DETERMINATION OF ULTRASONIC-BASED RHEOLOGICAL PROPERTIES OF DOUGH DURING FERMENTATION[1]

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(Manuscript received August 26, 2003; in final form February 9, 2004)

ABSTRACT

An ultrasonic technique was used to study the changes of the rheological properties of dough during fermentation at 37C and compared with the extensional properties of fermented dough obtained from tensile tests carried out in a Universal Testing Machine. The velocity and attenuation of a longitudinal wave (P-wave) propagated through the dough samples were measured and analyzed to obtain the viscoelastic moduli of the dough; the storage modulus M' *and the loss modulus* M''. *These moduli include both the bulk and the shear moduli. A wavelet analysis also was used to determine the effect of frequency on the ultrasonic-based viscoelastic moduli and the effect of the fermentation*

[1] Approved as Manuscript No. 17155 by the Agricultural Research Programs Division of Purdue University.

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process on the ultrasonic velocity dispersion. A decrease in ultrasonic velocity was observed with increasing fermentation times. Ultrasonic waves were strongly attenuated in the dough subjected to long fermentation times and fermentation had a large influence on the viscoelastic moduli of the dough. The ultrasonic velocity increased with increasing frequency, clearly showing the viscoelastic nature of the fermented dough. The analysis also showed significant ultrasonic velocity dispersion upon fermentation. Ultrasonic measurements yielded results that agreed with those obtained from conventional rheology commonly used to characterize the extensional properties of dough. Both tests clearly showed the loss of elasticity by the dough samples upon fermentation.

INTRODUCTION

Dough fermentation is an important stage in the bread-making process, in which the most distinctive change is the increase in the volume of dough. This stage produces the light, aerated, and spongy characteristics of bread. During the fermentation process, the forces exerted by the growing gas bubbles deform the dough matrix which resists the growth of these bubbles. Therefore, the deformation of the dough matrix produced by the growth of the bubbles is predominantly that produced by an extensional flow.

It is well known that the rheology of dough during the fermentation stage has a strong influence on the final quality of baked bread. Thus, and particularly during the fermentation stage, a knowledge of the extensional properties of dough would allow the fermentation process to be controlled so that the best quality of the baked products could be achieved. However, the measurement of the rheology of fermenting dough is not simple because fermenting dough is a complex and dynamic system. The dimensions and rheological properties of the dough vary with time during fermentation (Czuchajowska and Pomeranz 1993; Rasanen *et al.* 1997). In addition, fermenting dough is extremely fragile, making it extremely difficult to mount a sample in the rheological testing device without disturbing it.

In the past, several investigators have studied the changes in dough properties during fermentation. A rheofermentometer has been used to examine the gas production of fermented dough (Wehrle and Arendt 1998). Microscopy has also been utilized to investigate changes in gas distribution during dough fermentation (Rasanen *et al.* 1997; Shimiya 1997). Ito *et al.* (1992) developed a dielectric method to monitor gas production in fermenting bread dough. Collado and Deleyn (2000) used a test known as the Zymo-expansion-meter to measure the evolution and retention of gas in fermenting dough from 14 different types of wheat. While these methods have been valuable in providing useful information about gas distribution and gas evolution during dough fermentation, they do not provide information about dynamic changes in the rheological properties of dough during fermentation. It would be desirable to directly measure the rate and extent of deformation and the stresses during the entire fermentation process to fully understand the rheological properties of the fermenting dough. Furthermore, there is a need for on-line, rapid, and nondestructive methods for evaluating dough properties.

Rheologically, time-dependent viscoelastic materials, such as fermenting dough, can be studied by propagating ultrasonic waves through the material. The energy in these waves is very low and does not affect the structure of the dough. As they travel through the material, the waves are attenuated and the velocity of the waves changes in response to differences in physical properties. Ultrasound is a novel and challenging method that is being developed for use in the field of food science where nondestructive and noninvasive techniques are of great interest. However, only a few studies have evaluated the rheological properties of dough using ultrasonic techniques. Lee *et al.* (1992) evaluated the rheological properties of dough using ultrasonic waves and found a good agreement between the ultrasonic technique and conventional rheometry. Kidmose *et al.* (2001) obtained the rheological properties of dough during aging using measurements of the velocity of ultrasonic waves. In another study the physical properties of dough were analyzed using a theory that describes the propagation of ultrasonic waves through a viscoelastic material (Letang *et al.* 2001) who studied the effects of the dough water content and mixing time and found that the ultrasonic properties of dough were dependent on frequency which is the typical behavior of viscoelastic materials.

To our knowledge, the feasibility of using ultrasound to characterize the rheological properties of dough during fermentation has not been studied. Therefore, the main objectives of this work were (1) study the changes in the ultrasonic properties of dough throughout the fermentation process, (2) determine changes in dough rheological properties during fermentation using ultrasonic data, (3) evaluate extensional properties of dough upon fermentation and compare them with the ultrasonic-based rheological properties and (4) highlight the potential of ultrasound for dough testing during processing.

MATERIALS AND METHODS

Sample Preparation

Commercial all-purpose flour (14% moisture basis) was used to prepare the dough samples used in this study. The dough was prepared using 35 g of flour, 27 g distilled water, and 0.35 g dry yeasts (Red Star® Yeast, Milwaukee, WI). The ingredients were mixed in a 35 g mixograph mixer (National MFG. Co., Lincoln, NE) for 6 min (approximately optimum mixing time). No other ingredients beyond flour, water and yeast were used to prepare the dough samples.

