ADDIS ABABA UNIVERSITY

FACULTY OF NATURAL SCIENCE

FOOD SCIENCE AND NUTRITION PROGRAM

Effect of Cooking Methods on Nutritional Composition and Anti-Nutritional Factors of Hyacinth Bean (*Lablab Purpureus L.*) Sweet Varieties in Debati woreda, Ethiopia

BY

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A Thesis Submitted to the School of Graduate Studies of Addis Ababa University in Partial Fulfillment of the Requirements for the Degree of Master of Science in Food Science and Nutrition

June, 2012

Addis Ababa, Ethiopia

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SCHOOL OF GRADUATE STUDIES

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Acknowledgements

First of all I would like to thank the almighty God for giving me health, endurance and strength to become successful in my thesis work. He always stands in front of me in all things that I do.

I would like to express profound gratitude and sincere appreciation to my advisor Dr. Melese Abdissa for his endless cooperation, guidance, academic comments, constructive suggestion and his encouragement starting from the development of the proposal up to the thesis work. Really his keen interest, kindness and an expertise input made this work easier and feasible. It is intellectually stimulating and rewarding experience to work with him. Moreover, my endless gratitude goes to my advisor professor Negussie Retta for his expertise comments, supervision and useful suggestions throughout this research work.

My special thanks go to Dr. Gulelat Desse, Mr Ashagrie Zewdu, Mr Endale Amare, Mr Tilahun Bekele, Mr Adamu Belay, Mr Dilnesawu Zerfu, Mr Muluneh Kifle, Mr Debebe Hailu, Mr Tadele Tuba, Mrs Woinshet Abera and Mrs Hirut Assaye for their facilitation and help during various tasks.

I am also grateful to EHNRI for their good will to use their laboratory space and facilities. Unequivocally, I thank all Food Science and Nutrition laboratory, AAU and EHNRI laboratory staff for their cooperation and technical support during the laboratory analysis.

Finally, I am extremely grateful to my best half (Meseret Abebe), my son Giyon Anbissa, my brothers (Amsalu Muleta, Dessalgn Muleta, Defersha Ejeta, Getachew Abebe) for their financial, moral and psychological encouragement throughout the course and on this thesis work.

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LIST OF ABREVIATIONS

AA	Arachidonic acid
AAS	Atomic Absorption Spectrophotometer
ANFs	Antinutritional Factors
ANOVA	Analysis of variance
AOAC	Association of Official Analytical Chemists
Ca	Calcium
CHO	Carbohydrate
Cl	Chlorine
$CuSO_4$	Copper Sulphate
DHA	Docosahexanoic Acid
DNA	Deoxy ribonucleic Acid
EHNRI	Ethiopian Health & Nutrition Research Institute
EPA	Eicosapentaenoic Acid
F	Fluorine
FAO	Food and Agricultural Organization of United Nations
Fe	Iron
g	gram
GLM	General Linear Model
h	hour
HCl	Hydrochloric Acid

HDL	High Density Lipoprotein
IDA	Iron Deficiency Anemia
K	Potassium
K_2SO_4	Potassium Sulphate
Km	Kilometer
LA	Linolic Acid
$LaCl_3$	Lanthanum Chloride
LDL	Low Density Lipoprotein
mg	miligram
Mg	Magnesium
MI	Micronutrient Initiative
mL	milliliter
mm	millimeter
mmol	milimol
Mn	Manganese
Na	Sodium
NaOH	Sodium Hydroxide
NAS	National Academy of Science
NDC	Non-Digested Carbohydrate
NDO	Non- Digestible Oligosaccharides
nm	nanometer

NRC	National Research Council
NS	Non significant
Phy	phytate
PO_4^3	Phosphate ion
PPM	Parts per million
PUFA	Poly Unsaturated Fatty Acids
RDA	Recommended Daily Allowance
RS	Resistant Starch
S	Sulfur
SCFA	Short Chain Fatty Acids
SCN	Standing Committee on Nutrition
Se	Selenium
SE	Standard error
SPSS	Statistical Package for Social Sciences
UNDP	United Nations Development programme
UNICEF	United Nations Children's Fund
W/V	Weight by Volume
WHO	World Health Organization
Zn	Zinc
µL	micro liter

Abstract

Hyacinth bean is very important grain legume, of the family Leguminosae , have been utilized as crop and staple food source. Characteristically, legumes, especially beans are considered an important and inexpensive protein and dietary fiber sources in human nutrition. And this study was conducted with an objective to assess the effect of cooking methods on the nutritional composition and anti-nutritional factors of Hyacinth Bean (Lablab purpureus L.) sweet varieties. The raw and processed hyacinth bean (Lablab purpureus L.) sweet varieties *were studied and compared for their nutritional composition: moisture crude protein, total ash, crude fiber, crude fat, carbohydrate; minerals: (Ca, Fe, Zn, and P) and ANFs: (phytate and tannin) based on different processing techniques. Sensory acceptability test of cooked hyacinth bean varieties were also reported. Boiling, autoclaving and soaking treatments significantly ($p<0.05$) decreases the moisture, crude fat and ash contents respectively whereas they significantly ($p<0.05$) increases the fiber and carbohydrate contents. Cooking and soaking treatments were non significant ($P<0.05$) on crude protein content on both hyacinth bean varieties. Boiling and autoclaving treatments increases the Ca, Fe, Zn and P contents of Highworth variety while they significantly($p<0.05$) increases Ca, Fe and P and decreases Zn contents of Rongai variety. Boiling and autoclaving treatments significantly ($p<0.05$) very effective to reduce the phytate and tannin content of both Rongai and Highworth varieties. The phytate: Zn , phytate: Fe and Ca:phytate molar ratios and phytate*Ca:Zn millimolar ratios were determined in order to estimate the bioavailability of these minerals. The molar ratio of phy:Fe and Ca:phy was >1 and >6 respectively indicating that Fe and Ca absorption could be impaired. This study also revealed that, the sensory acceptability test on the major sensory attributes of boiled and autoclaved hyacinth bean varieties were reported. And there was no a significant difference observed on the sensory attributes of (color, aroma, taste, texture and over all acceptability) of the hyacinth bean products.*

Key words: *Hyacinth bean variety, soaking, boiling, autoclaving, nutritional composition, minerals, ANFs, phtatate:mineral molar ratio, sensory acceptability test*

1. Introduction

1.1. General Description

Hyacinth bean (*Lablab purpureus L.) sweet* is a very important grain legume, of the family Leguminosae , have been utilized as crop and staple food source. It is an Asian origin crop which, was probably domesticated in India as a cultivated grain legume. The crop has been documented in India prior to 1500 BC (Clapham and Rowley-Conwy, 2007). It is a legume crop widely grown throughout Africa, Asia and Latin America for its vegetable or pulse for human consumption. The crop is widespread in Egypt, Sudan, Madagascar and Ethiopia (Khalid *et al.*, 2008) Hyacinth bean is cultivated either as a sole crop or in mixed production systems. Its popularity can be demonstrated by its more than 150 local names reported by various authors and on data bases (Brigitte *et al.*, 2010).

In Ethiopia, the common name of hyacinth bean is Gerenga (Brigitte *et al.*, 2010) or locally it is called *"Aepo"* and *"Opa"* in both Shinashegna and Gumzegna respectively. It grows in a wide range of soils from deep sands to heavy clays where the pH varies from 4.5-7.5 (NAS/NRC, 1979). Its growth optimum requires annual rainfall range of 650-3,000 mm and an average temperature of 18-30°C. Hyacinth bean can also grow where annual rainfall is <500 mm and tolerates high temperature in a harsh conditions; with loses of leaves during prolonged dry periods (Morris, 2009).

There are two varieties of hyacinth beans which are called Rongai and Highworth often consumed in Ethiopia. Rongai is vigorous and leafy, with white flowers and two to four seeds per pod. Seeds are buff to pale brown (light brown) in colour, have a linear, white hilum whereas Highworth is similar in appearance to Rongai in vegetative growth, but has purple flowers, smaller seeds, black when mature, or with some dark brown when ripened rapidly (Cameron, 1988).

In Ethiopia, it is cultivated for human consumption in areas like Konso (southern Ethiopia), Gamo-gofa and around Gondar and Gojam in central, Metekel (North-West) and Northern areas (Institute of Biodiversity Conservation, 2007). The nutritive value of hyacinth bean primarily

depends on contents and availability of the nutrients and is subject to the presence or absence of anti-nutritional and /or toxic factors.

Nevertheless, legume processing methods such as soaking, germination, decortication, fermentation and cooking greatly influence their nutritive value. Of these, cooking and germination plays an important role as it influences the bioavailability and utilization of nutrients and also improves palatability which incidentally may result in enhancing the digestibility and nutritive value (Bark, 1996; Ramakrishna *et al.*, 2006). Given the general remark as to how to increase the nutrient availability, there is specific requirement to each species of the legumes. Information available on the nutritive value (content and availability) of hyacinth bean is limited (Ramakrishna *et al.*, 2006).

Characteristically, legumes, especially beans are considered an important and inexpensive protein and dietary fiber sources in human nutrition. Further, beans contain a considerable amount of vitamins and minerals (Osman, 2007). Therefore, knowledge on the nutrient content of lablab would have immense contribution on its nutrition.

In Ethiopia, hyacinth bean is usually consumed at the immature green and mature dry seeds after cooking as a popular staple food in some areas. The dry seeds can also be consumed as whole or decorticated after cooking and processing in different ways. In addition to these uses, the splits of decorticated hyacinth are used to make sauce (stew) in Ethiopia particularly in nationalities like Gumz, Shinasha and Konso. It is considered as very delicious staple food by Shinasha and Gumz nationalities

Utilization of legumes as food is often limited due to presence of several factors. The development of the hard to cook defect reduce the palatability and cooking quality of legumes. Seed size, swelling capacity, seed coat and texture of certain hyacinth bean are also associated with cooking quality.

However, hyacinth beans may contain compounds that can negatively affect their nutritional value, such as trypsin inhibitors, lectins, phytates, polyphenols and tannins. Some of these anti-nutritional factors can affect the bioavailability of divalent mineral ions. Other compounds such as lectin and tannin can cause human health problems if consumed without appropriate

processing techniques (Alajaji and El-Adawy, 2006). Lack of knowledge of optimum processing techniques might have resulted to underutilization of hyacinth beans for human consumption.

Enzyme inhibitors and phytic acid which reduces the bioavailability of divalent minerals, toxic factors such as tannins and haemaglutinins (Huma *et al.*, 2008) can be possible reasons for underutilization of hyacinth bean. Generally, legumes have been reported to have low nutritive value because of low amounts of sulfur-containing amino acids, low protein digestibility and the presence of anti-nutritional factors (ANF).

Legumes are usually cooked before being used in the human diet. This improves the protein quality by destruction or inactivation of the heat labile anti-nutritional factors (Chau *et al.*, 1997). However, cooking causes considerable losses in soluble solids, especially vitamins and minerals (Barampama and Simard, 1995). Finally, the main purpose of this paper is to assess the nutritional composition and anti-nutritional factors (ANF) of hyacinth bean (*lablab purpureus L.*) sweet as affected by cooking methods.

1.2 Statement of the problem

Hyacinth bean is" lost crop in Africa" (Brigitte *et al.*, 2010) regardless of its potential to maintain food security, protein energy malnutrition and public health benefits. Hyacinth bean plants, seeds, and pods have many uses including as vegetables and forage, as well as potential nutraceutical and pharmaceutical uses (Morris, 2003).

Most researches on hyacinth bean have been related to distribution and its agronomy (Brigitte *et al.*, 2010). A study of the composition and nutritive quality of hyacinth beans would therefore be of great interest, because the knowledge provided would help to orient the work of investigators involved in varietal selection and also reduce or eliminate anti-nutritional factors to make edible and nonedible hyacinth bean seeds more acceptable as an inexpensive source of protein. The data on nutritional /anti-nutritional content of hyacinth bean may help to select the specific variety or type of legume to grow on large scale in developing and developed countries. These kinds of studies would also help to increase the availability of food by processing underutilized varieties into edible forms through research and development.

Despite the fact that, legumes constitute an important source of nutrients, they show the presence of many components that can interfere with the bioavailability of minerals, proteins and starch and regulate metabolic functioning. Nevertheless, when processed and consumed in a variety of forms, legumes change in their nutritional potential (Osman, 2007).

Specifically, in the case of hyacinth bean optimum effects of processing factors were not published. No comparative studies were conducted on the nutrient content, availability and anti-nutritional factors masking expression of the nutrients in Rongai and Highworth varieties of hyacinth bean. Therefore, it is imperative to research and figure out optimum nutrient and anti-nutrient conditions of these varieties of hyacinth bean that were consumed by the society.

1.3. Significance of the study

- This study contributes to the vegetarian diet to select an appropriate food item.
- It also provides an empirical data about the nutritional composition and anti-nutritional factors in different cooking methods.
- It provides a base line for further analysis of other chemical species in hyacinth bean.
- It contributes helpful information to identify the gap and undertake further study on hyacinth bean. Moreover, this study was help stake holders, nutritionists, agricultural research institutes and food scientists to give an emphasis in the future.

Therefore, it is hypothesized that no effect of variety and cooking method on contents of the bean varieties.

1.4. Objectives

1.4.1. General objective of the study

To assess the effect of cooking methods on the nutritional composition and anti-nutritional factors of Hyacinth bean (*Lablab purpureus* L.) sweet varieties.

1.4.2. Specific objective of the study

- To study comparative values of proximate and anti-nutritional composition of hyacinth bean.
- To determine the effect of processing on nutritional and anti-nutritional factors in hyacinth bean.
- To estimate the major mineral composition (Ca, Fe, Zn and P) of hyacinth bean varieties.
- To evaluate the sensory acceptability of cooked hyacinth bean.

2. Literature review

2.1. Nomenclature of Hyacinth bean (*Lablab purpureus L.) sweet*

Hyacinth bean *(Lablab purpureus* L.) previously classified as *Dolichos lablab*, is known in different parts of the world by different names (Table 2.1). In fact, there is much disagreement in the literature as to names and varieties (Kay, 1979). This multiplicity of names is indicative of the range of forms available globally and the fact that it has long been cultivated for human food and as green manure. The widespread use of lablab for animal grazing is more recent (Cameron, 1988).

Table 2.1. Some common names used for *lablab purpureus*

Dolichos lablab	Lablab	Garbanzo	Lablab niger	Sim bean
Country bean	Frijol dolicho	Gerenga	Poor-man's bean	Field bean
Dolichos bean	Lubia bean	Egyptian bean	Chimbolo verde	Gallinita
Lablab valguris	Frijol jancito	Siem bean	Frijol de la tierra	Pig-ears
Hierba de conejo	Caballero	Proto japones	Bonavist bean	Batao,
Rongai dolichos	Fiwi bean	Indian butter bean	Tonga bean	Wal
Banner bean	Faselbohne	Caroata chwata	Quiquaqua	Helmbohne
Pois antaque				

Source: Murphy and Colucci(1999)

2.2. History and distribution

The wild forms of hyacinth bean are believed to have originated in India (Deka and Sarkar, 1990) and were introduced into Africa from Southeast Asia during the eighth century (Kay, 1979). Presently, hyacinth bean is common in Africa, extending from Cameroon to Swaziland and Zimbabwe, through Sudan, Ethiopia, Uganda, Kenya and Tanzania (Skerman *et al.*, 1991). As early as 1819, seeds of hyacinth bean from Egypt were planted in the Botanical Gardens in Sydney, New South Wales. However, it was not until after the release of the forage cultivar "Rongai" in 1962, that hyacinth bean became widely used as forage in Australia.

Despite its large agro-morphological diversity in South Asia, its origin, however, appears to be African (which is the only continent where wild plants of the species have been recorded to

occur naturally (Brigitte *et al.*, 2010). Fuller (2003) suggested its introduction to South Asia occurred from west to east so that archaeo-botanical finds have been dated, for example, from 2000 to 1700 BC at Hallur, India's earliest Iron Age site in the state of Karnataka, to 1200–300 BC at the Veerapuram excavation site in the state of Andhra Pradesh.

Hyacinth bean has been widely distributed to many tropical and subtropical countries where it has become naturalized (Purseglove, 1968). In South and Central America, East and West Indies, Asia, China and India. Hyacinth bean is grown as an annual or a short-lived perennial (Whyte *et al.*, 1953). In these areas, the seed and immature pods are used for human food while the herbage is used as green manure, for erosion control, and as a feed supplement for cattle grazing mature pasture in the dry season.

2.3. Plant description

Hyacinth bean is a summer growing annual or short-lived perennial fodder sown for grazing and conservation in tropical environments with a summer rainfall. It is a vigorously trailing, twining herbaceous plant, resistant to disease and insect attack (Cameron, 1988).

There are two crop types of hyacinth beans including the vine garden types and the erect, bushy field types (NAS/NRC, 1979). The plant produces many branches and is often grown as an annual crop reaching 6 m tall with well-developed roots. The leaves occur alternately and produce three leaflets (5--15 cm × 4–15 cm) (Shivashankar and Kulkarni, 1989). Leaflet vein colors range from green to purple, while flower colors range from white, pink, violet to purple (NAS/NRC, 1979) in (Figure 2.1). The seedpods vary in shape, color, and form. Some are flat or inflated, straight or curved (5–20 cm × 1–5 cm), and usually contain 3–6 seeds of variable colors and sizes (Shivashankar and Kulkarni, 1989) as indicated Figure 2.3. The seed coat colors range from cream to black (Pengelly and Maass, 2001) as shown in Figure 2.2.

Hyacinth bean grows well in several diverse areas and conditions including arid, semiarid, and humid regions (200–2,500 mm of annual precipitation). The plant is adapted to areas where temperatures range from 22 to 35°C, lowlands and highlands, and tolerates diverse soil types

(ranging from acid to alkaline) (NAS/NRC, 1979). The hyacinth bean is drought tolerant due to its deep rooting system; however, adequate moisture is needed early for establishment. Of the two hundred types of hyacinth bean recognized, only two varieties, Rongai and Highworth, are available commercially (Cameron, 1988).

(a) *(b)*

Fig 2.1. Two types of hyacinth bean.(a) is field type and (b) home garden type of hyacinth bean plants(Source: Factsheet - *Lablab purpureus*, 2007)

Fig 2.2. Different seed coat colors of hyacinth bean (Source: Factsheet - *Lablab purpureus*, 2007).

2.3.1. Rongai cultivar

The Rongai cultivar was derived from material from the Rongai district of Kenya (Cameron 1988) and was released in New South Wales, Australia in 1962 (Wilson and Murtagh, 1962). As described above, this is a summer growing, rampant and vigorously twining herbaceous annual or short-lived perennial. Stems trail, reaching 3 to 6 m in length; broad ovate-rhomboid leaflets acute at the apex, range between 7 and 15 cm and are arranged in a trifoliate manner. Leaves are almost glabrous on the upper surface and have short hairs on the lower surface. Petioles are long and slender and inflorescence lax, fascicled, of many flowered racemes on elongated peduncles. Pods are 4-5 cm in length containing 2-4 buff or pale brown seeds with a conspicuous white hilum. The brown, ovoid and laterally compressed seeds number 3600-4300 per kg (Barnard, 1972). Rongai is a late maturing white flowering cultivar that will continue to grow until cut or damaged by frosts. In the absence of frost, flowering may continue for several months (Cameron, 1988).

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Fig 2.3. Different types of seedpods of hyacinth bean in form, color and shape (Source: Factsheet - *Lablab purpureus*, 2007)

2.3.2. Highworth cultivar

The Highworth cultivar originated from Coimbatore, South India and is morphologically similar to Rongai. Contrasting with the green foliage, white flowers and light brown seeds of Rongai, foliage of Highworth has a purple band near the leaf axel, purple flowers and black seeds. Highworth is an early flowering line with high seed-yielding ability; it is suitable for pulse production and forage uses. It was originally intended for grain production in districts where early frosts prevented the seeding of Rongai (Cameron, 1988). It should be noted that the Rongai cultivar is most prevalent in the tropical forage legume literature.

