Effect of Processing Conditions on Protein Extraction and Composition and on Some Other Physicochemical Characteristics of Parboiled Rice

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ABSTRACT

The effect of parboiling on three rice varieties was evaluated. Even light parboiling practically eliminated breakage after milling. It caused a slight protein increase in the milled rice, increased grain length, and soluble starch. It also darkened the color of the milled and decreased water absorption of the cooked rice. It considerably altered protein solubility in various solvents. The decrease of protein solubility in an ionic-detergent solution clearly differentiates raw from parboiled rice and might be developed for use as an assessment of parboiling, especially for waxy rice where blue values cannot be determined.

The purpose of this investigation was to measure certain changes occurring in rice grains when they are parboiled. These changes may serve to indicate the severity of the parboiling treatment.

Roberts et al. (1) found that the level of soluble starch and the degree of expansion of the grain may be used as an objective evaluation of the degree of parboiling. It has also been reported (1,2,3) that light-reflectance measurements could be used for the same purpose. Kurien et al. (4) found that the water-absorption capacities were significantly lower for parboiled rice than for raw rice cooked for the same period. However, samples of raw and parboiled rice cooked to an equivalent degree of softness indicated that parboiled rice could absorb more water without losing its shape.

A number of papers have been published on the extraction and composition of the proteins of brown and milled raw rice (5,6,7,8,9,10), but as far as we know no data have yet been published on the protein composition of parboiled rice.

Hence, the main emphasis in the present investigation was placed upon the extraction and composition of the proteins from parboiled rice subjected to different processing conditions.

Kik (11) reported a protein equivalent ratio (PER) of 1.93 for parboiled rice and 1.74 for milled rice. This would indicate no loss of nutritional value as a result of parboiling.

McIntyre and Kymal (8) successfully used an anionic detergent of the alkyl aryl-sulfonate type as an extractant for removing the proteins from rice. This extraction was most efficient when sodium bisulfite and sodium carbonate were added to an aqueous solution of the detergent.

In the present work similar extractants were used for studying the effect of parboiling on the extraction and composition of rice proteins.

Although little is known about the exact mode of action of detergents with protein, the former are thought to react with hydrophobic groups (12). Using

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casein, recent spectroscopic work (13) has pointed to the possible involvement of tyrosine and tryptophan residues.

MATERIALS AND METHODS

Representative paddy samples of three varieties, Stirpe 136, K-7821, and (Rexoro X Red) X Century Patna (RRCP), all harvested in September 1969 from a rice-breeding farm of the Institute of Cereals, Thessaloniki, Greece, were used. Broken, undersized, and immature grains were removed by sieving. The samples were stored for 3 to 3.5 months at ordinary room temperature and humidity prior to parboiling. For purposes of comparison, three commercial samples of milled raw rice and six samples of parboiled rice of different origins were included in the samples tested.

To study the effect of steaming, 150-g. samples of paddy rice were steeped in water at 64°C. for 3.5 hr. and drained. The moisture content for Stirpe was 32.6%; for K-7821, 31.8%; and for RRCP, 30.1%. The samples were then steamed together for 5 min. in a Prestige pressure cooker (capacity 8 pints) at 5, 10, and 15 p.s.i. (equivalent to 108.4°, 115.2°, and 121.0°C.).

To study the effect of steeping, 150-g. samples of paddy were steeped in water at 64°C. for 3.5 hr.; 5 hr., 35 min.; 5 hr., 58 min.; and 7.5 hr., and drained. Samples of the three varieties with the same steeping time were steamed simultaneously at 10 p.s.i. After parboiling, all samples were air-dried in the absence of direct sunlight to avoid any stress in the kernels. All samples were milled in a Minghetti mill (Verzelli, Italy) (50 g. for 5 min.). At the time of milling, the raw and parboiled paddy samples had moisture contents of 11 to 12.7%. The rice flour required for analysis was milled in an 8-in. Christy and Norris laboratory hammer mill (Chelmsford, Engl.). The sieve perforations were 0.3 mm., and about 92% of the resulting flour would pass a 100-mesh (B.S.S.) sieve.

The breakage was defined as the amount in gram of grains of less than three-fourths size per 100 g. of milled rice, and was determined manually. Length and breadth of 20 whole grains selected at random from the raw and parboiled milled samples were also measured and mean values calculated in order to determine if parboiling affected these dimensions.

