Evaluating a New Rapid Technique to Assess Spring Wheat Flour Performance

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Research Questions

Variability in flour performance and consistency under manufacturing conditions remains an issue for processors. The reasons for variability are many, including genetics, environmental conditions, aging of wheat during the year, blending of wheats and the particular milling fractions that are blended to deliver flour of a certain specification. However, there is evidence that some wheat varieties maintain their quality despite variability in growing season. Wheat is transacted in the market based on protein content, but flour performance is based on protein quality – an attribute that remains difficult to measure or quantify. Recent research from our laboratory working with Eastern hard and soft wheats, demonstrated that a new technique – Glutopeak tester –can distinguish differences between flours with similar protein quantity. However, similar work has not been conducted with western wheat varieties. These observations led us to the next phase of research where we focus on individual wheat varieties. This project proposes to assess if this new technique has the potential predictive capabilities of assessing flour performance. The glutopeak tester is a high shear based technique that provides two attributes of gluten quality; torque that is an indication of strength of gluten and time to peak that is an indication of kinetics of gluten aggregation. The test is rapid (<5 min) and it requires only about 9 g of sample. If the technique has predictive capabilities, then it will be of great value to wheat breeders, elevators, millers and processors to asses wheat quality as opposed to merely protein quantity.

Results

We compared performance of flours from 32 spring wheat varieties grown in different environments during 2012 and 2013 using industry standard procedures to measure water absorption, mixing strength, loaf volume, etc. We subjected the same flours to the glutopeak test, a fast 5-10 min. test that could be used at grain elevators, to test its ability to predict mixing and baking properties. During the test, the sample is mixed with water (ratio of flour : water about 1:1) and subjected to intense mechanical action on the speed of the rotating element (from 1900 to 3000 rpm). These conditions - allowing for the formation of gluten - promote a strong increase in the consistency of the slurry up to a maximum peak. From that moment, the continuous mechanical stress causes the breakdown of the gluten network, a phenomenon recorded as a decrease in consistency (see Figure 1). Using this technique we were able to demonstrate that flours with similar protein content had different gluten aggregation kinetics and conversely wheat flours with different protein content had similar aggregation kinetics in terms of Peak Maximum Time and Peak Torque (Figure 1a). During gluten aggregation, some samples showed different peak torque and peak time but they exhibited the similar values for the area under the peak indicating that they required a similar energy for gluten aggregation and a similar volume of the corresponding bread (samples A and B in Figure 1b). Other samples were characterized by a different aggregation kinetics, but they were different in the energy necessary for gluten aggregation and therefore in loaf volume (samples C and D in Figure 1b). Further studies are underway to explain what is the driving force of these differences in gluten aggregation. Considering the data set analyzed (n=128), samples with high peak torque exhibited faster aggregation (low peak maximum time; $r=-0.62$; $p<0.01$; see Table 1) and they were able to keep high value of consistency even after prolonged mixing (2 min after the peak; $r = 0.90$; $p<0.01$; see Table 1).

The charts that report the min, max, average and median values of protein content, peak maximum time, peak torque, area under the curve of the flours of 128 spring wheat samples (see Figure 2) highlight that the set of samples is well distributed around the mean values (the mean and median values are very similar). Samples grown in Stephen, MN - characterized by relatively high protein content - show a faster aggregation (low peak maximum time) and strong gluten formation (high peak torque) on average. Considering the area under the curve - that takes into consideration both the peak torque and time for gluten aggregation - samples grown in Roseau, MN exhibited the lowest value, suggesting weaker gluten aggregation performances compared to the samples grown in Oklee and Stephen. As regard the effect of growing year, wheat grown in Stephen 2012 showed - on average - stronger protein network forming than samples grown in Stephen in 2013.

Taking into consideration all the samples (n=128), the peak maximum time from the Glutopeak test was correlated with the bake mixing time ($r=0.62$; $p<0.01$ see Table 2) and the farinographic stability ($r=0.48$; $p<0.01$ see Table 2). Interesting, samples that required higher energy for gluten aggregation exhibited higher mixograph pattern ($r=0.40$; $p<0.01$ see Table 2) and farinographic stability ($r=0.39$; $p<0.01$ see Table 2).

