Davis, P. L., Chace, W. G., Jr., Cubbedge, R. H., *HortScience* **2**, 168 (1967).
Davis, P. L., Harding, P. L., *Amer. Soc. Hort. Sci. Proc.* **75**, 271 (1960).
Davis, P. L., Smoot, J. J., *Citrus Ind.* **41**, 6 (1960).
Jahn, O. L., Yost, G. E., Soule, J., U. S. Department of Agriculture, Agricultural Research Service, 51-14 (1967).

Received for review November 3, 1972. Accepted February 12, 1973.

Canning and pH Stability of Epichlorohydrin-Treated Parboiled Rice

James E. Rutledge* and Mir Nazrul Islam

Rice starch in the parboiled grain was etherified by epichlorohydrin in an alkaline environment. Samples were evaluated after canning and retorting for 60 min at 240°F in semiliquid media. The treatment vastly improved the kernel stability for canning in excess water, even under acidic conditions. Costly agitation-type retorting, which allows less severe processing due to improved heat transfer, was not required for this rice because of its high thermal stability. Cross-linked samples showed approximately 68% less leaching at pH 7 and approximately 82% less at pH 5, as opposed to untreated samples. Taste panel evaluation involving color, cohesiveness, flavor, and doneness of cross-linked samples indicates a high potential for its incorporation in various canned formulations.

At the present time, very few commercially canned products contain rice. This is due to two primary reasons. The first involves the stability of the rice grain and the second involves the high cost of agitated retorting equipment. Most canned products which could use rice in the formulation require processing for approximately 60 min at 240°F or a somewhat shorter process at 250°F in conventional retorting equipment. In the course of processing, a point is reached where the hydrogen bonds responsible for the starch granular integrity are weakened and an irreversible swelling occurs. If processing is continued, the starch granules will eventually rupture, resulting in leaching of solids and grain distortion. With regard to the above, white rice is less resistant to thermal degradation than that which is parboiled. However, due to lengthy processing in conventional retorts, parboiled rice is still considered unsatisfactory. With agitated retorting, parboiled rice can be canned in various soup formulations in a fairly acceptable manner. The cost of such equipment, however, is the main limiting factor. Many food operations are seasonal or operate with limited capital and cannot afford the agitated type of process.

The purpose of this study was to produce a rice which could withstand processing conditions encountered in still retorting while maintaining the desirable organoleptic properties associated with rice.

MATERIALS AND METHODS

The cross-linking treatment imposed on parboiled rice consists of three main steps: activation, cross-linking, and neutralization.

Activation. One-hundred grams of a canner's quality parboiled Bluebelle rice (obtained from Uncle Ben's, Inc., Houston, Tex.) was weighed in a 500-ml Erlenmeyer flask. Two-hundred milliliters of a 0.1 *N* NaOH solution and 10 g of NaCl were added to the rice. The mixture was allowed to stand for 2 hr.

Cross-Linking. After soaking, 26 ml of freshly prepared 1% epichlorohydrin solution (1 ml of epichlorohydrin made up to 100 ml with 0.1 *N* NaOH) was added to the flask and allowed to react on a shaker for 4 hr. The flask was closed with a rubber stopper to prevent loss of volatile epichlorohydrin.

Neutralization. After cross-linking, the alkali–salt–epichlorohydrin mixture was decanted into a 1000-ml volumetric flask. The rice was washed with distilled water several times and the washing was added to the volumetric flask for subsequent determination of unreacted epichlorohydrin. The washed rice was resuspended in 100 ml of distilled water; about 4.5 ml of 4 *N* HCl was added slowly drop by drop throughout 5 hr. The pH was maintained above 4, otherwise the rice grain may undergo acid modification, which is characterized by heavy starch leaching during canning. The grains were neutralized to a pH of 6.5 and thoroughly washed with tap water and air dried at room temperature.

Estimation of Unreacted Epichlorohydrin. The reaction mixture and the washing, which were collected in the 1000-ml volumetric flask, were brought to volume with distilled water and filtered after standing in a cold room at 5° for 18 hr for retrogradation of the starch, which aided filtration; 50 ml of the filtrate was diluted to 100 ml and a 10-ml aliquot was pipetted into a 50-ml volumetric flask, and 1 ml of 2 *N* sodium hydroxide was added. An epichlorohydrin standard and a reagent blank were prepared in a similar manner. The stoppered flasks were heated on a water bath at 80° for 1.5 hr and then cooled, and 1 ml of 10 *N* sulfuric acid was added, followed by 5 ml of 0.1 *M* sodium periodate. The flasks were placed in the dark for 10 min, after which 5 ml of 1.0 *M* sodium arsenite was added and the solutions were diluted to 50 ml with distilled water. One-milliliter aliquots were then pipetted into test tubes; 10 ml of chromotropic acid reagent was added rapidly, accompanied by mixing of each of the tubes, which were then heated in a boiling water bath for 30 min. The tubes were removed, cooled to room temperature, and poured into cuvettes, and the absorbance was read at a wavelength of 570 mμ in a Bausch & Lomb Spectronic 20 using a water blank set at zero absorbance. The procedure was similar to that described by Hamerstrand *et al.* (1960).

