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Full Length Research Paper

# Chemical composition and nutritional evaluation of a cowpea protein concentrate

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Cowpea protein concentrate was prepared by isoelectric precipitation, under optimum conditions determined in a previous study. The concentrate contained 86.90 % of the flour total protein, and had 85.82 protein. Sugars and ash were in lower contents in the concentrate compared to the flour, while Fe and Zn contents were not significantly (p> 0.05) different. Phytates and polyphenols were eliminated in abundance during protein extraction and presence of trypsin and  $\alpha$ -amylase inhibitors in the concentrate suggested application of an adequate heat treatment before its use in food products. The concentrate in vitro protein digestibility was high (86.81-88.74 %), and had a balanced essential amino acid composition compared to the FAO/WHO reference pattern. Tryptophan and total sulphur amino acids were first and second limiting amino acids. The predicted protein efficiency ratio, essential amino acids index and biological value indicated the good quality of cowpea protein concentrate, although elimination of tryptophan and sulphurrich proteins during protein extraction affected the chemical score and protein digestibility corrected amino acid score.

Keywords: cowpea, protein concentrate, antinutritional factors, protein quality

#### INTRODUCTION

There is an increasing world demand of less expensive proteins with good nutritional and functional properties, particularly in developping and under-developped countries where the supply of food of animal origin is limited due to non-availability and high cost (Cheftel *et al.*, 1985). This situation result from constant increase of the human population and growing interest for protein to industry for application in food and non food markets.

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Legumes are connsidered as poor man's meat. They are generally rich in protein (18-25 %), and good sources of minerals and vitamins (Tharanathan and Mahadevamma, 2003). In addition, consumption of legumes has been related to many beneficial physiological effects in controlling and preventing various metabolic diseases such as diabetes mellitus, coronary heart disease and colon cancer (Simpson *et al.*, 1981). Therefore, use of grain legumes for food is restricted by their beany flavor and the presence of antinutritional and toxic factors. Traditional processing techniques such as soaking, cooking, sprouting or roasting have limited effects on elimination of antinutritional factors, and sometimes could decreased protein quality and affected certain functional



Figure 1. Schematic of cowpea protein concentrate preparation

properties (Friedman, 1992; Yusuf, Ayedun and Sanni, 2008). On the other hand, processing techniques employed during protein extraction are known to be effective in detoxification of seed material. Futhermore, protein extracts have superior functional properties than legumes flours, and are extensively used in industry as nutritional and functional ingredients (Neto *et al.*, 2001).

Cowpea (*Vigna unguiculata*) is a drought tolerant food crop, well adapted in varieties of climates and soils. This crop is widely cultivated throughout the tropics and subtropics, particularly in west and central Africa, with an annual production of 3 millions tones (Onyenekwe *et al.*, 2000). Its grains are used to prepare many traditional foods such as moin-moin or akara. cowpea grains have been used to fortify cereal-based weaning foods, in which they formed complementary amino acid profiles and improved protein quality (Bresani, 1985). Also, McWatters *et al.* (2003) prepared biscuits from cowpea composite flour, with good sensory quality.

Isoelectric precipitation is the most comon technique for extracting protein from grain legumes in food industry. This technique involves the variation of physico chemical parameters which affect product and protein yields (Berot and Davin, 1996). Our previous study reported the optimum combination of these parameters for the preparation of protein concentrate from cowpea seed flour (Mune *et al.*, 2008). In the present study, the chemical composition and the nutritional potential of the concentrate were investigated, in view of its possible use as a nutritional ingredient in the food industry.

#### MATERIALS AND METHODS

#### Materials

Cowpea seeds were purchased from Mokolo market (Yaoundé, Cameroon). The seeds were handpicked and stored in polyethylene bags in the refrigerator (~4°C) until used.