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Based on the results obtained using the GlutoPeak, 4 varieties (Knudson, Marshall, Prosper and Samson) were chosen for further investigation, because of their high variability (e.g. >20% for the area under the peak). Unlikely, the SDS-solubility data of the dough did not show any strong correlation with the indices from the GlutoPeak test. Samples with faster aggregation exhibited high solubility at the dough development time ($r=-0.48$; $p<0.1$). A larger set of samples should be investigated.

Interesting, a significant negative correlation between the maximum torque (that corresponds to the peak occurring as gluten aggregates) and the fluorescence intensity ($r=-0.71$; $p<0.01$) was observed, suggesting that flours with greater number of hydrophobic patches on the protein surface (great fluorescence intensity) create a weak gluten network during aggregation.

Application/Use

Results demonstrated that gluten aggregation kinetics of varieties grown in different locations and over different years were different. the new technique can be successfully used for characterizing spring wheat lines.

Material and Methods

Thirty-two varieties of spring wheat were grown in three locations (Roseau, Stephen and Oklee) in 2012-2013, for a total of 128 samples. On the basis of the results, 4 varieties (Knudson, Marshall, Prosper, Samson) were chosen for investigating the protein solubility and the hydrophobic interactions during dough-making.

The gluten aggregation properties of flours were measured using the GlutoPeak (Brabender GmbH and Co KG, Duisburg, Germany), according to (Kaur Chandi & Seetharaman, 2012). An aliquot of 8.5 g of flour was dispersed in 9.5 ml of 0.5M $CaCl_2$. Sample and solvent temperature was maintained at 34 °C by circulating water through the jacketed sample cup. The paddle was set to rotate at 1900 rpm and each test ran for 7 min. The following indices were determined: *i)* Maximum Torque expressed in Brabender Equivalents (BE) - corresponding to the peak occurring as gluten aggregates; *ii)* Peak Maximum Time expressed in minutes - corresponding to the time at peak torque; *iii)* Area under the peak expressed in arbitrary units (AU) - corresponding to the energy required for gluten aggregation; iv) loss of torque 2 minutes after the peak (%) related to the ability of strength of the gluten network during prolonged mixing.

A Farinograph-AT (C.W. Brabender Inc., South Hackensack, NJ, USA) equipped with a 10 g mixing bowl was used for preparing the dough (at 30°C and 63 rpm). All the dough samples were prepared at a constant water absorption level (60%). Samples were collected at the dough development time and at the end of the test (20 min mixing) and immediately transferred to liquid nitrogen followed by freeze-drying. The freeze-dried samples were ground using a pestle and mortar in order to have a powder sample with particle size less than 0.5 mm. Freeze-dried samples containing ~1 mg protein were suspended in 1 mL of a 0.05 M sodium phosphate buffer (pH 6.8) containing 2.0% sodium dodecyl sulfate (SDS) and transferred to a shaker for 60 min at room temperature. After centrifugation (10,000×g for 5 min), the amount of protein in the supernatant was determined colorimetrically using the RC-DC Protein Assay (Bio-Rad, Hercules, CA, USA) based on the Lowry assay (Lowry *et al*., 1951). Bovine serum albumin was used as a standard and results were expressed as mg soluble protein/g protein.

Rearrangement of hydrophobic patches on the protein surface of selected samples were measured by spectrofluorometric technique, monitoring the chemical environment of extrinsic flourophore1,8-anisolino naphthalene sulfonate (ANS), as described by Bonomi et al. (2004). A Farinograph-AT (C.W. Brabender Inc., South Hackensack, NJ, USA) equipped with a 10 g mixing bowl was used for preparing the dough (at 30°C and 63 rpm). All the dough samples were prepared at a constant water absorption level (60%). ANS (0.2 mM) was added to the water used for making the dough. Dough samples for analyses were collected at the dough development time, corresponding to the time from first addition of water to the point of maximum consistency range. Fresh samples were pulled from the farinograph with minimal additional physical manipulation, transferred to a fluorescence cell (quartz-windowed standard surface) and analyzed within 3 min. A fluorophotometer (LS 55, Perkin Elmer, Waltham, MA, USA) was used to measure emission spectra from 400-600 nm with excitation at 390 nm.

