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## Influence of damaged starch on cookie and bread-making quality

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**Abstract** In this study two wheat and one triticale cultivars were milled in a disc mill to obtain different levels of damaged starch. The effects of the damaged starch content on physicochemical flour tests and cookie and bread-making quality were analyzed. The grain milling conditions in disc mill and grain hardness influenced the amount of damaged starch. The solvent absorption of flours, as measured by Solvent Retention Capacity Profile (SRC) and Alkaline Water Retention Capacity (AWRC), was significantly incremented by the damaged starch content. There was a consistent loss in cookie quality as the damaged starch content increased. In spite of the fact that the proteins were not affected by flour milling, bread quality decreased as the damaged starch content increased.

**Keywords** Damaged starch · Bread · Cookie · Wheat · Triticale

### Introduction

During wheat milling a portion of the starch granules sustains mechanical damage. The level of the damage depends on wheat hardness and milling technique. Hard wheat flour is typically used for yeast-leavened pan breads, whereas soft wheat flour is used for pastries, cookies, and cakes [1].

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Hard wheat is more difficult to reduce to flour-sized particles; therefore, hard wheat flour has a larger mean particle size than that of soft wheat flour.

Hard wheat produces a higher proportion of damaged starch during the milling process. In turn, damaged starch causes a higher water absorption capacity and is more readily hydrolyzed by  $\alpha$ -amylase. In formulas containing little or no added sugar, damaged starch levels should be high enough for good yeast gas production to occur, but not so high that dough handling problems are encountered [2, 3].

Amylase activity also leads to dextrins, which have an important effect on water-holding ability and porosity of the dough, as well as on bread softness, an excessive dextrin production is observed and sticky dough is obtained. The rheological properties of the dough are slightly modified by amylolytic consumption of damaged starch. Damaged starch has a large hydration capacity and its disappearance leads to a decreasing dough consistency. Consequently, although the production of oligosaccharides represents the positive aspect of amylolytic activity, the negative effect of damaged starch consumption on the rheological properties of dough should not be neglected.

Soft wheat flours are used for cookies. Predictive cookie quality parameters are related to cookie size: diameter and the ratio between the width and height. Flours that produce larger diameter and lower height cookies are considered to have better quality. Another aspect of cookie test baking is the cracking that occurs on the top surface of the cookie referred to as cookie top grain. Good top grain (many surface cracks) results from recrystallization of sucrose at the cookie surface during baking [4].

In soft wheat products, especially in cookies and cakes, high levels of damaged starch are detrimental to quality. Gaines et al. [5] reported that damaged starch contributed to reduce the cookie size when flours from three soft wheats were evaluated. Donelson and Gaines [6] observed that the cookie diameter was negatively affected by damaged starch level of reconstituted flours from hard and soft wheat. In a previous work we studied the effect of damaged starch content on cookie quality elaborated with triticale flour

obtained from six different tempering conditions, however, a narrow range of damaged starch level was obtained [7].

There have been numerous studies about damaged starch, but the effect of damaged starch on baking quality has been evaluated using reconstitution procedures, flours with a narrow range of damaged starch or flours obtained from several wheat cultivars with different degrees of hardness, and consequently, with different damaged starch and protein levels, where the quality of the final products was strongly influenced, not only by the damaged starch level but also by the flour composition.

The more important factor in baking quality is the gluten characteristics of dough. Strong dough with an extensive gluten network is suitable for bread making [8]. In contrast, weak dough without an extensive gluten network is best for cookies and cakes [9]. For this reason the effect of damaged starch on flour quality is difficult to identify independently of protein influence, so we think that it is important to clarify damaged starch effects on baking quality limiting protein influence.

In this study two wheat and one triticale flours were re-milled to obtain different levels of damaged starch without changing the quality and quantity of protein. The aim of this work was to study the effects of the damaged starch content on physicochemical flour tests and cookie and bread making quality.

## Materials and methods

### Samples

Two wheat cultivars, Klein Don Enrique and Baguette, and one triticale cultivar, Tatú, were used. Tatú was grown in mid-level fertility soils at Campo Escuela de la Facultad de Ciencias Agropecuarias, Universidad Nacional de Córdoba, Argentine. Baguette and Klein Don Enrique were provided by the Instituto Nacional de Tecnología Agropecuaria (INTA), Estación Experimental Marcos Juárez, Córdoba, Argentine.

