

## RESEARCH ARTICLE

# Technological quality of wheat grains and flour as affected by nitrogen fertilization and weather conditions

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## ABSTRACT

The quality of grains and flour can be influenced by genetics, crop management, and the environment. The objective of this study was to evaluate the technological quality of wheat grains and flour influenced by the interaction among genotype, nitrogen (N) fertilization and weather conditions in different regions of wheat crop adaptation in southern Brazil, aiming to support farmers and the bakery industry sector in their decision-making processes. The experiment was carried out in three environments (Londrina in rainfed and irrigated conditions; and Ponta Grossa in rainfed) in a randomized block design with a  $10 \times 2$  factorial scheme. Ten wheat genotypes (BRS Sanhaço, BRS Graúna, BRS Gaivota, BRS Gralha-Azul, TBIO Sinuelo, TBIO Mestre, TBIO Sossego, TBIO Sintonia, TBIO Toruk, and Quartzo) and two N rates (40 and 120 kg ha<sup>-1</sup>) were assessed. The following technological quality analyses were performed in grains or flour: hectoliter weight, thousand-kernel weight, falling number, grain protein concentration, experimental flour extraction (i.e. flour yield), wet gluten concentration, and alveography. Increasing N rates from 40 to 120 kg ha<sup>-1</sup> enhanced the concentrations of grain protein and wet gluten. However, it did not influence dough gluten strength and the commercial classification of the flour. Nitrogen fertilization also influenced the flour yield, dough tenacity and elasticity index, depending on the genotype  $\times$  environment interaction. Environments with higher temperatures favored the flour yield and wet gluten concentration, while lower temperatures increased the thousand-kernel weight falling number. Water deficit increased the dough extensibility and grain protein concentration, whereas higher water availability favored the falling number, dough tenacity, and tenacity/extensibility ratio. Therefore, these outcomes are important drivers for farmers when choosing specific wheat genotypes for the environmental conditions of their farms, when they intend to meet the industrial requirements of mills and food companies that use wheat flour.

**Keywords:** Bakery; Environment; Genotypes; Gluten; Protein; *Triticum aestivum* L.

## INTRODUCTION

Wheat (*Triticum aestivum* L.) flour is a worldwide feedstock used by the food industry sector for the production of bread and other baked products such as noodles, pasta, cakes and biscuits, besides being an ingredient in many other processed food products. White flour is derived from the endosperm storage tissue of the grain and comprises mainly starch (about 75–85%) and protein (about 10%), which are the two major components that determine the flour quality, with the gluten proteins determining the viscoelastic properties of dough that underpin many of its uses, including bread making (Min et al., 2017).

For milling and bakery industries and other food companies, the technological quality of wheat grains and flour is mainly

based on protein concentration (particularly gluten), despite encompassing other traits such as  $\alpha$ -amylase activity (indirectly evaluated by the Hagberg falling number), dough alveography parameters (gluten strength, tenacity, and extensibility), hectoliter weight (HW) and thousand-kernel weight (TKW) (Guarienti et al., 2004; Hellemans et al., 2018; Xue et al., 2019).

The aforementioned technological quality traits are defined by wheat genotype, environment, and the genotype  $\times$  environment interaction, including soil and climate conditions, in addition to being influenced by crop management practices (e.g. fertilization, irrigation, plant growth regulators usage, and control of pests and diseases), harvest process, and drying and storage operations (Blumenthal et al., 1993; Guarienti et al., 2004; Yong et al.,

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2004; Franceschi et al., 2009; Denčić et al., 2011; Kaya and Akcura, 2014; Rozbicki et al., 2015; Ferreira et al., 2021; Rekowski et al., 2021; Faria et al., 2022). In this context, the supply of essential mineral nutrients by fertilizers is fundamental for the suitable growth and development of wheat plants, favoring the yield and quality of wheat grains and flour (Xue et al., 2016; Ma et al., 2019; Guerrini et al., 2020).

Among the nutrients supplied through fertilization, nitrogen (N) is emphasized as an element required in larger amounts by crops, especially by plants of the *Poaceae* family such as wheat (Bazzo et al., 2016; Souza et al., 2021; Marinho et al., 2022a). The importance of N for wheat crops is consensual, considering that N deficit results in smaller plants with fewer fertile tillers and lower N utilization efficiency, which leads to lower grain yield (Ferreira et al., 2021; Souza et al., 2021; Ferreira et al., 2022) and usually reduced grain protein concentration (Souza et al., 2019a; Xue et al., 2019; Lollato et al., 2021). In this context, well-nourished plants – particularly in relation to N – produce grains with better nutritional and morphological quality (i.e. grains with all structures well-developed and with suitable protein content) that are adequate for the consumer market and bakery industry (Pataco et al., 2015; Souza et al., 2019a; Lollato et al., 2021).

Guerrini et al. (2020), studying the effects of N fertilization in Italian wheat genotypes, verified that N fertilizer rates increased the grain protein concentration and dough gluten strength. The higher values for such wheat quality traits were observed in the treatments with higher N rate (135 kg ha<sup>-1</sup>) for all genotypes. Pinnow et al. (2013) also verified a positive effect of N fertilization on dough gluten strength and wet gluten concentration from harvested grains of a Brazilian wheat genotype (BRS Pardela). On the other hand, while investigating several Brazilian wheat genotypes, Schmidt et al. (2009) and Bassoi and Foloni (2012) did not observe any increase in dough gluten strength due to N fertilization, even with an increase in grain protein concentration. In this context, the increase of dough gluten strength due to increased grain protein concentration promoted by N fertilization, up to the measure that it modifies the commercial classification of wheat, is still uncertain (RCBPTT, 2016).

Ripe wheat grains present between 8% and 20% of proteins in their composition, including proteins that produce gluten, which is the main responsible for the rheological properties of the dough (Franceschi et al., 2009; Antunes et al., 2020). The concentrations of protein and gluten in the grains are determined by genetics and influenced by environmental conditions, such as temperature and availability of nutrients and water (Guarienti et al., 2004;

Rozbicki et al., 2015; Savill et al., 2018). For instance, during the grain-filling stage, the N accumulated in the plant biomass is preferentially remobilized to produce starch and, posteriorly, to increase the grain protein concentration (Silva et al., 2019; Lollato et al., 2021), which depends on the water supply throughout this period. Thus, in the condition of drought stress, the protein/starch ratio changes in the direction of protein, resulting in smaller wheat grains with a higher protein concentration (Blumenthal et al., 1993). Furthermore, some studies validated the influence of environmental temperature on the rheological properties of wheat dough from grains of Brazilian genotypes (Guarienti et al., 2004; Franceschi et al., 2009). Nevertheless, to the best of our knowledge, no scientific work evaluated the combined effects of N fertilization and weather conditions in tropical and subtropical regions on wheat plants concerning the technological quality of the produced grains and flour.