Ultrasonic Measurements

Experimental Set-up. Figure 1 illustrates the experimental set-up used for the ultrasonic measurements. A Panametrics (Waltham, MA), model 5800, pulser-receiver system connected to a PC was used for the ultrasonic measurements. LABVIEW software (National Instruments, Austin, TX) was used for data acquisition and analysis. The transducers used in this experiment were two Panametrics broad-band V609, 5 MHz piezoelectric transducers which were calibrated prior the tests by measuring the speed of sound through distilled water. In order to follow the physical changes of the dough samples during the fermentation process ultrasonic measurements were carried out using the transmission mode. As illustrated in Fig. 1, one of the two transducers was used as the source while the other was used as the receiver. A suitable measurement cell was designed for the test. It consisted of two Plexiglas™ plates having threaded holes for the insertion of the transducers. The region surrounded by the dotted circle in Fig. 1 has been expanded in the same figure to show the measurement cell in more detail. The sample was placed between the two transducers and inside the cell which was sealed by the two plates to prevent moisture evaporation during the fermentation process. During measurements the distance between the two transducers was held constant at 3 mm. Fermentation of the sample was facilitated by placing the measurement cell containing the dough sample in an oven at 37C. Ultrasonic signals were collected every 5 min for a total of 30 min. All experiments were carried out in triplicate.

Analysis of Ultrasonic Wave Propagation Through a Viscoelastic Material. The ultrasonic velocity and attenuation coefficient were determined from the propagation time or Time of Flight (TOF) and from the decrease in the amplitude of the ultrasonic wave that propagated through the sample of known thickness, respectively. The propagation time or TOF was calculated by measuring the time period between the peak of the emitted and transmitted pulse waves and the ultrasonic wave velocity as the sample thickness divided by the determined TOF.

For longitudinal waves propagating in a viscoelastic material, the longitudinal-wave complex modulus M^* can be calculated as $M^* = M' - iM'' = K^* + \frac{4}{3}G^*$

where K^* ($= K' - iK''$) and G^* ($= G' - iG''$) are the complex bulk and shear moduli, respectively (Marvin *et al.* 1954; Kono 1960). K' and G' are the bulk

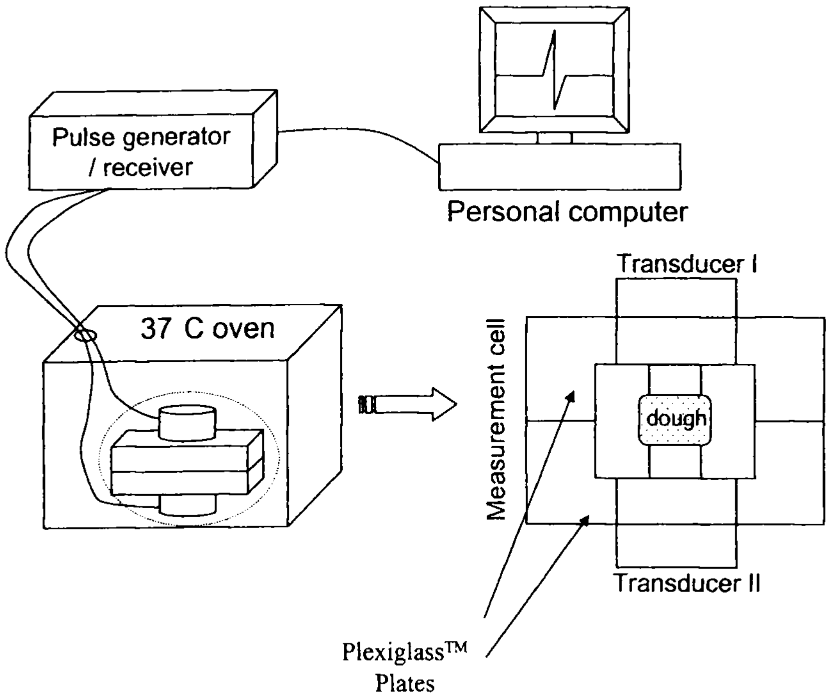

FIG. 1. ILLUSTRATION OF THE EQUIPMENT USED FOR THE ULTRASONIC MEASUREMENTS ON DOUGH DURING FERMENTATION

and shear storage moduli, whereas K'' and G'' are the bulk and shear loss moduli, respectively. The former give an indication of the elastic properties whereas the latter give the viscous properties of the dough. The relationships among the moduli, ultrasonic velocity, and attenuation at a given frequency (ω) are used to calculate the moduli M′ and M″:

$$M' = K' + \frac{4}{3}G' = \frac{\rho \upsilon^2 \left(1 - \frac{\alpha^2 \upsilon^2}{\omega^2}\right)}{\left(1 + \frac{\alpha^2 \upsilon^2}{\omega^2}\right)^2} \tag{1}$$

$$M'' = K'' + \frac{4}{3}G'' = \frac{2\rho\upsilon^2 \frac{\alpha\upsilon}{\omega}}{(1 + \frac{\alpha^2\upsilon^2}{\omega^2})^2} \tag{2}$$

where ρ, ν, and α are the density, the ultrasonic velocity, and the attenuation coefficient of the dough, respectively, and ω is the frequency of the wave.

The dough density, needed to calculate the moduli of dough using Eq. (1) and (2) was evaluated, separately from the ultrasonic measurements, during the fermentation process. The density of the fermented dough was found by placing the sample between two transparent Plexiglass™ plates separated by a fixed distance of 3 mm before it was placed in the 37C oven. During the fermentation process, pictures of the fermenting dough located between the plates were taken with a digital camera Kodak, DC290. As a consequence of the fermentation the dough spread radially between the plates and the cross-sectional area of the dough sample was determined from the digital picture using image processing software (Photoshop®, Adobe Systems, San Jose, CA). The density was calculated using the measured area, the thickness, and the weight of the dough piece. Density measurements were done in triplicate and a plot of measured dough densities as a function of fermentation times is illustrated in Fig. 2.

