ISSN : 0974 - 7508 Volume 9 Issue 10

Natural Products

An Indian Journal

Full Paper

NPAIJ, 9(10), 2013 [403-411]

Effect of temperature and pH on the functional properties of flours and protein isolate from dehulled black crowder cowpea (*Vigna unguiculata*) seeds

Charles C.Osobie[1,2]*, Ijeoma Olawuni[1,2], Angela Nwakudu[1,2], Nwokeke Blessing[1,2], Nwokenkwo Emilia[1,2], Amandikwa Chinyere[1,2]

[1]Department of Food Science and Technology, Federal University of Technology, Owerri (FUTO), Imo State, (NIGERIA)

[2]Department of Food Science and Technology, Imo State Polytechnic, Umuagwo, Ohaji, (NIGERIA)

E-mail: ccharles4@gmail.com

ABSTRACT

This study evaluated the effect of pH and temperature on the functional properties of dehulled Black Crowder Cowpea (BCC) seed flours. The dry seeds were soaked, dehulled and processed into full fat, defatted and protein isolates flours. The functional properties of the flours studied included the Water Absorption Capacity, Emulsion Capacity, Swelling Index and Foaming Capacity. Significant differences ($P<0.05$) existed in the Water Absorption Capacity and Emulsion Capacity of the samples treated with temperatures ranging from 40°C to 100°C, while the same samples given the same temperature treatment were insignificantly different in their Swelling Index and Foaming Capacity. However, the samples, when their pH were adjusted to a range of 4.5 to 14, showed no significant differences ($P<0.05$) in their Water Absorption Capacity, Swelling Index and Foaming Capacity. In general, the BCC seeds were found to compare favourably with other tropical legumes hence, it can find useful application in the food industry especially in food supplementation and in bakery and confectionary products as well as in other possible applications.

KEYWORDS

Black crowder cowpea;
pH;
Temperature;
Functional properties.

INTRODUCTION

Black Crowder Cowpea (BCC) is one of the many tropical species of theses tropical legumes or cowpeas that are used less. It is hardly ever known or utilized a good deal, except among localized communities in the Eastern part of Nigeria where it is known as "Akidi ojii", "Akidienu" or "Akidiani" and eaten in different combinations and forms with other staple foods.

It is prepared in this area as porridge with yam or maize. It is also cooked dehulled or undehulled and made into a spiced paste used to eat Tapioka. Further inquiries would identify other parts of Nigeria where it is consumed. With the knowledge of the profound nutritional benefits and significant functional potentials of these vegetable protein sources, speculations are that consistent research on other tropical legumes like Black Crowder Cowpea could come out with tremendous results that could greatly provide laudable choices for food supplementation and diversification[4].

For the purpose of this work, investigations are directed to the highly variable local cowpea species of

Full Paper

the tropics that are barely recognized or used, using Black Crowder Cowpea (*Vigna unguiculata*) as a case study. This particular work is aimed at providing information on the effect of temperature and pH variations on the functional compositions of the full-fat flour, defatted flour and protein isolate made from the Black Crowder Cowpea.

MATERIALS AND METHODS

Dry seeds of Black Crowder Cowpea (BCC) used in this study were sourced from Akwata market in Enugu, Enugu state, Nigeria. Laboratory equipments and other facilities used in the analyses were obtained from Central Laboratory Service unit of the National Root Crops Research Institute (NRCRI), Umudike, Abia state, Nigeria.

Chemicals and reagents

Chemicals and reagents used in the course of this work were of analytical grade (Analar). They included Sodium hydroxide (NaOH), Sulphuric acid (H2SO4---), Hydrochloric acid (HCl), Boric acid (H_3BO_3), Ethanol (C2H5OH), Hexane (C6H14), Selenium crystals, Methyl red, Bromocresol green, refined olive oil, etc.

Equipments

The equipments and apparatus used in this research work included the Cabolite electric oven, Authur Thomas laboratory mill, Satarious digital analytical balance, general laboratory glass wares (beakers, conical flasks, crucibles, Petri dishes, desiccators, etc.), Gallen Camp electric muffle furnace, Excello Kjedahl apparatus, Colab fume chamber, retort stand, stop watch, thermometer, Satarious digital pH meter, Colab electric centrifuge, manual sieve, etc.

Methods

The features of the dry seeds were determined by the method employed by Fashakin and Fasanya. The raw seeds were selected at random and examined by subjective methods for shape, seed coat texture, seed colour and eye colour. The testa was described as smooth or rough, depending on the appearance to the eye. The degree of attachment of the testa (seed coat) to the cotyledon was described as tough or loose, depending on the ease of separation.