A better understanding of the combined effects of environmental factors and wheat crop management (e.g. choice of genotypes, irrigation, and N fertilization rates) is essential for the production of high-quality grains and flour. In this context, the objective of this study was to evaluate the technological quality of grains and flour influenced by the interaction among genotype, N fertilization and weather conditions in different regions of wheat crop adaptation in Brazil, aiming to support farmers and the bakery industry sector in their decision-making processes.

## MATERIAL AND METHODS

### Environmental characteristics of the experimental sites

The study was carried out in three environments: Ponta Grossa in rainfed situation (PG<sub>rainfed</sub>), and Londrina in rainfed (L<sub>rainfed</sub>) and irrigated (L<sub>irrig</sub>) conditions, whose areas are located in the regions I and III of wheat crop adaptation in Brazil, respectively (RCBPTT, 2016). In PG<sub>rainfed</sub> (Fig. 1a), the experiment was established in an experimental station of the National Soybean Research Center (*Embrapa Soja*) (25°09'31" S, 50°04'22" W, 886 m a. s. l.), where the soil is classified as Rhodic Hapludox (*Latosolo Vermelho distroférrico*), with clay texture (526 g kg<sup>-1</sup> clay and



**Fig 1.** Wheat plots located in experimental stations of the National Soybean Research Center (*Embrapa Soja*) in Ponta Grossa (a) and Londrina (b), PR, Brazil.

397 g kg<sup>-1</sup> sand). The regional climate, according to the Köppen classification, is mesothermal humid subtropical (Cfb), with mild summers, average annual temperature and precipitation of 17.5 °C and 1,495 mm, evenly distributed rainfalls and frequent frosts. In Londrina (Fig. 1b), the experiments in L<sub>rainfed</sub> and L<sub>irrig</sub> were carried out side by side (separated by 10 m between edges) in other experimental station of *Embrapa Soja* (23°11'37" S, 51°11'03" W, 628 m a. s. l.), where the soil is classified as Rhodic Eutrudox (*Latosolo Vermelho eutrófico*), with clay texture (732 g kg<sup>-1</sup> clay and 107 g kg<sup>-1</sup> sand). The regional climate is humid subtropical (Cfa), with warm and rainy summer, average annual temperature and precipitation of 21.2 °C and 1,392 mm, sparse frosts, and no defined dry season.

### Experimental design and establishment of the treatments

The experiments were carried out in a randomized complete block design, with three replications, using a 10 × 2 factorial arrangement with ten wheat genotypes (cultivars: BRS Sanhaço, BRS Graúna, BRS Gaivota, BRS Gralha-Azul, TBIO Sinuelo, TBIO Mestre, TBIO Sossego, TBIO Sintonia, TBIO Toruk, and Quartzo; henceforth referred to as Sanhaço, Graúna, Gaivota, Gralha-Azul, Sinuelo, Mestre, Sossego, Sintonia, Toruk, and Quartzo, respectively) and two N rates (40 and 120 kg ha<sup>-1</sup>) applied in topdressing at the beginning of plant tillering. The N rates were determined according to Foloni et al. (2016) and RCBPTT (2016).

The genotypes have a predominantly medium development cycle, except Sintonia and Quartzo that have an early cycle, and Sinuelo which cycle ranges from medium to late. Regarding the commercial classification, Sintonia is classified as improver wheat flour, and Mestre and Toruk as bread/improver wheat. The remaining genotypes are considered bread wheat (Basso and Foloni, 2015a, b; Basso and Foloni, 2016; Basso et al., 2017; RCBPTT, 2016).

Each experimental plot comprised nine rows with 6 m length spaced by 0.18 m (9.6 m<sup>2</sup>). Wheat crop was sown in the 2016 crop season (L<sub>rainfed</sub> and L<sub>irrig</sub> = April 29; and PG<sub>rainfed</sub> = June 9) on soybean straw using a no-till system and a sowing density of 350 viable seeds m<sup>-2</sup>. Base fertilization was performed into the seeding furrow (at 4 cm depth) with 250 kg ha<sup>-1</sup> of the formulated fertilizer 08–28–16 (N–P<sub>2</sub>O–K<sub>2</sub>O) (RCBPTT, 2016; Foloni et al., 2016). At the beginning of plant tillering, the topdressing fertilization used ammonium nitrate (32% N) as the source of N.

In the environment of Londrina with water supplementation (L<sub>irrig</sub>), irrigations were performed according to soil moisture, indirectly measured by a set of tensiometers. Five 25-mm irrigations were applied throughout the crop

cycle using a self-propelled sprinkler irrigation system (Fig. 2). The water balance was calculated according to Thornthwaite and Mather (1955) method.

Phytosanitary management and other agronomic practices were conducted according to the recommendations of the Brazilian Commission of Wheat and Triticale Research (*Comissão Brasileira de Pesquisa de Trigo e Triticale* – RCBPTT, 2016).

The grain harvest (L<sub>rainfed</sub> and L<sub>irrig</sub> = September 16; and PG<sub>rainfed</sub> = November 3) was performed in the seven central rows with 6 m length using a self-propelled combine developed for small-plot of cereals (Fig. 3). Immediately after harvesting, 2 kg of grains from each experimental plot were sampled for wheat technological quality analyses. The samples were stored in a cold chamber (7–10 °C) until the laboratory analyses.

### Wheat technological quality analyses of grains and flour

The following technological quality parameters were obtained using specific instruments for analyses of wheat grains and flour (Fig. 4):

Hectoliter weight: corresponds to the mass of 100-liter grain (kg hL<sup>-1</sup>), which was obtained following the



**Fig 2.** Self-propelled sprinkler irrigation system used to apply five 25-mm irrigations throughout the crop cycle in the environment of Londrina with water supplementation (L<sub>irrig</sub>).



**Fig 3.** Wheat experimental plots at ripening stage (a) in the environment of Londrina, which grain harvest was performed by a self-propelled combine developed for small-plot of cereals (b).





