

Effect of Selected Preservatives on Nutritional Indices of *Phaseolus vulgaris* (Kidney Beans) and *Phaseolus lunatus* (Lima Beans)

Aworinde Olusola Abosedé¹, Nweze Carole Chibuzor²,
Bamidele Titilayo Oluwayemisi³, Nwefia Wilson Ifeanchó⁴

⁴corresponding author, PhD (in view), Department of Biochemistry and Molecular Biology, Nasarawa State University, keffi, Nigeria.

¹Biochemist, Nasarawa state University Keffi, Nigeria

²Nutritionist, Biochemist (PhD), Nasarawa state University Keffi, Nigeria.

³Biochemist (Professor of Nutritional Biochemistry), Nasarawa state University Keffi, Nigeria.

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Abstract: Background: The increasing necessity for all season food availability as well the importance of nutritional value being preserved properly have resulted in the use of both natural and artificial preservatives. This research aimed at evaluation of the effect of selected preservatives on the nutritive indices of *Phaseolus vulgaris* (Kidney Beans) and *Phaseolus lunatus* (Lima Beans).

Materials and methods: The *P. vulgaris* and *P. Lunatus* used for this research were obtained in sac bags. In accordance with the research protocol. The both beans seeds were prepared and sent for preliminary analysis in laboratory before storage with selected preservatives proceed. The selected preservatives were procured and prepared for the preservation purpose following the research guidelines. The beans seeds were divided into airtight vessels and label it group A to E which were preserved with selected preservatives, DDVP (sniper), birds eye pepper, Aluminum phosphide, the group D was mixed with ash respectively while the fifth (group E) served as control and were left untreated with any preservative. The beans mixed with preservative were left to stay for 6 months. Standard methods were used to assess the effect of the preservatives on the proximate constituents, mineral concentrations and amino acid contents respectively compared with the control which was not mixed with any of the preservatives.

Results: It was observed from the results that the groups preserved with sniper and wood ash had lowest and highest % moisture content in both beans respectively, the crude proteins of the aluminum phosphide preserved beans showed a non-significant improvement in it % composition compared with control, there was also a non-significant ($p \leq 0.05$) increase seen in the crude protein of the preserved groups when compared with control. For the crude fibre compositions it is observed that sniper preserved showed a significantly high content, with wood ash preserved group having the lowest % when compared with control. For the carbohydrate all group preserved with the selected showed a statistically significant improvement with sniper preserved with highest value of compared with control. It was observed from the result also that pepper preserved group had a significantly higher Ca concentration of while highest phosphorus significant value was observed in pepper preserved group. The results showed that kidney beans and lima beans are rich in essential amino acids with Lysine showing the highest concentration among the essential amino acid with a value of 8.17 g for the pepper preserved and was noticed to be same across all the essential amino acids while wood ash preserved group show dis-improvement in amino acid concentration.

Conclusion: The observed improvements in some parameters showed that the chemical in some groups are more effective at some aspect while natural preservatives at some other points. Its effectiveness for a longer shelf life and stop or delay the growth of bacteria, suppress the reaction when food comes in contact with oxygen or heat, they also prevent the loss of some essential amino-acids and some vitamins enhance the food flavors and colors. The results of this study improvements in shelf life and nutritional values.

Keywords: preservatives, nutrition, aluminum, artificial, proximate analysis, minerals, vitamins, amino acid.

I. INTRODUCTION

Legumes are plants belonging to the family Leguminosae also called as Fabaceae that produce seeds within a pod [1]. Common legumes used for human consumption include lima beans, peas, broad beans, lentils, soybeans, lupins, lotus, sprouts, mung bean, green beans and peanuts and are referred to as grain legumes or food legumes [2]. These seeds are valued worldwide as an inexpensive meat alternative and are considered the second most important food source after cereals [2]. Legumes are very essential especially when in terms of nutrition. They contain the essential amino acids that are needed for building proteins, complex carbohydrates, dietary fibre, unsaturated fats, vitamins and essential minerals for the needed for human diet [3], [4]. In addition to their nutritional superiority, legumes have also been ascribed economical cultural, physiological and medicinal roles owing to their possession of beneficial bioactive compounds [5].

Phaseolus lunatus is commonly known as Lima beans, butter bean, sieva bean or double bean, is a leguminous plant that originated from Peru in western South America. Its name was derived from Peru's capital city "Lima". In Nigeria it is known as "kapala" in Yoruba land, Waken rumfa" in Hausa language, "ukpa" (Igbo) in South western and South-eastern Nigeria respectively [6]. Lima beans are underutilized in many parts of tropical Africa in all likelihood due to its hard-to-cook phenomenon. There is also poor information on its potential as a good food source rich in protein [7]. The seeds contain 24% proteins, 61% carbohydrate and some minerals elements.

The ukpa variety is characterized by large rhomboid shaped flattened seeds. It has white cotyledons and the white seed coat bears black streaks radiating from the dark hilum giving it a zebra-skin coloration. *P. lunatus* production is hindered by pest infestation which reduces its quality and also its quantity. To this effect, pesticide comes into play to kill these pests. [8]

The use of pesticides in grains storage becomes more intensive in the Northern part of Nigeria especially at Doma grains market in Nasarawa State, which is viewed as the largest grain market in Nigeria. The *P. lunatus* seed farmers and traders' ignorance about pesticide toxicity led to its misuse: unguided application, inadequate usage, etc. Dichlorvos and Aluminum phosphide are one of the classes of insecticides referred to as organophosphates used to control households and stored products insects. It is effective against mushroom flies, aphids, spider mites, caterpillars, thrips, and white flies in greenhouse, outdoor fruits, and vegetable crops [9]. It acts against insects as a contact and stomach poison. Dichlorvos pesticide self-poisoning is an important clinical problem in the developing world, and kills an estimated 200,000 people every year. Concentrates of Dichlorvos is mildly irritating to skin and may cause localized sweating, involuntary muscle contractions and burning sensations or actual burns. When inhaled, the first effects are usually respiratory and may include bloody or runny nose, coughing, chest discomfort, difficult or short breath, and wheezing due to constriction or excess fluid in the bronchial tubes. When in contact with eyes will cause pain, bleeding, tears, pupil constriction, and blurred vision [10].

a. *Phaseolus Vulgaris* (Kidney Beans)

Phaseolus vulgaris is among the common bean species. It is an herbaceous annual plant grown worldwide for its edible dry seed. Its botanical classification, along with other *Phaseolus* species, is as a member of the legume family Fabaceae, most of whole member acquire the nitrogen they require through an association with rhizobia, a species of nitrogen-fixing bacteria [11]. It is brownish in colour and it was originated in central and south America. Small-seeded and climbing ecotypes are found in the wild in north Argentina and Central America.