Seed weight

Weight of randomly selected hundred (100) seeds of BCC was determined by weighing on the analytical balance. The average weight per seed was evaluated by dividing the net weight by the number of seeds weighed (100).

Preparation of samples

The Black Crowder Cowpea (BCC) seeds were first processed into full fat flour and defatted flour before the protein was isolated to get the Protein Isolate. The method described by Okezie and Bello was employed. The dry bean seeds were manually sorted to remove stones, residual vegetative components, insect-perforated and shrivelled seeds, and other extraneous materials. The wholesome seeds were used.

Production of full fat BCC flour

The dry seeds of the BCC were first washed in clean water to dislodge adhering dusts and other possible surface contaminants. The wash water was discarded after selectively separating the seeds from it. The seeds were then soaked in another clean water at 1:5 (w/v i.e. bean weight per water volume) ratio for 24hours. The seed coat of the BCC was found to be as thick and adhering as bread fruit's and so was difficult to remove manually. The soaked seed was therefore removed from the soaking water and slightly dried before it was dehulled using a loosely set manual mill as employed in local bread fruit dehulling process. The hulls were thus more easily separated. The dehulled seeds were dried in the oven at 30°C for about 48hours and milled thereafter into flour with the laboratory mill. The flour was then sieved through a 0.5mm sieve to obtain the full fat BCC flour.

Production of defatted BCC flour

The full fat flour sample was soaked overnight (about 18hours) in ethanol at 1:5 (w/v) ratio at room temperature. Thereafter, the extracted flour was filtered from the solvent-oil medium. The defatted flour was then air-dried for about 6hours and pulverized before it was sieved. Part of the defatted flour was then set aside for analysis while the rest were used for the production of Protein Isolate.

Full Paper

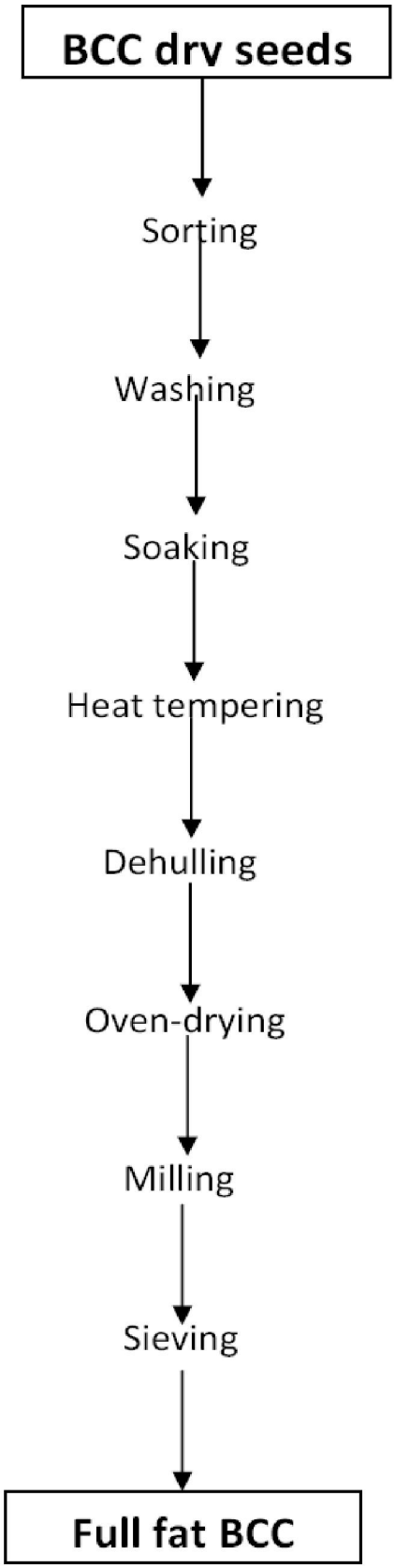

Figure 1 : Flow chart for the production of dehulled full fat BCC flour

Production of the protein isolate

The method described by Okezie and Bello (1988) was used in the protein isolation. About 70g of the defatted flour was mixed with 1400ml of water to form a 1:20 (w/v) ratio of slurry. The pH of the solution was brought to 6.37 and the solution was then allowed to settle for 3hours. The spent residue was separated from the dissolved protein extract by decanting, after which the dissolved protein was centrifuged. The pH of the extracted protein was then adjusted with dilute HCl to its isoelectric point between 4.0 – 4.3. The precipitate formed was afterwards recovered by centrifugation at room temperature and discarding the whey. The resulting curd (Protein Isolate) was then dried under air and cooled in desiccators before it was powdered and sieved.

