BAKING STUDIES WITH CASSAVA AND YAM FLOUR. I. BIOCHEMICAL COMPOSITION OF CASSAVA AND YAM FLOUR[1]

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ABSTRACT

Cassava (*Manihot utilissima*) and yam (*Dioscorea alata*) flour were analyzed to determine their biochemical composition. Definite differences were observed between these tuber flours with regard to their basic chemical composition. Yam flour had higher protein, ash, and fat content than did cassava flour, while the latter had higher starch and fiber content. The level of damaged starch in cassava flour was high, which may be an important consideration in the use of cassava flour for baking purposes. Sugar analysis revealed higher total sugar content in yam than in cassava flour. The individual free sugar pattern for these flours, although similar, differed in amounts for each sugar. The water-soluble nonstarchy polysaccharides (WSNP) extracted from the tuber flours differed in amount, protein content, and sugar composition. Diethylaminoethyl-cellulose chromatography of the yam WSNP suggested the presence of acidic polysaccharides in the extract. The lipids present in cassava and yam flour were markedly different with respect to their extractability and nature. The majority of the yam-extractable lipid was polar in nature, while the cassava was primarily nonpolar. Chemical score of the essential amino acids present in yam and cassava flour revealed that yam protein was of better quality than was cassava protein. The sulfur-containing amino acids were the first limiting amino acids for both tuber flours.

Cassava and yam flour have received considerable attention recently as potential components of composite flours (1–5). The biochemical constituents of these flours and their possible role and influence in dough and bread properties, however, have received little attention.

According to Rasper *et al.* (6), doughs containing cassava starch produced better loaves than did those containing yam starch. They suggested that the close gelatinization temperature and granule size of cassava and wheat starch may have been the cause of the results obtained. When flours instead of starches were used, however, yam flour produced the best loaves, indicating that other components in the nonwheat flours may have a greater effect than do the differences in functional properties of the starches. We (7) showed that the baking and farinograph response of tuber starch-reconstituted doughs depends on the physicochemical properties of the starch.

Hanh and Rasper (8) investigated the role of the water-soluble nonstarchy polysaccharides (WSNP) from yam and cassava flour in composite flours. Water-soluble fractions from all tubers investigated increased the loaf volume, whereas a detrimental effect was observed with the water-insoluble fraction. Rasper and MacDonald (9) concluded that the water-soluble fraction from cassava affected only the water absorption of the doughs, while the water solubles from some yam species exhibited a strengthening effect on the dough.

Hudson and Ogunsua (10) showed that the degree of starch damage of cassava

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flour had no effect on baking quality. They suggested that the presence of fiber in cassava flour was largely responsible for the inferior baking quality of cassava flour compared with that of cassava starch.

This study was undertaken to investigate the effect of cassava and yam flour in baking. The first part of this study involved an examination of cassava and yam flours to help to understand data obtained in dough and bread-baking studies using tuber flours.

MATERIALS

Tuber Samples

Commercial cassava (*Manihot utilissima*) flour of the variety Branca de Sta Catarina obtained from Brazil was used throughout this study. Yam (*Dioscorea alata*) flour of the variety Florido was also obtained from Brazil, but was prepared on laboratory scale from peeled and tunnel-dried yam tubers.

METHODS

Preliminary Tuber Flour Analysis

Protein, fat, ash, and diastatic activity of the tuber flours were determined according to AACC Methods 46-11, 30-10, 08-01, and 22-15, respectively (11).

Fiber was determined using the acid detergent method (12).

Starch content in the tuber flour was determined by the enzymatic procedure that Thivend *et al.* (13) described, with certain modifications. The sample (1.0 g) was dispersed in 25 ml of water, boiled with gentle stirring for 3 min, and heated at 20 lb of pressure at 127° C for 4 hr. Glucoamylase digestion was allowed to take place for 1 hr at 55° C, and starch content was determined on the liberated glucose using the glucose oxidase procedure.

Damaged starch was determined by its susceptibility to α-amylase hydrolysis according to the approved methods of the AACC (14).

Sugar Analysis

Extraction. The procedure of Ponte *et al.* (15) was used, with one modification. The sample was extracted twice using the ternary solvent system.