2.4. Agronomic characteristics

2.4.1. Environmental conditions

Hyacinth bean is a legume well suited to most tropical environments as it is adaptable to a wide range of rain fall, temperature and altitude. It is reported to grow well under warm, humid conditions at temperatures ranging from 18 to 30°C and is fairly tolerant to high temperatures (Hendricksen and Minson, 1985b; Kay, 1979; Cameron, 1988). Below 20° C the plant will have reduced growth; leaves begin to drop at minus 2°C but the plant can survive frost for a limited period (Kay, 1979; Mayer *et al.*, 1986). Hyacinth bean is drought hardy, and has been grown in arid, semi-arid and humid regions with rainfalls between 200 and 2500 mm (Hendricksen and Minson, 1985b; Cameron, 1988). It needs rainfall or irrigation (minimum of 10 to 20 mm) during germination and early establishment, although once established it is extremely resistant to drought (Mayer *et al.*, 1986). Being a hardy plant, hyacinth bean can be found throughout the tropics and subtropics; ranging from 30° South to 30°North Latitude. It is normally grown from sea level up to elevations of between 1800 and 2100 metres (Cameron 1988; Mayer *et al.*, 1986).

2.4.1.1. Soil

Hyacinth bean grows in a wide range of soil types, from deep sands to heavy black clays and can tolerate pH ranges of 5 to 7.5 (Kay, 1979). The plant can survive short periods of flooding thus growing well in alluvial planes (Menéndez *et al.*, 1985) but needs free drainage as it does not tolerate water logging (Kay, 1979). Saline conditions have been found to reduce populations and produce chlorotic leaves. Soil fertility is important; thus phosphate fertilizers may need to be applied at planting (Cameron, 1988).

2.5. Chemical composition

2.5.1. Protein

Protein is the most abundant nitrogen-containing compound in the diet and in the body. It is one of complex biomolecule present in cells and tissues (Gibney *et al.*, 2009). The need for protein during the period of skeletal and muscle growth of human being is high. An intake of 2.1g of high biological-value protein per kg of body weight permits nitrogen retention of about 45%, as

long as energy intake is adequate (Guthrie, 1989). If the protein is of low biological value the amount needed increases proportionately. Protein with high biological value of at least 70 to 85%, e.g. eggs, milk, meat, with almost half of amino acids being essential amino acids, have been recommended to be used for infants and children (Picciano, 1987).

Virtually all the food we consume, both plant and animal contains some protein. Some foods are considered as 'protein foods', which means that they contain a relatively high concentration of protein. The main examples are meat, fish, milk, cheese, eggs and legumes, especially soya beans. However, the importance of any food as a source of any nutrient also depends on the amount of that food that is eaten (Sanders and Emery, 2003).

Legumes are good sources of cheap and widely available proteins for human consumption. They are staple foods for many people in different parts of the world. Legume seeds have an average of twice as much protein as cereals and the nutritive value of the proteins are usually high (Doss *et al.*, 2011). They range between the highly utilized legumes such as soybeans, cowpeas to the lesser known ones like African yam beans (*Sphenostylis stenoscarpa*), *Mucuna conchinchinesis* and *Mucuna flagellipes* ("ukpo"). Studies have shown that the lesser known legumes together with other conventional legumes can be used for combating protein malnutrition prevalent in the third world. Among these legumes, hyacinth bean seeds and pods could be used as famine food worldwide where humans suffer from malnourishment and disease (Morris, 2009).

Legumes/ pulses are considered to be a very important group of plant foodstuff, particularly in developing world, as a cheap source of protein for vegetarians as well as economically poor people which could not afford to purchase animal source proteins. Being a cheap source of dietary protein, legumes are now successfully used in child feeding programmes and food and feed formulation (Kamatchi *et al.*, 2010). These include peas, beans and lentils as a good source of protein and are excellent substitutes for meat. Protein contents of legumes vary between 17 and 34 per cent. which include metabolic, structural and storage protein. Storage protein is made up to 80 per cent of the total protein (Huma *et al.*, 2008). According to Kamatchi *et al* (2010) the crude protein content of the five varieties of hyacinth bean ranged from 20.46-25.47% and also

dried hyacinth bean seeds contain 20–28% protein, 6.1% lysine content, and compliment cereal diets (Morris, 2009).

Legume seeds are rich in lysine and poorer in sulfur-containing amino acids (methionine and cystein) compared to cereals. As the study conducted by Kamatchi *et al.*(2010) the content of the sulfur containing amino acids and tryptophan seem to be deficient in all the varieties of *lablab purpureus;* whereas threonine, valine, isoleucine, leucine, phenylalanine, lysine, and histidine in all the investigated varieties were found to be higher compared to requirement pattern. Thus, hyacinth bean can provide the essential amino acids required for building block for our bodies including bones and muscles and enzymes, hormones and vitamins.

2.5.2. Carbohydrates

Carbohydrates are the single most abundant and economic sources of food energy in the human diet, constituting 40–80% of total energy intake in different populations. It is major classes of biomolecules and play several important roles in all life forms, including: sources of metabolic fuels and energy stores, structural components of cell walls in plants and of the exoskeleton of arthropods and integral features of many proteins and lipids (glycoproteins and glycolipids), especially in cell membranes where they are essential for cell–cell recognition and molecular targeting (Gibney *et al.*, 2009).

Carbohydrates constitute the main fraction of grain legumes, accounting up to 55-65% of the dry matter. Of these, starch and non- starch polysaccharides (dietary fiber) are the major constituents, with smaller but significant amounts of mono, di and oligosaccharides (*Bravo et al., 1998*). These legumes contain slow digested carbohydrates and high proportion of non-digested carbohydrates (NDC) that might be fermented in the large intestine. Non-digested carbohydrates reaching the colon include mainly resistant starch (RS), soluble and insoluble dietary fiber, and nondigestible oligosaccharides (NDO) (Henningson *et al.*, 2001).

The NDC are associated with a low glycemic response, low serum cholesterol levels, and a decrease of colon cancer risk factors (Serrano and Goni, 2004). The physiological effects of NDC from common beans may be related to colonic fermentation end products, short chain fatty acids (SCFA), such as acetic, propionic and butyric acids, and the content and distribution of

SCFA are dependent on the microflora and the carbohydrate substrate at the intestinal tract (Cummings and Englyst, 1987; Cumming and Macfarlane, 1997).

Beans contain some complex sugars of the raffinose family. These are the sugars that cause digestive issues with bean consumption. These sugars must be broken down by enzymes that are not available in the human digestive system and are therefore available for microbial action in the colon, resulting in gas production and flatulence. These sugars can be removed effectively from the beans by soaking the beans, and then cooking them, discarding the soaking and cooking liquids (Raatz, 2012).

2.5.3. Dietary fiber

No universally accepted definition of dietary fibre exists. A useful and generally accepted definition is that dietary fibers are plant substances not digested by human digestive enzymes, including plant cell wall substances (cellulose, hernicelluloses, pectin, and lignin) as well as intracellular polysaccharides such as gums and mucilages (Spiller, 2001). In some definitions of dietary fiber, resistant starch components like oligosaccharides and inulin and non-carbohydrate components like lignin, waxes, and chitins are also included (Aggett *et al.*, 2003). The early definition by Trowell of "the remnants of plant cells resistant to hydrolysis by the alimentary enzymes of man" remains as a key definition or it is chemically defined as the sum of the plant nonstarch polysaccharides and lignin (Spiller, 2001). Dietary fibres are also called "non-digestible carbohydrates", especially in relation to the physiological effects of these substances in infants and young children (Aggett *et al.*, 2003).

The major portion of dietary fiber in foods is derived from the plant cell walls in foods. A wide range of plant organs and types of tissue is consumed in the human diet, although highly lignified (woody) tissues are rejected during food preparation (Spiller, 2001). Despite the fact that, dietary fiber exerts a wide range of physiological effects when consumed and its complex nature is responsible for a range of physical and chemical properties that are responsible for these physiological effects. It has important therapeutic implications for certain conditions such as diabetes and hyperlipdemia and may have preventive implications for others such as hypertension, coronary heart disease and intestinal disorders (Anderson and Bridges, 1988). Because the fiber content of human foods from the plant source ranges from trace amount to

almost 50% of dry weight and because different types of fiber have different effects (Anderson and Bridges, 1988).

Beans provide an adequate amount of dietary fiber in human nutrition. Beans are rich in both soluble and insoluble fibers, so they provide the nutritional benefits of both fiber classes. The soluble fiber in beans dissolves in water, trapping bile which helps to lower blood levels of low density lipoprotein (LDL) cholesterol, especially if LDL cholesterol levels were high to begin with, without compromising the level of protective high density lipoprotein (HDL) cholesterol (Raatz, 2012). They also provide substantial amounts of insoluble fiber, which help attract water to the stool and enhance transit time of waste through the colon. This may help to combat constipation, colon cancer, and other conditions that afflict the digestive tract

The most fiber-rich plant foods are unrefined cereals and legumes, including soy beans, beans, lentils and peas. All plant foods contain both insoluble and water-soluble dietary fibers, although in varying quantities (Michaelsen *et al.*, 2008). Insoluble fibers, e.g. celluloses, some hemicelluloses and lignin, are indigestible or only partially fermented in the large intestine. An insoluble fiber in the diet causes soft stools and shortens intestinal transit time, which may reduce the digestibility and availability of nutrients (Michaelsen *et al.*, 2008). Soluble fiber, e.g. pectins, gums and mucilages are found in all plant foods, especially fruit and vegetables, but in varying amounts. A soluble fiber possesses water binding properties and is relatively rapidly fermented in the colon. Some soluble dietary fibers such as inulin can improve absorption of calcium (Coudray *et al.*, 2003; Abrams *et al.*, 2005; Wong *et al.*, 2006).

Diets with a high content of soluble dietary fibers may lead to flatulence due to the relatively rapid fermentation in the large intestine (Meance *et al.*, 1999). Especially a group of oligosaccharides, α- galactosides, typically found in legumes, are digested in the colon by bacteria resulting in the production of short-chain fatty acids and gases causing flatulence. High intake of soluble dietary fibers has been shown to lead to negative effects on energy intake in the short term (Rigaud *et al.*, *1998*) as well as in longer term studies in healthy subjects, and in malnourished children (Doherty and Jackson, 1992).

There are several studies and reviews dealing with the potential negative effect of dietary fibers on energy intake and growth in infants and children. Dietary fibers may reduce energy intake through a suppressing effect on appetite, and they may increase faecal losses of energy due to reduced absorption of fat and carbohydrate (Aggett *et al.*, 2003). In a study from the Netherlands on infants and young children receiving a "macrobiotic" diet with a high content of dietary fiber (13 g/d) the weight gain and linear growth was reduced considerably compared to a control group (Dagnelie *et al.*, 1989). The diet of these children was high in dietary fiber, low in fat content, contained no animal source foods and had an overall low energy density so the reason for this lower rate of weight gain in the children receiving the macrobiotic diet can not only be attributed to the high content of dietary fiber.

2.5.4. Dietary lipids

Nutrition contributes significantly to the well-being of people by innovative discovery of new food sources that can supply the essential nutrients in the diet. One of such nutrients is dietary lipids. Dietary lipids supply essential fatty acids that are necessary for the maintenance of cellular membrane integrity and the various biochemical functions associated with it (Odutuga *et al.*, 2008). Lipids are agents that, in situ or via the circulation, exert a broad spectrum of physiological and pharmacological effects on many target tissues (Odutuga *et al.*, 1997).

The essential fatty acids are usually limited in a diet and must be taken in from sources which are especially rich in them. The diets in most low-income countries consist mainly of basic staple foods - cereals, legumes and roots. Generally, the content of polyunsaturated fatty acids (PUFA) in these staple foods is low (except for peanut and soy). The cereal staples and peanut have a relative high content of n-6 PUFA and only very small amounts of n-3 PUFA (Michaelsen *et al.*, 2008). Fat content of most pulses is low (1 to 2%). Odutuga *et al.* (2008) reported that crude fat content of cooked beans as 10% of soyabean, 3.5% of cowpea and 0.5% of hyacinth bean respectively. Malnutrition is usually prevalent where too few different plant foods are available, especially in poor households which results from irregular and unbalanced diet (Olaofe *et al.*, 1998).

Dietary fat plays an important role in allowing adequate absorption of fat-soluble vitamins and an adequate supply of essential fatty acids. The differences between fat sources with respect to

absorption of the fat soluble vitamins, vitamin A, D and E, appear to be small (Michaelsen *et al.*, 2008). About 5 g of fat has been found to be needed per meal to provide good bioavailability of vitamin A. The absorption seems to be improved somewhat by fat rich in oleic acid (C18:1), but other oils are probably almost as good (Borel, 2003). Therefore, we assume the essential fatty acid issue to be the most relevant with respect to moderately malnourished children (Michaelsen *et al.*, 2008).

There are two types of essential fatty acids, the n-6 and the n-3 polyunsaturated fatty acids (PUFA), which in most diets are provided by vegetable oils in the form of linoleic acid (LA, C18:2n-6) and α-linolenic acid (ALA, C18:3n-3) respectively. Essential fatty acids may also be supplied from meat and fish in their long-chained forms, arachidonic acid (AA, C20:4n-6), eicosapentaenoic acid (EPA, C20:5n-3) and docosahexaenoic acid (DHA, C22:6n-3) (Michaelsen *et al.*, 2008).

2.6. Mineral elements

Mineral elements are generally classified as either microelements or macroelements. The macro elements are those present in relatively higher amounts (750ppm) in animal tissues. They include Ca, K, Na, Mg, S., $PO4^{3-}$, Cl. The microelements, also called trace elements, are present at less than 50ppm. Trace elements essential for human nutrition include among others Fe, Zn, Mn, Se, F, I_2. Mineral elements unlike proteins and vitamins, cannot be synthesized in the body, and so must be obtained through dietary means. Hence the amount of a mineral element in an animal tissue reflects the amount present in the food consumed by the animal, which is in turn a function of the element present in the soil, and the extent to which the plant concentrates it during growth (Mertz, 1980).

Minerals are involved in activation of intracellular and extracellular enzymes, in regulation of critical pH levels in body fluids necessary for the control of metabolic reactions and in osmotic balance between the cell and its environment. A deficiency of any one of the essential minerals can result in severe metabolic disorders and compromise the health of the organism. Some minerals deficiencies are common in developing countries, but mineral subdeficiencies may also occur in developed countries (Afinah *et al.*, 2010).

2.6.1. Functions and bioavailability of mineral elements

Minerals serve one or more functions in the body. They are constituents of skeletal tissues, cofactors to enzymes, carrier proteins, protein hormones and electrolytes in body fluids and cells (Okoye, 1992). Bioavailability of mineral elements is affected by a number of factors which includes their chemical form, the compositions of diet and health situation of the individuals. Foods and diets of animal origin have been shown to contain mineral elements in forms that are more readily absorbed (Pennington *et al.*, 1988), while those of plant origin are less available due to the presence of some anti-nutritional factors (FAO/WHO, 1998). For instance, Okoye (1992) observed that iron deficiency anaemia might result from poor iron content of staple diet, poor absorption from the gut lumen and excessive concentration of phytates and tannins in the diet or the form of iron present in the staple diet. Zinc absorption is also impaired by phytates and fibre (FAO/WHO, 1998). Dietary calcium deficiency in Nigeria is attributed to the consumption of diets with high phytate and phosphate levels (Okonofua, 2002). Children often have an inadequately low calcium intake thereby giving rise to high prevalence of nutritional ricket (Oginni *et al.*, 1996; Thacher *et al.*, 2000). Other factors include mineral – mineral interaction, processing methods that result into loss of minerals, and ignorance of mineral-rich foodstuff found in the communities. In developing countries, the diets of most people or families, which are basically plant-based, are marginal for micro nutritional adequacy.

However, many populations might ingest inadequate quantities of calcium, iron, and zinc and may be marginally deficient in magnesium. Thus, concern continues about the impact of dietary phytate upon the mineral status of certain vulnerable segments of the population including children, teenagers, pregnant women, and the elderly. For the first three groups, representing periods of prolific growth, the phytate/mineral ratios are critical. In the final group, the baby-boomers, inadequate nutrient intakes coupled with use of over-the-counter medications may further compromise mineral status. The changing face of the food supply through genetic modification, fortification, creation of functional foods, and increased use of supplements will affect mineral nourishment (Afinah *et al.*, 2010).

2.6.2. Iron

It is highly unlikely that life in any form can exist without iron because of its diverse functions. It is essential for the production of haemoglobin, which helps deliver oxygen from the lungs to body tissues, transport electrons in cells, and synthesis of iron-containing enzymes that are required to utilize oxygen (O_2) for the production of cellular energy (Cook, 1982). The body□s iron stores, iron absorption and iron loss determines iron balance. At least two-third of body iron is functional iron, mostly haemoglobin within circulating red blood cells, with some myoglobin in muscle cells and parts of iron-containing enzymes. Most of the remaining body iron is storage iron (existing as ferritin and haemosiderin), which serves as a deposit to be mobilized when needed (ACC/SCN, 2000). The reduction of body iron has three main stages;

i.Iron depletion, which refers to a decrease of iron stores measured by reduction in serum ferritin concentration.

ii. Iron deficient erythropoeisis, when storage iron is depleted and there is insufficient iron absorption to counteract normal body losses.

iii. Iron deficiency anemia, which is the most severe degree of iron deficiency that ensues if the haemoglobin concentration falls below normal (Gillespie and Johnson, 1998).

While the biochemical liabilities of deficiencies are evident, the efficacy of body iron conservation and iron's ability to generate reactive species should caution against supplying excess iron to those with adequate iron reserves (Gillespie and Johnson, 1998).

Iron deficiency and its anaemia (IDA) is the most common micronutrient malnutrition problem in the world as it affects more than 3.5 billion people globally, of which about 2 billion are children and women (UNICEF/UNU/WHO/MI, 1999). It is associated with an estimated 111,000 maternal deaths each year (SCN, 2004). IDA is a serious health condition that results from insufficient intake and/or poor absorption of iron in the diet and can decrease mental and psychomotor development in children. It can also increase both morbidity and mortality of mother and child at childbirth, decrease work performance and decrease resistance to infection (Mohammadi *et al.*, 2011).

Iron is present in food in both heme (in flesh foods such as meat, fish and poultry) and nonheme forms in dairy products and eggs, and in plant foods such as beans, cereals, nuts, fruits and

vegetables (Ruel and Levin, 2006). Heme iron is highly bioavailable (15 to 35 percent is absorbed), whereas nonheme iron is much less bioavailable, with absorption rates ranging from 2 to 20 percent. The factors that influence the amount of iron absorbed from a meal include the individual's iron status and requirements, the sources and content of iron in the meal, and the other meal constituents. Absorption of both heme and nonheme iron is affected by the individual's characteristics, but nonheme iron is particularly sensitive to the presence of inhibitors of iron absorption such as phytic acid, tannins, and selected dietary fibers (Hallberg, 1981). Ascorbic acid and even small amounts of meat and fish, on the other hand, are active promoters of nonheme iron absorption ((Ruel and Levin, 2006).

Staple crops provide a large proportion of the total daily intake of energy and micronutrients among poor populations who have limited access to animal foods (Allen *et al.*, 1992). The main sources of iron in these populations' staple cereals, starchy roots, tubers and legumes are in the nonheme iron form and have low bioavailability. Estimates indicate that cereals contribute up to 50 percent of iron intakes among households from lower socioeconomic groups in developing countries (Ruel and Levin, 2006). The main problem with diets based on non-animal staples is that they usually contain large amounts of phytic acid (Allen *et al.*, 1992), the most potent inhibitor of nonheme iron absorption. Strategies to reduce the phytic acid concentration of the diet should therefore be prioritized as one of the crucial food-based approaches to increase the bioavailability of iron from plant based diets. Experience with these strategies is reviewed in the following sections as well as the strategies that aim at increasing intake of animal products.

