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Molecular mobility in model dough systems studied by time-domain nuclear magnetic resonance spectroscopy

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Abstract

The microscopic distribution and dynamic state of water and molecular mobility in various model systems are investigated using time-domain NMR spectroscopy. Starch and gluten showed different continuous distribution populations in T_{21} (μsec range, obtained from One pulse experiments) and T_{22} (msec range, obtained from CPMG experiments) proving that starch and gluten have different water dynamics and molecular mobility. A starch/gluten mixture (76:12, w/w) and wheat flour dough exhibited similar patterns indicating that water and molecular mobilities in dough tended to be more representative of interactions with starch than gluten, even though both water–starch and water–gluten interactions are occurring in wheat flour dough. Increasing the water content did not influence the continuous distribution pattern of T_{21} but affected the relative amount of each fraction in T_{21} (i.e. an increase of the more mobile fraction and a decrease of the less mobile fraction with increasing moisture). Added water has an important role in the more mobile fraction but not in the less mobile fraction, which is in μs range. This indicates that model food systems contain multiple microstructural domains with various water and molecular mobilities that show correspondingly different water dynamics. Therefore, the dispersion of various relaxation time constants helped identify the distribution of independent microstructural domains. The manipulation of the composition of the model food system influences the water dynamics and molecular mobility and provides a basis for the application of the microstructural domain concept to real food systems.

Keywords: Starch; Gluten; Dough; Water mobility distribution; Time-domain NMR spectroscopy

1. Introduction

Water mobility has an important effect on the overall mobility and structural properties of metastable food polymer systems, such as wheat flour dough. Although water molecules interact with one another by hydrogen bonding, they are in a dynamic state and show very rapid translational and rotational movements. The population of water molecules that is associated with macromolecules through a variety of possible mechanisms shows thermo-dynamically or kinetically different behavior from those of bulk water (Given, 1991).

Since starch and gluten are the main structural components of wheat flour dough, their hydration behavior of significant practical interest. The effects of water on starch and gluten are manifested by the complex chemical nature of both polymers and hence their interaction with water is complex (Li et al., 1996). However, not all water is intimately associated with the starch or gluten, as these two polymers are poorly water-soluble. Although starch and gluten seem plasticized by water, water is not necessarily mixed homogeneously with these two polymers. Thus, a marked difference in water mobility may be observed between gluten and starch. For instance, although water in a glassy starch was proposed to be immobile and rigid like the polymer starch, it has more recently been reported to be highly mobile (Li et al., 1996). Umbach et al. (1992) also

Abbreviations: CPMG, Carr-Purcell-Meiboom-Gill; FID, free induction decay; NMR, nuclear magnetic resonance; T_2, spin–spin relaxation time; T_{21}, spin–spin relaxation time obtained from 90° pulse; T_{22}, spin–spin [redacted]

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found that a small fraction of water in dry starch was very tightly associated, but additional water did not interact with the starch and remained quite mobile. Further hydration led to more water–gluten interactions (Umbach et al., 1992). It was obvious that the molecular mechanism of water–gluten interaction is distinct from a water–starch interaction (Li et al., 1996).

Proton NMR relaxation times have been used extensively to investigate the state of water in dough (Assifaoui et al., 2006; Belton et al., 1995; Choi and Kerr, 2003; Given, 1991; Leung and Steinberg, 1979; Leung et al., 1983; Li et al., 1996; Richardson et al., 1986; Ruan et al., 1999; Toledo et al., 1968). The interpretation of NMR relaxation time measurements of food systems has been model dependent. Many researchers have used multiexponential models to analyze NMR relaxation data to represent complex food systems. In NMR relaxation data analysis using multiexponential models may suffice to interpret homogeneous or relatively simple model systems, but they do not provide satisfactory solutions for more heterogeneous food systems. Therefore, it has been suggested that continuous distribution of relaxation times is a better representation of the information contained in relaxation experiments (Lillford et al., 1980). A number of researches have been carried out using continuum models to follow the relaxation behavior in various systems (Araujo et al., 1992; Assifaoui et al., 2006; Bertram et al., 2002; Choi and Kerr, 2003; Cornillon, 2000; Hills et al., 1996; Kroeker and Henkelman, 1986; Le Botlan and Ouguerram, 1997; Lillford et al., 1980; Menon and Allen, 1991; Newcomb et al., 1990; Ruan et al., 1999; Tang et al., 2000, 2001; Tellier et al., 1993; Whittall and MacKay, 1989). Among them, to the authors' knowledge, only two reports regarding the use of continuous distribution of NMR relaxation data in dough system are available. They focused on the comparison between the multiexponential discrete model and the continuous model with varying moisture content (Ruan et al., 1999) and characteristics of water mobility in biscuit dough (Assifaoui et al., 2006).

