

	EFFECT OF TEMPERATURE AND CONSISTENCY ON WHEAT DOUGH PERFORMANCE
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12	Running title: Consistency of wheat flour along breadmaking
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ABSTRACT

- 26 The effects of the dough consistency (300-700 BU), temperature of mixing (16-32 °C) and temperature along fermentation (15-35 °C) on the wheat bread dough performance during
- 28 mixing, proofing, cooking and cooling have been studied through a central composite experimental design. Farinograph responses revealed the significant role of dough
- 30 consistency (α <0.001) and mixing temperature (α <0.001) on wheat bread dough elasticity. Fermentation responses obtained from the rheofermentometer showed that dough
- 32 consistency induces a significant positive linear effect on dough development, whereas gas development was mainly governed by the fermentation temperature. Wheat bread dough
- 34 behaviour subjected to a dual mechanical shear stress and temperature constraint showed that dough consistency had a significant linear and positive effect on the starch gelatinization
- 36 and gelling process. Therefore, breadmaking is highly governed for the dough consistency, namely dough hydration, which has a direct consequence on the mixing, fermenting, cooking
- 38 and cooling performance of the wheat bread dough.
- 40 **Key words**: bread dough, hydration, consistency, temperature mixing, proofing.

INTRODUCTION

- In breadmaking, mixing is one of the key steps that determines the mechanical properties of the dough, which have a direct consequence on the quality of the end product. The rheological properties of wheat flour doughs are largely governed by the contribution of starch, proteins and water. The protein phase of flour has the ability to form gluten, a continuous macromolecular viscoelastic network, but only if enough water is provided for hydration and sufficient mechanical energy input is supplied during mixing (Amemiya and Menjivar 1992, Gras et al 2000, Rojas et al 2000).
- 50 During wheat bread processing several physical changes are involved, in which gluten proteins are the main responsible for bread dough structural formation, whereas starch is mainly implicated in final textural properties and stability (Cuq et al 2003). Dough mixing 52 involves large deformations, that are beyond the linearity limit, which correlates with non 54 linear rheological properties. Although the characterisation of linear viscoelastic behaviour has received vey much attention through different shear small deformations, the studies 56 exceeding the linear viscoelasticity are guite limited, and always required a range of values of the studied factors (Collar and Bollaín 2005). During mixing, fermenting and baking, dough 58 is subjected to different shear and extensional large deformations (including fracture), which are largely affected by temperature and water hydration. Several studies have been 60 conducted to determine the effect of water content on the dough viscoelastic behaviour reflecting mainly the linear viscoelastic response (Hibberd 1970, Mani et al 1992, Lefebvre 62 and Mahmoudi 2007). However, elongational rheology studies of wheat flour dough are required for assessing the baking performance, since the oscillatory shearing is unable to 64 develop the dough (Gras et al 2000). In addition, small deformation rheology are sensitive to starch-starch, starch-protein and protein-protein interactions (Rosell and Foegeding 2007), but only large deformation measurements can provide information about the extent of the 66

contribution of long-range (protein-protein) and short range (starch-starch, starch-protein)

- 68 interactions to the viscoelastic behaviour of wheat flour dough (Amemiya and Menjivar 1992).
- Macroscopic changes in the dough properties are mainly consequence of biochemical modifications. Water absorption and both protein content and quality have a strong influence on the properties of dough during mixing and in consequence on the resulting dough consistency (Armero and Collar 1997, Sliwinski et al 2004). In particular, proteins mainly involved in the viscoelastic properties of the dough are the high molecular weight glutenins subunits, which affect dough viscoelasticity in a similar and remarkable way than the water
- 76 content (Lefebvre and Mahmoudi 2007). During mixing the structural and rheological changes are accompanied of changes in the gluten protein composition (Skerrit et al 1999).
- Namely, mixing process induces an increase in the amount of total unextractable polymeric protein and large unextractable monomeric proteins (Kuktaite et al 2004).
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Studies about the influence of hydration on mixing have provided very useful information about the viscoelastic changes and microstructure behaviour during the process (Uthayakumaran et al 2002). However, usually those researches have been carried out in wheat flour-water mixtures under very controlled conditions of temperature. Scarce information is available pertaining bread wheat dough (yeasted dough) behaviour during mixing and proofing under different real conditions of hydration and temperature.