#### Methods

#### Preparation of cowpea flour

Cowpea seeds were washed and rinsed in deionised water at room temperature ( $25\pm2$  °C). They were soaked for 1 h in water at room temperature ( $25\pm2$  °C), dehulled and dried in an air convection oven at 50 °C for 72 h. The dried seeds were ground into flour, passed through a 150 µm mesh sieve, stored hermetically in polyethylene bag in a refrigerator at about 4 °C.

#### Preparation of cowpea protein concentrate

Cowpea protein concentrate was prepared by the isoelectric precipitation method (Figure 1) as described by Mune *et al.* (2008). An aliquot (10 g) of cowpea flour was mixed with 100 ml of NaCl solution (0.15 M) and stirred at 35  $^{\circ}$ C for 120 minutes. The pH was adjusted to

Table 1.	Chemical	composition o	f cowpea flou	ur and protein	concentrate	(dry weight basis) <sup>a</sup>

	Flour	Protein concentrate
Nutritional factor (g/ 100 g)		
Moisture	5.77 ± 0.21 b	8.88 ± 0.46 a
Crude protein	21.95 ± 0.00 b	85.82 ± 1.08 a
Non-protein nitrogen	1.62 ± 0.04 a	1.63 ± 0.05 a
Protein nitrogen	20.33 ± 0.00 b	84.19 ± 1.08 a
Crude lipid	1.21 ± 0.09 b	2.39 ± 0.02 b
Fibre (NDF)	5.75 ± 0.15 a	1.73 ± 0.37 b
Starch (by difference)	57.01 ± 1.47 a	8.89 ± 1.48 b
Total sugars	9.41 ± 0.39 a	1.11 ± 0.00 b
Reducing sugars	0.30 ± 0.01 b	0.75 ± 0.06 a
Ash	4.67 ± 0.14 a	1.79 ± 0.38 b
Minerals (mg /100 g)		
lron Zinc	19.39 ± 1.33 a	19.50± 1.03 a
	9.95 ± 2.15 a	0.20 ± 0.31 a

<sup>a</sup> Means followed by different letters (a-b) in the same line are significantly (p< 0.05) different

9.91 using a Hanna Model HI 8521 pH-meter (Hanna Instruments, Portugal), and the mixture was further stirred at 4 °C for 30 min. The resultant slurry was then centrifuged at 2000 g for 30 min at 4 °C using a Jouan Model GR 4.11 centrifuge (Jouan, Saint Nazaire, 44600, France). The pellet obtained after recovering the supernatant, was dissolved in the initial NaCl solution at the above liquid to solid ratio under stirring. The pH was adjusted to the initial value and the slurry stirred for 30 min at 4 °C and then centrifuged as previously explained. The resultant supernatants of the two alkaline extractions were combined and one-part volume of 95 % (v/v) ethanol added. The pH was adjusted to 4.5 under stirring and the precipitated proteins were recovered by filtration under vacuum using a whatman N° 1 filter paper. The protein concentrate was dried at 50 °C for 48 h in an air convection oven, ground and passed through a 150 µm mesh sieve.

#### Proximate composition

Moisture, ash, total lipids and crude protein (Nx6.25) were determined according to AOAC (1990) methods. Dietary fibre was analysed using neutral acid detergent (Goering, and Van Soest, 1970). Non-protein nitrogen (NPN) was determined by the method of Bhatty and Finlayson (1973) as modified by Naczk *et al.* (1985) by which proteins were precipitated with 10 % trichloroacetic acid (TCA) solution, and the resultant non-protein nitrogen was determined according to the Kjeldahl procedure. Iron and zinc contents were determined by atomic absorption spectrophotometry using a Unicam

Model 969 atomic absorption spectrophotometer (Unicam Limited, York Street, Cambridge, CB1 2PX, United Kingdom), after digestion of 0.25 g sample with 6 ml of concentrated nitric acid at  $150 \pm 5$  °C for 6 h according to Laurent (1981). Total simple sugars were determined by the anthron method (Montreuil *et al.*, 1981) and reducing sugars by the 3,5-dinitrosalicylic acid (DNSA), following the sugars extraction in hot 80 % (v/v) ethanol (Cerning, and Guilhot, 1973). Starch (+maltodextrins) was determined by difference.

