

EFFECT OF PROTEIN DISRUPTION USING PROTEOLYTIC TREATMENT ON COOKED RICE TEXTURE PROPERTIES

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ABSTRACT

*Two long-grain rice (*Oryza sativa* L.) cultivars (Wells and Francis) were harvested from Stuttgart, AR and Essex, MO at high (21.0–22.0%) and low (12.8–16.3%) moisture contents (MCs). Rice samples were soaked in distilled water, sodium phosphate buffer and sodium phosphate buffer with a 0.2% protease (PS) at 37C for 30 min before cooking. A uniaxial compression test was performed to assess cooked rice texture properties. Results showed that protease treatment resulted in a significant decrease in cooked rice firmness for all cultivars across harvest locations and harvest MC. This indicated that proteins were involved in providing structural support to the rice kernel during cooking, restricting starch granule swelling and water hydration. The increase in cooked rice MC after PS treatment provides support for this finding. Stickiness significantly ($P < 0.05$) increased by soaking either in water or buffer solutions. This was attributed to a greater solubilization of starch, especially amylose leaching, during cooking. Protease treatment did not significantly increase cooked rice stickiness over the buffer treatment.*

PRACTICAL APPLICATIONS

The structural integrity of a rice kernel plays an important role in determining the textural properties of cooked rice. This study demonstrated that protein (i.e., through its disruption using protease) as well as lipid and starch (i.e., removal of starch and lipid during the draining and washing steps) all contribute in determining cooked rice texture properties. Results from this study indicated the possibility of reducing rice cooking duration to achieve texture properties similar to that with no protease treatment. Moreover,

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sticky rice products can be produced by protease treatment of rice before cooking.

KEYWORDS

Cooked rice, MC at harvest, protease, protein disruption, rice (*Oryza sativa* L.), soaking, texture

INTRODUCTION

Rice is consumed largely as cooked whole grain, which is produced after dehulling and milling processes by removing the hull and bran layers of the rough rice kernel, respectively. Milled rice predominantly contains approximately 80% starch, 6–9% protein and less than 1% lipids (Juliano 1985).

Rice texture is an important aspect of rice quality as it affects cooked rice acceptance by consumers. Further, expectations for specific textures in rice are known to widely differ in various parts of the world. Amylose content has long been recognized as the main driving force dictating the texture properties of rice – high-amylose cultivars being firmer and less sticky than low amylose cultivars (Juliano 1985). However, it is also reported that rice of similar amylose content can differ in their viscoelastic properties (Hamaker and Griffin 1990). Hamaker and Griffin (1990) reported the role of proteins in dictating rice texture, especially stickiness. They showed that the stickiness of cooked rice was increased when rice was cooked in water containing dithiothreitol, a compound known to disrupt proteins. This study clearly indicated the role of proteins in determining the stickiness of cooked rice. However, the study did not evaluate the role of proteins toward cooked rice firmness. Chrastil (1992) showed that changes in cooked rice functional properties during storage resulted from the increase in disulphide bonding of rice proteins during storage. In addition, the complexing of lipids (free fatty acids) with amylose (usually referred to as a LAM complex) is also thought to contribute to the rigidity of rice gels and could play a role in increasing cooked rice firmness (Eliasson and Krog 1985; Biliaderis and Tonogai 1991). The alteration of the rice kernel structure has been reported to affect rice functional properties. For instance, Marshall *et al.* (1990) indicated that partial removal of a rice kernel's lipid or protein resulted in significant changes in starch gelatinization. Shibuya and Iwasaki (1982), on the other hand, studied the effect of a proteolytic enzymatic treatment on the gelatinization properties of rice flour in aged and nonaged rice. Enzymatic treatment resulted in softer and stickier rice flour compared with nontreated rice flour.

