Full Length Research Paper

Physicochemical and functional properties of proteinates of two Bambara groundnut (*vigna subterranean*) landraces

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Bambara groundnut (*Vigna subterranea*), an underutilised, and until lately, under researched, is a crop with a high potential for the attainment of food security and poverty alleviation in Africa, as it shows considerable drought resistance and high nutritional qualities. Physico-chemical composition and key functional properties of proteinates for potential food product development of two Bambara groundnut landraces were determined. landraces recorded percent crude protein, crude fat, total ash, crude fibre, carbohydrate and moisture in the ranges of 21.80 to 23.99%; 4.84 to 6.07%; 3.22 to 3.30%; 3.16 to 3.67%; 52.69 to 54.51% and 11.16 to 11.59% respectively. The results of functional property determinations of isolates indicated that foam capacity, water binding capacity, oil binding capacity, emulsification property and emulsifying stability also ranged from 66.0 to 72.0%; 9.89 to 10.0 mg/g sample; 8.99 mg/g sample; 53.0 to 56.0% and 96.43 to 98.15% respectively. The results show that Bambara groundnut flour has great potential for incorporation into various human foods where it could provide quality useful plant proteins.

Keywords: Bambara groundnut, proximate composition, functional properties, proteinate

INTRODUCTION

Bambara groundnut (Vigna subterranea), an indigenous orphan, underutilised, and until lately, under researched crop, cultivated throughout sub-Saharan Africa, is a crop with a high potential for the attainment of food security and poverty alleviation in Africa, as it shows considerable drought resistance and potentially, high nutritional qualities. Though Smartt and Simmonds (1995) observed that the introduction of groundnut, an oily seed crop, into West Africa from Brazil outplayed the importance of Bambara as an oil seed crop, at an era where consumers are concerned about the danger of intake of more fatty and oily foods, the relatively low oil content of Bambara groundnut (5 - 6%) high carbohydrate (40 - 60%) and relatively high amount of protein (16 - 25%) make the crop a complete food (Goli, 1997; Brough and Azam Ali, 1992; Poulter and Caygil, 1980; Ankroyed and Doughty, 1982; Deshpande and Demodaram, 1990) capable of

reducing nutritional deficiencies and a probable raw material source for the canning industry (Doku and Karikari, 1971). The majority of Bambara groundnut is produced for home consumption although a small amount is grown as a cash crop (Coudert, 1982).

The crop has a number of production advantages in that it can yield on poor soils with little rainfall as well as produce substantial yields under better agronomic conditions (Linnemann and Azam-Ali, 1993; Anchirinah et al., 2001; Azam-Ali et al., 2001). Its nutritional superiority over other legumes renders it the potential to be preferred in terms of nutritional value and economic importance (Linnemann, 1990; Brough and Azam-Ali, 1992). Despite these important attributes, neither the agro-ecological and genetic potential as well as nutritional importance of Bambara groundnut have yet been fully realized nor its full economic significance determined. The crop is still cultivated from local landraces rather than varieties bred specifically for particular agro-ecological conditions or production systems (Ofori et al., 2006). According to Rowland

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(1993), apart from soybeans, Bambara is rated high in protein compared to other legumes and staples. It also contains methionine, which makes its protein more complete than any other bean. It also has the highest concentration of soluble fibre than any bean, which research has shown to reduce the incidence of heart disease and certain types of cancer (Tulimara, 2004). In addition to the good nutritional properties Bambara aroundnut also contains rich quantities of polyunsaturated fatty acids, with linoleic acid being the predominant fatty acid (Kay, 1979). Although Bambara groundnut dishes are nutritious, its preparation, processing and consumption is constrained by the hardto-cook characteristics, dehulling difficulty, antinuritional inhibitors, strong beany flavour and bitter after-taste (Enwere and Hung, 1996).

