Rheological Changes in Cracker Sponges During an 18-Hour Fermentation¹

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ABSTRACT

determined from the slope of the linear relationship between resistance to extension and time. The slopes showed that sponges at pH 4.1 and 3.5 had much greater enzyme activities than sponges at pH 4.7 and 2.7. Reconstituting an enzyme-free composited residue with enzyme extracts from several cracker flours showed that different flours differed in enzyme activity.

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Conventionally, the saltine cracker making process requires about 24 hr of fermentation. One purpose of this long fermentation is to obtain an optimum modification of the flour gluten. That modification is believed to be closely related to rheological changes in the cracker sponge that are important in determining dough handling properties and final product qualities (Rogers and Hoseney 1989). Physical testing methods have been used to measure the rheological changes in cracker sponges during fermentation (Faridi 1975; Pizzinatto and Hoseney 1980; Doescher and Hoseney 1985a,b). The influences of the various formula ingredients of cracker sponges on dough rheology also were studied by Pizzinatto and Hoseney (1980).

Of variables tested, the pH of cracker sponges was the most effective in reducing their resistance to extension. The effect was attributed to

proteolytic enzymes, because the optimum pH of 4.1 coincides with the reported optimum pH of indigenous flour protease. A mixture of

commercial gluten and wheat starch was used to study the effect of

proteolytic enzymes. Their optimum pH for decreasing the resistance to extension ranged from 3.8 to 4.1. Relative enzyme activities were

In this study, the major inherent factors in flours that caused rheological changes in cracker sponges were investigated. Of special interest was the importance of flour proteolytic enzymes on the rheological properties of cracker sponges.

MATERIALS AND METHODS

Flour

A series of soft wheat cracker flours with different baking properties was used: soft white wheat flour A (protein 9.5%, moisture 12.7%, ash 0.46%) and soft red wheat flour B (protein 10.1%, moisture 13.0%, ash 0.49%), flour C (protein 9.1%,

flour G (protein 10.1%, moisture 13.8%, ash 0.48%). The commercial cracker flours (flours C-G) were supplied by Nabisco Brands, Peavey, Lance, and Dixie-Portland. In addition, two soft wheats, a soft white wheat (SWW, A) and a soft red winter (SRW, B) were milled on the Kansas State University pilot mill. A commercial bread flour (protein 12.2%, moisture 12.6%, ash 0.47%) was also used in certain studies.

moisture 13.0%, ash 0.45%), flour D (protein 9.1%, moisture

12.6%, ash 0.46%), flour E (protein 9.6%, moisture 12.1%, ash

0.52%), flour F (protein 7.8%, moisture 13.1%, ash 0.42%), and

Yeast

Red Star commercial compressed baker's yeast (70% moisture) from Universal Food Corporation was used.

Gluten and Wheat Starch

Gluten and wheat starch samples were provided by Midwest Solvent Co., Atchinson, KS. The protein and moisture contents were 80 and 11.1%, respectively, for gluten and 0.3 and 7.9% for wheat starch.

Chemicals

All chemicals were reagent grade.

Preparation of Slurry

A slurry was prepared as described by Doescher and Hoseney (1985b). The mixture contained flour and water (1:2 ratio), 0.62% yeast, and 3% sucrose based on flour weight. It was fermented for 18 hr at 30°C and 90% relative humidity. A feed of 20% flour (based on sponge flour weight) was then added, and the slurry was given a second 18-hr fermentation.

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Rheological Studies by Extensigraph

To study the rheological changes occurring during fermentation of cracker sponges an extensigraph procedure (Pizzinatto and Hoseney 1980) was used. Sponges that contained yeast (0.36%based on flour weight), slurry (4.52% based on flour weight), and water (22% based on flour weight) were mixed for 3 min in a pin mixer (TMCO-National Mfg. Company, Lincoln, NE) modified to 32 rpm and allowed to ferment for 18 hr (30° C, 90% rh). After fermentation, the sponge was mixed for 5 min with shortening (11%), salt (1.8%), and soda (0.45%) but no additional flour, and divided into three pieces of 150 g each. Each piece was rounded, molded, allowed to rest for 45 min, and then measured with an extensigraph. Each treatment was duplicated.

To study proteolytic enzyme activity, a constant (or control) gluten and starch system was used. No shortening was added at the dough-up stage to aid in handling of the sponges. In all the rheological studies, soda was added at the dough-up stage, unless specifically mentioned otherwise.

