Bulgarian Journal of Agricultural Science, 20 (No 5) 2014, 1221-1226
Agricultural Academy

RHEOLOGICAL PROPERTIES OF GLUTEN OBTAINED FROM POLISH WHEAT CULTIVARS

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Abstract

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The aim of the study was to investigate the rheological properties of gluten obtained from several Polish wheat cultivars harvested in 2010, 2011 and 2012. Gluten was separated and its elasticity and spreadability were evaluated. The following rheological methods were used: uniaxial extension using a texture analyser, ultrasound viscosity measurements and small amplitude oscillatory shear measurements. The protein content varied between 14.24% and 11.74%. There were significant differences in rheological properties of gluten obtained from the same wheat cultivars at different harvest year. Gluten behaved as an elastic material similar to particulate gel with a tendency for decrease in elastic component in comparison to viscous component at higher frequencies, which could suggest breaking of some bonds. There was a linear correlation between ultrasound measured viscosity and maximum extensional force, and between ultrasound measured viscosity and storage modulus. All fundamental rheological methods used for gluten investigation gave results with an agreement with basic gluten elasticity measurements performed according to a Polish Standard method. Samples with better elasticity had higher viscosity, maximum extension force and storage modulus. Fundamental rheological methods can give better knowledge of the viscoelastic behavior of gluten, which can improve dough quality control.

Key words: gluten, protein, rheology, texture, wheat

Introduction

Wheat is currently the most important human food grain in the world. It ranks second after maize, which is the first in total production as a cereal crop. Only in the European Union wheat production in 2010/11 reached 650 million tones (Michalska-Klimczak, 2011). Wheat is an unique grain, because of the special properties of its flour. The unique viscoelastic properties of the wheat flour come primarily from the gluten-forming storage proteins. Gluten proteins are found in the endosperm of the mature wheat grain. They are largely insoluble in water or dilute salt solutions and are composed of monomeric gliadins and polymeric (extractable and unextractable) glutenins (Goesaert et al., 2005). Gluten can be defined as the viscoelastic residue remaining after starch granules and water-soluble constituents are washed out from the dough. It contains 75–85% protein and 5–10% lipids in dry mass (Wieser, 2007). Gluten is responsible for physical properties of dough and its quality is usually characterized by the degree of extensibility and elasticity (Juric et al., 2001). Considering the frequently implementation of oxidative improvers in the bakery industry, wheat cultivars with stronger gluten should be used. Lately there is a trend to use gluten to nonfood applications (cosmetic and hair products, detergents,

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rubber, and polymer products). Obtained new biodegradable, low cost and non-toxic materials can be an alternative for synthetic polymers (Lagrain et al., 2010).

Gluten is a complex system and its viscoelastic behaviour is not yet fully understood. Better knowledge of the viscoelastic properties of gluten is necessary for dough quality control to improve its water absorption capacity, cohesivity, viscosity and elasticity. Many empirical measurements of the rheological properties of gluten were used with such apparata as farinograph, mixograph, extensigraph, alveograph, amylograph or texturometer. Most of the methods were invented for industrial applications, as they are easy to perform in factory conditions. Obtained data are usually used for dough processing and quality control. There is still not many research done using fundamental rheological methods and finding existing correlations between measured rheological properties.

Gluten rheological properties can be affected by some factors such as wheat variety, crop location, temperature, rainfall, soil fertility and harvest year (Stoeva and Ivanova, 2009b; Delibaltova and Kirchev, 2010). Stoeva and Ivanova (2009a) noted, that the genotype differences with regard to sedimentation and wet gluten were markedly dependent on the year, while the variations of pharinographic values and bread loaf depended more on fertilization. They concluded that the agronomy practices affect the availability and uptake of nutrients, and causes increase of yield and suppression of sufficient gluten accumulation in the grain. Atanasova et al. (2010) observed, that the varieties with highest quality accumulated lowest gluten in grain, even in comparison to medium and low-quality wheats. Storage of the grain can deteriorate properties of gluten by free radical generation/oxidation and non-enzymatic glycosylation, which can lead to fragmentation or cross-linking of proteins, lipids and sugars (McDonald, 1999).