Water absorption was determined as follows: Five grams of milled rice was placed into a beaker 5 cm. in diameter and 10 cm. high, containing 40 ml. of distilled water. The beaker was then covered with a watch glass and immersed into a boiling-water bath for 23 min. The contents were then poured into a 6-cm. diameter Büchner funnel and drained for 5 min., the funnel being covered with a watch glass. The grains were then quickly transferred to a weighing dish and weighed. Each test was performed in duplicate. The water absorption was expressed as the milliliters of water absorbed by one gram of rice.

The gelatinization temperature of the three rice varieties was measured by the Brabender Amylograph with 100 g. of ground (40-mesh) milled raw rice (14) and 400 ml. of water. (Moisture content, 11.0 to 11.8%.)

The color of the dry whole-grain milled samples was measured by a Colormaster differential colorimeter (15) (MEECO Instruments, Hatboro, Pa.). Three measurements of green (G), red (R), and blue (B) reflectance were made. For measurement, the milled rice was placed into a specially prepared cell. This was constructed by attaching pieces of black perspex to the edges of a thin glass plate

and the reflectance of the surface layer of the grain measured at the bottom of the cell. Cell dimensions were 1 X 5 X 5.5 cm., providing a sample depth of 1 cm. The instrument was standardized by two white tiles. The calibration values of the tile placed on the left-hand sample aperture were G = 86.91, R = 86.98, and B = 84.69. Those of the tile placed on the standard aperture (right-hand) were G = 86.77, R = 86.96, and B = 84.25.

Soluble starch was calculated from the "blue values" as determined by Roberts et al. (1). These were determined at 600 nm. with a 1-cm. cell, but a 20-ml. aliquot of filtrate was used instead of the 10 ml. proposed by Roberts et al. For standardization, a graph was plotted at various concentrations of soluble starch (May and Baker No. 60039) against their corresponding "blue values". Two separate determinations were made.

The protein of the rice flour was extracted by a modification of the percolation method of Maes (16). The sample size had to be reduced to avoid plugging of the column by the samples, especially the parboiled ones. Five grams of flour was mixed with a metallic spatula on a glazed sheet of paper with 10 g. of finely ground pumice (200 mesh), taking care to avoid agglomeration of the flour. This was then thoroughly mixed on the same sheet of paper with 100 g. of sand (Merck No. 7712, acid-washed and ignited sea sand). The percolation column (25 X 3 cm.) was plugged with glass wool and packed with successive layers of quartz (Merck No. 7536), sand, sample mixture, sand, and quartz. The quartz and sand layers were about 1.5 cm. thick. The column was gently compacted by knocking, and covered at the top by glass wool held in place by a few glass balls. All extractions were then carried out in a constant temperature room at 25°C. At first the flour was defatted by extracting the column with 150 ml. of acetone and then successively with 250 ml. of distilled water, 250 ml. of 5% sodium chloride solution, 150 ml. of 60% ethanol, 250 ml. of 3% detergent (natural pH 4.6), and 250 ml. of a mixture of 3% detergent, 2% sodium carbonate, and 0.2% sodium bisulfite (natural pH 10.8). In this way fourteen columns could be extracted simultaneously.

The detergent used was a preparation of a straight-chain sodium alkyl benzene sulfonate (mean carbon chain length C_{11}) prepared from the powdered trade product NANSA S40/S (Albright and Wilson Ltd., Whitehaven, Engl.) by successive precipitations from ethanol and 1:1 acetone-ethyl ether (17). The composition of the detergent was claimed to be: Active material, 40%; sodium phosphate, trace; water, 3%; sodium sulfite, to 100%.

The relative extraction efficiency of four of the solvents used was determined as follows: 1 g. of rice flour was extracted with 20 ml. of solvent at 25°C. in 50-ml. plastic centrifuge tubes for 6 hr. A flask shaker was used to ensure thorough agitation with the tubes placed in a horizontal position. After extraction, the tubes were centrifuged at 2,500 r.p.m. for 8 to 10 min. in an M.S.E. "Minor" centrifuge with a 5.1-in. head, and nitrogen determined on a 10-ml. aliquot of the supernatant solution.