## Antinutritional factors

#### Polyphenols

Polyphenol content was determined according to Singleton and Rossi (1965) as gallic acid equivalents, after their extraction in 70 % (v/v) aqueous acetone (Shahidi, and Naczk, 1989).

#### Phytate

Phytate content was determined based on complex formation of phytic acid and Fe(III)-ion at pH 1-2 (Stone *et al.*, 1984), after extraction in 1.2 % HCl solution containing 10 % Na<sub>2</sub>SO<sub>4</sub> as described by Thompson and Erdman (1980). An excess of Fe(III)-ion present in the solution would react with thiocyanate ion to form a characteristic pink complex, Fe(SCN)<sub>3</sub>. The optical density at 465 nm was measured (Itabashi, 1985), and an inverse linear relation was found for phytate

Amino acid	Flour	Protein concentrate	FAO/WHO (1991)	
Histidine	3.60	3.45	1.9	
Isoleucine	4.30	4.49	2.8	
Leucine	8.61	9.45	6.6	
Lysine	6.81	6.50	5.8	
Thréonine	4.77	4.25	3.4	
Tryptophan	0.97	0.27	1.1	
Valine	5.54	5.46	3.5	
Méthionine	0.49	0.22		
Cysteine	1.61	1.24		
Total sulfur amino acids	2.10	1.46	2.5	
Tyrosine	1.44	2.13		
Phénylalanine	4.76	5.58		
Total aromatic amino acids	6.20	7.71	6.3	
Total essential amino acids	42.90	43.04	33.9	
Aspartic acid	12.63	13.03		
Serine	6.48	6.55		
Glutamic acid	15.04	15.56		
Proline	4.68	4.91		
Glycine	7.51	6.63		
Alanine	7.61	6.63		
Arginine	3.15	3.65		
Total non-essential amino acids	60.70	60.41		
Leucine / isoleucine ratio	2.00	2.10	2.36	

Table 2. Amino acid composition of cowpea flour and protein concentrate (g/16 g N)

concentration from 40 to 200 nmol/L.

#### Trypsin inhibitor activity

Trypsin inhibitor activity was determined based on the method described by Lgari et al. (2002) using soluble casein as substrate, after the extraction of protease inhibitor in a 0.02 M pH 8.0 Tris (trihydroxyaminomethan) buffer solution containing 0.02 M CaCl<sub>2</sub> (Griffiths, 1984). The assay mixture contained 2 ml of inhibitory solution, 2 ml of trypsin solution [2.5 mg of bovin trypsin (SIGMA, 15,900 u/mg) in 25 ml of 0.01 N HCl], and 5 ml of soluble casein solution (0.2 % in Tris-HCl buffer, pH 8.2, 0.05 M). Before the addition of the substrate, the mixture was incubated for 30 min at 37 °C, to allow the binding of the inhibitors to the protease. Casein solution was then added and the reaction incubated for 20 min at 37 °C. The reaction was stopped by addition of 2.25 ml of 25 % TCA solution. The samples were centrifugated at 2000 g for 30 min at 4 °C. A blank set was prepared by adding TCA before the addition of the protease solution. A control set was prepared in which inhibitory solution was deleted from the assay mixture. The released tryptophan was determined in an aliquot of the supernatant by the ninhydrin reaction as described by Panasuik et al. (1998). The trypsin inhibitory unit (TIU) was expressed as that

producing the inhibition of 1 mg pure trypsin.