The aim of this study was to investigate the rheological properties of gluten obtained from several Polish wheat cultivars harvested in 2010, 2011 and 2012.

Material and Methods

A field experiment in growing spring wheat was carried out in the period 2010-2012 at the Czesławice Experimental Farm, belonging to the University of Life Sciences in Lublin (Poland), using a split-block design with 3 replications, in 27 m^2 plots. The study included spring wheat cultivars Monsun harvested in 2010 (M10), 2011 (M11) and 2012 (M12); Korynta harvested in 2010 (K10), 2011 (K11) and 2012 (K12); Tybalt harvested in 2010 (T10), 2011 (T11) and 2012 (T12) and Zadra harvested in 2010 (Z10), 2011 (Z11) and 2012 (Z12). After harvest, the grain was brought to the same moisture content – 14%. The grain was stored at controlled conditions in a granary in piles, on a neutral substrate at a temperature range of 10-20^{o}C for 3, 15 or 27 months. During storage, the wheat grain was protected from the direct action of light (sunlight, artificial illumination) by means of special roller blinds as well as it was aerated (stirred) from time to time, thus preventing it's overheating. Protein concentration in the grain was determined using a Perten infrared analyzer and grain moisture was measured using the gravimetric method.

Before milling, grain samples were cleaned by a laboratory cleaner-separator Model SZD (ZBPP, Poland). To obtain flour, before milling, 1000 g wheat samples were tempered at 15% moisture at 22^{o}C for 24 h and milled with an adapted laboratory mill (QC109/2, Hungary) using 0.8 mm sieve. Moisture content of the flour was determined by oven drying at 130°C for 1 h by the AACC method 44-15A (AACC, 2000).

Gluten separation, elasticity and spreadability

To obtain gluten approximately 50 g of the flour was transferred into a mortar, approximately 25cm ^{3} of water at 15-20°C was added and the mixture was kneaded until it did not adhere to the walls. The ball of dough was formed and immersed in warm water for about 10 min. and kneaded with fingers to separate the starch. Spreadability and elasticity were determined by a Polish Norm (PN-77/A-74041., 1977). Spreadability was measured as the increase in diameter of the 5 g gluten ball placed in an oven at 30°C for 60 min. Elasticity was expressed as a permanent elongation in mm after return of 20 mm cylinder made of 5 g of gluten and extended to 50 mm. Elasticity was also expressed by 4-point scale: (I^{o} – permanent elongation 0-10 mm, II^{o} – permanent elongation 11-25 mm, III^{o} – permanent elongation 26-50 mm, IV^{o} – breaking before 50 mm extension.

Uniaxial extension

Gluten cylinders were fixed between two clamps 30 mm apart using the TA-XT2i Texture Analyzer (Stable Micro Systems, Godalming, UK). Before extension test, the gluten cylinder dimensions were 50 x 13 mm. The cylinders were extended for 50 mm with 1 mm/s speed. Six extensions were performed for each gluten sample.

Ultrasound viscosity

Viscosity x density in mPas x g/cm^3 of the gluten samples was measured using Unipan type 505 ultrasound viscometer (UNIPAN, Warsaw, Poland). Before each measurement, ultrasound signal level was checked. Measuring probe of the magnetostrictive vibrator was placed entirely in the gluten sample. Induced ultrasound waves were damped by tested material and the results were displayed as the product of viscosity and density. All measurements were performed in six replications.

Small amplitude oscillatory shear measurements

Dynamic oscillatory measurements were performed using RS300 (ThermoHaake, Karlsruhe, Germany) rheometer with a serrated parallel steel plate geometry (35 mm diameter) to limit the potentiality of sliding effects. Gluten samples (35 mm diameter and 3 mm thick) were analyzed by strain and frequency sweep. The temperature for all measurements was 22°C. For strain sweeps a frequency of 0.1 Hz was used. For frequency sweep 5% strain was used, which was in linear viscoelastic region.