The nitrogen was determined, always in duplicate, by the macro-Kjeldahl method using the catalyst proposed by Matveef (18). The only modification made was that the catalyst was dissolved in advance in sulfuric acid. This solution is readily prepared by heating a 500-ml. Kjeldahl flask containing 57 g. of anhydrous sodium sulfate, 3 g. of selenium powder, and 270 ml. of sulfuric acid till a clear solution is

obtained. Nine milliliters of this solution was used for the digestion of 1 g. of rice flour. The nitrogen eluted with the first three liquids and collected from the columns was determined on aliquots of 100 ml. by 15 ml. of the catalyst solution. Nitrogen of the two last liquid extracts containing the detergent was determined on 50-ml. aliquots with 30 ml. of the catalyst solution. Silicone (Hopkins and Williams Ltd. No. 996402) was added to eliminate foaming during digestion and distillation. Crude protein was calculated by using a Kjeldahl factor of 5.95. Moisture content was determined by heating the samples at 130°C. for 1 hr. in a Brabender oven. The UV spectra of the column eluates containing detergent were obtained with a Unicam SP 1800 recording spectrophotometer against pure solutions of the extractants.

RESULTS AND DISCUSSION

Parboiling slightly increased the protein content of the milled rice (Tables IA and IIA). This was not unexpected because the grains become harder and less polishings, which are high in protein content, are removed (19). The soluble starch is directly

TABLE IA. EFFECT OF STEAMING PRESSURE ON SOME CHARACTERISTICS AND ANALYTICAL DATA OF PARBOILED RICE (MEAN DATA FOR THE THREE RICE VARIETIES)

(steeping time 3.5 hr.; steaming time 5 min.)

Steaming Pressure p.s.i.	Milling Breakage %	Dimensions of Milled Grains		Color of Milled Grains (B reading)	Cooking Test-Water Absorption ml./g.	Protein (N × 5.95) % (d.m.)	Soluble Starch mg./g. (d.m.)
		Length mm.	Breadth mm.				
Raw	6.2	5.67	2.27	36.5	2.71	6.64	5.4
5	0.7	5.81	2.32	30.4	2.32	6.76	8.3
10	0.5	5.82	2.30	27.6	2.22	6.84	14.5
15	0.4	5.94	2.25	24.9	2.03	6.81	23.6
L.S.D. 5%							±8.49
1%							±12.85

TABLE IB. EFFECT OF STEAMING PRESSURE ON PROTEIN EXTRACTION OF PARBOILED RICE (MEAN DATA FOR THE THREE RICE VARIETIES)

Steaming Pressure p.s.i.	Fractionation by Percolation: % of total protein soluble in:						Extraction Efficiencies of Two Solvents on Rice Proteins by Shaking	
	Water	NaCl 5%	Alcohol 60%	Detergent 3%	Alkaline detergent containing bisulfite	Extraction efficiency	Detergent 3%	Alkaline detergent containing bisulfite
Raw	0.9	6.7	2.3	32.1	53.6	95.6	73.4	98.3
5	2.5	0.3	0.4	12.4	67.5	83.2	31.1	94.5
10	2.7	0.4	0.4	8.2	68.5	80.2	25.9	95.2
15	2.7	0.3	0.4	4.9	74.2	82.6	20.1	90.8
L.S.D. 5%	±0.55	±1.50	±0.34	±3.82	N.S.	N.S.	±7.15	N.S.
1%	±0.83	±0.52	±2.27	±5.79			±10.82	

TABLE IIA. EFFECT OF STEEPING TIME ON SOME CHARACTERISTICS AND ANALYTICAL DATA OF PARBOILED RICE (MEAN DATA FOR THE THREE RICE VARIETIES)

(Steaming pressure 10 p.s.i.; steaming time 5 min.)

Steeping Time hr.	Milling Breakage %	Dimensions of Milled Grains Length mm.	Dimensions of Milled Grains Breadth mm.	Color of Milled Grains (B reading)	Cooking Test-Water Absorption ml./g.	Protein (N X 5.95) % (d.m.)	Soluble Starch mg./g. (d.m.)
Raw	6.2	5.67	2.27	36.5	2.71	6.67	5.4
3.50	0.5	5.82	2.30	24.1	2.22	6.84	14.5
5.58	0.4	6.10	2.24	25.8	2.12	6.78	17.0
7.00	0.4	5.99	2.16	25.0	2.03	6.81	19.1
L.S.D. 5%							±4.87
1%							±7.37

TABLE IIB. EFFECT OF STEEPING TIME ON PROTEIN EXTRACTION OF PARBOILED RICE (MEAN DATA FOR THE THREE RICE VARIETIES)

(Steaming pressure 10 p.s.i.; steaming time 5 min.)