The importance of having this information is even more crucial for the processes involving retarded fermentation, like in the case of frozen dough. Nowadays, interrupted breadmaking processes are widely employed for decreasing the loss of consumers acceptance associated to loss of bread freshness (bread staling) during storage (Rosell and Gomez 2007). In those breadmaking process the mixing energy input, type of mixer, water amount in the formulation

and the presence of additives have great impact in the quality of bread (Nemeth et al 1996).

- 94 Breadmaking conditions must be specifically defined for this type of products.
- 96 The aim of this study was to determine the effect of both mixing and proofing temperature ranges and dough consistency on dough handling ability and fermentative performance of 98 wheat bread doughs in order to know the most appropriate experimental conditions to 98 optimize rheo-fermentative. For that purpose an experimental design was used. The 100 responses of the bread dough during thermal treatment were also determined by using the 102 Mixolab device.

102 MATERIALS AND METHODS

Commercial wheat flour for breadmaking was used in this study. The characteristics of the

- 104 flour were: 14.51% moisture content (ICC 110/1), 9.54% protein content (ICC 105/2), 1.09% fat content (ICC 136), 0.51 % ash content (ICC 104/1), 450s Falling Number (ICC 107/1) and
- ¹⁰⁶ 94.4 % gluten index (ICC 155). The alveographic parameters (ICC 121) were 57 mm, 141 mm and 237 10⁻⁴ J for tenacity (P), extensibility (L) and deformation energy (W), respectively.
- 108 The bread improver or processing aid (83.2% wheat flour, 15% Multec datem HP20, 1% fungal α -amylase and xylanase, 0.8% ascorbic acid) was provided by Puracor (Groot-
- 110 Bijgaarden, Belgium). The rest of the ingredients were acquired in the food market.

112 Bread dough sample

Basic wheat dough formula on 100 g flour basis consisted of the amount of water necessary

- 114 to give the required consistency, 5% (flour basis) baker's compressed yeast, 2% (flour basis) commercial salt, 1% (flour basis) bread improver. Bread dough was mixed in a Farinograph
- 116 (Brabender, Duisburg, Germany), following the ICC Method (ICC 115/1). In order to determine the effect of mixing and proofing conditions on the wheat bread dough parameters
- 118 an experimental design was used. Design factors (quantitative independent factors) included

dough consistency (from 300 to 700 BU), mixing temperature (from 16 to 32°C) and proofing

- 120 temperature (from 15 to 35°C). The model resulted in 16 different combinations of hydrated wheat dough mixed in a Brabender Farinograph (300g flour capacity) during four minutes.
- 122 Preliminary experiments were performed to determine the amount of water required for obtaining the dough consistency levels specified in the experimental design.

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Mixed dough samples were immediately transferred to the Mixolab device and Rheofermentometer vessel for further analysis.

128 Fermentation

The rheology of dough during fermentation was determined using a Rheofermentometer F3

- 130 (Tripette et Renaud, France) following the supplier specifications. Hydrated wheat bread dough (315g) was placed in the fermentation vessel at different temperatures (according to
- 132 experimental design) for three hours; a weight constraint of 2.0 kg was applied. The rheofermentometer measured and recorded simultaneously the parameters related to dough
- 134 development, gas production, and gas retention. A detailed description of this equipment and the parameters is reported by Erdogdu-Arnoczky et al (1996) and Wang et al (2002).

136 Mixolab measurements

Mixing and pasting behaviour of the wheat bread dough was studied using the Mixolab 138 (Chopin, Tripette et Renaud, Paris, France) which measures in real time the torque (expressed in Nm) produced by passage of dough between the two kneading arms, thus

- allowing the study of its physico-chemical behaviour (Bonet et al 2006, Collar et al 2007).Rosell et al (2007) reported a detailed description of the equipment and the parameters
- 142 registered. The instrument allows analysing the quality of the protein network, and the starch behaviour during heating and cooling. For the assays, 50 grams of wheat bread dough were
- 144 placed into the Mixolab bowl and mixed. The settings used in the test were 8 min at 30°C, temperature increase at 4°C/min until 90°C, 8 min holding at 90°C, temperature decrease at