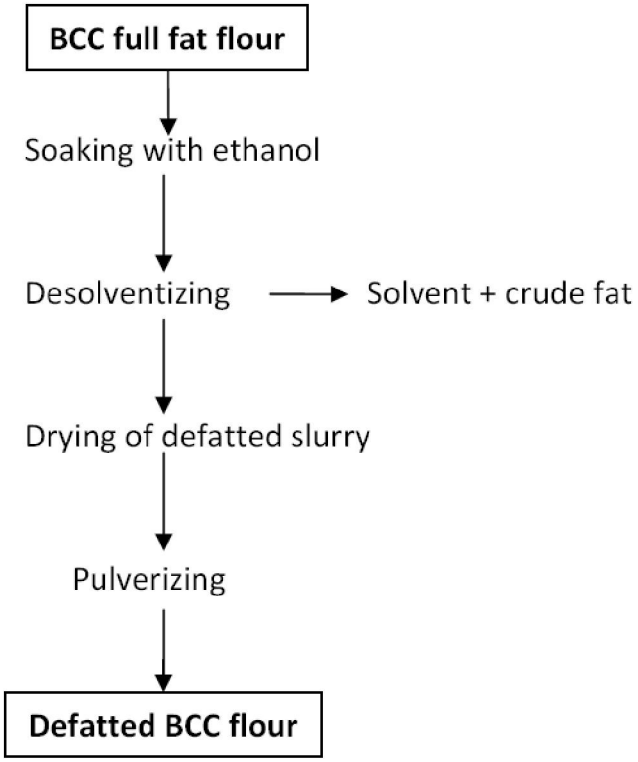

Figure 2 : Flow chart for the processing of full fat BCC flour into defatted BCC flour

Functional properties of flour samples

Bulk density

The bulk density of the flour samples was determined mimicking the method described by James[2]. Separate 10 ml graduated measuring cylinders were gently filled with 5g (W) of each of the samples ensur-

Full Paper

ing that the particles settled at the bottom. Thereafter, the volume (V) occupied by the sample of known weight is recorded.

Bulk density = W/V

Where: W = weight of sample in gram (g); V = Volume of sample in millilitre (ml)

Measurements were reported as means of triplicate determinations.

Emulsion capacity

The method of Okezie and Bello[7] was used. 1g of the flour samples were blended with 10ml distilled water at room temperature for 30 seconds. After complete dispersion, 10ml of refined vegetable oil was added and blended for another 30 s. The mixture was later transferred into a centrifuge tube and centrifuged at 1,600 rpm for 5 min. Emulsion capacity was calculated as:

Emulsion capacity = (EH/WH) X 100

Where: EH = Emulsion Height (Volume of whole solution in the centrifuge tube); WH = Water Height

Swelling index

The swelling index was determined using the method of Okezie and Bello[7]. It was determined as the ratio of height of a unit weight of the sample to the height of the swollen sample left in contact with excess water for 1hour. 1g of the sample was weighed and dispensed into a test tube, leveled, and the height was noted. 10ml of distilled water was added to the sample and the test