Wavelet analysis was also used to study the ultrasonic signals. Wavelet analysis is a time-frequency localization procedure that provides information on ultrasonic velocity dispersion and structural characteristics of the material that cannot be obtained with the traditional Fast Fourier Transformation (FFT). The use of this analysis is relatively new in the field of food science. It was used in the investigation of the properties of frozen food systems (Lee *et al.* 2004) using ultrasonic techniques. The analysis was implemented in the Matlab software (The MathWorks, Natick, MA) and is based on the method described in these previous studies (Nolte *et al.* 2000; Lee *et al.* 2004).

Extensional Properties Measurements

An Universal Testing Machine (MTS Systems Corp., Madison Heights, MI) was used to characterize the extensional properties of the dough samples during fermentation. The method introduced by Uthayakumaran *et al.* (2000) was modified and used for this work. An oven set at 37C was installed on the Universal Testing Machine to allow rheological measurement to be taken during the fermentation process. The dough sample was placed between two circular 3 cm diameter plates. The plates were serrated to increase the adhesion of the sample to the plates and the sample, and thus to ensure that the sample did not

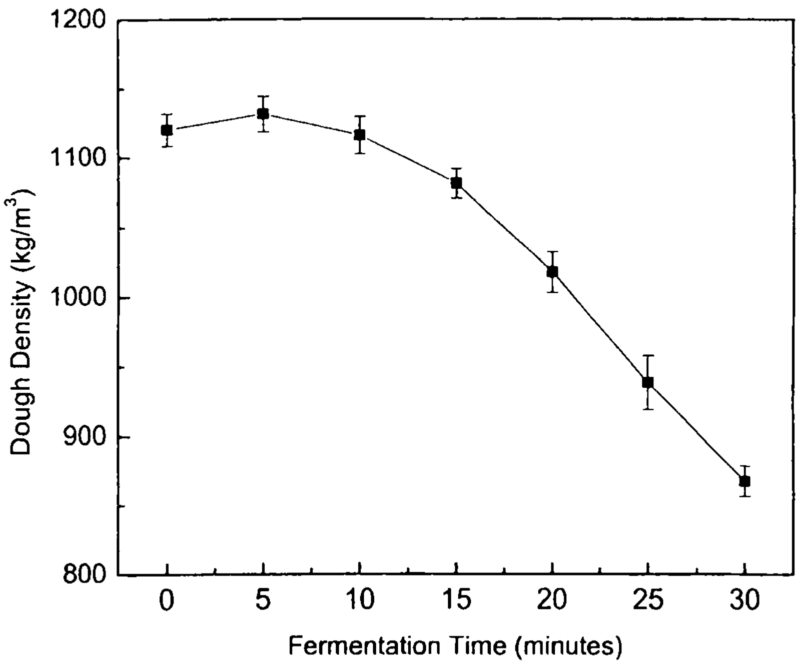

FIG. 2.THE EFFECT OF FERMENTATION TIME ON THE DOUGH DENSITY

separate from the plate during tensile testing. The lower plate was fixed and the upper plate was attached to the crosshead of the Universal Testing Machine. The crosshead and plate moved upwards with a velocity of 50 mm/min to elongate the dough sample until it was broken (Fig. 3). To avoid artifacts that could be caused by fracture of the sample during loading and/or lack of adhesion to the plates, only samples in which the breakage occurred in the midsection of the sample were analyzed. The force and distance recorded by the Universal Testing Machine software were used to obtain strain, strain rate, stress and extensional viscosity of the samples. As described by Uthayakumaran *et al.* (2000), stress and strain were calculated from the crosshead displacement and the sample momentary cross-sectional area, which was estimated assuming that the sample volume was kept constant and the sample shape was kept cylindrical during the measurement. The extensional viscosity of the samples was the ratio of the calculated stress during extension and the strain rate. All the measurements were done in triplicate using a different sample for each fermentation time. In this method the strain rate is not constant during the extension of the sample. A method to achieve constant strain rates during extensional flow tests is discussed by Uthayakumaran *et al.* (2000). It consists in varying the cross-head speed so that the imposed Hencky's strain rate is constant. Methods that use constant crosshead speeds, and thus time varying strain rates, as applied to squeezing flow viscometry are discussed by Campanella and Peleg (2003). The fact that the applied strain rate during the test is not constant may not qualify the applied extensional test used as a fundamental one. However, the main objective of this work was to introduce a novel technique to testing fermenting dough and compare it with results of commonly used techniques in dough research.

The SAS ANOVA test (SAS Institute, Cary, NC) was used to determine significant differences among the samples.