- ¹⁴⁶ 4°C/min until 55°C, and 6 min holding at 55°C; and the mixing speed during the entire assay was 73 rpm. Parameters obtained from the recorded curve were: initial consistency (C1),
- 148 stability (min) or elapsed time at which the torque produced is kept constant, minimum torque (Nm) or the minimum value of torque produced by dough passage subjected to mechanical
- and thermal constraints (C2), peak torque (Nm) or the maximum torque produced during the heating stage (C3), the minimum torque during the heating period (Nm) (C4) and the torque
- obtained after cooling at 50°C (C5). In addition, the slopes during heating were determined and referred to α (protein reduction) and β (starch gelatinization).
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Experimental design and statistical analysis design

A central composite design, consisting of three factors (DC, MT, FT) five level pattern with 16 design points was used (Table 1). Factors levels were coded as -1,68179, -1, 0, +1, +1,68179, and included dough consistency (from 300 to 700 BU), mixing temperature (from 16 to 32°C) and proofing temperature (from 15 to 35°C). For each dough characteristic (response) measured along mixing, fermenting, heating and cooling, analysis of variance was conducted using Statgraphics V.7.1 program (Bitstream, Cambridge, MN), to determine significant differences among the factors combination. Response surface plots were generated from the regression equations by using the Statgraphics program. Response surface plots were obtained by holding the independent variable with least significant effect

on the particular response at constant value and changing the other two variables.

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RESULTS AND DISCUSSION

- 168 Experimental data from the central composite design of wheat bread dough characteristics (responses) during mixing, fermentation and dual mechanical shear stress and temperature
- 170 constraint were statistically analyzed in order to determine the significance of design factors.Multiple regression equations were constructed to estimate the effect of dough consistency,

172 mixing temperature and proofing temperature (independent variables) on bread dough responses. The magnitude of the coefficients in second order polynomials shows the effect
174 of the concerned factor on the response.

176 Effect of consistency and temperature on Farinograph deformation responses of wheat bread dough

- 178 It is generally accepted that mixing characteristics are strongly related to dough rheological properties, and they can be recorded as torque versus time curves obtained from small scale
- 180 mixers (Dobraszczyk and Morgenstern 2003). For Farinograph responses, only dough consistency and mixing temperature were included as independent factors, since
- 182 fermentation conditions would not have any physical meaning. Regression coefficients and correlation coefficients or coefficients of determination (R²) indicated the regression
- 184 equations accounted for 58 to 98% of the variance in Farinograph responses (Table 2).Dough consistency, the most prominent factor affecting dough mixing parameters, had a
- 186 significant linear effect on most of the Farinograph responses, with the unique exception of development time, on which dough consistency induced a negative quadratic effect.
- 188 Elasticity defined as the bandwith of the farinogram, which in the case of the mixograph has been related to extensional properties of the dough during mixing and can be used to assess
- 190 indirectly the role of water in the lubrication during mixing (Gras et al 2000). Results obtained in the present study shows that in the Farinograph as well, the elasticity or bandwith is
- 192 significantly related to water hydration, and as the dough consistency increases (water added decrease) the elasticity increases and thus the extensional viscosity.

194 The temperature during mixing resulted in significant (α <0.01) negative linear effect on the development time and consistency at end. Similar effect was described by Farahnaky and

- Hill (2007), when used the water content, temperature and salt level, for developing a model that could compensate quantitative changes of any of those factors. Conversely, the mixing
- 198 temperature induced significant positive linear effect on elasticity. Therefore, performing the

mixing at 16C resulted in an increase (21%) of the elasticity, whereas a decrease in the
development time and consistency at end of 28% and 6%, respectively. The interaction of
both factors (dough consistency and mixing temperature) had a significant (α<0.01) positive
effect on dough elasticity.