Statistical analysis of results (standard deviation and analysis of variance) was performed using the statistical program Statistica 5.0 PL (StatSoft Polska, Warsaw, Poland). The significance of differences between means was determined using the Tukey's test at confidence level of $p \leq 0.05$ based on the least significant difference.

Results and Discussion

Figure 1 shows water content in grain of different cultivars harvested in different years and stored. Besides two cultivars harvested in 2012, there were no statistically significant differences in grain water content. It shows that the grain was stored in proper conditions, which did not trigger such negative phenomenon as e.g. overheating. There were no statistically significant differences in grain total protein content (Figure 2). Only in a case of a cultivar Korynta higher protein content in 2012 year grain was noted. This cultivar showed the highest protein content compared to other cultivars included in the study. The protein content varied between 14.24% and 11.74%. Strelec et al. (2010) found, that there were no significant changes in total protein content in wheat seeds during one year storage under different storage conditions, however changes in protein chemical properties were observed. Table 1 presents basic gluten characteristics (spreadability and elasticity) measured according to Polish Standard methods (PN-77/A-74041., 1977). Spreadability up to 10 mm meets gluten quality requirements and in most cases such a value was measured. Generally it was observed, that gluten with lower spreadability was characterized by higher elasticity (I°). Gluten with higher permanent elongation after extension spread more easily staying in an oven at 30°C for 60 min.

Gluten samples were subjected to uniaxial tensile test, which is one of the oldest and most widely used test methods to measure wheat dough. Tensile tests usually produce an ap-

Table 1
Flour moisture content, gluten spreadability and elasticity

Sample	Flour moisture content, %	Gluten spreadability, mm	Gluten elasticity (°), mm
K10	9.6	11.4	II (10 mm)
K11	8.7	10.2	I (6 mm)
K12	10.5	8.9	I (6 mm)
M10	11.0	9.5	I (5 mm)
M11	11.6	7.0	I (7 mm)
M12	11.6	7.3	I (5 mm)
T10	10.1	8.2	I (5 mm)
T11	10.7	7.8	I (5 mm)
T12	10.8	15.1	II (10 mm)
Z10	11.5	12.0	II (11 mm)
Z11	11.1	9.2	I (6 mm)
Z12	10.6	13.5	II (12 mm)

