Advances in Science and Technology, Vol. 1, No. 1, January - March 2007, pp. 9-14.

Phytochemical Composition of Vegetable Cowpea Genotypes

A.O. Ano [1] and C. I. Ubochi [2]

[1] Soil Science Laboratories, National Root Crops Research Institute, Umudike,Umuahia, Abia State, Nigeria.
[2]Department of Pure and industrial Chemistry, Imo State University, Owerri, Imo State, Nigeria.

(Submitted: August 25, 2006; Accepted: January 15, 2007)

Abstract

The study examined the levels of phytochemicals in assession of two vegetable cowpea genotypes. Mean alkaloid contents of these cowpea were 0.10±0.0% and 0.08±0.0%for the climbing genotype *Vigna unguculata* subspecies *sesquiped* and the postrate genotype, *Vigna unguiculata* subspecies *dekintana* and *mensensis* respectively. Flavonoids ranged from 0.26 to 0.41% and from 0.26 to 0.30% in the climbing and postrate vegetable cowpea genotype.

The phenolic compounds in the two genotypes were similar with each having a mean value of 0.12%. The climbing genotype had higher tannins with a mean value of 0.53 ±0.32% than the postrate genotype, which had a mean value of 0.03±02%. Saponins in the climbing genotype ranged from 0.13 to 0.22% and from 0.08 to 0.50% in postrate. It was concluded that the two vegetable cowpea genotypes studied were rich sources of these phytochemicals. Consumption of vegetable cowpea should be encouraged because of the health related properties of these phytochemicals.

Keywords: Phytochemicals; alkaloid, tannins, flavonoid, saponin,

1.0 Introduction

Legumes constitute one of mankinds important sources of food. In many developing countries, the legumes are the most important high –protein food, and they play the role played by meat and other animal products in rich countries. Legumes require specific soil and micro-climates for optimum performance. In Nigeria the most popular legumes are cowpea (*Vigna unguiculata* L. Walp) and Soybeans. *Vigna unguiculata* L. Walp commonly called 'beans' in Nigeria survives better under semi-arid and subtropical climatic conditions, while soybean does not tolerate acid soils. The specific requirements of these legumes make their cultivation in Southern Eastern Nigeria with humid climate to be unconducive. Consequently 'beans' eaten in South-Eastern Nigeria are imported from the North. This makes the commodity expensive and beyond the reach of many families. However, in Eastern Nigeria, the farming systems have evolved a particular type of cowpea called vegetable cowpea **"Akidi"** which thrives under humid climate ecology (Udealor, 2002). Vegetable cowpea **"Akidi"** has two genotypes: *Vigna unguiculata* subspecies *sesquiped alis* (Redden, 1981) commonly called **"Akidi enu"**. This genotype has a climbing habit, the other genotype with prostrate habit is called *Vigna unguiculata* subspecies dekintiana and mensensis (Steele and Mehra, 1980) commonly known as **Akidi ani"**. Each of these genotypes have accessions.

Nutrient composition analysis has revealed a high level of protein in the vegetable cowpea genotypes (Ubochi and Ano, 2006 unpublished). Apart from protein, legumes are known to be rich sources of phytochemical. Phytochemicals belong to a group of compounds known as secondary metabolites, they are not involved in the process of plant growth, but act as deterrent to insects and microbial attack (Okwu, 2004). Alkaloids, tannins flavonoids, terpeniods and phenolic compounds are members of this group. These compounds have biological effects as a consequence of their antioxidant properties. Tannins and flavonoids show anticarcinogenic and antimutagenic effects (Urquiagu and Luighton, 2000) Polyphenols interfere in several steps that lead to the development of malignant tumors, inactivating carcinogens and inhibiting the expression of mutagens (Urquiagu and Luighton

2000). There is no literature on the levels of phytochemicals in vegetable cowpea. The objective of this work was to quantify the phytochemicals in vegetable cowpea genotypes.

2.0 Materials and Method

Samples of accessions of vegetable cowpea genotypes were dried in the oven at 60°C to a constant weight. Each of the samples was milled using a Wiley Milling Machine. The milled samples were used for the analysis of phytochemicals. The accessions of each of the vegetable cowpea studied are shown in Table 1.

2.1 Quantitative determination of phytochemicals in vegetable cowpea.

A. Alkaloids

Two grams of each of the samples were defatted with 100ml of diethyl ether using a soxhlet apparatus for 2hrs. Five hundred milligram of each of the defatted samples was weighed into 250ml beaker and 200ml of 20% acetic acid in ethanol was added and covered to stand for 4hr. This was filtered and the extract concentrated to one-quarter of the original volume on a water-bath. Concentrated ammonium hydroxide was added drop-wise to the extract until precipitation was complete. The whole solution was allowed to settle and the precipate collected and weighed (Harborne, 1973, Obadoni and Ochuko, 2001).