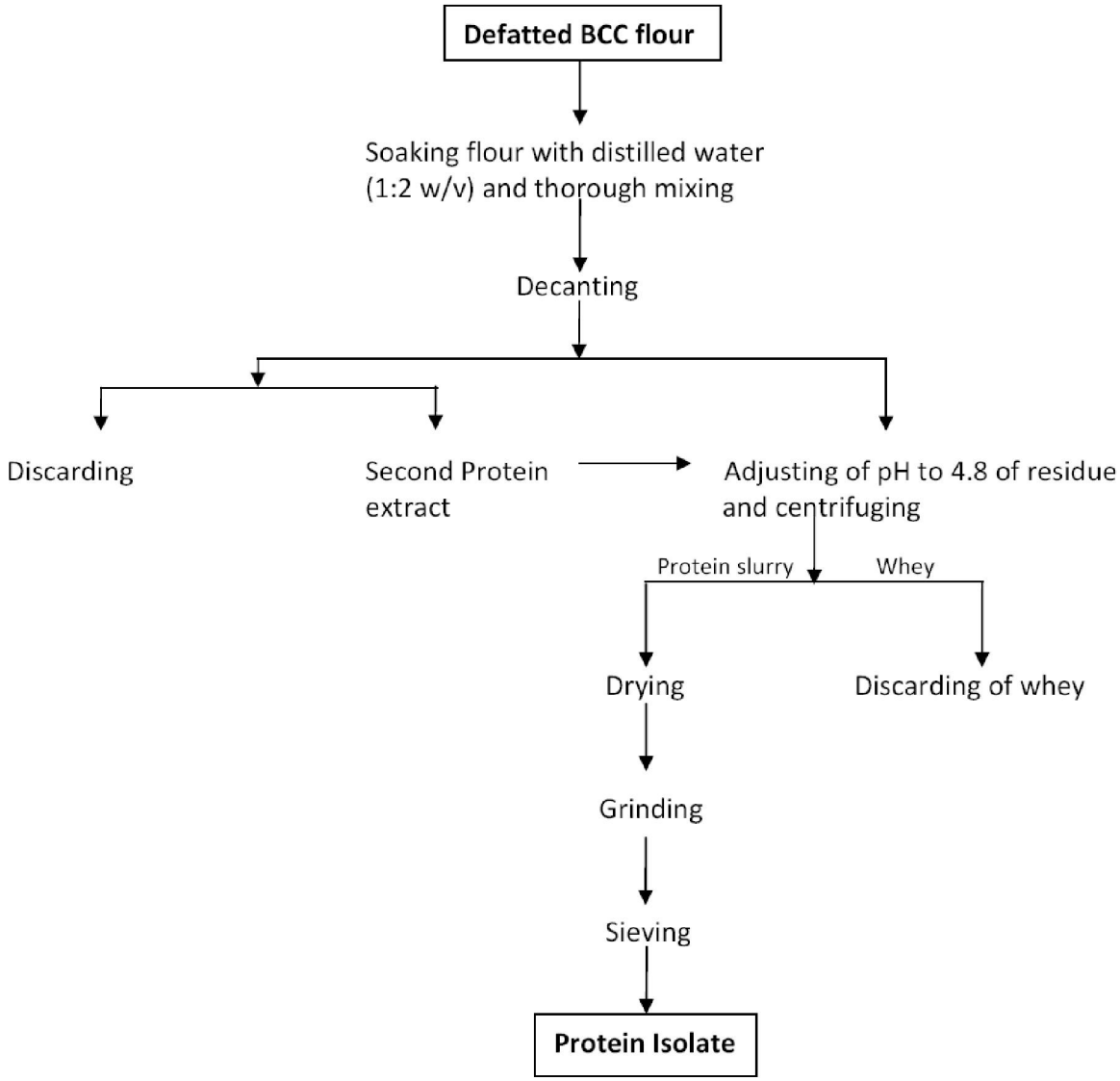

Figure 3: Flow chart showing the production of Protein Isolate from defatted BCC flour

tube was left to stand for 1hour. The height which the sample then occupied was recorded and the swelling capacity was calculated as:

Swelling capacity = H2/H1

Where: H2 = Height occupied by the sample after swelling; H1 = Initial height occupied by sample

Water/Oil absorption capacity (WAC/OAC)

The method of determination of water/oil absorption capacity described by Okezie and Bello[7] was used. 1g of sample was weighed and dispensed into a test tube and 10ml of distilled water/refined vegetable oil was added. The sample was later mixed thoroughly and allowed to stand for 30 min at room temperature. The mixture was centrifuged at 1,500 rpm for 30 min. The volume of free water or oil (the supernatant) was decanted and measured. Water/oil absorption capacity was determined thus:

WAC = (Vw1-Vw2)/mass of sample used

OAC = (Vo1-Vo2)/mass of sample used

Where: WAC = Water absorption capacity; Vw1 = Initial volume of water (10ml);Vw2 = Final volume of water decanted; OAC = Oil absorption capacity; Vo1 = Initial volume of oil (10ml); Vo2 = Final volume of oil decanted

Triplicate results were obtained for each sample and their mean values reported.

Wettability

This was determined as the time (in seconds) taken by a unit weight (1g) of the sample to get completely wetted on a sample of distilled water in a beaker under laboratory conditions. This method was described by Okezie and Bello[7]. A 600ml capacity clean beaker was used to measure and retain about 500ml of water. With the aid of a retort stand, a set-up was arranged such that a clean (dry) test tube was clamped in an inverted position over the water in the beaker. The clamped position was adjusted such that the distance from the mouth of the test tube to the surface of the water in the beaker was exactly 10cm. Both the water in the beaker and the clamped position on the test tube were marked with masking tape.

Thereafter, the marked test tube was detached and 1g of the sample was weighed into it and its mouth covered with a (dry) thumb. It was carefully inverted over the water and clamped with the retort stand at the marked spot without removing the thumb. With the stop watch set to read the time, the thumb was removed and the sample allowed to fall onto the water surface as the stop watch was put on simultaneously.

The samples were observed closely and the stop watch stopped just as the last few sample particles got wet. The time (in seconds) was read from the stop watch and recorded as the wetting time. This experiment was repeated three times for each sample and the mean values obtained.