The harvest moisture content (MC) of rough rice also plays a role in determining rice quality. Rice harvest MC indicates that rice kernel development with starch and protein syntheses are completed when rice MC reaches 27–29% (Siebenmorgen *et al.* 2004) Recently, Siebenmorgen *et al.* (2004) studied the impact of MC at harvest on rice flour pasting viscosity and reported that rice harvested at high MC yielded less viscous flour than those harvested at low MC. It has been hypothesized that the higher protein content and amylase activity of rice harvested at high MC (i.e., because of the greater proportion of less mature kernels) are the main causes of these pasting viscosity changes in rice flour. However, there are no reported studies on the impact of harvest MC on rice texture properties. Currently, most of the research dealing with establishing the role of protein and lipid fractions toward rice functionality has been performed on rice flour and it is yet to be determined if these findings can be translated to intact rice kernel. We hypothesize that the structure of a rice kernel plays a key role in determining its texture properties once cooked, with proteins restricting water absorption during cooking. Therefore, the objective of this study was to investigate the effect of surface protein disruption, denaturation or hydrolysis using a proteolytic treatment, on the texture properties of rice after cooking.

MATERIALS AND METHODS

Rice Samples and Rice Fractionation

Two long-grain rice cultivars (Francis and Wells) were harvested from two different locations (Stuttgart, AR and Essex, MO) in the fall of 2004 at various MCs as shown in Table 1. After transporting to the University of Arkansas rice processing laboratories, rice samples were air dried to approximately 12% MC before size fractionation, using a precision Sizer (Carter-Day Co., Minneapolis, MN), to large, medium and then fractions. The medium thickness fractions (1.69–1.72 mm of milled rice) from all cultivars were used in this study to provide samples as uniform as possible in size, as kernel size tends to affect rheological tests results such as uniaxial compression.

Rice Milling

Preliminary experiments were conducted to determine the milling durations necessary to achieve the desired surface lipid content (SLC) for each rice samples. Rice samples from each cultivar at each harvest MC were dehulled using a dehusker (THU-35, Satake, Hiroshima, Japan) and milled for 10, 20, 30, 40 and 50 s using a McGill No. 2 mill (Rapsco, Brookshire, TX). A double-tray sizing device (Grain-Man Machinery Mfg., Miami, FL) was used

TABLE 1.
MILLING QUALITY OF FRANCIS AND WELLS RICE CULTIVARS HARVESTED AT HIGH
AND LOW MOISTURE CONTENTS (MCs) FROM TWO LOCATIONS

Cultivar	Location	Harvest MC (%) [*]	Surface lipids (%)	Head rice yield (HRY) (%)	Degree of whiteness
Wells	Stuttgart, AR	13.7	0.42 ^{ab*}	67.33 ^a	39.53 ^{bc}
		21.4	0.42 ^a	68.33 ^a	40.67 ^a
	Essex, MO	16.3	0.41 ^a	64.58 ^b	38.70 ^c
		22.0	0.40 ^a	60.44 ^c	40.23 ^{ab}
Francis	Stuttgart, AR	14.7	0.40 ^a	61.22 ^c	41.1 ^a
		21.8	0.41 ^a	69.44 ^a	39.13 ^c
	Essex, MO	12.8	0.41 ^a	37.20 ^d	39.67 ^{bc}
		21.0	0.40 ^a	65.64 ^b	40.63 ^{ab}

* For the same cultivar harvested at various MCs, the means of surface lipids, HRY and degree of whiteness of milled rice with different letters are significantly ($P < 0.05$) different according to the least significant difference.

to separate head rice from the broken kernels and the milled rice SLC was determined using a Soxtec apparatus as described later. The relationship between milling duration and SLC was then determined for each cultivar at each location and harvest MC in order to achieve a target SLC of approximately 0.40% (wet base).

Initially, 150 g of rough rice from each cultivar (MC of approximately 12%) was dehulled using a dehusker (THU-35, Satake). The resulting brown rice was then milled using a McGill No. 2 (Rapsco) mill to various durations, based on the results of the preliminary milling experiment, to achieve the target SLC of approximately 0.40%. A double-tray sizing device (Grain-Man Machinery Mfg.) was used to separate head rice from the broken kernels. Head rice yield (HRY) was calculated by dividing the head rice weight by 150 g and is reported as means of duplicate analyses.

