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308 Lasani Town, Sargodha Road, Faisalabad - Pakistan
Mob: +92 300 3008585, Fax: +92 41 8815544
E-mail: editorpjn@gmail.com

Compositional Studies of Differently Processed Ornamental Plant Seed Flour: *Caesalpinia pulcherima*

Johnson O. Agbede

Division of Nutritional Biochemistry, Department of Animal Production and Health,
Federal University of Technology, P.M.B. 704, Akure, Nigeria
E-mail: joagbede@yahoo.com

Abstract: Seeds of *Caesalpinia pulcherima*, an ornamental plant, were subjected to different processing methods. Thereafter, the processed samples were chemically characterized with respect to the proximate composition, gross energy and mineral, and amino acid contents. The anti-nutritional factors were also determined. The seed contained: crude protein 353.9gkg⁻¹DM (range: 228 - 491gkg⁻¹DM); ash 122.9gkg⁻¹DM (range: 97 - 183gkg⁻¹DM); crude fibre 48gkg⁻¹DM (range: 34 - 68gkg⁻¹DM); ether extract 146.4gkg⁻¹DM (range 72-179gkg⁻¹DM) and gross energy 19.8MJkg⁻¹ (range; 17.9 - 20.6MJkg⁻¹). Dehull + other variables employed enhanced the crude protein; ether extract and gross energy but reduced the ash and crude fibre contents. The amino acids were generally low when compared with FAO/WHO (1973) recommended pattern and the seeds also contained some nutritional needed minerals. The levels of the anti - nutrients in the seeds were reduced or eliminated completely by dehull + thermal processing methods. It was concluded that the seeds of *C. pulcherima* could be included in dietary formulae for man or monogastric animals especially where protein availability is in short supply.

Key words: *Caesalpinia pulcherima*, ornamental plant, mineral elements

Introduction

Relevant to human existence and living is good nutrition (Agbede, 2000). This is often a major problem in most developing countries of the world. Consequently, the cases of under - nutrition is more rife in these countries. To be able to reduce the adverse effect of hunger and or starvation, it is pertinent that some lesser - known plants are investigated for their nutritive value in human or non-ruminant nutrition. Thus, one of such lesser-known plant that could be considered is an ornamental plant: *Caesalpinia pulcherima*, a grain legume seed plant.

Grain legumes are widely used as cheap protein sources for man and livestock (Oke *et al.*, 1995) and they have been adjudged to be of good nutritional value (Ologhobo, 1980; Del Rosario *et al.*, 1981; Agbede, 2000; Agbede and Aletor, 2003). *Caesalpinia pulcherima* are abundant in Nigeria where they are grown around the houses as ornamental plant. The tender seeds of *C. pulcherima* are known as "Eko omode" in SW Nigeria and children of between ages 4 and 10 years always eat it. It is a pod bearing plant with yellowish or pink colour flowers. It could grow as high as 4 metres and the seeds when dried are hard and brown in colour. Previous study by Agbede (2000) showed that the fresh seeds of *C. pulcherima* contained 34% crude protein, 6.8% crude fibre and 17.3% crude fat. Despite the nutritive value the local children are only consuming the seeds of this ornamental plant without knowing some of its nutrient and anti-nutritional compositions. Thus, this study was designed to provide some analytical information on the *C. pulcherima* when differently

processed with a view to expand its present utilization by man or animals.

Materials and Methods

Seed collection: The fresh pods *Caesalpinia pulcherima* were harvested from the residential quarters of the Federal University of Technology, Akure, Nigeria where they are grown as aesthetic plant. Thereafter, the seeds were manually removed from the pods. While some of the seeds were dried whole in the sun, some were manually dehulled before they were sun dried. Both the dehulled and whole seeds were kept in airtight containers and deep frozen prior to use.

Processing methods adopted: Six kilograms of *C. pulcherima* seeds were divided into eleven parts of about 500g which, corresponded to each of the 10 different processing methods: 1) one- stage cooking (OSC) (ii) two - stage cooking, (TSC) (iii) roasting (RS) (iv) soaking in urea (SU) (v) autoclaving, (AU) (vi) dehull - OSC, (vii) dehull + TSC; (viii) dehull + RS, (ix) dehull + SU and dehull + AU while the 11th part was unprocessed and considered as whole raw seeds.