Canning Evaluation. Forty cans of rice were used in the evaluation, 20 containing the epichlorohydrin-treated rice and the remaining 20 using the untreated parboiled sample. Both treated and control rice were canned at pH 7 and 5, which resulted in a 2 × 2 factorial arrangement of treatments. Fifteen grams of rice was used in each can. The cans were filled to a ½ in. headspace with boiling water of the appropriate pH and processed at 240°F for 60 min in a conventional still retort, after which the cans were water cooled.

Department of Food Science, Louisiana State University, Baton Rouge, Louisiana 70803.

Table I. Treatment Means for Percentage Solid Loss and Organoleptic Evaluation

	Treated		Control	
	pH 7	pH 5	pH 7	pH 5
% solid loss	7.32	6.62	23.14	36.06
Color	4.88	4.78	3.72	3.84
Cohesiveness	4.96	5.00	1.70	1.60
Flavor	4.96	4.78	4.30	4.40
Doneness[a]	3.16	3.18	1.14	1.20

[a] In the case of doneness, a score of 3 was excellent, whereas a score of 5 or 1 was considered underdone or mushy, respectively.

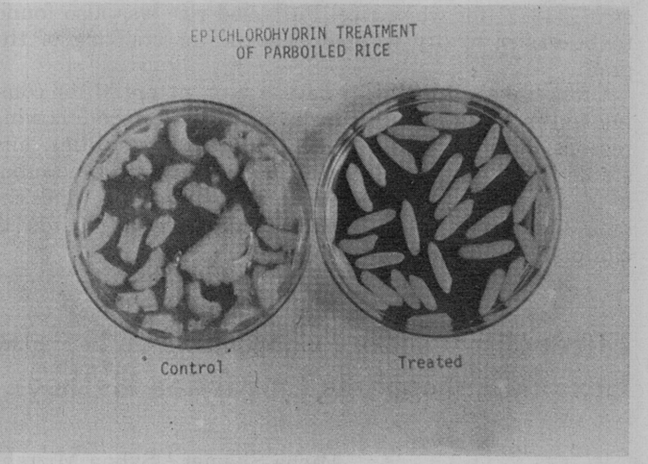

Figure 1. Canning stability of epichlorohydrin-treated parboiled rice.

Five cans from each treatment combination were used in the objective evaluation concerning percentage solid loss during canning. The solid loss was obtained by calculating the difference in weight between the dry matter content of the original sample and the dry matter content of the sample retained after washing over a 1.68 mm wire-mesh screen as outlined by Webb and Adair (1970). The remaining five cans in each treatment combination were evaluated subjectively by ten judges for color, off-flavor, cohesiveness, and doneness using a five point hedonic scale similar to that described by Batcher *et al.* (1956).

Data collected from the organoleptic evaluation were analyzed as a 2 × 2 factorial in a completely randomized design with five cans per treatment combination. The subjective evaluation was a 2 × 2 factorial in a randomized block design with ten judges as replications and five cans per treatment combination.

RESULTS AND DISCUSSION

The effects of cross-linking starch in the intact kernel of a grain such as rice appear to greatly increase its stability to thermal processing conditions. The treatment means concerning percentage solid loss are presented in Table I. Cross-linked samples showed approximately 68% less leaching at pH 7 and approximately 82% less at pH 5 as opposed to untreated samples. The striking point is that cross-linking brought the losses down to about the same value, regardless of the large differences in losses of the control samples at the two pH levels. Cross-linking treatment, pH, and the treatment × pH interaction were highly significant ($p < 0.01$) . After examining the treatment means in Table I and in view of the interaction, it is evident that low pH was more detrimental in the case of untreated samples.

Earlier work by Rutledge *et al.* (1972) has shown that rice receiving the alkaline treatment minus the cross-linking reagent showed even greater solids loss during canning than the control samples. Thus, the difference between cross-linked and untreated samples is no doubt related to increased starch granular stability toward thermal degradation and pH extremes. Figure 1 illustrates the well-defined grains of the cross-linked sample compared to the control, which shows considerable leaching with longitudinal splitting and fraying of edges and ends of the grains.

