 1.8% respectively. The reduction occurred may be as a result of leaching of soluble inorganic salt into the processing water during soaking of the sample for 8 hours.

The carbohydrate content also decreased as a result of soaking and boiling from 55.731% of unfermented locust bean to 36.528% of *dawadawa*. This result is in agreement with results of earlier workers (Addy *et al.*, 1995; Omafuvbe *et al.*, 2004; Osman, 2007). Loss in carbohydrate may be due to leaching of soluble carbohydrate like sugars into the soaking and cooking water; while loss in carbohydrate during fermentation may be as a result of utilization of some of the sugars by fermenting organisms for growth and metabolic activities.

The mineral contents of unfermented locust bean and fermented locust bean were determined by Atomic Absorption Spectrophotometer (AAS) and Flame Photometer machine. The mineral values obtained were determined from AAS standard graph (Appendix D).

The bulk density of unfermented was 6.25g/cm^3 and that of the fermented locust bean was 15.5g/cm^3. This shows that there was an increase in bulk density as a result of the sample subjected to soaking, heating and fermentation which in turn made the fermented locust bean to have a fine texture when ground.

The values obtained for pH of unfermented locust bean and fermented were 8.3 and 6.25 respectively. This obtained pH was not surprising because fermentation decreases the pH of substrates.

CHAPTER FIVE

5.0 CONCLUSIONS AND RECOMMENDATIONS

5.1 Conclusions

From the results obtained in this study, it has been shown that processing operations such as soaking and boiling have significantly improved the values of the product; however, the fermentation process achieved better quality that will enhance utilization of the product "*dawadawa*". The result showed that there was an increase in crude protein and lipid content of *dawadawa* which is highly needed for building body tissue and also enhance the organoleptic of soup and stew when added. The mineral contents such as Magnesium, copper and iron were reduced while potassium and zinc were increased as a result of processing. The raw and fermented locust bean contained no lead which signifies that the sample is not injurious to body health.

5.2 Recommendations

The following recommendations are made after completing this work:

1- *Dawadawa* is highly nutritious and should be encouraged in health diets.

2- Research should be carried out to carefully evaluate the microbial hazards of this fermented product.

3- Methods to improve the local method of processing of locust bean into *dawadawa* should be looked into.

4- The possibilities of extraction of fat from locust bean for consumption should be investigated.

5- The methods of preservation of *dawadawa* should be improved to prolong the shelf life of the food spices.

REFERENCES

Achi, O.K. (1992). Microorganisms associated with natural fermentation of *prosopis Africana* seeds for production of *okpiye* plant foods. Hum. Nutr., 42: 297-304

Achi, O.K. (2005). Traditional fermented protein condiments in Nigeria. African Journal of Biotechnology, 4(13): 1612-1621

Addy, E.O., L.I. Salami, L.C. Igboeli and S.H. Remawa, 1995. Effect of processing on nutrient composition and anti- nutritive substances of African locust bean (*Parkia biglobasa*) and baobab seed *(Adansonia digitata*). Plant foods Hum. Nutr., 48: 113-117

Adewakun, A.E (1988). Technological Improvement and Shelf-life studies of *"iru"* production from African locust bean (*Parkia biglobosa*). Department of Food Science, University of Ibadan. Pp. 1-3

Adewusi, S.R.A., B.O. Orisadare and O.L. Oke, (1992). Studies on Weaning Diets in Nigeria. Plant Foods for Human Nutr., 42: 183-192

Aidoo, K.E. (1986). Lesser-known fermented plant foods. Trop. Science, 26: 249-258

Ajisegiri, E.S.A. (2001). Solar Energy Application in Agricultural and Food Preservation. Pp. 102-104

Anon (1981). A new protein food from traditional process. Nutrition Review (39) pp. 9-11

AOAC, 1980. Official Methods of Analysis (13th edition). Association of Analytical Chemists, Washington, DC, USA, pp. 211-213

Barber, L.A. and S.C. Achinewhu (1992). Microbiology of *ogiri* production from melon seeds (*Citrullus vulgaris*). Nig. Food Journal, 10:129-135

Barimalaa, I.S., Acinewhu, S.C. and E.N Amadi (1989). Studies on the solid substrates fermentation of bambara nut (Vigna *subterranean*). J.Sci. Food Agric, 66:443-446

Campbell-platt, G. (1980). African locust bean (*parkia species*) and its West African fermented food products, *Dawadawa*. Ecology of Food and Nutrition, 9: 123-132

Dupriez, H. (1992). Land and Life African Gardens and Orchards. Macmillan. Pp. 304

Eka, O.U (1980). Effect of fermentation on the nutrient status locust bean. Food Chemistry. 5: 305-308

Fetuga, B.L., G.M. Babatunde and V.A. Oyenuga (1973). Protein quality of some nigerian foodstuff 1: Chemical assay of nutrients and amino acid composition, Journal of Science Food Agric, 24: 1505-1514

Hamid, A.D. (1994). The Indigenous Fermented Food of Sudan. Britain university press. Pp. 323-324

Ibitoye, A.A. (2005). Laboratory Manual on Basic methods in Plant Analysis. Foladave Publishing Co., Akure. Pp.6-27

Ihekoronye, A. and O. G. Patrick (1985). Integrated Food Science and Technology for the Tropics. Macmillan Education Ltd. pp. 92-93, 103-104, 127-128

Ikenebomeh, M.J. and R. Kok (1999). Mass balance of the processing and fermentation of the African locust bean (*Parkia species*). J. Can. Inst. Food Sci. Tech., 17: 48-50

Ikenebomeh, M.J. (1986). African locust bean fermentation some physico-chemical parameters. Nig. Journal Appl. Sci., 4: 101-108

Ikenebomeh, M.J. and C.N. Esenwah (2008). Processing Effects on the Nutritional and Anti-Nutritional Contents of African Locust Bean. Pakistan Journal of Nutr., 7(2): 214-217

Mayhew, S. and A. Penny (1988). Tropical and Sub-Tropical Foods. Macmillan Press. Pp. 36-37, 46-47

Mottram, R.F. (1979). Human Nutrition, 3rd edition. Edward Arnol publisher ltd. London.