Determination of the preliminary Cooking

Time/Cooking: Preliminary cooking was carried out to know the cooking time for the whole raw and dehulled *C. pulcherima* seeds. The cooking was done with aluminum pot using one part of either the raw or dehulled seeds to 15 parts of clean water on a Gallenkamp thermostat hot plate. The whole raw seeds

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of *C. pulcherima* got cooked after 37 mins while dehulled seeds cooked after 18 mins. The seeds were considered cooked when they become soft to touch on pressing between the thumb and other fingers. Thereafter, about 500g of the raw or dehulled seeds were subjected to one or two-stage cooking. At the end of the predetermined cooking time, the boiling water was drained and seeds sun-dried.

Dehulling and roasting: Five hundred grams (500g) of the raw seeds were manually dehulled while about 500g of the raw or dehulled seeds were roasted in fine sand and stirred, using the Gallenkamp thermostat hot plate until a characteristic brownish coloured seed was obtained which indicated complete roasting. Thereafter, the seeds were cleaned and cooled.

Soaking in urea and autoclaving: About 500g of the raw or dehulled seeds were soaked for 6 days in 4% urea solution. At the expiration of the 6th day, the urea solution was drained off and the seed rinsed with water before been sun dried. Also, 500g of the raw or dehulled seeds were autoclaved at 105°C at 1.2kg/cm³ pressure for 30 minutes and thereafter sun-dried.

At the end of the processing, all the raw or processed seed samples were finely ground using a laboratory hammer mill (DIETZ, 7311 Dettingen-Teck, West Germany). They were then kept in airtight container and deep frozen (-18°C) prior to chemical analysis and protein quality evaluation.

Proximate composition analysis: The raw and processed samples were analyzed for proximate composition by AOAC (1995) methods. Crude protein was calculated by multiplying the nitrogen content of the samples with the factor of 6.25. Nitrogen free extract content was determined by difference. Each sample was analyzed thrice.

Minerals and gross energy determination: The Na and K contents were determined by Flame photometry (Jenway Ltd, Dunmow, Essex, UK), and P by Vanadomolybdate method (AOAC, 1995). Ca, Mg, Fe, Zn, Cu and Mn were determined after wet digestion with a mixture of nitric, sulphuric and hydrochloric acids, using atomic absorption spectro-photometer (Buck Scientific, East Norwalk, CT06855, USA). The gross energy was determined against thermochemical-grade benzoic acid using a Gallenkamp Ballistic bomb calorimeter (Cam Metric Ltd, Cambridge, England). Each sample was analyzed thrice.

Analysis of anti-nutrients:

Tannin and phytin: Finely milled raw and processed samples (200mg in 10ml of 70% aqueous acetone) were extracted for 2 h at 30°C in water-bath using

Gallenkamp orbital shaker (Surrey, U.K) at 120 revolutions per minute. Pigments and fat were first removed from the samples by extracting with di-ethyl ether containing 1% acetic acid. Thereafter, the total polyphenols (as tannic equivalent) was determined in 0.05ml aliquot in test tubes by the addition of distilled water to make it to 1.0ml, followed by the addition of 0.5ml of the Folin Ciocalteu reagent (Sigma) and then 2.5ml of the sodium carbonate solution. The tubes were vortexed and the absorbance recorded at 725nm after 40min as described by Makkar and Goodchild (1996). The amount of total polyphenols (as tannic equivalent) was calculated from the standard curve. Duplicate samples of each seed type were analyzed.

For the quantification of phytin, eight (8) grams of each finely ground raw and processed samples was soaked in 200ml of 2% hydrochloric acid and allowed standing for three hours. The extract was thereafter filtered through two layers of hardened filter paper. 50ml of the filtrate was pipette in duplicate into 400ml capacity beakers before the addition of 10ml 0.3% ammonium thiocyanate solution as an indicator, and 107ml of distilled water to obtain the proper acidity (pH 4.5). The solution was then titrated with a standard iron chloride (FeCl₃) solution containing 0.00195gm Fe/ml until a brownish yellow colour persists for 5 minutes. Phytin-Phosphorus was determined and phytin content was calculated by multiplying the value of phytin-Phosphorus by 3.55 (Young and Greaves, 1940). Each milligram of iron is equivalent to 1.19 milligrams of phytin-Phosphorus. Duplicate samples of each seed type were analyzed.