Total sugar determination. The phenol sulfuric acid method that Dubois *et al.* (16) described was used to determine the amount of sugar present in the extract. Sucrose was used to establish a standard curve, and the results were expressed as percentage of sucrose.

Reducing and nonreducing sugars. Reducing and nonreducing sugars were determined by the alkali ferricyanide procedure according to the approved method of the AACC (17).

Ion-exchange chromatography of sugars. Free sugars in the extract obtained using the ternary solvent system of Ponte *et al.* (15) were determined with the Technicon Sugar AutoAnalyzer system (Technicon Instruments Corporation, Tarrytown, NY). Sugar analysis was performed as described by Abou-Guendia and D'Appolonia (18) using the buffer gradient system Method I as described by Hough *et al.* (19).

WSNP

Isolation and purification. The WSNP were isolated according to the procedure of D'Appolonia (20) that is used for wheat flour pentosans. The only difference was that a flour water ratio of 1:4 and 1:10 was used for the yam and cassava flours, respectively. The crude WSNP were purified according to D'Appolonia and MacArthur (21).

Diethylaminoethyl (DEAE)-cellulose chromatography. DEAE-cellulose chromatography, in the borate form, of the purified WSNP from yam was done using a 2 × 50-cm column. The column was prepared according to the procedure of Neukom and Kundig (22). The sample (125 mg) was dissolved in a small amount of water and applied to the top of the column. After the sample had been allowed to penetrate into the DEAE-cellulose, elution was accomplished stepwise with the following eluants: 1) distilled water, 2) 0.0025*M* $Na_2B_4O_7$, 3) 0.025*M* $Na_2B_4O_7$, 4) 0.125*M* $Na_2B_4O_7$, and 5) 0.4*N* NaOH. The nitrogen content of each fraction was determined by the method of Folin-Ciocalteu as modified by Lowry *et al.* (23).

Sugar composition. Each of the DEAE-cellulose fractions obtained (5–10 mg) was hydrolyzed with 5 ml of 1*N* H_2SO_4 for 5 hr at 100° C, neutralized with barium carbonate, and centrifuged. The sugars were transformed into alditol acetates (24) and analyzed by gas-liquid chromatography (GLC) using a column packed with 3% ECNSS-M on Gas Chrom Q, 100–120 mesh, using fucitol acetate as the internal standard.

Lipids

Extraction. Tuber flour (4 g) was extracted with petroleum ether (30–60° C bp) for 12 hr in a Soxhlet extractor. After petroleum ether extraction, the flour residue was allowed to dry and then extracted with 40 ml of water-saturated *n*-butanol (WSB) for 30 min as Lin *et al.* (25) described. The solids were removed by vacuum filtration and reextracted two more times with WSB. The WSB extracts were combined and purified according to Lin *et al.* (25). The petroleum ether-extracted lipids were used for free fatty acid (FFA) isolation or combined with the WSB extract for polar and nonpolar lipid fractionation.

Fractionation of polar and nonpolar lipids. The petroleum ether-WSB–extracted lipid was fractionated into polar and nonpolar fractions by preparative thin-layer chromatography (TLC) (25). Polar lipids were considered to be those having rf values less than the monoglyceride fraction, and nonpolar lipids as those having rf values greater than and including the monoglyceride fraction.

FFA extraction. FFAs were isolated from the petroleum ether-extracted lipids. The lipid material (20–50 mg) was streaked on a TLC plate coated with Adsorbosil-5 (0.50 mm thick). Linoleic acid was spotted as the reference lipid. During application, a continuous stream of nitrogen was maintained on the plate surface to avoid oxidation.

After development in petroleum ether/diethyl ether/acetic acid (70:30:1 v/v), the plates were visualized briefly with iodine vapor. The band corresponding to the FFA fraction was marked, the iodine removed with N_2, and the fraction scraped from the plate. The FFAs were eluted from the coating material with 50 ml of chloroform/methanol solution (1:1 v/v). During stirring, a nitrogen atmosphere was maintained. The mixture was centrifuged at 1,130 × *g* for 10 min, the liquid phase decanted, and the Adsorbosil-5 reextracted twice. The combined extracts were evaporated under reduced pressure, the vacuum released with nitrogen, and the FFAs dissolved in 20 ml of chloroform. After elimination of the fines by centrifugation at 33,000 × *g* for 10 min, the extract was evaporated to dryness and the flask weighed.

