

## Mixolab Versus Alveograph and Falling Number

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### Abstract

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Recently, in 2005, a new method for monitoring the rheological properties of the dough on the entire technological process of bread making became available through Mixolab at an international level. This laboratory equipment has amazing possibilities for the research and development, enabling a complex analysis of flour. It allows the analysis of flour proteins quality (water absorption, stability, elasticity, weakening), the analysis of starch behaviour (gelatinisation, gelatinisation temperature, the modification of its consistency on additives addition) and the analysis of enzymatic activities (proteolytic, amylolytic). The objective of this study is to establish a relation between the alveograph, Falling Number, and Mixolab values. Sixty flours, collected around the Romanian country, were analysed simultaneously on alveograph (standard protocol), for the Falling Number, and on Mixolab ("Simulator" and the standard option "Chopin+" protocol). A selection of principal factors based on the Principal Component Analysis (PCA) was applied which allowed the building of an efficient predictive model for each parameter. There were significant correlations between most of the Alveograph parameters: maximum pressure (P), deformation energy (W), extensibility (L), alveograph ratio (P/L) and Simulator Mixolab stability. Using the Mixolab standard option "Chopin+" protocol a close association was found between some Mixolab parameters: stability and protein weakening (C2, difference of the points C1–C2 abbreviated C12) and the alveograph values (P, W). From the point of view of the correlations established with the Falling Number index, very good results were obtained with the parameters obtained with Mixolab that measures starch gelatinisation (C3, difference of the points C3–C2 abbreviated C32), amylolytic activity (C4, difference of the points C3–C4 abbreviated C34), and starch gelling (C5, difference of the points C5–C4 abbreviated C54).

**Keywords:** Principal Component Analysis; Mixolab; Alveograph; Falling Number; protein content; wet gluten

In Romania, wheat represents one of the most important crops in agriculture, covering 38.5% of the whole cereal surface in 2007. Winter wheat is especially cultivated (about 99%), coming from different species of *Triticum vulgare*. Romanian types of flour have a very good protein content which exceeds 13%, and have gluten deformation indices which are situated within 5 ÷ 15 mm, showing a type of flour that is good for bread-making.

The technical information necessary for the evaluation of the flour quality is obtained on the basis of

some indices determined through the organoleptical, chemical, rheological and technological analyses. The number of the testing methods for defining the characteristics of wheat flours is rising continuously, as a result of the striking necessity to anticipate their technological behaviour (POPA *et al.* 2009).

Rheological characteristics, such as elasticity, viscosity, and extensibility, are important for the milling and baking industry in view of the prediction of the dough processing parameters and the end products quality (JIRSA *et al.* 2007). These

rheological characteristics change during the bread making process and are difficult to measure in definitive terms (HRUŠKOVÁ & ŠMEJDA 2003).

The investigations of flour and dough characteristics have been conducted using traditional instruments as Farinograph (SHUEY 1972), Alveograph (FARIDI & RASPER 1987) and Extensigraph (RASPER & PRESTON 1991), which provide practical information for the baking industry. The Mixolab technique developed by Chopin Technologies Company, launched on the market at the AACC annual meeting in 2005, performs a complex analysis of flour. It allows recording the mechanical changes due to mixing and heating simulating the mechanical work as well as the thermal conditions that might be expected during the baking process (ROSELL *et al.* 2006 cited by HAROS *et al.* 2006). This function makes Mixolab unique, by allowing the user to obtain through a single test the information on the water absorption capacity and kneading stability, as well as the gelatinisation temperature, amyolytic activity, or starch gelling.

A typical Mixolab curve is shown in Figure 1. From the technological point of view, the curve has five different stages. Stage one (dough development) corresponds to the farinographic curve which determines the development time, stability, dough weakening, and water absorption of the flour. Stage two (protein weakening) corresponds to the first stage of dough warming. Therefore, the extent to which the dough consistency decreases is an indication of protein quality and strength. Stage three (starch gelatinisation) corresponds to the second stage of dough warming when its temperature exceeds 50–55°C. During this stage, starch granules swell and absorb water and amylose molecules leach out which results in an increase in

viscosity. At the fourth stage, the dough consistency will decrease in relation with the increase of  $\alpha$ -amylolytic activity. At the fifth stage, starch gelling is achieved during the cooling of the dough which triggers the starch gelatinisation and its retrogradation, a phenomenon that leads to an increase of its consistency. It allows measuring the effects of those additives which delay the starch retrogradation and bread staling, respectively.