Milled Rice Whiteness

The degree of whiteness of the milled rice samples was determined in duplicate with a Kett whiteness tester specifically designed to measure the whiteness of rice (Model C-300-3, 1-8-1 Minami-Magome, Ota-Ku, Tokyo, Japan). The Kett whiteness tester was calibrated in accordance with the international whiteness standard using a calibration plate. Rice whiteness values were reported as a reflective index of the sample surface, where higher whiteness values indicate whiter milled rice.

Protein Content

Milled rice protein content for various SLCs was determined using the American Association of Cereal Chemists (AACC) method 46-11A (AACC 1996). Nitrogen content was measured in duplicate by the Kjeldahl procedure, and the protein content of rice flour was calculated by multiplying the nitrogen content by 5.95.

SLC

Milled rice SLC was determined in duplicate using a Soxtec system (Avanti 2055, Foss North America, Eden Prairie, MN) according to the AACC method 30-20 (AACC 1997) by modifying the washing duration from 30 to 20 min using petroleum ether as described by Matsler and Siebenmorgen (2005).

Soaking of Milled Rice before Cooking

Twenty grams of head rice was soaked into 100 mL of either distilled water, 0.03-M phosphate buffer (pH = 7.4) or 0.03-M phosphate buffer (pH = 7.4) containing 0.2% protease (*Streptomyces griseus*, Sigma EC 232-909-5, Sigma, St. Louis, MO) and was allowed to soak for 30 min at 37°C. The soaking medium was then drained, the rice was washed three times with distilled water and the amount of weight gained because of soaking was calculated. This treatment was performed on the two rice cultivars harvested from the two locations at various MCs.

Because soaking and washing of milled rice may account for losses of rice starch that might wash out during the draining and washing process, experiments were conducted where the soaking solution (i.e., distilled water, 0.03-M phosphate buffer or 0.03-M phosphate buffer [pH = 7.4] containing 0.2% protease [*S. griseus*, Sigma EC 232-909-5]) was used to cook rice samples after soaking of the milled rice. After this soaking treatment, the weight of the soaked rice and the soaking solution was adjusted to account for the evaporated amount of water. This analysis was performed only on one of the cultivars harvested at one of the locations.

Instrumental Texture Measurements of Cooked Rice

A rice-to-water ratio of 2:1, accounting for the amount of water absorbed by the rice during the soaking procedure, was used for cooking rice. Rice was cooked using a miniature precision rice cooker, featuring a heating mantle (TM 102, Glas-Col, Terre Haute, IN) controlled by a temperature controller (89000-10, Eutech Instruments Pte. Ltd., Singapore), for 20 min at a

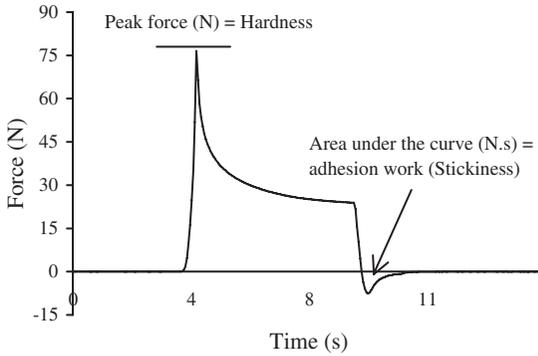


FIG. 1. TYPICAL FORCE-DEFORMATION CURVE AND TEXTURE ATTRIBUTE DETERMINATION

maximum cooking temperature of $98.5 \pm 1\text{C}$. The cooking conditions were identical for all rice samples to eliminate differences in cooked rice texture properties resulting from the cooking method. Texture attributes of cooked rice were determined by a uniaxial single compression method using a TA-XT2 plus texture analyzer (Texture Technologies, Scarsdale, NY). Ten cooked rice kernels placed on a nonlubricated flat aluminum plate were compressed using a 50-kg load cell to leave a gap measuring 0.3 mm between the two compression plates at the bottom of the compression cycle. The crosshead was also stopped for 5 s at its maximum travel distance before returning to an anvil separation of 20 mm. The crosshead speed of the texture analyzer was set at 10 mm/s, where the test speed was set at 5 mm/s and the posttest speed at 0.5 mm/s. Texture attributes were obtained using the texture exponent software (Stable Micro Systems, version 1,0,0,92 (2000) Godalming, U.K.). The maximum compression force (N) was used as an indicator of cooked rice firmness while the adhesion energy (i.e., area under the curve, N·s) measured during the upward travel of the compression plate was used as an indicator for cooked rice stickiness. Figure 1 shows a typical force-deformation curve. Each rice sample from each cultivar, harvest MC and soaking treatment was cooked in duplicate and six measurements were conducted for each.