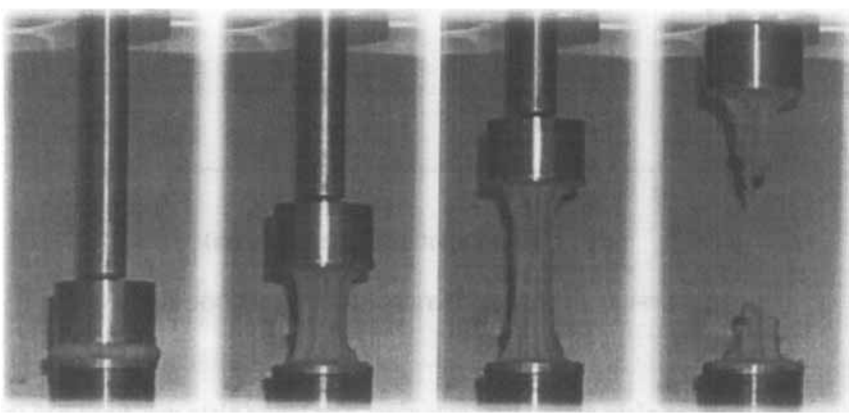

FIG. 3. PICTURES OF A DOUGH SAMPLE DURING AN UNIAXIAL EXTENSIONAL TEST

RESULTS

The measured ultrasonic velocity decreased significantly during fermentation (Fig. 4). The change in the density of the dough sample due the increase of the bubble volume during fermentation is a possible reason for the reduction in the ultrasonic velocity [Fig. 2]. Wehrle and Arendt (1998) showed that the presence of CO_2 bubbles produced by yeast in fermenting dough may affect the elastic dough network, resulting in a more viscous structure. This more viscous behavior of the fermenting dough would cause a decrease in the measured ultrasonic velocity.

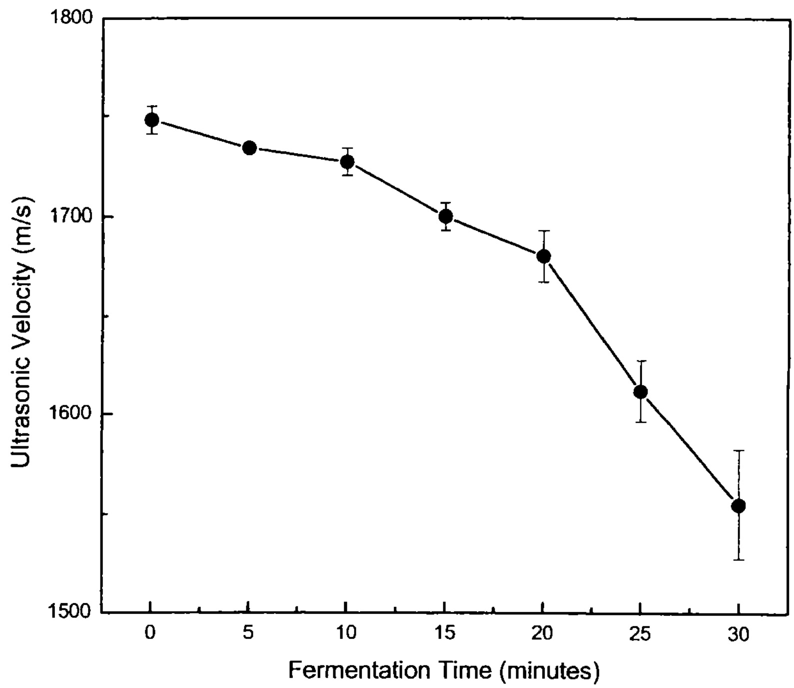

FIG. 4. ULTRASONIC VELOCITIES MEASURED IN A DOUGH SAMPLE DURING FERMENTATION

Figure 5 shows that the attenuation coefficient of the dough increased during fermentation. At the beginning of the fermentation process, the ultrasonic attenuation, as well as ultrasonic velocity, appeared to be nearly constant or decreased slightly. Figure 5 shows that ultrasonic waves were highly attenuated after 10 min showing the effect of the bubble growth. The CO_2 production by yeasts does not increase the number of gas bubbles but instead increases the size of the bubbles already formed (Baker and Mize 1941). The lag period observed in the ultrasonic properties of the fermented dough has also been reported during the measurement of CO_2 production in dough fermentation and has been attributed to a phenomenon associated with the solubility of CO_2 in the dough aqueous phase surrounding the bubble. At the start of the fermentation process CO_2 diffuses through the aqueous phase of the dough matrix until it becomes saturated with CO_2. After the aqueous phase has been saturated the CO_2 is available to leaven the system (Hoseney 1994). As fermentation proceeds the CO_2 cannot diffuse out of the dough matrix and both the pressure inside the bubbles and their volume increase. As noted in Fig. (4) and (5) this phenomenon is detected by the measured ultrasonic properties because the expansion of gas bubbles increases the interfacial area between the gas cells and the continuous dough phase giving rise to more scattering of the ultrasonic waves. This decreases the ultrasonic velocity and increases the ultrasonic wave attenuation.