#### α-Amylase inhibitor activity

α-Amylase inhibitory activity was determined based on the method described by Bandary et al. (2008), after extraction of a-amylase inhibitors as described by Lonstaff and Mc Nab (1991). The assay mixture contained 2 ml of inhibitory solution, 50 µL of porcine pancreatic α-amylase[5 mg/ml in Tris-HCl 0.05; pH 6.9, containing 0.01 M CaCl<sub>2</sub>], 2 ml of the Tris-HCl buffer pH 6.9, 2 ml of soluble starch (5 mg/ml in Tris-HCl buffer pH 6.9) soaked in boiling water for 5 min. Before the addition of the substrate, the mixture was incubated for 60 min at 37 °C, to allow binding of the inhibitors to the enzyme. Starch was added and the reaction incubated at 37 °C for 20 minutes. The reaction was stopped by addition of 6 ml ethanol 90 % (v/v), and the test tubes plunged in and ice bath for 10 minutes. The samples were centrifuged at 2000 g for 15 min. The release of reducing groups (calculated as maltose equivalents) was determined in an aliquot of the supernatant by the 3,5-DNSA method. A blank set was prepared by adding ethanol 90 % (v/v) before the addition of the  $\alpha$ -amylase solution. A control set was prepared in which inhibitory solution was deleted from the assay mixture. The  $\alpha$ -amylase inhibitory unit

Materials	Antinutritional fa	ictors	IN VITRO protein Digestibility (%)			
	Polyphenol	Phytate	α-AI <sup>D</sup>	TI	Pepsin-	Pepsin-
	(mg/100 g)	(g/100 g)	(AIU/g)	(TIU/g)	Trypsin	Pancreatin
Cwpf	162.98 ± 7.61 a	1.01 ± 0.03 a	1.35 ± 0.13 a	5.32 ± 0.20 a	51,53 ± 0,00 b	59,58 ±1,62 b
Cwpc	122.26 ± 7.46 b	0.71 ± 0.01 b	1.40 ± 0.13 a	2.93 ± 0.31 b	86,81 ± 2,41 a	88,74 ±0,63 a

Table 3. Antinutritional factors and in vitro protein digestibility of cowpea flour and protein concentrate a

<sup>a</sup> Cwpf, cowpea flour; Cwpc, cowpea protein concentrate;  $\alpha$ -AI,  $\alpha$ -amylase inhibitor; TI, trypsin inhibitor; Mean in the same column followed by different letters (a-b) are significantly (p< 0.05) different

<sup>D</sup> Expressed as mg pure trypsin inhibited (TIU) per g sample (in dwb)

<sup>c</sup> Expressed as 1 µmol maltose equivalents/min inhibited (AIU) per g sample (in dwb)

(AIU) was defined as one unit of  $\alpha$ -amylase activity (1  $\mu$ mol maltose/min) inhibited.

#### Amino acids

Amino acids were determined using a BECKMAN 6300 amino acid analyser according to the method of Spackman *et al.* (1958). Hydrolysis of samples was performed in the presence of 6 M HCl, trifluoroacetic acid (TFA, 2:1, v/v) and 5 % thioglycolic acid, for 24 h at 100 °C. Cystein was determined by the method of Beveridge *et al.* (1974) using 5,5'-dithio-2-nitrobenzoate (DTNB), and the absorbance read at 412 nm. Tryptophan was determined by ultraviolet molar absorption coefficient of each sample in a Tris-Gly buffer (0.086 M Tris, 0.09 M glycine, 0.04 M EDTA, pH 8) containing 8 M urea as described by Pace *et al.* (1995).

#### In vitro protein digestibility and available lysine

*In vitro* digestibility was determined using trypsin-pepsin (Chavan *et al.*, 2001) and pepsin-pancreatin (Akeson, and Stahman, 1964) enzymatic systems using bovin trypsin (SIGMA, 15900 U/mg), pepsin (SIGMA, 1mAnson-E/mg) and pancreatin (SIGMA, 4 usp). The nitrogen content of the TCA-soluble matter was determined by the Kjeldahl method (AOAC, 1990). Protein digestibility was expressed as the percentage of the soluble TCA 10 % nitrogen, with respect to the total nitrogen content of the undigested sample.