Since it is a new instrument the information related to its utilisation in view of different aspects of wheat flour quality is quite limited (KAHRAMAN *et al.* 2008; OZTURK *et al.* 2008). The Mixolab is a Standard method ICC No. 173 and the company that produces Chopin is waiting for new information in order to determine the quality parameters that this device establishes.

For this purpose, a range of Romanian white wheat samples were analysed using the Falling Number and Alveograph dough rheology equipment, and the data were compared to those derived from the Mixolab.

## MATERIALS AND METHODS

Sixty samples of commercial wheat flour (harvest 2007) whose ash contents varied between 0.63–0.67 with an average value of 0.65 were obtained from different Romanian milling companies. Deionised water was used in all experiments. For each sample, the following specific qualities were determined: wet gluten content (ICC Standard No. 155), protein content (ICC Standard No. 202), ash content (AACC Standard No. 08-21), and the falling number (ICC Standard No. 107/1).

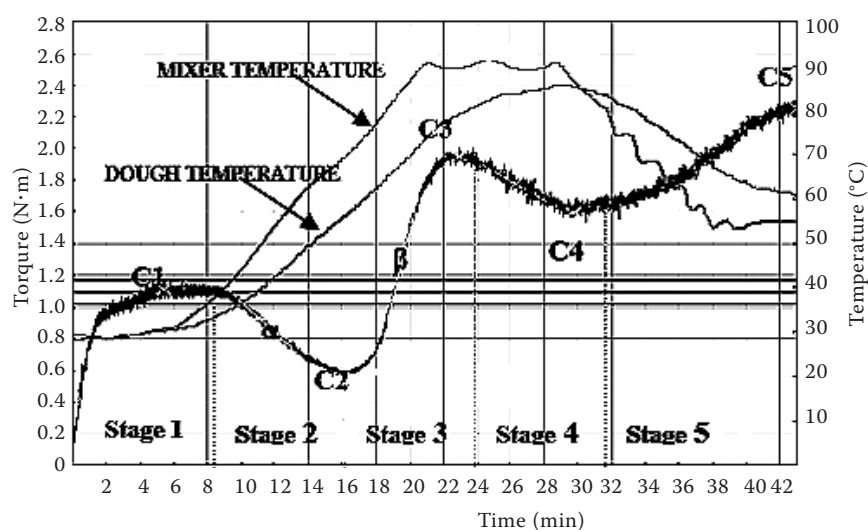


Figure 1. A typical Mixolab curve for dough (HAROS *et al.* 2006)

The rheological properties of the flours were determined by a Chopin Alveograph (according AACC Standard No. 54-30A) and Mixolab (CHOPIN, TRIPETTE and RENAUD). Each alveograph chart was analysed for five factors: P – the maximum over pressure needed to blow the dough bubble – expresses dough elasticity, L – the average abscissa at the bubble rupture – expresses dough elasticity, P/L – alveograph ratio, W – the deformation energy.

The rheological behaviour of dough was analysed in the first stage using the standard option “Chopin+” protocol (thirty flour samples – first batch) on the Mixolab.

The procedure employed for the analysis of the mixing and pasting behaviour of the Mixolab is the following: mixing speed  $80 \text{ min}^{-1}$ , tank temperature  $30^\circ\text{C}$ , heating rate  $2^\circ\text{C}/\text{min}$ , total analysis time 45 minutes. The parameters obtained from the recorded curves involve: water absorption (%) or percentage of water required for the dough to produce a torque (C1) of  $1.1 \text{ N}\cdot\text{m}$ , mixing stability (min) or elapsed time at which the torque produced is kept at  $1.1 \text{ N}\cdot\text{m}$ , protein weakening (C2,  $\text{N}\cdot\text{m}$  and difference of the points C12,  $\text{N}\cdot\text{m}$ ), starch gelatinisation (C3,  $\text{N}\cdot\text{m}$  and difference of the points C32,  $\text{N}\cdot\text{m}$ ), amylolytic activity (C4,  $\text{N}\cdot\text{m}$  and difference of the points C34,  $\text{N}\cdot\text{m}$ ), starch gelling (C5,  $\text{N}\cdot\text{m}$  and the difference between the points C54,  $\text{N}\cdot\text{m}$ ).

In the second stage, for another thirty flour batch samples, the option “Simulator” protocol of the Mixolab was used. The Simulator is a device that enables farinographic measurements to be taken directly with the Mixolab.