GLC of fatty acid methyl esters. The methylated FFA fraction (26) and the methylated fatty acid from the petroleum ether-extracted lipid (27) were qualitatively and quantitatively determined by GLC. A Barber Colman Series 5000 Gas Chromatograph equipped with a flame ionization detector and an 8-ft glass column packed with gas-chrom P, 100–120 mesh, coated with diethylene glycolsuccinate, 10% by weight, was used.

Amino acid analysis. Basic, acidic, and neutral amino acids were determined on a Beckman Model 120B amino acid analyzer after acid hydrolysis of the tuber flour samples. The procedure of Spackman *et al.* (28) was used as modified by Benson and Patterson (29) and shortened to a 2 hr procedure by Beckman Instruments Inc. (Fullerton, CA).

RESULTS AND DISCUSSION

Preliminary Analysis of Cassava and Yam Flour

Data representing the preliminary analysis of the tuber flours are presented in Table I. Protein, ash, and fat content were higher in yam than in cassava flour. The higher protein content in yam compared with that in other tubers has been reported previously (30,31). Cassava flour contained higher levels of fiber, starch, diastatic activity, and damaged starch than did yam flour. The high content of damaged starch present in cassava flour may cause some restrictions on the amount of this flour that could be used in blends with wheat flour for bread-baking purposes.

Sugar Analysis

Total sugar content, reducing and nonreducing sugar values, and individual free sugar analysis for both tuber flours are presented in Table II. The total sugar content was similar to the sum of the individual free sugars, indicating that the total sugar content was composed primarily of individual free sugars and little if any oligosaccharide-type material. Both tuber flours had higher amounts of total

TABLE I
Preliminary Tuber Flour Analysis[a]

Tuber Source	Protein[b] (%)	Ash (%)	Fat (%)	Starch (%)	Fiber (%)	Diastatic Activity (%)	Starch Damage (%)
Yam	7.8	3.18	1.30	70.9	2.10	1.63	5.7
Cassava	1.2	0.75	0.67	88.4	4.66	4.30	45.6

[a]All results reported on dry basis.
[b]Calculated on basis of N × 6.25.

sugar than did wheat flour (32), with yam flour having the highest amount. The ratio of reducing to nonreducing sugars was approximately 1 for both tuber flours. The individual free sugar analysis for yam and cassava flour was similar, although the amounts differed. Sucrose was the main individual free sugar, followed by glucose and fructose. Besides these three sugars, traces of maltose and raffinose were detected in cassava flour. These results were somewhat similar to those reported for cassava (33) and yam (34). In the previous studies, however, higher levels of fructose than of glucose were reported for the particular species of cassava and yam investigated.

WSNP

Data on the isolation, purification, and sugar composition of the WSNP from cassava and yam flour are shown in Table III. Cassava yielded a higher amount of crude WSNP than did yam, but the protein content present in the yam-extractable material was about 11 times higher than that found in cassava. As noted in Table I, the protein content of the yam flour was considerably hieher than that of the cassava flour. Cassava crude WSNP contained essentially

TABLE II
Total Sugar Content, Reducing and Nonreducing Sugar Values, and Free Sugar Composition of Cassava and Yam Flour[a]

Tuber Source	Total Sugar (%)	Reducing Sugar (%)	Nonreducing Sugar (%)	Free Sugar Composition: Sucrose (%)	Glucose (%)	Fructose (%)	Maltose (%)	Raffinose (%)
Cassava	3.10	1.97	1.71	1.69	0.92	0.43	Trace	Trace
Yam	5.23	2.55	2.75	3.59	1.09	0.65	0.00	0.00

[a]All results are reported on dry basis.