Lectin (haemagglutinin) and trypsin inhibitor activity (TIA):

The lectins in raw or processed seeds were extracted from the defatted seed flours by the method of Huprikar and Sohoni (1965). The lectin titres of the extract were subsequently determined using 0.25% saline - washed trypsinized rabbit red blood cells in a two-fold serial dilution technique of Kabat and Mayer (1961) while the nitrogen content of the extract was determined by micro-kjedahl method (AOAC, 1995). Duplicate samples of each seed type were analyzed.

The TIA was assayed in terms of the extent to which an extract of the defatted flour inhibit the action of Bovine Trypsin (EC 3.4. 21.4) on the substrate benzoyl-DL-arginine-p-nitroanilide (BAPNA) hydrochloride (Kakade *et al.*, 1969) as modified by Smith *et al.* (1980). Raw or processed seed (1g) each were extracted continuously at ambient temperature for 3 hours with 50ml, 10mM NaOH using a mechanical shaker (Gallenkamp orbital shaken Surrey, UK). The pH of the resulting slurry was adjusted to 9.4 - 9.6 with 1M NaOH. After extraction, the suspension was shaken and diluted with distilled water such that 1cm³ of the extract produced trypsin inhibition of 40 - 60% at 37°C. The respective dilutions were noted

Table 1: Proximate composition (gkg⁻¹DM) and Gross energy (MJkg⁻¹) of differently *Caesalpinia pulcherima*

Treatment	Crude protein	Ash	Crude fibre	Ether extract	N-free extract	Gross energy
Whole raw seed (WRS)	336.0	123.0	68.0	173.0	300.0	20.4
One stage cooking (OSC)	294.0	106.0	61.0	133.0	406.0	20.1
Two stage cooking (TSC)	264.0	93.0	57.0	126.0	460.0	19.4
Roasting (RS)	228.0	102.0	54.0	146.0	470.0	19.4
Soaking in urea (SU)	300.0	237.0	48.0	163.0	252.0	20.1
Autoclaving (AU)	239.0	99.0	54.0	72.0	536.0	17.9
Dehull + OSC	491.0	99.0	37.0	179.0	194.0	20.6
Dehull + TSC	454.0	97.0	37.0	158.0	254.0	20.1
Dehull + RS	482.0	100.0	40.0	135.0	243.0	20.3
Dehull + SU	373.0	183.0	34.0	171.0	239.0	19.5
Dehull + AU	432.0	113.0	38.0	154.0	263.0	20.2
Mean	353.9	122.9	48.0	146.4	328.8	19.8
Std dev.	97.8	45.5	11.5	30.2	116.7	0.8
Coefficient of variation	27.6	37.0	24.0	20.6	35.5	4.0

and in this assay dilution of 10 were suitable. Consequently, TIA was calculated in terms of mg pure trypsin (Sigma type III, lot 20 H0868) per gram as weighed (Smith *et al.*, 1980).

$TIA = 2.632.D. A_t / S$ mg pure trypsin inhibited g⁻¹ sample
Where D is the dilution factor, A_t is the change in absorbance at 410nm due to trypsin inhibition per cm³ diluted sample extract, and S is the weight of the sample. Duplicate samples of each seed type were analyzed.

Hydrogen cyanide (HCN): Eight grams (8g) of the raw or differently processed seeds was dissolved in 0.1 M H₃PO₄ and blended with home blender for 2 - 3 minutes followed by a 2M H₂SO₄ hydrolysis at 100°C for 50 minutes. This hydrolytic process was followed by the reaction with chloramine-T/pyridine barbituric acid (Konig Reaction) as developed by Bradbury *et al.* (1991). Potassium cyanide (KCN) dried over concentrated H₂SO₄ was used to calibrate the standard curve from a stock solution containing 75mg KCN per 100ml. Duplicate samples of each seed type were analyzed.