This simplified protocol implies not using the heating section of the Mixolab and is equivalent to the following settings: kneading speed  $80 \text{ min}^{-1}$ , target torque  $1.1 \text{ N}\cdot\text{m}$ , dough weight 75 g, mixer temperature  $30^\circ\text{C}$ , hydration water temperature  $30^\circ\text{C}$  and kneading time 30 minutes. Following the tests, the curves are mathematically processed in order to acquire data close to those obtained on the Farinograph for the following settings: water absorption (%), development time (min), weakening ( $\text{N}\cdot\text{m}$ ) and stability (min).

Analytical and rheological characteristics were used for PCA, multivariate analyses. Principal Component Analysis (PCA) is based on the transformation of the primary data. The result is a new set of variables (principal components) that are linear combinations of the original variables and

are uncorrelated. The new variables thus generated are smaller in number, and yet account for the inherent variation of the data to the maximum possible extent. In fact, in this way, a new space (factor space) is generated onto which the cases and the variables can be projected and classified into categories. The significance of the components decreases progressively; however, the contribution of a single principal component to the explanation of each variable variation can not decrease as does the average of the contribution sum over all variables. For PCA results interpretation, plots of variables and objects in the projection of either PC onto axis  $x$  and  $y$  are necessary (ŠVEC *et al.* 2007).

## RESULTS AND DISCUSSION

**Analytical characteristics.** To illustrate the characteristics of the sample sets, the mean and range with abbreviated names are given in Table 1. The quality of wheat flours corresponded to the Romanian standard (SR 877:1996 – Wheat flours) for the mill products of 650 flour type. Almost the whole set is represented by bread making flours which cover a wide range of bread making quality.

**Rheological characteristics.** As the alveograph data (Table 2) show, the flours used in the experiments had a variable bread making potential, from poor to very good.

The values obtained with the standard option “Chopin+” protocol for the parameter indicating stability were much lower than the values obtained for the same parameter with the Mixolab, using the option “simulator” test (Table 3).

In the first stage, the correlation between the Mixolab with the standard option “Chopin+” protocol, the Alveograph and the Falling Number was analysed using the Principal Component Analysis (PCA) – Figure 2.

The correlation between the results of the Falling Number device and the variables obtained with Mixolab device, the points from C1 to C5, and also the difference between the points C12, C32, C34, C54 are represented in Figure 2a. The first two main components explain 100% of the total variance (PC1 = 99.96% and PC2 = 0.028%). Thus, it results that the first main component can express all parameters from the informational point of view, without the loss of any information. These things are normal because there is a very good correlation between the Falling Number index

Table 1. Flour analytical parameters

Parameters	Abbreviated name	Mean	Range		CV (%)
			min.	max.	
<b>Set 1</b> (for the sample flours analysed on the Mixolab using “Chopin+” protocol)					
Falling Number (s)	FN	349.30	103	485	33.10
<b>Set 2</b> (for the sample flours analysed on the Mixolab using simulator test)					
Wet gluten (%)	WG	27.13	23.6	29.9	6.53
Protein (%)	PR	14.66	11.7	16.3	8.41
Falling Number (s)	FN	438.6	251	499	14.95

and the following parameters that measure starch gelatinisation – C3 ( $r = 0.877$ ) and the difference between the points – C32 ( $r = 0.878$ )), amylolytic activity – C4 ( $r = 0.794$ ), starch retrogradation – C5 ( $r = 0.907$ ), and the difference between the points C54 ( $r = 0.953$ )).

From the point of view of the Falling Number index, there is a good correlation ( $r = 0.878$ ) between the index and the difference between the points C32. The dependence of the difference between the points C32 and the Falling Number indicates a strong relationship between the two parameters. The increase of the difference between C3 and C2 leads to an increase of the Falling Number index. A very good correlation ( $r = 0.953$ ) was obtained between the difference between the points C54 and the Falling Number, showing a relatively determinist connection. The increase of the difference between the measured parameters starch gelling (C5) and the amylolytic activity (C4) leads to an increase of the Falling Number index. A negative correlation was obtained between the Falling Number index and the difference between the points C34 ( $r = -0.54$ )

and C12 ( $r = -0.482$ ). The increase of the value of the slope that expresses the enzymatic degradation speed and of the slope that expresses the speed of the weakening of the protein network due to the effects of heat will lead to a decrease of the Falling Number index. This is explainable because the degradation speed of starch depends on its damage degree and on the quantity of  $\alpha$ -amylase in the flour. As a consequence, the lower is the amylolytic activity, the higher is the dough consistency.