B. Tannins

Five hundred milligram of each of the milled samples was weighed into 100ml plastic bottle, 50ml of distilled water was added and shaken for 1 hour in a mechanical shaker. This was filtered into 50ml volumetric flasks and made up to the mark. Thereafter 5ml of the filtrate was pipetted into a test tube and 3ml of 0.1M $FeCl_3$ in 0.1M HCl and 0.008M potassium ferrocyanide were added. Standards were prepared using tannic acid. The absorbance of the samples and standards were measured using a spectrophotometer at 120nm wavelength (Van-Burden and Robinson, 1981).

C. Saponin

Twenty grams of each of the milled samples were dispersed in 200ml of 20% ethanol. The suspension was heated over a hot water bath for 4 hours with continuous stirring at about 55°C. The mixture was filtered and the residue re-extracted with another 200ml of 20% ethanol. The combined extracts were reduced to 40ml over water bath at about 90°C. The concentrate was transferred into 250ml separating funnel and 20ml of diethyl ether was added and shaken vigorously. The aqueous layer was recovered while the organic layer was discarded. The purification process was repeated.60ml of n-butanol was added. The solution was washed twice with 10ml of 5% aqueous sodium chloride. The remaining solution was heated in a water bath. After evaporation, the compound (Saponin) was dried in the oven to a constant weight, and thereafter weighed (Obadoni and Ochuko, 2001).

D. Flavonoid

10g of each of the milled sample were extracted repeatedly with 100ml of 80% aqueous methanol at room temperature. The whole solution was filtered through whatman filter paper No. 42. The filtrate was later transferred into a crucible and evaporated to dryness over a waterbath, and thereafter weighed (Boham and Kocipal, 1974).

3.0 Result and Discussion

A. Alkaloids

Alkaloids are natural organic compounds present in plants. These compounds have basic characters and contain at least one nitrogen atom in a heterocylic ring. Among these compounds are dipiperdine, pyrrolizidine, B-carboline and phenyl ethylamine. Alkaloids have therapeutically significant plant substances. Pure, isolated plant alkaloids and their synthetic derivatives are used as basic medicinal agents for their analgesic, antispasmodic and bactericidal effects (stray 1998). They exhibit marked

...physiological activity when administered to animals even at very small doses (Finar, 2(

The mean alkaloid content of each of the two vegetable cowpea genotypes were : with the climbing genotype (Vigna unguiculata sesquiped alis) having a value of 0. and the postrate genotype (Vigna unguiculate subspecies dekintana and mensensis, a value of 0.08±0.00% (Table 1).

Alkaloid contents of the vegetable cowpea were higher than 0.78 mg/100g report leaves of spondias mombin (Okwu and Okwu, 2004) but lower than 0.22% found in indigenous spices of South-Eastern Nigeria (Okwu, 2004).

B. Phenols

The phenolics form a large group of naturally occurring compounds. They are characteri by the presence of an aromatic ring with one or more hydroxyl groups (Finar, 2002). Th compounds have quite a diverse range of biological activities depending on their structu concentration and degree of polymerization (Urquiaga and Leighton, 2000). Phenol compounds have therapeutic, antiseptic, antifungal and bactericidal properties (Ajao an Shounkan, 1984; Rodriguez and Samuels, 1999, Rodriguez and Hesses, 2000).

The levels of phenolic compounds in the two vegetable cowpea genotypes were similar. The mean values of phenols in the two genotypes, *Vigna unguiculata* subspecies *sesquiped alis* **"Akidi anu"** and *Vigna unguiculata subspecies dekintana and mensensis* **"Akidi ani"** were 0.12±0.03% and 0.12±0.05% respectively. Among **"Akidi enu"** and **"Akidi ani"** genotype, accessions AKEB and AKAB had the highest amounts of phenolic compounds respectively.

Table 1: Alkaloids, Flavonoids and Phenols Content of Vegetable Cowpea Genotypes.