Extraction of Proteolytic Enzymes

One kilogram of flour was extracted with 6 L of ammonium sulfate solution (18% saturation), by stirring for 1.5 hr at 4° C. The slurry was then centrifuged at $1,000 \times g$ for 15 min. The residue was lyophilized and prepared for reconstitution studies. The supernatant was adjusted to 40% saturation by adding solid ammonium sulfate, allowed to stand for 24 hr at 4° C, then centrifuged. The supernatant was then adjusted to 80% saturation and allowed to set for another 24 hr at 4° C. The precipitate was collected by centrifugation and lyophilized. This preparation was used as the crude proteolytic enzyme in reconstitution studies. The isolation procedure is essentially that reported by Salgo (1981).

Reconstitution Studies

In studying the rheological effects, different amounts of the crude proteolytic enzyme extract were added back: zero, one third, two thirds, and all.



Fig. 1. Rheological changes of sponges made from gluten and starch set at different pH levels for 0 or 12 hr.

RESULTS AND DISCUSSION

Proteolytic Enzyme Effects in a Commercial Gluten and Starch System

To study the rheological effects of proteolytic enzymes we needed an enzyme-free system. We chose a system of commercial gluten and starch. However, it was found that the resistance to extension of this system decreased during 12 hr of rest at pH 4.1. This indicated that at least part of the proteolytic enzymes remained associated with the gluten or that the low pH changed the sponge rheology.

To examine the possible effects of acid, two series of sponges were adjusted with lactic acid to pH values between 2.7 and 4.7 and allowed to stand for 0 hr and 12 hr. Resistance to extension was



Fig. 2. Effect of pH on the activity of the proteolytic enzymes.



Fig. 3. Activity of indigenous proteolytic enzymes at different pH levels.

measured after the sponges were neutralized to pH 7.0. At zero time, the acid greatly changed sponge rheology (Fig. 1).

The effect of the acid was accounted for by subtracting the effects at 0 hr from the results at 12 hr. With this technique, the



Fig. 4. Changes in the rheological properties of cracker sponges at pH 4.1 during an 18-hr setting time. Soft white wheat (SWW) = complete flour; SWW no enzyme = extracted with ammonium sulfate.



Fig. 5. Rheological effects after setting 18 hr at pH 4.1 with different amounts of enzyme extract expressed as percent of total extract.

largest rheological changes were found to be in the range from pH 3.8 to 4.1 (Fig. 2), which was the optimum range reported for the proteolytic enzymes (Salgo 1981). This evidence further supported the theory that proteolytic enzymes were playing important roles in changing the sponge rheology.

Sponges were adjusted to pH 2.7, 3.5, 4.1, and 4.7, and then allowed to set for different times. As shown in Figure 3, good linear relationships were found for decreases in resistance to extension at all pH values. At pH 3.5 and 4.1, much steeper slopes were obtained, which indicated greater enzyme activities in that pH range.

The slope at pH 3.5 was slightly greater than the slope at pH 4.1; however, this changed after subtracting the effect of the acid. Sponges at pH 4.1 showed a more rapid rheological change than the others.

Rheological Effects of the Proteolytic Enzymes Extracted from Soft White Wheat Flour

Sponges made from the soft white wheat residue after enzyme extraction showed no rheological change (Fig. 4) during 18-hr "fermentation" at pH 4.1. This indicated that the proteolytic enzymes, presumably completely extracted from the residue, did have a very important effect on sponge rheology.

A reconstitution study, combining the enzymes and the residue to give a reconstituted sponge, gave resistance to extension curves measured after 18 hr of fermentation similar to those of the unextracted control flour. This showed that the enzymes were active when added back to the residue. The resistance to extension of this sponge was 432 Brabender units, so about 82% of the enzyme activity was recovered.

As shown in Figure 5, the rheology of the sponge was closely related to the amount of the extract added. Therefore, the rheological change caused by fermentation can be materially influenced by increasing the proteolytic enzyme in the cracker sponge.

Rheological Effects of the Proteolytic Enzymes Extracted from Different Cracker Flours

The proteolytic enzymes were extracted from seven cracker flours with different baking properties. The residues from six flours were combined to study the relative enzyme activities. As shown in Table I, reconstituted flours made from the combined residue and the enzyme extract from each flour were made into sponges and allowed to stand for 18 hr. The proteolytic enzyme extracts from the different flour sources showed different abilities to change sponge rheology.