PCA loadings of the Falling Number, Mixolab parameters and Alveograph values are represented in Figure 2b. The two plots represented here 82.59% and 17.06% of the total variance. The plot of PC1 vs. PC2 loadings shows, along the PC1 axis, a close association between the rheological parameters indicating: protein weakening (C2), deformation energy (W) and stability (S). This is due to the presence of large quantities of the grain enzymatic systems, thus of the proteolytic enzymes hydrolysing gliadine and glutenine (the moment opposed by the dough in the first warming stage decreases, resulting in a retrogradation of gluten strength to

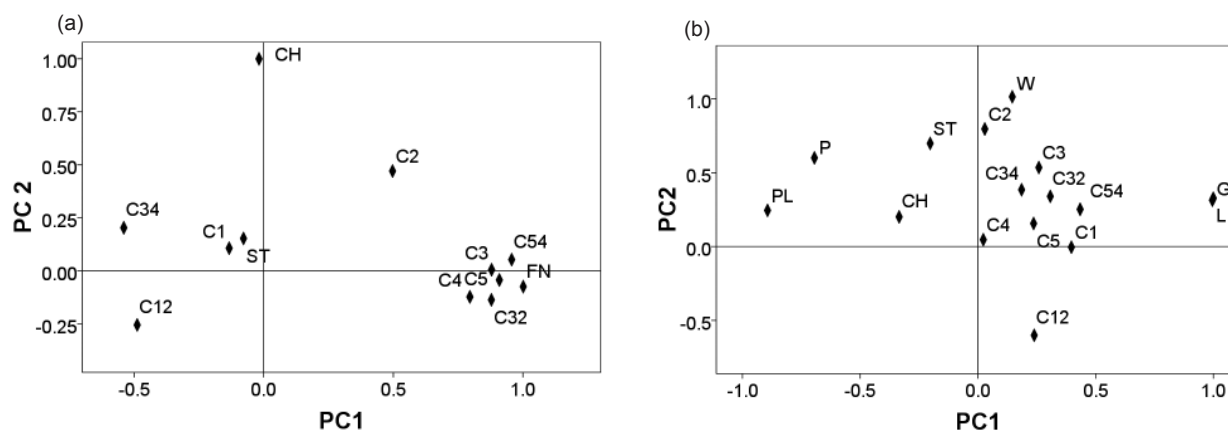


Figure 2. (a) PC1 and PC2 score for the Mixolab rheological characteristics and Falling Number; (b) PCA loadings of Mixolab, Alveograph and Falling Number

Table 2. Mixolab characteristics of flour samples

Parameters	Abbreviated name	Mean	Range		CV (%)
			min	max.	
<b>Set 1</b> (Mixolab characteristics of flour samples using standard option “Chopin+” protocol)					
Water absorption (%)	CH	58.09	54.6	62.4	3.01
Stability (min)	ST	6.33	3.06	10.46	37.70
Maximum consistency during – phase 1 (N·m)	C1	1.10	1.01	1.25	4.66
– phase 2 (N·m)	C2	0.31	0.09	0.49	35.01
– phase 3 (N·m)	C3	1.90	1.37	2.32	12.67
– phase 4 (N·m)	C4	1.42	0.33	1.87	25.72
– phase 5 (N·m)	C5	2.18	0.47	2.81	29.77
Difference of the points – C1–C2 (N·m)	C12	0.79	0.59	1.03	16.67
– C3–C2 (N·m)	C32	1.59	0.99	2.01	17.57
– C3–C4 (N·m)	C34	0.36	0.05	1.04	64.93
– C5–C4 (N·m)	C54	0.76	0.14	1.22	43.91
<b>Set 2</b> (Mixolab characteristics of flour samples using “simulator” test)					
Water absorption (%)	CH	56.54	55.0	64.7	4.87
Development time (min)	DT	3.10	1.03	8.07	62.59
Weakening (N·m)	WE	0.23	0.06	0.59	46.10
Stability (min)	ST	11.04	0.43	18.24	42.37
Maximum consistency during kneading $C_{\max}$ (N·m)	CM	1.12	0.67	1.36	15.67

different stresses). However, the value deformation energy (W) is inversely correlated with the value of the difference between the points C12 ( $r = -0.883$ ). PC2 distinguishes between maximum pressure (P), alveograph ratio (P/L), stability (S), and difference between the points C12. Stability (S) shows a positive effect on maximum pressure (P), alveograph ratio (P/L), and a negative one on the difference between the points C12.