The kernels were milled to  $58 \pm 2\%$  flour yield on a 4-roller laboratory mill (Agromatic AG AQC 109, Laupen, Switzerland). Then each of those three flours was intentionally re-milled in a Whisper Series Bench Top disc mill (Rocklabs, Auckland, New Zealand). Wheat flours were re-milled for 0, 2.0, and 5.0 min and triticale flour was re-milled for 0, 3.5, and 7.0 min. The re-milling times were chosen according to a previous analysis in order to obtain three flours of each cultivar with low, medium and high damaged starch levels. Moisture was determined by the AACC 44-19 standard method [10].

### Grain hardness

Grain hardness was determined by the Particle Size Index (PSI), following the AACC 55-30 method [10], using the Agromatic AG 109 mill (Laupen, Switzerland). The result was calculated as the relative weight of sieved flour  $\times 100$ ,

and compared with a table to appoint the relative hardness. Determinations were made in duplicate.

### Protein

The nitrogen content was determined by the AACC 46-13 Micro Kjeldhal Method modified with boric acid [10]. The sample was digested in a Technicon II digester (Dublin, Ireland), for four hours, then, the distillation was done in a VELP Scientifica UDK126A unit of distillation (Milan, Italy), the nitrogen was collected in a boric acid solution and the crude protein was calculated  $N \times 5.7$ . Determinations were made in triplicate.

### Damaged starch

The content of damaged starch was determined according to the AACC 76-30A method [10]. A fungal enzyme from *Aspergillus oryzae* (A6211, Sigma Chemical Co., St. Louis, MO, USA) was used. Determinations were made in triplicate.

### Determination of flour quality

Alkaline water retention capacity (AWRC) was determined according to the AACC 56-10 method [10]. Flour (1 g) was suspended in 5 ml of  $8.4 \text{ g l}^{-1} \text{ NaHCO}_3$ , hydrated for 20 min and centrifuged at  $1000 \times g$  for 15 min at room temperature. The sediment obtained was weighed and the AWRC was calculated.

Sodium dodecyl sulfate sedimentation index (SDS-SI) values were determined using 1 g of flour moistened with 8 ml of Coomassie Blue solution in a 25 ml cylinder. The sample was left to stand for 3 min, 40 s; and vortexed for 5 s; then, left to stand for 1 min, 55 s; and vortexed again. SDS and lactic acid (12 ml) were added immediately and agitated for 1 min in a horizontal agitator. The resulting suspension was left to stand 14 min, and the volume of moistened flour was measured. Results were expressed in cubic centimeters [11].

Solvent retention capacity profile (SRC) was obtained according to the AACC 56-11 method [10]. White flour samples (5 g) were suspended with 25 g of water,  $500 \text{ g l}^{-1}$  sucrose,  $50 \text{ g l}^{-1}$  sodium carbonate and  $50 \text{ g l}^{-1}$  lactic acid. The samples were hydrated for 20 min and centrifuged at  $1000 \times g$  for 15 min. Each precipitate obtained was weighed and the SRC for each solvent was calculated according to the following equation (Gaines, personal communication):

$$\% \text{ SRC} = \left[ \left( \frac{\text{PW}}{\text{FW}} \right) \times \left( \frac{86}{100 - \%M} \right) - 1 \right] \times 100$$

PW is the precipitate weight; FW the flour weight;  $\%M$  the flour moisture content. Determinations were made in triplicate.

## Preparation of cookies

Cookies were prepared according to León et al. [12]. The ingredients used were: flour (45 g); caster sugar (27 g); shortening (20 g); powdered milk (2.25 g);  $\text{NaHCO}_3$  (0.50 g);  $\text{NaCl}$  (0.42 g) and 8.5 ml of water. Shortening, sugar and water were mixed to form a creamy butter. The rest of the other ingredients were added and the dough was kneaded for 5 min. The ingredients were mixed in a spiral arm mixer (Philips HR 1495, Argentine). Baking was performed for 10 min at 200 °C in a forced convection oven (Continental 2001, Brazil) equipped with a temperature controller. Six cookies were obtained by batch and 4 cookies (more homogeneous) were selected to determine cookie factor. To determine cookie quality the term cookie factor was introduced as the ratio between the width and height of four cookies taken at random. The higher value was correlated to better quality. Cookie production was made in duplicate.