The objectives of this study were to investigate the role of moisture and constituent concentrations on molecular water dynamics and molecular mobility in wheat flour dough using the continuous distribution analysis of NMR relaxation data (spin–spin, T_2) and to gain insight into the nature of the molecular interactions of water–starch, water–gluten and water–dough systems.

2. Materials and methods

2.1. Materials

Wheat starch (Gemstar 100 Plus, 11% moisture), wheat gluten (Gem of the West Vital Wheat Gluten, 7% moisture) and wheat flour (76% starch, 12% protein, 12% moisture) were kindly donated from Manildra Co. (Hamburg, IA). Wheat flour was used to make model dough (33.1–47.2% moisture). Wheat starch and wheat gluten were used to make mixtures of starch/water (41.1% moisture), gluten/water (41.1% moisture), and starch/gluten/water (41.1% moisture). In the case of the starch/gluten/water mixture, starch and gluten (76:12, dry basis) were mixed first and then water was added to achieve the intended moisture content. Duplicate samples were used for all NMR experiments.

2.2. NMR analysis

Approximately 10 g of sample was placed in a disposable glass test tube (13 mm O.D. × 100 mm length, Fisher Scientific, Pittsburgh, PA). The tube was sealed with Parafilm M (Fisher Scientific, Pittsburgh, PA) to prevent moisture loss during the measurement. A 20 MHz PCT 20/20 NMR Analyzer (Process Control Technology Corporation, Ft. Collins, CO) was used to perform all NMR experiments at 30 °C. The 90° pulse (One pulse) sequence and the Carr-Purcell-Meiboom-Gill (CPMG) pulse sequence were used for acquisition of the free induction decay (FID) data for spin–spin relaxation times, T_{21} and T_{22}, respectively. The pulse width and sequence repetition time for both pulses were 20 μs and 1 s, respectively. The dwell time between data was 0.7 μs for the One pulse and 100 μs for the CPMG. The number of data points acquired was 120 for the One pulse and 60 for the CPMG. Sixteen scans were accumulated to increase the signal-to-noise ratio. The FID obtained from a 90° pulse sequence (T_{21}) and the Carr–Purcell–Meiboom–Gill (CPMG) pulse sequence (T_{22}) were analyzed as a continuous distribution of exponentials using RI WinDXP software (version 1.2.2, Resonance Instruments Ltd., Oxfordshire, UK).

3. Results and discussion

3.1. Effects of constituents

Fig. 1 depicts the continuous distribution of spin–spin relaxation time, T_{21} (obtained from One pulse experiment) of water in starch, gluten, starch/gluten mixture (76:12, dry basis) and dough samples at 41.1% moisture content. Continuous distributions of T_{21} of all samples showed two peaks suggesting that there are two groups exhibiting distinctively different mobilities. Because T_{21} values of these two groups are distributed in the microsecond range, proton signals falling into these two groups can be regarded as from the solids (gluten and starch) or water molecules closely associated with the solids (Ruan et al., 1999). Two proton populations were also observed from a single pulse for biscuit dough at 19.4% moisture content (at 25 °C) (Assifaoui et al., 2006). They suggested that the population associated with the less mobile region may correspond to protons in solid like components, such as starch, proteins and water molecules tightly associated with those of solids. The second population may correspond to more mobile protons in the biscuit dough. However, they