i. Nutritive values of *p. Vulgaris* (kidney beans)

P. vulgaris are a rich source of protein and carbohydrates, as well as minerals and vitamins. This diet complements the mainly cereals diet in countries that grow cowpea as a major food crop. Kidney beans are rich in calcium, magnesium, phosphorus, most especial it also contains iron. It also has a small amount of sodium, zinc, copper, manganese [4]. They

are also an excellent source of resistant starch which is broken down by bacteria in the large intestine to produce short-chain fatty acid used by intestinal cells for food energy [12]. It contains calories of 378.9 kcal, total carbohydrates of 59.0 g, fat of 1.2 g, protein of about 25.0 g, vitamin c of 1.3 mg, calcium of 40 mg and iron of about 1.8 mg. It has a better nutritive value than cereal straws because it has higher protein content 61% DM of NDF vs 51% for a leaf-rich straw (Callaway, 2004).

b. *Phaselous Lunatus* (Lima Beans)

There are many varieties of *Phaseolus lunatus*, each with feature colour characteristics such as black, dark red, mottle black, white, brown, cream, mottle brown and dark brown. The pod of lima bean is flat, oblong and slightly curved, averaging about three inches in length. Within the pod sides, are four to six flat kidney-shaped seeds that are generally referred to as lima beans. In Nigeria, lima bean is one of the legumes cultivated in the eastern and northern part of the country [4]. However, the hard to cook phenomenon experienced with lima bean, especially after long storage period and the toxic component cyanogenic glycosides) are the major constraints to the cultivation and utilization of this legume. The cooking water must be discarded and replaced at intervals during cooking to reduce its toxic component [7].

i. Nutritive Value of Lima Bean

Lima bean is nutrient dense and their richness generates feelings of satiety. These sweet tasting bean has a rich, starchy, meaty texture and a creamy, distinctive flavour. In fact, lima beans are a good source of B-complex vitamins (vitamins B6, niacin and folate), protein (including the important amino acid lysine), fiber, especially soluble fibre in the form of pectin, iron, potassium and magnesium. They have very little fat. Studies, suggest that intake of lima bean lowers low density lipoprotein cholesterol levels, probably due to their soluble fibre content. Lima bean contains the phytochemicals coumestrol and saponins, compounds that may impart anticancer benefits [13]. National Academy of Science - NAS (1979) assessed the nutrient value of some lima bean varieties and concluded that protein content of lima bean ranged from 21.4 to 36.1%, fat 1.00 to 2.2%, ash 03.4 to 17.9%, fibre 01.7 to 04.9% and carbohydrate 55.6 to 73.2% depending on the variety. Lima bean, like many other legumes constitute a rich source of niacin, thiamin and riboflavin (Sathe *et al*; 1984). Lima bean contains high levels of potassium, phosphorus, calcium and iron. The major form in which phosphorus exists in the bean is in the form of phytate phosphorus [14,15].

ii. Health Benefits of Lima Bean

Lima bean is very rich and a good source of cholesterol-lowering fibre, as are most other legumes. In addition to lowering cholesterol, lima bean has high fibre content that prevents blood sugar levels from rising too rapidly after a meal. This qualifies these beans an 9 especially good choice for individuals with diabetes, insulin resistance or hypoglycemia [16]. When combined with whole grains such as rice or wheat, lima bean provides virtually fat free high quality protein [18]. It is a good source of protein for vegetarians because its combination with whole cereals provides protein comparable to that of meat or dairy foods without high calories or saturated fat, found in these foods.

c. Antioxidant

Lima beans have antioxidant properties, reported in unprocessed beans [13]. protein hydrolysate, dietary fibre [14], and lectin. Many phytochemical compounds present in lima beans have been reported to have antioxidant properties such as bioactive peptides, and phenolic compounds including quercetin [14], gallic acid, catechin, rutin. DPPH radical scavenging activity of the fresh seed ranged from around 2.6 to 94.4 %. The ·OH scavenging activity of baby lima bean was found to be as high as 74.97 %. The lectin, PLUN of a Brazilian lima bean variety (P. lunatus var. cascavel) showed antioxidant activity [7], which may be attributed to the structure of the lectin and its constituent amino acids [9]. Dietary fibre is another constituent of lima bean that has antioxidant properties. It works by trapping free metals and forming metal complexes that act as reducing agents. The antioxidant activity of *P. lunatus* fibrous residue was found to be 35.6% at 72 h. This is comparable to the 35% antioxidant activity of vitamin E [8].

d. Food Preservation

Preservation of Food is the process of treating and handling food to stop or greatly slow down spoilage (loss of quality, edibility or nutritive value) caused or accelerated by micro-organisms. Some methods, however, use benign bacteria, yeasts or fungi to add specific qualities and to preserve food (e.g. cheese, wine). Maintaining or creating nutritional value, texture and flavour is important in preserving its value as food. This is culturally dependent, as what qualifies as food fit

for humans in one culture may not qualify in another culture. Preservation usually involves preventing the growth of bacteria, fungi, and other micro-organisms, as well as retarding the oxidation of fats which cause rancidity. [15].

Natural Preservation: From ages humans have been using preservatives to extend the shelf life of various foods, making them last longer and keeping their colour, taste and nutrients intact. These days' foods come with a lot of imitation preservatives, but there are several natural preservatives that you can use to preserve food as well. Conference Series Ltd invites all the participants from all over the world to attend Food & Nutrition conference during May 22-24, 2017 at Las Vegas, Nevada, USA which includes prompt Keynote Presentations, Oral talks, Poster Presentations Young Research Forum and Exhibitions. Some well-known speaker like Zhaowei Zhang, Ministry of Agriculture is going to deliver his speech regarding "Simultaneous detection for multiplexed mycotoxins by using immunoassay and confirming methods in food" [18].

e. Artificial Preservatives

The artificial food preservatives sometimes act as the antioxidants, They make their food more acidic, They reduce the moisture level of the food, They slow down the ripening process and they prevent the microbes growth, Not all of these additives are 100-percent safe and artificial food preservatives help the food stay for longer period. You have to know that there are many food additives particularly the nitrites, the aspartame, the saccharin and the benzoates which have been linked to the cancer because they produce the carcinogenic compounds when they are metabolized. [19]. Example the use of sodium and Aluminum phosphide.

f. Pest and Control of *P. Lunatus*

Insects are one of the important factor in the low yields of crops developing countries like Nigeria, and they affect each tissue component and developmental stage of the plant. In bad infestations, insect pressure is responsible for over 90% loss in yield. The legume pod borer, *Maruca vitrata*, is the main pre-harvest pest of the *P. lunatus*. *M. vitrata* causes the most damage to the growing *P. lunatus* due to their large host range and distribution. It causes damage to the flower buds, flowers, and pods of the plant, with infestations resulting in a 20–88% loss of yield [20].