## Baking procedure

The recipe and bread making process followed here are those currently employed in our country in the preparation of bread. The dough formulation used in this study comprised: 1 kg wheat flour, 30 g compressed yeast, 18 g sodium chloride, 2 g sodium propionate, 0.15 g ascorbic acid, and 600 ml water. The water addition was based on a farinograph test using the 500 BU line. The ingredients were mixed in an Argental L-20 mixer (Argental, Santa Fe, Argentine). Yeast and salt were previously dissolved in water and the remaining ingredients were added as solids. The resulting dough was allowed to rest for 15 min in a fermentation cabinet at 30 °C and 70% RH and, then, the bulk dough was sheeted in a Mi-Pan vf roller (Mi-Pan, Córdoba, Argentine) containing two rolls of 50 cm × 12.7 cm. The dough was then divided in 80 g pieces and molded into a loaf shape (Braesi MB 350, Brazil). Dough pieces were immediately proofed at 30 °C (96% RH) up to maximum volume increment (about 90 min) [13] and baked at 200 °C for 18 min. Bread-making production was made in duplicate.

## Bread properties

**Bread volume:** Bread loaf specific volumes were determined by rapeseed displacement and weight, 4 h after baking. Form ratio was measured as height/width in each loaf. Determinations were made in duplicate.

**Texture of crumb:** Three bread pieces were cut into 2 slices (2.5 cm thick) and the ends were discarded. Each slice was subjected to a compression test in a TA-XT2i texturometer (Stable Microsystems, Surrey, UK), under the following conditions: compression cell 5 kg; crosshead speed, 100 mm  $\text{min}^{-1}$ ; maximum deformation, 40%; grip dimension, 3.6 cm. The hardness of the crumb was reported as the force required to compress samples to 25% of their original width. Six slices were

analyzed per point, and average values were reported. Determinations were made in duplicate.

## Wet gluten

Wet gluten balls from wheat and triticale flours were obtained by gluten hand-washing method following the AACC standard method 38-10 [10]. Gluten balls were made in duplicate.

## Protein extraction and electrophoresis

Proteins were extracted with a buffer solution (flour:liquid 1:30) (pH 6.8) containing 0.063 M Tris-HCl, 20 g  $\text{l}^{-1}$  SDS, 100 g  $\text{l}^{-1}$  glycerol, and 0.1 g  $\text{l}^{-1}$  bromophenol blue, without 2-mercaptoethanol [14]. A multistacking SDS-PAGE procedure was used to determine the size distribution of polymeric proteins under nonreducing conditions. Three stacking gels (pH 6.8) of 40 g  $\text{l}^{-1}$  T, 27 g  $\text{l}^{-1}$  C; 60 g  $\text{l}^{-1}$  T, 27 g  $\text{l}^{-1}$  C; 80 g  $\text{l}^{-1}$  T, 27 g  $\text{l}^{-1}$  C were laid on top of a 120 g  $\text{l}^{-1}$  T, 27 g  $\text{l}^{-1}$  C resolving gel [15].

Gels were analyzed by densitometry in an Image Master VDS (Pharmacia Biotech Inc., Uppsala, Sweden).

## Statistical analysis

Results were expressed as mean values  $\pm$  SD. The data were statistically treated by analysis of variance, the means were compared by the LSD Fisher test at a significance level of 0.05, and the relationships between measured parameters were assessed by Pearson's test, in all cases using the INFOSAT statistical software (Facultad de Ciencias Agropecuarias, UNC, Argentine).

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## Results and discussion

In order to obtain wheat and triticale flours with different levels of damaged starch, the samples were re-milled at different periods of time in a disc mill, consequently, the damaged starch content increased with the milling time for each sample. The protein content did not show significant differences between different milling times (Table 1). Both wheat cultivars, Baguette and Klein Don Enrique, showed higher grain hardness and damaged starch levels than triticale cultivar Tatú because soft grain offered a lower resistance to milling and the flour obtained had a lower content of damaged starch.

Solvent retention capacity (SRC) establishes a practical flour quality and functionality profile useful for predicting baking performance [10]. The lactic acid SRC is associated with glutenin characteristic, sodium carbonate SRC with levels of damaged starch and sucrose SRC with pentosan and gliadin characteristics; water SRC is influenced by all the flour constituents [16].