Comparison of Rheological Changes in Cracker Flours After 9 hr

Wu and Hoseney (1989) showed that the rapid change in rheology that occurs during the first 5 hr of fermentation (Doescher and Hoseney 1985b) has essentially no influence on the cracker dough. In addition, they showed that sponges made with yeast plus slurry have a pH that favors proteolytic enzyme action after about 9 hr of fermentation. Therefore, the actual period of time for proteolytic enzyme activity would be from about 9 hr until

TABLE I Rheological Effects of the Proteolytic Enzymes Extracted from Different Cracker Flours^a

Extracted from	Resistance to Extension (BU) ^b
Control (no extract)	1,150
Flour D	280
Flour C	336
Flour G	370
Soft white wheat flour A	453
Soft red wheat flour B	462
Flour E	474
Flour F	493

^aEnzymes were added to a constant residue and fermented for 18 hr.

 $^{b}LSD_{0.05} = 32.$

TABLE II		
Comparison of Rheological Changes in Different Cracker Flours		
After Setting for 9 hr at pH 4.1		

	Resistance to Extension	
Samples	(BU) ^a	
Flour B	970	
Flour D	864	
Flour C	811	
Flour E	476	
Flour F	425	
Flour G	252	

 a LSD_{0.05} = 33.



Fig. 6. Comparison of the change in rheological properties of different flours with short standing times at pH 4.1. SWW = soft white wheat, SRW = soft red wheat; flours B-D = commercial cracker flours.

the end of 18 hr of fermentation. Thus, 9 hr of reaction time could be used to simulate cracker fermentation.

The differing activities of enzymes on a composited residue flour extracted from different cracker flours raised the question of how those enzymes would function on each flour's own residue. As shown in Table II, after setting for 9 hr at pH 4.1, sponges made from different flours showed different decreases in resistance to extension. This may be a useful indicator to determine flour quality and predict baking results.

In studying the rate of the rheological changes with standing time (Fig. 6), it was found that although the rheological properties were the same for several sponges after 9 hr of standing, they had different rates of change in the reduction of resistance to extension. A particularly high rate was found for the sponge mixed from the soft red winter flour.

The sponge mixed from bread flour is also shown in Figure 6. The high resistance to extension infers either a low proteolytic enzyme activity or a very strong residue that was not affected by enzyme activity. It would be difficult to bring the resistance to extension of that sponge to a reasonable (low) value for cracker production.

CONCLUSIONS

Sponge pH 4.1 was found to be the most effective in reducing the resistance to extension. The effect was attributed to proteolytic enzymes. A constant system using commercial gluten and wheat starch was made for studying the effect of proteolytic enzymes. The optimum pH was in the range 3.8–4.1. Relative enzyme activities determined from the slope of the linear relationship between resistance to extension versus time showed that the sponges at pH 4.1 and 3.5 had much greater enzyme activities than at pH 4.7 and 2.7. Rheological properties of sponges made with flours from which enzymes had been extracted did not change during fermentation. The rheological changes returned when enzyme extracts were added back to the sponges. Thus, proteolytic enzymes have important effects on sponge rheology.

Reconstituting a constant residue with enzyme extracts from different flours showed that enzyme activity differed in different flours. The proteolytic enzymes also exhibited different abilities to change sponge rheology when acting on their own flour residue. This may be a useful indicator to determine flour quality and to predict baking performance, as Rogers and Hoseney (1989) have shown that dough rheology and baking performance are related.

LITERATURE CITED

- DOESCHER, L. C., and HOSENEY, R. C. 1985a. Saltine crackers: Changes in cracker sponge rheology and modification of a cracker baking procedure. Cereal Chem. 62:158.
- DOESCHER, L. C., and HOSENEY, R. C. 1985b. Evaluation of commercial cracker flours by test baking and study of changes occurring during sponge fermentation. J. Cereal Sci. 3:261.
- FARIDI, H. 1975. Physical and chemical studies of saltine cracker lengthy fermentation. Ph.D. dissertation, Kansas State University, Manhattan.
- PIZZINATTO, A., and HOSENEY, R. C. 1980. Rheological changes in cracker sponges during fermentation. Cereal Chem. 57:185.
- ROGERS, D. E., and HOSENEY, R. C. 1989. Effects of fermentation on saltine cracker production. Cereal Chem. 66:6-10.
- SALGO, A. 1981. Study of wheat proteases. Ph.D. dissertation, Technical University of Budapest.
- WU, J. Y., and HOSENEY, R. C. 1989. Rheological changes in cracker sponges during the first five hours of fermentation. J. Food Process. Preserv. In press.

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