The beans weevil *Acanthoscelides obtectus* Say (Coleoptera: Bruchidae) commonly known as bruchids is a significant pest of legumes, especially beans, in some part of the world attacking crops in the field and in warehouses. Ahmed *et al.*, reported that *A. obtectus* is a serious Neotropical origin insect pest on kidney beans, *Phaseolus Lunatus*. and other legumes seeds. It is a destructive pest of stored *P. Lunatus*. The insect larvae begin feeding from the embryo and eventually consumes the entire seed, making the grain hollow and only the seed coat [21]. In severe infestation, the infested seeds are filled with frass, cast skins and excreta, which adversely deteriorate the quality of the grain. Hence, a common trend of zero-tolerance by buyers is increasing. Infestation by insect produces unpleasant odors, dirty appearance and abhorrent taste due to contamination by insect fragments and excretion. Severe infestation also makes seeds unpalatable. The quality of the grain may decrease due to nutrient depletion. Because of the damaging effect of this weevil on stored beans, farmers are indirectly forced to make use of preservatives to prevent the negative effects of the pests and avoid post-harvest price collapse, marked seasonal price fluctuation and reduction in market value.

g. Some Selected Preservatives

i. Sniper Pesticide (Dichlorvos)

Dichlorvos or 2,2-dichlorovinyl dimethyl phosphate (DDVP) is an organophosphate, widely used as an insecticide to control household pests, in public health, and protecting stored product from insects. The compound has been commercially available since 1961 and has become controversial because of its prevalence in urban waterways and the fact that its toxicity extends well beyond insects [22]. It is used as a fumigant and has been used to make pet collars and pest strips. It is available as an aerosol and soluble concentrate. Dichlorvos is an insecticide with contact, respiratory and stomach action. Like many organophosphorus insecticides, it also inhibits the enzyme cholinesterase, which results in disruption to the nervous and muscular system.

As a synthetic organophosphorus but which many Nigerians have converted to an indoor insecticide. A person may be exposed to the associated risk of DDVP through inhalation, absorption through the skin, ingestion and eye contact. According to CDC effects of exposure may include irritation of eyes, skin, miosis, ache eyes; rhinorrhea, (discharge of thin nasal mucus); headache; chest tightness, wheezing, laryngeal spasm, salivation, cyanosis, anorexia, nausea, vomiting,

diarrhea, sweating, muscle fasciculation, paralysis, dizziness, ataxia; convulsion; low blood pressure, cardiac irregularity. The popularity of sniper used among Nigerians as an insecticide has grown in recent times owing to its effectiveness in killing insect better than well-established brand of insecticide. The demand is also fueled by its affordability.

ii. Aluminum phosphide

Aluminum phosphide are fumigants used primarily for indoor fumigation of raw agricultural commodities, animal feeds, processed food commodities, and non-food commodities in sealed containers or structures to control insects, and for outdoor fumigation of burrows to control rodents and moles in non-domestic areas, no crop land, and agricultural areas. Aluminum phosphide also is used to control rodents which can carry various diseases including sylvatic plague.[24]

a. Aluminum phosphide as perseverative

Aluminum phosphide is registered for use on the following food/feed crops: temporarily stored feed/food commodities, barley, buckwheat, corn, millet, oats, rice, wild rice, rye, sorghum, triticale, wheat, peanuts, soybeans, beans, vegetables, dates, figs, almonds, brazil nuts, cashews, filberts, pecans, pistachios, walnuts, grass forage/fodder/hay, popcorn, rape, sesame, cocoa, coffee, legume vegetables, safflower seeds, and sunflower. It is also registered for use in orchards, commercial transportation facilities and commercial shipping containers, food processing plants (nonfood contact), silos, commercial storehouses/warehouses, and grain/cereal/flour bins and elevators. It is also used on diseased or empty beehives.

h. Target Pests:

The pests that are controlled by aluminum phosphide are africanized honey bee, almond moth, angoumois grain moth, bean weevil, diseased bees, cadelle, cereal leaf beetle, cigarette beetle, confused flour beetle, dermestid beetles, driedfruit beetle, driedfruit moth, european grain beetle, flat grain beetle, flatheaded grain beetle, fruit flies, granary weevil, greater wax moth, hairy fungus beetle, hessian fly, honey bee, indian meal moth, khapra beetle, lesser grain borer, maize weevil, Mediterranean flour moth, mosquitos, pea weevil, pink bollworm, raisin moth, red flour beetle, rice weevil, rust red grain beetle, rusty grain beetle, sawtoothed grain beetle, spider beetles, tobacco moth, tracheal mite, wax moth, yellow meal worm, chipmunks, gophers, ground squirrels, house mice, marmot, meadow vole, mice, moles.

i. Acute Toxicity

Phosphine gas produced from aluminum phosphide has been tested for acute toxicity by the inhalation route of exposure. No significant exposure to phosphine gas are expected via the oral or dermal routes. Results of the acute inhalation toxicity study (MRID# 41377001) found that the LC50 is greater than 11 ppm (approximately 0.014 mg/L), placing it in Toxicity Category I.

j. Dietary Risk Assessment

Dietary exposure to aluminum phosphide can potentially occur via residues of phosphine gas remaining in treated commodities. For all data submitted to the Agency for establishment of food tolerances, residues of phosphine gas have been typically reported as non-detectable.

However, because the use of aluminum phosphide is considered a food use, tolerances for phosphine gas are required. The tolerances are intended for enforcement purposes, i.e., to monitor and safeguard against misuse, not for risk assessment.

II. MATERIALS AND METHODS

The study involved the use of Dichlorvos, Aluminum phosphide, ash and pepper as preservatives of *P. vulgaris* and *P. Lunatus*. This is to determine the effect of the use of these preservatives on the Nutritional indices of *P. vulgaris* and *P. Lunatus*. In all cases the effects in preserved groups are compared with that of control for appropriate decisions to be taken.