**Table 1** Particle size index (PSI), damaged starch, protein content and wet gluten of flour samples

	PSI (%)	Disc mill time (min)	Damaged starch (%) <sup>a</sup>	Protein (%) <sup>a</sup>	Wet gluten (%)
Baguette	15.3 ± 0.2	0.0	9.3 ± 0.9 b	11.6 ± 0.1 b	30.7 ± 1.8 a,b
	Hard	2.0	14.7 ± 1.1 e	11.9 ± 0.5 b	28.9 ± 0.5 a
		5.0	17.2 ± 0.6 f	11.7 ± 0.2 b	28.7 ± 0.2 a
Klein Don Enrique	21.2 ± 0.8	0.0	8.4 ± 0.3 b	14.3 ± 0.2 c	32.9 ± 0.3 c,d
	Medium soft	2.0	12.8 ± 0.6 d	14.1 ± 0.3 c	33.6 ± 2.5 d
		5.0	17.7 ± 1.0 f	13.9 ± 0.4 c	32.6 ± 0.7 c,d
Tatú	25.6 ± 0.1	0.0	6.1 ± 0.2 a	9.7 ± 0.6 a	nd
	Soft	3.5	10.4 ± 0.6 c	10.0 ± 0.4 a	nd
		7.0	14.0 ± 0.6 e	9.8 ± 0.3 a	nd

The values are the mean of three measurements with the standard deviation. Values followed by different letters are significantly different ( $p < 0.05$ )

nd, no development

<sup>a</sup>Expressed in dry- weight of flour

**Table 2** Physicochemical tests of flour samples

Sample (min)	AWRC (%)	H <sub>2</sub> O SRC (%)	Na <sub>2</sub> CO <sub>3</sub> SRC (%)	Sucrose SRC (%)	Lactic SRC (%)	SDS-SI (cm <sup>3</sup> )
Baguette (0)	65.6 ± 2.2 b	67.4 ± 0.4 c	78.7 ± 0.1 b	86.3 ± 0.5 a	109.9 ± 1.0 d	10.7 ± 0.3 a
Baguette (2)	76.1 ± 1.2 c	75.6 ± 0.2 f	92.3 ± 1.7 d	100.1 ± 0.1 b	125.0 ± 1.0 f	9.8 ± 0.3 b
Baguette (5)	79.7 ± 1.5 d	79.3 ± 0.2 g	99.0 ± 0.2 e	106.5 ± 0.5 c,d	126.6 ± 1.9 f	9.4 ± 0.1 c,d
Klein DE (0)	67.3 ± 1.6 b	65.0 ± 0.1 b	78.1 ± 0.9 b	90.6 ± 1.1 a	102.5 ± 0.4 c	9.3 ± 0.1 a
Klein DE (2)	73.9 ± 0.8 c	72.4 ± 0.6 e	90.5 ± 0.0 c	102.3 ± 0.1 b,c	116.1 ± 2.3 e	11.4 ± 0.7 b,c
Klein DE (5)	85.4 ± 0.5 e	83.9 ± 1.2 h	109.6 ± 0.4 f	121.7 ± 0.4 e	132.6 ± 2.1 g	13.9 ± 0.6 e
Tatú (0)	63.0 ± 0.4 a	60.9 ± 0.5 a	72.8 ± 1.0 a	88.2 ± 1.1 a	77.9 ± 0.9 a	8.6 ± 0.1 a
Tatú (3.5)	82.0 ± 1.3 d	70.3 ± 0.6 d	99.0 ± 0.0 e	107.5 ± 0.6 d	89.5 ± 1.5 b	7.3 ± 0.4 d
Tatú (7)	92.9 ± 2.8 f	78.6 ± 1.2 g	113.0 ± 0.3 g	124.4 ± 6.2 e	100.9 ± 1.7 c	7.0 ± 0.5 e

The values are the mean of three measurements with the standard deviation. Values followed by different letters are significantly different at  $p < 0.05$

Water, carbonate, sucrose, and lactic SRC values of each cultivar increased significantly ( $p < 0.05$ ) as disc mill time increased (Table 2).