Available lysine (g/16 g N) was determined by dye binding procedure using 1-phenylazo-2-naphtol-6-sulfonic acid (Orange 12), as described by Hurrell *et al.* (1979). A sample aliquot containing 15 mg of 'Arg + His + Lys' was mixed with 4 ml of half saturated sodium acetate, and 40 ml of Orange 12 reagent were added directly for 'Arg + His + Lys' determination; or after propionylation of lysine with propionic anhydride for 'Arg + His' determination. Difference in absorbance between the two at 475 nm after 2h reaction in the dark at ambient temperature was used for calculating reactive lysine. Absorbance measurements were performed using a Spectronic Model 601 spectrophotometer (Milton Roy company, Rochester, NY, 14625, USA).

#### Determination of nutritional parameters

Nutritional parameters were determined on the basis of the amino acid profiles. A chemical scoring of amino acids was calculated using the FAO/WHO (1991) reference pattern. Essential amino acid index (EAAI) was calculated according to Oser (1959) using as standard the amino acid composition of the whole egg protein published by Cheftel *et al.* (1985). Protein efficiency ratio (PER) was estimated according to the regression equations developed by Alsmeyer *et al.* (1974), as given below:

Biological value was calculated according to Oser (1959) using the following equation:

BV= 1.09xEAAI - 11.7

#### Statistical analysis

Results are expressed as mean value ± standard deviation of three different determinations, except for amino acid contents. The data were statistically analysed by the Student-Newman-Keuls test. The computer software used in this study was SPSS (version10.1, 2000, SPSS Inc., USA).

#### **RESULTS AND DISCUSSION**

# PREPARATION OF COWPEA PROTEIN CONCENTRATE

The chemical composition of cowpea flour is presented in

Materials	Chemical	Limiting am	ino acids	PDCAAS	EAAI	PER		BV	Available lysine
	score (%)	First (CS, %)	Second (CS, %)	(%)	(%)	PER <sub>1</sub>	PER <sub>2</sub>	(%)	(g/16 g N)
Cwpf	84.00	Cys + Met (84 %)	Trp (88.18 %)	50.05	74.27	3.02	3.29	69.26	1.00 ± 0.04 b
Сwpc	24.55	Trp (24.55 %)	Cys+Met (58.40 %)	21.78	61.85	3.39	3.60	55.71	1.29 ± 0.07 a

Table 4. Nutritional parameters and available lysine of cowpea flour and protein concentrate a

<sup>a</sup> Cwpc, cowpea protein concentrate; Cwpf, cowpea flour; CS, Chemical score; PDCAAS, protein digestibility corrected amino acid score; EAAI, essential amino acid index; PER, protein efficiency ratio calculated according to Alsmeyer *et al.* (1974) equations; BV, biological value; Means in the same column with different letters (a-b) are significantly (p< 0.05) different

Table 1. The high protein content (20.33 %) shows that this flour is a good raw material for the preparation of protein concentrate. Since starch is the major macromolecule (57.01 %), protein concentrate preparation in an industrial scale should take in account starch isolation to improve economical rentability.

Cowpea protein concentrate was obtained with a protein yield (86.90 %) higher than those found by Mwasaru et al. (1999) (36.4-53.5 %), Chavan et al. (2001) for beach pea (67.9-77.3 %), and Sanchez-Vioque et al. (1999) for chickpea (65.9 %). The loss of acidsoluble proteins may in part explain incomplete recovery of protein from the flour. Sanchez-Vioque et al. (1999) observed that 18 % of protein extracted from chickpea flour (80.9 %) in alkaline conditions remained soluble at the isoelectric pH. In addition, protein could be retained in the residue due to complexation with other seed material, particularly fibre. Moure et al. (2002) showed that enzymatic hydrolysis of fibre from chilean hazelnut seeds favoured protein extractability, and Ma (1983) observed that the fibre-rich fraction obtained after protein extraction from oat seed contained 10-20 % protein.