Sample Collection and Processing

i. Collection of Samples:

The matured *P.vulgaris* and *P. Lunatus* were collected from the area of cultivation in Doma, Doma Local Government Area, Nasarawa State, Nigeria. The sample was collected in bags and transported to laboratory. The samples was cleaned and sorted to remove stones and dirt.

ii. Identification of Samples

The samples were identified by an academic staff of the department of Plant science and Biotechnology, Faculty of Science, Nasarawa University Keffi, Nigeria.

iii. Wood-ash preparation

The stem from neem tree (*Azadirachta indica*) was burnt to ash. The cooled ash was then sieved to remove dirt and 150g weighed and packed into nylon bags.

iv. Pepper Preparation

Fresh birds eye pepper (*Capsicum frutescens*) purchased from the market was dried in the sun. 150g of the sun dried pepper was weighed and packed in to nylon bags.

v. Procurement of Chemicals

Sniper (Dichlorvos or 2,2-dichlorovinyl dimethyl phosphate (DDVP)) and Aluminum phosphide were purchased from standard agro-allied store in Keffi, Nasarawa state.

vi. Seed preservation

The cleaned seeds were divided into two sections, one for *phaseolus lunatus* and the other for *phaseolus vulgaris*. The first section containing *phaseolus lunatus* was divided into four parts, each having a bucket with tight lids that contained seeds weighing about 200g.

A bucket of lima beans seed mixed with DDVP, the second contained lima beans mixed with birds eye pepper, the third bucket contained lima beans mixed with ash. The fourth bucket contained lima beans with no preservative/treatment served as control.

The second section which contained *phaseolus vulgaris* were divided into five parts, each having a bucket with tight lids that contained seeds weighing about 200g.

The first bucket contained kidney beans mixed with DDVP, the second, kidney beans mixed with birds eye pepper while the third bucket contained kidney beans mixed with Aluminum phosphide. The fourth bucket also contained kidney beans mixed with ash and the fifth kidney beans seed with no preservative/ treatment which served as control. The seeds were stored for a period of six (6) months and properly labeled. During the storage period the seeds were checked periodically.

vii. Processing of the beans

Each treatment including the control was milled into powder, packed in a clean polythene bag, labeled and sealed. The powdered bean was kept for analysis.

A. PROXIMATE COMPOSITION ANALYSIS

The powdered samples were analyzed for moisture, protein, fat, ash, fiber and nitrogen free extract using methods as described by AOAC 2005 and Carbohydrate was obtained by subtracting the sum of other proximate parameters from 100.

i. Determination of Moisture content.

Principle

The principle of the thermogravimetric method of moisture content determination is defined as the weight loss of mass that occurs as the material is heated. The sample weight is taken prior to heating and again after reaching a steady-state mass subsequent to drying.

Method

The moisture content was determined using AOAC (2006) by weighing out 2g into glass petri dish, which has been previously dried and weighed. The dish including the sample inside it was placed in hot air oven for 5 hours at $130 \pm 3^{\circ}\text{C}$. Finally, the sample was dried to constant weight, cool for ten minutes in a desiccator each time before weighing.

$$\% \text{ Moisture} = \frac{\text{Wt loss on drying, (g)} \times 100}{\text{Wt test portion, g()}}$$

W_t = Weight of sample

g = gram

ii. Determination of Ash Content

Principle

When foods and food products are heated to temperatures of 500-600°C, the water and other volatile constituents are evolved and the organic constituents are burned in the presence of oxygen of the air to carbon dioxide and oxides of nitrogen and also eliminated together with hydrogen as water. The mineral constituents remain in the residues as oxides, sulfates, phosphates and chlorides and this inorganic residue constitutes the ash of food.

Method

Ash content was determined using AOAC (2005). A clean empty crucible was placed in a muffle furnace at 600°C for an hour, cooled in desiccator and then weight of empty crucible is noted (W_1). 2g of each of sample was taken into the crucible (W_2). Then the crucible was placed in muffle furnace at 600°C for 2 hours. The crucible was cooled in a desiccator and weighed (W_3). Percent ash was calculated by following formula:

$$\% \text{ Ash} = \frac{\text{weight of test portion g} - \text{weight loss on ashing g}}{\text{weight of test portion}} \times 100$$

iii. Nitrogen determination by micro Kjeldahl method (crude protein)

Principle

The nitrogenous compounds of the food material are converted into ammonium sulphate by digesting with concentrated Sulphuric acid in presence of digestion mixture which acts as a catalyst. The clear digest after dilution is made basic with strong alkali, and the liberated ammonia is steam distilled into boric acid.

Method

A known weight (200mg) of sample was placed in Kjeldahl flask and about 200 milligram of catalyst mixture (potassium sulphate, copper sulphate and selenium powder) was added.

10.0cm² of concentrated sulphuric acid was also added to the content of the flask. Heated gently for few minutes until frothing ceases and increase the heat to digest for 1 hour. Allowed to cool and made to 100cm³ volume with distilled water.

10.0cm³ aliquot of the dilute solution of the digest was distilled by pipetting the volume into distillation chamber of micro Kjeldhal distillation apparatus. 10.0cm³ of 40% sodium hydroxide solution was added and steam distil into 10.0cm³ of 4% boric acid containing mixed indicator (note colour from red-green) titrate with standard 0.01N or 0.02N hydrochloric acid to grey end point.

$$\% \text{ N} = \frac{(a-b) \times 0.01 \times 14.0057 \times c \times 100}{d \times e}$$

a = titre value for the sample

b = titre value for the blank

c = Volume to which digest is made up with distilled water

d = Aliquot taken for distillation

e = Weight of dried sample (mg)

To convert to % crude protein, multiply by necessary conversion factor (6.25).

iv. Fat Determination of Crude Fat

Principle:

Fats are characterized by the readiness with which they are extracted by ethyl ether, petroleum spirit, carbon disulfide etc. Hence these organic solvents are used for extracting fats from food samples. These solvents also extract small amounts of substances other than the fat and the result is generally designated as “crude fat” or “ether extract”.

Method:

A soxhlet extraction apparatus was used and a 250ml quickfit flask which was previously dried in the oven. 5gm of sample was weighed and transferred to a fat-free extraction thimble, plugged lightly with cotton wool. The thimble was placed in the extractor and about 150cm³ of petroleum ether (B.P. 40-60°C) added into the flask until it siphons over once. The source of heat was adjusted (electrothermal heating mantle) so that the ether boils gently and was left to siphon over for at least 6 hours. The flask (which now contains all the oil) was detached. The extract (oil) was filtered through Whatman filter paper into a previously weighed beaker, washing paper finally with small portion of hot fresh ether. Solvent was evaporated at 100°C and the beaker containing residue dried in an air oven for 1 hour at 100-105°C.

$$\% \text{ Crude Fat} = \frac{\text{Weight of ether extract} \times 100}{\text{Weight of sample}}$$

v. Crude Fibre Determination

Principle

The sample is allowed to boil with 1.25% dilute H₂SO₄, washed with water, further boiled with 1.25% dilute sodium hydroxide and the remaining residue after digestion was taken as crude fibre.