2.6.3. Zinc

Zinc is an essential trace mineral that is a component of over 200 enzymes and is known to be necessary for normal collagen synthesis and mineralization of bones, and is involved in vital processes such as mitosis, synthesis of DNA and protein, and gene expression and activation (McCall *et al.*, 2000). Zinc deficiency is a public health problem, and is associated with poor growth, decreased immune function, increased susceptibility to and severity of infections, adverse outcomes of pregnancy, and neurobehavioral abnormalities (Walingo, 2009). In animals, zinc deficiency has been associated with abnormalities in bone growth, bone formation, and

mineralization whereas in humans zinc deficiency has been associated with poor bone health and low bone mass in women (Walingo, 2009).

It is well known that zinc is present in many foods, but in most developing countries children have a low intake of foods rich in readily absorbable zinc, such as liver, red meat, poultry, fish, oysters, and crabs(Walsh *et al.*, 1994). Traditional staple foods, such as cereals, legumes, and tubers, contain zinc. Whereas the zinc content of beans is one of the highest among vegetable sources; it is nearly equal to that of dairy products but is far inferior to that of meats. Evaluation of the bean core collection revealed a range of 21 to 54 ppm in zinc content, with an average value of 35 ppm (Beebe *et al.*, 2000).

Zinc nutrition status influences the absorption, transport and utilization of vitamin A. The enzyme that plays a major role in the oxidative conversion of retinol to retinal is zinc dependent, and may be adversely affected in zinc deficiency. Total zinc content from the diet and bio-availability from the diet's food combination also influence the efficiency of zinc absorption (Walingo, 2009). Zinc inhibitors like phytates , fibre, oxalate, EDTA, and polyphenols such as tannins are present in higher amounts in plant foods, especially cereals and legumes, and influence zinc absorption (bioavailability). These substances form insoluble complexes with zinc, preventing its absorption(Walsh *et al.*, 1994 ; Walingo, 2009). Cow's milk, because of its high concentrations of calcium and casein, and soymilk, because of its phytate content, may further reduce the absorption of zinc from the diet. In contrast, zinc in breast milk is well absorbed. Vegetables and fruits contribute very little to dietary zinc intake, but fruits eaten with cereals may increase the bioavailability of zinc (Bell *et al.*, 1987)

2.6.4. Calcium

Calcium is a divalent cation with an atomic weight of 40. In the elementary composition of the human body, it ranks fifth after oxygen, carbon, hydrogen, and nitrogen, and it makes up 1.9% of the body by weight (Nordin, 1976). Carcass analyses show that calcium constitutes 0.1–0.2% of early fetal fat-free weight, rising to about 2% of adult fat-free weight. In absolute terms, this represents a rise from about 24 g (600 mmol) at birth to 1300 g (32.5 mol) at maturity, requiring

an average daily positive calcium balance of 180mg (4.5mmol) during the first 20 years of growth (WHO/ FAO, 2004).

Calcium is an essential nutrient that plays a vital role in neuromuscular function, many enzyme-mediated processes and blood clotting, as well as providing rigidity to the skeleton by virtue of its phosphate salts (WHO/FAO, 2004). Calcium (Ca) is an important mineral in which more than 99% of which is found in the bones, and teeth to keep them strong, and support their structure (Shils, 1999 ; WHO/FAO, 2004). The rest is stored in blood, muscles, and cells. It is obtained from the foods including: Milk, Cheese and Yogurt, Green Vegetables etc. Calcium has a very important role in bone and tooth structure, in blood clotting, muscle contraction. Those of us who do not consume enough Calcium should take Calcium supplements. The exact amount of Calcium depends on age and other factors; however, children, and teenagers need more Calcium compared to adults (NIHODS, 2007). Aged women need Calcium to prevent Osteoporosis, which weakens the bones that are likely to get broken. Half of women and men under 50 get their bones broken due to Osteoporosis. Therefore, a diet rich in Ca and vitamin D keep bones strong. Calcium forms a vital part of bone and tooth structure, and is also important as a positive ion (Ca^{2+}) in blood clotting, muscle contraction, and nerve impulse transmission. It also participates in Glycogen metabolism (Heydon, 1983).

Inadequate intake of Calcium increases the risk of Osteoporosis (bone loss with no apparent cause). Adequate Calcium nutrition during childhood has important implications for bone growth, and development, and is thought to reduce the incidence of Osteoporosis in later life. Excess intake of Calcium may cause kidney stones and reduces mineral absorption in general (Wardlaw and Insel, 1996). Phytic acids markedly decrease Calcium bioavailability, and the Calcium/ Phytate molar ratio has been proposed as an indicator of Calcium bioavailability. The critical molar ratio of Calcium/Phy is reported to be 6:1 (Oladimeji *et al.*, 2000).

Calcium is the outstanding single constituent of bones and teeth, and the most abundant mineral in the body with recommended daily allowance(RDA) for adults 1200 milligrams or approximately one gram. Calcium and $PO4^{3-}$ are properly utilized in the body by vitamin D (Anderson and smith, 2002). The best natural sources are Sea Vegetables, Low-Fat Yogurt, Skim

Milk, Beans, Seeds, Nuts, Green Vegetables, etc. Intakes over 2000 milligrams per day may lead to Hypocalcaemia, induce constipation, and inhibit the intestinal absorption of Iron, Zinc, and other essential minerals (Banerji, 2005).

Many factors influence the availability of calcium for absorption and the absorptive mechanism itself. In the case of the former, factors include the presence of substances which form insoluble complexes with calcium, such as the phosphate ion (WHO/FAO, 2004). The relatively high calcium–phosphate ratio of 2.2 in human milk compared with 0.77 in cow milk may be a factor in the higher absorption of calcium from human milk than cow milk (Nordin, 1976). And also Phytates, present in the husks of many cereals as well as in nuts, seeds, and legumes, can form insoluble calcium phytate salts in the gastrointestinal tract. Excess oxalates can precipitate calcium in the bowel but are not an important factor in most diets (WHO/FAO, 2004).

2.6.5. Phosphorus

Phosphorus (as phosphate) is an essential constituent of all known protoplasm and is uniform across most plant and animal tissues. It is part of the phospholipids, an essential functional component of cell membranes, and is part of high energy phosphate compounds like e.g. adenosine triphosphate (ATP) and creatine phosphate, the biological energy conservation molecule which is essential to all vital processes (Michaelsen *et al.*, 2008). Phosphorus is also an essential component of hydroxyapatite, the main structural bone mineral. Deficiency of phosphorus is common in malnourished children and severe hypophosphatemia is associated with increased mortality in kwashiorkor (Manary *et al.*, 1998). Phosphorus deficiency is also likely to cause rickets-like bone changes in malnourished children (Golden, 2009).

Because of its wide distribution in plant and animal cells, phosphorus is present in most foodstuffs. Some of the phosphorus present in plant foods, notably legumes and whole grain cereals, is in the form of phytic acid (inositol hexaphosphate) which forms insoluble complexes with divalent cations such as calcium, magnesium, zinc and iron, thereby inhibiting their absorption from the gut. Plasma phosphate concentration appears to be regulated mainly by

urinary excretion. Parathyroid hormone and calcitonin both increase phosphate excretion (Gibney *et al.*, 2009).

Absorption of dietary phosphorus is high (55-70%), relatively independent of dietary composition, and does not appear to be up-regulated at low intakes (Michaelsen *et al.*, 2008). Dairy products, meat, poultry, eggs, fish, nuts, and legumes are generally good sources of highly available phosphorus. However, the main form of phosphorus from plant material is phytate which is resistant to digestion unless enzymatically degraded by phytase. Thus, phosphorus from phytate is only absorbed to a minor degree under normal conditions and the phytate fraction of phosphorous should therefore be discounted from the calculations of the total phosphorous requirements (Golden, 2009).

2.7. Hyacinth bean as a food

Hyacinth bean seeds and pods are popular foods in South Asia , India, China, West Africa, Japan, and the Caribbean Islands (Morris, 2009). The young hyacinth bean pods, seeds, leaves, and flowers are cooked and eaten as vegetables. Dried seeds are cooked and eaten or processed into bean cakes, fermented as tempeh or before cooking, sprouted and eaten fresh as bean sprouts (Morris, 2009). The dry seeds are cooked together with rice after soaking water for one night to supply protein. However, the mature dark colored seeds require boiling prior to consumption because they contain a trypsin inhibitor broken down by heat and a toxic cyanogenic glucoside soluble in boiling water (NAS/NRC,1979).

According to Mahesh and Shridhar (2007), the young fresh green pods of hyacinth bean are used as a vegetable, while dry seeds are used to prepare cooked *dhal*. For this latter purpose, whole seeds after brief soaking in water may be used, or may be soaked in water overnight and the seed coats removed prior to cooking. Alternatively, seeds are soaked in water overnight and germinated for 24–48 h prior to using the sprouts (with or without the removal of seed coats) in a variety of dishes including soups, salads, mixed stir-fried vegetables, and *dhal* preparations.

Nutrition is advocated as the first medicine in the fight against AIDS (FAO/WHO, 2002).Many neglected legume crops can enrich soil while providing high quality human food and fodder. They can also provide environmental adaptation, nutritional value, and versatile uses (Mal, 1994). The hyacinth bean is one of these underutilized legumes because it is more nutritious than many major legume species (FAO/UNDP, 2004). Many amino acids, micronutrients, macronutrients, and vitamins for human health have been identified in hyacinth bean seeds and pods (Morris, 2009). Hyacinth bean is a legume proposed to be used in the fight against malnutrition because of its high amino acid, crude protein, carbohydrate (Chau *et al.*,1998), and mineral contents (Morris, 2009). Historically, tan to yellow colored hyacinth bean seeds and pods have been used as vegetables.

2.8. Medicinal value of hyacinth bean

Hyacinth beans are naturally rich in carbohydrates, proteins, fat and fibers as well as minerals including calcium, phosphorus and iron. Furthermore, several legumes have tremendous potential as nutraceuticals because of their healing properties (Morris, 2003). Hyacinth bean *(Lablab purpureus* L.) has great potential as medicinal legume. Among the legumes, hyacinth bean constitutes an important source of therapeutic agents used in the modem as well as traditional systems of medicine (Morris, 2003). It carries tremendous healing potential. The seeds are used as laxative, diuretic, anthelmintic, antispasmodic, aphrodisiac, anaphrodisiac, digestive, carminative, febrifuge and stomachic (Naeem *et al.*, 2009). Hyacinth beans contain fiber which is known to prevent cancer, diabetes, heart disease, obesity and is used as a laxative (Beckstrom-Sternberg and Duke, 1994). Hyacinth bean contains the potential breast cancer fighting flavonoid known as kievitone. The flavonoid, genistein found in hyacinth bean may play a role in the prevention of cancer and as a chemotherapeutic and/or chemo preventive agent for head and neck cancer (Kobayashi *et al.*, 2002) .

Tyrosinase (polyphenol oxidase) is present in plant tissue and is important in fruit and vegetable processing as well as storage of processed foods. Prevention of browning of foods, enzymatic or nonenzymatic, has long been a concern of food scientists. Hyacinth bean contains tyrosinase, which has potential for the treatment of hypertension in humans (Naeem *et al.*, 2009).

2.9. Nutraceuticals in hyacinth bean

However, additional phytochemicals have been identified in hyacinth beans through literature reviews with potential use as nutraceuticals and/or pharmaceuticals since 2003. These phytochemicals can be used for hyperlipidemia, as an antimicrobial, appetite suppressant, for osteoporosis, in hypertension, and possibly in pancreatic cancer (Morris, 2009).

Several phytochemicals for use as nutraceuticals have been discovered (Bisby *et al.*, 1994) in hyacinth beans and discussed (Morris, 2003). Numerous additional phytochemicals found in hyacinth bean seeds have potential nutraceutical use. For example, lauric acid found in hyacinth bean seeds is a potential nutraceutical (Duke, 2008). Interestingly, a clinical trial regarding lauric acid consumption by healthy men and women gave a more favorable serum lipoprotein pattern than consumption of partially hydrogenated soybean oil rich in trans-fatty acids (de Roos *et al.*, 2001). In addition, the glycerol monoester of lauric acid mixture with glycerol monolaurate has demonstrated antimicrobial activities against *Staphylococcus aureus* (Zhang *et al.*, 2007).

Myristic acid found in hyacinth bean plants (Duke, 2008) has clinically been proven to have beneficial lipidic effects in males and enhances docosahexaenoic acid of cholesteryl esters (Dabadie *et al.*, 2005). Oleic acid has been found to range from 950 to 7,832 ppm in hyacinth bean seeds (Duke, 2008). A recent clinical trial concluded that oleic acid empties from the stomach more slowly and suppresses appetite in healthy human beings (Little *et al.*, 2007). The carbohydrate myoinositol found in hyacinth bean seeds (Bisby *et al.*, 1994) has been discovered via clinical trial to significantly reduce blood pressure safely and is well tolerated in humans. The flavonoid genistein found in hyacinth bean hypocotyls (Bisby *et al.*, 1994) has recently been studied in a clinical trial regarding its effects on bone metabolism in osteopenic women. Their results indicated that bone mineral density increased in genistein recipients (Marini *et al.*, 2007). As noted earlier, hyacinth bean seed contains the amino acid leucine, which is a potential nutraceuticals. Promising clinical results have been shown for greater synthesis of muscle proteins in human males when resistance exercise is followed by ingestion of leucine enriched amino acids plus carbohydrate (Dreyer *et al.*, 2008). In addition, isoleucine found in hyacinth bean seeds has also been clinically proven to stimulate hepatic and muscle protein synthesis by

improving the amino acid composition of the upper gastrointestinal bleed by simultaneous intravenous isoleucine administration (Olde Damink *et al.*, 2007).

2.10. Antinutritional factors (ANFs)

Nature has endowed plants with the genetic capacity to synthesize substances that are toxic, and thus to ensure their survival against predators whether they be insects, fungi or animals including humans. Humans have learnt which foods are safe to eat or how such foods can be treated in order to destroy their toxicity use, and have developed suitable techniques for detoxifying the foods before consumption. One of these toxic materials are anti-nutritional factors (Shanthakumari *et al.*, 2008). Antinutrients are chemicals which have been evolved by plants for their own defense, among other biological functions. Anti-nutrients reduce the maximum utilization of nutrients especially proteins, vitamins, and minerals, thus preventing optimal exploitation of the nutrients present in a food and decreasing the nutritive value (Ugwu and Oranye, 2006).

There is a wide distribution of biologically-active constituents throughout the plant kingdom, particularly in plants used as animal feeding stuff and human nutrition. The knowledge that these compounds elicit both toxic and advantageous biological responses has given rise to several investigations in recent times as to their possible physiological implications in various biological systems (Soetan and Oyewole, 2009).

One major factor limiting the wider food utilization of many tropical plants is the ubiquitous occurrence in them of a diverse range of natural compounds capable of precipitating deleterious effects in man and animals. Compound which act to reduce nutrient utilization and/or food intake are often referred to as anti-nutritional factors. The biological effects of all these chemicals are diverse, and complex. When man ingests plant foods to meet nutritional needs, a wide variety of these non nutrient phytochemicals are ingested at the same time. Food crops regularly eaten have many beneficial nutrients but there are traces of ant nutritional factor components such as cyanides, oxalates, phytate, phenolics, tannin, protease inhibitors, heavy metals etc (Omoruyi *et al.*, 2007).

Grain legumes, such as soybean (*Glycine max* (L.) Merr.), pea (*Pisum sativum* L.), faba bean (*Vicia faba* L.), lupins (*Lupinus* spp.), common vetch (*Vicia sativa* L.), grass pea (*Lathyrus sativus* L.) and hyacinth bean (*Lablab perpureus L.) sweet*, represent one of the most quality and least expensive solutions for a long-term demand for plant protein in animal and human nutrition (Mikić *et al.*, 2009). One of the demerits to an increased use of grain legumes as feed and food is the presence of ANFs, that both decrease nutritive value of grain legumes and, if taken in larger amounts, cause health problems that may be fatal for both human and the animals. By this reason, food processing of all grain legumes is aimed at decreasing the content of anti-nutritional factors to a safe extent, leading to an increased proportion of grain legumes in diets for all species and categories of animals and human beings (Mikić *et al., 2009).*

Although legume seeds contain a moderately high amount of macro and micro nutrients in which their use in food and feed is still limited by the presence of several ANFs. These include tannins, phytic acid, trypsin inhibitors and flatulence causing oligosaccharides. Among all the anti-nutritional components, phytic acid and tannin are more dominant in legumes as one of prime concern for human nutrition and health management (Mohamed *et al.*, 2011). The amount of phytic acid ranged from 233.33 to 599.67 mg/100g, whereas range for tannic acid is in between 164.70 and 371.67 mg/100g of the legume used in current study (Huma *et al.*, 2008). The amount of these antinutrients is higher in pigmented legumes as compare to white colored legumes (Huma *et al.*, 2008).

2.10.1. Phytic acid

Phytic acid (myoinositol, 1, 2, 3, 4, 5, 6 hexakisdihydrogen phosphate) and phytate (salts of phytic acid) are widespread in plant seed grains (also including cereals), roots, tubers (and legumes (Mohamed *et al.*,2011). Phytate accumulates in the seeds during the ripening period and is the main storage form of both phosphate and inositol in plant seeds and grains.

The phytate molecule is negatively charged at physiological pH and is reported to bind with essential, nutritionally important divalent cations such as Fe^{2+}, Zn^{2+}, Mg^{2+}, Ca^{2+}.... etc., and forms insoluble complexes, thereby making minerals unavailable for absorption (Mohamed *et al.*,2011). It also forms complexes with proteins, amino acids and starch and inhibits their

digestion (Oatway *et al.*, 2001). Figure (2.4a) shows the example of a phytate interaction with iron and a protein. The dephosphorylation of phytate is a prerequisite for improving nutritional value because removal of phosphate groups from the inositol ring decreases the mineral binding strength of phytate. These results increased bioavailability of essential dietary minerals (Sandberg *et al.*, 1999). Solubility is a prerequisite for absorption of most minerals, although solubility at neutral pH has been shown to be less important for calcium absorption. The chemical structure of phytic acid is indicative of strong chelating potential (Figure 2.4b).

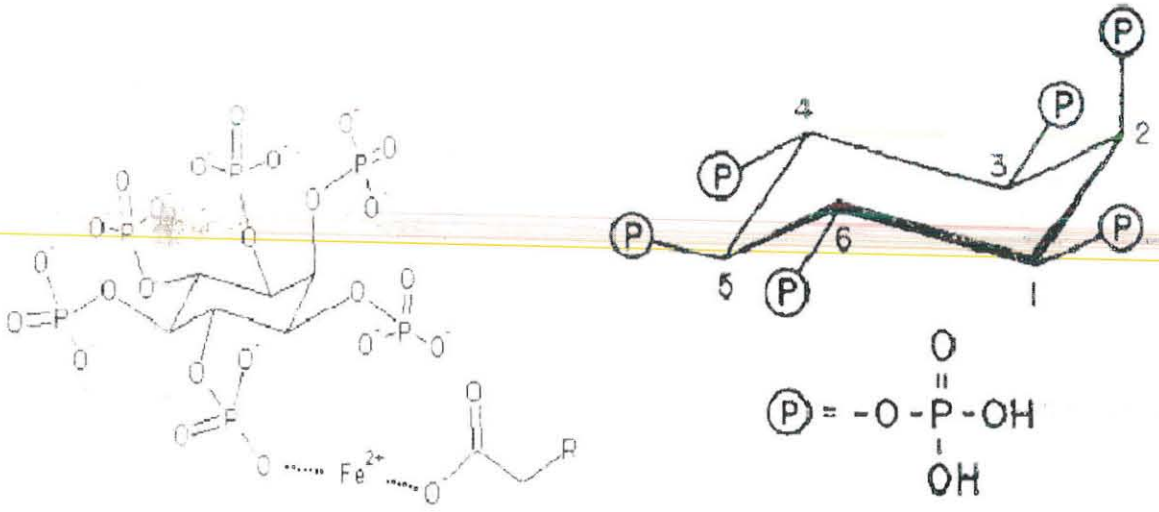
Besides, phytate has also been reported to form complexes with proteins at both low, and high pH values. These complex formations alter the protein structure, which may result in decreased protein solubility, enzymatic activity, and proteolytic digestibility. The phytate degrading enzyme, phytase, is in vogue for degradating phytate during food processing, and in the gastrointestinal tract. The major concern about the presence of phytate in the diet is its negative effect on mineral uptake (Greiner *et al.*, 2006).