204 Response surface plots were drawn for the significant dough mixing responses (Figure 1), where it can be observed the predominant effect of dough consistency on all the mixing 206 responses, and the sinergistic effect that dough consistency and mixing temperature induced on the dough elasticity. Mixing of flour and water is associated with the hydration of flour 208 particles, where wheat gluten proteins pass through their glass transition phase, which increase protein molecular chain mobility (Cuq et al 2003). The input of mechanical energy 210 that takes places during kneading confers the necessary energy for distributing flour components, favoring the proteins interaction and the formation of covalent bonds between 212 them, which finally leads to the formation of a continuous macromolecular viscoelastic structure (Cuq et al 2003). In the range of dough consistency (directly related to water 214 content) and mixing temperature tested, wheat gluten proteins pass from the glassy state to the rubbery state, since at water contents above 15-20% glass transition of the gluten 216 proteins occurs at room temperature (Cug et al 2003). The effect of water content is guite small in the range 36.5-42.5% as revealed some stress relaxation studies within the linear 218 viscoelasticity (Phan-Thien and Safar-Ardi 1998). The studies carried out in shear, that is within the linearity limit, show that the water content has a large magnitude effect on the 220 viscoelastic behaviour of the dough (Manik et al 1992, Lefebvre and Mahmoudi 2007). An increase of the water content resulted in a decrease of the elastic (G') and viscous (G") 222 moduli, having the hydration in the range (43-47%) an identical effect on both moduli (Hibberd 1970). Results from the present study showed that water content in the range 49-224 63% also has also a predominant effect on bread dough (in the presence of baker's yeast) shear deformation responses.

Effect of consistency and temperature on the fermentation responses of wheat bread 228 dough

One of the main objectives of mixing is to develop a three dimensional viscoelastic structure
with sufficient gas retaining properties for holding the carbon dioxide released during the fermentation. During fermentation, the expansion of the air bubbles previously incorporated
during mixing will provide the characteristic aerated structure of bread. Dough used for breadmaking involving freezing has usually greater consistency than the conventional dough
because the water amount is reduced (Sahlstropm et al 1999, El-Hady et al 1996). This consistency will affect the subsequent operations like fermentation and baking. The changing
properties of dough during fermentation stage were continuously measured by the

- rheofermentometer, which provided information about dough development, gas production and gas retention (Bloksma 1990).
- Bread dough corresponding to the different runs of the experimental design showed large
 differences in their behaviour during fermentation (Figure 2). Different groups were classified
 according to their dough development trends during fermentation. Some bread doughs (runs
 3, 5, 11, 12) showed very low stability during fermentation, which was related to the bread
 doughs with the lowest consistency and high either mixing or proofing temperature. That
 effect was particularly extreme for the run 11 that corresponded to the bread dough with the
 lowest dough consistency (300 BU). In addition, there was a group of samples with very low
 rate of volume increase, which corresponded to the runs (8, 10, 13, 14, 15) with the lowest
- the highest consistency bread dough, mixed at 24 °C and fermented at 25 °C (run 2).
- All the recorded parameters during the fermentation of wheat bread dough were significantly dependent on the wheat bread dough consistency and fermentation temperature (Table 3 and 4). The temperature during dough mixing did not have any significant single effect on the

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fermentation responses. The regression equations obtained for dough development responses showed very high correlation coefficients, ranged from 87 to 95% (Table 3). The wheat bread dough consistency, and in extension the water hydration, had a significant and

256 positive effect on the volume of bread dough (Hm and h) and time to reach them (T1, T2), whereas a linear negative effect on (Hm-h)/Hm, which has an inverse relationship with the dough stability during fermentation. Dough consistency at the highest level tested (700 BU) resulted in 33% increase of the maximum bread dough volume and pertaining the bread dough stability the increase went up to 67%.

Fermentation temperature, although it had not a significant effect on the maximum dough 262 volume, provoked a linear negative effect on the other dough development responses, with the exception of dough stability where the effect was positive. It must be stressed that the 264 combination of dough consistency and fermentation temperature resulted in a significant antagonistic effect on dough stability along fermentation. Response surface plots (Figure 3) 266 showed the main effect of the dough consistency and the fermentation temperature on the dough development behaviour. Bread dough volume during fermentation increased with the 268 dough consistency and temperature increase. Conversely, the bread dough volume at the end of the fermentation (h), the dough stability and the time to reach the maximum volume 270 (T1) were affected in opposite manner by these two independent variables. Therefore, from Figure 3 it can be extracted that bread dough stability can be increased by performing high 272 consistency bread dough. As it was expected, fermentations can be accelerated by raising the temperature and when high temperature is needed for fermenting bread dough 274 increasing bread dough consistency can overcome the problems associated to low stability. Conversely, when a retarded fermentation is required, high bread dough consistency 276 associated to low temperatures are advisable.