Method

The defatted ground sample was transferred from fat determination into 250ml quickfit flask, 150ml of 1.25% sulphuric acid was added and fit into reflux condenser. Reflux was done for 30 minutes, cooled and filtered using Buchner funnel fitted with Whatman filter paper. Rinsed three times with hot distilled water, dried and carefully transferred the residue into quickfit flask. 150ml of 1.25% Sodium hydroxide was added and reflux for 30 minutes. Filtered using Buchner funnel, rinsed three times with hot distilled water, once with 1.25% sulphuric acid and finally with 95% ethanol. The filter paper containing residue was removed and placed into porcelain crucible. It was dried in oven for 2 hours at 130°C. Cooled in dessicator, ash at 550⁰±10⁰C in murfle furnance, cooled in dessicator and weighed.

$$\% \text{ Crude Fiber} = \frac{W_1 - W_2 \times 100}{W_0}$$

vi. Micronutrient Analysis

Principle

Atomic absorption spectrometry (AAS) detects elements in either liquid or solid samples through the application of characteristic wavelengths of electromagnetic radiation from a light source. Individual elements will absorb wavelengths differently, and these absorbances are measured against standards. In effect, AAS takes advantage of the different radiation wavelengths that are absorbed by different atoms.

vii. Determination of Minerals

Determination of minerals such as Iron (Fe), Zinc (Zn), Calcium (Ca), Potassium (K) and Magnesium (Mg) was carried out using Atomic Absorption Spectrometry. Bean samples used were analyzed following the methods of the AOAC method. Samples was first acid-digested and analyzed using the atomic absorption spectrophotometer as described below. To 5.0g of powdered samples was added 30ml of concentrated Nitric acid (HNO) ≥ 95.1% purity. The flask was placed in the dark overnight. Afterwards, 40ml of perchloric acid (HClO₄) ≥ 98% purity was added. The mixture was then

initially heated on a hot plate at 50°C for 15 min. afterwards, the temperature of heating was gradually increased to 200°C. Heating continued until the white dense fumes of perchloric acid disappeared. After digestion, the contents were cooled, filtered through What man filter paper, filtrate transferred to a 100 ml volumetric flask and then made up to the 100 ml mark with deionized water.

Preparation of the standard curve: Dissolve 0.2195g of pure dry KH_2PO_4 in 1 litre of distilled water. This solution contains 50 µg P/ml. Preserve this as a stock standard solution of phosphate. Take 10 ml of this solution and dilute it to 0.5 litres with distilled water. This solution contains 1 µg P/ml (0.001 mg P/ml). Put 0, 1, 2, 4, 6 and 10 ml of this solution in separate 25-ml flasks. Add to each flask, 5 ml of the molybdate reagent; and dilute with distilled water to about 20 ml. Add 1 ml of dilute SnCl_2 solution, shake again and dilute to the 25-ml mark. After 10 minutes, read the blue colour of the solution on the spectrophotometer at a wavelength of 660 nm. Plot the absorbance reading against “µg P” and connect the points.

Sample preparation: Take 1 g of plant sample and digest with 10 mL concentrated nitric acid, filter the digest and make the volume up to 100 ml.

Development of colour: Take 5 ml of the digested solution a 25-ml volumetric flask; deliver 5 ml of the molybdate reagent with an automatic pipette, dilute to about 20 ml with distilled water, shake and add 1 ml of the dilute SnCl_2 solution with a bulb pipette. Fill to the 25-ml mark and shake thoroughly. Read the blue colour after 10 minutes on the spectrophotometer at 660 nm after setting the instrument to zero with the blank prepared similarly but without the sample.

C = concentration of P (µg/ml) as read from the standard curve;

df = dilution factor, which is $100 \times 5 = 500$, as calculated below:

1 g of sample made to 100 ml (100 times); 5 ml of sample solution made to 25 ml (5 times).

1 000 000 = factor for converting µg to g.

viii. Determination of Amino Acid Profile [26]

Principle

PTH amino acid Analyzer (HPLC) automatically analyzes phenylthiohydantoin (PTH) amino acids derived from Edman degradation of proteins and peptides.

Edman degradation, developed by Pehr Edman, is a method of sequencing amino acids in a peptide. In this method, the amino-terminal residue is labeled and cleaved from the peptide without disrupting the peptide bonds between other amino acid residues.

Method

The sample was dried at 70°C to constant weight, defatted, hydrolyzed, evaporated in a rotary evaporator and loaded into the Applied Biosystems PTH Amino Acid Analyzer.

Defatting Sample:

The sample was defatted using chloroform/methanol mixture of ratio 2:1. About 4g of the sample was put in extraction thimble or wrapped in filter paper and extracted for 15 hours in soxhlet extraction apparatus.

III. RESULTS

a. PROXIMATE COMPOSITION OF KIDNEY BEANS

Table 2 presents the effect of selected preservatives on the proximate composition of kidney beans. The crude protein value of lima beans showed significant differences with ranges from 19.14-21.52%. The highest crude protein was found in the sample preserved with aluminum phosphate which is followed closely with the control showing a value of 21.49% and 20.82% with kidney beans preserved with wood ash.

Table 1. Effect of selected preservative on proximate composition of Kidney beans (%)

	Crude protein	Crude fat	Crude fibre	Moisture	Ash	Carbohydrate
KA	21.31 ± 0.07 ^c	4.35 ± 0.02 ^a	5.15 ± 0.04 ^b	1.21 ± 0.09 ^a	5.11 ± 0.03 ^e	62.87 ± 0.12 ^d
KB	19.14 ± 0.03 ^a	4.56 ± 0.02 ^b	9.92 ± 0.21 ^d	2.28 ± 0.09 ^b	4.42 ± 0.05 ^d	59.68 ± 0.16 ^b
KC	21.52 ± 0.05 ^d	4.64 ± 0.04 ^b	8.38 ± 0.25 ^c	2.89 ± 0.07 ^c	4.07 ± 0.04 ^c	58.50 ± 0.21 ^a
KD	20.82 ± 0.04 ^b	5.82 ± 0.10 ^c	3.43 ± 0.04 ^a	8.06 ± 0.14 ^e	3.15 ± 0.03 ^a	58.72 ± 0.16 ^a
KE	21.49 ± 0.09 ^c	4.57 ± 0.03 ^b	3.83 ± 0.06 ^a	4.75 ± 0.12 ^d	3.43 ± 0.02 ^b	61.93 ± 0.21 ^c

Mean ± SEM of three replications, KA - Kidney beans preserved with sniper

KB - Kidney beans preserved with pepper, KC - Kidney beans preserved with aluminum phosphate, KD - Kidney beans preserved with wood ash, KE - Kidney beans preserved with control.

b. Proximate Composition of Lima Beans

The results of proximate composition of Lima beans preserved with some selected preservatives is shown in table 2.