References

Kaur Chandi, G., Seetharaman, K. (2012) Optimization of gluten peak tester: a statistical approach. Journal of Food Quality 35, 69-75.

Lowry, O.H., Rosebrough, N., J., Farr, A., Randall, R.J. 1951. Protein measurement with the folin phenol reagent. Journal of Biological Chemistry, 193, 265-275.

Related Research

Using this technique, Dr. Seetharaman was able to demonstrate that flours with similar protein content had different gluten aggregation kinetics and conversely wheat flours with different protein content had similar aggregation kinetics. Given that commercial flours are a blend of different wheat varieties and/or types, he conducted a flour blending study to assess the impact on gluten aggregation kinetics. Results suggested that different proportions of

wheat types in a given blend influence gluten aggregation kinetics. A further exploration to understand reasons for these observations was conducted by isolating gliadin and glutenin from different wheat types (eastern soft, eastern hard, and western hard). The results showed that gliadin and glutenin proteins in the different wheat types were inherently different and blending of gliadin and glutenin from different wheat types did not result in similar gluten aggregation kinetics.

The project is a collaboration with Dr. Jim Anderson, Wheat Breeder, University of Minnesota and used wheat varieties developed by other scientists in the region.

Recommended Future Research

- Relating gluten aggregation properties and hydrophobic interactions testing a larger number of varieties, years and locations
- Applying proteomics and thiolomics approach to understand variability in flour performance from wheats grown in different environmental conditions.

