

Vitamin C, Fibre, Lignin and Mineral Contents of Some Edible Legume Seedlings

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Abstract: Seedling of 5 species of legumes were analysed for their vitamin C, fibre, lignin and mineral contents using standard analytical techniques. *Vigna sinensis* (Oloyin), *Vigna sinensis* (Ife-brown), *Vigna unguiculata* (Mala), *Vigna unguiculata* (Sokoto) were analysed at day 7 and *Cajanus cajan* at day 14. The ascorbic acid contents were found to be 10.6, 26.4, 31.4, 34.6 and 38.4 mg/100 g, respectively; the fibre contents gave values of 22, 20, 20, 21.5 and 31%, while the lignin contents were 0.057, 0.062, 0.082 and 0.18%, respectively. The results of trace mineral analysis gave values ranging from 3.36-7.84 mg/100 g for calcium, 0.72-2.16 mg/100 g for copper, 5.40-18.00 mg/100 g for iron, 81.00-82.26 mg/100 g for potassium and 93.00 -123.60 mg/100 g for magnesium in the 5 species. The values obtained for potassium and magnesium were high for all the species.

Key words: Fiber, lignin, mineral, edible, seedling, vitamin C

INTRODUCTION

The large protein rich seeds of plants belonging to the family of the legumes also called pulses constitute one of mankind important sources of food. Peas, kidney beans, lima beans, lentils, chickpeas, mungbeans, cowpea, peanuts and broad beans are the principal food legumes (Bishop *et al.*, 1983).

Green vegetable are living entities and are respiring when they are freshly harvested. They have a high content of water and an abundance of cellulose. The cellulose is in a form which although not digested serves a useful purpose on the intestine as roughage, thus promoting normal elimination of waste products.

The chief nutritive significance of green vegetables is their richness in minerals and vitamins. Leaves in general are important sources of iron vitamin A, thiamin, riboflavin and ascorbic acid. The protein in leaves is low, but what is present is usually of the highest grade.

Although, bean seeds do not contain vitamin C (ascorbic acid), the vitamin is formed in useful amount in sprouted bean seeds. Green vegetables are especially valuable when eaten raw, as they will suffer no cooking losses, there are however, considerable loss of vitamin C in wilted vegetables (Durust *et al.*, 1997).

Green vegetables lose their eating quality very rapidly after harvesting Sugar content declines and the amount of cellulose increases. At this stage also vitamin C content of leaves begin to decrease as the structure of the living cells become disorganized.

The aim of the research is to determine the suitability of leguminous seedlings as a source of vitamin C, fibre

Table 1: Species names of this research

Botanical name	Local name	English name
<i>Vigna sinensis</i>	Oloyin	Cowpea (red)
<i>Vigna sinensis</i>	Ife-Brown	Cowpea (red)
<i>Vigna unguiculata</i>	Mala	Cowpea (red)
<i>Vigna unguiculata</i>	Sokoto	Cowpea (white)
<i>Cajanus cajan</i>	Feregede	Pigeon pea

and mineral. It has been discovered abroad that leguminous sprouts are good sources of vitamin C and it is being consumed as a result of it.

MATERIALS AND METHODS

Seed collection: Seed of different species of legumes used for this research were obtained from the open market in Akure and Ibadan except for the *Phaseolus vulgaris* from Jos. Those selected were the harvest from immediate past planting season and were also made sure that they were not infested with weevils to ensure their viability and germinability.

In all, 5 species were collected as shown in Table 1 for this research.

Planting preparation and planting: Boiling test tubes (50 cm) were used for planting the seeds. These test tubes were filled half way with vermiculite (sterilized) as the planting medium for growth and watered up to it's level and labelled according to the species of beans to be planted inside it.

The required number of seeds to be planted were carefully selected for each species and sterilized (by soaking) in 1% v v⁻¹ hypochlorite solution for 1 h to prevent fungal growth during germination.

The seeds are drained, blotted with cotton wool and then 2 seeds are placed in each of the already prepared test tubes just on top of the vermiculite and kept in a carton. They are kept in the dark for 2 days to allow them sprout after which they are brought to the open to continue germinating inside a cage until they are required.

Sample collection and preparation for analysis: Samples were collected at day 7 for the 4 cowpea species and day 14 for pigeon pea. The plants were carefully uprooted (followed by the removal of the cotyledons) and rinsed with water to remove adhering vermiculite and material that will interfere with the analysis.

The well rinsed samples are weighed in the required quantities depending on the analysis to be carried out and chopped into small pieces with a clean razor. The samples at this stage is ready for ascorbic acid analysis but further treatment on the samples is required for crude fibre, ligning and mineral analysis.