The four solvents of SRC test had high and significant correlation with the damaged starch level (Table 3). The correlation observed with sodium carbonate SRC and water SRC values is expected, because these solvents measure the relative contributions of damaged starch [16]. The correlation between lactic acid SRC and damaged starch suggests that this parameter is not only influenced by glutenin, because the higher damaged starch level increased acid lactic retention.

AWRC is a test to select flours of good cookie quality. The flour fractions consisting of pentosans, proteins, glycoproteins, and damaged starch is thought to be responsible for the retention of alkaline water [17]. Accordingly, AWRC values were increased with damaged starch level (Table 2) and correlated with carbonate, sucrose and water SRC (Table 3). Excellent cookie-baking flours produce large cookie diameters and low AWRC values [16]. The flours under study showed a negative correlation between AWRC and cookie factor (Table 3) in concordance with several authors who have found a negative correlation between AWRC and cookie quality [12, 18, 19]. Consistent with this result, negative correlations between cookie factor

**Table 3** Correlation between SRC, AWRC, damaged starch and cookie factor of flour samples

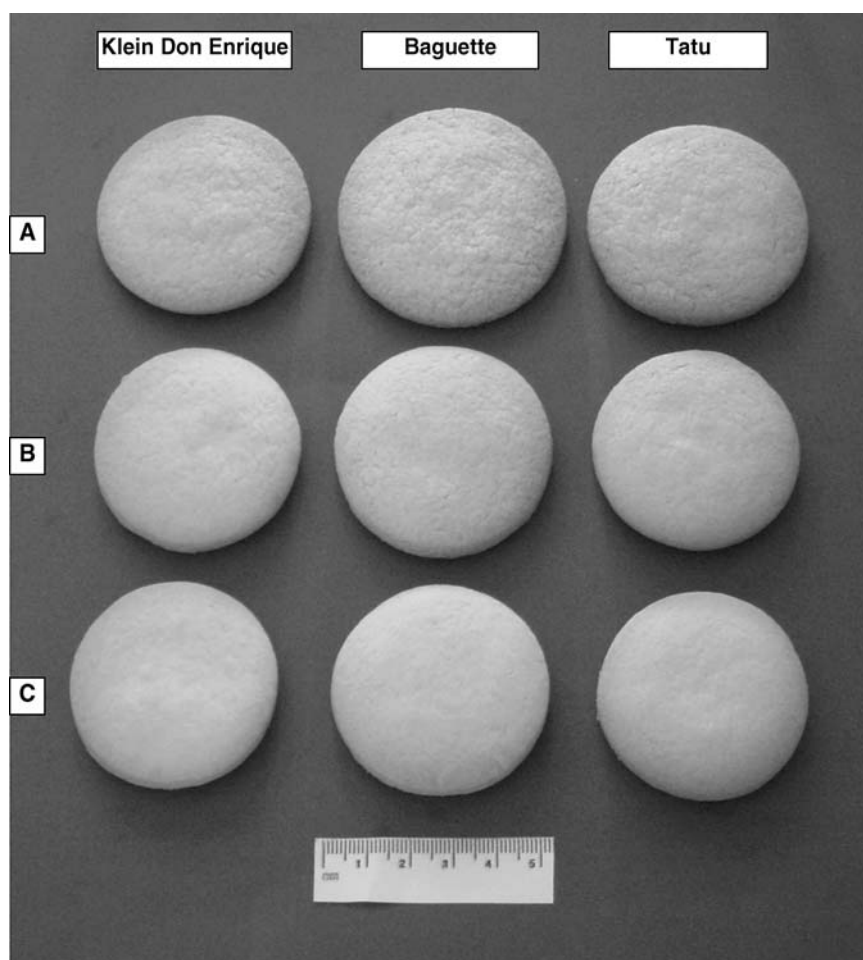
	H <sub>2</sub> O SRC	Na <sub>2</sub> CO <sub>3</sub> SRC	Sucrose SRC	Lactic acid SRC	AWRC	Damaged starch	Cookie factor
Na <sub>2</sub> CO <sub>3</sub> SRC	0.90**						
Sucrose SRC	0.84**	0.97**					
Lactic acid SRC	0.77**	0.44	0.36				
AWRC	0.83**	0.98**	0.97**	0.34			
Damaged starch	0.97**	0.82**	0.75**	0.82*	0.72**		
Cookie factor	-0.70**	-0.87**	-0.87**	-0.18	-0.89**	-0.67**	
Protein	0.19	-0.05	-0.06	0.58*	-0.10	0.26	0.21

\*Significant at  $p \leq 0.05$

\*\*Significant at  $p \leq 0.01$



**Fig. 1** Cookies elaborated with Baguette, Klein Don Enrique and Tatú flour containing different levels of damaged starch. **A** Lower level. **B** Middle level. **C** Higher level



with sodium carbonate, sucrose and water SRC were observed (Table 3). Guttieri et al. [20] also found that cookie diameter and top grain score correlated negatively with sodium carbonate, sucrose, and lactic SRC.