As an edible legume with good nutritional attributes the potential to develop many food products could be unprecedented. However. information on physicochemical and functional properties which is prerequisite for enhanced utilization of the crop in Ghana is inadequate, hence this study. While many food products (eg milks, low fat spread, meat replacers, emulsifiers, etc) have been developed from soy, peanut, and cowpea because considerable support in both material and financial were received from government and international organisations, not enough similar research has been done on Bambara groundnut.

The objective of this work therefore is to determine the physicochemical and functional properties of proteinates of two Bambara groundnut landraces to increase its utilization base.

MATERIALS AND METHODS

Two Bambara groundnuts landraces for the study namely 'Kpalulu' and 'Dagati achi' were obtained from farmers at Wenchi in the Brong Ahafo region of Ghana. Samples were dehulled and winnowed. The dehulled seeds were dried in hot air oven at 40 °C for 8 hours. About 1000g each of the two landraces were then milled to flour, kept in black polyethylene bags and stored in a freezer until when needed. The flour was used for the proximate analyses using standard AOAC (1990) methods.

Determination of Functional Properties of Bambara groundnut Protein

Protein Extraction

Proteins from the flours of the two landraces were extracted by manipulating temperature, pH and acid-base coagulation treatments according to the method of Soestrisno and Holmes, (1992) with slight modification. To 100 g of defatted Bambara groundnut flour was added

1000 mL of distilled deionised water and thoroughly mixed in the Kenwood electronic mixer for 1 minute. The pH of the suspension was adjusted to between 10 and 11 pH units using 2 N NaOH. The temperature of the suspension was kept constant during the extraction at 50 ℃ by means of a thermostatically regulated shakerwaterbath (Eyela Shaker S8-8, Rikakikai Co. Ltd., Tokyo). Slurry was allowed to incubate for about 30 minutes, centrifuged at constant temperature of 4°C using a refrigerated centrifuge (Centikon T-42k, Kentron Instruments) and speed, 5000 rpm for 10 minutes. The supernatant was decanted and flour residue resuspended for a repeat of the extraction process. The two supernatants were pulled together and coagulated by adjusting the pH to the iso-electric point (pl) of the Bambara groundnut (4.0 pH units) determined earlier with 2N HCI. Proteins were separated from the whey by centrifuging at 5000 rpm for 10 minutes at 4°C, freezedried in CHRIST Freeze Drver. Yields were calculated from total weight of freeze-dried products per 100 g of defatted Bambara groundnut flour. The freeze-dried protein isolates were kept in polyethylene and stored at 4 °C until required.

Protein Solubility Determination

Protein solubility profile of the isolates was determined by the method of Sathe and Salunkhe (1981). Ten milligrams of isolate was dissolved in 10 mL 1N NaOH and the pH adjusted to desired level (2 - 12) with 1N HC1. Slurry was stirred for 30 minutes and centrifuged at $5000 \times g$ for 15 minutes. The protein content of the supernatant was determined by the method of Lowry *et al.*, (1951), using Folin Ciocalteu's reagent and egg albumin as a standard.

Foaming Properties Determination

This was determined according to the method of Howell and Taylor (1991) and Murphy and Howell (1990). Freeze-dried Bambara groundnut protein isolate (1.25 g) was weighed into 250 mL distilled water and whipped for 5 minutes at top speed in a Kenwood electronic blender. The entire contents of the blender were rapidly transferred into a 500 mL measuring cylinder and foam volume and drainage measured after 30 seconds followed by 5 minute intervals for 30 minutes. Foam expansion and foam volume stability were calculated in duplicate and the mean values estimated.