Gelatinization temperature

5g of the sample was suspended in a beaker containing 20ml of water and heated while continuously stirring it. The temperature at which the suspension gels was recorded as the gelatinization temperature.

Foaming capacity

The method described by Onwuka[8], was adopted. One gram (1g) of the sample was blended with 10ml of distilled water in a warring blender for 5 minutes at room temperature. The mixture was quickly but carefully transferred to measuring cylinder and the foam volume was measured. The volume of foam formed was then recorded and used to calculate the foam capacity in percentage.

% Foaming Capacity (FC) =[(Va – Vb) / Vb]x 100

Where: Va = volume after whipping; Vb = volume before whipping

A final observation is made after 15, 30, 60, and 120 s to obtain the foam stability (in %).

Statistical analysis

The data obtained from the experiment were analyzed using Analysis of Variance (ANOVA) and the data were evaluated for significant differences ($P<0.05$) in their means. Differences between means were separated using the Fisher's Least Significant Difference (LSD) test procedure.

RESULT AND DISCUSSION

Seed characteristics

After the examination of the dry seeds, the results of the features were collected as shown in TABLE 1. The shape was oblong and its testa (seed coat) was

Full Paper

black in colour, white eye-coloured, and smooth in appearance. The seed coat was also hard in texture and tough in its attachment to the cotyledon. The weight per 100 seeds was estimated at 15.23 ±0.26 g, and the average seed weight was 0.15 ±0.02 g.

TABLE 1 : Seed characteristics of black crowder cowpea (BCC)

Average seed weight	Seed colour	Testa characteristics	Testa Attachment to cotyledon
0.15±0.02g	Black	Oblong shape, white eye, mostly smooth in appearance	hard in texture and tough in its attachment to the cotyledon

Functional properties of flour

TABLE 2 shows results of the functional properties of the full fat flour, defatted flour and protein isolate. Taha and Ibrahim (2002) stated that bulk density, BD (stated as g/ml or g/cm^3) is an important factor since it helps in choosing the appropriate packaging units. Bulk density of the BCC protein isolate (0.08 g/cm^3) was observed to be lower than those of the isolate of other legumes like African Yam Bean isolate (BD = 0.62 g/cm^3) and soy bean isolate (0.43 g/cm^3).

TABLE 2 : Functional properties of full fat flour, defatted flour and protein isolate made from black crowder cowpea

Samples	BD	WAC	OAC	EC	SWI	GT	FC	W
Full fat flour	1.68 ±0.02	2.43 ±0.05	1.89 ±0.04	62.06 ±0.94	1.51 ±0.02	78.50 ±0.41	20.50 ±0.35	30.67 ±1.25
Defatted flour	1.36 ±0.02	2.53 ±0.05	1.49 ±0.04	67.00 ±0.82	1.71 ±0.03	76.50 ±0.41	2.55 ±0.47	63.67 ±3.30
Protein Isolate	0.08 ±0.00	3.26 ±0.05	2.62 ±0.04	62.06 ±0.47	1.61 ±1.61	76.70 ±0.47	47.68 ±1.02	40.00 ±1.63

***All values are expressed as mean ± SD of their evaluations. Where: BD = Bulk density (g/cm^3); WAC = Water absorption capacity (ml/g); OAC = Oil absorption capacity (ml/g); EC = Emulsion capacity (%); SWI = Swelling Index (g/cm); GT = Gelling temperature (°C); FC = Foaming capacity (%); W = Wettability = (sec); SD = Standard deviation**

A 40°C to 100°C range temperature variation in a liquid medium produced a decrease in the Foaming Capacity (FC) and Water Absorption Capacity (WAC) but an increase in the Swelling Index (SWI) and Emulsion Capacity (EC) of the samples respectively (see TABLES 3.1A, 3.2A, 3.3A and 3.4A). Significant differences (P<0.05) were obtained in the results of each of some of the above functional properties treated to the temperature range (40 – 100°C). These differences (P<0.05) were observed in the Water Absorption capacity (WAC) and Emulsion capacity (EC) of the three samples. However, the same temperature treatment on the samples produced insignificant differences (P<0.05) in their Swelling Index and Foaming Capacity. These functional properties of the samples were obtained in the order WAC: Protein Isolate > Defatted flour > Full fat flour; OAC and FC: Isolate > Full fat flour > Defatted flour; EC: Defatted flour > Full fat flour = Protein Isolate. Gelling property is known to be important in comminuted sausage products and is the basis of many Oriental textured food e.g. tofu. The values were about 78.50°C, 76.50°C and 76.70°C for the full fat flour, defatted fat flour and Protein Isolate respectively.