Table 2. Effect of preservative on proximate composition of Lima beans (%)

	Moisture	Crude Protein	Crude Fibre	Crude Fat	Ash	Carbohydrate
LA	4.37 ± 0.05 ^a	18.44 ± 0.05 ^a	4.32 ± 0.01 ^b	4.13 ± 0.04 ^c	4.89 ± 0.01 ^b	63.85 ± 0.10 ^c
LB	5.49 ± 0.11 ^b	19.30 ± 0.11 ^b	3.39 ± 0.09 ^a	2.72 ± 0.02 ^b	4.72 ± 0.02 ^a	64.37 ± 0.13 ^d
LC	4.56 ± 0.03 ^a	19.59 ± 0.07 ^c	5.07 ± 0.03 ^c	2.59 ± 0.04 ^a	5.73 ± 0.03 ^d	62.47 ± 0.10 ^b
LD	6.91 ± 0.11 ^c	18.39 ± 0.09 ^a	4.53 ± 0.06 ^d	4.36 ± 0.03 ^d	5.10 ± 0.04 ^c	60.70 ± 0.12 ^a

Mean ± SEM of three replications, KA - Kidney beans preserved with sniper

KB - Kidney beans preserved with pepper, KC - Kidney beans preserved with aluminum phosphate, KD - Kidney beans preserved with wood ash, KE - Kidney beans preserved with control.

Mineral Composition of Kidney Beans

Table 3 shows the effect of selected preservative on mineral composition of kidney beans.

The calcium content of kidney beans showed the lowest in kidney beans preserved with aluminum phosphate (7.42mg/1000g) and the highest in kidney beans preserved with sniper (12.92mg/1000g).

Table 1. Effect of selected preservative on mineral composition of kidney beans (mg/1000g)

	Ca	Fe	Zn	K	Mg
KA	12.92 ± .06 ^e	4.37 ± 0.03 ^c	0.63 ± 0.002 ^b	104.89 ± 0.33 ^a	73.46 ± 0.04 ^d
KB	9.74 ± .02 ^c	3.98 ± .07 ^b	0.55 ± 0.01 ^a	192.23 ± 0.10 ^d	55.71 ± 0.05 ^a
KC	7.42 ± .03 ^a	3.78 ± .02 ^a	1.48 ± 0.01 ^d	126.54 ± 0.17 ^c	137.93 ± 0.55 ^e
KD	7.75 ± .04 ^b	5.24 ± .02 ^e	0.63 ± 0.02 ^b	107.61 ± 0.12 ^b	64.44 ± 0.10 ^b
KE	10.58 ± .02 ^d	4.08 ± .02 ^b	0.71 ± 0.01 ^c	104.38 ± 0.09 ^a	69.35 ± 0.06 ^c

Mean ± SEM of three replications, KA - Kidney beans preserved with sniper, KB - Kidney beans preserved with pepper, KC - Kidney beans preserved with aluminum phosphate, KD - Kidney beans preserved with wood ash, KE - Kidney beans preserved with control.

Table 2. Effect of selected preservatives on mineral composition of Lima beans (mg/1000g)

	Ca	Fe	Zn	Mg	K
LA	8.27 ± 0.08 ^a	2.58 ± 0.21 ^a	0.46 ± 0.01 ^a	64.26 ± 0.19 ^a	107.44 ± 0.24 ^b
LB	16.66 ± 0.25 ^d	3.87 ± 0.32 ^c	0.57 ± 0.03 ^b	72.64 ± 0.28 ^c	105.39 ± 0.11 ^a
LC	10.92 ± 0.15 ^c	3.10 ± 0.08 ^a	0.46 ± 0.02 ^a	74.92 ± 0.35 ^d	109.40 ± 0.09 ^d
LD	8.85 ± 0.16 ^b	3.38 ± 0.02 ^b	0.49 ± 0.01 ^a	67.06 ± 0.16 ^b	108.44 ± 0.07 ^c

Mean $\pm$ SEM of three replications, LA -Lima beans preserved with sniper, LB- Lima beans preserved with pepper, LC - Lima beans preserved with wood ash, LD -Lima beans preserved with control

Amino Acid Composition of Lima Beans

Table 5 represents the effect of selected on amino acid composition of lima beans. Lima beans is rich in amino acids. Cysteine has the lowest concentration which is expected in legumes while glutamic acid has the highest concentration. Lima beans preserved with pepper shows the highest concentration of amino acids when compared with lima beans preserved with sniper, wood ash and the control except for phenylalanine, lysine, tryptophan and cysteine. Leucine shows the highest concentration amid the essential amino acid.

Table 3. Effect of selected preservatives on amino acid composition of Lima beans (g/100g)

	LA	LB	LC	LD
Leucine	7.69 $\pm$ 0.09 ^{ab}	7.97 $\pm$ 0.07 ^b	7.61 $\pm$ 0.13 ^{ab}	7.48 $\pm$ 0.14 ^a
Isoleucine	6.10 $\pm$ 0.11 ^{bc}	6.30 $\pm$ 0.12 ^c	5.83 $\pm$ 0.06 ^{ab}	5.54 $\pm$ 0.15 ^a
Phenylalanine	6.74 $\pm$ 0.11 ^a	6.92 $\pm$ 0.16 ^a	6.56 $\pm$ 0.22 ^a	6.47 $\pm$ 0.17 ^a
Lysine	6.95 $\pm$ 0.17 ^a	7.06 $\pm$ 0.08 ^a	6.87 $\pm$ 0.08 ^a	6.66 $\pm$ 0.09 ^a
Histidine	3.23 $\pm$ 0.11 ^{ab}	3.46 $\pm$ 0.12 ^b	3.19 $\pm$ 0.06 ^{ab}	3.02 $\pm$ 0.08 ^a
Threonine	2.20 $\pm$ 0.11 ^{bc}	2.44 $\pm$ 0.13 ^c	2.03 $\pm$ 0.09 ^{ab}	1.78 $\pm$ 0.08 ^a
Valine	5.96 $\pm$ 0.27 ^a	6.46 $\pm$ 0.07 ^b	5.77 $\pm$ 0.16 ^a	5.67 $\pm$ 0.16 ^a
Methionine	2.04 $\pm$ 0.04 ^{bc}	2.28 $\pm$ 0.08 ^c	1.89 $\pm$ 0.09 ^{ab}	1.73 $\pm$ 0.07 ^a
Tryptophan	1.17 $\pm$ 0.07 ^a	1.32 $\pm$ 0.09 ^a	1.18 $\pm$ 0.17 ^a	1.11 $\pm$ 0.10 ^a
Proline	4.71 $\pm$ 0.09 ^b	4.70 $\pm$ 0.21 ^b	4.67 $\pm$ 0.04 ^b	4.09 $\pm$ 0.09 ^a
Arginine	6.56 $\pm$ 0.13 ^{ab}	6.75 $\pm$ 0.18 ^b	6.38 $\pm$ 0.15 ^{ab}	6.20 $\pm$ 0.11 ^a
Tyrosine	1.89 $\pm$ 0.09 ^{ab}	2.30 $\pm$ 0.13 ^b	1.73 $\pm$ 0.17 ^a	1.69 $\pm$ 0.14 ^a
Cysteine	1.25 $\pm$ 0.11 ^a	1.42 $\pm$ 0.08 ^a	1.17 $\pm$ 0.11 ^a	1.18 $\pm$ 0.13 ^a
Alanine	4.31 $\pm$ 0.11 ^b	4.48 $\pm$ 0.15 ^b	3.98 $\pm$ 0.09 ^a	3.91 $\pm$ 0.07 ^a
Glutamic acid	13.64 $\pm$ 0.19 ^b	14.38 $\pm$ 0.16 ^c	13.47 $\pm$ 0.12 ^{ab}	13.07 $\pm$ 0.10 ^a
Glycine	5.01 $\pm$ 0.11 ^{ab}	5.27 $\pm$ 0.11 ^b	4.99 $\pm$ 0.06 ^{ab}	4.74 $\pm$ 0.10 ^a
Serine	3.92 $\pm$ 0.06 ^{ab}	4.23 $\pm$ 0.12 ^b	3.79 $\pm$ 0.08 ^a	3.67 $\pm$ 0.10 ^a
Aspartic acid	10.09 $\pm$ 0.13 ^a	10.47 $\pm$ 0.11 ^b	9.89 $\pm$ 0.10 ^a	9.92 $\pm$ 0.10 ^a