Appendix

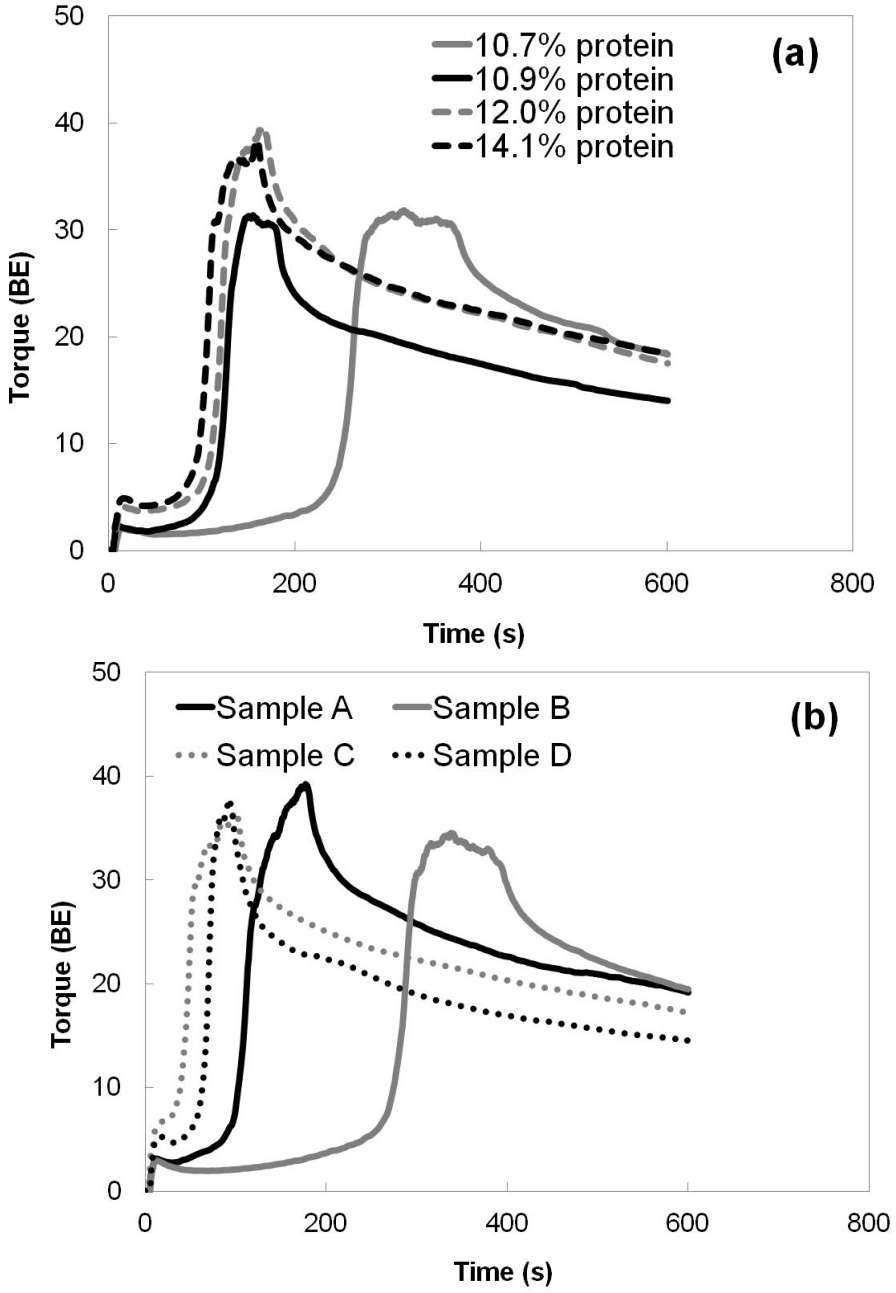

Figure 1. Examples of GlutoPeak curves

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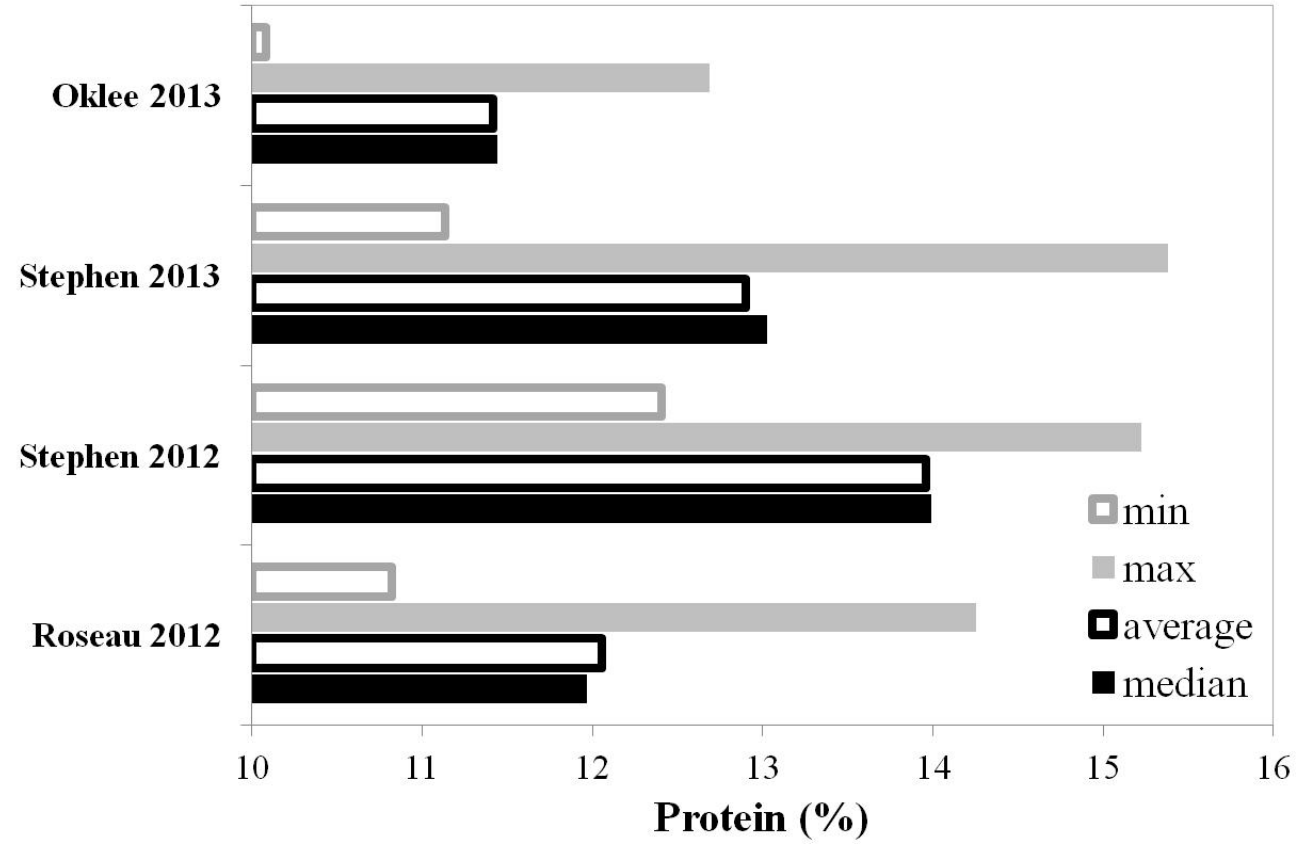