Defatting (fat removal)-crude fibre samples: Ten gram of each sample was soaked in hexane and covered with aluminium foil (to prevent evaporation of the solvent) for 24 h. They were filtered and spread on a sheet in the open until no smell of hexane can be perceived.

They were then dried in the oven at a temperature of 50°C until they are completely dry after which they were ground into fine powder using mortar and pestle. These samples were kept in a dessicator until required.

Extractives (organic and aqueous soluble substances) removal-lignin determination samples: About 10 g of each sample was soaked in hexane and ethanol 1:1 for 24 h. The same procedure was followed for the extraction as that of defatting repeating all the above steps to the last.

Drying-mineral analysis samples: The samples were dried in the oven at a temperature of 40°C. They were then ground into fine powder with mortar and pestle and kept in a dessicator until required.

Ascorbic acid determination (AOAC, 1990)

Preparation of reagents

Extraction solution: About 15 g of trichloroacetic acid was dissolved in 40 mL acetic acid and 200 mL distilled water. It was diluted to 500 mL and filtered.

Standard solution: About 0.05 g of ascorbic acid was dissolved in 60 mL of the extraction solution and made up to 250 mL with distilled water. It was always prepared immediately before use.

Indophenol standard solution: About 0.05 g of 2, 6-dichlorophenolindophenol (sodium salt) was dissolved in 100 mL of distilled water and filtered.

The 2,6-dichlorophenolindophenol was standardized by titrating against 10 mL of acid stock solution until a faint pink colour was obtained. The concentration of the ascorbic acid in mg was expressed as equivalent to 1 mL of dye solution.

$$V \text{ mL of dye} = 0.05 \text{ g Ascorbic acid (mg)}$$

$$1 \text{ mL} = \frac{0.05}{V}$$

where,

V = Titre value.

To about 5 g of each of the fresh and chopped samples were added 60 mL of the extraction solution and the mixture was homogenized with an electric high speed homogenizer. The mixture was filtered under suction. The filtrate was poured into 250 mL volumetric flask and made up to the mark with distilled water.

Ten milliliter of the resulting solution was pipetted into a conical flask and titrated against the standard indophenol solution and the titre value (Y) recorded. This was done 4 times for each sample.

$$10 \text{ mL of sample solution} = Y \times \frac{0.05}{V}$$

- 100 mL of sample contained K mg of ascorbic acid.
- 100 g of sample contained 20 K mg ascorbic acid.
- In 100 g leaf sample = 20 K mg ascorbic acid.

$$K \text{ (mg)} = Y \times \frac{0.05}{V} \times \frac{250}{10} \times \frac{100 \text{ mg}/100 \text{ g}}{W}$$

Crude fibre determination (AOAC, 1990): One gram portion of each of the dried defatted sample was weighed and transferred into a conical flask. Added to it was 70 mL of 1.25% v v⁻¹ H₂SO₄ solution and the mixture boiled gently for 30 min, maintaining a constant volume by rotating the flask every few minutes in order to mix the content and remove particles from the sides.

At the end of the 30 min, the acid was allowed to stand for 1 min and then filtered. The insoluble matter washed with hot water; it was then washed back into the original flask with 1.25% w v⁻¹ NaOH solution. Seventy milliliter of 1.25% NaOH solution was added to the residue in all.

The mixture was boiled for another 30 min with the same precautions as those used in the earlier boiling acid treatment. It was allowed to stand for 1 min and then

filtered immediately. The residue was rinsed with hot water, into a crucible, evaporated on a hot plate, dried in the oven and weighed.

The dried residue was ashed at 59°C for 30 min and weigh again. The weight of dried residue before ashing less the ash itself is referred to as crude fibre.

Lignin determination (gravimetric method): One gram of portion of the dried extracted sample was weighed and discharged into a round bottom flask and added to it was 12.5 mL of 72% v v⁻¹H₂SO₄ solution, stirring for 2 h at room temperature.

The mixture was diluted to 4% H₂SO₄ and boiled under reflux for 4 h. The reaction is cooled, filtered and the residue washed several times with hot water. The residue is dried in a dessicator for 3 days and weighed. The determination was done in duplicate for each sample.

Mineral analysis (atomic absorption spectrophotometric method): Eleven gram of portion of each of the samples was weighed and placed in a crucible. They were then ashed in the muffle furnace at 550°C for 3 h and cooled in a dissicator. The resulting white ash was dissolved subsequently in 5 mL of 20% v v⁻¹ HCl solution heating gently for 30 min. A clear solution was filtered and diluted to a concentration suitable for working range of the instrument and kept in plastic containers.

The mineral analysis of samples were done using atomic absorption spectrophotometer (Buck Scientific Model 110).

