SHORT COMMUNICATION article

Nutritional evaluation of processed cocoyam, soya bean flour, and their blends by feeding trials using Albino rabbits

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Abstract: Food nutrients are essential for the growth and development of human. In several parts of the world, Cocoyam (*Colocasia esculenta*), is a tropical root crop cultivated for food formulations and consumption by humans. This study was conducted to assess the nutritional evaluation (weight gain, protein efficiency ratio, net protein retention, and biological value) of the processed cocoyam, soya bean (*Glycine max*) flours and their blends by feeding trials using Albino rabbits. The sun-dried samples were milled to size (sieve with 25 µm aperture size) to obtain homogenous flours. Blends were obtained with various ratios according to the percentage of soya bean/cocoyam flours. The biological value, protein efficiency ratio, and net protein retention were analysed using standard methods for the calculations. Net protein retention of soya bean flour was higher than that for cocoyam flour. The highest biological value was given by the 50% blend, while the lowest was given by the control diet. The higher the protein efficiency ratio values, the higher the protein quality. The highest protein efficiency ratio value was given by soya bean flour, while the lowest value was from Cocoyam flour. The best blend of Cocoyam and soya bean flours obtained based on chemical and nutritional evaluations was the 50% blend of soya bean and Cocoyam flours.

Introduction

Micronutrients are used to build and repair tissues and regulate body processes, while macronutrients are converted to smaller sugars (glucose, fructose and galactose) from carbohydrates, protein to amino acids and fats to fatty acids and glycerol [1]. Animals have specialized digestive systems that work to break down macronutrients to provide energy and utilize micronutrients for metabolism and anabolism (constructive synthesis) in the body [2]. Organic nutrients consist of carbohydrates, fats, proteins (or other building blocks, amino acids) and vitamins [3]. Inorganic chemical compounds such as dietary minerals elements and water may also be considered nutrients [4]. Malnutrition may result from an inadequate or unbalanced diet when the body does not get enough diet [5]. Cocoyam (*Colocasia esculenta*) is an annual herbaceous plant with a long history of usage in traditional medicine in the tropical and subtropical regions. Cocoyam is rich in vitamin B6 and potassium. It contains dietary fibre and higher protein content than the majority of tropical root crops [6]. The herb has been known since ancient times for its curative properties and has been utilized for the treatment of various ailments such as asthma, arthritis, diarrhoea, internal haemorrhage, neurological disorders, and

internal skin disorders. Its juice is widely used for the treatment of body aches and baldness [7]. Soya bean *(Glycine max)* is the key benefits are its high protein content, vitamins, mineral elements and insoluble fibre. Soya bean contains vitamin K, riboflavin, foliate, vitamin B6, thiamine, and folic acid [8]. A variety of health benefits, including protection against breast cancer and treatment have been attributed to soya bean food consumption primarily because of soya bean isoflavone (genistein, daidzein, and glycitein) [9]. There is also evidence that soya bean alleviates hot flashes in menopausal women [10]. Considerable data suggest that the soya bean isoflavone is largely responsible for many of the proposed benefits of soya bean foods [11]. The aim of this study was to examine the nutritional evaluation of processed cocoyam, soya bean flours and their blends by feeding trials using albino rabbits. Protein efficiency ratio (PER), NPR (net protein retention), and BV (biological value), were assessed in the animal feeding studies as nutritional assessment methods.

Materials and methods

Cocoyam (*Colocasia esculenta*) and soya bean (*Glycine max*) were sourced locally from the New Benin market within Benin City, Southern Nigeria and were identified by H. A. Akinnibosun at the Plant Biology and Biotechnology Department of the University of Benin, Benin City, Nigeria with herbarium numbers of UBH-C582 and UBH-G628 for *Colocasia esculenta* and *Glycine max*, respectively, and Albino rabbits bought from Aduwawa Market, in Benin City, Nigeria.

Preparation of samples: The cocoyam samples were peeled, sliced into pieces and sun-dried for five days. soyabean bought from New Benin Market, was sun-dried for five days. The dried cocoyam and soya bean were milled separately. Wire whisk sieve of 25 µm aperture size was used to sieve the flours to obtain homogenous flours, packaged in different airtight containers, labelled prior to the laboratory for analyses.