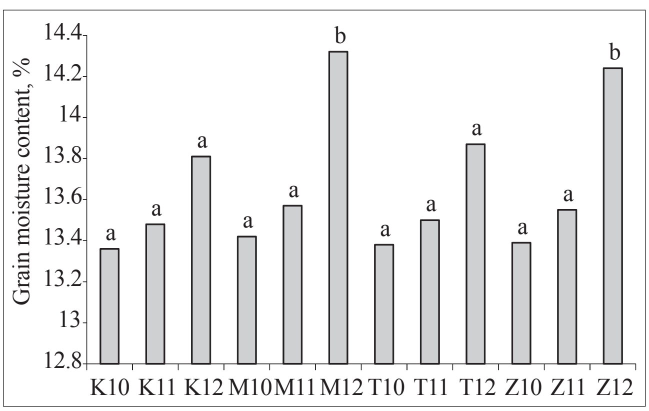

Fig. 1. Grain moisture content

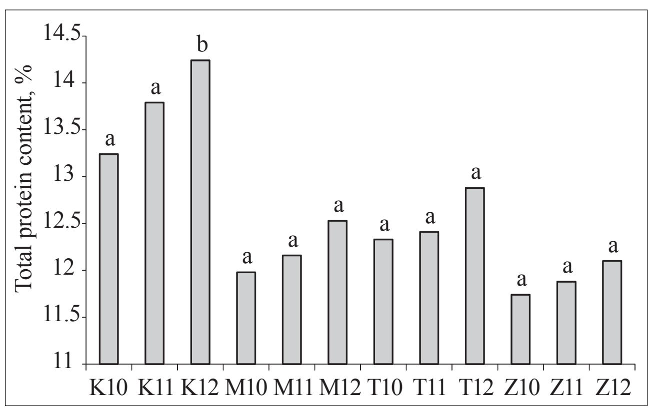

Fig. 2. Grain total protein content

proximately uniform extension of a sample providing necking does not occur (Dobraszczyk and Morgenstern, 2003). Figure 3 shows extension curves obtained for wheat cultivar Tybalt. In all cases a strain hardening was observed. For the sample T10 a peak was noted at the breaking point with a small strain hardening just before sample fracture. Usually strain hardening was observed after 30 s of extension. Studies have shown that the strain hardening properties exhibited during elongational measurements of different flours relate to the baking performance of those flours and are a requirement of gas retension (van Vliet et al., 1992). Uthayakumaran et al. (2002) showed that gluten dough possessed much larger elongational viscosities than flour. Furthermore, pure gluten doughs also possessed greater elongational viscosity than glutenstarch doughs, whose elongational viscosity decreased with increasing proportion of starch. Dobraszczyk and Morgenstern (2003) observed that the stress-strain curves showed considerable strain hardening, and strain and stress at rupture was considerably less for poor quality flours than good quality flours. Nicolas et al. (2003) suggested that the increase in stress at fracture at extension reflects that more entanglements were formed between gluten strands. Maximum extension force differed for different cultivars and harvest years (Table 2). For cultivar Zadra no significant changes in maximum extension force was found. Maximum extension force is associated with extensive hydrogen bonding both within and between high molecular weight protein subunits in addition to entropic and energetic contributions (Tatham et al., 2008). Increased strain hardening was attributed to entanglement of long-chain molecules during extensional flow, whereas in simple shear they remain coiled and can slip past each other, giving rise to observed shear thinning at higher strains (Cogswell, 1981). Bonding between gluten protein molecules influences its viscoelastic properties. Gluten is treated as a good material for fundamental rheological characterization, as the linear region of its viscoelastic behavior is extended up to 10%, whereas the linearity limit of dough is extremely low (0.2%) (Lefebvre et al., 2003). In our research linear viscoelastic domain was between 5.76% and 8.97% (data not shown). Figure 4 shows frequency sweeps for gluten samples obtained from cultivar Tybalt. Gluten behaved as an elastic material with storage moduli values 2-4 times higher than loss moduli. Gluten should be viewed as a kind of colloidal or particulate gel, a network formed by the aggregation of particles stuck together by hydrogen bonds and hydrophobic interactions (Lefebvre et al., 2003). There was an increase in storage and loss moduli with frequency, but

Table 2
Rheological properties of gluten

Sample	Viscosity x density, mPas x g/cm^3	Maximum extension force, N	Storage modulus at 10 Hz, Pa
K10	147 (12) a*	0.20 (0.01) a	1810 (23) a
K11	273 (17) b	0.25 (0.01) b	2421 (44) b
K12	321 (24) b	0.27 (0.02) b	2403 (11) b
M10	159 (12) a	0.19 (0.03) a	1942 (54) a
M11	451 (21) c	0.32 (0.07) c	2850 (122) c
M12	273 (15) b	0.21 (0.01) a	2276 (56) b
T10	426 (23) c	0.30 (0.02) c	2722 (131) c
T11	385 (11) c	0.26 (0.01) b	2435 (121) b
T12	280 (29) b	0.20 (0.01) a	1987 (70) a
Z10	182 (9) a	0.17 (0.01) a	1886 (18) a
Z11	280 (13) b	0.20 (0.02) a	2305 (54) b
Z12	175 (13) a	0.17 (0.01) a	1882 (39) a

*Means within a column followed by various letter (a-c) are significantly different at (P = 0.05)