With the cross-linked samples no clumping or matting on the bottom of the can occurred; however, with the control samples approximately ¼ of the rice in the cans was matted. Figure 2 clearly illustrates the matting occurring in the control samples, which is no doubt due to the heavy starch leaching.

The treatment means for the organoleptic evaluation concerning color, flavor, cohesiveness, and doneness are presented in Table I. Treatment was highly significant ($p < 0.01$) with regards to all quality attributes, indicating the high potential for epichlorohydrin-treated parboiled rice in heat processes formulations. The pH of the canning liquor was nonsignificant ($p > 0.05$) with regard to the various organoleptic factors; however, there was a significant ($p < 0.05$) treatment × pH interaction concerning flavor, which indicates that pH has a greater influence on flavor with cross-linked samples as opposed to untreated samples. Regardless of the pH of the treated samples, the panel members consistently preferred the flavor of the cross-linked rice. The panel members preferred the texture of the cross-linked rice, whereas the control samples were considered overdone or mushy.

Cross-linking the starch in rice appears slightly more difficult than is the case with powdered or granular starch, evidently due to the higher degree of organization within the kernel or grain. In the same respect, the authors have noted that white rice appears to be more difficult than parboiled rice to cross-link, which has resulted in slight modifications of the treatments applied on parboiled rice as opposed to those described on white rice by Rutledge *et al.* (1972). The quantity of alkali added was increased from 1:1 (w/v) to 1:2 (w/v) for parboiled rice as compared to white rice previously reported to adjust for the greater swelling tendency associated with parboiled rice. Evidently, the greater water absorption tendency of parboiled rice reduced the neutralization time substantially. In support of the observation that white rice is somewhat more difficult to cross-link, data collected indicate that 61.8% of the epichlorohydrin added to parboiled rice reacted, as compared to 51.3% reported by Rutledge

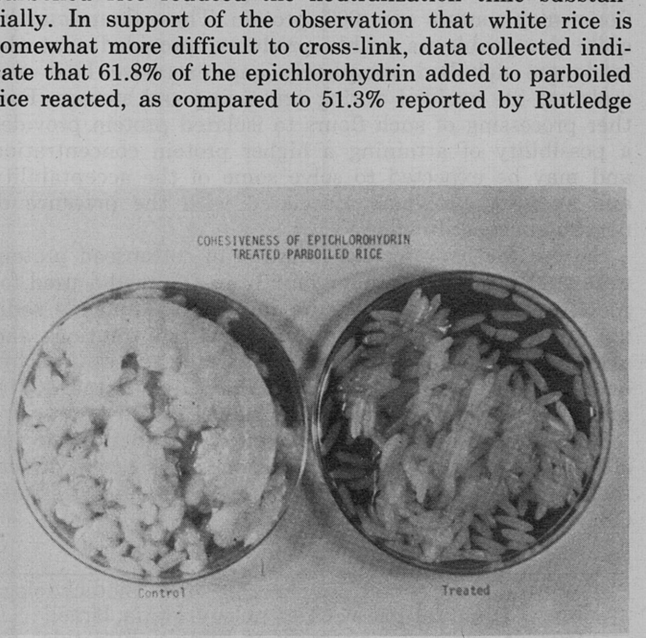

Figure 2. Cohesiveness of epichlorohydrin-treated parboiled rice after canning.

et al. (1972) for white rice. Parboiled rice was also found to be easier to dry without checking or cracking of the grain.

Cross-linked rice should have a high potential for commercial incorporation in canned products for the following reasons: less solid loss; sound organoleptic qualities; and stable at pH 5. Hence, it is suitable for tomato-type products, no blanching is required before canning, and the cross-linking operation is very simple with regards to equipment and processing temperature.

LITERATURE CITED

Batcher, O. M., Helmintoller, K. F., Dawson, E. H., *Rice J.* **59**, 4 (1956).

Hamerstrand, G. E., Hofreeter, B. T., Mehltretter, C. L., *Cereal Chem.* **37**, 519 (1960).

Rutledge, J. E., Islam, M. N., James, W. H., *Cereal Chem.* **49**, 4 (1972).

Webb, B. D., Adair, C. R., *Cereal Chem.* **47**, 708 (1970).

Received for review August 14, 1972. Accepted January 22, 1973. MNI is a Graduate Student with a International Rice Research Institute Fellowship.