Cooked Rice MC

Triplicate measurements of cooked rice MC were carried out after each cook. Approximately, 5 g of cooked rice was dried at 130C for 24 h using a drying oven (Precision Winchester, VA). The cooked rice MC was calculated as the amount of water removed after 24 h.

Statistical Analysis

Analysis of variance was performed using JMP (release 5.1.2, SAS Institute, Cary, NC), to determine the significance of the differences (least significant difference at a 5% level of probability) in the physicochemical properties among rice harvested at various MCs from different locations and subjected to various soaking treatments. Two replicates of samples from each cultivar, location and harvest MC were processed and evaluated and values are reported as averages of the replicates.

RESULTS AND DISCUSSION

Rice Milling and Quality Characteristics

The whiteness, HRY and dimensions of rice kernels from the two cultivars harvested at each MC from the two locations are presented in Table 1. The results illustrate milling quality differences of the cultivars across harvest MC and location. Cultivars harvested at low MC had lower HRY than those harvested at high MC. Francis harvested from Stuttgart, AR at 21.8% MC had the highest HRY (69.44%), while Francis harvested from Essex, MO at 12.8% MC had the lowest HRY (37.20%). This result is not surprising as rice dried in the field and harvested at low MC is usually subjected to moisture readsorption during rainfall or morning dew (Siebenmorgen *et al.* 1992). In addition, samples harvested at high MC were dried under gentle drying conditions that might positively impact HRY. This agrees with Wongpornchai *et al.* (2004) who indicated that hot-air drying ($T = 70^{\circ}\text{C}$) yielded lower HRY than samples dried using modified air at $T = 30$ and 40°C or hot air at $T = 40$ and 50°C .

The degree of milling (DOM), which indicates the amount of lipids remaining on the surface of the rice kernel after milling, has long been reported to play a role in determining rice sensory quality (Champagne *et al.* 1997). Because different rice cultivars are known to mill differently (i.e., milling duration varies from cultivar to cultivar to achieve a target DOM), preliminary milling experiments were conducted to determine the milling durations necessary to achieve an SLC of approximately 0.40% of the sample. As a result, SLC did not vary significantly among cultivars with various harvested MCs and from different locations (Table 1). The whiteness of the samples, which is highly correlated to SLC, ranged from 39.1 to 41.1. This indicated that the differences observed in cooked rice texture properties were not because of the lipid fraction.

Soaking of rice kernels significantly ($P < 0.05$) decreased the SLC of rice kernels (Table 2). In a similar study, Monsor and Proctor (2002) found that simple water washing significantly reduced milled rice surface lipids and free

TABLE 2.
SURFACE LIPID AND PROTEIN CONTENTS BEFORE AND AFTER VARIOUS SOAKING TREATMENTS OF FRANCIS AND WELLS MILLED RICE HARVESTED AT HIGH AND LOW MOISTURE CONTENTS (MCs) AT TWO LOCATIONS