Michael, A.M. and T.P.Ojha (2006). Principle of Agricultural Engineering. Vol. 2. Sunil Kumar Jain. pp. 342

National Research Council (NRC) (1996). Lost Crops of Africa. Vol. 1. Grain National Accademic Press. Washinton D.C. pp. 3-6

Norman G.M. and B. Robert (2006). Principle of Food Sanitation. Springer Science+ Business Media, Inc., pp. 32-33

Obeta, J.A.N. (1983). A note on the microorganisms associated with the fermentation of seeds of African oil bean seed (*Pentaclethra macrophylla*). Journal of Appl. Bacteriol, 54: 217-222

Odunfa, S.A. (1981a). A note of microorganisms associated with the fermentation of African locust bean (*parkia species*) during *iru* production. Journal of Plant Foods, 3: 245-250

Odunfa, S.A. (1981b). Microbiology and amino acid composition of ogiri food condiment from fermented melon seeds. Die Nahrung, 25: 811-816

Odunfa, S.A. (1985a). Biochemical changes in fermenting African locust bean (*Parkia biglobosa*) during *iru* fermentation. Journal Food Technology, 20: 295-303

Odunfa, S.A. (1985b). Microbiological and Toxicological aspect of fermentation of castor oils for *ogiri* production. Journal Food Science, 50: 1758-1759

Odunfa, S.A. (1986). Dawadawa. In: Legumes based fermented foods eds. Redy N.R, Pierson M.D. , Salunkhe D.K.R.C Press Boca Raton. pp. 173-189

Ogbonna, D.N., T.G. Sokari and S.C. Achinewhu (2001). Development of an owoh- type product from African yam beans (*Sphenostyllis stenocarpa*) by solid substrate fermentation. Plant Food Hum. Nutr., 56: 183-194

Ogunbunmi, E.M. and O. Bashir (1980). Protein and amino acid content of some Nigerian food condiment. Nutr. Rep. Int., 22: 497

Omafuvbe, B.O., O.S. Falade, B.A. Osuntogun and S.R.A. Adewusi (2004). Chemical and biochemical changes in African locust bean (*Parkia biglobosa*) and melon seeds (*Citrullus vulgaris*) during fermentation to condiments. Pak. J. Nutr., 3: 140-145

Onwuemeh, I.C. (1979). Crop Science. Camelot Press Ltd. pp. 58-59, 232-234

Osman, M.A. (2007). Effect of different processing methods on nutrient composition, antinutrtional factor and *in vitro* protein digestibility of *Dolichos lablab* bean. Pak. J. Nutr., 6: 299-303

Popoola T.O.S. (1980). Micro organisms associated with soya bean (*Glycine max*) and a fermented soya bean product *dawadawa* Unversity of Jos, Nigeria.

Popoola, T.O.S. and C.O. Akueshi (1984). Nutrtional evaluation of *dawadawa* , a local spice made from soya bean (*Glycine max*). MIRECEN Journal, 3: 405-409

Sanni, A.L and D.N.Ogbonna (1991). The Production of *Owoh* A Nigeria Fermented Seasonning Agent from Cotton Seed (Gossypium hirsitium). Food Microbiol. 8: 223-229

Sarkar, P.K., P.E. Cook and J.D. Owens (1993). Bacillus fermentation of soya beans. World Journal Microbial Biotech., 9: 295-299

Tahir, F., A. Uba and G.G. Chigbo (1998). Studies on the bacteriological quality of locally prepared *dawadawa* in Bauchi metropolis. African Journal of Natural Sciences, 2: 32-35

Vickery, M.L. and B. Vickery (1979). Plant Production of Tropical Africa. Macmillan Tropical Agric. Horticulture and Applied Ecology Series. pp. 32

APPENDIX A

Moisture Content of Unfermented and Fermented Determination

Weights taken during moisture content of Unfermented and Fermented determination

Sample	W_1	W_2	W_3
Unfermented	44.723	46.72	46.58
Fermented	42.45	51.45	48.11

W_1 = Mass of empty crucible (g)

W_2 = Mass of crucible and sample (g)

W_3 = Mass of crucible and sample after drying (g)

$$\%moisture = \frac{w_3 - w_1}{w_2 - w_1} x100$$

$$Unfermented\ sample: \%moisture = \frac{46.72 - 46.58}{46.72 - 44.723} x100$$

$$= 7.01\%$$

$$Fermented\ Sample: \%moisture = \frac{51.45 - 48.11}{51.45 - 42.45} x100$$

$$= 37.1\%$$

Crude Fibre of Unfermented and Fermented Determination

Weights obtained during crude fibre content of Unfermented and Fermented determination.