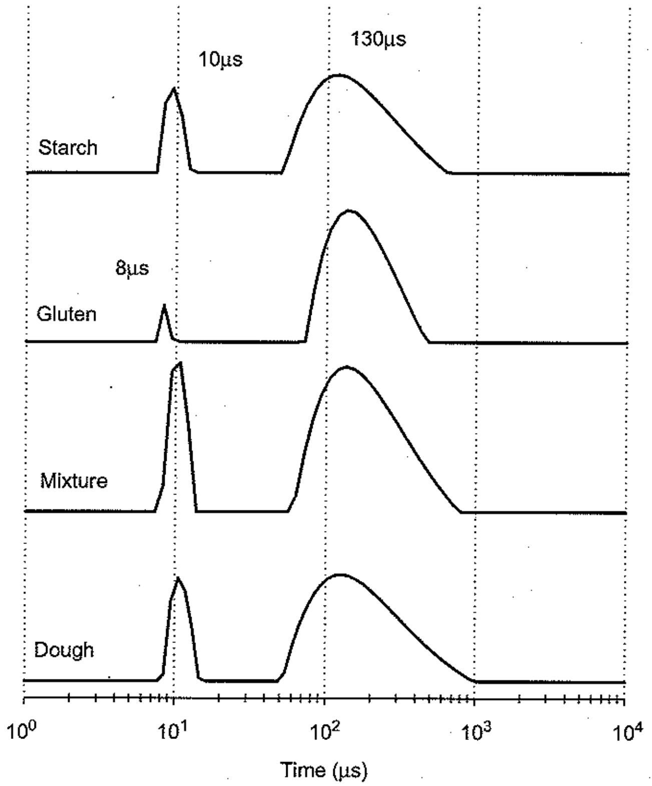

Fig. 1. Continuous distribution of spin–spin relaxation times (T_{21}, obtained from one pulse experiment) of various samples at moisture content of 41.1%.

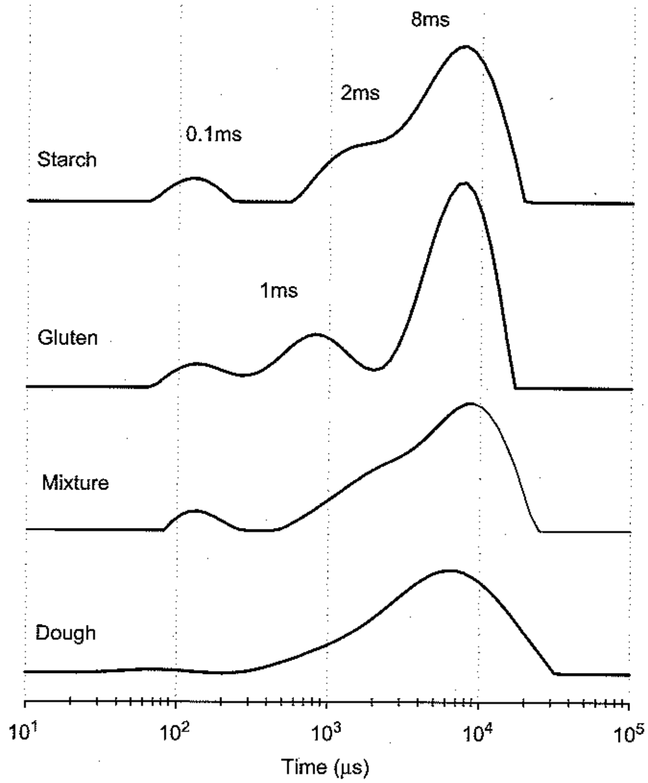

Fig. 2. Continuous distribution of spin–spin relaxation times (T_{22}, obtained from CPMG experiment) of various samples at moisture content of 41.1%.

proffered that the high time constant values T_2 measured using FID are not true spin–spin relaxation time because the FID signal contains not only the spin–spin relaxation but also the lost signal due to local inhomogeneities in the magnetic field. Therefore, caution must be taken when comparing and using T_2 data (Assifaoui et al., 2006).