Effect of Processing Conditions on Isolation of Cottonseed Protein by Sodium Hexametaphosphate Extraction Method

Micha Shemer,[1] Sylvia Mizrahi, Zeki Berk,* and Shoshana Mokady

A method for the isolation of cottonseed protein based on extraction with sodium hexametaphosphate (SHMP) and precipitation with acid was investigated. Maximum yields were obtained when extraction was carried out at pH 7.0, with 2% SHMP, at 60° for 30 min, and the curd was precipitated at pH 2–3. The isolate contained up to 94.8% protein, with the main impurity being phosphates. The nutritive value of the isolate was not significantly different from that of degossypolized cottonseed flour.

Present world production of cottonseed is in excess of 20 million tons (Tropical Product Quarterly, 1970), representing a protein potential of some 6 million tons per annum. Although cottonseed was recognized long ago as a possible source of protein for human consumption (Martinez *et al.*, 1970), actual utilization in this direction is negligible. The main reasons for this situation are the presence of gossypol, an antinutritional pigment, and failure to convert cottonseed protein to readily acceptable edible products. The solution to the gossypol problem in processing cottonseed for human food involves at present three different types of approaches: development of gossypol-free (glandless) seed through plant breeding (McMichael, 1959); multisolvent extraction (King *et al.*, 1961; Krishnamoorthi, 1965); and physical separation of the gossypol glands; *e.g.*, by the liquid cyclone process (Gastrock *et al.*, 1969). Glandless or degossypolized cottonseed flours may contain over 60% protein. These flours may be quite acceptable as edible products, provided that the seeds are dehulled completely and excessive heat and moisture are avoided during processing and storage. Further processing of such flours to isolated protein provides a possibility of attaining a higher protein concentration and may be expected to solve some of the acceptability and stability problems associated with the presence of nonprotein constituents.

Several methods for the isolation of cottonseed protein were suggested. They differ mainly in the media used for protein extraction. The media investigated include sodium sulfite (Arthur and Karon, 1948), salt solutions and alkali (Olcott and Fontaine, 1939), water and sodium hydroxide (Berardi *et al.*, 1969; Lawhon and Cater, 1971), and polyphosphates (Chajus and Chajus, 1964).

The purpose of the present work was to determine the effect of various processing conditions on isolation of cottonseed protein, using sodium hexametaphosphate (SHMP) as the extraction medium.

Department of Food Engineering and Biotechnology, Technion, Israel Institute of Technology, Haifa, Israel.

[1] Present address: University of Illinois, Burnsides Research Laboratory, Department of Food Science, Urbana, Illinois.

EXPERIMENTAL SECTION

Preparation of Cottonseed Flour. Fat-free and degossypolized flour was prepared by a two-stage solvent extraction process. First, the decorticated seeds (meats) were gound in aqueous acetone containing 30% water to remove gossypol. This medium is recommended by Pons and Eaves (1967) for the removal of gossypol with minimum extraction of other constituents. Grinding in solvent was done by an Ultra-Turrax colloid blender (Janke & Kunkel A.G., Stauffen, Germany). Subsequently the solvent was filtered off and the meats were reextracted with anhydrous acetone to remove the oil. The extracted flour was dried in open air.

Laboratory Isolation of Proteins. Two-hundred grams of degossypolized fat-free flour was extracted for 1 hr, at 50° and pH 7, in a solution of 40 g of SHMP in 2 l. of water. The extract was separated from the insoluble residue by centrifugation and subsequent filtration. The filtered extract was acidified with diluted HCl to the appropriate pH (usually 2–3). The precipitate (curd) was allowed to settle and the supernatant (whey) was decanted off. The curd was washed with water by resuspension and centrifugation. Finally the washed curd was dried in a vacuum oven at 50°.

Pilot Plant Isolation of Protein. Batches of 1 kg of flour were extracted with 10 l. of water and 200 g of SHMP. The procedure for extraction and precipitation was the same as described above, but drying of the curd was different. A suspension of curd in water was spray-dried, using a Niro-Production Minor apparatus (Niro Atomizer A/S, Copenhagen, Denmark). Air temperature was 190° at the inlet and 90–100° at the outlet.

Analytical. Moisture, fat, total nitrogen, free and total gossypol were determined following methods described by the American Oil Chemists' Society (1959). Phosphorus was determined according to the colorimetric molybdovanadate method described in AOAC (1970).

Nutritive Value. Net protein utilization (NPU) was determined according to Miller and Bender (1955) on individual animals. Weanling albino rats of the Charles River C.D. strain, age 21 days, were used. Three males and three females were used for each test material. The feeding period was 10 days.