Sample	W_1	W_2	W_3
Unfermented	5.0	13.268	13.033
Fermented	5.0	10.021	9.921

W_1 = Weight of the sample used (g)

W_2 = Weight of the sample and crucible (g)

W_3 = Weight of the sample crucible and ash (g)

$$\%\,crude\ fibre = \frac{w_2 - w_3}{w_1} x100$$

$$Unfermented\ Sample: \%Crude\ Fibre = \frac{13.268 - 13.033}{5.0} x100$$

$$= 4.7\%$$

$$Fermented\ Sample: \%crude\ fibre = \frac{10.021 - 9.921}{5.0} x100$$

$$= 2.0\%$$

Crude Lipid Content of Unfermented and Fermented Determination

Weights obtained during Crude fat of Unfermented and Fermented determination

Sample	W_1	W_2	W_3
Unfermented	0.757	2.429	2.269
Fermented	0.926	2.121	1.868

W_1 = Weight of the thimble (g)

W_2 = Weight of the thimble and sample (g)

W_3 = Weight of the sample thimble after dryed at $50^{O}C$ (g)

$$\%crude\ fat = \frac{w_2 - w_3}{w_2 - w_1} x100$$

$$Unfermented\ Sample: \%Crude\ Fat = \frac{2.429 - 2.269}{2.429 - 0.757} x100$$

$$= 9.5695\%$$

$$Fermented\ Sample: \%crude\ fat = \frac{2.121-1.868}{2.121-0.926} x100$$

$$= 21.172\%$$

Crude protein of Unfermented and Fermented Determination

Titration volumes of Unfermented and Fermented determination

Sample	Titre volume (cm^3)
Unfermented	1.8
Fermented	2.4
Blank	0.2

Control titre(T) = (Unfermented - Blank) = (1.8 – 0.2) = $1.6cm^3$

(T)= (Fermented - Blank) = (2.4 – 0.2) = $2.2cm^3$

Molarity of HCl(M) = 0.1M

Weight of the wet sample = 0.5g

Used volume of NaOH = 10ml

$$\%Nitrogen(N) = \frac{M\ xT\ x0.014x10}{W} 100$$

Percentage Protein = % Nitrogen x 6.25

$$Unfermented\ Sample: \%N = \frac{0.1x1.6x0.014x10}{0.5} x100$$

$$= 4.48\%$$

% Protein = 4.48 x 6.25

$$= 28\%$$

$$Fermented\ Sample \%N = \frac{0.1x2.2x0.014x10}{0.5} x100$$

$= 6.16\%$

% Protein = 6.16 x 6.25

$= 38.5\%$

Ash Content of Unfermented and Fermented Determination

Weights obtained during Ash content of Unfermented and Fermented determination

Sample	W_1	W_2	W_3
Unfermented	14.65	16.65	14.69
Fermented	14.662	17.889	14.72

W_1 = Weight of the crucible (g)

W_2 = Weight of the crucible and sample (g)

W_3 = Weight of the crucible sample after subjected to temperature of $350^{O}C$ to $450^{O}C$ (g)

$$\%ash = \frac{w_3 - w_1}{w_2 - w_1} x100$$

$$Unfermented\ Sample: \%Ash = \frac{14.69 - 14.65}{16.65 - 14.65} x100$$

$$= 2\ \%$$

$$Fermented\ Sample: \%Ash = \frac{14.72 - 14.662}{17.889 - 14.662} x100$$

$$= 1.8\ \%$$

Carbohydrate Content of Unfermented and Fermented Determination

	Unfermented	Fermented
Percentage Crude Protein Content =	28%	38.5%
Percentage Crude Fibre Content =	4.7%	2%
Percentage Crude Fat Content =	9.569%	21.172%
Percentage Ash Content =	2%	1.8%
Total	44.269%	63.472%

Percentage Carbohydrate for Unfermented = 100% - 44.269%

= 55.731%

Percentage Carbohydrate for Fermented = 100% - 63.472%

= 36.528%

Mineral Content of Unfermented and Fermented Determination

The mineral contents such as: Magnesium, iron, copper, zinc and lead were determined by Atomic Absorption Spectrophotometer while sodium and potassium were determined by Flame Photometer. The values of unfermented and fermented locust bean were obtained on the standard graph of AAS (Appendix D).

Bulk density of Unfermented and Fermented Determination

$$Bulk\ density = \frac{Loose\ Volume\ x\ Initial\ Weight\ of\ the\ Sample}{Packed\ Volume}$$

Loose volume for Unfermented = $10cm^3$

Loose volume for Fermented = $17cm_3$

Initial weight of sample = 50g

Packed volume for Unfermented = $80cm^3$

Packed volume for Fermented = $55cm^3$

$$Unfermented\ Sample: Bulk\ density\left(g/cm^3\right)=\frac{10x50}{80}=6.25g/cm^3$$

$$Fermented\ Sample: Bulk\ density\left(g/cm^3\right)=\frac{17x50}{55}=15.5g/cm^3$$

pH of Unfermented and Fermented Determination

The pH of Unfermented and Fermented were 8.3 and 6.52 respectively determined by pH meter.