Emulsion Capacity

Freeze-dried Bambara groundnut protein isolate (3.5 g) was dispersed in 50 mL water and 50 mL of vegetable oil

Variety	Moisture	Ash	Crude Protein	Crude Fat	Crude Fibre	Carbohydrate
Kpalulu	11.16	3.30	21.80	6.07	3.16	54.51
	(0.06)	(0.13)	(0.25)	(0.23)	(0.25)	(0.05)
Dagati achi	11.59	3.22	23.99	4.84	3.67	52.69
-	(0.20)	(0.15)	(0.13)	(0.19)	(0.44)	(0.26)

Table 1. Proximate Composition (%) of Bambara Groundnut

Values in parenthesis are standard deviation

 Table 2. Yield and Protein Content from Extraction of Bambara Groundnut (Kpalulu and Dagati achi)

Variety	Yield of Protein (%)	Protein(%)
Kpalulu	27.05	61.17
Dagati achi	29.41	65.22

using a magnetic stirrer (Stuart Scientific Magnetic Stirrer Hotplate, Stuart Scientific Co. Ltd., UK), and emulsified. Emulsion was divided into two 50 mL centrifuge tubes and centrifuged at 1300 x g for 5 minutes. The height of emulsified layer and height of whole layer were measured and emulsion capacity calculated.

Emulsion Stability

The produced emulsion was heated at 80 $^{\circ}$ C in a water bath for 30 minutes, cooled under tap water for 15 minutes and centrifuged at 1300 x g for 5 minutes. Height of remaining emulsified layer and the initial emulsified layer were measured and percent emulsion stability calculated.

Water and Oil Absorption Capacities

The method of Beuchat, (1977) was used to determine water and oil absorption capacities of proteinates. Sample (0.5 g) was thoroughly mixed with 5mL distilled water / oil for 5 minutes using a magnetic stirrer. The mixtures were allowed to stand for 30 minutes at ambient temperature, centrifuged at 5000 x g for 30 minutes and the volume of the supernatant measured using a 10 mL graduated cylinder. Density of water was assumed to be 1 g/mL and that of oil 0.89 g/mL. Percent water and oil absorbed were calculated .

RESULTS

Results of proximate composition determinations are presented in table 1.

Yield of Protein

Table 2 below shows the yields of extracted proteins at 50°C extraction temperature, pH of 10 and coagulating pH of 4 (iso-electric point). Yields were calculated from total weight of freeze-dried products per 100 g of Bambara groundnut flour. Non-destructible protein yields

on dry solids basis were 27.05% and 29.41% for Kpalulu and Dagati achi respectively with respective percent protein contents of 61.17% and 65.22% for Kpalulu and Dagati achi.

Protein Solubility

The solubility profile of the two Bambara groundnut proteins is shown in Figure 1. Maximum solubility of 578 μ /mL for Kpalulu and 428 μ /mL for Dagati achi occurred at pH 10 and pH 12 respectively. As the acidity is increased, solubility dropped rapidly and a minimum value of 67 μ /mL and 72 μ /mL for Kpalulu and Dagati achi respectively were observed at pH 4. pH 4 was therefore considered as the isoelectric point (pl) of Bambara groundnut protein isolates.

Foam Capacity

Table 3 below shows data on foaming potential of the Bambara groundnut proteinates as a function of time.

Foam Volume Stability

Foam volume stability decreased significantly (P<0.05) with time (Figure. 2). The foams broke down rapidly within 5 minutes after which they began to stabilize. The stabilities of Bambara groundnut which were 27.57 % and 26.76 % for Kpalulu and Dagati achi respectively were lower than that of egg albumin(40.93 %) after 30 minutes of foam formation.

Emulsion Capacity and Emulsion Stability

Results of emulsion capacities and emulsion stability of protein isolates of landraces are as shown in Table 4.

Water and Oil Absorption Capacity

Results of water and oil absorption capacity of protein isolates of landraces are as shown in Table 5.