RESULTS AND DISCUSSION

Seedling growth: The 5 species of legumes were planted for analyse.

Ascorbic acid (vitamin C): Titration method by procedures based on methods as described in the AOAC (1990) was used. Table 2 presents the results of the vitamin C present in the seedlings of the various legumes.

From Table 1, it can be seen that the 5 species of pulses contain moderate quantities of ascorbic acid of 10.6, 26.4, 31.4 and 38.4 mg/100 g as these values are close

to that of orange which is about 50 mg/100 g (Kirschamann, 1979). It could be deduced from these value that the white cowpea species (*Vigna unguiculata*) are better sources of vitamin C than the brown species (*Vigna sinensis*). Also, the values might increase with increase in age of the plants as reflected in the highest value of 38.4 mg/100 g obtained for *Cajanus cajan* at day 14. This suggests that more vitamin C can be got from older plants but on the contrary the lignin content will be higher.

Virtually all animal species including those domesticated animal, posses the necessary enzymes for the conversion of glucose to ascorbic acid (Ihekoronye and Ngoddy, 1992). Only man and a handful of other species require a dietary source of this vitamin. Apart from the primary function of the vitamin in maintaining collagen and in healing wounds and burns, it also aids in forming red blood cells and in preventing haemorrhaging. Vitamin C fight bacteria infection (Mudambi and Rajagoval, 1987) and it may also have a protective role to play against cancer (Muller, 1988).

Vitamin C has significant relationship with other nutrients. It aids in the metabolism of the amino acid phenylalanine and tyrosine. Vitamin C converts the inactive form of folic acid to the active form, folinic acid and may have a role in calcium metabolism. In addition, vitamin C protects thiamin, riboflavin, folic acid, panthotenic acid and vitamin A and E against oxidation (Mudambi and Rajagoval, 1987). It also detoxifies lead, cadmium copper, DDT, mercury and many other environmental toxins.

Crude fibre: The method of AOAC (1990) was used, Table 3 presents the result of the crude fibre content of the seedlings of the various legumes.

The crude fibre content as seen from Table 2, are almost the same except for *Cajanus cajan* which contains more fibre. And this is understandable as it was analysed a week after other species. The result of the determination prove that the plants can be good sources of fibre in the diet as the values of between 20 and 22% compare favourably with 20% fibre content in cabbage.

Fibre is the part of food that is not digested by the human body. The normal functioning of the intestinal

Table 2: Ascorbic acid contents of some edible legumes

Sample	Weight of samples (g)	Titre values (cm ³)				Average litre	Ascorbic acid mg/100 g
		1	2	3	4		
<i>Vigna sinensis</i> (Oloyin)	5.58	0.10	0.15	0.15	0.10	0.125±0.029	10.6
<i>Vigna sinensis</i> (Ife-brown)	5.35	0.30	0.30	0.30	0.30	0.30±0.00	26.4
<i>Vigna unguiculata</i> Mala	5.65	0.40	0.35	0.30	0.45	0.375±0.065	31.4
<i>Vigna unguiculata</i> Sokoto	5.45	0.40	0.40	0.40	0.40	0.40±0.00	34.6
<i>Cajanus cajan</i> (Feregede)	5.53	0.40	0.50	0.40	0.50	0.45±0.058	38.4

Table 3: Crude fibre contents of some edible legumes

Sample	Weight of crude fibre (g)		Mean weight of crude fibre	Percentage
	1st determination	2nd determination		
<i>Vigna sinensis</i> (Oloyin)	0.21	0.23	0.22±0.01	22.0
<i>Vigna sinensis</i> (Ife-brown)	0.20	0.20	0.20±0.00	20.0
<i>Vigna unguiculata</i> (Mala)	0.20	0.20	0.20±0.00	20.0
<i>Vigna unguiculata</i> (Sokoto)	0.00	0.22	0.215±0.007	21.5
<i>Cajanus cajan</i> (Feregede)	0.32	0.30	0.30±0.01	31.0

Table 4: Lignin contents of some edible legumes

Sample	Weight of lignin (g)		Mean weight of lignin	Percentage
	1st determination	2nd determination		
<i>Vigna sinensis</i> (Oloyin)	0.25	0.27	0.26±0.01	0.050
<i>Vigna sinensis</i> (Ife-brown)	0.31	0.31	0.31±0.00	0.062
<i>Vigna unguiculata</i> (Mala)	0.31	0.30	0.305±0.007	0.061
<i>Vigna unguiculata</i> (Sokoto)	0.36	0.40	0.38±0.03	0.082
<i>Cajanus cajan</i> (Feregede)	0.60	0.62	0.61±0.01	0.189