It is interesting to note that the two peaks of gluten have relatively narrow dispersions compared to the other samples, which are coincidental with the two peaks of starch. This suggests that proton signals from the solids or water molecules very close to the solids might not be easily distinguishable in the case of starch and gluten.

Fig. 2 shows the continuous distribution of the spin–spin relaxation time, T_{22}, for various samples at 41.1% moisture content obtained from CPMG experiments. Gluten shows three distinct populations and the distributions of the other samples show two populations with one merged broad population in the range of 0.4–30 ms. Therefore, only among the three peaks in gluten is the peak near 1 ms distinguishable from the merged peak of starch in the range of 0.5–20 ms. However, this little difference would not be enough to differentiate the water and molecular mobility in starch and gluten.

It is generally recognized that starch and gluten have different water binding capacities. However, the location and behavior of water during dough preparation is still controversial. One group suggested that there was more water associated with the gluten than with the starch in the dough and water moves from the gluten to the starch during conventional baking (Umbach et al., 1992; Willhoft, 1971). The other group proposed that more water is associated with the starch than with the protein, thus there would be more hydrogen atoms associated with starch-interacting with water than protein interacting with water (Bushuk and Hlynka, 1964). This group also reported that 46% of the water in dough is associated with starch, 31% with protein, and 23% with pentosan gum (Bushuk, 1966).

According to those reports, water mobility in wheat flour dough would not be solely manifested by one component because both water–starch and water–gluten interactions would influence the overall water mobility in a dough system. They should exhibit the mixed spectra representative of the different components. In this work, gluten showed little differences in its continuous distribution of T_{21} and T_{22} (peak shape and range) compared to starch. However, the continuous distribution of T_{21} and T_{22} of the starch/gluten mixture and the wheat flour dough were not strongly influenced by gluten, but they were greatly affected by starch, which is the major component (88% dry basis) of those systems. This indicated that water and molecular mobility in dough is more representative of starch than gluten, even though both water–starch and water–gluten interactions are occurring in wheat flour dough. Because water mobility in dough is highly influenced by moisture content, more work is needed to prove this hypothesis. Therefore, further research to determine the effect of moisture content on continuous distribution of T_{21} and T_{22} of starch, gluten and starch/gluten mixtures is ongoing to support this hypothesis.

3.2. Effects of moisture

Fig. 3 shows the effect of moisture content on continuous distribution of spin–spin relaxation time, T_{21} of wheat flour dough obtained from One pulse experiment. At moisture content of 33.1–47.2%, two peaks appear on each distribution, which have distinctively different mobility. As mentioned earlier, this T_{21} distribution would represent the proton signals from solids or water molecules closely associated with the solids. It has been reported that two peaks in T_{21} distribution were observed in wheat flour dough at moisture content of 12–23% and only one peak was observed above 23% moisture content (Ruan et al., 1999). On the other hand, two peaks in T_{21} distribution were observed in wheat starch at a moisture content of 0–22% and less mobile fraction shifted from 7.1 to 10.3 μs with increasing moisture content (Choi and Kerr, 2003). They suggested that the greater mobility of starch protons with increased moisture is possibly due to the plasticizing effects of water on starch.