Vegetable Cowpea Genotype	Vegetables Cowpea Accession	Alkaloids %	Phenols %	Flavonoids %
Vigna unguiculata	AKER	0.10	0.10	0.41
subspecies *sesquiped*	AKEP	0.10	0.10	0.41
alis **"Akidi enu"**	AKEB	0.10	0.16	0.26
(Climbing Vegetable Cowpea)	Mean ± SD*	0.10±00	0.12± 0.03	0.36± 0.08
igna unguiculata	AKAB	0.08	0.16	0.26
bspecies *dekintana*	AKAS	0.08	0.08	0.30
d *mensensis*	Mean ±	0.08±0.00	0.12±	0.28±
"Akidi ani"	SD*		0.05	0.03
strate Vegetable Cowpea)				

= Standard Deviation

Flavonoids

The term flavonoids embrases all compounds whose structures are based on flavone. Anthocyanins belong to this group of compounds. Anthocyanins are water-soluble natural plant pigments.

They are responsible for the large variety of colours in flowers. Flavonoids exhibit estrogenic, antioxidant and anticancer activities. Flavonoids are antioxidants and free radical scavengers which prevent oxidative cell damage and also protects against all stages of carcinogensis (Salah et al; 1995 and Del Rio eta al 1997) Flavonoids in intestinal tract lower the ri...

Table 2: Saponins and Tannis Content of Vegetable Cowpea Genotypes

Vegetable Cowpea Genotype	Vegetables Cowpea Accession	Saponins mg/100g	Tannis mg/100g
Vigna unguiculata	AKER	0.22	0.30
subspecies *sesquiped*	AKEP	0.13	0.90
alis **"Akidi enu"**	AKEB	0.31	0.39
(Climbing Vegetable Cowpea)	Mean±	0.22±	0.53±
	SD*	0.09	0.32
Vigna unguiculata	AKAB	0.08	0.01
subspecies *dekintana*	AKAS	0.50	0.05
and *mensensis*	Mean ±	0.29±	0.03±
"Akidi ani"	SD*	0.21	0.02
(Postrate Vegetable Cowpea)			

SD* = Standard Deviation

heart disease. As antioxidants, flavonoids provide anti-inflammatory action (Okwu, 2001a, 2001b). The anti-inflammatory properties of some flavonoids have been attributed to their ability to inhibit the production of NO by activated macrophages (Scuro et al 2004).

The flavonoids content of these vegetable cowpea genotypes were high ranging from 0.26% to 0.41% and from 0.26% to 0.30% in *Vigna unguiculata subspecies* sesquiped *alis* **"Akidi enu"** and *Vigna unguiculata* subspecies *dekintana* and *mensensis* **"Akidi ani"** respectively. The levels of flavonoids in this vegetable cowpea were higher than 0.11% reported for *Monodora myristica. Monodora myristica* is being used for the treatment of arthritis in herbal medicine principally due to its flavonoids content (Okwu, 2001b). It therefore follows that humans feeding on vegetable cowpea may not suffer from arthritis.

D. Saponins

Saponins are naturally occurring compounds that are widely distributed in all cells of legume plants. Saponins which derive their names from their ability to form stable, soaplike foam in aqueous solutions, constitute a complex and chemically diverse group of compounds. In chemical terms, saponins contain a carbohydrate moiety attached to a triterpenoid or steroids. Clinical studies have suggested that saponins affect the immume system in ways that help to protect the human body against cancers and also lower cholesterol levels (Sodipo et al, 2000, Shi et al 2004). A high saponin diet can be used in the inhibition of dental caries and platelet aggregation, in the treatment of hypercalciuria in humans, and as an antidote against acute lead poisoning (Shi et al, 2004).

In the legumes studied saponins ranged from 0.13% to 0.31% in *Vigna unguiculata* subspecies *sesquiped* alis **"Akidi enu"** and from 0.08% to 0.50% in *Vigna unguiculalata* subspecies *dekintana* and *mensensis* **"Akidi ani"**. The levels of saponins in the legume studied were higher than 0.01% and 0.03% reported for *monodora myristica* and *Xylopia aethiopica* respectively (Okwu, 2004).

E. Tannins

Condensed tannins also known as proanthocyanidins are polymers or oligomers of flavan-3-ol units. Their structure can be quite variable, among the commonest being a series of four to eight linked (-)-epicatechin units. Tannins have astringent properties, hasten the healing of wounds, and inflammed mucous membrane (Okwu and Okwu, 2004).

Tannin contents in the vegetable cowpea ranged from 0.30 to 0.90% in the climbing genotype and from 0.01 to 0.05% in the prostrate genotype. The climbing genotype of the cowpea contained higher tannins than the prostrate genotype (Table 2). Tannin contents of the climbing vegetable cowpea genotype was within the range obtained for *Spondias mombin* barks. Barks of *Spondias* mombin are used in treating wounds, varicose ulcers hemorrhoids in herbal medicine (Morton, 1987a, 1987b, Stary, 1996, Kozioc and Marica, 1998) because of the presence of tannins in the bark of the plant. *Vigna unguiculata* subspecies *sesquiped* alis **"Akidi enu"** could also be used for the treatment of those infirmities because the levels of tannins in this vegetable cowpea genotype and barks of *S.mombin* are comparable.