278 In addition, temperature along fermentation significantly (α <0.001) affected gas development responses of bread dough (Table 4). The regression equations explained more than 94% of

- 280 the variance in the gas development responses during fermentation, as indicated the values of the correlation coefficients, R². Fermentation temperature had a linear effect on all the gas 282 development responses, and in addition, this factor induced a quadratic effect on the time to reach maximum gas production (T'1), the time when appears dough porosity (Tx) and the 284 volume of retention. However, the effect of the fermentation temperature was positive on the maximum gas development (H'm), total volume, volume of carbon dioxide lost and volume of 286 retention, whereas was negative on T'1, Tx and retention coefficient. Gas retention is of considerable interest due to its repercusion on the crumb structure and volume of bread 288 (Giannou et al 2003). Those effects were related to an increase in the yeast fermenting activity and also revealed an increase of the dough permeability to carbon dioxide (Wang et 290 al 2002). Second order interaction was observed between the temperature of bread dough during mixing and that during fermentation. Presumably, when the rheofermentometer vessel 292 equilibrated the initial dough temperature (mixing temperature) to the one fixed in the experimental design for the fermentation temperature (the third independent variable), the 294 effect of the initial temperature was masked by the fermentation temperature. There was a significant effect of the combination initial dough temperature and fermentation temperature 296 on the initial stage of yeast fermentation, whereas, no significant effect of the fermentation temperature was observed on the maximum volume of bread dough (Hm). The temperature 298 during mixing can affect the activity of the yeasts, modifying their fermenting ability, and a dramatic effect on the loaves baked from frozen dough has been observed when a final 300 mixing temperature over 20°C was used (Zounis et al 2002). However, some authors found better results with high temperatures at the end of mixing and with a reduction of water 302 content (Sahlstropm et al 1999) although differences in results could be ascribed to the diversity of formulations tested.
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Mixing and thermal responses of wheat bread dough derived from the Mixolab device

306 In studying the baking performance, mixing and proofing are not the only stages that should be analyzed. Changes in the viscosity of highly hydrated starch-based systems such as 308 doughs during baking are known to affect the viscoelastic behavior and texture and keeping quality of finished bread (Collar 2003). It has been already stated that the presence of 310 biochemical constituents like the added ingredients, additives, and technological aids in dough formulation favor viscosity changes in dough influencing baking performance and 312 bread staling (Andreu et al 1999, Collar et al 2005, Collar et al 2006). In order to determine the possible relevance of the mixing conditions on the further baking process, wheat bread 314 dough was subjected to a dual mechanical shear stress and temperature constraint using the Mixolab device. Information concerning mechanical and thermal protein weakening, starch 316 gelatinization and starch gelling can be extracted from the recorded curves (Rosell et al

2007, Collar et al 2007).

318 The plots in Figure 4 portray the various recorded curves in the Mixolab device corresponding to runs of the experimental design. The first part of the curves recorded the 320 bread dough behaviour during overmixing, showing a decrease in the dough consistency, as a consequence of the continuous mechanical input that produces the protein breakdown with 322 a reduction in the average molecular weight of the proteins (Skerrit et al 1999). Despite, the great variation observed in the plots of the bread dough samples obtained from different 324 processing conditions, no groups of samples could be distinguished during the mixing. Regardless run 2 that corresponded to the highest consistency (700 BU), small differences 326 were observed during the mixing stage, which was expected since the bread samples were transferred from the Farinograph to the bowl of the Mixolab after completing bread dough 328 hydration. Nevertheless, the largest dispersion was observed during the gelatinization and gelling stages. A group of bread samples with very close behaviour during gelatinization and 330 gelling were observed, which corresponded to the runs with the lowest bread dough consistency (3, 5, 8, 11, 13) and the lowest mixing temperature (run 4). At this stage, the 332 starch gelatinization is the main responsible for the torque variations, which includes starch