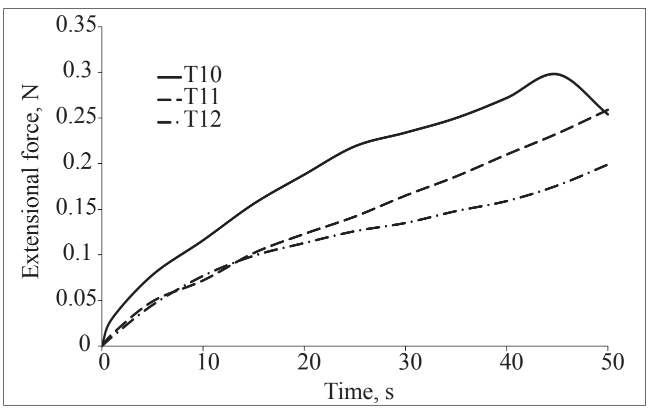

Fig. 3. Extensional force curves for Tybalt cultivar gluten

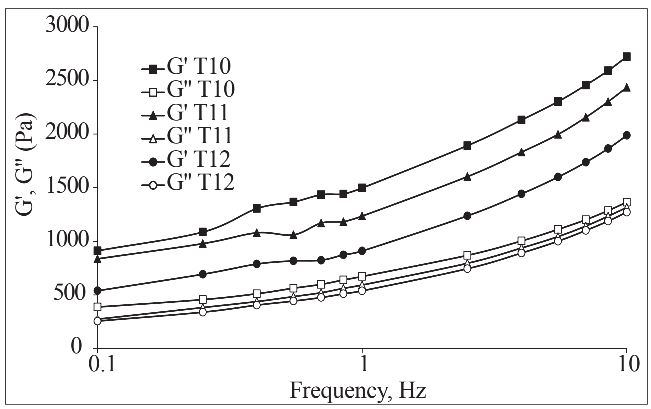

Fig. 4. Oscillatory frequency sweeps of gluten obtained from Tybalt cultivar

tangent delta increased (Figure 5). Higher frequency allowed more energy into the sample and probably some bonds between protein molecules broke, as a decrease in elastic component in comparison to viscous component was observed. It is in agreement with strain weakening observed at initial stage of gluten extension (Figure 3). Santos et al. (2005) also noted a decrease in the loss tangent values at higher frequencies. There were differences in storage modulus measured at 10 Hz for different cultivars and harvest years (storage time). Modenes et al. (2009) obtained results indicating that during 5 months storage period of wheat grain the gluten force and falling number did not change. Edwards et al. (2003) noticed that the rheological properties of wheat dough were influenced greatly not only by cultivar genotype but also by the environmental conditions faced by the plants during grain ripening. Gluten isolated from stronger bread wheat cultivars has exhibited higher storage modulus and lower loss tangent than gluten isolated from weak wheat cultivars. In our research wheat was harvested at different years and for the same cultivar other factors than storage time could affect the rheological properties of gluten.

Gluten viscosity was measured using an ultrasound device. Measuring magnetostrictive vibrator was placed in the gluten sample and ultrasound waves were damped. The results were displayed as the product of viscosity and density and are presented in Table 2. This method was not previously used for measurements of the rheological properties of gluten. Product of dynamic viscosity and density measured using ultrasounds was correlated with other rheological measurements: maximum extensional force and storage modulus G' measured at 10 Hz. Figure 6 presents linear correlation between ultrasound measured viscosity and maximum extensional force with R^2 = 0.82. Gluten samples with higher product of viscosity and density reacted with greater resistance to tensile forces, producing higher values of extensional force. Similarly gluten characterized by higher values of the product of viscosity and density were more elastic as a strong linear correlation (R^2 = 0.89) was noted between viscosity and storage modulus (Figure 7). Both methods are non-destructive. In oscillatory measurements applied stress does not affect the rheological characteristics of the test material, as the measurements are performed in the linear viscoelastic region. Assessing viscoelastic properties of wheat flour dough and gluten can be very significant because they affect the dough handling behavior during processing and influence the interactions among dough components (Upadhyay et al., 2012). All fundamental rheological methods used for gluten investigation gave results with an agreement with basic gluten elasticity measurements performed according to a Polish Standard method (Table 1). Samples with better elasticity had higher viscosity, maximum extension force and storage modulus.