AWRC values and cookie factor were affected by the damaged starch level. The cookie diameter dramatically decreased with the higher level of damaged starch in both wheat and triticale flours (Fig. 1) because damaged starch absorbs more water than does undamaged starch. These results were in agreement with other authors [5, 6] who demonstrated that increased damaged starch decreases cookie diameter. In a previous work [7], we found correlation between damaged starch content and cookie factor ( $r = -0.52$ ) when flours obtained from triticale with six different tempering conditions were analyzed.

Good cookie and cracker flours hold water poorly [21]. The main hydrophilic components of a cookie formula are flour and sugar. Lower water absorption by flour provokes higher water absorption by sugar that increments syrup and decreases dough viscosity during baking; consequently dough could spread farther producing larger diameter cookies [22]. Flours with excessive water retention require increased baking times and increased energy costs in bakeries [20].

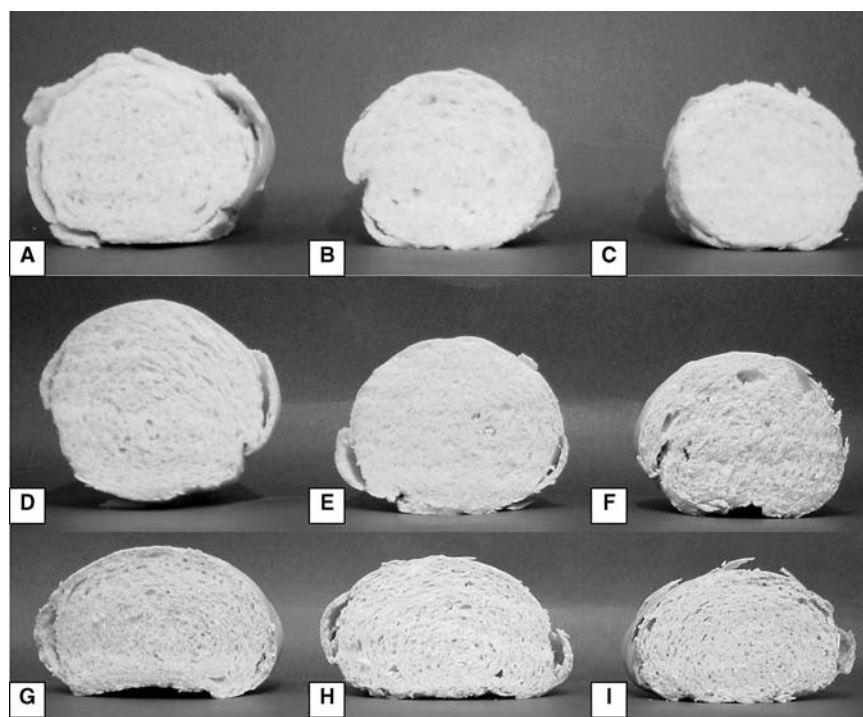
The best cookie factor was obtained from Baguette flour (Table 4) without disc mill time (Baguette 0 min), but it did not show the least damaged starch content. This fact evidenced the contribution of other flour components as protein in cookie quality. The influence of protein quality on cookie quality parameters has been studied. The protein to gluten formation, gliadin and glutenin, are functional during cookie baking, even though little, if any, of the gluten