APPENDIX B

ANOVA: Single Factor

SUMMARY

Groups	*Count*	*Sum*	*Average*	*Variance*
Moisture	2	44.11	22.055	452.70405
Crude fibre	2	6.7	3.35	3.645
Lipid	2	30.741	15.3705	67.3148045
Crude protein	2	66.5	33.25	55.125
Ash	2	3.8	1.9	0.02
Carbohydrate	2	92.259	46.1295	184.377605
Cu	2	12	6	24.5
Fe	2	3.765	1.8825	1.1689205
Pb	2	0	0	0
Mg	2	15.54	7.77	0.02
K	2	104.9	52.45	224.2962
Na	2	100.875	50.4375	1904.98781
Zn	2	9.76	4.88	3.993138
Bulk Density	2	21.75	10.875	42.78125
pH	2	14.82	7.41	1.5842

ANOVA

Source of Variation	*SS*	*df*	*MS*	*F*	*P-value*	*F crit.*
Between Groups	9849.788	14	703.55628	3.55748533	0.010084	2.4243
Within Groups	2966.518	15	197.76787			
Total	12816.31	29				

(P<0.05)

APPENDIX C

Atomic Absorption Spectrophotometre Standard Table

	Zinc	*Iron*	*Copper*	*Chromium*	*Lead*	*Magnesium*
1ppm	0.040	0.017	0.002	0.455	0.208	0.544
2ppm	0.055	0.033	0.004	0.772	0.238	0.743
3ppm	0.115	0.050	0.006	0.869	0.361	0.816
4ppm	0.150	0.075	0.008	0.941	0.377	0.883
5ppm	0.200	0.080	0.010	1.092	0.388	0.915
6ppm	0.225	0.102	0.012	1.134	0.404	0.968
7ppm	0.265	0.120	0.013			0.994
8ppm	0.300	0.137	0.017			1.000
9ppm	0.361	0.143	0.018			
10ppm	0.375	0.173	0.020			

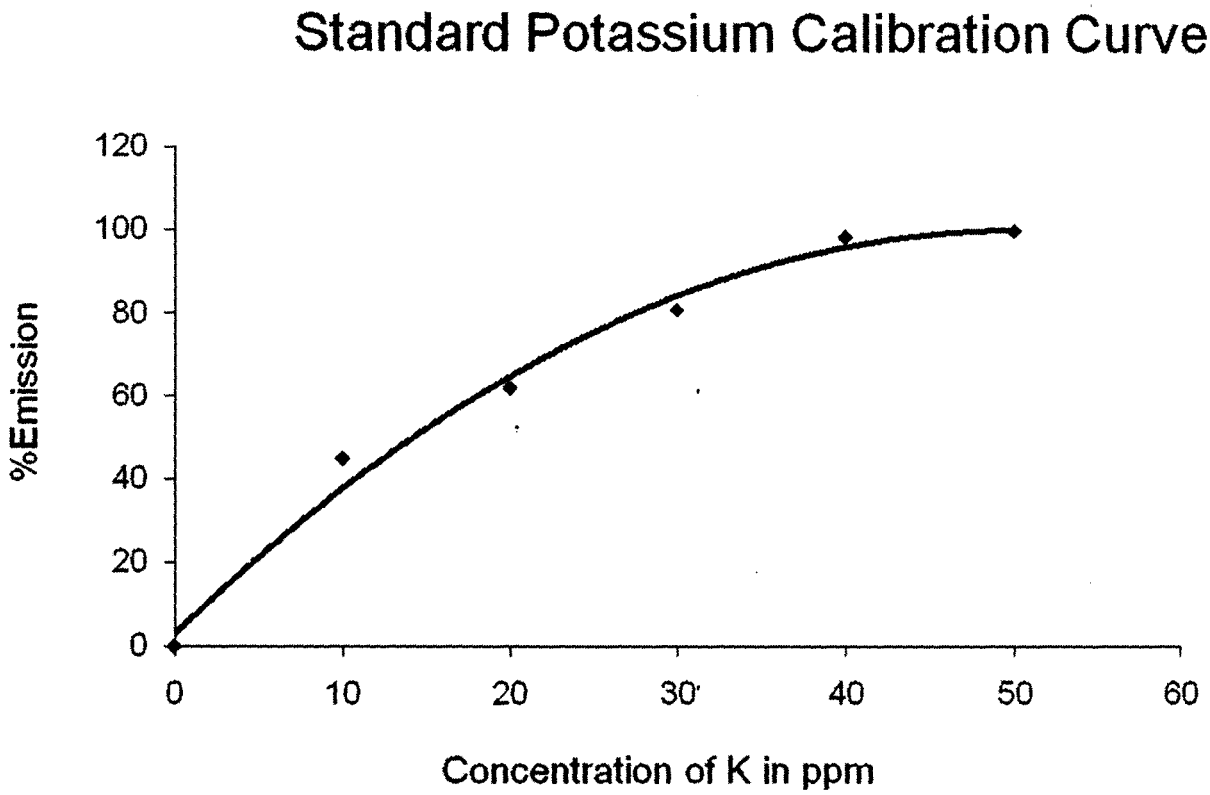
Standard Potassium and Sodium % Emission Reading with Flame Photometer.