## **Chemical composition**

The chemical composition of cowpea protein concentrate is presented in Table 1. The protein content was in the same range as those reported by Mwasaru et al. (1999), and higher than those of Bambara bean (80.2-81.5 %), benniseed (78.5-80.5 %) and mucuna bean (78.3 %) (Yusuf et al., 2009; Adebowale and Lawal, 2003) protein concentrates. The concentrate (2.39 %) contained two times more lipid than the flour (1.21 %). This concentration of lipids with the protein fraction was also observed by Chavan et al. (2001) and Sanchez-Vioque et al. (1999). It was suggested that polar lipids were extracted in weak-alkaline conditions, and could interact with proteins. The fibre content of cowpea concentrate (1.73 %) was significantly (p< 0.05) lower than that of the flour (5.75 %), and similar to those of beach pea (1.51-1.83 %) and pigeon pea (1.54-1.63 %) protein extracts (Chavan et al., 2001; Mwasaru et al., 1999). Generally, water-soluble sugars and minerals are eliminated in

abundance during protein concentrate preparation. The ash and total sugars contents of the concentrate were significantly (p< 0.05) lower than those of the flour. Therefore, the non-significant (p> 0.05) Fe and Zn contents of cowpea concentrate and flour suggested that divalent cations could be associated to proteins. There is a 93.48 % reduction in the non-reducing sugars content (difference between total sugars and reducing sugars content) of the cowpea protein concentrate (0.36 %), compared to the flour (9.11 %). This suggested abundant elimination of flatus-causing oligosaccharides such as raffinose, stachyose and verbascose, which are non-reducing and found in cowpea seeds by Onwuliri and Obu (2000) and Onyenekwe *et al.* (2000).

## Amino acid composition

The amino acid compositions of cowpea flour and protein concentrate are presented in Table 2. In terms of essential amino acids, cowpea protein concentrate was rich in leucine (9.45 %) and lysine (6.50 %), and poor in tryptophane (0.27 %) and total sulphur amino acids (1.46 %). The major non-essential amino acids were glutamic (15.56 %) and aspartic (13.03 %) acids. Protein concentrate preparation concentrated aromatic amino acids, leucine and isoleucine. The remaining essential amino acids were in lower contents in the concentrate compared to the flour. Sanchez-Vioque et al. (1999) related the lower contents of these amino acids in chickpea protein extracts compared to the flour to the high reduction of albumins, which are rich in lysine, cysteine and methionine. Albumins were probably loosen during isoelectric precipitation, since it has been reported that the acid-soluble fraction of legume protein contain a higher proportion of nutritionally essential sulphur amino acid-rich proteins, than does the acid precipitable fraction (Cerletti et al., 1985; Oomah and Bushuk, 1983).

The quality of proteins as source of amino acids can usually be assessed by comparison with the FAO/WHO (1991) recommended pattern of essential amino acids. Cowpea protein concentrate had higher total essential amino acids than the FAO/WHO (1991) reference pattern. In addition, histidine, isoleucine, leucine, lysine, threonine and valine contents met the FAO/WHO (1991) requirements for infants, while tryptophan and total sulphur amino acids were in non-adequate levels. Generally, higher lysine and leucine, lower tryptophan and sulphur amino acids contents are common to most legume proteins. These results showed that cowpea protein concentrate could be used to complement cereal proteins, which contain high amount of total sulphur amino acids and are low in lysine. The leucine/isoleucine ratio of cowpea concentrate (2.1) was in ideal range suggested by FAO/WHO (1991). Deosthale *et al.* (1970) showed that excess leucine in foods interfered with the utilization of isoleucine and lysine.