Conversely, as a strong chelator of iron and zinc, phytate in plant foods actually can serve as an antioxidant to reduce free radical formation mediated by these metals. The formation of insoluble mineral-phytate complexes at physiological pH values is regarded as the major reason for the poor mineral bioavailability, because these complexes are essentially nonabsorbable from the human gastrointestinal tract (Greiner *et al.*, 2007).

The major past anxiety over seed-derived dietary phytic acid has been its role in mineral diminution and deficiency. Human populations that manage to survive on whole grain and/or legume staple foods consume large amounts of phytic acid (Table 4.2), and this may contribute to their risk for mineral depletion and deficiency. However, dietary phytic acid may also have vital positive roles, for example, as an antioxidant and an anticancer agent. The recognized benefits of dietary phytic acid may be a more important consideration in certain populations than concerns over mineral deficiency. The question of seed-derived dietary phytic acid in human nutrition and health is more complicated by the fact in the cereal grain, phytic acid is deposited in the aleurone and germ, which is also the site for the grain's main mineral stores. Removal of

these tissues during milling or polishing removes most phytic acid and most of the grain's mineral deposits (Afinah *et al.*, 2010).

Because phytate binds essential minerals and can prevent their absorption, most human nutritionists view the compound negatively. However, its unique chelating action with iron provides phytate with antioxidant characteristics (Burgess and Gao, 2002). More recently, studies indicate that phytic acid is a natural antioxidant important for seed viability. Evidence also indicates that the inhibitory effects of phytate on mineral absorption are not seen in varied diets containing animal protein. Phytate may actually be beneficial as a dietary antioxidant in an animal protein diet. There may be nutritional advantages, or at least no disadvantage, to addition of phytate to meat products (Afinah *et al.*, 2010).



(a) (b)

Figure 2.4: (a) Phytate showing an example of an interaction with iron and protein and (b) Structure of phytic acid in dilute solution. (Adapted from Afinah *et al.*, 2010)

Table 2.2: Seeds/grains/fruits with seeds that are commonly eaten by humans that contain phytic acid concentrations of one percent or more on a weight basis.

Plant	Structure	% phytic acid
Sesame	Dry seed	4.17
Pumpkin/squash	Embryo	4.08
Flax (linseed)	Dry seed	3.69
Rape seed	Dry seed	2.50
Sunflower	Embryo	2.10
Mustard	Dry seed	2.00
Cashew	Embryo	1.97
Brazil and other tree nut	Embryo	1.80
Hemp	Dry fruit	1.74
Peanut	Seed in shell	1.70
Tomato	Dry Seed	1.66
Soybean	Dry seed	1.55
Almond	Dry Embryo	1.42
Egg plant	Seed only	1.42
Beans	Dry seed	1.41
Pistachio	Embryo	1.38
Water melon	Seed only	1.36
Kiwi fruit	Fleshy fruit	1.34
Broad beans	Dry seed	1.11
Cucumber	Immature seed	1.07
Sorghum	Dry grain	1.06
Cocoa bean	Dry seed	1.04
Barley	Dry grain	1.02
Oats	Dry grain	1.02
Wheat	Dry grain	1.02
Peas	Dry seed	1.00

(Adapted from Afinah *et al.*, 2010).

Phytic acid has six strongly dissociated protons (pKs 1.1 to 2.1) and six weakly dissociated protons (pKs 4.6 to 10.0). The effect on minerals is observed through the formation of phytate-mineral or peptidemineral-phytate complexes. These complexes have stoichiometries of the $M^{+(n)}$-phytate type (n = 1–6). Phytate forms a wide variety of insoluble salts with divalent and trivalent cations. Usually, the divalent cations (e.g.: Zn^{2+}, Ca^{2+}, and Mg^{2+}) form insoluble penta- and hexa-substituted salts. The insolubility of these complexes is regarded as the major reason for the reduced bioavailability of minerals due to diets high in phytic acid. Several factors determine the effect of phytate on mineral bioavailability: pH, size and valence of the mineral, mineral and phytate concentrations and ratios, and food matrix that include the presence of enhancers and/or inhibitors (Weaver and Kannan, 2002).

2.10.2. Tannins

Plant tannins are ubiquitous in nature and although a lot of attention has been given to their study, the term "tannin" continues to be difficult to define precisely. Indeed, whereas related phenolic compounds such as simple phenolics, neolignans and flavonoids are characterised and classified according to their chemical structure. Tannins are a diverse group of compounds that are related primarily in their ability to complex with proteins (Fahey and Jung, 1990). Thus, tannins are usually defined as water-soluble polyphenolic substances that have high molecular weight and that possess the ability to precipitate proteins. Furthermore, researchers proposed that the molecular weight of tannins should lie between 500 to 3000, permitting the tannin molecule to easily orientate itself between the protein chains, but having sufficient phenolic groups to crosslink efficiently with protein (Goldstein and Swain, 1965; McLeod, 1974). However, the definition of tannins is regularly modified in the light of new findings (Mueller-Harvey and McAllan, 1992). As a consequence, the list of polymers bound by tannins has been extended to include polysaccharides (cellulose, hemicellulose and pectin) and nucleic acids, steroids, alkaloids, and saponins (Haslam, 1986). While this working definition is useful in describing some chemical and physical characteristics of tannins, it is nevertheless vague and could obfuscate the research on tannins (Ayres *et al.*, 1997).

Tannins are naturally occurring plant polyphenols that can have a large influence on the nutritive value of forage legumes. The definition that is based solely on the ability of polyphenols to precipitate proteins is too restrictive for consideration of the nutritional effects of this diverse group of plant compounds (Reed, 1995). Horvath (1981) developed a broader definition: "any phenolic compound of sufficiently high molecular weight containing sufficient phenolic hydroxyls and other suitable groups (i.e., carboxyls) to form effectively strong complexes with protein and other macromolecules under the particular environmental conditions being studied." The ability to complex with minerals could also be added to the definition. This definition allows for the fact that tannins may form complexes with starch and cellulose as well as protein. Tannins exist in mixtures with many other classes of plant phenolic compounds. Inconsistent relationships between the definition of tannins and the analysis of mixtures of phenolic compounds complicate research on the nutritional toxicology of tannins in forage legumes (Reed, 1995).

Although tannins are chemically a diverse and ill-defined group it is usual to divide them into two main classes: (a) the hydrolysable and (b) the condensed tannins (Mangan, 1988). *Hydrolysable tannins* are polyesters of phenolic acids such as gallic acid, *m*-digallic acid (gallotannins) or hexahydroxydiphenic acid (ellagitannins) and D-glucose or quinic acid, the latter serving as a polyalcohol core (Mueller-Harvey and McAllan, 1992). *Hydrolysable tannins* receive their name because they are readily cleaved by enzymes (i.e. *Penicillium* tanninase) as well as by dilute acid to give a sugar such as glucose and a phenolcarboxylic acid such as gallic acid (Strumeyer and Malin, 1975).

Tannin is an astringent, bitter plant polyphenolic compound that either binds or precipitates proteins and various other organic compounds including amino acids and alkaloids (Harold, 2004). The compounds are widely distributed in many species of plants, where they play a role in protection from predation, and perhaps also in growth regulation. Tannins have traditionally been considered as ANFs but it is now known that their beneficial or anti-nutritional properties depend upon their chemical structure and dosage and the total acceptable tannin daily intake for a man is 560 mg (Anonymous, 1973). The new technologies used to analyze molecular and chemical structures have shown that a division into condensed and hydrolyzable tannins is far too

simplistic and readily form indigestible complexes with proteins and other macromolecules under specific environmental conditions (Mole and Waterman, 1987).

Recent studies have demonstrated that products containing chestnut tannins included at low dosages (0.15-0.2 %) in the diet can be beneficial (Schiavone *et al.*, 2008). The most abundant polyphenols are the condensed tannins, found in virtually all families of plants, and comprising up to 50% of the dry weight of leaves. Condensed tannins inhibit herbivore digestion by binding to consumed plant proteins and making them more difficult for animals to digest, and by interfering with protein absorption and digestive enzymes. Tannins had been reported to affect tprotein digestibility, adversely influencing the bioavailability of non-haem iron leading to poor iron and calcium absorption, also carbohydrate is affected leading to reduced energy value of a diet containing tannins (Reed *et al.*, 1990).

Certain polyphenoles are able to bind Fe, which make the complex-bound Fe unavailable for absorption. The amount of Fe-binding phenol galloyl groups in foods roughly corresponds to the degree of inhibition of Fe absorption. All major types of food polyphenoles can strongly inhibit dietary non-haem iron absorption. The negative influence on Fe absorption is nutritionally the most important, especially in industrial products such as infant formulas, but more importantly in many developing countries where the diet is based on cereal and legume products. Cereals contain varying amounts of polyphones and generally the amounts are considered higher in the colored seeds (Sandberg, 2002).

Tannins in forage legumes have both negative and positive effects on nutritive value (Reed *et al.*, 1990; Mueller-Harvey and McAllan, 1992). Tannins in high concentrations reduce intake, digestibility of protein and carbohydrates, and animal performance (Reed *et al.*, 1990). Tannins in low to moderate concentrations prevent bloat and increase the flow of non-ammonia nitrogen and essential amino acids from the rumen (Waghorn *et al.*, 1987; McNabb *et al.*, 1993). The positive effects of tannins on protein utilization have practical importance because problems associated with extensive proteolysis and(or) deamination in the rumen limit production in modern feeding systems (Beever *et al.*, 1989).

2.11. Strategies for reducing anti nutritional factors

The removal of undesirable components is essential to improve the nutritional quality of legumes. In this way, these could effectively be utilized to their full potential as human food. It is widely accepted that simple and inexpensive traditional processing techniques are effective methods of achieving desirable changes in the composition of seeds. Soaking, cooking, fermentation and germination may improve the quality of legumes due to the removal of some anti-nutritional factors. In many instances, usage of only one method may not impart the desired removal of anti-nutritional compounds and a combination of two or more methods is required (Ibrahim *et al.*, 2002).

Different individual traditional processing or combinations of them are pronounced of reducing anti-nutritional factors and raising nutrients bioavailability. How efficient processing might affect composition of nutrients and to which extent it will decrease ANFs are still considered as vital issue to be searched (Dhurandhar And Chang, 1990). Such work will be of potential benefits, offers recommendations to manufacturers and helps to formulate accurate diets to humans and animals.

Generally, legumes have been reported to have low nutritive value because of low protein digestibility and the presence of ANFs. Legumes are usually cooked before being used in the human diet. This improves the protein quality by destruction or inactivation of the heat labile anti-nutritional factors (Alajaji and El-Adawy, 2006). However, cooking causes considerable losses in soluble solids, especially vitamins and minerals (Alajaji and El-Adawy, 2006).

The *Dolichos lablab* bean is one of the lesser-known legumes of arid and semi-arid land. The bean classified by the National Academy of Science (NAS) as potential source of protein that has not been explored yet. Studies on nutrient composition showed that the bean is good source of protein, carbohydrate and energy (Duke, 1983; Deka and Sarkar, 1990). The ANFs level of theses bean have been studied by Deka and Sarkar(1990). Deka and Sarka (1990) reported that the trypsin inhibitors activity level ranged from 11.8 to 29.0 TIA /gm, Phytic acid level varied from 100.0 to 313.4 mg/100gm and tannin content of untreated lablab bean has been reported to be high in a *lablab purpueus*. In order to utilize bean effectively as human food, it is essential to inactivate or remove these ANFs. Different processing methods are employed on hyacinth bean as studied by Osmaman (2007). From the study conducted by Osman, trypsin inhibitor level is

reduced by 66.66% during cooking, 23.05% by roasting and 19.39% by germination whereas roasting causes greatest reduction (60.69%) on phytic acid content followed by autoclaving (52.29%), germinating (48.94%), cooking (44.85%) and soaking (22.19%). Generally, adequate heat processing inactivates the trypsin and phytate in hyacinth bean (Osman, 2007). Heat stable compounds in cereal and legumes such tannins and pytates are easily removed after germination (Reddy *et al.*, 1985) and fermentation (Osman, 2004). A better understanding of the effect of different traditional processing methods on nutritive value, may lead to wider use of this legume at home and in food industries (Osman, 2007).

Various food processing and preparation techniques, such as decortication, soaking, cooking, germination and fermentation, are the major efforts made to reduce the amounts of phytate in foods (Liang *et al.*, 2008; Khattab and Arntfield, 2009; Wang *et al.*, 2010; Kumar *et al.*, 2010). The most effective treatments are fermentation and germination (Honke *et al.*, 1998) but their application remains limited because of the additional workload they imply or the particular organoleptic properties they induce.

Khokhar and Chauhan (1997) reported the ANFs in Moth bean (*Vigna aconitifolia*). The dry seeds were given different treatments including soaking, sprouting and cooking and the changes in the level of the anti-nutritional factors were estimated. Soaking the seeds in plain water and mineral salt solution for 12 h decreased phytic acid to the maximum (46–50%) whereas sprouting for 60 h had the most pronounced saponin lowering effect (46%). The other methods of processing were less effective in reducing the levels of these ANFs. The processing methods involving heat treatment almost eliminated trypsin inhibitor activity while soaking and germination partly removed the activity.

El-Adawy (2002) reported the nutritional composition and ANFs of chickpeas (*Cicer arietinum L.*) undergoing different cooking methods and germination. The studies involved the effects of cooking treatments (boiling, autoclaving and microwave cooking) and germination on the nutritional composition and ANFs of chickpeas. Cooking treatments and/or germination caused significant ($p < 0.05$) decreases in ANFs. Germination was less effective than cooking treatments in reducing trypsin inhibitor, hemagglutinin activity, tannins and saponins; it was more effective in reducing phytic acid, stachyose and raffinose.

3. Materials and methods

3.1. Study area

This study was conducted in Debati woreda, Metekel zone of Beneshangul Gumz Regional State, Western Ethiopia. The region has a total area of approximately 50,380 km^2 with altitude ranging from 580 to 2,731 meters above sea level. Agro-ecologically, it is divided into lowlands (below 1500 meter), medium altitude (between 1,500-2,500 meter) and highland (above 2,500 meter) above sea level. Annual rainfall varies between 800 and 2000 mm. The temperature reaches a daily maximum of 20^0C to 25^0C in the rainy season and rises to 35^0C to 40^0C during dry season. The hottest period is from February to April. The minimum daily temperatures range from 12^0C to 20^0C, depending on season and altitude.

The capital city of the region is named Assosa and is 687 km from Addis Ababa while Debati woreda is located at 269 km south west of Assosa. The woreda has twenty nine kebeles and the majority of the kebeles have a good weather condition to cultivate the hyacinth bean for self consumption in the woreda.

3.2. Preliminary data collection

A technical questionnaire was prepared for diagnostic survey and collection of data. It was to get adequate initial information about the hyacinth bean. Three groups of respondents were selected namely hyacinth bean cultivators, processors and consumers from both sexes. These were women processing hyacinth bean (*Lablab purpureus L.*) sweet for various products, local community consuming different products of the crop and farmers involved cultivation of the crop. The data was used to structure the actual study and as a supplementary for the main study part in this research work.

3.3. Sampling and sample preparation

Two varieties of hyacinth bean (*Lablab purpureus L.) sweet* (Rongai and Highworth) were purchased from local markets of Debati and Berber in which most of the farmers in the woreda participate. The samples were purchased from the sellers by the random sampling technique and hand-sorted to remove wrinkled, moldy seeds and foreign materials, then packed in polyethylene bags and transported to Laboratory of Food Science and Nutrition Graduate Programme.

3.4. Processing cooking treatments

The collected samples were divided into two parts. One part was analyzed without further treatment whereas the other portion was subjected to various treatments.

Soaking: Hyacinth bean seeds were soaked in distilled water (1:10, w/v) for 12 h at room temperature (25 ^{0}C). The soaked seeds were drained and rinsed three times with 600 mL distilled water and then either boiled or autoclaved as described below before analysis:

Boiling: The rinsed and soaked seeds were cooked in tap water and the cooking time measured according to (Alajaji and El-Adawy, 2006). Briefly, 250 mL of tap water was brought to boiling point in an aluminium container and then a 25 g seed sample was added. After 80 min, one seed was withdrawn without interrupting the boiling. The degree of its cooking was tested by pressing the seed between the forefinger and thumb. If the seed felt uncooked, another seed was tested after further 5 min. At each interval the degree of cooking of the boiled seed was determined by testing up to five seeds and this was kept on until final cooking indicated its doneness. The time at which boiling stopped was recorded as time of doneness.

Autoclaving: The rinsed and soaked seeds were autoclaved using vertical autoclave (Systec, Model Systec V-150, Wettenberg, Germany) at 15 lb pressure (121 ^{0}C) in tap water (1:10, w/v). Briefly, 250 mL of tap water was added to an aluminum container and then a 25 g seed sample was added. Finally the cooking was checked until 50% of the seeds were soft when pressed between the fingers (at 35 min). After the samples were withdrawn from the autoclave, it was immediately dried by oven at a temperature of 65^{0}C for 6 hours. Therefore, from the above cooking procedures four processing categories: raw, soaked, boiled and autoclaved were achieved for the two varieties of the Hyacinth bean studied.

3.5. Analytical methods

Proximate, mineral and anti-nutritional analyses were made on raw, soaked, boiled and autoclaved seeds of Hyacinth beans following the method indicated respective to each nutrient and anti-nutrients.

3.5.1. Proximate and mineral analyses

3.5.1.1. Determination of moisture content

Moisture content of raw and processed hyacinth bean samples were determined according to AOAC (2000), using the official method 925.05.

The moisture content was determined by drying oven method. The empty dishes used for the moisture determination were cleaned and dried at 105°Cfor 1 hr (Wagtech Britania drying oven of model DHG-9055A) and transported into desiccators (with granular Silica gel) for about 30 min. The mass of each dish was measured (M_i) and regulated until constant weight was obtained. About 5 g of the hyacinth bean flour was weighed into the dishes and recorded as (M_2). The sample was mixed thoroughly and dried at 105 ^{0}c for 4 hours. And it was taken and kept in desiccators until it gets cool. After cooling, the mass was measured and recorded as (M_3). The dishes and their contents were cooled in desiccators to room temperature and reweighed. The moisture content was determined by measuring the weight of a sample before and after the water was removed and calculated from the equation:

$$\text{Moisture (\% w/w)} = \frac{M2-M3}{M2-M1} X100$$

Where: M_1 = Mass of the dish

M_2 = Mass of the dish and the sample before drying

M_3 = Mass of the dish and the sample after drying

3.5.1. 2. Determination of crude protein content

Protein content of the raw and processed hyacinth bean samples was determined according to AOAC (2000), using the official method 979.09.

The crude protein content in the sample of hyacinth bean flour was quantified by Kjeldahl Methods. About 0.5 g of hyacinth bean flour was added to the Kjeldahl digestion flask and weighed by analytical balance (model LA 204). Then 6 mL of concentrated sulphuric acid was added and 3.5 mL of 30% hydrogen peroxide solution was added to the digestion flask step by step. Thereafter, the digestion flasks were shaked until the violet reaction disappeared. About 3 g of the catalyst mixture (i.e, copper sulphate($CuSO_4$) and potassium sulphate (K_2SO_4) were added in to the digestion flask and the digested by heating at 370^OC for four hours. After digestion was completed, formed clear solution was cooled for 30 minutes and neutralized by addition of 25 mL 40% NaOH(sodium hydroxide) and diluted using 25 mL of distilled water. 25 mL of distilled water, 25 ml of boric acid (4%) and 3 drops of Methyl red indicator were added into receiving flask 250 mL capacity connected to a distiller with a tube. The distillation process was terminated when the volume of the receiving flask reached between 200 and 250 mL. Note: all the reagents were added to the blank except the sample. The nitrogen content was estimated by titration of the borate anion formed with 0.1N HCl. The amount of nitrogen was calculated using the formula:

$$\text{Nitrogen (\% w/w)} = \text{NHCl}\,\frac{(Vs-Vb)X14}{W}\,X100$$

Where

V_S = Volume (mL) standard HCl solution used in the titration of the sample

Vb = Volume (mL) standard HCl solution used in the titration of the blank

W = Sample weight

N = Normality hydrochloric acid

14 = The molecular weight of nitrogen.