Proximate analysis: Proximate compositions (moisture, ash, crude fibre, crude protein, ether extract, and nitrogen-free extract), the percentage of the cocoyam and soya bean flour and their blends at different proportions (10%, 20%, 30%, 40%, and 50%) were determined by the Standard methods of the Association of Official Analytical Chemists [12].

Moisture content: 2.0 gm of the various samples were dried in an oven at 105°C until constant weight was obtained. The drying was for 24 hrs. The loss in weight was taken as the water or moisture content which can be expressed as a percentage of the material. The dry matter content was determined [12].

Determination of crude protein: The sample (0.5 g) was weighed into a kiedhal flask and 15.0 ml of sulphuric acid was added. The catalyst was made from a mixture of potassium sulphate and copper sulphate (ratio 1: 9) and a pinch of selenium were added. The material was placed on a heater in a fume cupboard and digested until the mixture became clear. That is, when all the nitrogen, except that in the form of nitrate and nitrite, present in the material would have been converted to ammonium sulphate. This corresponded with the time when the digest was clear, having changed from dark brown to light green or golden yellow colour. The next stage was distillation at which the digest was diluted by adding 50.0 ml of water into the kjedhal flask in the fumes cupboard to reduce the acidity. The digest was made up to 100 ml. 5.0 ml of the digest (aliquot) was placed inside the digester tube, 5.0 ml of 40% sodium hydroxide was added and placed on the heater and distilled off into a flat-bottom flask containing 5.0 ml of boric acid solution and distilled until 150 ml distillate was collected. The distillate obtained was titrated with 0. 01 M hydrochloric acid to the endpoint. The end point was obtained when the distillate colour turned to the initial colour of boric acid. This step was the titration stage. The nitrogen content of the material was calculated and the crude protein content was obtained by multiplying the nitrogen content by a factor (6.25). The factor was used because of the assumption that the average protein contains 16% nitrogen (100/16=6.25). The nitrogen and crude protein contents were calculated [12].

Determination of ether extracts (crude fat): 2.0 g of the sample was weighed into already weighed fat-free filter paper and placed inside a Soxhlet extractor. A flask containing the petroleum ether was placed on a heating mantle maintained at a low temperature (70°C). As the solvent heated up, the hot solvent rose and dripped through the samples and in the process extracted the fat which discharged into the flask. This process continued until all the fat had been extracted from the material. After extraction, the samples and filter paper were brought out and dried in an oven at 105°C for 2.0 hrs to complete the drying. The filter paper and the samples obtained were then weighed, and crude fat content was calculated [12].

Ash content: 2.0 g of the sample was weighed into a crucible of known weight. The crucible and sample were then put into a muffle furnace and ignited at 550°C until all organic content was removed [12].

Determination of crude fibre: 2.0 g of the sample was weighed into a standard flask into which 100 ml of 1.25% sulphuric acid was added and boiled on a heating mantle for 30 min. During boiling, the 100 ml mark was maintained by constantly adding warm water using a wash bottle. After 30 min, it was removed from the heating mantle, cooled and filtered; the residue was washed to neutrality (testing with litmus paper). The residues were transferred back into the standard flask containing 1.25% potassium hydroxide, and then made up to 100 ml, with the potassium hydroxide. It was again boiled for 30 min by maintaining the 100 ml, mark, after which it was removed, cooled and filtered and washed to neutrality with water and then, confirmed with litmus paper to be neutral. The residue was then transferred into a crucible of known weight with 50: 50% acetone/ethanol mixture and dried to constant weight in the oven at 105°C, and the weight was recorded. The sample from the oven was placed inside a muffle furnace at 300°C for 1 hr to ash the residue. The sample was then removed from the furnace after aching kept in a desiccator to cool and then weighed and the percentage of crude fibre was calculated [12].

Nitrogen-free extracts (NFE): The NFE was obtained by difference that is by subtracting the amounts of all five components above from 100%, and, thus, NFE was determined [12].