Circle et al. (1972) pointed out that the oil binding capacity of protein materials is important factor that de-

TABLE 3.1A : Effect of temperature on foaming capacity

Samples	FC Affected By Temperature Variations (%)					
	40°C	60°C	80°C	100°C	Total	Mean ±SD
Full-fat	16.19	13.21	2.83	0.00	32.23	8.06 ±6.7996
Defatted	2.38	0.95	0.47	0.00	3.80	0.95 ±0.8913
Protein Isolate	19.00	4.00	0.00	0.00	23.00	5.75 ±7.8222
Total	37.57	18.16	3.30	0.00	59.03	
Mean	12.52	6.05	1.10	0.00		
SD	±7.2636	±5.2115	±1.238	±0.00		

***Using ANOVA for row and column, the effect due to variation in temperature is not significant, and cannot be further separated using Fisher's LSD Test.**

TABLE 3.1B : Table of Two-Way ANOVA for effect of temperature on foaming capacity+

Source of variance	SS	DF	MS	F_{cal}	$F_{tab = 0.05}$
SSA	105.18	2	52.59	2.2668	5.14
SSB	293.68	3	97.90	4.2198	4.76
SSE	139.18	6	23.20		
Total	538.04	11			

TABLE 3.2A : Effect of temperature on swelling index

Samples	FC Affected By Temperature Variations (%)					
	40°C	60°C	80°C	100°C	Total	Mean ±SD
Full-fat	2.50	2.70	2.70	3.00	10.90	2.73 ±0.1785
Defatted	2.70	2.80	2.90	2.90	11.30	2.83 ±0.6042
Protein Isolate	2.70	2.90	2.70	2.70	11.00	2.75 ±0.0866
Total	7.90	8.40	8.30	8.60	33.20	
Mean	2.63	2.83	2.77	2.87		
SD	±0.0943	±0.0816	±0.0934	±0.1247		

***Using ANOVA for row and column, the effect due to variation in temperature is not significant, and cannot be further separated using Fisher's LSD Test.**

TABLE 3.2B : Table of Two-Way ANOVA for effect of temperature on swelling index of samples

Source of variance	SS	DF	MS	F_{cal}	$F_{tab = 0.05}$
SSA	0.0217	2	0.0109	0.6646	6.94
SSB	0.0867	3	0.0289	1.7622	6.94
SSE	0.0983	6	0.0164		
Total	0.2067	11			

TABLE 3.3A : Effect of temperature on water absorption

Samples	WAC Affected By Temperature Variations (%)					
	40°C	60°C	80°C	100°C	Total	Mean ±SD
Full-fat	3.00	2.30	2.10	1.10	8.50	2.13^{b} ±0.6796
Defatted	2.80	2.50	2.30	1.20	8.80	2.20^{a}±0.6042
Protein Isolate	2.60	2.20	1.80	0.60	7.20	1.80^{c} ±0.7483
Total	8.40	7.00	6.20	2.90	24.50	
Mean	2.80^{a}	2.33^{b}	2.07^{b}	0.97^{c}		
SD	±0.16	±0.13	±0.21	±0.26		

***a – c means with uncommon superscript along the rows and column differ significantly at p < 0.05.**
LSD (R) = 0.020; LSD (C) = 0.333

TABLE 3.3B : Table of Two-Way ANOVA for effect of temperature on water absorption

Source of variance	SS	DF	MS	F_{cal}	$F_{tab = 0.05}$
SSA	0.3617	2	0.1809	11.03	5.14
SSB	5.4492	3	1.8164	110.76	4.76
SSE	0.0983	6	0.0164		
Total	5.9092	11			

TABLE 3.4A : Effect of temperature on emulsion capacity

Samples	EC Affected By Temperature Variations (%)					
	40°C	60°C	80°C	100°C	Total	Mean ±SD
Full-fat	0.6458	0.6853	0.7014	0.7014	2.7339	0.6835^{b} ±0.0227
Defatted	0.6549	0.7183	0.7343	0.7483	2.8558	0.7140^{a} ±0.0357
Protein Isolate	0.6296	0.6618	0.6691	0.6838	2.6443	0.6611^{c} ±0.0198
Total	1.9303	2.0654	2.1048	2.1335	8.234	
Mean	0.6434^{b}	0.6885^{a}	0.7016^{a}	0.7112^{a}		
SD	±0.0105	±0.0232	±0.0266	±0.0272		

***a – c means with uncommon superscript along the rows and column differ significantly at p < 0.05.**
$LSD\ (R)_{0.05}$ = 0.015; $LSD\ (C)_{0.05}$ = 0.026