The protein content was calculated from the equation:

Protein content (% w/w) = 6.25 X %N

3.5.1.3. Determination of crude fat content

Crude fat content of the raw and processed hyacinth bean flours were determined according to AOAC (2000), using the official method 4.5.01.

Crude fat was determined by semi-continuous solvent extraction method (Soxhlet methods). The flasks used for the extraction was cleaned by placing them in Wagtech Britania drying oven (model DHG-9055A) at 105^0c for 1 hr and cooled in desiccators. The masses of the cooled flasks was measured by analytical balance (model LA 204). Accordingly, for all samples of different process undertaken in the experiment, about 2g of powdered hyacinth bean flour were placed in the cellulose extraction thimble and the thimble was covered with fat free cotton. The thimble was placed in an extraction chamber which is suspended above the flask containing the solvent (50 mL of diethyl ether) and below a condenser. The flask which had dried in a drying oven at 105^0C containing boiling chips was placed inside the extraction chamber and heated at 55^0C and the solvent evaporates and move up into the condenser where it is converted into a liquid that trickles into the extraction chamber containing the sample. At the end the extraction process, which typically lasts for four hours, the flask containing the solvent and the lipid was removed, the solvent was evaporated in drying oven at 70^0C and the mass of lipid remaining was quantified gravimetrically and calculated from the difference in weight of the extraction flask before and after extraction as percentage. The crude fat in the initial sample was calculated as :

$$\text{Fat content (\% w/w)} = \frac{Weight\ of\ fat}{Weight\ of\ sample}\ x\ 100$$

3.5.1.4. Determination of crude fiber content

Crude fiber content of raw and processed hyacinth bean flour samples were determined according to AOAC (2000), using the official method 920.169.

About 1.6 g of the sample was weighed in each of 600 mL beaker. 200 mL of 1.25% sulfuric acid solution was added to each beaker and allowed to boil for 30 minute by rotating and stirring periodically. During boiling the level was kept constant by addition of hot distilled water. After 30 minutes 20 mL of 28% potassium hydroxide solution was added into each beaker and allowed to boil for another 30 minute, and then the solution found in each of the beaker was filtered through crucibles containing sand by placing each of them on Buchner funnel fitted with No.9

rubber stopper. During filtration the sample was washed with hot distilled water and finally the residue was washed with 1% sulphuric acid solution, hot distilled water, 1% sodium hydroxide solution and at last with acetone. Each of the crucibles with their contents were dried for 2 hour at about 130^0C and cooled in desiccators and weighed and recorded as (M_1). Then, they were ashed for 30 minute at 550^0C in furnace and were cooled in desiccators. Finally the mass of each crucible was weighed and recorded as (M_2). The content of crude fiber was calculated from the equation:

$$\text{Crude fiber (\% w/w)} = \frac{M2-M1}{W} \; x\; 100$$

Where

M_1 = Mass of the crucible, the sand and wet residue

M_2: = Mass of the crucible and the sand

W = Sample weight.

3.5.1.5. Determination of total ash content

Total ash content of the raw hyacinth bean and processed flour samples were determined according to AOAC (2000), using the official method 941.12.

For the determination of ash, clean empty crucible was placed in a Wegtech Britania drying oven (model DGH 9055A) dried at 105^0C for an hour, cooled in desiccators with (granular silca gel) and then weight of empty crucible was measured by sensitive analytical balance of model A214 and noted (W_1). About 2.5 gram of each of sample was taken in crucible (W_2). The sample was ignited over a hot plate of model B212 burner until it is charred completely, and the crucible was transported into Carbolite muffle furnace of a model CSF 1200 at 550^0C for 4 hours. The appearances of gray white ash indicate complete oxidation of all organic matter in the sample. After ashing the crucible was cooled in a desiccators and weighed (W_3). Percent ash was calculated by following formula:

$$\%\text{Ash(w/w)} = \frac{Differnce\ in\ Wt.Ash}{Wt.of\ a\ sample}\ x100$$

Where: Difference in Wt. ash = $W_3 - W_1$

3.5.1.6. Determination of carbohydrate content

The carbohydrate was calculated by difference with the inclusion of crude fiber. This is mathematically expressed as:

Utilizable Carbohydrate (% w/w) = 100 – (Crude fiber + Crude protein + Crude ash +

Crude fat + moisture)

3.5.1.7. Calcium, Zinc and Iron determination

Total mineral content of raw and processed hyacinth bean flour samples was determined according to Osborne and Voogt (1978) by using Buck Scientific Atomic Absorption Spectrophotometer (AAS) (model 201 VGP, Canada).

The ash was obtained from dry ashing method of powdered hyacinth bean flour. The ash was wetted completely with 6 mL of 6N HCl and dried on a low temperature hot plate. 15 ml of 3N HCl was again added to the dried ash and heated on hot plate until the solution boils. The ash was let to cool at room temperature in a hood and filtered into 100 mL graduated flask using 125 mm filter paper (Whattman 42). Again 10 ml of 3N HCl was added into each crucible dishes and heated until the solution just boils, cooled, and filtered into the flask. The crucible dishes were again washed three times with deionized water, the washing was filter into the graduated flask. 5 mL of 10% Lanthanum chloride solution was added into each graduated flask of the sample containing. Then the solution was cooled and diluted to 100 mL with deionized water. A blank which contains 25 mL of 3 N HCl and 5 mL of 10% $LaCl_3$ were added into 100 mL volumetric flask. Four series of working standard metal solutions were prepared by appropriate dilution of the metal stock solutions (nitrates of the metals) with deionized water containing 2.4 mL 3 N HCl in a 10 mL volumetric flask. Calibration graph (concentration Vs absorbance) for each element using the prepared standard solutions. The sample concentration was determined using Flame Atomic Absorption Spectrophotometer;' (Varian Spectra AA-20 Plus, Varian Australia, Pty, Ltd, Australia). A single mineral hollow cathode lamp was used for each element. Series of

working standard solutions for iron (0.00, 2.00, 6.00, 10.00) for zinc (0.00, 0.60, 1.00, 1.40, 1.80) for calcium (0.00, 1.00, 1.50, 2.50, 3.00) concentration of the standards (µg/mL) were prepared. Reading was taken from the graph, which illustrated the metal concentrations that correspond to the absorption values of the samples, and the blank. The metal contents were calculated by using the following formula:

$$\text{Metal content (mg/100g)} = \frac{(A-B)X\,V}{10W}$$

Where :

W = weight of the sample (g)

V = volume of the extract (mL)

A = Concentration (µg/mL) of the sample solution

B = Concentration (µg/mL) of the blank solution

3.5.1.8. Determination of phosphors

Phosphors was determined by the colorimetric method using ammonium molybdate . phosphorous was converted into phosphomolybdate, which was reduced to a blue molybdenum compound by aminonaphtholsulphonic acid to give a blue molybdenum compound. A sample solution was obtained from mineral analysis (iron, zinc and calcium). One mL of a clear extract was taken from the sample solution and diluted to 100 ml with deionized water in 100 mL of volumetric flask. A 5 mL of a duplicate sample dilution was added into test tubes. A 0.5 mL of molybdate and a 0.2 mL of aminonaphtholsulphonic acid was added into the test tube (sample solution) and mixed thoroughly step by step. A 0.20 mL of aminonaphtholsulphonic acid was added into the test tube repeatedly each time until the solution becomes clear. The solution was allowed to stand for 10 minutes. The absorbance (reading A) of the solution was measured at 660 nm against distilled water. Simultaneously with sample phosphorous, standard and blank analysis were carried out. Standard and blank were prepared as above but 5 mL of working standard and 5 mL of deionized water (reading B) in place the sample dilution was used respectively. Standard curve was made from absorbance versus concentration. Phosphorous was calculated using the following formula:

$$P\ (\text{mg}/100\text{g}) = \frac{(A-B)X\ 50\ X\ 100}{Slope\ X\ W\ X\ 10}$$

Where : A = Reading of the sample solution

B = Reading of the blank solution

W = Weight of fresh sample

3.5.2. Anti-nutritional factors:

3.5.2.1. Determination of phytate content

Phytate was determined by the method of Latta and Eskin (1980) and later modified by Vantraub and Lapteva (1988). About 0.03g of hyacinth bean samples were extracted with 10ml 0f 2.4% HCl in a mechanical shaker (Eberbach) for 1hour at an ambient temperature and centrifuged at 3000rpm for 30 minute. The clear supernatant was used for phytate estimation. A 2mL of Wade reagent (containing 0.03% solution of $FeCl_3.6H_2O$ and 0.3% of sulfosalicilic acid in water) was added to 3mL of the sample solution (supernatant) and the mixture was mixed on a Vortex for 5 seconds. The absorbance of the sample solutions were measured at 500 nm using UV- VIS spectrophotometer (Beckman DU-64- spectrophotometer, USA).

A series of standard solution were prepared containing 0, 4.5, 9, 18, 27 and 30 µg/ml of phytic acid (analytical grade sodium phytate) in 0.2N HCl. A 3mL of standard was added into 15ml of centrifuge tubes with 3mL of water which were used as a blank. A 1mL of the Wade reagent was added to each test tube and the solution was mixed on a Vortex mixer for 5 seconds. The mixtures were centrifuged for 10 minutes and the absorbances of the solutions (both the sample and standard) were measured at 500nm by using de ionized water as a blank. A standard curve was made from absorbance versus concentration and the slope and intercept were used for calculation. The phytate content was calculated by using the following formula:

$$\text{Phytic acid}\ (\text{mg}/100\text{g}) = \frac{\text{Absorbance}-\text{intercept x 10}}{\text{Slope x density x wt x 3}}$$

3.5.2.2. Determination of tannin content

The amount of condensed tannin was determined by the Vanillin assay of Burns (1971) as modified by Maxson and Rooney (1972). About 0.5g of hyacinth bean sample was weighed,

extracted with 10mL 1% HCl in methanol, at room temperature in mechanical shaker for 24 h. The mixture was centrifuge at 1000G for 10 minutes. 1mL supernatant was mixed with 5mL of vanillin-HCl reagent in another test tube. When the reaction was completed (after 20 minute), the absorbance was read at 500nm using spectrophotometer (BECKMAN, DU-64, USA). D-catechen was used as standard value of tannin in mg D-catechen per gram of sample. 40 mg D-catechen was dissolved in 100mL of 1%HCl in methanol and from this 0, 0.2, 0.4, 0.6, 0.8 and 1ml were taken in a test tube and the volume was adjusted to 1mL with 1% HCl in methanol. 5mL of vanillin-HCl reagent in each test tube was added. After 20 minutes the absorbance was read at 500 nm. The absorbance of the blank was subtracted from the absorbance of the corresponding vanillin-containing sample. A standard curve has been constructed (Absorbance vs. D-catechin) and the linear portion of the curve was extrapolated to produce the standard curve. Values of tannins were expressed in milligram of D-catechin equivalent per gram of sample by the following formula.

$$Tannin\ (100g) = \frac{Abso - Intercept}{Slope * density * wt.sample} * dilutionfactor$$

3.6. Determination of molar ratio of phytate/mineral

The mole of phytate and minerals was determined by dividing the weight of phytate and minerals with its atomic weight (phytate: 660 g/mol; Fe: 56 g/mol; Zn: 65 g/ mol; Ca: 40 g/mol). The molar ratio between phytate and mineral was obtained after dividing the mole of phytate with the mole of minerals (Norhaizan & Nor Faizadatul Ain, 2009). Phytate phosphorous was calculated by assuming phytate contains 28% phosphorus, i.e. [Phytate P = phytate * 0.28] and accordingly non phytate phosphorous = total phosphorous - phytate phosphorous.

3.7. Sensory evaluation

The products of hyacinth bean that were produced by boiling and autoclaving methods were evaluated for their acceptability by consumers (panels). A panel of 20 semi-trained panelists was selected from the students as well as teachers of the Food Science and Nutrition Graduate Programme of Addis Ababa University based on their availability, interest and their physical

conditions during the sensory evaluation period. An orientation was given for the panelists about the parameters to be tested in boiled and autoclaved products of hyacinth bean varieties. Sensory evaluation was done using 9 point hedonic scale for color, aroma, taste, texture and overall acceptability in such an order that value 1 was assigned to lower acceptability and value 9 to higher acceptability test.

The sensory evaluation was conducted at room-temperature and bottled drinking water was provided to clean their mouths in between the test of each product. Four batches of hyacinth bean products were prepared from two varieties of the bean and coded with three-digit randomized numbers. The order of presentation of the products to each panel was different in a random sequence after 20 t0 30 minute interval of the first product served. Then, the panelists were asked to state their judgment of the products' acceptability on the given attributes on the score sheet. Finally, panelists recorded their responses on an evaluation sheet designed to indicate the value of the sample of each product.

3.8. Design and statistical analysis

Factorial treatment arrangement as per CRD was used. The data was analyzed using GLM of SPSS (v. 15) by two way analysis of variance (two-ANOVA) and significant mean separation followed Dunkan multiple range test (DMRT). Results were expressed as the mean value $\pm$ standard error (S.E.) of triplicates except for the mineral contents, which were determined in duplicate. Significant differences was determined at the $P< 0.05$ level.

4. Result and Discussion

4.1. Production and utility of the Hyacinth bean

The preliminary study indicated that (questionnaire attached at appendix part) hyacinth bean is the staple food crop of the area. Invariably, respondents comprised of farmers, women processing the crop for various products and local community further confirmed that the crop is not only staple diet but also serve as fictional food.

Moreover, agriculturally the farmers are cultivating the hyacinth bean for two purposes. a) They cultivate when they want to supply a green manure for their soil. Murphy and Colucci (1999) reported that hyacinth bean is used as a green manure, by adding organic matter as well as nitrogen and minerals to the soil. The natural action of converting atmospheric nitrogen into forms available for the plant soil system improves productivity in an inexpensive, environmentally friendly manner. Therefore, it is used as natural fertilizer which enables small landholders to improve their soil and increase productivity. b) As staple food crop in the area. Morris (2009) suggested that hyacinth bean seeds and pods are popular foods in South Asia, India, China, West Africa, Japan, and the Caribbean Islands. The young hyacinth bean pods and seeds are cooked and eaten as vegetables.

They predominantly cultivate two varieties and they call them as " *misim aepa*" and " *end aepa*" respectively. They sow the seed starting from May- June like other crops in Ethiopia and needs high amount of rain fall for maximum production of the hyacinth bean seeds. According to the farmers the crop favors sandy and clay soil types with hot climatic condition for maximum production. But, with the absence of such weather conditions and soil types the production of the crop is not attractive. The full maturity of the seed will take five to six months. The farmers in the area commonly grow it by inter-cropping with other crops like cotton, maize, millet, sorghum, common beans (i.e, kidney red beans, pinto beans, kidney black beans).

Women process the hyacinth bean seed as "stew", " niffro" and cook with or without cereals. For the preparation of beans, the women were first roasted and decorticate to remove the hulls. The main advantage of roasting in this case is to make the hull easily removed during the dehusking. The women's processed the seed as a ' niffro" firstly by sorting moldy, foreign materials and others manually and then washed at least three times before cooking of the seed. Then after the

water of the boiled seed was drained and prepare other *watt* commonly used in the area which is called "*kimma watt*" by it. Because the women and the society were said that, the drained water from hyacinth bean had a special taste for" *kimma watt*". Therefore, the drained water was not discarded but they consider it as an ingredient for the preparation of the special watt in the community.

Currently, the predominant usage of the seed is not limited as a food but it has also other potential functions for different diseases and prevention of malnutrition. The societies believed that, consumption of the hyacinth bean prevent malaria, facilitates healing of wound, building of broken bones, feeding very emaciated infants and children (generally prevention of malnutrition) and feeding of lactating mothers for the strength of their bone and high production of milk for their infant. Similarly, Duke (2008) reported that, many amino acids, micronutrients, macronutrients and vitamins for human health have been identified in hyacinth bean seeds and pods.

4.2. Proximate and mineral composition of hyacinth bean

4.2.1. Proximate composition of hyacinth bean.

4.2.1. 1. Crude protein content

Effect of processing on the chemical composition of hyacinth bean varieties (i.e Rongai and Highworth) was presented in (Table 4.1). The mean total protein content of raw, soaked, boiled and autoclaved were found to be 20.69, 20.56, 17.60 and 20.87g/100g for Rongai and 22.78, 21.84, 20.38, and 20.82g/100g for Highworth varieties respectively. Soaking, boiling and autoclaving had no significant ($P>0.05$) effect on total protein mean values for both Rongai and Highworth varieties as compared to the raw statistically. However, there was a decreasing trend in the total protein content during soaking, autoclaving and boiling of both Rongai and Highworth varieties observed as compared to the raw. Such reduction of the mean crude protein contents might be attributed to leaching into water and denaturation of protein during cooking treatments and soaking. Observations in the present study were in agreement with those reported by Mubarak (2005) for cooked mung bean and Alajaji and El-Adawy (2006) for cooked chickpea respectively. Also, Khatoon and Prakash (2004) reported that microwave cooking and pressure cooking do not affect the nutrient composition of eight legumes. In a similar research conducted

on jack bean, there is no a significant (P>0.05) difference in crude protein content by different treatments such as cooking, autoclaving and soaking (Doss *et al., 2011).*

Controversially, the varieties were significantly (p<0.05) different in their mean total protein contents. Observations from present study revealed that, the mean protein value of Highworth variety had higher (22.78g/100g) as compared to Rongai (20.69g/100g) variety. In spite of their crude protein difference in both varieties, the results of these two varieties have comparable protein content with those reported by Kamatchi *et al.*(2010) for raw five different varieties of *Lablab purpureus (L.)* Sweet that is ranged from 20.46 – 25.47%. Similar researches conducted on certain legumes indicated also comparable value of protein. These includes chickpea (20.7%) (Bravo *et al.*, 1999), pigeonpea (22.01%) and peas (22.95%) respectively as reported by Masood and Rizwana (2010) and *Phaseolus vulgaris* (22.4%) and *cajanus cajan* (22.7%) (Apata and Ologhobo, 1994). Thus, higher level of protein content of seed materials of Hyacinth bean varieties has nutritional significance, since moderate intake of these seeds will greatly increase the total dietary protein intake of the consumers. Its utilization as a protein ingredient in the human food will reduce the over-dependence on the conventional protein supplements notably soybean and other common legumes.

In contrast, the mean crude protein content of raw hyacinth bean varieties were found to be lower when compared to an earlier reports on certain common legume grains such as Jack bean (29.8 g/kg) (Doss *et a.,2011)*, mucuna pruriens var.pruriens (24.9 g/kg) (Udedibie and Carlini, 1998) and gila bean (26.82 g/kg) (Vadivel *et al.*, 2008). In a similar fashion, these results were found to be lower in total protein when compared to *Dolichos Lablab bean (26.86%)* as reported by Osman (2007).

The present study showed that variety had a significant effect on mean protein content where as processing had no significant (p>0.05) effect on the crude protein contents of hyacinth bean. While the interactive effect of variety x process on protein content of hyacinth bean was not significantly (p>0.05) different.

4.2.1.2. Moisture content

The mean moisture content of raw and processed hyacinth bean varieties was shown in (Table 4.1). The mean moisture content of raw, soaked, boiled and autoclaved hyacinth bean varieties were 9.38, 9.60, 8.65 and 7.56g/100g for Rongai and 9.57, 9.33, 8.91 and 6.73g/100g for Highworth varieties respectively. The mean moisture content of soaked, boiled and autoclaved were significantly ($p<0.05$) affected as compared to raw (control) in both varieties of hyacinth bean. Boiling and autoclaving showed a decrease in the moisture content of both varieties; this might be the breaking down of the seed in cooking process which facilitates easily removal of moisture in oven drying at a temperature of 65^0C for 6 hours. Especially high reduction of moisture content was observed in autoclaving process in both varieties. These results show a close agreement with Doss *et al.*(2011) for jack bean. Incontrast, Mubark (2005) reported that soaking, autoclaving and boiling of mung bean increases the moisture content.