# Antinutritionnal factors and in vitro protein digestibility

Antinutritionnal factors and in vitro protein digestibility of cowpea flour and protein concentrate are presented in Table 3. A 25 % reduction in polyphenolic content was observed in cowpea protein concentrate compared to the flour. The polyphenolic content of the concentrate was 122.26 mg/100g. Polyphenolic compounds can interact with protein and reduce their digestibility, as well as alter amino acid availability and functional properties (Lin et al., 1974). In this regards, low levels of polyphenol are desirable. Shahidi et al. (1999) and Chavan et al. (2001) observed a 94 % reduction in polyphenolic content of beach pea protein extracts compared to the flour, and the concentrates contained 50.5-76.4 mg/100 g polyphenol. Cowpea protein concentrate was significantly (p < 0.05) lower in phytate content (0.71 %) than the corresponding flour (1.01 %). Phytates influence the nutritional and functional properties of grain legumes and their derivatives by forming complexes with proteins, amino acids (Reddy and Salunkhe, 1981) and trace minerals (Erdman, 1981). For nutritional application of cowpea protein concentrate, inactivation of enzymes inhibitors by an adequate heat treatment will be required, since 55 % of the trypsin inhibitor activity of the flour was found in the concentrate, and  $\alpha$ -amylase inhibitory activity was not significantly (p> 0.05) different in cowpea flour and concentrate.

*In vitro* protein digestibility of cowpea protein concentrate (86.81-88.74 %) was in the same range than that obtained *in vivo* by Rangel *et al.* (2004) (86.90 %), and those of sweet and bitter lupin (86.90-90.80 %) protein extracts, obtained with a trypsin-pancreatin enzymatic system (EI-Adawy *et al.*, 2001). Protein digestibility of cowpea concentrate was significantly (p< 0.05) higher than that of the flour (51.53-59.58 %). Improvement in protein digestibility was attributed to the denaturation of proteins and the reduction of antinutritionnal factors. Since protease inhibitors are albumins (Richardson, 1991), they were removed in the protein isolation procedures. Sanchez-Vioque *et al.* 

(1999) showed that the dissociation of globulin due to action of pH, facilited their accessibility to digestive proteases, and improved the hydrolysis.

#### Nutritional parameters and available lysine

The nature and quantity of amino acids contained in a dietary protein, determined the efficiency with which an organism could use the protein. Nutritional parameters and available lysine of cowpea flour and protein concentrate are presented in Table 4. The chemical score and the protein digestibility corrected amino acid score (PDCAAS) of the concentrate (24.55-21.78 respectively) were lower than those of the flour (84.00 and 50.05 %, respectively). Based on chemical score, the first and second limiting amino acids of the concentrate were tryptophane and total sulphur amino acids, respectively. Protein efficiency ratio of cowpea protein concentrate (3.39-3.60) was higher than those reported for beach pea (2.75-2.81) and lupin protein extracts (2.22-2.32) (El-Adawy et al., 2001; Chavan et al., 2001). Essential amino acids index (EAAI) and biological value were lower in the cowpea concentrate (66.85 and 55.71 %, respectively) compared to the flour (74.27 and 69.26 %, respectively). This could be attributed to the high reduction of certain essential amino acid content, particularly tryptophan, cysteine and methionine.

Available lysine was significantly (p < 0.05) higher in cowpea protein concentrate (1.29 g/16 g N) compared to the corresponding flour (1.00 g/16 g N). These values remained lower than those obtained by the amino acid analysis (6.81 g/16 g N for the flour and 6.50 g/16 g N for the concentrate). Nevertheless, Walker and Feather (1983) showed that a fraction of non-available lysine could be recovered *in vivo* after acid hydrolysis.

#### CONCLUSIONS

Cowpea protein concentrate prepared under optimum conditions (Mune *et al.*, 2008) does appear to have good potential as a source of low-cost protein, with nutritional quality comparable to those of other grain legume protein concentrates. Presence of enzyme inhibitors indicated that appropriate heat treatment should be necessary before incorporation of cowpea concentrate in food products. The ultimate success of utilizing any seed protein as a food ingredient depends largely on its functional properties. Further work will be conduct to evaluate and improve functional properties of cowpea concentrate, and then promote the use of this seed to alleviate protein malnutrition in developing and under developing countries.

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