**Fig 4.** Scientific instruments used to analyze the technological quality of wheat grains and flour: Test weight scale (Dalle Molle®), for hectoliter weight analysis (a); semi-analytical balance (Ohaus®), for the thousand-kernel weight determination (b); Quadrumat Senior mill (Brabender®), for experimental flour extraction (c); Falling Number System (Perten Instruments®), for analysis of  $\alpha$ -amylase enzyme activity (d); Chopin Alveograph (Chopin Technologies®), for evaluation of dough gluten strength, tenacity, extensibility, and elasticity index (e); NIR XDS analyzer (Foss®), for grain protein concentration (f); and Glutomatic system (Perten Instruments®), for wet gluten evaluation (g).

methodology proposed by the American Association of Cereal Chemists (AACC, 1999).

Thousand-kernel weight: obtained by counting and weighing eight repetitions of 100 wheat grains per plot. Afterwards, the TKW was determined following the Rules for Seed Analysis (*Regras para Análise de Sementes – RAS*; Brasil, 2009).

Falling number: determined in milled grains passed through a 0.8 mm sieve (Perten® LabMill), using the Falling Number Perten Instruments®, following the method 56-81.03 of the AACC (1999).

Experimental flour extraction (i.e. flour yield): determined with an experimental mill (Quadrumat Senior mill, Brabender®), according to the method 26-10.02 of the AACC (1999).

Alveography: determined with a Chopin Alveograph, according to the method 54-30.02 of the AACC (1999). The following dough parameters were evaluated: gluten strength, tenacity (P), extensibility (L), the ratio between tenacity and extensibility (P/L), and elasticity index.

Grain protein concentration: determined by near-infrared reflectance (NIR) using the NIR XDS analyzer (Foss®), according to the method 39-10.01 of the AACC (1999).

Wet gluten concentration: determined following the method 38-12.02 of the AACC (2000), using the Glutomatic system (Perten®).

### Statistical analysis

The statistical analyses were performed using the R statistical software (R Core Team, 2020), individually for

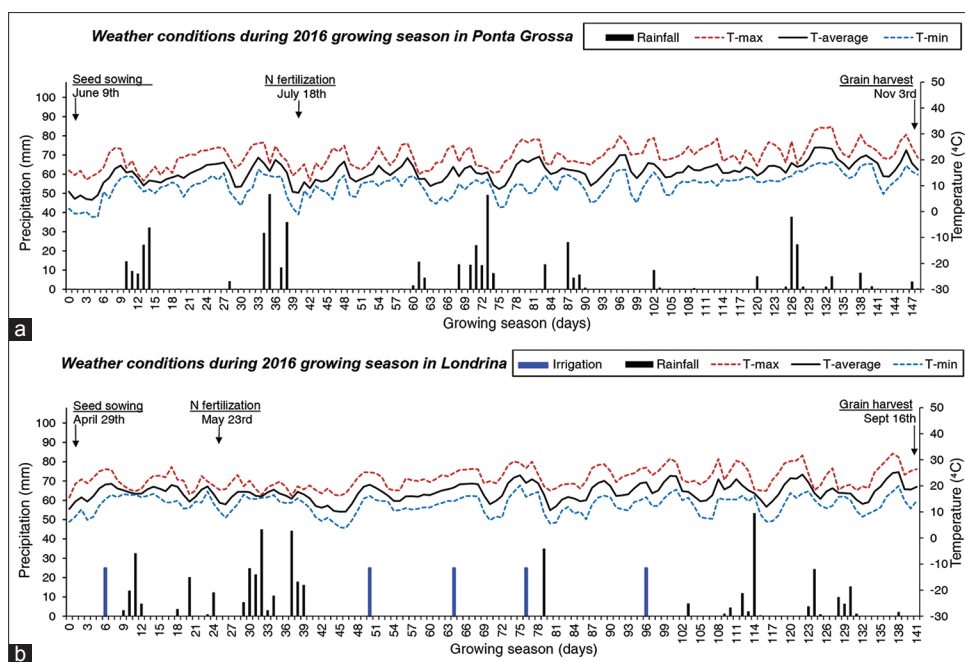
each environment. The Shapiro-Wilk's test evaluated the model's assumptions regarding the normality of residuals, and the Bartlett's test assessed the variance homogeneity. Then, the analysis of variance (ANOVA) was employed. When the ANOVA resulted in a significant  $P$ -value ( $P \leq 0.05$ ), the means of the two treatments from the factor 'N rate' were compared by the Student-Newman-Keuls' test ( $P < 0.05$ ). The means from the qualitative factor 'wheat genotype' were grouped by the Scott-Knott clustering algorithm ( $P < 0.05$ ). The variables evaluated in the three environments were combined and evaluated for Pearson correlation analysis.

## RESULTS

### Weather conditions during the wheat-growing season

The wheat-growing season in PG<sub>rainfed</sub> had 148 days with means of average, maximum, and minimum temperatures of  $15.4 \pm 3.9$ ,  $21.6 \pm 4.5$ , and  $10.3 \pm 4.5$  °C (mean  $\pm$  standard deviation), and total precipitation of 520.4 mm (Fig. 5a). The rainfall distribution was relatively regular, resulting in an accumulated water deficit of 5.7 mm. The wheat sowing was followed by an eight-day drought period (with only 0.2 mm of rain). After that, there was 87.8 mm throughout five consecutive days. The topdressing N fertilization was carried out on the 40<sup>th</sup> day of the cycle, preceded by 125.4 mm of rainfall in six successive days. After this fertilization, there was a 20-day drought period (with 2.8 mm of rainfall). Throughout the grain-filling period (from flowering to ripening), there was an accumulated precipitation of 95 mm and means of average, maximum, and minimum temperatures of  $18.0 \pm 3.1$ ,  $24.2 \pm 3.9$ , and  $13.7 \pm 3.0$  °C.

In Londrina, the wheat-growing season had a cycle of 141 days, with means of average, maximum, and minimum



**Fig 5.** Precipitation (rainfall and irrigation), maximum (T-max), average (T-average), and minimum (T-min) temperatures during the growing season in Ponta Grossa (from June 9<sup>th</sup> to November 3<sup>rd</sup> = 148 days; graphic a) and Londrina (from April 29<sup>th</sup> to September 16<sup>th</sup> = 141 days; graphic b). Note: The treatments in Londrina with water surplus received five 25-mm irrigations (at 6<sup>th</sup>, 50<sup>th</sup>, 64<sup>th</sup>, 76<sup>th</sup>, and 96<sup>th</sup> days). Dates of major field operations are also provided in the figure.