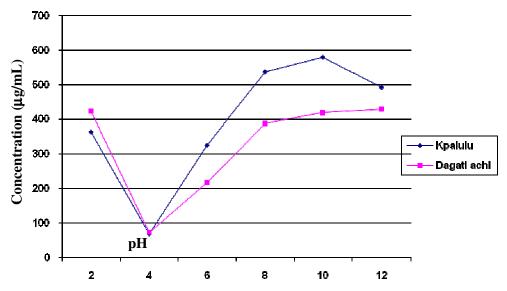


Figure 1. Solubility Profile of Protein Isolate from Kpalulu and Dagati Achi as a Function of pH

Table 3. Foam Capacity of Protein Isolate of Kpalulu and Dagati Achi

Variety	Foam Capacity(%)
Kpalulu	72.0
	(2.8)
Dagati achi	66.0
	(0.0)

Values in parenthesis are standard deviation

DISCUSSION

Proximate Composition (%) of Bambara Groundnut. The results agreed closely with those reported by Nwanekezi *et al.*, (1994) on Bambara groundnut and African yam bean and are similar to that of cowpea as reported by Garti, (1999) with the exception of crude fat which was relatively higher for Bambara groundnut because Bambara is a high fat legume relative to cowpea. Apart from crude protein, fat and carbohydrates which registered some differences within landraces, moisture, ash and fibre had similar amounts within landraces. Dagati achi had slightly higher proteins (23.99 %) than Kpalulu (21.80%). Kpalulu recorded slightly higher carbohydrates (54.51 %) than Dagati achi (52.69 %). These differences may be attributed to varietal properties.

Protein Solubility

Solubility of Kpalulu and Dagati achi varied significantly (P<0.05) at all the pH values except at pH of 4. The variation in the solubility values for Kpalulu (67 μ /mL) and

Dagati achi (72 μ /mL) at pH 4 were not statistically significant.

From the graph, as pH values increase beyond the isoelectric point (pl), the negative charges on the protein molecules increase and water may interact with these charges, thus contributing to solubilization. Also, protein molecules with the same net charges have the tendency to repel each other and to dissociate or unfold (Cheftel *et al.* 1985). The trend of solubility observed for the protein isolate suggests their potential use in beverages as vegetable milk for lactose intolerant individuals (Abbey and Ibeh, 1988). However, the practical significance of protein solubility at higher pH values (9 - 12) may be limited as most foods are rarely at this pH.

Foam Capacity

Results suggest that the foaming capacities for both Kpalulu and Dagati achi were higher than those reported for raw (40%) and heat processed (24%) cowpea flour (Abbey and Ibeh, 1988) and for winged bean flour (29%) but were lower than the values reported for winged bean isolate (82%) and soy isolate (84%) (Okezie and Bello, 1988).

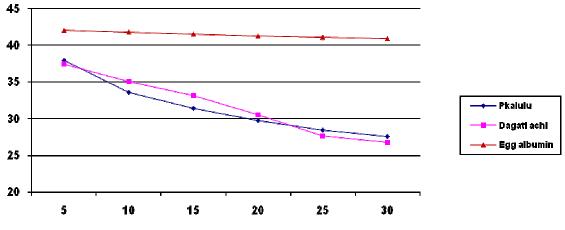


Figure 2. Foam Volume Stability of Acid Proteinates as a Function of Time

 Table 4. Emulsion Properties of Kpalulu and Dagati Achi Protein

Emulsion Properties	Kpalulu	Dagati Achi
Emulsion Capacity (%)	53.0	56.0
Emulsion Stability (%)	98.15	96.43

Foam Volume Stability

As the volume of foam decreased, the rate of foam collapse decreased concomitantly. This could be attributed to the concentration of the protein molecules which increased as the volume decreased. The stability of egg albumin was significantly (P<0.05) higher than foams of Kpalulu and Dagati achi at all the times. The ability of extracted Bambara groundnut proteinates to foam and maintain the foam was inferior to egg albumin (Figure. 2). However the foaming stabilities of Bambara groundnut protein were higher than that of cowpea acid proteinate (16.64%) extracted at pH 10 and temperature of 50 $^{\circ}$ C (Garti, 1999).