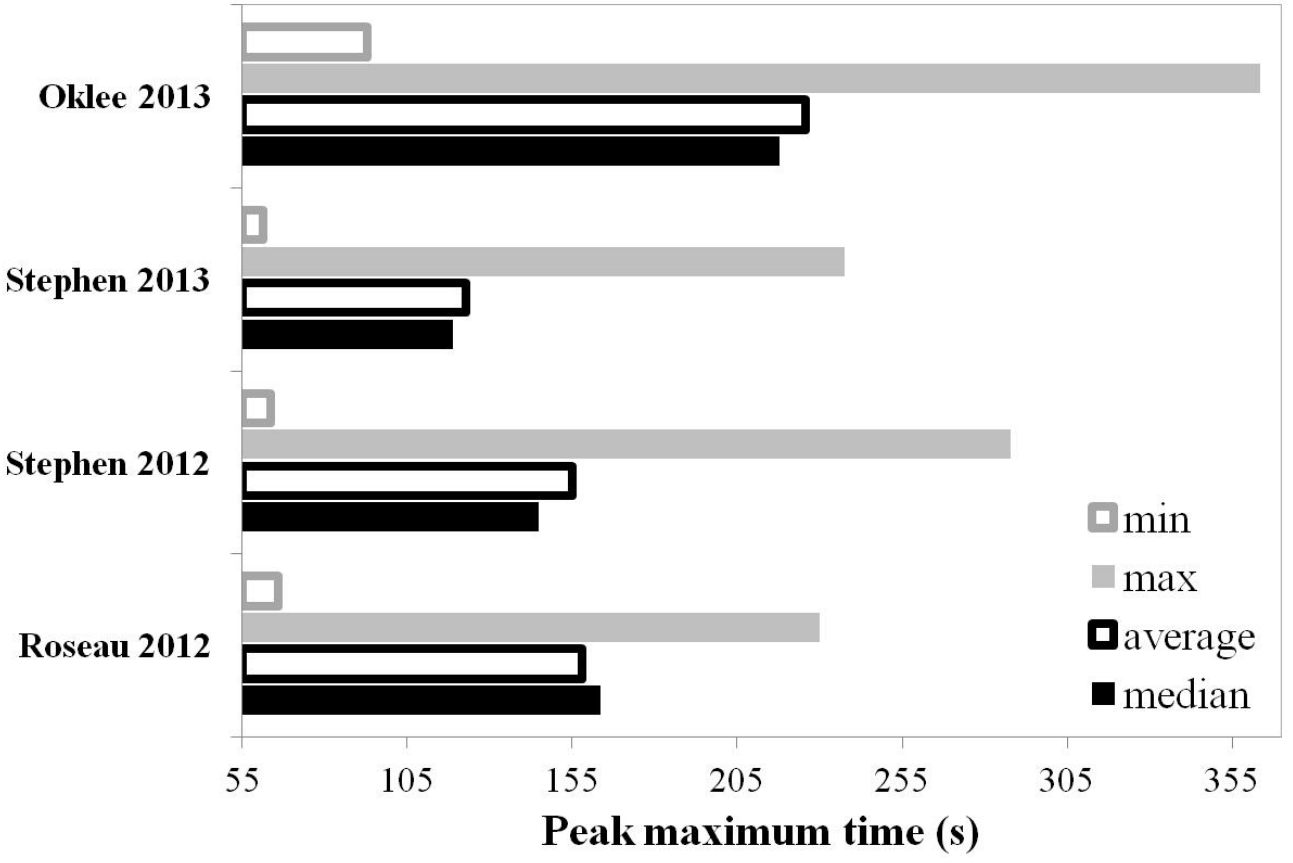

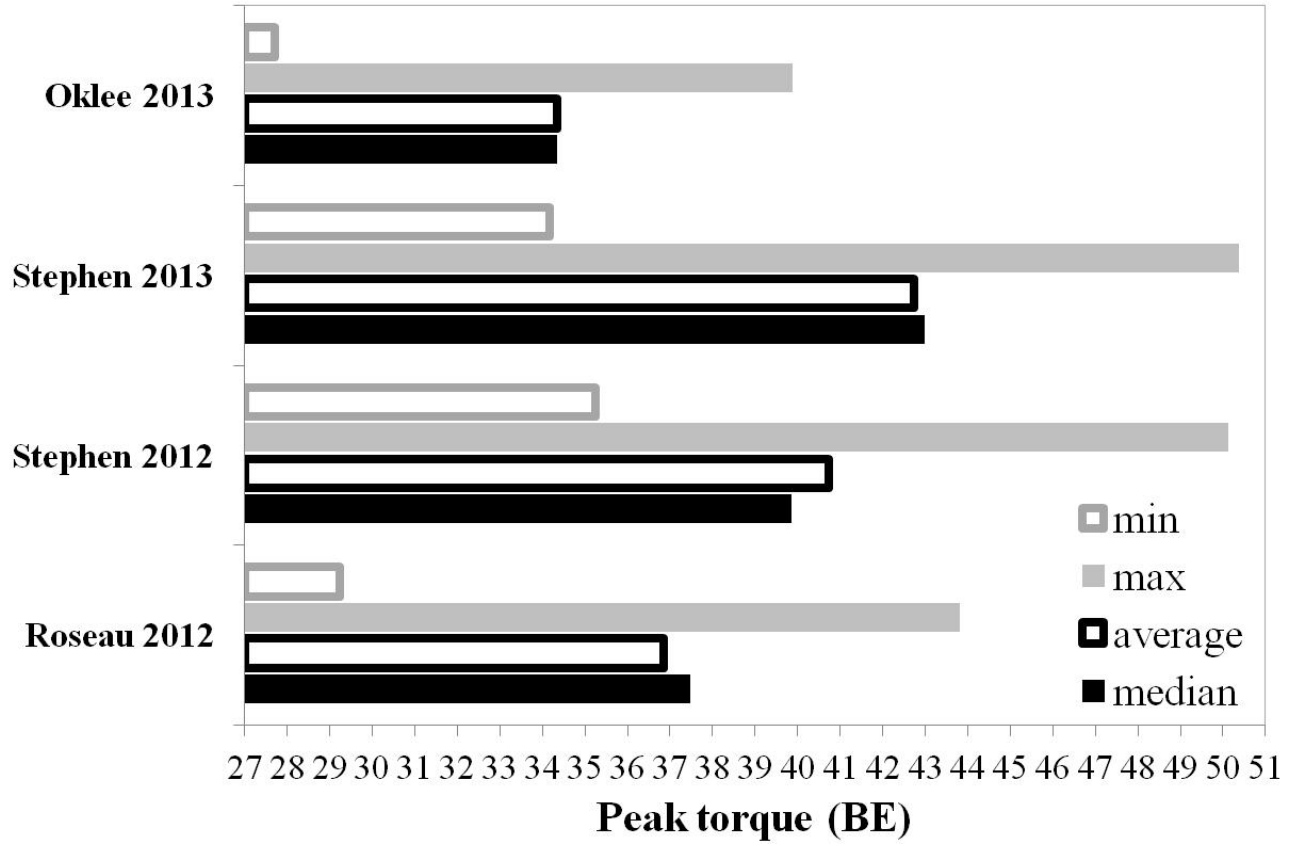

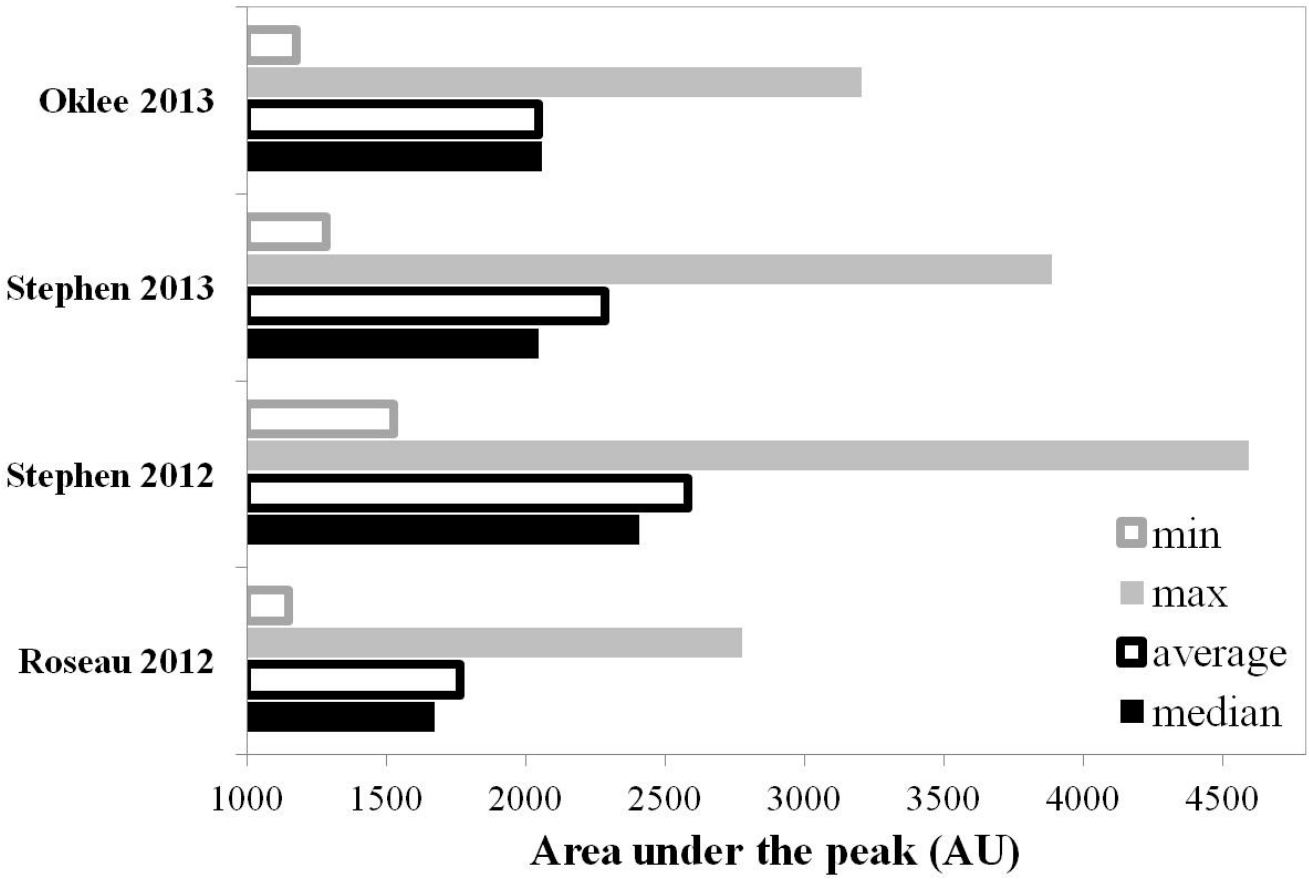

Figure 2. Min, max, average and median values for protein and GlutoPeak of 32 spring wheat varieties grown in different environments.

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Table 1. Glutopeak indices: Pearson coefficients

	Peak Maximum Time	*Peak Torque*	*Energy peak*	*Torque 2 min after peak*
Peak Maximum Time	1			
Peak Torque	-0.62	1		
Energy peak	0.28	0.37	1	
Torque 2 min after peak	-0.46	0.90	0.45	1
n.s. not significant				

Table 2. Mixing profile: Pearson coefficients

	Peak Maximum Time	*Peak Torque*	*Energy peak*	*Torque 2 min after peak*
Mixograph Pattern	0.28	n.s.	0.40	0.37
Bake mixing time	0.62	-0.34	n.s.	n.s.
Farinographic stability	0.48	n.s.	0.39	n.s.
n.s. not significant				