Amino acid analysis: For the amino acids determination, the raw seeds of *Canavalia ensiformis* or *Mucuna pruriens* (50-75mg) were hydrolyzed by refluxing for 24 hours in a heating block previously heated at 110±1°C. The hydrolysate was cooled and quantitatively transferred to a -50ml flask and diluted to volume with water. After filtration, a 10ml aliquot of the filtrate was heated in a rotary evaporator (40°C) to remove excess acid before analysis using high-performance liquid chromatography (HPLC) with a Varian HPLC system (Palo Alto, CA) and a Shimadzu RF-535 Fluorescence detector (Tokyo, Japan) set at an excitation wavelength of 325nm and an emission wavelength of 465nm. Separation was achieved in adsorbosphere OPA-HR (150 x 4.6 mm) column (Alltech, Carnforth, UK). The

mobile phase was 1,4-dioxan and 2-propanol (HPLC grade). Duplicate samples of each seed type were analyzed.

Results and Discussion

Table 1 shows that the crude protein (CP), ether extract (EE) and gross energy (GE) contents of *Caesalpinia pulcherima* were enhanced by dehulling + other processing variables. On the contrary, the ash content and in some cases, the crude fibre (CF) were reduced by dehulling + other processing variables. In general, there were wide variations between the raw and processed samples for the proximate compositions as evident in the CV values obtained (21 - 37%). With regards essentially to the CP (353.9gkg⁻¹DM), EE (146.4gkg⁻¹DM) and GE (19.8 MJ kg⁻¹) and low CF (48gkg⁻¹DM), *C. pulcherima* seeds could be an important source of nutrients in foods for man and non-ruminants in developing countries where hunger or starvation is prevalent. The values of CP, ash, EE and GE obtained in this study were higher than those reported for some varieties of cowpeas (Oke *et al.*, 1995), *Manihot glaziovii* (Odetokun and Ayesanmi, 1998) and more recently, *Mucuna pruriens* and *Canavalia ensiformis* (Agbede and Aletor, 2003). It is also conceivable that dehulling + other processing techniques would be preferred for the processing of the seeds.

The study further showed that the differently processed *C. pulcherima* contained some macro and micro mineral elements (Table 2). Higher levels of these mineral elements were found in the raw seeds over the processed seeds, thus corroborating the earlier reports of Apata and Ologhobo (1990) and Agbede and Aletor (2003). Also, it appears that Calcium, Magnesium and Zinc were more concentrated in the cotyledon than the whole seeds. However, the values obtained in these studies fell within the range reported for some gain

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Table 2: Mineral composition of differently processed *Caesalpinia pulcherima*

Treatments	Major minerals (gkg ⁻¹)					Minor minerals (mgkg ⁻¹)			
	Na	K	Ca	Mg	P	Zn	Mn	Fe	Cu
Whole raw seed (WRS)	19.6	11.6	9.7	5.3	7.8	1.26	ND	145.9	ND
One stage cooking (OSC)	17.0	6.7	5.2	1.5	4.7	1.21	ND	68.4	ND
Two stage cooking (TSC)	12.6	6.5	4.4	0.8	3.2	1.07	ND	60.7	ND
Roasting (RS)	12.2	7.8	3.9	2.3	4.8	0.82	ND	83.1	ND
Soaking in urea (SU)	12.4	7.1	3.3	1.7	2.4	1.15	ND	54.9	ND
Autoclaving (AU)	13.1	9.8	3.3	1.6	4.9	1.23	ND	53.0	ND
Dehull + OSC	18.5	5.6	6.4	3.6	9.7	2.49	ND	91.8	ND
Dehull + TSC	14.9	5.3	4.7	3.4	8.4	2.24	ND	73.8	ND
Dehull + RS	16.7	9.5	6.4	5.3	6.2	2.06	ND	28.7	ND
Dehull + SU	7.8	8.9	5.3	1.9	1.9	1.85	ND	54.9	ND
Dehull + AU	5.4	9.2	8.1	1.3	1.2	1.99	ND	53.0	ND
Mean	13.7	8.0	5.5	2.6	5.0	1.58	-	69.8	-
Std Dev.	4.3	2.0	2.0	1.6	2.8	0.56	-	30.4	-
CV	31.4	25.0	36.5	61.5	56.0	35.4	-	43.6	-