Conc. Na^+/ppm	*% Emission*	*Conc. K^+/ppm*	*% Emission*
0	0.00	0	0.00
20	27.20	10	44.80
40	54.90	20	62.00
60	68.60	30	80.90
80	79.70	40	98.50
100	100.00	50	100.00

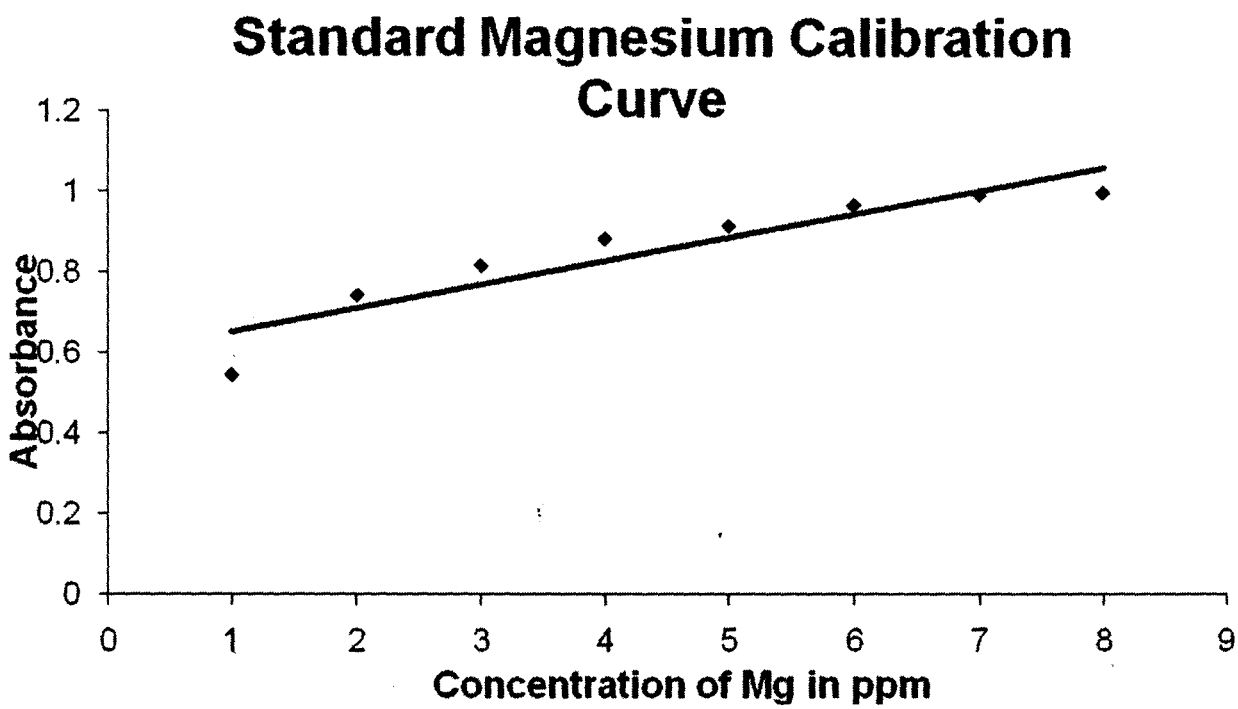
The values for minerals obtained for Fermented and unfermented locus beans

Sample	**Mg**	**Cu**	**Fe**	**Zn**	**Pb**	**Na**	**K**
Unfermented	1.338	0.023	0.090	0.147	0.00	30.70	152.2
Fermented	1.308	0.009	0.064	0.253	0.00	113.00	103.5
Blank	0.180	0.004	0.045	0.017	0.00	4.6	7.2

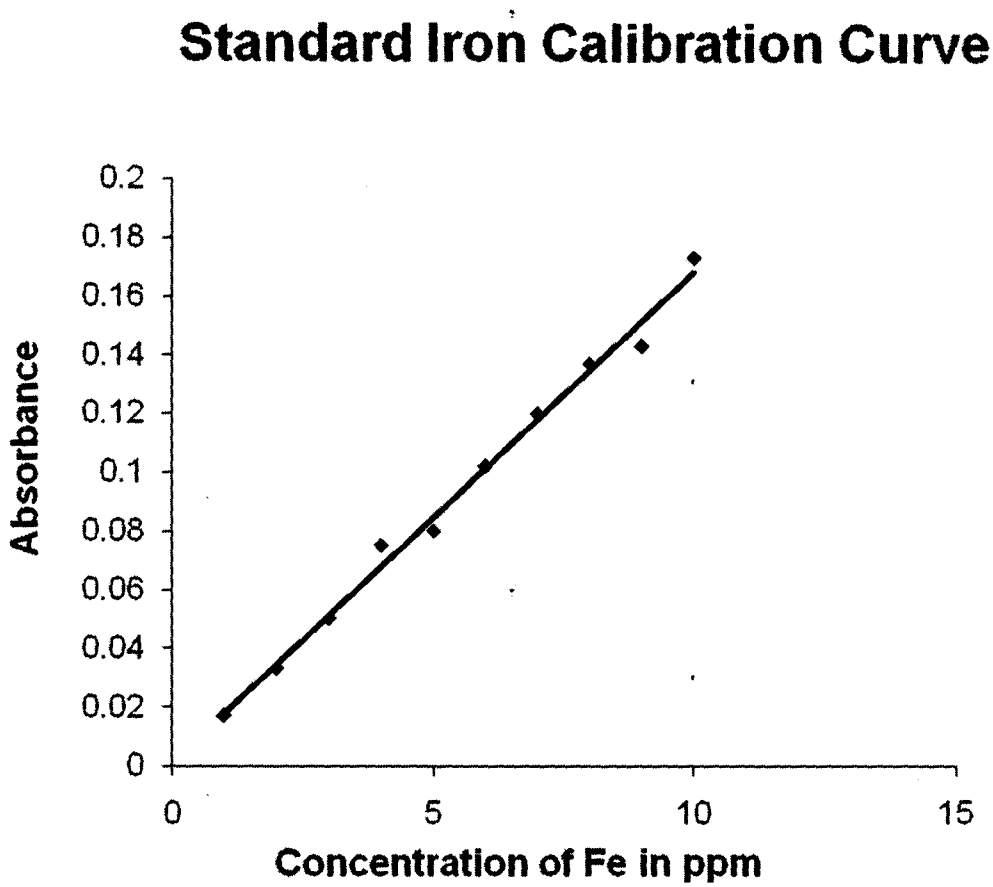
APPENDIX D1


Standard Potassium Calibration Curve
%Emission
120
100
80
60
40
20
0
0
10
20
30
40
50
60
Concentration of K in ppm