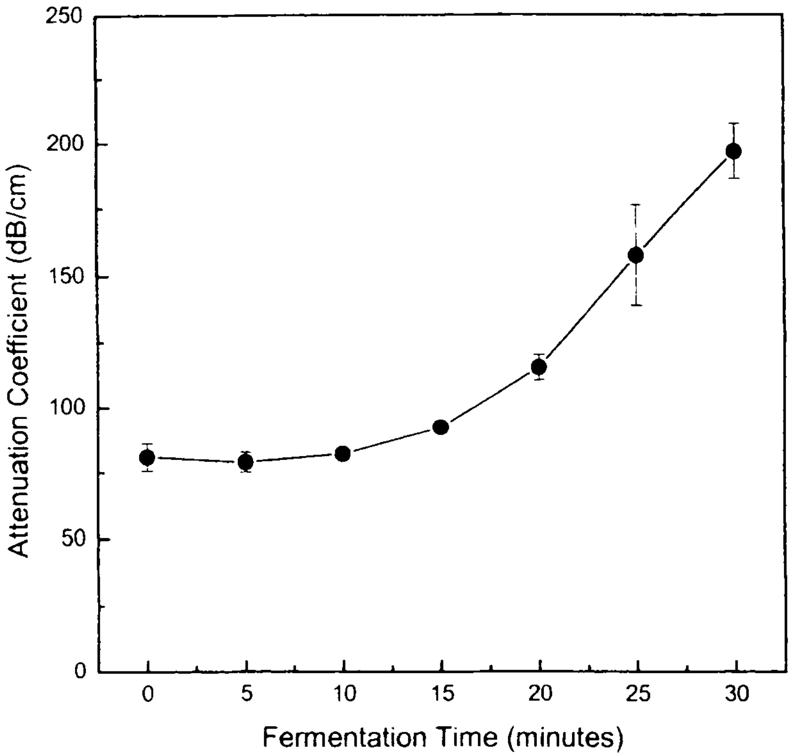

FIG. 5. ATTENUATION COEFFICIENTS FOR A DOUGH SAMPLE DURING FERMENTATION

The rheological properties of dough varied distinctly during fermentation. The viscoelastic moduli M' and M'' calculated from Eq. (1) and (2) are illustrated in Fig. 6. The elastic nature of the dough samples apparently makes the storage modulus (M') significantly larger than the loss modulus (M''). The storage modulus (M') decreases during the fermentation process ($P < 0.01$) indicating that the dough properties become less elastic during fermentation. The loss modulus (M'') did not change significantly during fermentation ($P > 0.05$) so the ratio of M'' to M' increased during fermentation. That ratio known as *tanδ*, is a rheological parameter often used in studies of foods to identify transition phenomena induced by temperature, notably the glass transition (*Tg*) range. The results obtained in this study clearly show that the dough sample became less elastic during fermentation.

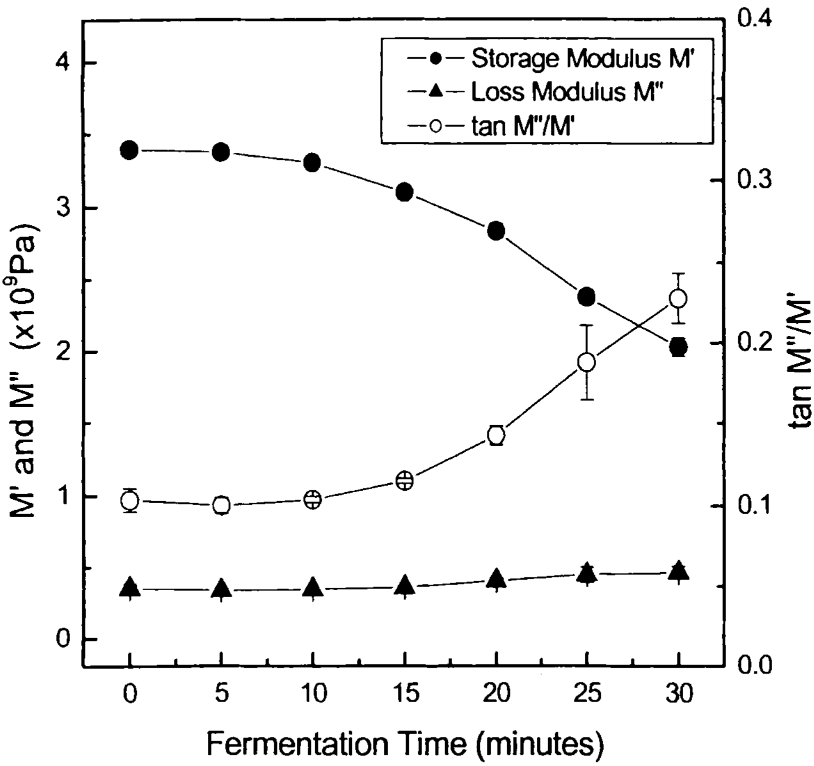

FIG. 6. THE EFFECT OF THE FERMENTATION PROCESS ON THE VISCOELASTIC MODULI M' AND M" OF A DOUGH

Wavelet analysis was used to investigate the dependence of the ultrasonic velocity on frequency and the dispersion of the velocity of ultrasonic waves propagating through the fermenting dough samples. Velocity dispersion is often associated with the physical structure of a material (i.e., defects, discontinuities, fractures, etc.) that causes ultrasonic waves with different frequencies to propagate at different velocities. This behavior depends on the physical properties of the material being tested. Group wavelet transforms of the ultrasonic signals produced by the dough samples during fermentation are shown in Fig. 7. Transmitted wave amplitude as a function of time for different fermentation stages are depicted on the left side of Fig. 7. The X and Y axes in the two-dimensional wavelet transforms (right plots) are frequency and time, respectively. The contour plots of the transform amplitudes represent different levels of energy of the ultrasonic waves. Small circular contours represent waves with high energy whereas large circular contours represent waves with lower energy. One of the distinct features in the transformation illustrated in Fig. 7 is the shape of the energy profile upon fermentation. Waves of low energy arrive later than those with high energy, implying velocity dispersion. Moreover, the waveform at the beginning of fermentation has a single wave packet. After 20 min, the waveform began to divide into several wave packets, and became more complex. As illustrated in Fig. 7, each wave packet had a different arrival time, that is, a different velocity. This suggests that the ultrasonic wave was scattered as it propagated through the sample. Spatial heterogeneity in the dough can arise throughout the fermentation process from changes in the dough volume and moduli that are caused by growth and retention of the gas produced by yeasts. For this analysis a fermentation time of 25 min was used because after 30 min of fermentation the attenuation of the ultrasonic waves was high and an accurate wavelet analysis could not be made, even though ultrasonic velocity and attenuation could be estimated.