ND = Not Detected

Table 3: Amino acid profile (g/16gN) of *Caesalpinia pulcherima*

Amino acid	<i>C. pulcherima</i>	FAO/WHO (1973) recommended pattern	Whole egg*
Aspartic acid	8.37		
Serine	9.99		
Glutamine	5.46		
Histidine	2.95		2.4
Glycine	2.89		
Threonine	3.10	4.0	5.1
Arginine	7.61		6.1
Alanine	3.40		
Methionine	1.59		3.2
Cystine	1.27		1.8
Cystine + methionine	2.86	3.5	5.0
Isoleucine	3.58	4.0	5.6
Leucine	5.08	7.0	8.3
Lysine	1.63	5.5	6.3
Phenylalanine	6.28		5.1
Tyrosine	4.04	6.0	4.0
Valine	6.03	5.0	7.6

*Cited by Robinson (1987)

legumes (Oke *et al.*, 1995; Agbede, 2000). The Ca, Mg and P required for bone formation are relatively low in *C. pulcherima* seeds. Conceivably, dietary formulation that would be based on these seeds would require supplementation with either seeds that are high in these mineral elements or be provided as part of the diet, so as to avert the precipitation of deficiency symptoms.

Table 3 shows the amino acid profile of *C. pulcherima* seed flour in comparison with FAO/WHO (1973) recommended pattern and whole egg (Robinson, 1987). With exception of valine, the amino acid contents of *C. pulcherima* was lower than the FAO/WHO (1973) recommended pattern while only phenylalanine and histidine compared favourably with that of whole egg (Robinson, 1987). Thus by implication, dietary formulae

based on *C. pulcherima* will require amino acid supplementation, especially the essential amino acids (EAAs) such as methionine, leucine and lysine.

Table 4 shows that phytin and phytin-P were completely removed by dehull + two stage cooking, lectin by all the processing variables (except roasting), trypsin inhibitor activity (TIA) by all the processing variables (except raw seed + cooking), tannic acid by dehull + autoclaving and HCN by dehull + other processing variables. Thus, the relatively high CV values (36.3 - 187.4%) suggest that the processing variables employed in this study enhanced the inactivation of all the anti-nutritional factors quantified to a large extent. Generally, anti-nutritional factors have been reported to have the capable of retarding growth (Martinez *et al.*, 1995) and lowered

Table 4: Processing effects on some anti-nutritional constituents of *C. pulcherima* seed flour

Treatments	Phytin (mg/100g)	Phytin- phosphorus (mg/100g)	Lectin (Hu/mg)	Trypsin inhibitor activity (mg/g sample)	Tannic acid (g/100g)	HCN (mg kg ⁻¹)
Whole raw seed (WRS)	33.8	9.5	1	92.6	33.8	17.2
One stage cooking (OSC)	23.5	6.6	ND	1.6	23.5	8.6
Two stage cooking (TSC)	16.1	4.5	ND	0.9	16.1	5.7
Roasting (RS)	33.8	9.5	2	ND	33.4	5.7
Soaking in urea (SU)	28.4	8.0	ND	ND	16.5	ND
Autoclaving (AU)	33.8	9.5	ND	ND	28.4	12.2
Dehull + OSC	19.8	5.6	ND	ND	19.8	ND
Dehull + TSC	ND	ND	ND	ND	19.1	ND
Dehull + RS	27.2	7.7	ND	ND	27.2	ND
Dehull + SU	8.2	2.3	ND	ND	8.2	ND
Dehull + AU	16.5	4.6	ND	ND	ND	ND
Mean	21.1	6.8	1.5	24.3	22.6	9.9
Std Dev.	8.8	2.5	0.7	45.5	8.2	4.9
CV %	36.5	36.8	46.7	187.4	36.3	49.5

CV = Coefficient of variation; ND = Not Detected

digestibility and absorption of dietary nutrients (Pusztal *et al.*, 1995). Thus, by implication, for *C. pulcherima* seeds to be incorporated in human or monogastric feeds, there is a need to dehull and subject it to other thermal processing techniques as shown in this study.

Conclusion: The study shows that the seeds of *C. pulcherima*, an ornamental plant, could be used as feeding material for human or non-ruminant animals judging from the high protein content, ether extract and energy content and low crude fibre. Though low in mineral contents, its consumption with ingredients high in the macro and micro mineral elements would enhance its utilization. The seeds contained some anti-nutritional factors but dehulling + thermal processing would help to reduce or completely eliminate some of the anti-nutritional factors to sub lethal level. It is therefore suggested that in most countries where protein under - nutrition is prevalent, the use of processed *C. pulcherima* seed flour could be used for man and/or monogastric animals feeding.

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