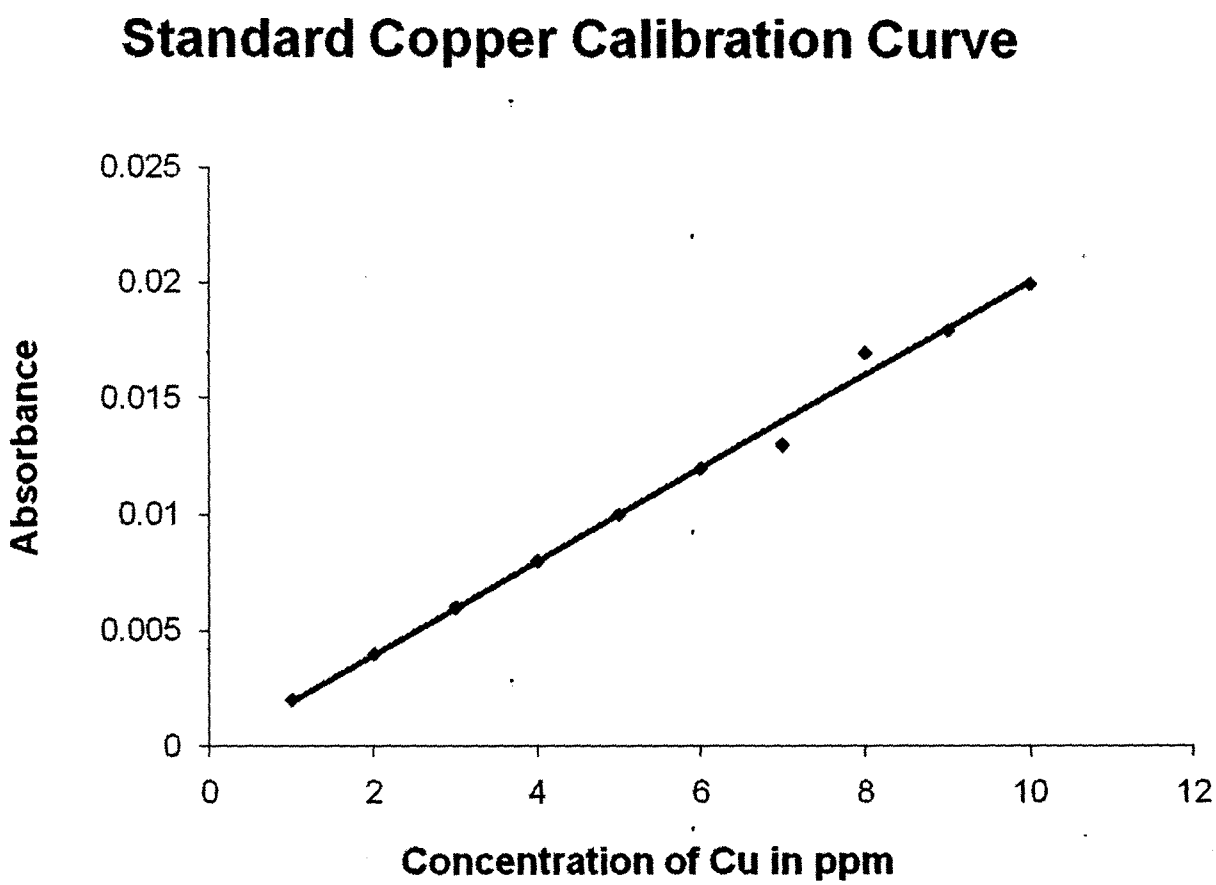
APPENDIX D2


Standard Magnesium Calibration Curve
Absorbance
1.2
1
0.8
0.6
0.4
0.2
0
0
1
2
3
4
5
6
7
8
9
Concentration of Mg in ppm