TABLE 3.4B : Table of Two-Way ANOVA for effect of temperature on emulsion capacity

Source of variance	SS	DF	MS	F_{cal}	$F_{tab = 0.05}$
SSA	0.0056	2	0.0028	28.00	5.14
SSB	0.0081	3	0.0027	27.00	4.76
SSE	0.0007	6	0.0001		
Total	0.0144	11			

termines how well the material will perform as meat analogue or extender. Oil Absorption Capacity of 4.08ml/g has been reported for African Yam Bean (AYB) Protein Isolate (Adebowale et al., 2009). This was found to be only slightly higher than that obtained for the BCC Protein Isolate (2.62ml/g). However, the Foaming Capacity of the BCC Protein Isolate (47.68%) was much higher than the value obtained for AYB isolate (5.23%). This property is the capacity to form stiff, stable foam and is an important requirement of proteins to be incorporated into a gel cakes, whipped toppings, and deserts. Hence, this relatively high foamability of BCC isolate may enhance its utilization as a functional ingredient in some Nigerian food products.

Effect of temperature

Results show that the 40°C - 100°C temperature variations brought about reduction in the mean Water Absorption Capacity of the full fat and defatted flour samples from 2.43ml/g to 2.13ml/g and from 2.53ml/g to 2.20ml/g respectively. Similar reduction of the mean Water Absorption Capacity was also obtained for the protein isolate (from 3.26ml/g to 1.80 ml/g). These reduction in WAC may be as a result of the denaturation of the protein content of the samples by heat of temperatures above 60 – 70°C resulting in the disruption of hydrogen bonding and non-polar hydrophobic interactions. As most other functional properties are protein-related, a reduction in their values was expected at temperature treatment tending to 100°C. It was however observed that temperature variations from 40°C to 100°C produced an increase in the Emulsion Capacity and Swelling Index of the full fat and defatted flours as well as the protein isolate. These results are tabulated in TABLES 3.1A - 3.4B below.

Effect of pH

The result of the effect of pH on the functional properties of the BCC flours and isolate samples is as shown in TABLES 4.1A – 4.3B. Unlike for the temperature treatment, pH variations from 4.5 to about 10.0 produced a progressive increase in the WAC, FC and SWI of the full fat flour, defatted flour and protein isolate of the BCC. These increases were found to be consistent with similar results obtained for wheat. Beyond this pH (10.0), the functional properties of the samples were

Full Paper

observed to produce a decreasing trend, with those of the isolate showing greater tendency to reduce in value. Okezie and Bello[7] reported that the pH affects the solubility of protein. In turn, the solubility is known to be a critical factor that influences functional properties.

TABLE 4.1A : Effect of pH on foaming capacity

Samples	FC Affected By Temperature Variations (%)						
	4.5	6.0	10.0	12.0	14.0	Total	Mean ±SD
Full-fat	4.85	5.96	9.09	9.72	8.51	38.13	7.63^{a} ±1.8863
Defatted	7.02	5.43	8.59	8.75	9.65	39.44	7.89^{a} ±1.4924
Protein Isolate	2.89	5.15	3.93	3.69	3.55	19.21	3.84^{b} ±0.7394
Total	14.76	16.54	21.61	22.16	21.71	96.78	
Mean	4.92	5.51	7.20	7.39	7.24		
SD	±1.69	±0.34	±2.32	±2.64	±2.65		

***a, b means with uncommon superscript along the rows differ significantly at p < 0.05 but that of column are not significant and cannot be further separated using Fisher's LSD Test. LSD $(R)_{0.05}$ = 2.078**

TABLE 4.1B : Table of Two-Way ANOVA for effect of pH on foaming capacity of samples

Source of variance	SS	DF	MS	F_{cal}	$F_{tab = 0.05}$
SSA	51.26	2	25.63	12.63	4.46
SSB	15.85	4	3.96	1.95	3.84
SSE	16.22	8	2.03		
Total	83.33	14			

TABLE 4.2A : Effect of pH on swelling index

Samples	SWI Affected By Temperature Variations (%)						
	4.5	6.0	10.0	12.0	14.0	Total	Mean ±SD
Full-fat	1.30	1.79	1.35	1.39	1.18	7.01	1.40^{b} ±0.2064
Defatted	1.25	1.29	1.30	1.39	1.32	6.55	1.31^{b} ±0.0460
Protein Isolate	1.69	2.34	2.12	1.62	1.52	9.29	1.86^{a} ±0.3163
Total	4.24	5.42	4.77	4.40	4.02	22.85	
Mean	1.41	1.81	1.59	1.47	1.34		
SD	±0.20	±0.43	±0.38	±0.11	±0.14		