4.0 Conclusion

The present study determined the levels of phytochemicals in various assessions of two vegetable cowpea genotypes. The results obtained indicated that the vegetable cowpea are rich sources of alkaloids, flavonoids, saponins, phenols and tannins. Consumption of vegetable cowpea should be encouraged because these phytochemicals have beneficial health effects.

References

Ajoa, A. O and Shonukan, A 1984, Antibacterial effect on aqueous and alcoholic extracts of *S.mombin* and *Alchornea Cordifolia,* International Journal of Crude Drug Research **23** (2), 67 – 72.

Boham, B. A and Kocipai, A. C. (1974) Flavonoids and condensed tannins from leaves of *Hawaiian Vaccinium Vaticulatum* and *V. Calcycinium*. Pacific Science **48**, 458 – 463.

Del –Rio, A; Obdullo, B. G., Castillo, J, Marin F. R. and Ortuno, A. 1997, Uses and Properties of citrus flavonoids. Journal of Agricultural and Food Chemistry **45**,4505 – 4515.

Finar, I. L. 2002, Stereochemistry and the chemistry of Natural Products, 5th edition. Vol. 2. P. 698 – 790.

Harborne, J. B. 1973, Phytochemical methods, Chapman and Hall, London, PP 110 – 113.

Kozioe, M. J. and Marcia, M. J. 1998, Chemical Composition, nutritional evaluation and economic prospects of *Spondias purpurea* (*Anarcardiacea*) Economic Botany **52** (4),373-380.

Morton, J. F. 1987a, *Purple Mombin* Fruits of Warm Climates. Julia F. Morton, Miami Publishers; New York P. 245.

Morton, J. F. 1987b, *Yellow Mombin* Fruits of Warm Climates. Miami Publishers, New York P. 245 – 248.

Obadoni, B. O and Ochuko, P. O. 2001, Phytochemical studies and comparative efficacy of the crude extracts of some homeostatic plants in Edo and Delta States of Nigeria, Global Journal of Pure and Applied Sciences **8**, 203 – 208.

Okwu, D. E. 2001a, Improving the nutritive value of cassava tapioca meal with local spices. Journal of Nutraceutical Functional and Medical Food **3**, 43-51.

Okwu, D. E. 2001b, Evaluation of the Chemical composition of indigenous species and flavouring agents, Global Journal of Pure and Applied Sciences **7**, 455 – 459.

Okwu, D. E. 2004, Phytochemicals and vitamin content of indigenous spices in South-Eastern, Nigeria. J. Sustainable Agric Environ **6**(1), 30 – 37.

Rodriguez, K. F. and Hesses, M. 2000, Antimicrobial activities of secondary metabolites produced by endophytic fungi from *S. Mombin,* Journal of Basic Microbiology **40**(4), 261 – 267.

Rodriguez, K. F. and Samuels, G. F. 1999, Fungal endophytes of *S mombin* leaves in Brazil, Journal of Basic Microbiology **39**(2), 131 – 135.

Salah, N, Miller, N.J. Paganga, G., Tijburg, L. Botwell, G. P, Rice, E and Evans, C. 1995, Polyphenotic flavonols as scavenger of aqueous phase radicals as chain breaking antioxidants, Arch. Biochem. Bioph **2**, 339 – 346.

Shi, J. Arunasalam, K, Yeung D, Kakuda, Y Mittal, G AND Jiang Y 2004, Saponins from edible legumes: Chemistry, Processing and Health Journal of Medicinal Food, **7**(1), 67 – 78.

Sodipo, O. A, Akiniyi, J. A and Ogunbamoru, J. V. 2000, Studies on certain characteristics of extracts of bark of *Pansinystalia macruceras* (K. Schemp) Pierre Exbeille, Global Journal of Pure and Applied Sciences **6**, 83 – 87.

Stray, F 1998, The Natural Guide to Medicinal Herbs and Plants, Tiger Books International, London. P. 12 – 16.

Urquiaga, I and Leighton, F 2000, Plant Polyphenol antioxidants and oxidative stress. Biological Research **33**, 159 – 165.

Van. Burden, T. P and Robinson, W. C. 1981, Formation of complexes between protein and tannin acid. J. Agric Food Chem. **1**, 77 – 82.