temperatures of  $17.6 \pm 3.2$ ,  $22.9 \pm 3.8$ , and  $12.9 \pm 3.5$  °C, and total precipitation of 465.3 mm (Fig. 5b). Furthermore, the treatments in  $L_{irrig}$  received five 25-mm irrigations (at 6<sup>th</sup>, 50<sup>th</sup>, 64<sup>th</sup>, 76<sup>th</sup>, and 96<sup>th</sup> days of the growing cycle), raising the total water supply to 590.3 mm and reducing the accumulated water deficit from 31.4 mm ( $L_{rainfed}$ ) to 4.3 mm ( $L_{irrig}$ ). After wheat sowing, there was a seven-day drought period, followed by a four-day rainy period (accumulation of 55.4 mm). In addition, there were two outstanding drought periods: the first lasting 39 days (without rainfall from 40<sup>th</sup> to 78<sup>th</sup> day) and the second with 32 days (from 80<sup>th</sup> to 111<sup>th</sup> day with an accumulated precipitation of 12.4 mm). The topdressing N fertilization was carried out on the 25<sup>th</sup> day, preceded by 33.7 mm of rainfall (in five consecutive days), and received another 190.8 mm in the 14 days following the fertilization. Throughout the grain-filling period, there was an accumulated precipitation of 95 mm (rainfall + irrigation) and means of average, maximum, and minimum temperatures of  $18.7 \pm 3.4$ ,  $24.6 \pm 4.3$ , and  $13.4 \pm 3.5$  °C.

#### Wheat technological quality of grains and flour

The HW had high environmental adaptability, with averages of 77.3, 78.7, and 78.3 kg hL<sup>-1</sup> in  $PG_{rainfed}$ ,  $L_{rainfed}$ , and  $L_{irrig}$ , respectively (Table 1). However, the HW was influenced by the main factor ‘genotype’ in all three environments. In  $PG_{rainfed}$ , the genotypes Gralha-Azul, Sanhaço, Sintonia, and Gaivotá had higher values of HW, and Toruk had the lower one. On the other hand, in  $L_{rainfed}$ , Sinuelo presented

the higher HW, followed by Sanhaço, Sossego, and Toruk. In contrast, the genotypes Graúna and Sintonia, followed by Mestre, had lower values of HW. In  $L_{irrig}$ , the behavior of the genotypes was similar to that of  $L_{rainfed}$ .

The TKW averaged 38.5, 34.9, and 34.4 g in  $PG_{rainfed}$ ,  $L_{rainfed}$ , and  $L_{irrig}$  (Table 1). Therefore, it was 11.3% high in  $PG_{rainfed}$  compared to Londrina (average of  $L_{rainfed}$  and  $L_{irrig}$ ). The TKW evaluated in  $L_{rainfed}$  and  $L_{irrig}$  was influenced only by genotype (Table 1). In  $L_{rainfed}$ , the genotypes Toruk, Quartzo, Sanhaço, Gralha-Azul, and Graúna had higher TKW as compared to the other cultivars. In  $L_{irrig}$ , the genotypes Graúna, Sanhaço, Toruk, and Gaivotá achieved the higher TKW.

The falling number averaged 409, 327, and 390 s in  $PG_{rainfed}$ ,  $L_{rainfed}$ , and  $L_{irrig}$  (Table 1). Therefore, it was 25.3% higher in  $PG_{rainfed}$  than that in  $L_{rainfed}$ . Furthermore, irrigation increased the falling number by 19.3% in Londrina. The factor genotype influenced the falling number in the three environments (Table 1). In  $PG_{rainfed}$ , most genotypes (except Toruk and Sinuelo) presented high values of falling number (i.e. low activity of  $\alpha$ -amylase enzyme). In  $L_{rainfed}$ , Graúna, Sintonia, Toruk, and Gaivotá had lower values of falling number (Table 1). In  $L_{irrig}$ , Sintonia and Gaivotá presented the lower falling number, followed by the genotypes Toruk, Quartzo and Graúna, which showed intermediate results.

**Table 1: Summary of the analysis of variance and means of hectoliter weight, thousand-kernel weight, dough falling number, and experimental flour extraction from grains of ten wheat genotypes cultivated with two nitrogen (N) rates in three environments: Ponta Grossa in rainfed (PG<sub>rainfed</sub>), and Londrina in rainfed (L<sub>rainfed</sub>) and irrigated (L<sub>irrig</sub>) conditions**

Source of variation	DF	Mean square											
		Hectoliter weight			Thousand-kernel weight			Falling number			Experimental flour extraction		
		PG <sub>rainfed</sub>	L <sub>rainfed</sub>	L <sub>irrig</sub>	PG <sub>rainfed</sub>	L <sub>rainfed</sub>	L <sub>irrig</sub>	PG <sub>rainfed</sub>	L <sub>rainfed</sub>	L <sub>irrig</sub>	PG <sub>rainfed</sub>	L <sub>rainfed</sub>	L <sub>irrig</sub>
Block	2	2.95	0.58	4.27	3.18	3.38	1.15	3164	2890	1731	0.98	2.13	7.27
Genotype (G)	9	23.9***	8.91***	4.87**	5.23 <sup>ns</sup>	13.04***	9.01**	15405***	7143***	18021***	24.8***	21.4***	40.4***
N rate (N)	1	0.38 <sup>ns</sup>	0.04 <sup>ns</sup>	0.00 <sup>ns</sup>	5.46 <sup>ns</sup>	3.31 <sup>ns</sup>	0.17 <sup>ns</sup>	1050 <sup>ns</sup>	459 <sup>ns</sup>	516 <sup>ns</sup>	9.26 <sup>ns</sup>	11.5**	24.4 <sup>ns</sup>
G x N	9	1.04 <sup>ns</sup>	0.51 <sup>ns</sup>	0.77 <sup>ns</sup>	1.95 <sup>ns</sup>	3.02 <sup>ns</sup>	1.40 <sup>ns</sup>	2854 <sup>ns</sup>	599 <sup>ns</sup>	2837 <sup>ns</sup>	3.42 <sup>ns</sup>	1.76 <sup>ns</sup>	1.15 <sup>ns</sup>
Error	38	1.95	0.27	1.31	3.85	2.80	2.54	2151	1546	1945	4.00	1.06	6.15
CV (%)		1.81	0.66	1.46	5.09	4.79	4.64	11.3	12.0	11.3	3.40	1.65	4.04