Cultivar	Harvest mc (%)	Surface lipids (%)				Protein content (%)			
		NS	DWS	SPBS	PS	NS	DWS	SPBS	PS
Stuttgart, AR									
Francis	14.7	0.40 ^{a*}	0.10 ^b	0.11 ^b	0.04 ^c	7.25 ^a	7.18 ^a	7.15 ^b	6.41 ^c
	21.8	0.41 ^a	0.10 ^b	0.11 ^b	0.06 ^c	7.96 ^a	7.93 ^a	7.57 ^b	7.43 ^c
Wells	13.7	0.42 ^a	0.11 ^b	0.13 ^b	0.05 ^c	7.55 ^a	7.44 ^b	7.43 ^b	6.60 ^c
	21.4	0.42 ^a	0.11 ^b	0.11 ^b	0.03 ^c	7.67 ^a	7.65 ^a	7.30 ^b	6.92 ^c
Essex, MO									
Francis	12.8	0.41 ^a	0.11 ^b	0.11 ^c	0.04 ^d	7.24 ^a	6.99 ^b	6.59 ^c	6.31 ^d
	21.0	0.40 ^a	0.10 ^b	0.11 ^c	0.04 ^d	6.57 ^a	6.36 ^b	5.86 ^c	5.68 ^d
Wells	16.3	0.41 ^a	0.11 ^b	0.12 ^b	0.06 ^c	7.52 ^a	7.23 ^b	6.73 ^c	6.66 ^d
	22.0	0.40 ^a	0.10 ^b	0.11 ^b	0.07 ^c	7.55 ^a	7.45 ^a	7.42 ^a	7.06 ^b

* For the same row, the means of surface lipids and protein content of NS, DWS, SPBS and PS treatments with different letters are significantly ($P < 0.05$) different according to the least significant difference.

NS, no soaking; DWS, distilled water soaking; SPBS sodium phosphate buffer soaking; PS, protease soaking.

fatty acids. The localization of these lipids on the outer caryopsis coat, aleurone and subaleurone layers of rice kernels (Juliano 1985) allowed surface lipids to wash out during draining of the soaking solution. The protease treatment probably resulted in disruptions on some rice proteins. This was manifested in the significant ($P < 0.05$) decrease in protein content as a result of draining and washing of soaked rice kernel before cooking. However, the decrease in protein content after protease treatment varied from 6.5 to 13.5%, which was much lower than the levels of 33–38% reported by Marshall *et al.* (1990). This may be a result of the differences in the soaking treatments applied. For instance, our samples were soaked for 30 min at 37C, while Marshall *et al.* (1990) soaked samples for 30 min with no indication of the soaking temperature or the type of protease used.

Cooked Rice MC

Table 3 shows the cooked rice MC of samples soaked with the various treatments. Cooked rice MC for the samples ranged from 67.61 to 71.28%. The cooked rice MC data indicated that the PS resulted in significant ($P < 0.05$) increased water absorption during cooking. Although the difference

TABLE 3. MEANS OF HARDNESS, STICKINESS AND COOKED RICE MOISTURE CONTENT (MC) AFTER VARIOUS SOAKING TREATMENTS FOR WELLS AND FRANCIS HARVESTED AT HIGH AND LOW MCs FROM TWO LOCATIONS