The moisture content of raw hyacinth bean was found to be 9.38g/100g and 9.57g/100g for Rongai and Highworth varieties respectively. There was a slight significant ($P<0.05$) difference on the moisture content of hyacinth bean varieties. In similar study conducted on nutrient and chemical evaluation of raw seeds of five varieties *lablab purpureus* by Kamatchi *et al.* (2010), moisture content of the bean was in the range of 7.25g/100g to 8.45g/100g. Other earlier researchers reported comparable results for some legumes which include cow pea (9.20g/100g), pigeon pea (8.45g/100g) respectively and jack bean (8.41g/100g) respectively (Olalekan and Bosede, 2010; Doss *et al.*, 2011). And also, both varieties had almost similar moisture content compared with certain mung bean (8.30%) and peas (9.05%) (Masood and Rizwana, 2010). In contrast, the moisture content of raw Rongai and Highworth varieties were found to be lower when compared to pigeonpea (11.07%) and cow pea (10.39%), respectively as reported by (Masood and Rizwana, 2010). Furthermore, the interactive effect of variety x processing significantly affects the moisture content of the hyacinth bean.

4.2.1.3. Crude fat content

The mean crude fat content of raw, soaked, boiled and autoclaved hyacinth bean seeds were presented in (Table 4.1). Thus, their mean crude fat contents were 1.17, 1.06, 0.66, and 0.93g/100g for Rongai and 1.16, 1.03, 0.49 and 0.88g/100g for Highworth varieties respectively. Raw, soaking, boiling and autoclaving showed a significant ($p<0.05$) difference on the mean crude fat contents of both Rongai and Highworth varieties as compared to the raw. In fact all the treatments (soaking, boiling and autoclaving) resulted in lossing crude fat in the hyacinth bean varieties. This reduction during soaking, boiling and autoclaving treatments might be attributed to their diffusion into the cooking water, while high reduction of crude fat was observed after boiling treatment (i.e, 0.66g/100g and 0.49g/100g) in (Table 4.1) in both Rongai and Highworth

Table 4.1. Effect of processing on proximate composition of two varieties of hyacinth bean*(Lablab purpureus L.) sweet in g/100g*

Variety	Process	Moisture	Protein	Fat	Ash	Fiber	CHO
	Raw	9.38 ± 0.03^{b}	20.69 ± 1.31^{a}	1.17 ± 0.09^{a}	3.53 ± 0.05^{a}	7.95 ± 0.06^{c}	57.28 ± 1.3^{a}
Rongai	Boiled	8.65 ± 0.04^{c}	17.60 ± 0.70^{a}	0.66 ± 0.07^{c}	2.79 ± 0.04^{c}	8.94 ± 0.08^{b}	61.36 ± 0.81^{a}
	Autoclave	7.56 ± 0.02^{d}	20.87 ± 1.38^{a}	0.93 ± 0.05^{b}	2.86 ± 0.01^{bc}	9.90 ± 0.21^{a}	57.54 ± 1.61^{a}
	Soaked	9.60 ± 0.05^{a}	20.56 ± 1.02^{a}	1.06 ± 0.01^{ab}	3.04 ± 0.12^{b}	8.41 ± 0.23^{c}	57.33 ± 1.25^{a}
	Raw	9.57 ± 0.04^{w}	22.78 ± 1.17^{x}	1.16 ± 0.03^{w}	3.59 ± 0.01^{w}	8.69 ± 0.23^{y}	54.21 ± 0.97^{z}
Highworth	Boiled	8.91 ± 0.04^{y}	20.38 ± 0.46^{x}	0.49 ± 0.04^{z}	2.80 ± 0.01^{z}	9.65 ± 0.20^{x}	57.76 ± 0.23^{xy}
	Autoclave	6.73 ± 0.05^{z}	20.82 ± 0.27^{x}	0.88 ± 0.02^{y}	2.88 ± 0.03^{y}	9.41 ± 0.13^{x}	59.28 ± 0.34^{x}
	Soaked	9.33 ± 0.04^{x}	21.84 ± 0.75^{x}	1.03 ± 0.03^{x}	3.33 ± 0.03^{x}	8.44 ± 0.13^{y}	56.03 ± 0.80^{yz}
Variety* processing		Sig	Sig	NS	Sig	NS	Sig

Values in the column per variety followed by different superscripts are significantly different ($p<0.05$). Each value is a mean of three replicates ± SE

varieties, in agreement with the results of Osman(2007) who suggested that the reduction could be attributed to high lipolytic enzyme activity which break down the triglyceride to simple fatty acids sterol esters and polar lipids, especially with presoaked and cooked *dolichos lablab bean.* The crude fat content was not easily affected after soaking treatment in both varieties of the bean. Observations in the present study were also in agreement with those reported by (Alajaji and El-Adawy, 2006) for chickpea seeds.

There was no significant ($p<0.05$) difference between Rongai and Highworth varieties in their mean fat contents, however, Rogai was slightly higher (1.17g/100g) than highworth (1.16 g/100g) variety. These results were found to be lower than mung bean (1.85g/100g), *dolichos lablab bean* (1.90g/100g), sword bean (2.8-3.8g/100g), jack bean (4.20g/100g) and chickpea (6.48g/100g) respectively (Mubarak, 2005; Pugalenthi and Vadivel, 2005; Alajaji and El-Adawy, 2006; Osman, 2007; Doss *et al.*, 2011). Moreover, the interactive effect of variety x processing had no significant ($p<0.05$) effect on the crude fat content in the bean.

4.2.1.4. Total ash content

The mean total ash content of raw, soaked, boiled and autoclaved hyacinth bean varieties were shown in (Table 4.1). The mean total ash content of raw, soaked, boiled and autoclaved hyacinth bean varieties were 3.53, 3.04, 2.79 and 2.86g/100g for Rongai and 3.59, 3.33, 2.80 and 2.88g/100g for Highworth varieties respectively. Soaking, boiling and autoclaving treatments significantly ($p<0.05$) reduced the total ash content of both hyacinth bean varieties as compared to the raw. The cooking treatments such as boiling and autoclaving resulted in a significant reduction of the total ash content in both varieties of the hyacinth bean. This decrease in ash content might be attributed to diffusion of certain minerals into the soaking or cooking water (Mubark, 2005; Alajaji and El-Adawy, 2006). The observations in the present study were in agreement with those findings of Akaerue and Onwuka (2010) which reported that boiling results in reduction in ash content on mung bean. In a similar manner, earlier studies reported that there was a reduction of mineral contents on legumes such as chickpea, mung bean and jack bean due to boiling and autoclaving treatments (Mubarak, 2005; Alajaji and El-Adawy, 2006 ; Doss *et al.*, 2011). However, the ash content of the *dolichos lablab bean* had increased during autoclaving as reported by Osman (2007). Soaking treatment significantly decreased the total ash content of the bean but when compared to other treatments, the lowest reduction in ash content

was observed by it. This is because, there was a combined effect employed in boiled and autoclaved hyacinth bean varieties. Therefore, the combined effects make the more leaching out of the minerals in hyacinth bean in particular and pulses in general, which was confirmed by (Mubarak, 2005; Alajaji and El-Adawy, 2006; Osman, 2007).

The raw hyacinth bean varieties (Rongai and Highworth) were significantly ($P<0.05$) different in their mean ash contents. From the present study, the mean ash content of Rongai variety was significantly lower (3.53g/100g) than that of Highworth (3.59g/100g) variety. Hence Highworth seeds might contribute high concentration of mineral elements, which have an advantage to speed up the metabolic processes and improve growth and development. The ash content in the present study were found to be lower as compared to other legumes such as mung bean (3.76g/100g), chickpea (3.72g/100g), *Dolichos lablab bean* (3.96g/100g), jack bean (4.48g/100g), five varieties of *Lablab purpureus (L.) sweet* (3.97-4.48g/100g), and locust bean (4.24g/100g) respectively (Mubarak, 2005; Alajaji and El-Adawy, 2006; Osman, 2007; Kamatchi *et al.*, 2010; Doss *et al.*, 2011; Gloria *et al.*, 2011). Based on the investigation, both variety and processing had a significant ($p<0.05$) effect on ash content of hyacinth bean, confirming the results reported by Wang *et al.*(2008) and also, the interactive effect of variety x processing on ash content was significant in hyacinth bean *(Lablab purpureus L.) sweet.*

4.2.1.5. Carbohydrate content

From Table 4.1 the mean carbohydrate content of raw, soaked, boiled and autoclaved hyacinth bean varieties were found to be 57.28, 57.33, 61.36 and57.54g/100g for Rongai and 54.21, 56.03, 57.76 and59.28g/100g for Highworth varieties respectively. Soaking, boiling and autoclaving treatments significantly increased the mean carbohydrate content of Highworth variety as compared to the raw. Even though the processing had no a significant ($p>0.05$) effect on the mean carbohydrate content of Rongai variety, it showed a trend of increasing the mean carbohydrate contents. Thus, from the present study results, it is possible to say cooking methods (boiling and autoclaving) and soaking were very effective to increase the mean carbohydrate content of the hyacinth bean varieties in particular and legumes in general. These observation had an agreement with those reported by Mubark (2005) mung bean, Alajaji and El- Adawy (2006) on chickpea, Osman (2007) on *dolichos lablab bean* and Doss *et al.*(2011) on jack bean.

Variety was significantly ($p<0.05$) different in their carbohydrate content. From this study result, Rongai had higher (57.28g/100g) mean carbohydrate content than Highworth (54.21g/100g) variety. Thus, the present study findings are consistent with studies on certain grain legumes such as *dolichos lablab bean* (67.23%), mungbean (62.03%), jack bean (57.83%), pigeon pea (56.63%), cow pea (56.60%) and jack bean (50.80%) respectively (Mubark, 2005; Osman, 2007; Olalekan and Bosede, 2010; Doss *et al.*, 2011). Cooking method (boiling and autoclaving) and variety were significant on carbohydrate content of hyacinth bean and also the interactive effect of variety x processing was significant ($p<0.05$) on the carbohydrate content of the hyacinth bean (*Lablab purpureus L.*) sweet.

4.2.1.6. Crude fiber content

Effect of soaking, boiling and autoclaving on the level of crude fiber content in two varieties of hyacinth bean was shown in (Table 4.1). From the table, the mean crude fiber content of raw, soaked, boiled and autoclaved hyacinth bean varieties were 7.95, 8.41, 8.94, 9.90mg/100g for Rongai and 8.69, 8.44, 9.65 and 9.41g/100g for Highworth varieties respectively. According to the result obtained in the study, hyacinth bean (*lablab purpureus* L.) provides an adequate amount of dietary fiber which is essential in human nutrition. Raatz(2012) reported fiber in beans helps to lower blood levels of low density lipoprotein(LDL) cholesterol, especially if LDL cholesterol levels were high to begin with, without compromising the level of protective high density lipoprotein (HDL) cholesterol.

Cooking methods (boiling and autoclaving) significantly ($p<0.05$) affected the mean crude fiber content of the hyacinth bean varieties while soaking was not significant ($p>0.05$) as compared to raw. Thus, boiling and autoclaving highly improves by increasing the mean crude fiber content in both varieties whereas soaking slightly increases the fiber content. This is due to heat treatment (cooking) may modify the structure of both cell wall and storage polysaccharides of pulses possibly by affecting the intactness of tissue histology and disrupting the protein–carbohydrate integration, thus reducing the solubility of dietary fiber (Siljestrom *et al.*, 1986). Similarly, Wang *et al.*(2008) suggested that, this increase might be due to protein–fiber complexes formed after possible chemical modification induced by the cooking of dry seeds. As revealed from the present study result, cooking and soaking treatments resulted in increasing the mean fiber contents in the bean. These observation had an agreement with those reported by

(Mubark, 2005) for mung bean and (Alajaji and El-Adawy, 2006) for chickpea. However, Doss *et al.*(2011) reported that, there is a little decrease in crude fiber content of jack bean after cooking (i.e, boiling and autoclaving) treatments. Even though fiber content decreased, the report illustrated that; when considering the effect of various common processing methods on the fiber content of jack bean, boiling and autoclaving treatment have not exhibited any significant reduction of crude fiber in jack bean seeds.

As per the study results indicated, the mean crude fiber content of two varieties of raw hyacinth bean (*Lablab purpureus L.*) sweet were not significantly ($p>0.05$) different statistically, but the mean crude fiber content of Highworth variety was higher (8.69g/100g) when compared to Rongai (7.95g/100g) variety. The present study results were found to be higher when compared to an earlier studies on common legume grains such as cowpea (0.97%), jack bean (1.07%), pigeon pea (1.10%), chickpea (3.82%), mung bean (4.63%), *Lablab purpureus* (L.) sweet (4.98-6.90%) and jack bean (7.37%) respectively(Mubark, 2005; Alajaji and El-Adawy, 2006; Olalekan and Bosede, 2010; Kamatchi *et al.*, 2010; Doss *et al.*, 2011). And also, these study result on crude fiber content has a consistency with that of earlier reports on *Canavalia gladiata* (9.32%), *C. virosa* (10.47%) and *Mucuna monosperma* (8.9-9.2%) consequently (Siddhuraju and Becker, 2001; Pugalenthi *et al.*, 2003).

Variety and processing had a significant ($p< 0.05$) effect on crude fiber content, however, the interactive effect of variety x processing (soaking, boiling and autoclaving) on crude fiber content was not significant ($p>0.05$) in hyacinth bean *(Lablab purpureus L.) sweet*. These observations agree with findings of Wang *et al.*(2008) who reported that variety x cooking had a significant effect on total dietary fiber level of field pea.

4.2.2. Mineral composition of hyacinth bean

Minerals are involved in activation of intracellular and extracellular enzymes, in regulation of critical pH levels in body fluids necessary for the control of metabolic reactions and in osmotic balance between the cell and its environment. A deficiency of any one of the essential minerals can result in severe metabolic disorders and compromise the health of the organism. Some minerals deficiencies are common in developing countries, but mineral sub deficiencies may also occur in developed countries (Lopez *et al.*, 2002). Thus, this study was highly emphasized on the

major minerals which are required nutritionally and their deficiency in the diet brings about severe health problems. These include calcium, iron, zinc and phosphorus.

Table 4.2. Effect of processing on mineral composition of two varieties of hyacinth bean*(Lablab purpureus L.) sweet in mg/100g*

Variety	Process	Ca	Fe	Zn	P
	Raw	125.95±1.01^{c}	20.52±0.05^{a}	2.85±0.09^{a}	872.78±8.12^{a}
Rongai	Boiled	141.03±5.35^{c}	19.37±1.08^{a}	2.31±0.05^{b}	891.21±66.24^{a}
	Autoclaved	149.27±4.64^{b}	21.79±0.71^{a}	1.95±0.10^{d}	887.17±24.97^{a}
	Soaked	149.71±3.41^{a}	20.86±0.52^{a}	2.04±0.25^{c}	828.47±45.13^{a}
	Raw	148.05±0.96^{x}	20.66±0.94^{x}	2.34±0.03^{x}	813.41±39.12^{y}
Highworth	Boiled	149.28±3.21^{x}	22.28±0.88^{x}	2.69±0.09^{x}	982.40±24.76^{x}
	Autoclaved	148.45±0.10^{x}	22.17±0.05^{x}	2.67±0.22^{x}	969.28±12.09^{x}
	Soaked	146.72±2.10^{x}	21.42±0.44^{x}	2.15±0.05^{x}	774.20±0.37^{y}
Variety*process		Sig	NS	Sig	NS

Values in the column per variety followed by different superscripts are significantly different ($p<0.05$). Each value is a mean of duplicates ± SE

4.2.2.1. Calcium content

Calcium is an essential nutrient that plays a vital role in neuromuscular function, many enzyme-mediated processes and blood clotting, as well as providing rigidity to the skeleton by virtue of its phosphate salts. Its non-structural roles require the strict maintenance of ionized calcium concentration in tissue fluids at the expense of the skeleton if necessary and it is therefore the skeleton which is at risk if the supply of calcium falls short of the requirement (WHO/FAO, 2004). The mean calcium content of raw, soaked, boiled and autoclaved hyacinth bean varieties were 125.95, 149.71, 141.03 and 149.27mg/100g for Rongai and 148.05, 146.72, 149.28 and 148.45mg/100g for Highworth varieties respectively as shown in (Table 4.2). The findings

indicated that cooking treatment significantly ($P<0.05$) increased the mean calcium content of Rongai variety. Even though cooking treatments and soaking have no significant ($P>0.05$) effect on the mean calcium content of Highworth variety, the trend showed an increment on the calcium content. Thus, cooking method (boiling and autoclaving) and soaking improves the calcium quality of hyacinth bean (*Lablab purpureus L.*) sweet by increasing its content against the raw, confirming the result reported by Wang *et al.*(2008). This increment in the calcium content might be attributed by the reduction of anti-nutritional factors after the cooking treatment which binds divalent minerals particularly phytic acid. The maximum increment in mean calcium content was observed after autoclaving (149.27mg/100g) and soaking (149.71mg/100g) treatments of Rongai variety respectively. Mubark (2005) reported that soaking, germination, boiling, autoclaving and microwave cooking have no significant ($p>0.05$) effect on the calcium content of mung bean. In contrast (Mubark, 2005; Alajaji and El-Adawy, 2006) indicated that, cooking methods (boiling, autoclaving and microwave cooking) caused significant reduction of calcium content of chickpea after the treatment against the raw.

The mean calcium content between raw hyacinth bean varieties were significantly ($P<0.05$) different. Highworth variety contains higher (148.05mg/100g) mean calcium content than Rongai (125.95mg/100g) variety. The present study were in agreement with those reported by Wang *et al.*(2008) on six varieties of field pea. This study result of raw hyacinth bean calcium content was found to be higher when compared to other previous reports on certain legumes such as mung bean (84.00mg/100g), jack bean (0.18mg/100g), pigeon pea (0.65mg/100g), cow pea (0.44mg/100g) and *lablab purpureus* (115.3mg/100g) respectively (Mubark, 2005; Olalekan and Bosede, 2010; Ragab *et al.*, 2010) but lower than *vigna unguiculata* (1537mg/100g), chick pea (176mg/100g), mung bean(216mg/100g), five varieties of *lablab purpureus* (364.67-575.03mg/100g) and African locust bean (222.2mg/100g) respectively (Thangadurai, 2005; Aljaji and El-Adawy, 2006; Habibullah *et al.*, 2007; Kamatchi *et al.*, 2010; Gloria *et al.*(2011). Moreover, the interactive effect of variety x processing had significant ($p<0.05$) influence on the calcium content of hyacinth bean. According to Wang *et al.*(2008) variety x processing has an influence on the calcium content in pulses in general.

4.2.2.2. Iron content

The effect of processing on raw and cooked hyacinth bean varieties was presented in (Table 4.2). The mean iron content of raw, soaked, boiled and autoclaved hyacinth bean varieties was 20.52, 20.86, 19.37 and 21.79mg/100g for Rongai and 20.66, 21.42, 22.28 and 22.17mg/100g for Highworth varieties respectively. Cooking method (boiling and autoclaving) and soaking had no significant($p>0.05$) effect on the iron content of hyacinth bean varieties against the raw but there was an increment in the iron content in all treatments except boiling in Rongai variety. This finding was consistent with other research studies on legumes such as chickpea and mung bean (Mubark, 2005; Alajaji and El-Adawy, 2006) for boiling and autoclaving treatments. Mubark (2005) and Alajaji and El-Adawy (2006) suggested that processing was not significant on iron content but it decreases iron content due to leaching into boiling water.