Table 5: Mineral contents of edible legumes

Sample	Ca mg/100 g	Cu mg/100 g	Fe mg/100 g	K mg/100 g	Mg mg/100 g
<i>Vigna sinensis</i> (Oloyin)	3.36	0.72	05.40	81.00	100.50
<i>Vigna sinensis</i> (Ife-brown)	5.04	2.16	18.00	81.54	096.30
<i>Vigna unguiculata</i> (Mala)	3.92	0.72	06.75	82.26	093.00
<i>Vigna unguiculata</i> (Sokoto)	5.60	0.92	08.10	82.26	096.15
<i>Cajanus cajan</i> (Feregede)	7.84	0.72	12.15	82.08	123.60

tract depends upon the presence of adequate fibre. A low fibre diet has been associated with heart diseases, cancer of the colon and rectum, varicose veins, phlebitis, obesity, appendicitis, diabetes and even constipation.

The relationship between dietary fibre and disease has been the subject of much medical interest recently. Dietary fibre is no longer thought of as roughage as there are many constituents which are not fibrous e.g., pectin and the dietary fibre in pulses. Only plant foods have dietary fibre. It is however, best to get this dietary fibre from natural plant foods rather than from plant extracts. There is some evidence that dietary fibre can interfere with the absorption of minerals in the gut. However, vegetables are rich in minerals and if fibre is eaten in this form, there is very little chance of mineral deficiency.

As most natural sources of fibre are also bulky foods, they will help you control your weight. This is not because of any magic property fibre has in slimming, it is simply because these foods are low in calories and often high in water so that they help to satisfy the appetite before too many calories has gone down.

The natural laxative effect of fibre is partly due to its ability to absorb water and increase the bulk of faeces and partly to the production of volatile fatty acids (acetic, propionic and butyric) in the gut as a result of microbial breakdown of some of the cellulose (Ihekoronye and Ngoddy, 1992).

Lignin: Gravimetric method which is a direct method of lignin determination was used. Table 4 presents the results of the lignin contents of the seedlings of the various legumes.

The results show low lignin contents. This is expected as the plants are young and tender. The progressive lignification in growing plants is also confirmed in the value obtained for *Cajanus cajan* which has a higher lignin content than other. The low lignin contents of the plants will ensure digestibility of the legume seedling if consumed. Also, the greatest laxative effect is produced by fibre low in lignin. Cabbage for example, containing 3% lignin, is more effective than wheat bran which contain 8% lignin.

Mineral analysis: Atomic absorption spectrophotometric method was used. The result of the mineral content of the seedlings of various species of legumes is presented in Table 5.

The results show that the vegetable pulses are good sources of nutritive metals. It shows that the plants are particularly rich in potassium and magnesium. The calcium content is however, low. The low values obtained for copper and iron is desirable as large quantities in foods has been reported to have destructive effects on ascorbic acid.

Minerals are considered to be essential in human nutrition. These minerals are vital to overall mental

physical well being and are important constituents of bones, teeth, tissues, muscles, blood and nerve cell. They generally help in maintenance of acid-base balance, response of nerves to physiological stimulation and clotting of blood in the body.

CONCLUSION

Foods does matter in our lives. Most of the time we are encouraged to eat so we can live. What we really need is sound advice to help us eat so we can live.

It has become obvious in recent years that the major diseases of affluent societies are diet related, consequently, nutrition has emerged as an important part of preventive medicine. Diet is recognized as a preventive factor in the cardiovascular diseases, dental caries osteoporosis, cancer, diverticular disease and anaemia to identify a few. Therefore, when a dietary factor is identified as either therapeutic or it exhibit a casual relationship to a public health problem, it's role in nutrition must be thoroughly explored. Conversely, if a dietary component has undergone a major change in its dietary contribution, its role must be carefully re-evaluated to identify the effect of the change on health.

Vegetables generally have a higher mineral and vitamin content than fruits. Against this background, vegetable intake must be explored as a source of the essential body nutrients and particularly the vegetable pulses (or leguminous vegetables) which can be referred to as the undiscovered vital food in this part of the world, because of all it has to offer us as seen in this research.

The minerals, fibre and vitamin C with their nutritional food values and even more are present and obtainable from the leguminous vegetables which have hitherto

being neglected and unexplored in this part of the world. This would further help us in our quest for healthier eating because most nutrients that are desired in our diet are obtainable from this 'undiscovered vital food'.

Food related illness is usually associated with disease of shortage or deficiency of nutrients now found mostly in the 3rd World countries. Against this background, the leguminous vegetables provide a cheap option to vitality and good health. It is really an accessible and affordable source of essential nutrients for the human body. The incorporation of legume seedling into the diet is desirable.

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