Emulsion Capacity

Emulsion capacities of Kpalulu (53.0 %) and Dagati achi (56.0 %) were slightly lower as compared to that of yellow pea as reported by Soestrisno and Holmes (1992) and that of cowpea (Ahenkora, 1989). However, these values were slightly higher than that of full-fat soy flour (Plahar *et al.*, 1977) and that of winged bean flour (Okezie and Bello, 1988).

Emulsion Stability

From Table 4, the emulsions from Bambara groundnut proteinates were more stable compared to those of full-

fat soy (42.8 - 50.7 %) (Plahar et al., 1977), winged bean isolate (22.2 %), winged bean flour (21.1 %), soy isolate (20.9 %) (Okezie and Bello, 1988) and for raw cowpea flour (78 %) (Ahenkora, 1989). This high emulsion stability of proteinates may be attributed to high globular fraction of Bambara groundnut protein. Sathe et al. (1982), attributed the high emulsion stability of winged bean protein to the globular nature of the major proteins of winged bean. Such high emulsion properties of Bambara groundnut flour will have the advantage of imparting emulsification and emulsion stability properties to products to which they are added better than other legumes like the full-fat soy flour which has emulsion properties ranging between 42.8 - 50.7 % (Plahar et al., 1977) and for winged bean flour which also has emulsion properties of 21.1% (Okezie and Bello, 1988). The comparatively good emulsion properties of Bambara groundnut isolate has the potential for use in meat, ice creams, baking and texturized or engineered high protein food industries as reported by Garti, (1999) on cowpea.

Water and Oil Absorption Capacity

Water absorption capacity is a useful indicator of whether flours or isolates can be incorporated into aqueous food formulations. Water absorption capacities of Kpalulu and Dagati achi isolates were 9.89 mg/g and 10.0 mg/g respectively. These values (Table 5.) are consistent with cowpea isolates as reported by Garti (1999). They are Table 5. Water and Oil Absorption Capacities of Kpalulu and Dagati Achi Protein

Property	Kpalulu	Dagati Achi
Water Absorption Capacity (mg/g)	9.89	10.0
Oil Absorption Capacity (mg/g)	8.99	8.99

twice the water absorption capacities of winged bean protein isolates (5 mg/g) (Okezie and Bello, 1988) and four times higher than African yam bean flour (2.2mg/g) and Bambara groundnut flour (2.6mg/g) (Nwanekezi et al., 1994). Such properties of these Bambara groundnut isolates and flour suggest that they may perform very well in terms of physicochemical properties when used for development of vegetable milk. High oil absorption capacity of protein isolates is a requirement for ground meat formulation, meat replacers and extenders, doughnuts, pancake, baked goods and soups (Abbey and Ibeh, 1988). Oil absorption capacities of Bambara groundnut (Table 5) compared favourably with that of cowpea varieties asetenapa and adom as reported by Addo (2004) and three times that of heat processed African yam bean and Bambara groundnut flour (Nwanekezi et al., 1994). The high oil absorption capacity of proteinates observed might probably be due to denaturation of protein during extraction resulting in the exposure of lipophilic or hydrophobic residues forming increased lipid-protein complexes (Kinsella, 1979).

CONCLUSION

This study has shown that the compositional and physicochemical characteristics of the two landraces of Bambara groundnut are similar to those of other legumes such as cowpea. The emulsion properties of Bambara groundnut isolate will have the advantage of imparting emulsification and emulsion stability to products to which they are added. The comparatively good emulsion properties have the potential for use in meat, ice creams, baking and texturized or engineered high protein food industries as reported by Garti, 1999 on cowpea (variety Ayiyi). The results show that Bambara groundnut has great potential for incorporation into various human foods where it could provide useful plant proteins. Such properties as high water and oil absorption capacities of these Bambara groundnut isolates is an indication that they will do very well in terms of physicochemical properties in the development of vegetable milk.

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