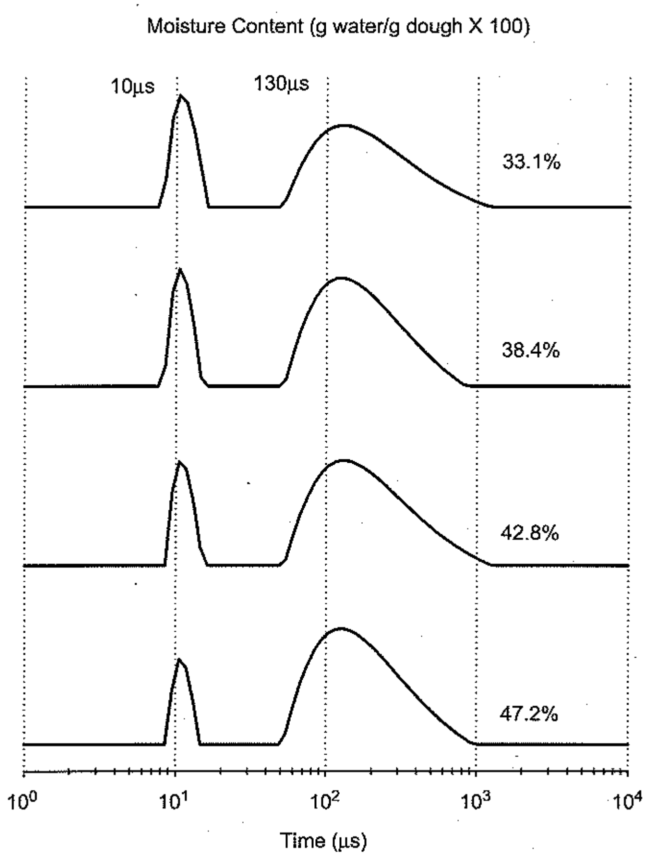

Fig. 3. Effect of moisture content on continuous distribution of spin–spin relaxation time (T_{21}, obtained from one pulse experiment) of wheat flour dough.

Fig. 4 shows the change of peak area of T_{21} with varying moisture content. The less mobile fraction (which is in the 10 μs range) decreased and the more mobile fraction (which is in the 130 μs range) increased with increasing moisture content, respectively. Similar results were observed in wheat starch at a moisture content of 0–22% (Choi and Kerr, 2003). At a certain moisture level, all of the water binding sites of the wheat flour solids would be hydrated. Thereafter, any additional water would be two or three layers away from the solid binding sites. Thus, those water molecules would exchange and relax more slowly than the water molecules closely associated with the wheat flour solids (Ruan et al., 1999). However, this more mobile component is susceptible to decay through field inhomogeneity, and several researchers have noted that it is not a reliable measure of water mobility (Choi and Kerr, 2003; Tang et al., 2000).

Fig. 5 shows the continuous distribution of spin–spin relaxation time, T_{22} (obtained from CPMG experiment) of wheat flour dough with varying moisture content. This T_{22} distribution obtained from CPMG experiment would exhibit relatively longer spin–spin relaxation time than those from the One pulse experiments. As moisture content increases, the more mobile fraction becomes broader and the mean value of the peak increases from 3 to 10 ms; whereas the less mobile fraction (which is in the 0.1 ms range) did not change with varying moisture. The broadening of the more mobile fraction suggests that there is an increase of variation in the chemical and physical states of

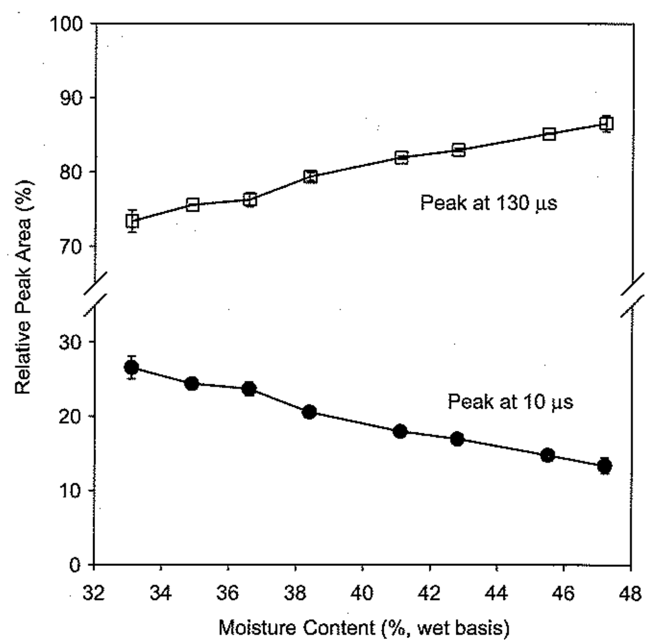

Fig. 4. Changes in peak area of spin–spin relaxation time (T_{21}, obtained from one pulse experiment) for wheat flour dough with varying moisture content.