Cultivar	Soaking treatment	Stuttgart, AR					
		Hardness (N)		Stickiness (N·s)		Cooked rice MC	
		HMC = 14.7%	HMC = 21.8%	HMC = 14.7%	HMC = 21.8%	HMC = 14.7%	HMC = 21.8%
Francis	NS	101.40 ^{ab,†}	93.47 ^{abb}	9.13 ^{4a}	7.32 ^{8b}	69.23 ^{cA}	69.09 ^{bA}
	DWS	102.87 ^{4a}	96.28 ^{4a}	17.35 ^{4a}	18.69 ^{4a}	69.10 ^{cA}	69.22 ^{bA}
	SPBS	97.34 ^{bA}	91.66 ^{bA}	15.30 ^{bA}	15.14 ^{bA}	71.29 ^{4a}	70.37 ^{abB}
	PS	85.19 ^{cA}	80.29 ^{cA}	13.24 ^{cA}	11.60 ^{cA}	70.25 ^{bA}	70.68 ^{aA}
Wells	NS	99.16 ^{4a}	97.35 ^{bA}	7.89 ^{cA}	7.76 ^{cA}	HMC = 13.7%	HMC = 21.4%
	DWS	100.44 ^{4a}	102.91 ^{4a}	13.04 ^{4b}	15.82 ^{4a}	69.05 ^{bca}	69.10 ^{bA}
	SPBS	99.08 ^{4a}	96.10 ^{bA}	12.14 ^{4b}	16.22 ^{4a}	68.27 ^{cA}	68.79 ^{bA}
	PS	78.12 ^{bA}	79.14 ^{cA}	9.16 ^{4b}	11.75 ^{bA}	69.80 ^{4ba}	69.18 ^{bA}
						70.73 ^{4a}	70.51 ^{aA}
Essex, MO							
Francis	NS	HMC = 12.8%	HMC = 21.0%	HMC = 12.8%	HMC = 21.0%	HMC = 12.8%	HMC = 21.0%
	DWS	99.35 ^{4a}	90.21 ^{bb}	8.72 ^{4a}	8.58 ^{4a}	68.34 ^{cA}	69.22 ^{bA}
	SPBS	93.11 ^{bb}	97.42 ^{4a}	13.48 ^{4b}	18.56 ^{4a}	67.61 ^{dA}	68.72 ^{bA}
	PS	95.76 ^{4ba}	93.61 ^{4ba}	15.22 ^{4a}	15.22 ^{4a}	69.67 ^{bA}	70.02 ^{aA}
Wells	NS	71.94 ^{cA}	68.40 ^{cA}	9.74 ^{bA}	8.79 ^{cA}	71.02 ^{4a}	70.71 ^{4b}
	DWS	HMC = 16.3%	HMC = 22.0%	HMC = 16.3%	HMC = 22.0%	HMC = 16.3%	HMC = 22.0%
	SPBS	100.41 ^{bA}	99.20 ^{4ba}	7.82 ^{cA}	7.97 ^{cA}	69.30 ^{bA}	69.37 ^{bA}
	PS	105.39 ^{4a}	97.23 ^{4b}	18.11 ^{4a}	16.55 ^{4a}	68.29 ^{cA}	68.97 ^{bA}
	101.67 ^{4ba}	102.61 ^{4a}	16.22 ^{4a}	18.45 ^{4a}	69.47 ^{4ba}	69.87 ^{4ba}	
	83.79 ^{cA}	81.39 ^{cA}	14.04 ^{bA}	12.77 ^{bA}	70.21 ^{4a}	70.62 ^{aA}	

* For the same harvest MC, the means of hardness, stickiness and cooked rice MC of NS, DWS, SPBS and PS treatments with different letters (lowercase) are significantly ($P < 0.05$) different according to the least significant difference.
 † Means of hardness, stickiness and cooked rice MC of the same cultivar and soaking treatment harvested at different MCs with different letters (uppercase) are significantly ($P < 0.05$) different according to the least significant difference.
 NS, no soaking; DWS, distilled water soaking; SPBS, sodium phosphate buffer protease soaking; PS, protease soaking; HMC, MC of rough rice at harvest.

between sodium phosphate buffer soaking (SPBS, buffer alone) and PS (buffer plus protease) was not always significant ($P > 0.05$), the trend for PS was toward an increase in water absorption during cooking. This was probably because of structural changes in the rice kernels as a result of protein structural alteration using proteolytic treatment. Proteins are known to provide a support network for starch granules (i.e., formation of starch proteins gel matrix) (Chrastil 1990, 1994) and have been postulated to restrict starch granule swelling (Hamaker *et al.* 1991). Protease treatment probably partially disrupted that network and starch granules were allowed to swell to a fuller extent, binding more water than when the network is unadulterated. Results also indicated that rice samples harvested at low MC had higher water uptake than that harvested at high MC during cooking. However, the trend was not significant at $\alpha = 0.05$.

Cooked Rice Hardness

Rice harvested at high (21.0–22.0%) MC was softer after cooking than those harvested at low (12.8–16.3%) MC (Table 3). This was more profound for Francis before the soaking treatment, where samples harvested at 12.8 and 14.7% MC were significantly ($P < 0.05$) firmer than those harvested at 21.8 and 21.0% MC, respectively. However, the significance between high and low harvest MC samples disappeared after SPBS and PS treatments. This could be a result of the drying conditions, where samples harvested at high MC were dried gently at controlled air conditions, compared with samples harvested at low MC that was left to dry in the field.