**Table 4** The quality parameters of cookie and bread samples

Sample (min)	Cookie factor	Specific volume (cm <sup>3</sup> /g)	Crumb hardness (g)
Baguette (0)	6.4 ± 0.0 g	3.29 ± 0.03 e	542 ± 31 a,b
Baguette (2)	5.1 ± 0.1 d	2.68 ± 0.07 d	859 ± 56 c
Baguette (5)	4.6 ± 0.0 b,c	2.36 ± 0.02 a	890 ± 71 c
Klein DE (0)	5.9 ± 0.2 f	3.52 ± 0.09 f	567 ± 16 a,b
Klein DE (2)	4.9 ± 0.1 c,d	3.01 ± 0.04 d	854 ± 70 c
Klein DE (5)	4.5 ± 0.3 b	2.55 ± 0.05 b	1333 ± 68 e
Tatú (0)	5.5 ± 0.1 e	3.86 ± 0.09 f	501 ± 43 a
Tatú (3.5)	4.3 ± 0.3 a,b	3.36 ± 0.05 e	599 ± 19 b
Tatú (7)	4.1 ± 0.1 a	2.55 ± 0.05 b	1201 ± 62 d

The values are the mean of three measurements with the standard deviation. Values followed by different letters are significantly different at  $p < 0.05$

**Fig. 2** Breads elaborated with Baguette A–C, Klein Don Enrique D–F and Tatú G–I flour containing different levels of damaged starch. A, D, G Lower level. B, E, H Middle level. C, F, I Higher level



network is produced [9]. Bettge et al. [23] found that alveograph deformation work (W), a measure of gluten strength, was negatively correlated with the sugar snap cookie diameter; however, our results did not show correlation between cookie factor and protein content, lactic SRC, or SDS-SI.

The baking quality of wheat and triticale flours with different levels of damaged starch were evaluated (Fig. 2). The specific volume and the damaged starch level were strongly negatively correlated (Table 5). It is known that a high level of damaged starch reduces baking performance [24, 25]; however, SDS-SI, and wet gluten were independent of the damaged starch level (Table 5).

The effect of damaged starch on bread crumb hardness is shown in Table 4. The data showed an inverse relationship between bread loaf volume and crumb hardness (Table 5), probably because more entanglements and interactions occur between the more densely packed polymers in samples derived from low-volume breads.

**Table 5** Correlations between damaged starch, flour protein, SDS-SI, wet gluten, bread volume and crumb hardness of flour samples

	Damaged starch	Protein	SDS-SI	Wet gluten	Bread volume
Damaged starch	1				
Protein	0.33	1			
SDS-SI	0.39	0.62**	1		
Wet Gluten	-0.29	0.63**	0.40	1	
Bread volume	-0.95**	-0.19	-0.28	0.52	1
Hardness	0.82**	0.44	0.35	0.12	-0.81**

\*Significant at  $p \leq 0.05$

\*\*Significant at  $p \leq 0.01$

The potential protein alteration in flour during milling in disc mill was analyzed by means of wet gluten and multistacking gel electrophoresis. Wet gluten content did not show significant differences between different milling times for each flour (Table 1). SDS-soluble protein aggregates extracted from the flour samples were separated in four fractions (4, 6, 8 and 12% of polyacrylamide) on the basis of their migration in a multistacking polyacrylamide gel. Densitometric analysis did not show significant differences ( $p > 0.05$ ) among the electrophoretic patterns corresponding to samples milled for different periods of time. These results evidence that gluten protein functionality was not affected by milling time in disc mill and that gluten proteins were able to form aggregates stabilized by disulfide bonds. Consequently, the damaged starch increase was responsible for the negative effect on loaf volume. This effect might be explained by the competence for the water between damaged starch and protein that prevents optimum gluten formation during mixing, although another plausible explanation could be that damaged starch increments initial flour absorption and that after starch degradation (via enzymatic hydrolysis), dough consistence decreases, which in turns causes loss of gas-retention capacity.

## Conclusions

Several investigations were made in this subject; generally, researchers have utilized flours with different damaged starch content, obtained from reconstitution procedures or several wheat cultivars with different degrees of hardness. The aim of this work was to obtain flours with different damaged starch levels from each cultivar.

This study confirmed the influence of damaged starch content on flour quality. The degree of SRC and IRAA solvent absorption of wheat and triticale flours were significantly incremented by damaged starch content and cookie quality decreased as a consequence of damaged starch content increment.

Damaged starch reduced baking performance. Two possible explanations were suggested: (i) damaged starch increases initial water absorption and prevents optimum gluten formation during mixing; (ii) dough consistency decreases and loses gas-retention capacity after starch degradation during the fermentation phase.

Damaged starch content should be a parameter of relevance to optimize the process of cookie and bread manufacture.

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