Feeding trials on the Albino rabbits: The test rabbits were fed daily with cocoyam and soya bean flours and their various blends including the control diet. PER, NPR, weight gain and BV were determined during the feeding trials and nutritional evaluation of the products. In the feeding trial, measurements such as the weight of the rabbits, and weight of feed intake were made at regular intervals. 40 Albino rabbits were used; the rabbits were divided into seven groups and randomly allotted to seven different dietary treatments namely, cocoyam flour, soya bean flour, 10%, 20, 30, 40, and 50% blend of soy bean and cocoyam flour and control diet. The first seven days were used for acclimatization and the feeding trial with the experimental feeds was conducted for 28 days. The feeds were formulated to conform with the rabbit nutrient/energy requirement (ENERGY - 2,500 - 2,700 kcal/kg, crude protein 16%-22%) [13].

The protein efficiency ratio (PER): PER is based on the weight gain of a test subject divided by its intake of a particular food protein during the test period.

Net protein retention (NPR): NPR is a measure of the percentage of ingested nitrogen intake (N) that is retained in the body. It is a combined test of digestibility and biological value and is defined as NPR= digestibility x BV [13].

Results and discussion

Table 1 shows the proximate composition of formulated experimental feeds. Thus, there are no differences regarding the dry and moisture contents of unblended cocoyam flour, unblended soya bean flour, 10%-50% blended soya bean/cocoyam flours, and control diet. However, the crude protein was varied among blended soya bean/cocoyam flours. This is also true for nitrogen-free extracts and crude fat. The value obtained for

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PER where the lowest efficiency ratio was for cocoyam flour, followed by control diet, 10% to 50% blend, and soya bean flour, respectively. PER is founded on the weight gain of a test matter divided by its intake of a specific food protein during the test period. It remains widely used for assessing the value of food proteins. A smaller value indicates a lower protein quality. The higher the PER values, the higher the protein quality. The results indicate the trend of protein quality in the cocoyam and soya bean products to be soya bean flour >50% to >10% blends of soya bean and cocoyam flours more than control diet more than cocoyam flour. The BV for unblended cocoyam flour, unblended soya bean flour, 10% to 50% blends and control were varied. The highest BV value was given by a 50% blend, followed by 40%, 20%, and 30% blends of soya bean and cocoyam flours, unblended soya bean and a 10% blend of soya bean and cocoyam flours. The supplementary action of one food protein on another is of special interest, since rarely does a human being or animal consume a single protein in his or her pattern of feeding. When two proteins are fed together, the biological value is greater than when either protein is fed alone and higher than the calculated average value would indicate [14]. Protein with high BV implies that a high number of amino acids are needed by the body. The BV of unblended soya bean flour was considerably higher than that of the unblended cocoyam flour. This is in agreement with the well-known good amino acid profile of soya bean. It is observed in the results that as the percentage of soya bean flour in the various blends increased, the BV and PER increased. This can also be ascribed to the well-documented good amino acid profile of soya bean which would tend to increase the BV of the blends as the content of soya bean flour increased in the blends. NPR is a practical method of evaluating differences in protein quality for human nutritional purposes. As for the NPR results, it is obvious that, once more, the unblended soya bean flour had a higher NPR value than the unblended cocoyam flour. Additionally, NPR values increased as the percentage of soya bean flour in various blends increased correspondingly, reflecting the higher quality protein in soya bean flour.

Samples	Dry matter content	Moisture content	Crude protein	Ether extract	Crude fat	Ash	Nitrogen free extracts
Unblended	89.55	10.45	04.00	0.35	0.68	2.30	82.23
cocoyam flour	±0.64	±0.64	± 0.28	± 0.07	± 0.04	± 0.28	± 0.18
Unblended	90.30	09.70	44.3	20.75	7.10	7.45	10.70
soya bean flour	±0.28	±0.28	±0.99	±0.92	± 0.42	± 0.78	±0.99
10% blend of soya bean/cocoyam	89.75 ± 0.35	10.25 ± 0.35	09.22 ± 1.10	$\begin{array}{c} 0.66 \\ \pm 0.08 \end{array}$	3.20 ±0.14	2.45 ±0.21	74.05 ± 1.15
flours							
20% blend of sova bean/cocovam	88.55	11.45	12.80	0.74	3.38	2.66	68.98
flours	±0.64	±0.64	±0.57	±0.02	±0.04	±0.03	±0.01
30% blend of soya bean/cocoyam	90.35 ±0.21	09.65 ±0.21	15.65 ±0.21	$\begin{array}{c} 0.96 \\ \pm 0.06 \end{array}$	3.55 ±0.35	2.73 ±0.04	67.47 ±0.05
flours							
40% blend of soya bean/	89.00 ± 0.00	11.00 ± 0.00	24.95 ±0.64	2.05 ± 0.07	3.60 ±0.28	4.40 ±1.13	54.00 ± 1.98
cocoyam flours							
50% blend of soya bean/	89.70 ±0.57	10.30 ±0.57	39.82 ±1.86	7.30 ±1.13	4.48 ±0.04	6.50 ±0.14	31.61 ±0.34
Control diet	90.15 ±0.07	90.85 ±0.07	05.95 ±0.21	0.46 ±0.08	1.25 ±0.07	1.00 ±0.00	81.50 ±0.15