Food legumes in general and beans in particular contain appreciable quantities of iron and other minerals (Beebe *et al.*, 2000). Based on the present investigation, the hyacinth bean contained an adequate amount of dietary iron in both varieties. Statistically, variety had no significance influence on iron content of hyacinth bean but Highworth variety contained slightly higher (20.66mg/100g) value than rongai (20.52mg/100g) variety. The present study finding was a close agreement with the report of Wang *et al.* (2008). The iron content of raw hyacinth bean is greater when compared with other legumes including *cajanus cajan, lablab purpureus and vigna unguiculata as 10.53, 11.41 and 13.31mg/100g* by Ragab *et al.*(2010) respectively. Similar research results were reported by Alajaji and El-Adawy (2006) as 7.72mg/100g for chickpea, Mubark (2005) as 9.70mg/100g for mung bean and Kamatchi *et al.*(2010) in the range of 6 55-10.33mg/100g for five varieties of *Lablab purpureus(L.)*. Furthermore, the interactive effect of variety x processing was not significantly different, confirmed with Wang *et al.* (2008)

4.2.2.3. Zinc content

From Table 4.1 the mean zinc content of raw, soaked, boiled and autoclaved hyacinth bean was 2.85, 2.04, 2.31 and 1.95mg/100g for Rongai and 2.34, 2.15, 2.69 and 2.67mg/100g for Highworth varieties respectively. Cooking treatments (boiling and autoclaving) and soaking significantly ($p<0.05$) affected the mean zinc content of Rongai while they did not exhibit any significant effect on Highworth variety. Generally, treatment had a significant ($p<0.05$) influence on the zinc content of hyacinth bean. Autoclaving, boiling and soaking decreased zinc content in

Rongai variety which might be attributed to splitting of the seed during boiling and autoclaving and made the zinc easily leached into water. This observation matches with those reported by Alajaji and El-Adawy (2006). In contrast, the mean zinc content of Highworth variety showed an increment during boiling and autoclaving treatments.

Zinc content of beans is one of the highest among vegetable sources, and is nearly equal to dairy products but is far inferior to meats (Beebe *et al.*, 2000). The present study indicated that hyacinth bean has an appreciable amount of zinc. Statistically, the mean zinc content of raw hyacinth bean varieties was not significantly ($P>0.05$) different but Rongai variety has higher (2.85mg/100g) zinc content than Highworth(2.34mg/100g) variety. The absorption and the amount of zinc in a bean may be affected by the chemical form of zinc in the soil, the nature of the environment and other components such as anti-nutritional factors and dietary fiber found in the bean (Beebe *et al.*, 2000). However, the zinc content of hyacinth bean was found to be lower as compared to other legumes such as chickpea (4.32mg/100g), *cajanus cajan* (3.64mg/100g), *lablab purpureus* (3.36mg/100g), *vigna unguiculata* (4.31mg/100g), and African locust bean(3.8mg/100g) respectively (Alajaji and El-Adawy, 2006; Ragab *et al.*, 2010; Gloria *et al.*, 2011) but higher than jack bean, pigeon pea and cow pea (1.58, 1.54 and 1.62mg/100g) Olalekan and Bosede (2010) respectively. Moreover, Kamatchi *et al.* (2010) reported that five varieties of *lablab purpureus (L.)* sweet has similar mean values (i.e, 2.69-2.73mg/100g) as hyacinth bean varieties under the investigation. Finally, the interactive effect of variety x processing was significant on zinc content of the hyacinth bean.

4.2.2.4. Phosphorus content

The mean phosphorus content of the raw, soaked, boiled and autoclaved hyacinth bean varieties were 872.78. 828.47, 891.21 and 887.17mg/100g for Rongai variety and 813.41, 774.20, 982.40 and 969.28mg/100g for Highworth variety respectively as shown in (Table 4.2). As the study results indicated, the soaking treatment caused significant ($p<0.05$) effect on Highworth variety of phosphorus as compared to the raw. Generally, cooking (boiling and autoclaving) treatments would show an increment of phosphorus content from (872.78 to 891.21) of Rongai and (813.41 to 982.40mg/100g) of Highworth variety consequently while the soaking treatment decreased the content from (872.78 to 828.47g/100g) for Rongai variety and (813.41 to 774.20g/100g) for Highworth varieties as compared to the raw. This study result is consistent with Wang *et*

al.(2008) who reported that cooking treatment increases the phosphorus content in pea. In contrast, Mubark(2005) and Alajaji and El-Aldawy(2006) reported that cooking treatment(boiling and autoclaving) and soaking decreases the phosphorus content in mung bean and chickpea subsequently.

In the present study there was a significant ($p<0.05$) difference in the mean phosphorus content between raw hyacinth bean varieties. Thus, Rongai variety had higher (872.78mg/100g) mean value of phosphorus than Highworth (813.41mg/100g) variety. The mean phosphorus contents in the present study was higher when compared with other legumes reported by Kamatchi *et al.*(2010) for five varieties of *Labalab purpureus* (500.27-733.00mg/100g) and (Mubark, 2005; Alajaji and El-Adawy, 2006) for mung bean and chickpea which was 391mg/100g and 222mg/100g respectively. Moreover, the interactive effect showed that there was a significant ($p<0.05$) effect on variety x processing on the level of phosphorus which is disagreed with those reported by Wang *et al.*(2008) on six varieties of field pea.

4.3. Antinutritional factors in hyacinth bean

4.3.1. Phytate content

Pulses are important sources of protein in the diets of millions of people in the world. However, their contribution to the nutrition of the consumer is limited, due to poor digestibility and ANFs (Ramakrishna *et al.*, 2006). The influence of soaking and cooking (boiling and autoclaving) on the level of phytate content present in the seeds of hyacinth bean (*Lablab purperus L.*) sweet was shown in (Table 4.3). The mean phytate content of raw, soaked, boiled and autoclaved hyacinth bean varieties were found to be 331.63, 306.30, 299.28 and 293.63mg/100g for Rongai and 335.08, 281.22, 274.65 and 299.48mg/100g for Highworth variety respectively. The commonest process of preparing pulses for consumption at the household level is to cook them by boiling in water (Ramakrishna *et al.*, 2006). Cooking is usually done before the use of legumes in a human diet. This improves the protein quality by either destruction or inactivation of heat-labile ANFs (Mubarak, 2005). The effect of different cooking methods (*i.e.* boiling and autoclaving) and soaking on the level of phytate content of hyacinth bean indicated a significant ($p<0.05$) reduction as compared to the raw hyacinth bean (*Lablab purperus L.*) sweet. The decrease of phytate content by soaking and cooking treatments (i.e, boiling and autoclaving) of the hyacinth bean may be due to leaching out of this compound into water (Doss *et al.*, 2011). Boiling of

hyacinth bean seeds brought about a significant decrease in phytic acid content with a percentage loss of the mean phytate contents by 9.75% and 18.03% for Rongai and Highworth varieties Consequently, whereas autoclaving shows the reduction of 11.56% and 10.62% of phytate level in both Rongai and Highworth varieties as compared to the raw. Therefore, it possible to say that boiling and autoclaving treatments are very effective to reduce the phytate contents of the bean. Soaking resulted in 7.64 and 16.07% loss of phytate in both Rongai and Highworth variety. In the present study the phytate content was significantly reduced by soaking and boiling and these treatments were twice effective in Highworth as compared to Rongai variety. These observations were in agreement with results of Mohamed *et al.*(2011) for cooked soybean, mung bean and kidney bean with the percentage reduction of 7.7, 15.4 and 16.5% subsequently. Similarly, Wang *et al.* (2008) reported that cooking caused a significant reduction (5.3–10.8%) of phytate contents in six field pea varieties. However, Osman (2007) reported a maximum reduction of phytate in *dolichos lablab bean* is that (44.85%) and (52.29%) in both cooking and autoclaving treatments respectively.

The mean phytate content of raw hyacinth bean varieties were not significantly ($p>0.05$) different statistically but the Rongai variety contains smaller (331.63mg/100g) mean phytate value than Highworth (335.08mg/100g) variety as indicated in (Table 4.3). The raw mean value of phytate is higher than Indian bean (82mg/100g) and chickpea (121mg/100g) respectively (Ramakrishna *et al.*, 2006; Alajaji and El-Adawy, 2006) but lower than those reported by (Osman, 2007) for *dolichos lablab bean*(605.39mg/100g). Moreover, these finding has similar mean value as compared to Kamatchi *et al.*(2010) with the range of 314mg/100g to 421mg/100g of five varieties of *lablab purpureus*. The high level of phytate is of nutritional significance as not only in the phytate phosphorus unavailable to humans, but it also lowers the availability of many other essential minerals (Wang *et al.*, 2008). Based on the the present study findings, processing significantly affected the mean phytate content whereas the variety was not significant on it. However, the interactive of variety x treatment was significantly ($p<0.05$) different on the phytate content of hyacinth bean. This study result was consistent with those reported by Wang *et al.*(2008) on the phytate content of six pea varieties.

Table 4.3. Effect of processing on phytate and tannin content of two varieties of hyacinth bean *(Lablab purpureus L.) sweet.*

Variety	Process	Phytate ($mg100g^{-1}$)	% Reduction	Tannin ($mg100g^{-1}$)	% Reduction
	Raw	331.63 ± 3.34^{a}	0.00	0.27 ± 0.03^{a}	0.00
	Boiled	299.28 ± 5.38^{b}	9.75	0.18 ± 0.04^{ab}	33.33
Rongai	Autoclaved	293.63 ± 4.30^{b}	11.56	0.12 ± 0.04^{b}	55.55
	Soaked	306.30 ± 6.52^{b}	7.64	0.24 ± 0.02^{a}	11.11
	Raw	335.08 ± 5.25^{x}	0.00	0.51 ± 0.02^{x}	0.00
Highworth	Boiled	274.65 ± 3.30^{z}	18.03	0.33 ± 0.01^{z}	35.29
	Autoclaved	299.48 ± 3.76^{y}	10.62	0.32 ± 0.02^{z}	37.25
	Soaked	281.22 ± 7.52^{z}	16.07	0.44 ± 0.03^{y}	13.73
Variety*process		Sig		NS	

Values in the column per variety followed by different superscripts are significantly different ($p< 0.05$). Each value is a mean of three replicates ± SE

4.3.2. Tannin content

Tannins are widely cited as an important anti-nutrient that precipitates iron in food preparation or in the gut (Beebe *et al.*, 2000). Effect of processing on the mean tannin content of raw and processed hyacinth bean varieties were presented in (Table 4.3). The mean tannin content of raw, soaked, boiled and autoclaved hyacinth bean varieties were found to be 0.27, 0.24, 0.18 and 0.12mg/100g for Rongai and 0.51, 0.44, 0.33 and 0.32mg/100g for Highworth variety consequently. Cooking method (boiling and autoclaving) and soaking treatments on the mean tannin content of hyacinth bean varieties caused a significant ($p<0.05$) reduction as compared to the raw. This reduction in tannin during soaking, boiling and autoclaving treatments might be attributed to the leaching of polyphenols in soaking and boiling water and the loss of compounds while treating at high temperature (Nithya *et al.*, 2006). From the study results, autoclaving

treatment greatly reduced tannin content (55.55%) of Rongai and (37.25%) for Highworth varieties followed by boiling (33.33%) for Rongai and (35.29%) of Highworth varieties respectively. The present study results were confirmed with those earlier reports which reduced the tannin content by boiling and autoclaving on some grain legumes such as chichpea (48.04% and 50.10%) (Alajaji and El-Adawy, 2006), Indian bean (76.47%) (Ramakrishna *et al.*, 2006), mung bean (45.5% and 51.5%) (Mubark, 2005) and jack bean (64% and 83%) (Doss *et al.*, 2011). In contrast, other studies indicated that the tannin content of *dolichos lablab bean* increased after cooking treatments as reported by Osman (2007). Therefore, both boiling and autoclaving treatments are very effective in the reduction of the tannin contents in the hyacinth bean in order to enhance the bioavailability of iron and protein in human body. In other hand, soaking treatment showed the lowest reduction (11.11%) for Rongai and (13.73%) for Highworth varieties respectively. Controversially, a maximum percentage loss during soaking treatment was reported in mung bean and jack bean (39.4% and 45.0%) respectively (Mubark, 2005; Doss *et al.*, 2011); while Osman(2007) reported that soaking treatment increased the tannin content in *dolichos lablab bean.*

The mean tannin content of raw hyacinth bean varieties was significantly ($p<0.05$) different to each other. Thus, Highworth variety indicated higher (0.51mg/100g) mean value of tannin than Rongai (0.27mg/100g) variety. This difference in tannin content is may be attributed by the seed color difference (Beebe *et al.*, 2000; Sandberg, 2002) who reported that the black beans consists higher tannin than others. The mean tannin content of hyacinth bean was lower than chickpea (4.85mg/g), mung bean (3.30mg/g), Indian bean (0.85mg/g) and jack bean (82.5mg/100g) respectively (Mubark, 2005; Alajaji and El- Adawy, 2006; Ramakrishna *et al.*, 2006; Doss *et al.*2011) and similar *dolichos lablab bean (0.*42%) and five varieties of *lablab purpureus L sweet* (0.35- 0.66%) (Osman, 2007; Kamatchi *et al.*, 2010) consequently.

4.4. Phytate and non-phytate phosphorus contents

The phytate phosphorus and non-phytate phosphorus content of the raw, soaked and cooked (boiled and autoclaved) hyacinth bean varieties were presented in (Table 4.4). The mean phytate phosphorus content of raw, soaked, boiled and autoclaved hyacinth bean varieties was 93.20, 86.49, 84.02 and 82.55mg/100g for Rongai and 92.69, 79.18, 77.31 and 84.14mg/100g for Highworth variety respectively while the mean non phytate phosphorus content was 779.58,

741.98, 807.19 and 804.63mg/100g for Rongai and 720.72, 695.02, 905.10 and 885.14mg/100g for Highworth variety respectively. From the result obtained in these experiment, cooking treatments (boiling and autoclaving) significantly ($p<0.05$) affected the phytate phosphorus of hyacinth bean varieties when compared to the raw. However, soaking treatment was not significant on the phytate phosphorus content of both hyacinth bean varieties. The non phytate phosphorus content significantly ($p<0.05$) increased after boiling and autoclaving treatments for Highworth variety when compared to raw whereas it was not significant in Rongai variety in

Table 4.4. Effect of processing on Phytate and non phytate phosphorus of hyacinth bean varieties.

Variety	Process	Phytate P[a] mg/100g	Non phytate P[b] mg/100g	Proportion of phytate P (%)
	Raw	93.20±1.51[a]	779.58±1.63a	10.68
Rongai	Boiled	84.02±2.58[ab]	807.19±1.44a	9.50
	Autoclaved	82.55±2.00[b]	804.63±1.76a	9.32
	Soaked	86.49±2.90[ab]	741.98±3.57a	10.45
	Raw	92.69±9.63[x]	720.72±37.49[y]	11.41
Highworth	Boiled	77.31±68.82[y]	905.10±26.20[x]	7.88
	Autoclaved	84.14±30.98[xy]	885.14±10.34[x]	8.68
	Soaked	79.18±42.23[y]	695.02±3.94[y]	10.23
Variety*process		NS	NS	

a= phytate phosphorous calculated by assuming phytate contains 28.18 % phosphorus (phytate * 0.28)

b= non phytate phosphorous = total phosphorous - phytate phosphorous

all treatments employed on it. The interactive effect of variety x treatment had no any significant ($p<0.05$) effect on the phytate phosphorus and non phytate phosphorus contents of the bean. Phytate had been considered an ANFs because of its ability of chelate minerals and impede their absorption and because of the limited capacity of monogastric species to hydrolyze and utilize phosphorus from this molecule. Moreover, the phosphorus content of phytate has been considered to be unavailable to the organism (Ferna´ndez *et al.*, 1997). Generally diets are regarded as being adequate in bioavailable phosphate. However, the high proportion of phosphate as phytate has consequences for bioavailability of minerals and trace elements (Umeta *et al.*, 2005). In this study, the raw and processed hyacinth bean varieties have low proportion of phosphate as phytate and hence, phosphorus, iron, zinc and calcium from these hyacinth bean varieties could be bioavailable for human body.

4.5. Phytate to mineral molar ratio of hyacinth bean

The molar ratios for calcium, zinc, iron and phytate were calculated to evaluate the effects of elevated levels of phytate in the bioavailability of dietary minerals. Bioavailability is a general term that refers to how well a nutrient can be absorbed and used by the body. Phytic acid exerts its inhibitory effect on the absorption of zinc, iron and calcium by forming insoluble complexes in the gut under physiological condition. The formation of such chelates depends on the ratio of the content of zinc, iron or calcium relative to that of phytate in the food (Umeta *et al.*, 2005). The calculated molar ratio values are also compared with the reported critical toxicity values for these ratios. Thus, the suggested critical values for the inhibitory effect of phytate on the bioavailability of minerals: Phytate/Iron >1, Phytate/Zinc >15, Phytate/Calcium >0.24 and [phytate]×[Calcium]/Zinc >200 (Norhaizan and Faizadatul, 2009). Therefore, the molar ratio phytate:mineral seems to be important to estimate the absorption, especially of iron, zinc and calcium. The calculated Ca: Phy, Phy: Zn, Phy: Fe and [Ca] [Phy]/ [Zn] molar ratios of raw and processed hyacinth bean were shown in Table 4.5.

4.5.1. Phytate: Zinc (Phy:Zn) molar ratio

The mean phy: Zn molar ratio of raw, soaked, boiled and autoclaved hyacinth bean varieties were found to be 11.49, 15.08, 11.29 and 14.95 for Rongai variety and 17.86, 12.96, 10.10 and 11.15 for Highworth variety respectively as shown from (Table 4.5). Except boiling of Highworth, all the treatments did not significantly ($p>0.05$) affected the mean molar ratio of

Phy:Zn as compared to the raw in both hyacinth bean varieties. All the mean molar ratio values (phy: Zn) were below the critical toxicity level (Phy:Zn >15) except soaking and raw for Rongai and Highworth varieties respectively. Therefore, the calculated phy:Zn molar ratios as an index for the potential zinc bioavailability due to the reduction of phytate after processing the hyacinth bean. Zinc has been described as the essential mineral most adversely affected by phytate and phytate to zinc molar ratios has been proposed as an indicator of zinc bioavailability.

Table 4.5. Effect of processing on phytate/mineral molar ration of hyacinth bean varieties

Variety	Process	Phy:Zn[1]	Phy:Fe[2]	Ca:Phy[3]	[Ca]*[Phy]:Zn[4]
	Raw	11.49 ± 0.06^{a}	1.38 ± 0.03^{a}	6.25 ± 0.15^{b}	0.37 ± 0.01^{a}
	Boiled	11.29 ± 1.28^{a}	1.32 ± 0.11^{a}	7.77 ± 0.53^{a}	0.40 ± 0.07^{a}
Rongai	Autoclaved	14.95 ± 0.88^{a}	1.15 ± 0.01^{a}	8.37 ± 0.46^{a}	0.56 ± 0.02^{a}
	Soaked	15.08 ± 1.81^{a}	1.26 ± 0.01^{a}	8.00 ± 0.09^{a}	0.57 ± 0.08^{a}
	Raw	17.86 ± 3.49^{x}	1.36 ± 0.05^{x}	7.38 ± 0.25^{y}	0.66 ± 0.08^{x}
Highworth	Boiled	10.10 ± 0.52^{y}	1.05 ± 0.03^{y}	8.92 ± 0.04^{x}	0.37 ± 0.03^{y}
	Autoclaved	11.15 ± 0.69^{xy}	$1.15\pm.0.04^{y}$	8.15 ± 0.24^{xy}	0.42 ± 0.03^{y}
	Soaked	12.96 ± 0.23^{xy}	1.12 ± 0.04^{y}	8.58 ± 0.72^{y}	0.48 ± 0.01^{xy}
Variety		Sig	NS	NS	Sig
***process**					

Values in the column followed by different superscripts are significantly different ($p< 0.05$).

Each value is a mean of two duplicates ± SE

[1]mg of phytate/molecular weight of phytate: mg of zinc/molecular weight of zinc.

[2]mg of phytate/molecular weight of phytate: mg of iron/molecular weight of iron.

[3]mg of Calcium/molecular weight of Calcium: mg of phytate/molecular weight of phytate.

[4](mg of Calcium/molecular weight of Calcium)* (mg of phytate/molecular weight of phytate)/ (mg of zinc/molecular weight of zinc) devided by 100.