Figure 8 shows the effect of frequency on the ultrasonic velocities calculated from the wavelet analysis. For all frequencies, the velocity of ultrasonic waves was lower in more fermented dough, which agrees with the results shown in Fig. 4. For all samples the ultrasonic velocity increased with an increase in frequency and the effect was more noticeable in the more fermented dough. These results indicate the viscoelastic nature of the fermented dough and the more viscous behavior of samples fermented for longer fermentation times. The sample dispersed more of the ultrasonic waves after fermentation. Dispersion of the ultrasonic velocity was more significant in more fermented samples. For instance, it was estimated that the ultrasonic velocity increased about 60% in the range of frequencies 2-6 MHz for the more fermented sample (25 min) whereas for the unfermented sample the ultrasonic velocity only increased about 20% in the same range of frequencies. For frequencies greater than 7 MHz, ultrasonic velocities were nearly constant at all fermentation times, indicating that at those frequencies the dough was not dispersive and its behavior was more elastic.

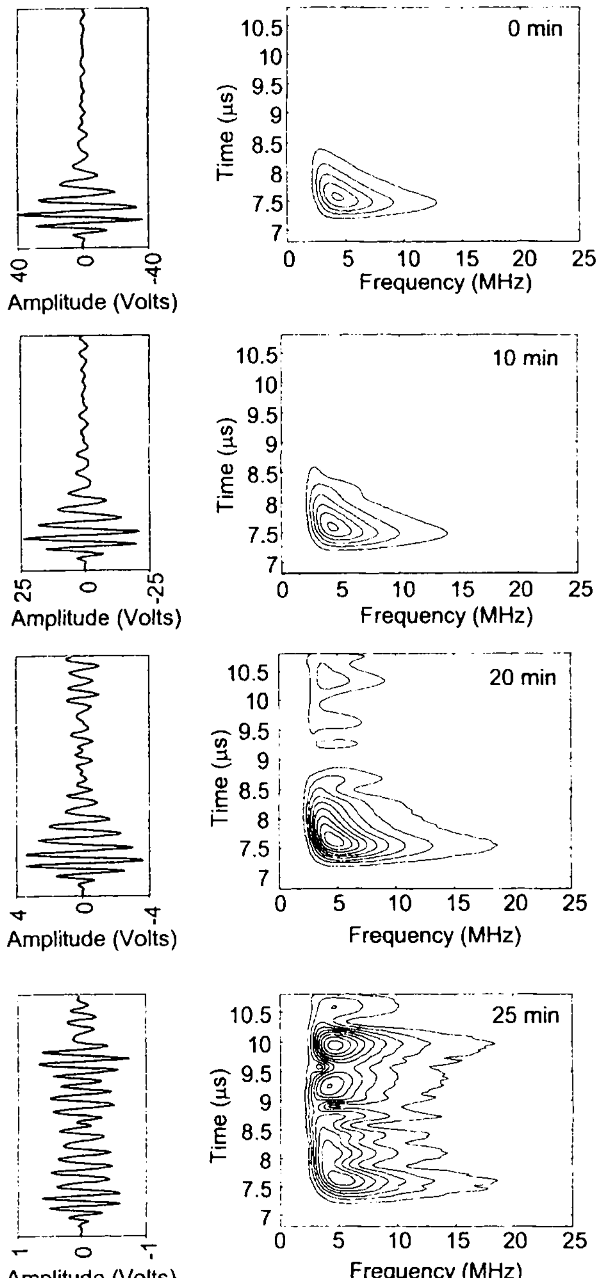

FIG. 7. WAVELET TRANSFORMS OF ULTRASONIC SIGNALS DURING FERMENTATION OF A DOUGH SAMPLE
(The time in the upper corner of the wavelet transforms indicates fermentation times).