Mean $\pm$ SEM of three replications, LA - Lima beans preserved with sniper, LB - Lima beans preserved with pepper, LC - Lima beans preserved with wood ash, LD - Lima beans preserved with control.

Amino Acid Composition of Kidney Beans

Table 6 shows the effect of selected preservatives on amino acid composition of kidney beans.

Kidney beans contains high amounts of amino acids. The result shows high amounts of essential amino acids in kidney beans preserved with pepper when compared with others except in tryptophan and methionine where there is no significant difference across the groups. Lysine showed the highest concentration among the essential amino acid with a value of 8.17g for kidney beans preserved with pepper.

Glutamic acid constituted the highest concentration of all amino acids and closely followed by aspartic acid which is expected in legumes while Cystine showed the lowest concentration. Kidney beans preserved with wood ash showed the lowest amino acid concentration while kidney beans preserved with pepper showed the highest amino acid concentration for most the amino acids.

Red kidney beans provide 10.2g glutamic acid, 9.5g aspartic acid, 1.2g cysteine, 1.7g methionine, 3g histidine, 4.4g alanine, 5.2g glycine, 3.4g threonine, 3.3g proline, 3.7g isoleucine, 3.1g tyrosine, 4.6g phenylalanine, 4.1g valine, 3.1g serine, 6.9g arginine, 7glysine and 7.2g leucine per 100g (Audu and Aremu, 2011).

Table 4. Effect of selected preservatives on amino acid composition of Kidney beans (g/100g)

	KA	KB	KC	KD	KE
Leucine	4.56 ± 0.09 ^{ab}	5.25 ± 0.17 ^c	4.93 ± 0.10 ^{bc}	4.25 ± 0.12 ^a	4.41 ± 0.21 ^a
Lysine	7.65 ± 0.20 ^b	8.17 ± 0.17 ^b	7.40 ± 0.18 ^{ab}	6.72 ± 0.11 ^a	7.77 ± 0.45 ^b
Isoleucine	5.12 ± 0.12 ^b	5.56 ± 0.13 ^c	5.23 ± 0.09 ^b	4.63 ± 0.10 ^a	5.07 ± 0.02 ^b
Phenylalanine	5.21 ± 0.15 ^a	6.08 ± 0.13 ^c	5.64 ± 0.10 ^b	5.52 ± 0.10 ^{ab}	5.15 ± 0.02 ^a
Tryptophan	1.09 ± 0.13 ^a	1.24 ± 0.11 ^a	1.13 ± 0.16 ^a	0.91 ± 0.11 ^a	1.05 ± 0.01 ^a
Histidine	2.74 ± 0.11 ^a	3.61 ± 0.13 ^b	2.62 ± 0.15 ^a	2.27 ± 0.14 ^a	2.65 ± 0.15 ^a
Threonine	4.25 ± 0.12 ^{ab}	5.03 ± 0.15 ^c	4.62 ± 0.12 ^b	4.17 ± 0.11 ^a	4.51 ± 0.12 ^{ab}
Valine	5.82 ± 0.10 ^{bc}	6.19 ± 0.15 ^c	5.62 ± 0.15 ^b	4.97 ± 0.14 ^a	5.88 ± 0.06 ^{bc}
Methionine	1.11 ± 0.12 ^a	1.16 ± 0.14 ^a	1.00 ± 0.10 ^a	0.78 ± 0.12 ^a	1.06 ± 0.01 ^a
Proline	3.12 ± 0.14 ^{bc}	3.46 ± 0.20 ^c	2.75 ± 0.16 ^{ab}	2.46 ± 0.15 ^a	3.07 ± 0.01 ^{bc}
Arginine	6.18 ± 0.09 ^b	6.78 ± 0.14 ^c	6.07 ± 0.15 ^b	5.64 ± 0.16 ^a	6.15 ± 0.02 ^b
Tyrosine	4.80 ± 0.09 ^b	5.53 ± 0.13 ^c	4.32 ± 0.16 ^a	3.91 ± 0.15 ^a	4.77 ± 0.10 ^b
Cystine	0.82 ± 0.15 ^a	0.97 ± 0.14 ^a	0.77 ± 0.13 ^a	0.66 ± 0.15 ^a	0.77 ± 0.06 ^a
Alanine	4.42 ± 0.13 ^b	4.94 ± 0.17 ^c	4.42 ± 0.18 ^b	3.84 ± 0.12 ^a	4.41 ± 0.15 ^b
Glutamic acid	12.07 ± 0.13 ^a	13.07 ± 0.13 ^b	12.77 ± 0.16 ^b	11.56 ± 0.16 ^a	11.86 ± 0.17 ^a
Glycine	2.33 ± 0.16 ^a	2.91 ± 0.14 ^b	2.53 ± 0.11 ^{ab}	2.18 ± 0.13 ^a	2.38 ± 0.17 ^a
Serine	2.55 ± 0.17 ^{ab}	3.09 ± 0.13 ^b	2.83 ± 0.12 ^{ab}	2.33 ± 0.18 ^a	2.52 ± 0.18 ^a
Aspartic acid	11.62 ± 0.12 ^b	12.32 ± 0.17 ^c	11.78 ± 0.13 ^b	10.94 ± 0.11 ^a	11.45 ± 0.12 ^b

Mean ± SEM of three replications, KA - Kidney beans preserved with sniper KB - Kidney beans preserved with pepper, KC - Kidney beans preserved with aluminum phosphate, KD - Kidney beans preserved with wood ash, KE - Kidney beans preserved with control.