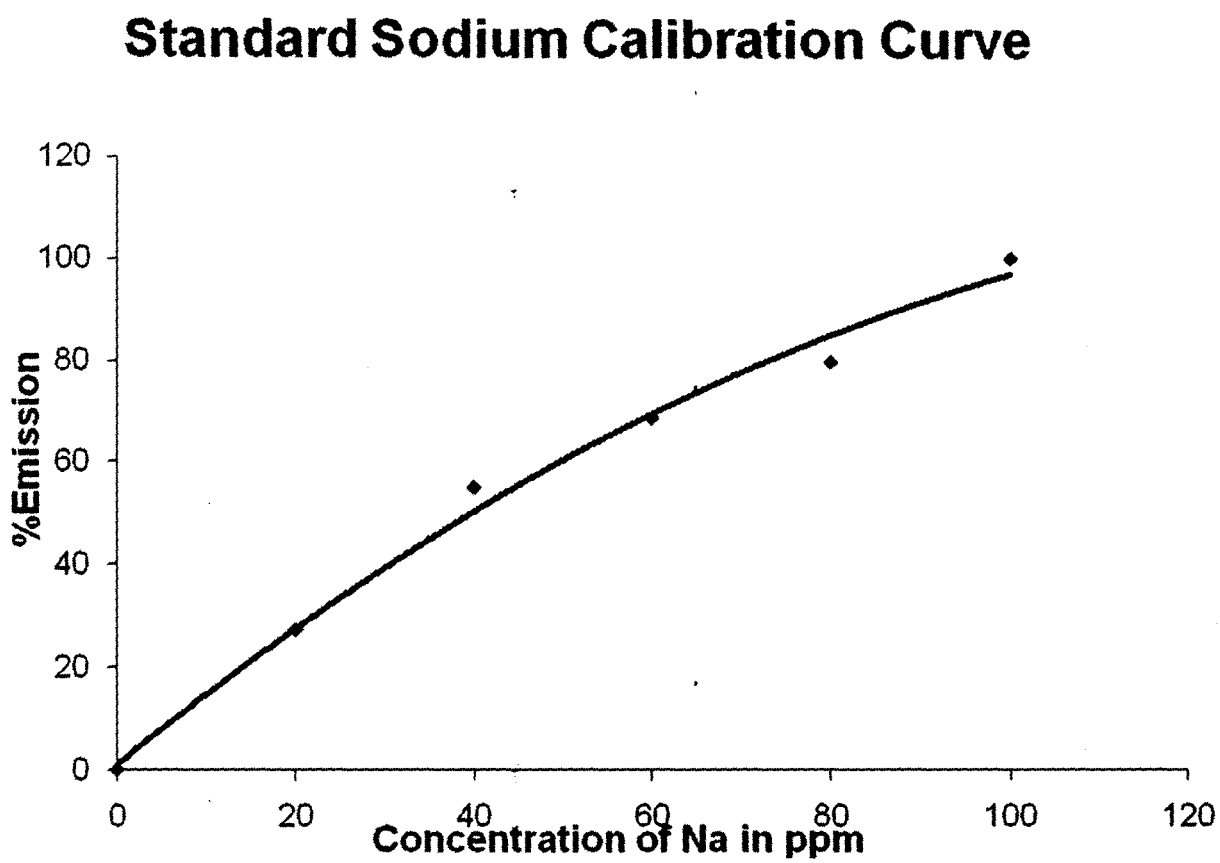
APPENDIX D3


Standard Iron Calibration Curve
Absorbance
0.2
0.18
0.16
0.14
0.12
0.1
0.08
0.06
0.04
0.02
0
0
5
10
15
Concentration of Fe in ppm