Differences in drying conditions have been reported to affect cooked rice texture as well as rice flour pasting properties. For instance Daniels *et al.* (1996, 1998) indicated that low-temperature-dried ($T = 33\text{C}$) rice had greater water absorption and volume expansion than high-temperature-dried ($T = 54\text{C}$) rice. Elevated drying temperatures (i.e., field drying temperature) are believed to affect rice proteins (i.e., through denaturation of the rice protein oryzenin) that in part influences the texture properties of cooked rice compared with rice dried under gentle temperatures (Siebenmorgen and Meullenet 2004).

Cooked rice hardness of rice samples harvested at various MCs ranged between 68.40 and 105.39 N for various soaking treatments (Table 3). Instrumental cooked rice hardness significantly ($P < 0.05$) decreased after PS treatment (protein disruption) across cultivars, locations and harvest MC. This is in agreement with Juliano (1985), who indicated that the cooked texture of rice high in protein tends to be tougher and chewier than for rice low in protein.

Protein distribution in the rice endosperm is such that a network of proteins is formed around swollen starch granules. This suggests that during cooking, proteins bind water, causing the concentration of the dispersed phase

(i.e., primarily a mixture of amylose and proteins) to increase, providing suitable conditions for starch–protein complex formation (i.e., a gel matrix that provides mechanical support for starch, which reduces leaching during cooking) that results in restricting starch granule swelling (Hamaker *et al.* 1991; Hamaker and Griffin 1993; Fitzgerald *et al.* 2003). Therefore, the partial removal of proteins by protease treatment before cooking would result in the reduction of the protein's intermolecular interactions and also increase the amount of water available for starch. This probably resulted in the greater swelling of starch granules in the absence of the native protein structure. This is probably why the firmness of cooked rice decreased after protease treatment. The increase in cooked rice MC after protease treatment provides evidence of the role of proteins in restricting water absorption during cooking. This agrees with Kim *et al.* (2001), who indicated that water absorption was related to protein content of rice cultivars. Yanase *et al.* (1984) also indicated that rice with higher protein content exhibited reduced water uptake ratios.

The decrease in cooked rice firmness after protein hydrolysis indicated that proteins play an important role in maintaining rice kernel integrity, thus influencing the functional properties of rice during cooking. In addition to proteins, lipids are also believed to influence the texture of cooked rice (Kim *et al.* 1986). Lipids have been reported to form inclusion complexes with amylose (Bank and Greenwood 1975). These complexes were reported to decrease water uptake (Eliasson and Krog 1985; Seneviratne and Biliaderis 1991) and also provide a barrier to starch granule hydration (Biliaderis and Tonogai 1991). However, the removal of SLC that was observed by soaking the sample in deionized water or in the buffer solution did not significantly impact cooked rice firmness measurements.

Cooked Rice Stickiness

The stickiness of the rice samples ranged from 7.32 to 18.69 N-s for the no soaking (NS)-, distilled water soaking (DWS)-, SPBS- and PS-treated rice cultivars. Results indicated that the stickiness of cooked rice significantly ($P < 0.05$) increased after soaking across cultivars, harvest locations and MC (Table 3). The soaking process probably allows the soaking solutions to pre-hydrate the kernel, resulting in a greater amount of starch leaching out from the starch granule during cooking. Furthermore, the disruption and the partial removal of proteins, because of the soaking process, resulted in the greater contribution of starch to cooked rice stickiness. This disruption and partial removal of proteins by protease treatment increased the amount of water available for starch granules to fully hydrate. This was indicated by the significant ($P < 0.05$) increase of cooked rice MC for PS compared with NS rice samples (Table 3).