granules absorption of the water available in the medium, their swelling, and the amylose
chains leaching out into the aqueous intergranular phase promoting the increase in the
torque, till the physical breakdown of the starch occurs (Rosell et al 2007). The following
decrease in temperature produces an increase of the torque associated to the gelation
process of the starch (Collar et al 2007). The swelling of the starch is greatly dependent on
the water available in the medium, which controls the gelatinization behaviour (León et al
1997) and that effect is magnified in the dough mixture used in the Mixolab device were
limited amount of water is available for the starch gelatinization (Rosell et al 2007).
Therefore, high consistency bread doughs (low hydrated) derived in limited gelatinization and
also limited gelling.

344 As occurred in the analysis of the mixing behaviour of the dough carried out in the Farinograph, when the Mixolab responses were analyzed, only dough consistency and 346 mixing temperature were included as independent factor, since the inclusion of fermentation conditions would not have any physical meaning. Dough consistency significantly affected 348 almost all the responses during mixing, heating and cooling of wheat bread dough, with the exception of stability, time to reach the minimum torque (time to C2) and the rate of 350 gelatinization (β). However, only initial consistency (C1), minimum torque value (C2), minimum torque during heating (C4) and the torque after cooling (C5) showed high square 352 coefficients. Dough consistency had a positive and linear effect on the amplitude of the curve that is related to dough elasticity, which agrees with the result obtained when dough elasticity 354 was measured in the Farinograph. Dough consistency at the maximum level tested (+1.68179) induced an increase of 37% of the minimum torque (C2) resulted from the protein 356 weakening induced by temperature increase, which is related to the aggregation and further denaturation of the proteins (Rosell et al 2007, Rosell and Foegeding 2007). Dough 358 consistency also resulted in a significant effect on both starch gelatinization (C3) and gelling (C5), during heating and cooling, respectively; and also the minimum torque during heating

360 (C4) (Figure 5). Dough consistency, significantly affected the gelatinization and gelling process of the starch, which shows the consequences of the dough hydration on the baking 362 process. The temperature during mixing only induced a significant effect on the minimum torque during the heating period (C4), which is related to the cooking stability (Rojas et al 364 1999). A sinergistic effect on this response resulted with the combination of dough consistency and the mixing temperature (Figure 5), which might be related to some alteration 366 of the starch granules when different temperatures were applied along mixing. Pasting performance of wheat flours during cooking and cooling involves many processes such as 368 swelling, deformation, fragmentation, disintegration, solubilization, and reaggregation that take place in a very complex media primarily governed by starch granules behaviour 370 (Alloncle and Doublier 1991). The behaviour of bread dough during cooking and cooling, namely peak viscosity, pasting temperature and setback during cooling, have been highly 372 correlated with bread staling kinetic parameters (Collar 2003), being good predictors at dough level of bread firming behavior during storage and high sensory scores of fresh bread 374 (Collar 2003). Therefore, the significant Mixolab responses obtained with variable bread dough consistency reveal the important consequences of bread dough consistency along the 376 breadmaking process.

378 CONCLUSION

Bread dough consistency, and therefore dough water hydration, significantly affected dough
responses during mixing, fermentation, cooking and cooling. Farinograph responses
revealed the significant role of dough consistency (α<0.001) and mixing temperature
(α<0.001) on wheat bread dough elasticity. Fermentation responses obtained from the
rheofermentometer showed that dough consistency induces a significant positive linear effect
on dough development, whereas gas development was mainly governed by the fermentation
temperature. Therefore, water was a limiting factor during the breadmaking process for
protein hydration and later for starch gelatinization and gelling. Factors like dough

consistency and temperature of mixing and/or fermenting should always be defined as a combination to reach optimum performance of dough along breadmaking.