At the second stage, PCA was performed on physical-chemical parameters of flour (wet gluten, protein content, Falling Number), Mixolab simulator parameters and Alveograph parameters values are to used test, the variations among the flour samples (Figures 3a–d).

The first two PCs (PC1 and PC2) explain 90.49% of the variation, with PC1 explaining 53.38% (Figure 3a). In Figure 3b, the first two PCs explained 99.79% of the variation, with PC1 explaining 99.14%. The protein is positioned in both plots at the same distance, a fact that indicates a similar correlation both between the rheological parameters resulting from the Alveograph and between those obtained with Mixolab. The content of protein naturally correlates with the quantity of wet gluten. There

is a positive correlation between the content of protein and the Falling Number ( $r = 0.713$ ) which is essentially connected with the second principal component PC2. Along the PC1 axis, the parameters water absorption (CH) and stability (ST) are well correlated and are indirectly correlated with maximum consistency during kneading (GM). As a consequence, in cereals richer in proteins, amylolytic activity decreases. This is due to the deactivation of  $\alpha$ -amylase which is retained by glutenine in quantities that become larger as glutenine becomes larger.

In Figure 3c, the first principle component PC 1 accounts for 82.27% of the total variance and the second component PC 2 accounts for 17.02% of the total variance. The parameters: stability (ST) and maximum pressure (P), are very well correlated ( $r = 0.763$ ) and inversely correlated with dough weakening (WE). Water absorption (CH) and alveograph ratio (PL) are well correlated ( $r = 0.554$ ) and are inversely correlated with dough weakening (WE). The second main component opposes the parameter alveograph ratio (PL) to the parameters extensibility (L) and swelling index (G), between which there is a good correlation ( $r = 0.875$ ).