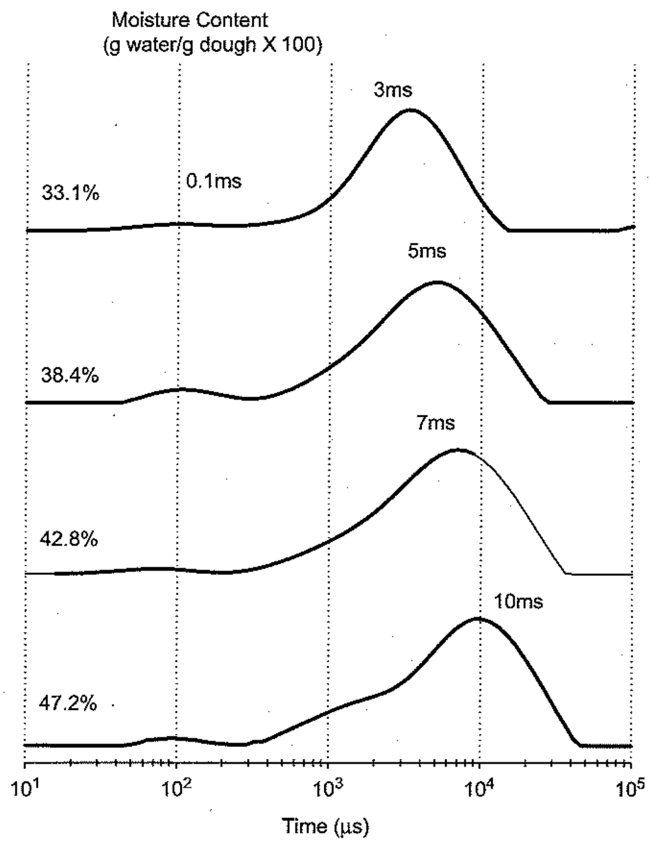

Fig. 5. Effect of moisture content on continuous distribution of spin–spin relaxation time (T_{22}, obtained from CPMG pulse experiment) of wheat flour dough.

water molecules in wheat flour dough. No change in the less mobile fraction (which is in the 0.1 ms range) might indicate that this fraction would represent the water molecules closely associated with solids in wheat flour dough. Therefore, the addition of water greatly influenced the more mobile fraction of wheat flour dough in both peak shape and the range of relaxation time.

4. Conclusion

The continuous distribution of the spin–spin relaxation times (T_{21} from One pulse experiment and T_{22} from CPMG experiment) of starch, gluten and starch/gluten mixture (76:12, dry basis) at 41.1% moisture content and wheat flour dough with varying moisture content were determined using time-domain NMR spectroscopy. Gluten showed a slightly different continuous distribution of T_{21} and T_{22} compared to starch, starch/gluten mixture and wheat flour dough. This suggests that gluten has different water dynamics than starch, and water and molecular mobility in starch/gluten mixture and wheat flour dough are predominated by the interaction of starch and water. The addition of water did not influence the continuous distribution pattern of T_{21} (i.e., two populations for T_{21} without changing relaxation time range) but affected the amount of less mobile and more mobile fractions in T_{21}, which showed a relative increase of the more mobile fraction and a relative decrease of the less mobile fraction with increasing moisture level. The more mobile fraction in the T_{22} distribution showed broadening and shifted to longer relaxation time, but the less mobile fraction did not show any differences associated with the increasing moisture content. This result indicates that the addition of water at the present levels more strongly affects the more mobile fraction than the less mobile fraction. Wheat flour dough is a complex and heterogeneous system and water molecules in dough exist in a number of states and these states correspond to water that is bound to different sites of the flour constituents or in exchange with bound water in various ways. The manipulation of the water dynamics in model food systems provides a basis for the application of the microstructural domain concept to real food systems. The dispersion of various relaxation time constants provided an indicator that discerned the distribution of independent microstructural domains. Future investigation is warranted on microstructural domains in real foods to establish a stronger connection between microstructural domains and food safety, quality and stability.

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