Data from this work can be used for optimising the mixing and fermentation conditions of

- 390 bakery products, especially those subjected to low and sub-zero temperatures that required retarded fermentation during breadmaking process.
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FIGURE CAPTIONS

- Figure 1. Response surface plots of the significant Farinograph responses obtained from the regression equations. DC: bread dough consistency, MT: mixing temperature, WA: water
 absorption.
- **Figure 2**. Plots of wheat bread doughs behaviour during fermentation recorded in the Rheofermentometer device. Numbers in legend are referred to experimental runs. Detailed

information about the runs is described in Table 1.

- Figure 3. Response surface plots of the significant dough development responses during fermentation obtained from the regression equations. Mixing temperature variable held at
 level 0. DC: dough consistency, FT: fermentation temperature.
- Figure 4. Plots of bread dough behaviour during mixing, heating and cooling recorded in the Mixolab device. Numbers in legend are referred to experimental runs. Detailed information
 about the runs is described in Table 1.
- **Figure 5.** Response surface plots of the significant Mixolab responses obtained from the regression equations. DC: bread dough consistency, MT: mixing temperature.

Table 1. A central composite design, consisting of a three factors (DC, dough consistency;MT, mixing temperature; FT, fermentation temperature) and five level pattern with 16 runs.

RUN	DC (BU)	MT (°C)	FT (°C)
1	0	0	0
2	1.68179	0	0
3	-1	1	1
4	0	-1.68179	0
5	-1	-1	1
6	0	1.68179	0
7	1	1	1
8	-1	-1	-1
9	1	-1	1
10	1	-1	-1
11	-1.68179	0	0
12	0	0	1.68179
13	-1	1	-1
14	1	1	-1
15	0	0	-1.68179
16	0	0	0

Table 2. Regression equation^a coefficients for bread dough mixing responses.

	Farinograph parameters						
Coefficient	WA	development time	elasticity	consistency at end			
	%	min	BU	BU			
b ₀	54.41	1.51	80.93	490.87			
b ₁ (DC)	-2.29 **	0.05	30.39 ***	107.97 ***			
b ₂ (MT)	-0.84	-0.25 *	10.05 ***	-17.54 **			
b ₁₁	-1.21	-0.14	5.77 *	-1.33			
b ₁₂	0.13	-0.13	8.75 **	-4.38			
b ₂₂	0.61	0.04	0.47	-6.64			
R-SQ (%)	74.54	57.72	96.29	97.62			

^a $y = b_0 + b_1 x_1 + b_2 x_2 + b_{11} x_1^2 + b_{22} x_2^2 + b_{12} x_1 x_2$

where $x_1 = DC$, $x_2 = MT$

(*), (**), (***) significant at α <0.05, α <0.01 and α <0.001, respectively.

534	Table 3. Regression equation ^a coefficients for bread dough development responses during
	fermentation.

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_	Dough development							
Coefficient	Hm	h	(Hm-h)/Hm	T1				
	mm	mm	%	min				
b ₀	53.87	37.37	32.75	105.93				
b ₁ (DC)	14.65 **	18.33 ***	-20.93 **	22.96 *				
b ₂ (MT)	2.20	2.21	-1.54	2.29				
b ₃ (FT)	3.52	-14.52 **	32.99 ***	-36.01 **				
b ₁₁	-2.40	0.96	4.68	2.39				
b ₁₂	-2.09	-2.68	1.93	1.13				
b ₁₃	1.84	5.23	-13.80 *	7.13				
b ₂₂	2.87	2.55	-3.03	8.75				
b ₂₃	-0.08	-2.08	1.93	-0.75				
b ₃₃	-0.68	-5.48	4.67	8.75				
R-SQ (%)	87.22	93.21	94.76	89.15				

^a $y = b_0 + b_1x_1 + b_2x_2 + b_3x_3 + b_{11}x_1^2 + b_{22}x_2^2 + b_{33}x_3^2 + b_{12}x_1x_2 + b_{13}x_1x_3 + b_{23}x_2x_3$

where $x_1 = DC$, $x_2 = MT$, $x_3 = FT$

(*), (**), (***) significant at α <0.05, α <0.01 and α <0.001, respectively.