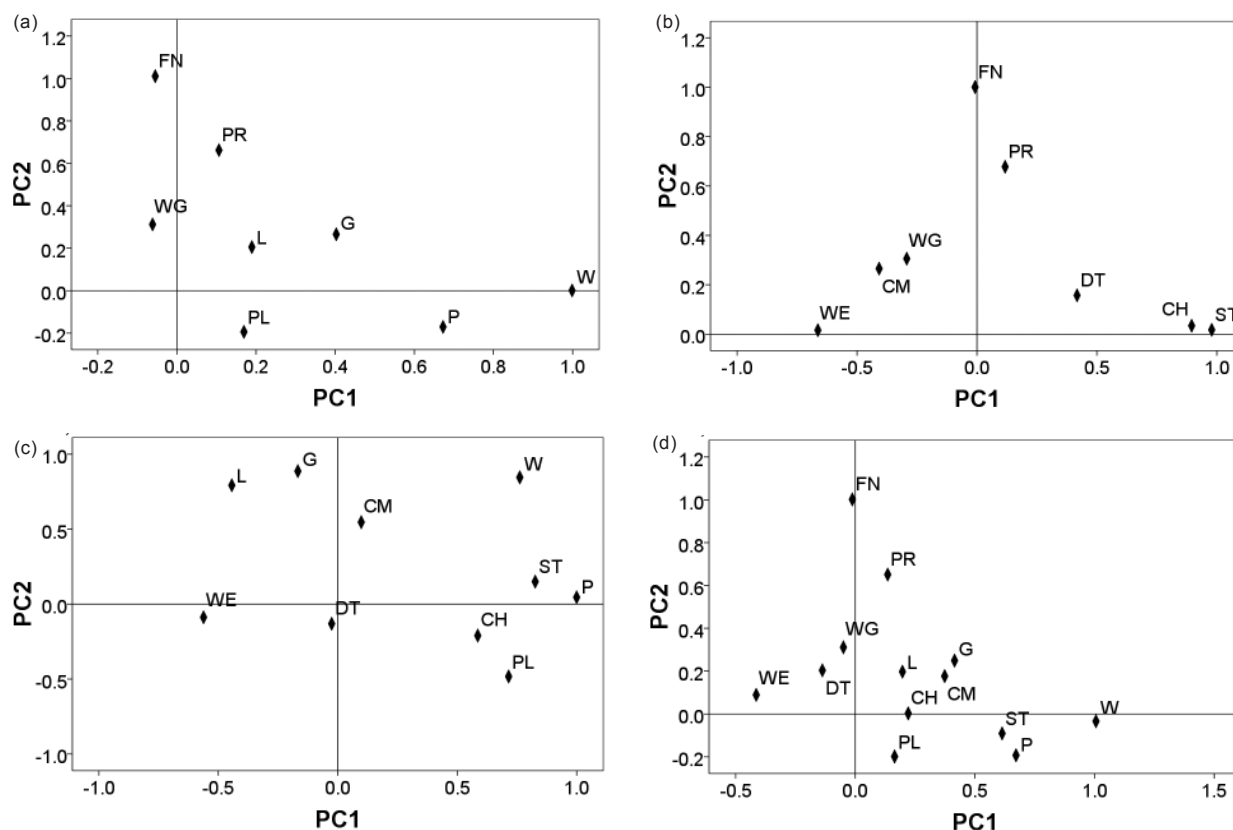


Figure 3. (a) PC1 and PC2 score for the Alveograph rheological characteristics and analytical characteristics (protein content, wet gluten, Falling Number); (b) PC1 and PC2 score for the simulator Mixolab rheological characteristics and analytical characteristics (protein content, wet gluten, Falling Number); (c) PC1 and PC2 score for the Alveograph and simulator Mixolab rheological characteristics; (d) PCA loadings of simulator Mixolab, Alveograph and analytical characteristics (protein content, wet gluten, Falling Number)

The rheological properties of the dough are improved up to a certain value of the water content, corresponding to the maximum swelling of

proteins, followed by a decrease in their value. The best consistence is obtained when the dough contains enough water to bloat the flour components.

Table 3. Alveograph characteristics of flour samples

Parameters	Abbreviated name	Mean	Range		CV (%)
			min.	max.	
<b>Set 1</b> (for the sample flours analysed on the Mixolab using “Chopin+” protocol)					
Maximum Pressure (mm)	P	61.63	21	127	45.38
Extensibility (mm)	L	71.96	13	126	43.20
Swelling Index (mm)	G	18.30	8.03	25.0	24.50
Deformation energy (10 <sup>-4</sup> J)	W	149.73	15	300	64.28
Alveograph ratio P/L	PL	0.99	0.36	2.75	55.13
<b>Set 2</b> (for the sample flours analysed on the Mixolab using simulator test)					
Maximum Pressure (mm)	P	90.8	36	127	27.59
Extensibility (mm)	L	72.6	37	126	33.91
Swelling Index (mm)	G	19.32	13.5	25.0	18.68
Deformation energy (10 <sup>-4</sup> J)	W	228.33	72	341	28.81
Alveograph ratio P/L	PL	1.47	0.51	3.32	54.56

During the flour components swelling, which is the best for bread making, the dough has the best resistance and elasticity.

PCA loadings of physical-chemical parameters of flour, Mixolab simulator parameters, and Alveograph parameters values are represented in Figure 3d. The first two PCs explain 90.26% of the variation, with PC1 explaining 53.24%. The plot of PC1 *vs.* PC2 loadings shows along the PC1 axis a high relationship between the stability (S), maximum pressure (P), and deformation energy (W) values, which are inversely correlated with the dough weakening value.

### CONCLUSIONS

Principal Component Analysis of the data set shows a high association between some analytical flour properties (protein-Falling Number), simulator Mixolab rheological properties (stability-hydration capacity), and Alveograph-Mixolab rheological properties (maximum pressure-stability). When Mixolab with the standard option “Chopin+” protocol and Alveograph were used, a strong direct relationship was found between maximum pressure (P), deformation energy (W), and protein weakening (C2). Also, stability (S), maximum pressure (P), and deformation energy (W) were inversely associated with the difference between the points C12. For Falling Number, a close direct positive relationship was found with starch gelatinisation (C3, difference between the points C32), amylolytic activity (C4), starch gelling (C5, difference between the points C54), and an indirect association with difference between the points C34.

The correlation established with PCA between the parameters determined with the three devices has highlighted the best correlation existing between the Falling Number and the parameters of the Mixolab device using the standard option “Chopin+” protocol. Also, by means of Alveograph and mixolab devices using Simulator and “Chopin+” protocols,

different but complementary data have been obtained. Therefore, the Alveograph and the mixolab provide distinct but complementary results.

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