TABLE III
Yield, Protein, and Sugar Composition of the Water-Soluble Nonstarchy Polysaccharides (WSNP) From Cassava and Yam Flour

Tuber Source	Yield (%)	Protein (%)	Sugar Composition: Arabinose (%)	Xylose (%)	Glucose (%)	Mannose (%)	Galactose (%)
			Crude WSNP				
Cassava	4.1[a]	4.6	Trace	Trace	100.0	Trace	Trace
Yam	0.9[a]	52.4	10.0	Trace	22.1	54.0	14.0
			Purified WSNP				
Cassava	1.2[b]	5.8	36.5	7.3	15.3	8.7	32.3
Yam	15.0[b]	11.0	17.2	Trace	Trace	54.0	28.7

[a]Yield from flour expressed on dry basis.
[b]Yield expressed on basis of crude WSNP.

glucose as component sugar. This result in conjunction with the dramatic decrease in WSNP yield on α-amylase treatment indicated the presence of high levels of soluble starch in the water extract. The cassava α-amylase–treated WSNP were mainly made up of arabinose and galactose followed by glucose, mannose, and xylose. The yam WSNP extract contained primarily mannose, followed by arabinose and galactose. The water extract from cassava or yam flour, unlike the wheat flour water extract (35), did not have appreciable amounts of xylose.

Table IV shows pertinent data for the DEAE-cellulose fractions of the WSNP extracted from yam. Cassava WSNP were not fractionated due to the extremely small amounts obtained after α-amylase treatment (Table III). The recovery from the DEAE-cellulose column was low, suggesting that some material was strongly bound to the column support and was not eluted with the solvent system used. Fraction IV had lower protein than did fraction V; it was composed primarily of arabinose and galactose, which suggested the presence of an arabinogalactan in the WSNP extracted from yam flour. D'Appolonia *et al.* (35) found an arabinogalactan in fraction IV when the water-soluble polysaccharides from wheat flour were fractionated by DEAE-cellulose. The sugar composition of fraction V resembled that of the original extract; mannose was the predominant sugar, followed by galactose, glucose, arabinose, and xylose.

TABLE IV
DEAE-Cellulose Column Chromatography of Water-Soluble Nonstarchy Polysaccharides (WSNP) Isolated From Yam

			Sugar Composition				
Fraction[a]	Recovery[b] (%)	Protein (%)	Arabinose (%)	Xylose (%)	Glucose (%)	Mannose (%)	Galactose (%)
F_{IV}	16.0	8.9	37.1	0.0	00.0	10.9	52.1
F_V	12.0	21.7	15.7	6.0	15.2	37.1	27.1

[a]F_I, F_{II}, and F_{III} were not analyzed due to limited amount of material recovered.
[b]Based on amount of WSNP fractionated.

TABLE V
Extractability and Nature of Cassava and Yam Flour Lipids

Tuber Source	Petroleum Ether Extract		Petroleum Ether + WSB Extract		
	Yield[a] (%)	FFA[b] (%)	Yield[a] (%)	Polar[b] (%)	Nonpolar[b] (%)
Cassava	0.24	22.7	0.57	26.0	74.0
Yam	0.29	35.0	0.43	66.3	33.7

[a]Yield from flour expressed on dry basis.
[b]Expressed as percent of extracted lipid.

Lipids

Lipids of cassava and yam flour were analyzed with respect to their extractability and kind; Table V shows the results. For the sake of clarity, total extractable lipid was obtained by successive extractions with a nonpolar solvent (petroleum ether) and a polar solvent (WSB). The majority of the cassava lipid determined by acid hydrolysis (Table I) was extractable, while that from yam was only partially extractable. The amount of petroleum ether-extracted lipid, however, was similar for both flours. The total extractable lipid from cassava was mainly nonpolar, while that from yam flour was polar in nature. The petroleum

TABLE VI
Fatty Acid Composition of Cassava and Yam Flour

Fatty Acid	From FFA[a] Fraction		From Petroleum Ether Extract	
	Cassava (%)	Yam (%)	Cassava (%)	Yam (%)
Palmitic	25.4	23.3	34.7	27.9
Stearic	2.1	3.0	3.1	3.0
Oleic	35.6	5.8	39.0	5.5
Linoleic	27.9	53.1	17.6	53.5
Linolenic	9.0	9.1	5.6	9.9

[a]FFA = free fatty acid.