The decrease in cooked rice stickiness after protein disruption (PS) (less sticky), relative to rice soaked with water or buffer (DWS or SPBS) (stickier), may be a result of the loss of starch during washing of the sample before cooking. We hypothesize that the loss of additional starch during the washing and draining steps resulted in a change in the amount of starch leached out during rice cooking. To prove this hypothesis, the same experiments were conducted without rinsing the sample after soaking. Figure 2a shows the hardness and Fig. 2b shows the stickiness of cooked rice after soaking without rinsing the sample three times with water before cooking. Results indicated a significant decrease in the instrumental texture hardness of cooked rice after PS treatment compared with other soaking treatments. Moreover, cooked rice stickiness was also influenced by the soaking treatments – PS-washed rice being the stickiest among all treatments, a result opposite to that obtained when rinsing the sample with water prior to cooking.

Our results suggest that PS resulted in the disruption of rice protein bodies into the soaking solution along with starch leaching (as indicated by the increase in total amylose content concentration in the soaking solution [results are not shown]). The increase in cooked rice stickiness after protein disruption supports the hypothesis that protein–starch gel matrix formation during cooking is the major factor limiting cooked rice stickiness. Prudencio-Ferreira and Areas (1993) studied soy protein and found that at high-temperature exposures, protein–protein network was formed through disulfide bonding.

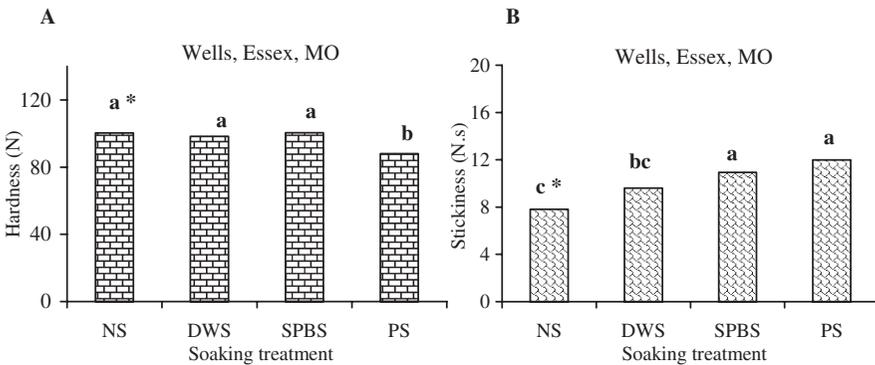


FIG. 2. (A) INSTRUMENTAL HARDNESS AND (B) AND STICKINESS (N) OF COOKED WELLS RICE SAMPLES OF ROUGH RICE HARVESTED AT 22.0% MOISTURE CONTENT FROM ESSEX, MO AFTER DIFFERENT SOAKING TREATMENTS (WITH NO DRAINING OR RINSING OF THE SOAKED SOLUTIONS)

*Columns with different letters are significantly ($P < 0.05$) different according to the least significant difference.

NS, no soaking; DWS, distilled water soaking; SPBS, sodium phosphate buffer soaking; PS, protease soaking.

The formation of these intermolecular bonds between protein molecules probably resulted in decreasing the contribution of proteins to rice stickiness and to more water available for starch to absorb. Moreover, the increase in disulfide bonding during cooking probably reduced the amount of water bound by proteins, providing more water being available for starch to swell further, leaching of more soluble amylose and increased rice stickiness.

CONCLUSIONS

The structural integrity of a rice kernel plays an important role in determining the textural properties of cooked rice. This study demonstrated that protein (i.e., through its disruption using protease), as well as lipid and starch (i.e., removal of starch and lipid during draining and washing steps), all contribute in determining cooked rice texture properties. Protein–starch interactions could be the major factor responsible for the changes in cooked rice texture properties as a consequence of changes in the structure and properties of proteins. During rice cooking, the formation of a starch–protein gel network that surrounds starch granules is considered an important factor that determines rice texture properties.

Proteolysis caused a significant reduction in protein content, although not all proteins were removed from the kernel by the protease treatment. Our results suggested that the disruption of proteins by protease before cooking resulted in increasing the amount of water available for starch, resulting in the greater swelling of the starch granules, and increase in the susceptibility of starch granules to leach out amylose–amylopectin. This ultimately decreased cooked rice firmness and increased stickiness.

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