IV. DISCUSSION

The nutritive importance of edibles foods or food products are determined through proximate composition analysis. Any factor that affects the compositions will also influence the overall nutritive output of such foods. Improving nutritive value and reduction in spoilage rate of foods have resulted to use of both natural preservatives and other chemicals. [27]. The results of the proximate contents of the lima beans treated with selected preservatives as presented in Table 2. The result showed the proximate composition of lima beans. It was observed that the selected preservatives (sniper, Pepper, and ash) had varying effect on the proximate composition of the lima beans. The moisture contents of the lima beans of the treated groups were significantly ($p < 0.05$) lower when compared with the control group with the sniper preserved group having the lowest percentage moisture. The Low values of moisture is an indicative values for good storage quality. Moisture contents of foods determines its shelf life (how long it can last with spoilage). This is because under moist condition microbial and enzymatic activity is the most and this predisposes food to easy spoilage [28]. The decrease in moisture composition observed in the preserved groups compared to control is any indication that the selected preservative bind with the lima beans and dry it moisture contents. This also may suggest that all the preservatives can prevent the spoilage of the lima beans over time. Also, the result of the crude fat, crude fibre, protein and carbohydrate composition showed a non-significant ($p < 0.05$) reductions on it percentage composition respectively in some of the selected preservative treated groups. Carbohydrates provide the necessary calories in the diet of most people of the world [29]. High value of carbohydrate in date makes it a very useful fruit for consumers. However, pepper preserved showed significant ($p < 0.05$) improvement in crude fat compared to control and other treated groups. Same for carbohydrates where ash preserved group showed significant improvement compared to control.

For kidney beans, the effect of the selected preservative on proximate composition is presented in Table 1. The result showed the proximate composition of the kidney beans preserved with sniper to have the lowest moisture content while the wood ash preserved has the highest moisture content. This suggests that sniper possess drying effect on the kidney beans and will therefore regulate microbial activities and will consequently prevent spoilage (Rosa, 1974). It was observed also that the aluminum phosphide preserved had the highest % protein content compared with other preserved groups and the control. Also, it was observed that the pepper and aluminum phosphide preserved group had improved

crude fibre contents when compared with control. Same for carbohydrate, all the preservatives caused significant ($p < 0.05$) increase in the treatment groups compared with control.

The effect of selected preservatives on mineral composition of lima beans and kidney beans are presented in Table 3 and 4. The result showed varying effect on the mineral elements concentrations of treatment group compared with the control. For the lima beans, the selected preservatives caused a significant ($p < 0.05$) increase in Ca, in the pepper treated group compared with control but that is not noticeable for Fe, Zn, K and Mg, with any of the preservatives as there was no significant difference observed in any of them respectively in its concentrations. For the kidney beans as shown in table 3, it was observed that some of the preservative improved the mineral elements concentrations while some significantly decreased the concentrations. Calcium is the most abundant mineral in the body. It regulates many cellular processes and has other vital roles in living organisms [30]. The Dietary Reference Intake (DRI) value for calcium is 1000 mg/day (institute of medicine 2004). The result of this work showed that calcium was the improved with concentration of 12.92 ± 0.6 mg/ 100g and the control 10.58 ± 0.20 mg/ 100g for sniper. Zinc plays a vital role in cellular membrane structure and function, and helps to maintain adequate levels of vitamin A in the body [31]. It acts as a potent antioxidant and is essential for growth and development of body tissues, proper immune function and regulation of insulin [32].

Amino acids are important biomolecules that both serve as building blocks of proteins and are intermediates in various metabolic pathways. They serve as precursors for synthesis of a wide range of biologically important substances including nucleotides, peptide hormones, and neurotransmitters. Moreover, amino acids play important roles in cell signaling and act as regulators of gene expression and protein phosphorylation cascade [33], nutrient transport and metabolism in animal cells [33 and innate and cell-mediated immune responses.

The effect of selected preservative amino acid composition of lima beans is presented in Table 5 for its essential and nonessential amino acid concentrations. The result showed that Glutamic acid was the most predominant amino acid with concentration of 14.64 ± 0.16 g/100g for sniper preserved group. Tryptophan, Threonine and methionine were in the lowest levels in lima beans. A total of nineteen amino acids was identified in the soya beans, Nonessential amino acid; Leucine, Lysine, Isoleucine, Phenylalanine, Tryptophan, Valine, Methionine, Histidine, Threonine and the essential amino acid; Proline, Arginine, Tyrosine, Cysteine, Alanine, Glutamic acid, Glycine, Threonine, Serine, and Aspartic acid. Soya bean is a good natural source of this compounds. On the quest not only to preserve lima beans to extend its shelf life but as well to avoid spoilage, several chemical and preservative having been selectively used to store lima beans after harvest [11] From the result in table 5 and 6 respectively it was observed that the selected preservative had varying effect on the amino acid composition of the treatment groups when compared with control that was well preserved with no addition of preservative, it was observed that some of the preservatives had improvement effect on the amino acid composition while some reduced the concentration when compared with the control resulting to imbalance in amino acids compositions of the soya beans when compared to FAO [35] recommended levels. These varying concentration may be associated with nature of the preservative which may have bind with amino acid side chains to deactivate or activate it in the lima beans making it abundant or reduced respectively [35].

on the other hands, the effect of some selected preservatives on Kidney beans amino acid composition is presented in table 6 for its essential and nonessential composition. The results of the table showed that the selective preservatives had caused significant ($p < 0.05$) reduction in the amino acid compositions of both the essential and non-essential amino acid respectively. However, among the identified amino acid glutamic acid composition of the kidney beans was the highest followed by aspartic acid while cysteine and methionine are the lowest. It was observed clearly that the each of selected preservatives had its separate effect on each group compare with control. It was seen from the study that some preservative both helps in both preventing spoilage and enhance nutritive indices while some compromises the nutritive value as it prevents spoilage.

V. CONCLUSION

Preservatives are important as it helps to sustain the quality of food for longer time. It has been reported that chemicals which are used as preservatives have some possible influence to the nutritive indices of foods. Even though natural preservatives have been used over the years, artificially produced chemical like sniper and aluminum phosphide were comparatively studied and compared some of the natural preservatives pepper and wood ash. It was observed that the chemical more effective at some aspect while natural preservatives at some other points.

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