Factor	Treatment	Hectoliter weight (kg hL <sup>-1</sup> )			Thousand-kernel weight (g)			Falling number (s)			Experimental flour extraction (%)		
		PG <sub>rainfed</sub>	L <sub>rainfed</sub>	L <sub>irrig</sub>	PG <sub>rainfed</sub>	L <sub>rainfed</sub>	L <sub>irrig</sub>	PG <sub>rainfed</sub>	L <sub>rainfed</sub>	L <sub>irrig</sub>	PG <sub>rainfed</sub>	L <sub>rainfed</sub>	L <sub>irrig</sub>
		PG <sub>rainfed</sub>	L <sub>rainfed</sub>	L <sub>irrig</sub>	PG <sub>rainfed</sub>	L <sub>rainfed</sub>	L <sub>irrig</sub>	PG <sub>rainfed</sub>	L <sub>rainfed</sub>	L <sub>irrig</sub>	PG <sub>rainfed</sub>	L <sub>rainfed</sub>	L <sub>irrig</sub>
Genotype (G)	Sanhaço	79.2 <sup>a</sup>	79.8 <sup>a</sup>	79.4 <sup>a</sup>	39.5	36.2 <sup>a</sup>	35.3 <sup>a</sup>	433 <sup>a</sup>	336 <sup>a</sup>	440 <sup>a</sup>	57.3 <sup>b</sup>	60.4 <sup>d</sup>	60.6 <sup>c</sup>
	Graúna	77.6 <sup>b</sup>	76.7 <sup>d</sup>	77.2 <sup>b</sup>	39.9	35.6 <sup>a</sup>	36.8 <sup>a</sup>	402 <sup>a</sup>	281 <sup>b</sup>	377 <sup>b</sup>	56.0 <sup>b</sup>	62.3 <sup>c</sup>	59.5 <sup>d</sup>
	Gaivota	78.2 <sup>a</sup>	78.9 <sup>b</sup>	79.4 <sup>a</sup>	38.1	33.0 <sup>b</sup>	34.8 <sup>a</sup>	438 <sup>a</sup>	317 <sup>b</sup>	322 <sup>c</sup>	60.1 <sup>a</sup>	62.1 <sup>c</sup>	62.5 <sup>b</sup>
	Gralha-Azul	79.6 <sup>a</sup>	78.7 <sup>b</sup>	77.7 <sup>b</sup>	38.3	35.7 <sup>a</sup>	34.1 <sup>b</sup>	448 <sup>a</sup>	364 <sup>a</sup>	416 <sup>a</sup>	57.3 <sup>b</sup>	60.2 <sup>d</sup>	57.7 <sup>d</sup>
	Sinuelo	76.4 <sup>b</sup>	80.0 <sup>a</sup>	79.3 <sup>a</sup>	38.0	33.1 <sup>b</sup>	33.5 <sup>b</sup>	350 <sup>b</sup>	329 <sup>a</sup>	402 <sup>a</sup>	61.2 <sup>a</sup>	63.9 <sup>b</sup>	64.3 <sup>a</sup>
	Mestre	77.1 <sup>b</sup>	77.6 <sup>c</sup>	77.4 <sup>b</sup>	39.1	33.7 <sup>b</sup>	32.8 <sup>b</sup>	455 <sup>a</sup>	336 <sup>a</sup>	457 <sup>a</sup>	58.3 <sup>b</sup>	60.6 <sup>d</sup>	58.3 <sup>d</sup>
	Sossego	76.7 <sup>b</sup>	79.5 <sup>a</sup>	78.6 <sup>a</sup>	38.3	34.0 <sup>b</sup>	33.4 <sup>b</sup>	462 <sup>a</sup>	379 <sup>a</sup>	457 <sup>a</sup>	58.7 <sup>b</sup>	61.2 <sup>d</sup>	61.0 <sup>c</sup>
	Sintonia	78.6 <sup>a</sup>	76.8 <sup>d</sup>	77.1 <sup>b</sup>	37.5	34.3 <sup>b</sup>	33.2 <sup>b</sup>	412 <sup>a</sup>	269 <sup>b</sup>	298 <sup>c</sup>	56.3 <sup>b</sup>	64.0 <sup>b</sup>	61.5 <sup>c</sup>
	Toruk	72.5 <sup>c</sup>	79.5 <sup>a</sup>	78.8 <sup>a</sup>	37.2	37.2 <sup>a</sup>	35.3 <sup>a</sup>	302 <sup>b</sup>	308 <sup>b</sup>	358 <sup>b</sup>	61.2 <sup>a</sup>	65.9 <sup>a</sup>	66.2 <sup>a</sup>
	Quartzo	77.2 <sup>b</sup>	79.1 <sup>b</sup>	78.3 <sup>a</sup>	39.7	36.4 <sup>a</sup>	34.4 <sup>b</sup>	391 <sup>a</sup>	349 <sup>a</sup>	370 <sup>b</sup>	61.1 <sup>a</sup>	63.5 <sup>b</sup>	62.1 <sup>b</sup>
N rate (N)	40	77.4	78.6	78.3	38.8	34.7	34.3	414	324	387	58.3	62.0 <sup>b</sup>	60.7
	120	77.2	78.7	78.3	38.2	35.1	34.4	405	330	393	59.1	62.8 <sup>a</sup>	62.0

DF = degrees of freedom; CV = coefficient of variation. ns, \*, \*\*, and \*\*\* = not significant, significant at  $P \leq 0.05$ ,  $P \leq 0.01$ , and  $P \leq 0.001$ , respectively, by the F test. Means followed by the same letters in the column, for the factors genotype or N rates, belong to the same cluster by the Scott-Knott's test ( $P \leq 0.05$ ) or by the Student-Newman-Keuls' test ( $P \leq 0.05$ ), respectively.

The experimental flour extraction averaged 58.7%, 62.4%, and 61.4% in PG<sub>rainfed</sub>, L<sub>rainfed</sub>, and L<sub>irrig</sub> (Table 1). Thus, it was 3.2 percentage points higher in Londrina (average of L<sub>rainfed</sub> and L<sub>irrig</sub>) than that in PG<sub>rainfed</sub>. Moreover, it was influenced by genotype in all environments; and increasing N rates from 40 to 120 kg ha<sup>-1</sup> increased the flour extraction by 0.8 percentage points in L<sub>rainfed</sub>. The genotype Toruk showed high flour extraction in the three environments, being the best genotype for this trait in L<sub>rainfed</sub>. In L<sub>irrig</sub>, the genotype Sinuelo also had high flour extraction. Finally, in PG<sub>rainfed</sub>, the genotypes Sinuelo, Quartzo, and Gaivota also stood out positively for this wheat dough trait. Concerning N rates, the application of 120 kg ha<sup>-1</sup> favored the increase of flour extraction only in L<sub>rainfed</sub>.