***a, b means with uncommon superscript along the rows differ significantly at p < 0.05 but that of column are not significant and cannot be further separated using Fisher's LSD Test. LSD $(R)_{0.05}$ = 0.293**

TABLE 4.2B : Table of Two-Way ANOVA for Effect of pH on swelling index of samples

Source of variance	SS	DF	MS	F_{cal}	$F_{tab = 0.05}$
SSA	0.8613	2	0.4307	10.68	4.46
SSB	0.4011	4	0.1003	2.49	3.84
SSE	0.3225	8	0.0403		
Total	1.5849	14			

TABLE 4.3A : Effect of pH on water absorption

Samples	WAC Affected By Temperature Variations (%)						
	4.5	6.0	10.0	12.0	14.0	Total	Mean ±SD
Full-fat	1.68	1.87	1.98	2.01	1.39	8.93^{c}	1.79 ±0.2293
Defatted	1.83	1.97	2.12	2.19	2.17	10.28^{b}	2.06 ±0.1368
Protein Isolate	2.63	3.96	3.73	3.48	2.54	16.34^{a}	3.27 ±0.5787
Total	6.14	7.80	7.83	7.68	6.10	35.55	
Mean	2.05	2.60	2.61	2.56	2.03		
SD	±0.42	±0.96	±0.79	±0.66	±0.48		

***a, b, c means with uncommon superscript along the rows differ significantly at p < 0.05 but that of column are not significant and cannot be further separated using Fisher's LSD Test. LSD $(R)_{0.05}$ = 2.078; LSD $(C)_{0.05}$ = 0.499**

TABLE 4.3B : Table of Two-Way ANOVA for effect of ph on water absorption of samples

Source of variance	SS	DF	MS	F_{cal}	$F_{tab = 0.05}$
SSA	6.2303	2	3.1150	26.59	4.46
SSB	1.0935	4	0.2734	2.33	3.84
SSE	0.9372	8	0.1172		
Total	8.2610	14			

CONCLUSION

Effect of temperature variation

From the outcome of the research, it can be concluded that variations in temperature from 40 °C to 100 °C produces increase in Emulsion Capacity and Swelling Index of Full-fat and Defatted flour samples as well as protein isolate of dehulled Black Crowder Cowpea. On the contrary, the same temperature variations bring about reduction in their Water Absorption Capacity and Foaming Capacity.

Effect of pH variation

pH variation from 4.5 to about 10.0 generates a progressive increase in the WAC, FC and SWI of the full fat flour, defatted flour and protein isolate of Black Crowder Cowpea (*Vigna unguiculata*), and beyond pH 10.0, the functional properties of the samples produce a decreasing trend, with those of the isolate showing greater tendency to reduce in value.

REFERENCES

[1] Association of Analytic Chemists; Standard Official Methods of Analysis of the Association of Ana-

lytical Chemists. 14th edition, S.W. Williams (Ed), Washington DC, **(1984)**.

[2] C.S.James; Experimental Methods in Analytical Chemistry of Foods. Chapman and Hall. New York, 28 **(1995)**.

[3] D.E.Kay; Crop and Product Digest No. 3 – Food Legumes. London: Tropical Products Institute, 86–101 **(1979)**.

[4] J.E.Kinsella; Functional Properties of Protein in Foods. A survey CRC Critical Review. Food Sci. and Nutr., **7**, 219–280 **(1979)**.

[5] A.Moure, J.Sineiro, Domínguez, J.C.Parajó; Functionality of oilseed protein products: A review. Food Research International, **39**, 945-963 **(2006)**.

[6] T.U.Nwabueze, M.O.Iwe; Mass flow rate, nutrient composition and some functional properties of single screw extruded Africa breadfruit (Treculia africana) blends. Journal of Food Technology., **4(1)**, 50–58 **(2006a)**. 90 Food Bioprocess Technol, **4**, 80–91 **(2011)**.

[7] B.O.Okezie, A.B.Bello; Physico–chemical and functional properties of winged bean flour and isolate, compared with soy isolate. Journal of Food Science, **53**, 450–454 **(1988)**.

[8] G.I.Onwuka; Food Analysis and Instrumentation. Theory and Practice. Naphtali Prints, Lagos, Nigeria, 133-137 **(2005)**.

[9] F.S.Taha, M.A.Ibrahim; Effect of degree of hydrolysis on the functional properties of some oilseed proteins: Fats and Oils Department, National Research Center, Cairo, Egypt. Grasas y Aceites 273, Fasc. 3, **53**, 273-281 **(2002)**.