Table 1: Proximate composition of formulated experimental feeds

N.B.: 10% blend means 10 g of soya bean flour mixed with 90 g of cocoyam flour,

and 20.0% blend means 20 g of soya bean flour mixed with 80 g of cocoyam flour, etc.

Table 2: Protein efficiency ratio, net-protein retention and biological values of soya bean and cocoyam flour and their blends

Samples	Nitrogen intake (g)	Retained nitrogen (g)	Absorbed nitrogen (g)	Total Diet consumed (g)/ (rabbit for 28 days)	Protein in Diet (%)	Protein consumed g/28days	Net- protein retention (%)	Biological value (%)	Weight gain (g)	Protein efficiency ratio
Unblended cocoyam flour	08.97 ± 0.49	01.26 ± 0.03	03.36 ± 1.25	1402.08 ± 33.61	4.00	350.52	14.05	37.50	155 ± 76.80	0.44 ± 0.02
Unblended soya bean flour	97.50 ± 3.30	33.55 ± 2.80	65.73 ± 5.38	1375.12 ± 26.50	44.30	31.04	34.41	51.04	292 ± 22.40	9.41 ± 0.48
10% blend of soya bean and cocoyam flour	20.42 ± 2.15	04.14 ± 1.15	09.79 ± 3.15	1379.40 ± 27.30	9.22	149.61	20.27	42.29	217 ± 8.50	1.45 ± 0.67
20% blend of soya bean and cocoyam flour	27.49 ± 2.03	08.85 ± 3.12	14.68 ± 4.16	1340.85 ± 2 3.30	12.80	104.35	32.19	60.29	285 ± 11.13	2.73 ± 0.01
30% blend of soya bean and cocoyam flour	35.00 ± 2.05	07.98 ± 2.50	14.42 ± 4.12	1400.03 ± 36.61	15.65	89.46	22.80	55.34	267 ± 17.10	2.98 ± 0.53
40% blend of soya bean and cocoyam flour	55.22 ± 3.15	16.19 ± 3.14	22.70 ± 2.18	1383.99 ± 33.70	24.95	55.47	29.32	71.34	183 ± 31.18	3.30 ± 0.45
50% blend of soya bean and cocoyam flour	89.25 ± 4.12	30.82 ± 3.18	42.27 ± 3.56	1401.08 ± 42.60	39.82	35.19	34.53	72.91	317 ± 23.57	9.01 ± 0.54
Control diet	13.36 ± 1.18	01.41 ± 0.04	04.50 ± 1.85	1406.81 ± 26.50	5.95	236.44	10.55	31.33	150 ± 15.40	0.63 ± 0.03

Concerning rabbit feeding trials, the results of PER for animals fed on the eight formulated experimental diets: Unblended cocoyam flour, unblended soya bean flour, 10% blend of soya bean and cocoyam flours, 20% blend of soya bean and cocoyam flours, 30% blend of soya bean and cocoyam flours, 40% proportion of soya bean and cocoyam flours, 50% proportion of soya bean and cocoyam flours and control diet.

Conclusion: This study revealed that cocoyam flour has the lowest protein quality as reflected in the value of the protein efficiency ratio, and lower biological value. The least biological value was recorded by the control diet. The best blend of cocoyam and soya bean flours obtained based on chemical and nutritional evaluations was the 50% blend of soya bean and cocoyam flours.

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