The dough tenacity averaged 77.8, 66.9, and 75 mm in PG<sub>rainfed</sub>, L<sub>rainfed</sub>, and L<sub>irrig</sub> (Table 2). Thus, it was 12.4% lower in the environment with the lowest water availability (i.e. L<sub>rainfed</sub>) as compared to the average of the other environments. In PG<sub>rainfed</sub>, the rate of 40 kg ha<sup>-1</sup> N increased the dough tenacity as compared to that of 120 kg ha<sup>-1</sup> N. In the three environments, the genotype Gralha-Azul had the

highest values of dough tenacity, whereas Sinuelo, Quartzo, and Toruk had the lowest ones (Table 2).

The dough extensibility averaged 70.5, 90, and 82.8 mm in PG<sub>rainfed</sub>, L<sub>rainfed</sub>, and L<sub>irrig</sub> (Table 2). Therefore, it was 19.4% and 8.7% higher in L<sub>rainfed</sub> than that in PG<sub>rainfed</sub> and L<sub>irrig</sub>, respectively, i.e. the dough extensibility was favored by environmental conditions of lower water availability. This alveography parameter was influenced by the genotype × N rate interaction in L<sub>rainfed</sub>, and by the genotype factor in PG<sub>rainfed</sub> and L<sub>irrig</sub> (Table 2). Thus, in PG<sub>rainfed</sub>, the genotypes Gaivota, Sintonia, Mestre, and Graúna had higher dough extensibility. In L<sub>irrig</sub>, Toruk, Sintonia, Gaivota, Sinuelo, and Graúna also produced flour with high dough extensibility. In L<sub>rainfed</sub>, increasing N rates from 40 to 120 kg ha<sup>-1</sup> increased the dough extensibility of the genotype Sinuelo (Table 4). But an opposite effect occurred with Toruk. Furthermore, the genotypes Sintonia, Toruk, Graúna, and Quartzo presented higher dough extensibility values within the N rate of 40 kg ha<sup>-1</sup> (Table 4).

The P/L ratio of the alveography averaged 1.24, 0.77, and 0.98 in PG<sub>rainfed</sub>, L<sub>rainfed</sub>, and L<sub>irrig</sub> (Table 2). Therefore, it was 30.6% lower in L<sub>rainfed</sub> (i.e. the environment with



**Table 2: Summary of the analysis of variance and means of dough tenacity (P), extensibility (L), and the ratio between tenacity and extensibility(P/L) from grains of ten wheat genotypes cultivated with two nitrogen (N) rates in three environments: Ponta Grossa in rainfed (PG<sub>rainfed</sub>), and Londrina in rainfed (L<sub>rainfed</sub>) and irrigated (L<sub>irrig</sub>) conditions**

Source of variation	DF	Mean Square								
		Dough tenacity			Dough extensibility			P/L ratio		
		PG <sub>rainfed</sub>	L <sub>rainfed</sub>	L <sub>irrig</sub>	PG <sub>rainfed</sub>	L <sub>rainfed</sub>	L <sub>irrig</sub>	PG <sub>rainfed</sub>	L <sub>rainfed</sub>	L <sub>irrig</sub>
Block	2	104	36.5	242	263	257	442	0.14	0.05	0.29
Genotype (G)	9	2467***	701***	2325***	1001*	391***	776***	0.91**	0.20***	0.93***
N rate (N)	1	1067*	77.1 <sup>ns</sup>	79.4 <sup>ns</sup>	32.3 <sup>ns</sup>	0.60 <sup>ns</sup>	91.3 <sup>ns</sup>	0.30 <sup>ns</sup>	0.01 <sup>ns</sup>	0.00 <sup>ns</sup>
G x N	9	185 <sup>ns</sup>	80.0 <sup>ns</sup>	158 <sup>ns</sup>	230 <sup>ns</sup>	199*	25.2 <sup>ns</sup>	0.28 <sup>ns</sup>	0.04 <sup>ns</sup>	0.04 <sup>ns</sup>
Error	38	233	82.6	142	463	75.2	187	0.29	0.02	0.10
CV (%)		19.6	13.6	15.9	30.5	9.64	16.5	43.6	18.5	32.9

Factor	Treatment	Dough tenacity (mm)			Dough extensibility (mm)			P/L ratio		
		PG <sub>rainfed</sub>	L <sub>rainfed</sub>	L <sub>irrig</sub>	PG <sub>rainfed</sub>	L <sub>rainfed</sub>	L <sub>irrig</sub>	PG <sub>rainfed</sub>	L <sub>rainfed</sub>	L <sub>irrig</sub>
Genotype (G)	Sanhaço	88.8 <sup>b</sup>	72.2 <sup>b</sup>	90.5 <sup>b</sup>	69.2 <sup>b</sup>	90.3	72.7 <sup>b</sup>	1.45 <sup>a</sup>	0.80 <sup>b</sup>	1.31 <sup>b</sup>
	Graúna	84.3 <sup>b</sup>	62.7 <sup>c</sup>	65.8 <sup>c</sup>	79.8 <sup>a</sup>	89.2	86.7 <sup>a</sup>	1.11 <sup>b</sup>	0.71 <sup>b</sup>	0.78 <sup>c</sup>
	Gaivota	62.3 <sup>c</sup>	68.5 <sup>b</sup>	72.3 <sup>c</sup>	87.2 <sup>a</sup>	90.8	91.3 <sup>a</sup>	0.73 <sup>b</sup>	0.75 <sup>b</sup>	0.80 <sup>c</sup>
	Gralha-Azul	113 <sup>a</sup>	86.3 <sup>a</sup>	114 <sup>a</sup>	61.7 <sup>b</sup>	87.8	70.7 <sup>b</sup>	1.90 <sup>a</sup>	1.01 <sup>a</sup>	1.71 <sup>a</sup>
	Sinuelo	51.2 <sup>c</sup>	56.3 <sup>c</sup>	56.7 <sup>c</sup>	59.2 <sup>b</sup>	93.8	88.3 <sup>a</sup>	1.00 <sup>b</sup>	0.61 <sup>c</sup>	0.67 <sup>c</sup>
	Mestre	91.7 <sup>b</sup>	71.7 <sup>b</sup>	88.8 <sup>b</sup>	83.3 <sup>a</sup>	95.5	81.5 <sup>b</sup>	1.16 <sup>b</sup>	0.76 <sup>b</sup>	1.16 <sup>b</sup>
	Sossego	94.7 <sup>b</sup>	77.0 <sup>b</sup>	88.7 <sup>b</sup>	61.5 <sup>b</sup>	70.2	63.3 <sup>b</sup>	1.83 <sup>a</sup>	1.12 <sup>a</sup>	1.42 <sup>b</sup>
	Sintonia	78.8 <sup>b</sup>	47.8 <sup>d</sup>	59.0 <sup>c</sup>	84.3 <sup>a</sup>	101.8	93.3 <sup>a</sup>	1.00 <sup>b</sup>	0.47 <sup>c</sup>	0.65 <sup>c</sup>
	Toruk	57.5 <sup>c</sup>	64.3 <sup>c</sup>	51.2 <sup>c</sup>	48.3 <sup>b</sup>	89.2	100 <sup>a</sup>	1.32 <sup>b</sup>	0.76 <sup>b</sup>	0.52 <sup>c</sup>
N rate (N)	Quartzo	55.8 <sup>c</sup>	62.5 <sup>c</sup>	63.3 <sup>c</sup>	70.8 <sup>b</sup>	91.0	79.8 <sup>b</sup>	0.87 <sup>b</sup>	0.69 <sup>b</sup>	0.80 <sup>c</sup>
	40	82.0 <sup>a</sup>	65.8	73.8	71.3	90.1	81.5	1.31	0.75	0.97
	120	73.6 <sup>b</sup>	68.1	76.1	69.8	89.9	84.0	1.17	0.78	0.99