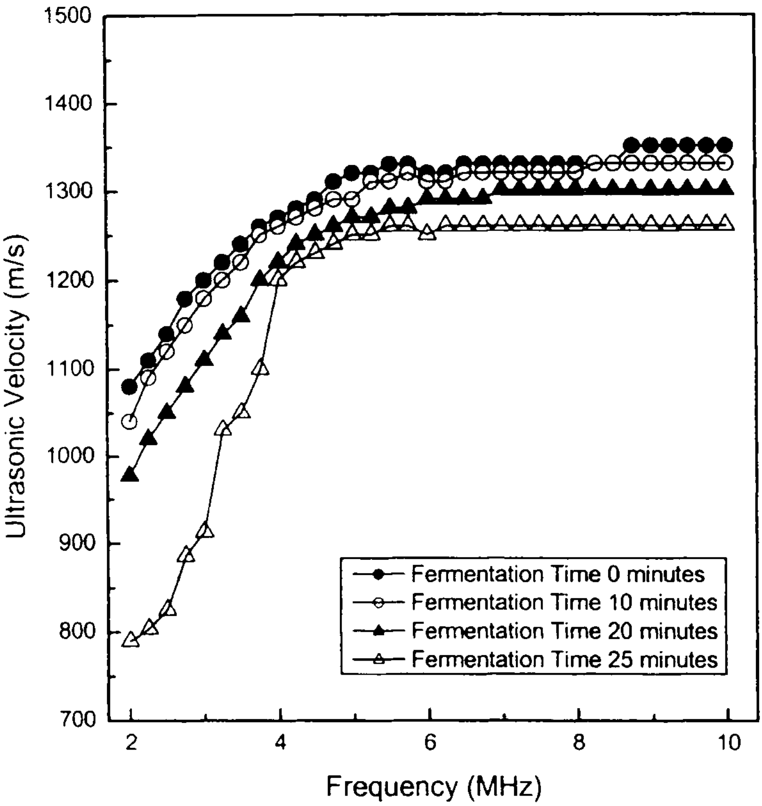

FIG. 8. VELOCITIES CALCULATED FROM WAVELET ANALYSIS AS A FUNCTION OF A FREQUENCY DURING FERMENTATION OF A DOUGH SAMPLE

Using the ultrasonic velocities and attenuation coefficients obtained from the wavelet analysis, the variations of the dynamic moduli of dough were determined as a function of frequency at different fermentation times [Fig. 9]. Figure 9a illustrates that for all frequencies, the more fermented dough had lower values of M'. This is consistent with the results presented in Fig. 6. More fermented doughs exhibited more frequency-dependent properties, as demonstrated by the differences in M'' for frequencies between 2 and 4 MHz [Fig. 9b]. Figure 9c illustrates the changes of *tan* M''/M' with frequency. In general the results follow the expected trends for fermentation times between 0 to 20 min. However there was an unexpected increase in the *tan*M''/M' for 25 min fermentation time in the range of 2-4 MHz. As discussed previously the viscous behavior of fermented dough could be attributed to changes in the rheology of the dough matrix and the increase in the volume of the gas phase upon fermentation. It appears that the dispersion of the ultrasonic waves in the range of frequencies 2-4 MHz for highly fermented dough is related to the growth of bubbles and to a lesser extent by the changes in the rheology of the dough matrix.

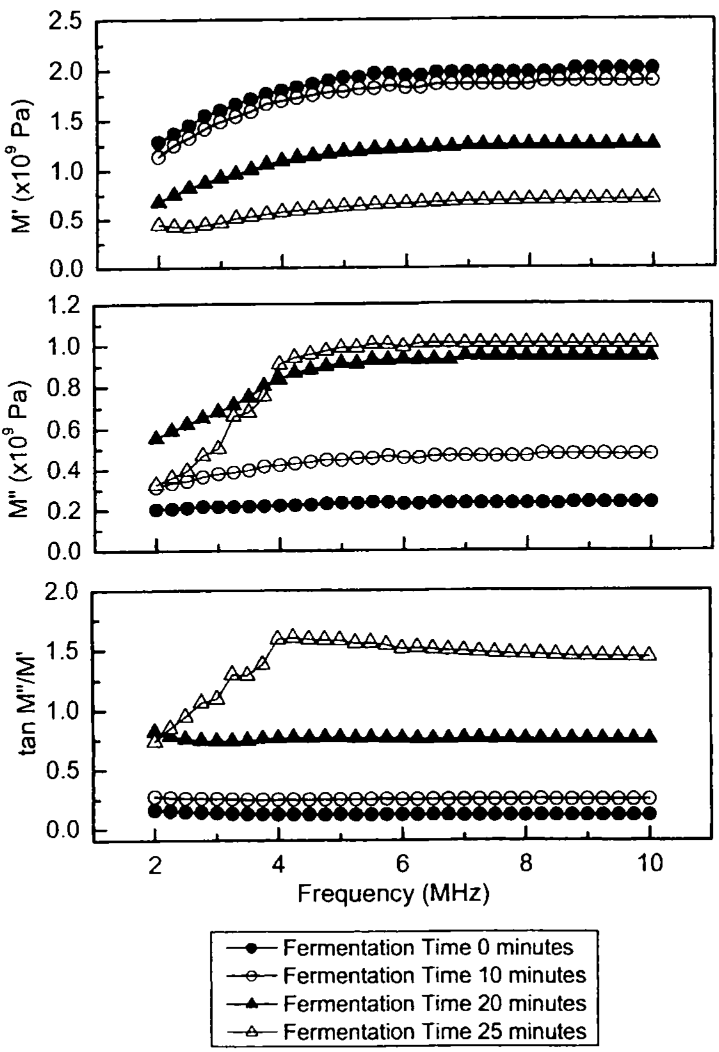

FIG. 9. CHANGES OF THE VISCOELASTIC PARAMETERS M', M", AND M"/M' AS A FUNCTION OF A FREQUENCY DURING FERMENTATION OF A DOUGH SAMPLE

A typical uniaxial extension curve for dough is shown in Fig. 10 (a). The extensional viscosity increased with increasing strain. This increase can be attributed to the strain hardening properties of dough that have been reported by others (Uthayakumaran *et al.* 2000). The sharp drop in the extensional viscosity curve indicates that for a strain higher than the peak strain the dough breaks. Thus, the value of the peak viscosity corresponds to the dough maximum resistance to extension.

As shown in Fig. (10b) the peak extensional viscosity decreased with increasing fermentation times. For the sake of comparison, values of storage modulus M' were superimposed on the same graph. Figure (10b) indicates nearly identical trends in both conventional and ultrasonic-derived rheological properties with time of fermentation. The experimental error in the conventional extensional test was higher as is illustrated in Fig. (10b). Therefore, the statistical significance of the fermentation time effect on the ultrasonic measurements was greater ($P < 0.01$) than the statistical significance of its effect on the extensional measurements ($P < 0.05$). This indicates that an ultrasonics sensor has great potential for testing dough properties during processing.