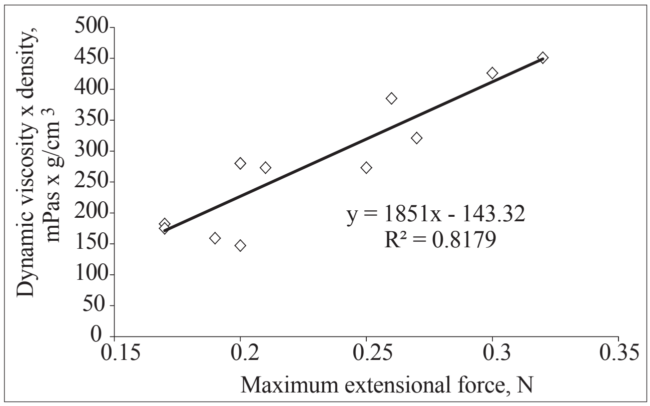

Fig. 6. Correlation between maximum extensional force and product of dynamic viscosity and density

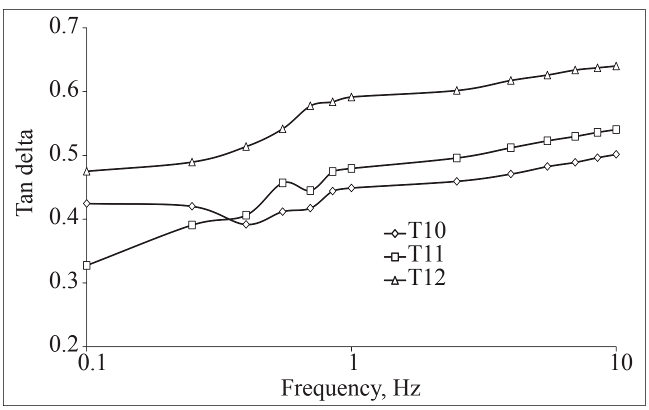

Fig. 5. Changes of tangent delta with oscillatory frequency for gluten obtained from Tybalt cultivar

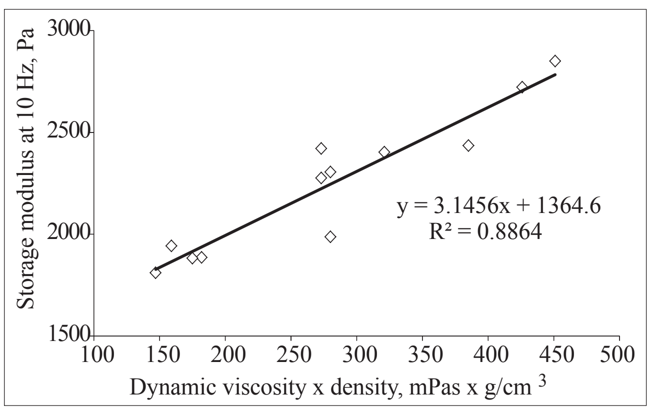

Fig. 7. Correlation between product of dynamic viscosity and density and storage modulus measured at 10 Hz

Conclusion

There were significant differences in rheological properties of gluten obtained from the same wheat cultivars at different harvest year. Probably environmental conditions faced by the plants during grain ripening influenced the differences. Gluten behaved as an elastic material similar to particulate gel with a tendency for decrease in elastic component in comparison to viscous component at higher frequencies, which could suggest breaking of some bonds. Product of dynamic viscosity and density measured using ultrasounds was correlated with other rheological measurements: maximum extensional force and storage modulus There was a linear correlation between ultrasound measured viscosity and maximum extensional force and between ultrasound measured viscosity and storage modulus. All fundamental rheological methods used for gluten investigation gave results with an agreement with basic gluten elasticity measurements performed according to a Polish Standard method. Fundamental rheological methods can give better knowledge of the viscoelastic behavior of gluten, which can improve dough quality control.

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Received February, 2, 2014; accepted for printing September, 10, 2014.