DF = degrees of freedom; CV = coefficient of variation. ns, \*, \*\*, and \*\*\* = not significant, significant at  $P \leq 0.05$ ,  $P \leq 0.01$ , and  $P \leq 0.001$ , respectively, by the F test. Means followed by the same letters in the column, for the factors genotype or N rates, belong to the same cluster by the Scott-Knott's test ( $P \leq 0.05$ ) or by the Student-Newman-Keuls' test ( $P \leq 0.05$ ), respectively.

the lowest water availability) as compared to the average of the other environments. The P/L ratio was influenced only by the factor genotype in the three environments. Gralha-Azul genotype presented the highest P/L ratio in the three environments, together with Sanhaço and Sossego in PG<sub>rainfed</sub>, and Sossego in L<sub>rainfed</sub>.

The grain protein concentration averaged 13.5%, 14.7%, and 13.6% in PG<sub>rainfed</sub>, L<sub>rainfed</sub>, and L<sub>irrig</sub>, i.e. it was higher in the environment with lower water availability (Table 3). The genotype and N rate factors influenced this trait in all environments (Table 3). Gaivota presented high grain protein concentration in the three environments, particularly in L<sub>rainfed</sub>. In L<sub>irrig</sub>, the genotype Mestre also showed high protein concentration. In PG<sub>rainfed</sub>, besides both genotypes aforementioned, Graúna and Toruk also stood out by the high grain protein concentration. In contrast, the genotype Sinuelo produced grains with the lowest protein concentration in all environments, whereas Quartzo also had lower values for this trait in PG<sub>rainfed</sub> and L<sub>irrig</sub>. Increasing N rates from 40 to 120 kg ha<sup>-1</sup> enhanced the grain protein concentration in all environments (Table 3). Nevertheless, such increases were of low magnitude (lower than 1%).

The wet gluten concentration averaged 24.1%, 30.1%, and 28.5% in PG<sub>rainfed</sub>, L<sub>rainfed</sub>, and L<sub>irrig</sub> (Table 3). Thus, it was lower in

the environment with lower temperatures, i.e. PG<sub>rainfed</sub> (Fig. 5a). In L<sub>rainfed</sub> and L<sub>irrig</sub>, the genotype Gaivota produced grains with a higher gluten concentration (Table 3). On the other hand, the lowest values of grain gluten concentration were verified for the genotypes Sinuelo and Quartzo in L<sub>rainfed</sub> and L<sub>irrig</sub>, Sossego in L<sub>rainfed</sub>, and Sintonia in L<sub>irrig</sub>. Increasing N rates from 40 to 120 kg ha<sup>-1</sup> increased the gluten concentration in L<sub>rainfed</sub> and L<sub>irrig</sub> (Table 3). In PG<sub>rainfed</sub>, the wet gluten concentration was influenced by genotype, N rate, and by the interaction between these factors (Table 3). In this environment, the gluten concentration of the genotype Sinuelo was reduced by increasing N rates (Table 4). Within 40 kg ha<sup>-1</sup> N, the genotypes Sanhaço, Graúna, Gaivota, Sinuelo, and Mestre produced grains with higher gluten concentration; whereas considering 120 kg ha<sup>-1</sup> N, Sanhaço, Graúna, Mestre, and Sintonia stood out positively.

The dough gluten strength had high environmental adaptability, with averages of with averages of 211, 212, and 211.5 × 10<sup>-4</sup> J in PG<sub>rainfed</sub>, L<sub>rainfed</sub>, and L<sub>irrig</sub> (Table 3). Such alveography parameter was influenced only by the factor genotype in PG<sub>rainfed</sub> and L<sub>irrig</sub> (Table 3). In PG<sub>rainfed</sub>, most genotypes presented high gluten strength, except Quartzo, Sinuelo and Toruk. On the other hand, in L<sub>irrig</sub>, Gralha-Azul had the highest gluten strength.

**Table 3: Summary of the analysis of variance and means of grain protein concentration, wet gluten concentration, dough gluten strength and elasticity index from grains of ten wheat genotypes cultivated with two nitrogen (N) rates in three environments: Ponta Grossa in rainfed (PG<sub>rainfed</sub>), and Londrina in rainfed (L<sub>rainfed</sub>) and irrigated (L<sub>irrig</sub>) conditions**

Source of variation	DF	Mean square											
		Grain protein concentration			Wet gluten concentration			Dough gluten strength			Dough elasticity index		
		PG <sub>rainfed</sub>	L <sub>rainfed</sub>	L <sub>irrig</sub>	PG <sub>rainfed</sub>	L <sub>rainfed</sub>	L <sub>irrig</sub>	PG <sub>rainfed</sub>	L <sub>rainfed</sub>	L <sub>irrig</sub>	PG <sub>rainfed</sub>	L <sub>rainfed</sub>	L <sub>irrig</sub>
Block	2	0.17	0.23	0.22	0.37	1.63	5.68	1759	825	1044	21.3	21.7	28.2
Genotype (G)	9	2.19***	8.61***	3.69***	41.2***	67.6***	32.4***	23160***	3456 <sup>ns</sup>	8575***	63.1***	27.4 <sup>ns</sup>	50.1***
N rate (N)	1	2.18**	3.67***	8.09***	39.7*	6.41*	54.2***	7216 <sup>ns</sup>	13.1 <sup>ns</sup>	960 <sup>ns</sup>	51.8*	0.82 <sup>ns</sup>	1.73 <sup>ns</sup>
G x N	9	0.21 <sup>ns</sup>	0.08 <sup>ns</sup>	0.15 <sup>ns</sup>	15.6*	2.10 <sup>ns</sup>	2.25 <sup>ns</sup>	2231 <sup>ns</sup>	1250 <sup>ns</sup>	819 <sup>ns</sup>	26.1**	7.78 <sup>ns</sup>	6.84 <sup>ns</sup>
Error	38	0.18	0.13	0.18	5.68	1.54	1.76	2459	1834	1050	8.34	15.0	9.11
CV (%)		3.18	2.44	3.12	9.90	4.12	4.65	23.5	20.2	15.3	4.73	6.64	5.32