TABLE VII
Amino Acid Composition of Cassava, Yam, and Wheat Flour[a]

Amino Acid	Cassava	Yam	Wheat (HRS)[b]
Lysine	5.37	4.47	1.9
Histidine	2.32	2.61	2.1
Arginine	11.86	9.94	3.4
Aspartic acid	10.40	11.56	3.9
Threonine	3.56	3.81	2.6
Serine	4.57	6.71	4.9
Glutamic acid	22.45	13.90	37.2
Proline	3.17	4.30	12.5
Glycine	3.19	3.66	3.2
Alanine	4.76	4.56	2.9
Cystine	0.00	0.33	2.2
Valine	4.33	4.89	4.3
Methionine	0.63	1.37	1.5
Isoleucine	3.22	4.19	3.9
Leucine	5.25	7.12	6.8
Tyrosine	0.89	2.68	2.7
Phenylalanine	3.26	5.42	5.2

[a]Expressed as grams of amino acid per 16 g of nitrogen, calculated to 100% N recovery from chromatographic analysis. Percent of nitrogen recovered from column for cassava was 80.4% and for yam 83.1%.

[b]Values from Tkachuk (38). HRS = hard red spring.

ether extract from either cassava or yam flour contained an appreciably higher amount of FFA than did wheat flour (36).

The fatty acid composition from the isolated FFA and from the petroleum ether-extracted lipid is shown in Table VI. Palmitic, stearic, oleic, linoleic, and linolenic acids were present in the isolated FFA fraction as well as in the petroleum ether-extracted lipid. Palmitic and linoleic acids were present to the largest extent in yam, while cassava contained, in addition to these two fatty acids, oleic acid in relatively high amounts. The fatty acid composition for cassava agreed with previously reported results (37).

Amino Acid Composition and Protein Quality

The levels of essential, semiessential, and nonessential amino acids for yam and cassava flour are summarized in Table VII. For comparison, the composition for wheat protein (38) is included in Table VII. Tryptophan was not included, since this amino acid is destroyed on acid hydrolysis.

From the data in Table VII, one may observe that both cassava and yam flours contain low amounts of methionine and cystine. The amino acid composition pattern for cassava and yam was somewhat similar. Glutamic acid, aspartic acid, and arginine represented the main amino acids. Yam contained higher amounts of all of the amino acids than did wheat except for proline, glutamic acid, and the sulfur-containing amino acids (38). In cassava, only lysine, arginine, aspartic acid, threonine, and alanine were higher than in wheat.

Protein quality was evaluated in terms of chemical score. Chemical score is defined as the percent of each essential amino acid compared with the percent of the same amino acid present in an "ideal" protein pattern. The "ideal" protein pattern used in this study was the 1971 FAO/WHO provisional amino acid score pattern (39). Chemical scores of the essential amino acids (except tryptophan) for cassava, yam, and wheat flour are given in Table VIII.

The sulfur-containing amino acids were the first limiting amino acids for both cassava and yam protein, which agrees with previously reported results (30,40,41). The second and third limiting amino acids for yam were lysine and

TABLE VIII
Chemical Score[a] of the Essential Amino Acids for Cassava, Yam, and Wheat Flour

Amino Acid	Cassava	Yam	Wheat
Isoleucine	81	105	95
Leucine	75***	102	96
Lysine	98	81**	42*
Methionine + cystine	18*	49*	123
Phenylalanine + tyrosine	69**	135	125
Threonine	89**	95**	70**
Valine	87	98	90***

[a]Calculated using provisional scoring pattern of Food and Agriculture Organization of the United Nations (39).
*First limiting amino acid.
**Second limiting amino acid.
***Third limiting amino acid.

threonine, respectively. Phenylalanine plus tyrosine, and leucine were the second and third limiting amino acids, respectively, for the cassava protein.

Protein quality of yam was somewhat better than was wheat protein in terms of chemical score, while cassava protein was poorer than wheat protein.

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