	Fractionation by Percolation % of total protein soluble in:						Extraction Efficiencies of Two Solvents on Rice Proteins by Shaking	
Steeping Time hr.	Water	NaCl 5%	Alcohol 60%	Detergent 3%	Alkaline detergent containing bisulfite	Extraction efficiency	Detergent 3%	Alkaline detergent containing bisulfite
Raw	0.9	6.7	2.3	32.1	53.6	95.6	73.4	98.3
3.50	2.7	0.4	0.4	8.2	68.5	80.2	25.9	95.2
5.58	2.7	0.3	0.4	6.1	69.0	78.5	24.5	92.1
7.00	3.2	0.3	0.4	6.8	66.0	76.7	22.2	91.5
L.S.D. 5%	±0.97	±1.44	±0.36	±4.29	N.S.	±12.25	±7.62	N.S.
1%	±1.48	±2.18	±0.54	±6.49		...	±11.54	

proportional to steaming pressure (Table IA), but depended strongly on the sample. No correlation appeared to exist between the soluble starch produced by parboiling and the gelatinization temperatures of the corresponding raw starches. For example, at 15 p.s.i. the former was 15.3, 34.6, and 20.9 mg. per g. for Stirpe 136, K-7821, and RRCP, respectively. The latter were found to be 64.5°C. for Stirpe 136, and 71.5°C. for both K-7821 and RRCP.

The water absorption of the cooked rice decreased progressively as the severity of the parboiling treatment increased (Table IA). The improvement in milling quality of paddy owing to parboiling is well known. Table IA shows that even light treatment practically eliminated milling breakage. The average breadth of the lightly parboiled grains was slightly greater and that of the fully gelatinized grains slightly less than that of the raw milled rice (Tables IA and IIA). The length of the parboiled rice grains was greater than that of raw rice grains. This observation is in disagreement with the results obtained by Kurien et al. (4), perhaps because of different parboiling conditions and different rice samples.

The visual appraisal of kernel color of the milled-rice samples appeared to correlate satisfactorily with the blue reflectance readings (B readings). These tend to decrease with steaming pressure (Table IA) (i.e., darkening of color).

In Fig. 1 the percentages of total protein soluble in 3% detergent solution by either the percolation or the shaking procedure are plotted against steaming pressure. Protein solubility decreases rapidly at first and then rather more slowly as the steaming pressure increases. The UV absorption peak of the detergent extracts obtained by percolation was at 285 nm. and that of the alkaline detergent extracts at 290 nm. As is apparent from Tables IB, IIB, and IIIB, the sum of protein extracted with the first four extractants in sequence through the percolation method is much less than the protein extracted by detergent solution alone with the shaking technique. Nevertheless, when direct percolation of samples 1 and 4 (Table IIIB) was carried out with 250 ml. of detergent solution, the percentages of total protein extracted were 70.33 and 16.90, respectively. These extractions are similar to those obtained with 6-hr. shaking, namely 73.22 and 19.51%. It appears that an insolubilizing effect of some column extractants, possibly acetone (20) or the aqueous alcohol (21), plays a role.

TABLE IIIA. ANALYTICAL DATA OF COMMERCIAL RICE SAMPLES

Sample No.	Grain Type	Treatment	Source	Color of Milled Grains (B-reading)	Protein (N X 5.95) % (d.m.)	Soluble Starch mg./g. (d.m.)
1	Long	Raw	U.S.	30.1	8.55	5.7
2	Long	Raw	Thailand	34.4	7.93	4.8
3	Short	Raw	China (Shoonan)	36.1	7.94	7.3
4	Long	Parboiled	U.S.	19.4	8.03	22.8
5	Long	Parboiled	Thailand	19.8	7.88	15.8
6	Long	Parboiled	"Uncle Ben's"	20.9	7.54	17.0
7	Long	Parboiled	"Whitworth's"	21.1	8.04	25.4
8	Long (waxy)	Parboiled	Thailand	19.5	7.38	...
9	Medium (waxy)	Parboiled	China	20.0	9.35	...