CONCLUSIONS AND DISCUSSION

The rheological properties of fermenting dough were investigated using ultrasound. The fermentation process had a great influence on the ultrasonic properties of dough, which were frequency-dependent. In general, the elastic behavior of dough decreased during fermentation. Ultrasonic measurements exhibited a trend similar to that measured by conventional extensional rheology. However, ultrasonic measurements were more consistent and had a significantly lower experimental error. This is probably attributable to the nondestructive nature of the ultrasonic measurement, which allows one to utilize the same sample for testing at all fermentation times. Conversely, a new sample has to be tested for each fermentation time when the conventional rheological test is used.

Ultrasonic longitudinal waves were used in this research. This enabled the calculation of the moduli M' and M''. These moduli incorporate both the bulk and shear properties of the sample. The bulk and shear properties could be separated if the ultrasonic measurements were conducted using shear waves. This would allow one to calculate the shear storage modulus G' and the shear loss modulus G''. Unfortunately dough has a very high attenuation, particularly when shear waves are used, and it is difficult to obtain reliable data from the tests carried out using shear wave transducers. However, it has been shown (Zheng *et al.* 2000) that the properties of dough may respond in a similar

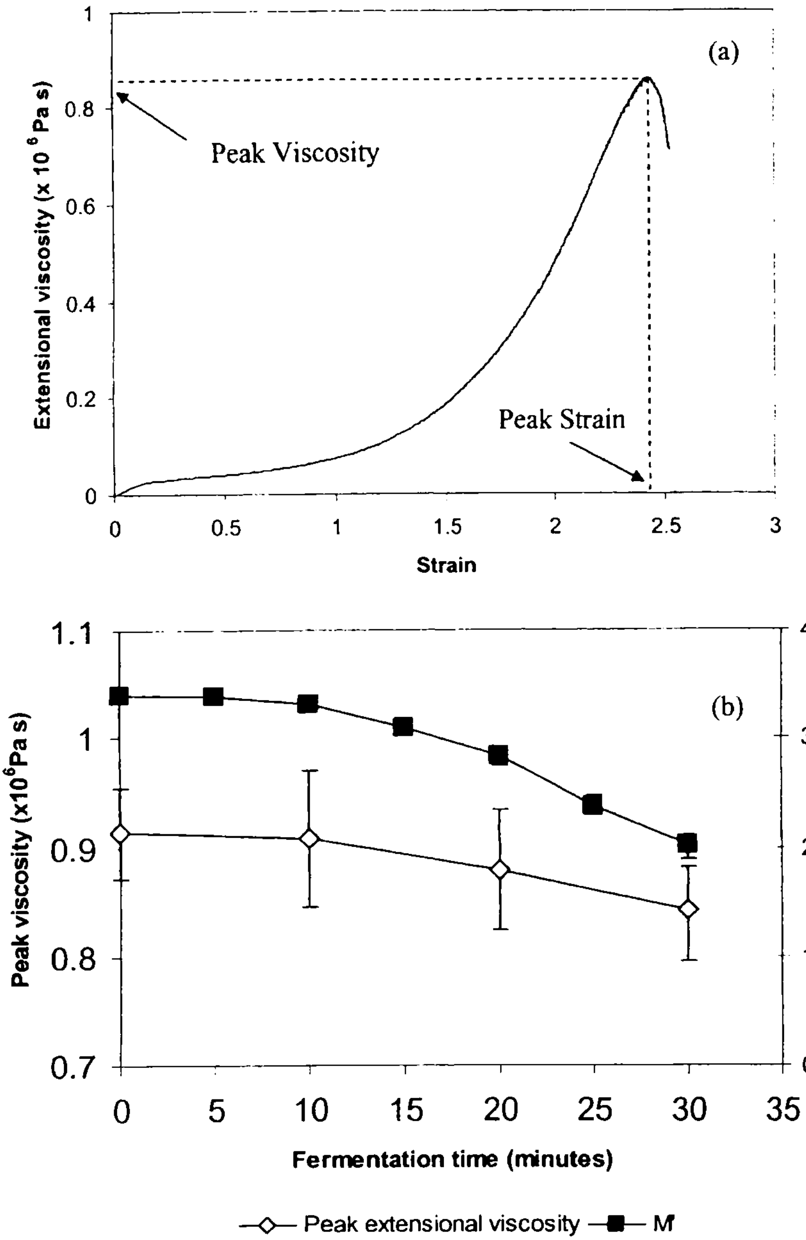

FIG. 10. EXTENSIONAL PROPERTIES OF DOUGH DURING FERMENTATION
(a) Typical uniaxial extension of a dough sample obtained using a Universal Testing Machine.
(b) Changes in the extensional viscosity during fermentation and its correlation with ultrasound measurements.

manner to shear or extensional tests or a combination of them. The trend observed in the rheological properties obtained from the ultrasonic-based rheology was almost identical to the trend obtained using conventional rheology. The ultrasonic tests were even able to detect the lag phase of the fermentation process. Therefore, ultrasonic methods can be used to evaluate the rheological properties of dough during fermentation. Wavelet analysis provides time (kinetic) and frequency (dynamic) information at the same time, thus giving a better understanding of the physical properties of a system and how it is changing with the time. Wavelet analysis has been generally used in the fields of mathematics, geophysics and physics. This is a new and promising approach to ultrasonic-based rheology research.

Ultrasonic methods have the potential to be used on-line to investigate nondestructive dough fermentation because ultrasonic velocity and attenuation are sensitive to the physical changes in dough caused by the fermentation process.

ACKNOWLEDGMENTS

This research was supported by Purdue Research Foundation and the Whistler Carbohydrate Research Center. Thanks also go to Dr. Richard Stroshine and Dr. Byoung-Kwan Cho for their critical reading of the manuscript.

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