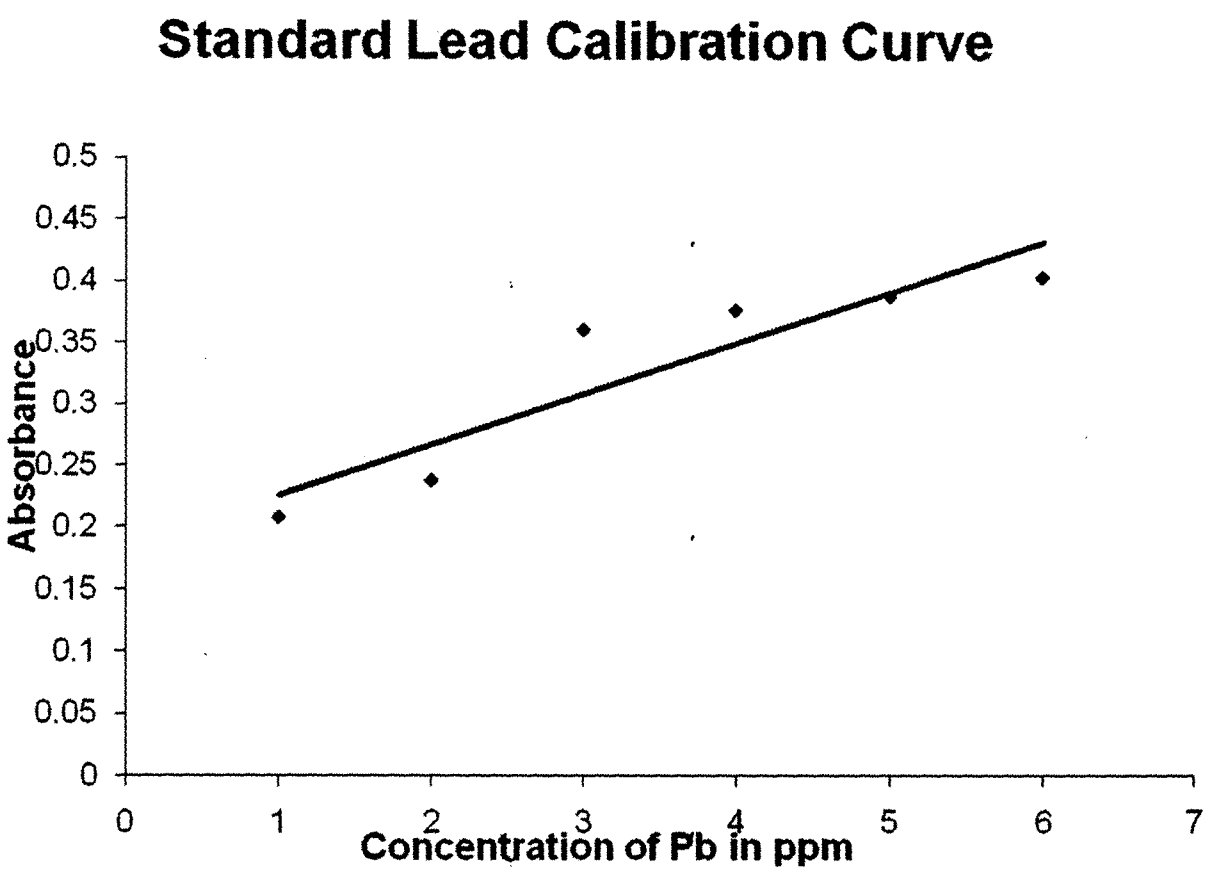
APPENDIX D4


Standard Copper Calibration Curve
Absorbance
0.025
0.02
0.015
0.01
0.005
0
0
2
4
6
8
10
12
Concentration of Cu in ppm

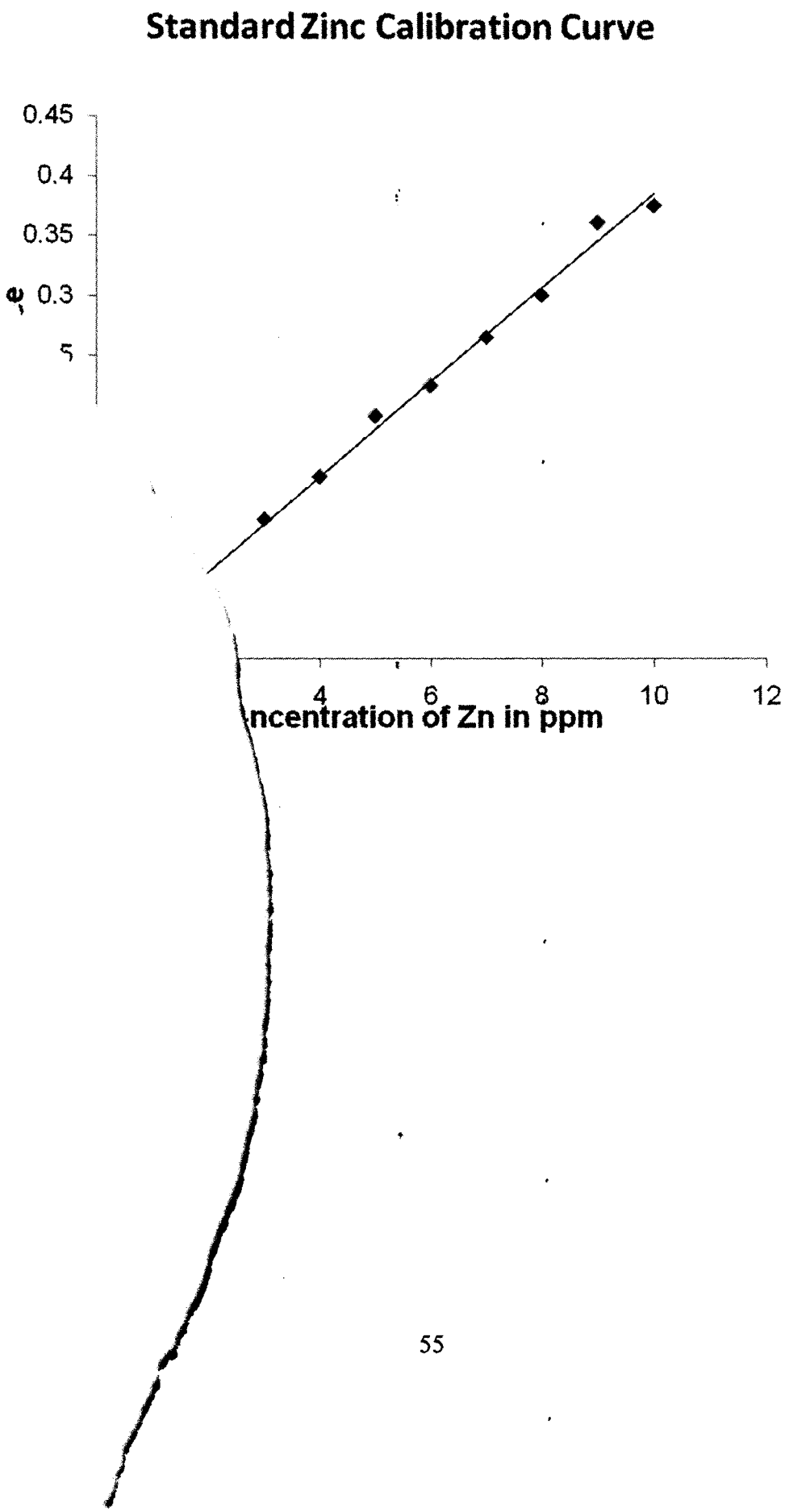
APPENDIX D5


Standard Sodium Calibration Curve
%Emission
120
100
80
60
40
20
0
0
20
40
60
80
100
120
Concentration of Na in ppm

APPENDIX D6


Standard Lead Calibration Curve
Absorbance
0.5
0.45
0.4
0.35
0.3
0.25
0.2
0.15
0.1
0.05
0
0
1
2
3
4
5
6
7
Concentration of Pb in ppm

APPENDIX D7


Standard Zinc Calibration Curve
0.45
0.4
0.35
0.3
4
6
8
10
12
ncentration of Zn in ppm