Especially Phytate forms chelating conjugates with nutritionally important zinc which results in insoluble complexes difficult for humans to hydrolyze during digestion, and thus, typically are nutritionally less available for absorption (Afinah *et al.*,2010). It has been observed that variety was non significant in its Phy:Zn molar ratio but Highworth variety contains(17.86) which was above the index for the potential zinc bioavailability and this suggests that the phytate present in raw Highworth could impair the absorption of zinc present in the grain and contribute to zinc deficiency. Moreover, the present study result indicated that, the interactive effect of variety x processing was significant ($p<0.05$) for the phy: Zn molar ratio of hyacinth bean.

4.5.2. Phytate : Iron ([Phy]/ [Fe]) molar ratio

The mean phytate: iron molar ratios of raw, soaked, boiled and autoclaved hyacinth bean varieties were 1.38, 1.26, 1.32 and 1.15 for Rongai and 1.36, 1.12, 1.05 and 1.15 for Highworth variety as shown in Table 4.5. The mean phy:Fe molar ratio of cooked (boiled and autoclaved) and soaked hyacinth bean seeds were not significant ($p>0.05$) in Rongai while they were significant ($p<0.05$) in Highworth variety as compared to the raw. Even though the treatments did not show a consistency on phy:Fe molar ratio of both hyacinth bean variety, they indicated a pattern of reduction. From the result obtained in the present study all mean values of phy: Fe molar ratios were higher than the critical toxicity level (i.e phy/Fe >1). Similarly Umeta *et al.*(2005) reported that the phy:Fe molar ratio of whole kidney bean stew was 2.8±0.4 which adversely inhibits the absorption of iron. The iron was poorly absorbed by the body after ingestion of hyacinth bean in a meal. So people who consume only the hyacinth bean products may be prone to deficiency of iron due to the inhibitory effect of phytate unless other advanced treatment which can reduce the phytate content in adequate amount is used. Usually, the divalent cations including iron form insoluble penta- and hexa-substituted phytate salts. The insolubility of these complexes is regarded as the major reason for the reduced bioavailability of iron in particular and minerals in general due to diets high in phytate (Afinah *et al.*, 2010).

The mean values of phy:Fe molar ratios between raw hyacinth bean varieties were not significantly($P<0.05$) different to each other but Rongai was slightly higher(1.38) than Highworth (1.36) variety. This result implies that both Highworth and Rongai varieties adversely inhibit the bioavailability of iron due to high contents of phytate. Similarly, the interactive effect of variety x processing was found to be non significant for phy:Fe molar ratio of hyacinth bean.

4.5.3. Calcium:Phytate ([Ca]/[Phy]) molar ratio

The effect of processing on the molar ratio of Ca:Phy for hyacinth bean varieties was presented in Table 4.5. The Ca: Phy mean molar ratio of raw, soaked, boiled and autoclaved hyacinth bean varieties were 6.25, 8.00, 7.77 and 8.37 for Rongai and 7.38, 8.58, 8.92 and 8.15 for Highworth variety respectively. Cooking treatments (boiling and autoclaving) and soaking significantly ($p<0.05$) increased the Ca: Phy molar ratio in Rongai variety whereas boiling treatment only significantly ($p<0.05$) increased the Ca: Phy molar ratio in Highworth variety as compared to the raw. Though autoclaving and soaking had no significant ($p<0.05$) effect on the Ca:Phy molar ratio in Higworth variety, but they would show a pattern of increment on the molar ratio. When the Ca: Phy molar ratios were greater than six, indicates poor calcium bioavailability (Oladimeji *et al.*, 2000). From the study result obtained, all the mean values of the Ca:Phy was found to be higher than the critical molar ratio, therefore, calcium absorption is adversely affected by phytate. Rongai and Highworth varieties were significantly ($p<0.05$) different in their raw Ca: Phy molar ratios. These revealed that Highworth variety contained slightly higher (7.38) mean value of Ca: Phy molar ratio than the Rongai (6.25) variety. Moreover, the interactive effect of the variety x processing was significant ($p<0.05$) on the Ca:Phy molar ratio of the hyacinth bean.

4.5.4. [Cacium]*[Phytate]/[Zinc molar ratio

The effect of processing on the [Ca]*[Phy]:[Zn] millimolar ratio of hyacinth bean varieties was presented in (Table 4.5). The mean [Ca]*[Phy]:[Zn] millimolar ratios of raw, soaked, boiled and autoclaved hyacinth bean varieties were found to be 0.37, 0.57, 0.40 and 0.56 for Rongai and 0.66, 0.48, 0.37 and 0.42 for Highworth variety respectively. Cooking treatments (boiling and autoclaving) and soaking were not significant ($p>0.05$) on the [Ca]*[Phy]/[Zn] millimolar ratio of Rongai variety but there was a significant ($p<0.05$) difference observed in boiling and autoclaving treatment for Highworth variety as compared to the raw. The potentiating effect of calcium on zinc absorption in the presence of high phytate intakes has led to the suggestion that the [Phy]*[Ca]/[Zn] millimolar ratio may be a better index of zinc bioavailability than the [Phy]/[Zn] molar ratio alone (Obah and Amusan, 2009). High calcium levels in foods can promote the phytate-induced decrease in zinc bioavailability when the [Ca]*[phytate]/[Zn] millimolar ratio exceeds 0.5 mol/kg (Umeta *et al.*, 2005). Similarly, Adetuyi *et al.*(2011) suggested that the calculated [Ca]*[Phytate]/[Zn] molar ratio is considered a better index for predicting zinc bioavailability compared with the phytate:zinc ratio because of the calcium to

phytate interaction. In this study, except autoclaved and soaked in Rongai variety and raw in Highworth varieties the mean values of [Ca]*[phytate]/[Zn] millimolar ratio were below the critical level. Therefore, cooking treatment and soaking of hyacinth bean enhances the bioavailability of zinc particularly in Highworth variety.

4.6. Effect of processing methods on sensory characteristics of hyacinth bean

Uncooked dried legumes are virtually indigestible, tasteless, and too hard to eat anyway. Processing is essential to make beans edible and to improve the nutritional quality, sensory and digestibility of the bean (Mann and stewart Truswell, 2002). The hyacinth bean is not consumed in its uncooked form rather it is edible only in its processed (i.e, boiled, autoclaved, microwave cooked and other thermal processes) to enhance the nutritional quality and increase the palatability of the products.

Sensory characterization of hyacinth bean as affected by different processing method is shown in (Table 4.6). The major sensory attributes of the boiled and autoclaved hyacinth bean products were indicated with respective orders as follows: taste 5.15-6.10, color 6.60-7.30, aroma 6.05-6.35, texture 6.85- 7.55 and overall acceptability 6.20-6.60 respectively. Cooking methods (boiling and autoclaving) on the sensory attributes of the hyacinth bean had no significant ($p > 0.05$) difference when they compared to each other. The mean score values for color and texture for both treatments (i.e, boiling and autoclaving) in both hyacinth bean varieties were more as compared to other sensory attributes, whereas the values for taste in both treatments were accounts the least of all by the panelist. In fact the panelists liked the taste of boiled Highwoth variety than others in both Highworth and Rongai. However, most of the panelist dislikes the taste of both varieties because they never consume this product before now and they said that there was a bitter taste after swallowing. As the sensory characteristics are important in consumer point of view, the prepared product must possess good sensory attributes except taste. Generally, it is true that processing improves nutrient value and enhances the palatability of the product for consumption.

Table 4.6. Effect of processing on sensory attributes of hyacinth bean varieties

Variety	Process	Attributes				
		Taste	Color	Aroma	Texture	Overall acceptability
Rongai	Boiled	5.15± 0.47[a]	7.30±0.23[a]	6.05±0.28[a]	7.05±0.32[a]	6.35±0.31[a]
	Autoclaved	5.55±0.46[a]	6.60±0.35[a]	6.35±0.31[a]	6.85±0.33[a]	6.20±0.46[a]
Highworth	Boiled	6.10±0.44[x]	6.75±0.26[x]	6.05±0.33[x]	7.45±0.28[x]	6.40±0.33[x]
	Autoclaved	5.50±0.43[x]	6.60±0.28[x]	6.10±0.33[x]	7.55±0.23[x]	6.60±0.22[x]
Variety*process		NS	NS	NS	NS	NS

Values in the column per variety followed by same superscripts are not significantly different ($p < 0.05$). (N = 20)

5. Conclusion and Recommendation

5.1. Conclusion

The study of the hyacinth bean attempted to get adequate information about its chemical composition (crude protein, crude fat, total ash, carbohydrate, moisture and crude fiber contents). And also, this paper provides data on the content of zinc, iron, calcium, phosphorus, phytate and tannin on the relative bioavailability with particular reference to their index of absorption in the body. In addition, the effect of cooking and soaking on the nutritional and sensory qualities of hyacinth bean products after the processing had been studied.

The results of this study showed that raw hyacinth bean (*Lablab purpureus L.) sweet* contains appreciable quantity of crude protein, carbohydrate, crude fiber, calcium, iron, zinc and phytate and low levels of crude fat and tannin. Thus, the consumption of hyacinth bean products alleviates protein-energy malnutrition caused by inadequate intake of these nutrients in the world in general and in Africa and Ethiopia in particular. Moreover, since hyacinth bean contributes an appreciable amount of zinc and iron, then it could protect micronutrient deficiency diseases related to zinc and iron and they enhances the growth of children and infants.

As indicated in this study, cooking method (boiling and autoclaving) improved the nutritional qualities of protein, carbohydrate and crude fiber and sensory qualities whereas it caused slight reduction in the amount of total ash and crude fat contents in the hyacinth bean varieties. Moreover, these traditional household practices such as boiling and cooking treatments such as autoclaving are very effective to decrease the phytate and tannin content significantly. Since boiling and autoclaving treatment increased the calcium, iron and phosphorus contents significantly by reducing the anti-nutritional factors in the hyacinth bean, then the consumption of the hyacinth bean varieties which need to encouraged to address the problem of iron, calcium zinc and protein deficiency not only in Ethiopia but also elsewhere.

5.2. Recommendation

The following recommendations are drawn from the thesis work on hyacinth bean.

- Both boiling and autoclaving methods are effective in improving the nutritional qualities of Rongai and Highworth varieties.
- Boiling treatment at household level is sufficient to enhance nutritive value and to reduce the anti-nutritional factors in the hyacinth bean.
- Since both varieties of hyacinth bean have an appreciable amount of protein, carbohydrate and micronutrients such as iron and zinc, thus, the consumption of the bean is advisable to combat protein energy malnutrition and micronutrient deficiency diseases related to iron and zinc in developing countries in general and Ethiopia particularly.

The following are indentified as future works to be done on the hyacinth bean by nutritionists and food scientists.

- Determination of other chemical species in the hyacinth bean varieties (total polyphenols, lectins, trypsin inhibitor, etc.)
- Determination of antioxidant properties of hyacinth bean and its products with other cereals
- Estimation of nutritional composition, anti-nutritional factors and antioxidants in pods and immature green hyacinth bean.
- Formulation of new products with its unique micronutrients and essential amino acids.
- Indentifying the medicinal value of both Rongai and Highworth varieties.

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Appendix

Technical questionnaire

The purpose of this questionnaire is to gather information about the hyacinth bean from the local community. The questionnaire gives an emphasis on how the local people process hyacinth bean for their consumption at home level and some uses of the bean in the community.

This questionnaire is filled by the local community and farmers that cultivates a hyacinth bean. Every questionnaire should be answered in appropriate way.

1. Address

Name _____

Sex male ☐ female ☐ Age _____

Region _____ woreda _____ kebele _____

2. Information about hyacinth bean

a) What was the geographical origin of hyacinth bean ?

_____

b) When was it introduced in the region?

_____

c) What are the existing varieties of hyacinth bean ?

3.. Information On Agronomic Characterstics Of hyacinth bean

a) When do the farmers sow hyacinth bean?

Mehir ☐ summer ☐

Spring ☐ Others_____

b) What is the sowing date (month)?

_____

_____

b) What altitude range is suitable for its growth?

_____

c) What is the total rain fall amount it requires?

---

d) What climatic (weather) conditions are suitable for its harvest?

---

e) What type of soil favors its growth?

---

f) Is any fertilizer enhancement required for its harvest?

Yes ☐ No ☐

g) Does it require pesticide for its harvest?

Yes ☐ No ☐

h) Comparing with other commonly harvested legumes, does hyacinth bean have an advantage?

Yes ☐ No ☐

i) If the answer for the above question is yes, what are the advantages it has over the others?

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4. The Consumption Of Hyacinth Bean In The Society

a) Can hyacinth bean be edible food source?

Yes ☐ No ☐

b) If the answer for the above question is no, what limits its consumption?

---

c) If the answer for the question (4a) is yes, what are the main food items that can be prepared from it?

_____

_____

d) in what forms does the hyacinth bean consumed?

1. Cooked bean (niffro) 2. Stew (watt) 3. Shiro watt 4. Cooked with other cereals 5. Any _____

e) What are the main traditionally used processes to make it edible?

_____

_____

f) What are the effects of these processes on the raw seed? (i.e. palatability, reduction of unwanted taste, reducing flatulence, etc)

_____

_____

g) What are the other uses of hyacinth bean for the local community?

_____

_____

In general can hyacinth bean have a medicinal value? For what kind of diseases?

_____

_____

Any information that you would like to add

_____

_____

THANK YOU

Sensory Acceptability Test

The purpose of this sensory test is to check which processing treatment are more relevat in its acceptability by panalists.

Instructions

Please rinse your mouth with water before starting and between each tasting.

Taste the samples according to the numbers indicated and give value from 1-9 for each attribute based on the key given.

If you have any question please ask the server.

Keys

1. Dislike extremely
2. Dislike very much
3. Dislike moderately
4. Dislike slightly
5. Neither like nor dislike
6. Like slightly
7. Like moderately
8. Like very much
9. Like extremely

	Sample code			
Attributes	**021**	**032**	**045**	**061**
Taste				
Colour				
Aroma				
Texture				
Over all acceptability				

General Comments:

_____

_____

_____

Tests of Between-Subjects Effects for proximate

Source	Dependent variable	Type III sum of square	Df	Mean square	F	Sig.
Variety	Protein	13.939	1	13.939	5.045	.039
	Fat	0.026	1	0.026	3.465	.081
	Ash	0.056	1	0.056	7.123	.017
	Moisture	0.154	1	0.154	33.241	.000
	FIBER	0.368	1	0.368	4.229	.056
	CHO	14.555	1	14.555	4.688	.046
Processing	Protein	25.559	3	8.520	3.083	.057
	Fat	1.176	3	0.392	52.247	.000
	Ash	2.176	3	0.725	92.159	.000
	moisture	21.594	3	7.198	1557.729	.000
	FIBER	7.727	3	2.576	29.636	.000
	CHO	52.614	3	17.538	5.649	.008
Variety*processing	Protein	6.655	3	2.218	.803	.510
	Fat	0.024	3	0.008	1.080	.386
	Ash	0.076	3	0.025	3.240	.050
	Moisture	1.146	3	0.382	82.705	.000
	FIBER	1.570	3	0.523	6.020	.006
	CHO	26.106	3	8.702	2.803	.073
Error	protein	44.209	16	2.763		
	fat	0.120	16	0.008		
	ash	0.126	16	0.008		
	moisture	0.074	16	0.005		
	FIBER	1.391	16	0.087		
	CHO	49.670	16	3.104		
Total	protein	10365.430	24			
	fat	21.900	24			
	ash	233.075	24			
	moisture	1846.843	24			
	FIBER	1922.255	24			
	CHO	79768.033	24			
Corrected total	protein	90.362	23			
	fat	1.347	23			
	ash	2.435	23			
	moisture	22.968	23			
	FIBER	11.055	23			
	CHO	142.945	23			

Tests of Between-Subjects Effects for minerals

Source	Dependent variable	Type III sum square	df	Mean square	F	Sig.
Variety	Fe	3.971	1	3.971	4.175	.075
	Zn	.124	1	.124	4.890	.058
	Ca	176.181	1	176.181	8.980	.017
	P	889.688	1	889.688	.370	.560
Processing	Fe	4.405	3	1.468	1.544	.277
	Zn	.603	3	.201	7.940	.009
	Ca	356.420	3	118.807	6.055	.019
	P	52302.630	3	17434.210	7.248	.011
Variety*processing	Fe	4.958	3	1.653	1.737	.237
	Zn	.820	3	.273	10.804	.003
	Ca	389.771	3	129.924	6.622	.015
	P	20637.926	3	6879.309	2.860	.104
Error	Fe	7.610	8	.951		
	Zn	.202	8	.025		
	Ca	156.959	8	19.620		
	P	19242.208	8	2405.276		
Total	Fe	7166.947	16			
	Zn	92.000	16			
	Ca	336591.302	16			
	P	12409391.275	16			
Corrected total	Fe	20.943	15			
	Zn	1.749	15			
	Ca	1079.330	15			
	P	93072.452	15			

Tests of Between-Subjects Effects for ANFs

Source	Dependent variable	Type III sum square	df	Mean square	F	Sig.
variety	phytate	611.656	1	611.656	7.747	.013
	tannin	.248	1	.248	98.412	.000
Processing	phytate	7829.969	3	2609.990	33.057	.000
	tannin	.119	3	.040	15.769	.000
Variety*processing	phytate	1310.716	3	436.905	5.534	.008
	tannin	.007	3	.002	.902	.462
Error	phytate	1263.281	16	78.955		
	tannin	.040	16	.003		
Total	phytate	2209489.436	24			
	tannin	2.594	24			
Corrected total	phytate	11015.621	23			
	tannin	.415	23			

Tests of Between-Subjects Effects for phytate/mineral molar ratios

Source	Dependent variable	Type III sum square	df	Mean square	F	Sig.
Variety	Phy:Zn	.134	1	.134	.029	.870
	Phy:Fe	.044	1	.044	10.075	.013
	Ca:Phy	1.767	1	1.767	8.295	.021
	[Ca][Phy]/Zn	.000	1	.000	.066	.804
Processing	Phy:Zn	36.548	3	12.183	2.600	.124
	Phy:Fe	.117	3	.039	9.000	.006
	Ca:Phy	6.633	3	2.211	10.379	.004
	[Ca][Phy]/Zn	.044	3	.015	1.978	.196
Variety*processing	Phy:Zn	60.859	3	20.286	4.330	.043
	Phy:Fe	.047	3	.016	3.633	.064
	Ca:Phy	1.230	3	.410	1.925	.204
	[Ca][Phy]/Zn	.119	3	.040	5.330	.026
Error	Phy:Zn	37.484	8	4.686		
	Phy:Fe	.035	8	.004		
	Ca:Phy	1.704	8	.213		
	[Ca][Phy]/Zn	.059	8	.007		
Total	Phy:Zn	2885.348	16			
	Phy:Fe	24.179	16			
	Ca:Phy	1017.024	16			
	[Ca][Phy]/Zn	3.858	16			
Corrected total	Phy:Zn	135.024	15			
	Phy:Fe	.243	15			
	Ca:Phy	11.335	15			
	[Ca][Phy]/Zn	.223	15			

Tests of Between-Subjects Effects for sensory evaluation

Source	Dependent variable	Type III sum square	df	Mean square	F	Sig.
Variety	taste	4.050	1	4.050	1.012	.318
	color	1.513	1	1.513	.930	.338
	aroma	.312	1	.312	.160	.690
	texture	6.050	1	6.050	3.499	.065
	overall	1.012	1	1.012	.439	.510
Processing	taste	.200	1	.200	.050	.824
	color	3.613	1	3.613	2.222	.140
	aroma	.613	1	.613	.314	.577
	texture	.050	1	.050	.029	.865
	overall	.013	1	.013	.005	.942
Variety*processing	taste	5.000	1	5.000	1.249	.267
	color	1.513	1	1.513	.930	.338
	aroma	.313	1	.313	.160	.690
	texture	.450	1	.450	.260	.611
	overall	.613	1	.613	.265	.608
Error	taste	304.300	76	4.004		
	color	123.550	76	1.626		
	aroma	148.250	76	1.951		
	texture	131.400	76	1.729		
	overall	175.350	76	2.307		
Total	taste	2800.000	80			
	color	3843.000	80			
	aroma	3163.000	80			
	texture	4314.000	80			
	overall	3441.000	80			