	Gas development								
Coefficient	H'm	T'1	Тx	total volume	volume of CO ₂ lost	volume of retention	retention coefficient		
	mm	min	min	ml	ml	ml	%		
b ₀	77.78	53.45	62.39	1506.57	267.08	1240.04	82.57		
b ₁ (DC)	-0.25	3.23	4.71	1.44	-2.01	3.58	0.38		
b ₂ (MT)	2.64	-3.37	-4.58	17.30	12.95	4.09	-0.58		
b ₃ (FT)	31.73 ***	-28.31 ***	-41.49***	465.96 ***	249.90 ***	215.78 ***	-12.45 ***		
b ₁₁	-0.50	1.87	2.01	28.69	7.96	20.61	0.08		
b ₁₂	-0.04	0.00	-3.00	-10.13	-12.38	2.50	0.36		
b ₁₃	-0.34	-1.50	-0.38	26.13	8.13	18.25	-0.39		
b ₂₂	0.58	0.28	0.16	41.42	27.59	13.71	-0.94		
b ₂₃	4.02 *	ns	1.88	2.13	4.88	-2.75	-0.09		
b ₃₃	0.26	13.00 **	17.66**	-97.35	17.51	-114.98 **	0.79		
R-SQ (%)	99.20	97.22	98.13	96.77	95.02	96.60	94.65		

Table 4. Regression equation^a coefficients for bread dough volume responses during fermentation.

^a y= b₀ + b₁x₁ + b₂x₂ + b₃x₃ + b₁₁x₁² + b₂₂x₂² + b₃₃x₃² + b₁₂x₁x₂ + b₁₃x₁x₃ + b₂₃x₂x₃

where $x_1 = DC$, $x_2 = MT$, $x_3 = FT$

(*), (**), (***) significant at $\alpha{<}0.05,$ $\alpha{<}0.01$ and $\alpha{<}0.001,$ respectively.

	Mixolab parameters										
Coefficient	time C1	ampl.	stability	C1	time C2	C2	C3	C4	C5	α	β
	min	Nm	min	Nm	min	Nm	Nm	Nm	Nm		
b ₀	1.45	0.073	9.025	1.038	17.91	0.362	1.791	1.955	2.818	-0.094	0.019
b ₁ (DC)	-0.49 *	0.011 *	-0.784	0.166 ***	0.13	0.076 ***	0.110 *	0.224 ***	0.306 ***	0.001	0.012 *
b ₂ (MT)	-0.07	0.000	0.151	0.022	0.13	0.017	0.008	0.075 **	0.082	-0.013	0.005
b ₁₁	0.02	-0.002	0.151	0.023	0.08	0.002	-0.030	-0.035	-0.076	-0.007	0.002
b ₁₂	0.11	-0.005	0.263	0.005	0.10	0.005	0.079	0.075 *	0.035	-0.002	0.001
b ₂₂	-0.42	-0.005	0.575	-0.015	0.05	-0.010	-0.044	-0.019	-0.071	0.004	0.003
R-SQ (%)	52.63	47.64	29.14	85.59	39.25	84.20	55.65	91.96	87.69	15.99	39.92

Table 5. Regression equation^a coefficients for mixing and thermal bread dough responses.

^a y= $b_0 + b_1x_1 + b_2x_2 + b_{11}x_1^2 + b_{22}x_2^2 + b_{12}x_1x_2$

where $x_1 = DC$, $x_2 = MT$

(*), (**), (***) significant at α <0.05, α <0.01 and α <0.001, respectively.

Figure 1. Response surface plots of the significant Farinograph responses obtained from the regression equations. DC: bread dough consistency, MT: mixing temperature, WA: water absorption.





Figure 2. Plots of wheat bread doughs behaviour during fermentation recorded in the Rheofermentometer device. Numbers in legend are referred to experimental runs. Detailed information about the runs is described in Table 1.



Figure 3. Response surface plots of the significant dough development responses during fermentation obtained from the regression equations. Mixing temperature variable hold at level 0. DC: dough consistency, FT: fermentation temperature.



Figure 4. Plots of bread dough behaviour during mixing, heating and cooling recorded in the Mixolab device. Numbers in legend are referred to experimental runs. Detailed information about the runs is described in Table 1.



Figure 5. Response surface plots of the significant Mixolab responses obtained from the regression equations. DC: bread dough consistency, MT: mixing temperature.