TABLE IIIB. SOLUBILITY FRACTIONATION OF PROTEINS OF COMMERCIAL RICE SAMPLES

	Fractionation by Percolation % of total protein soluble in:						Extraction Efficiencies of Two Solvents on Rice Proteins by Shaking	
Sample No.	Water	NaCl 5%	Alcohol 60%	Detergent 3%	Alkaline detergent containing bisulfite	Extraction efficiency	Detergent 3%	Alkaline detergent containing bisulfite
1	1.0	6.7	1.9	34.9	52.4	96.9	73.2	96.8
2	2.4	1.7	1.6	...	...	...	67.1	96.8
3	...	...	...	...	...	...	77.9	100.0
4	2.4	0.3	0.3	5.9	69.2	78.2	19.5	88.1
5	0.8	0.3	0.4	...	...	...	16.5	85.2
6	1.8	0.5	0.4	9.2	70.7	82.5	20.0	93.7
7	...	...	...	...	...	...	21.3	84.2
8	...	...	...	...	...	...	21.6	92.0
9	2.7	0.5	0.3	7.5	60.2	71.2	18.0	73.9

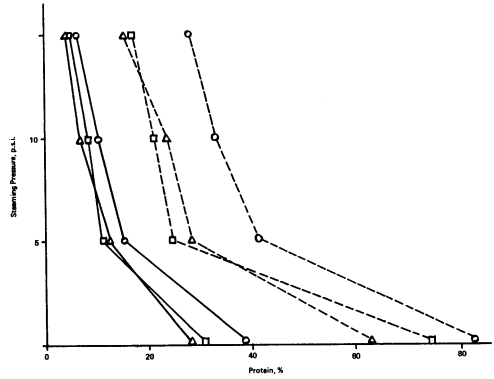

Fig. 1. Effect of steaming pressure on the percentage of total protein soluble in 3% detergent solution. Circles = Stirpe 136; triangles = K-7821; squares = RRCP; straight lines = percolation; and broken lines = shaking.

An almost quantitative extraction of the proteins, especially those of the raw-rice samples, was obtained with 6-hr. shaking in the presence of alkaline detergent solution and bisulfite (Table IB). The extraction efficiency on the column with five successive extractants is also very high, especially for the raw-rice samples. Such high column efficiencies have never been described in the literature before with other extractants (9,10). Column extraction of the crude glutelin fraction of parboiled samples by means of 0.1N sodium hydroxide (NaOH) could not be used because the column became impermeable. This was presumably due to the swelling effect of the NaOH on the already gelatinized starch.

The effect of parboiling on the protein fractions extracted successively on the column appears to cause an increase in the water-soluble fraction (albumin), a sharp decrease in the salt-soluble fraction (globulin), and a decrease in the alcohol-soluble fraction (prolamin), as compared with the raw-rice samples. The acetone extracts were free of nitrogen.

A protein-extraction trial of the raw and parboiled RRCP samples by shaking them with 0.1N NaOH showed that it was almost as effective for the raw sample (but considerably less so for the parboiled ones) as using the alkaline detergent solution containing bisulfite (mean extraction efficiencies 81.9 and 93.0, respectively).

A similar extraction trial of the same samples with 4% lactic acid (w./v.) had an extraction efficiency of 52.1 for the raw sample and 17.1 for the parboiled ones (mean value).

Although perhaps varietal or at least sample influence was pronounced, there seems to be a correlation between soluble starch, kernel color (B reading), and the percentage of protein soluble in 3% detergent solution by either the percolation or shaking technique, when different steaming pressures are used. The influence of steeping time on the different characteristics of parboiled rice appears to be of minor importance (Table IIA and IIB). Nevertheless, it appears that with increasing steeping times, the steaming needs to be less severe to obtain optimum parboiling treatment.

Different physicochemical characteristics of the commercial rice samples are apparent from Tables IIIA and IIIB. The behavior of these samples appears to be similar to that of the three varieties discussed so far. The parboiled samples are easily differentiated from the raw ones by low solubility in 3% detergent solution (Table IIIB).

The results reported here indicate that measurements of protein solubility in 3% detergent solution, either by percolation or preferably by shaking, clearly differentiate raw from parboiled rice and might be developed for use as an assessment of the degree of parboiling. Such measurements might be especially useful for waxy-rice samples where blue values cannot be determined. Nevertheless, the results of color, soluble-starch, or soluble-protein measurements should be interpreted with caution in connection with the evaluation of the degree of parboiling, since varietal or at least sample influence seemed to be of considerable importance.

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