Factor	Treatment	Grain protein concentration (%)			Wet gluten concentration (%)			Dough gluten strength (× 10 <sup>-4</sup> J)			Dough elasticity index(%)		
		PG <sub>rainfed</sub>	L <sub>rainfed</sub>	L <sub>irrig</sub>	PG <sub>rainfed</sub>	L <sub>rainfed</sub>	L <sub>irrig</sub>	PG <sub>rainfed</sub>	L <sub>rainfed</sub>	L <sub>irrig</sub>	PG <sub>rainfed</sub>	L <sub>rainfed</sub>	L <sub>irrig</sub>
Genotype (G)	Sanhaço	13.5 <sup>b</sup>	15.4 <sup>b</sup>	13.2 <sup>c</sup>	25.2	32.6 <sup>b</sup>	28.7 <sup>c</sup>	223 <sup>a</sup>	223	233 <sup>b</sup>	57.2	57.0	55.5 <sup>b</sup>
	Graúna	13.7 <sup>a</sup>	15.5 <sup>b</sup>	14.1 <sup>b</sup>	29.1	32.1 <sup>b</sup>	30.1 <sup>b</sup>	233 <sup>a</sup>	189	200 <sup>c</sup>	57.4	55.1	58.2 <sup>b</sup>
	Gaivota	14.4 <sup>a</sup>	17.0 <sup>a</sup>	14.7 <sup>a</sup>	24.9	36.6 <sup>a</sup>	32.3 <sup>a</sup>	217 <sup>a</sup>	222	223 <sup>b</sup>	63.2	60.1	56.6 <sup>b</sup>
	Gralha-Azul	13.2 <sup>b</sup>	14.5 <sup>c</sup>	14.1 <sup>b</sup>	22.9	29.3 <sup>c</sup>	28.5 <sup>c</sup>	270 <sup>a</sup>	262	288 <sup>a</sup>	60.1	58.1	57.2 <sup>b</sup>
	Sinuelo	12.9 <sup>c</sup>	13.0 <sup>e</sup>	12.6 <sup>d</sup>	23.5	25.5 <sup>d</sup>	26.3 <sup>d</sup>	121 <sup>b</sup>	180	173 <sup>c</sup>	60.1	57.7	57.1 <sup>b</sup>
	Mestre	14.2 <sup>a</sup>	15.3 <sup>b</sup>	14.5 <sup>a</sup>	26.9	31.6 <sup>b</sup>	31.7 <sup>a</sup>	293 <sup>a</sup>	234	250 <sup>b</sup>	65.1	58.9	57.4 <sup>b</sup>
	Sossego	13.2 <sup>b</sup>	13.5 <sup>d</sup>	13.2 <sup>c</sup>	21.3	26.6 <sup>d</sup>	27.6 <sup>c</sup>	230 <sup>a</sup>	196	203 <sup>c</sup>	62.5	55.9	52.2 <sup>c</sup>
	Sintonia	13.3 <sup>b</sup>	14.4 <sup>c</sup>	13.5 <sup>c</sup>	24.3	28.7 <sup>c</sup>	26.5 <sup>d</sup>	259 <sup>a</sup>	202	201 <sup>c</sup>	66.7	59.4	62.2 <sup>a</sup>
	Toruk	14.0 <sup>a</sup>	14.7 <sup>c</sup>	13.4 <sup>c</sup>	21.3	30.7 <sup>b</sup>	28.1 <sup>c</sup>	115 <sup>b</sup>	212	175 <sup>c</sup>	57.9	62.4	58.2 <sup>b</sup>
	Quartzo	12.5 <sup>c</sup>	13.5 <sup>d</sup>	12.4 <sup>d</sup>	21.1	27.0 <sup>d</sup>	25.2 <sup>d</sup>	149 <sup>b</sup>	201	170 <sup>c</sup>	60.7	59.5	52.4 <sup>c</sup>
N rate (N)	40	13.3 <sup>b</sup>	14.4 <sup>b</sup>	13.2 <sup>b</sup>	24.9	29.7 <sup>b</sup>	27.5 <sup>b</sup>	222	212	208	60.2	58.5	56.5
	120	13.7 <sup>a</sup>	14.9 <sup>a</sup>	13.9 <sup>a</sup>	23.2	30.4 <sup>a</sup>	29.4 <sup>a</sup>	200	212	216	62.0	58.3	56.9

DF = degrees of freedom; CV = coefficient of variation. ns, \*, \*\*, and \*\*\* = not significant, significant at  $P \leq 0.05$ ,  $P \leq 0.01$ , and  $P \leq 0.001$ , respectively, by the F test. Means followed by the same letters in the column, for the factors genotype or N rates, belong to the same cluster by the Scott-Knott's test ( $P \leq 0.05$ ) or by the Student-Newman-Keuls' test ( $P \leq 0.05$ ), respectively.

**Table 4: Mean values of wet gluten concentration, dough extensibility, and elasticity index from grains of ten wheat genotypes cultivated with two nitrogen (N) rates in Ponta Grossa (PG<sub>rainfed</sub>) and Londrina (L<sub>rainfed</sub>) in rainfed conditions**

Genotype	N rate (kg ha <sup>-1</sup> )					
	Wet gluten concentration (%)		Dough extensibility (mm)		Dough elasticity index (%)	
	PG <sub>rainfed</sub>		L <sub>rainfed</sub>		PG <sub>rainfed</sub>	
	40	120	40	120	40	120
Sanhaço	25.16 <sup>aA</sup>	25.21 <sup>aA</sup>	85.0 <sup>bA</sup>	95.7 <sup>aA</sup>	54.00 <sup>cB</sup>	60.43 <sup>aA</sup>
Graúna	30.16 <sup>aA</sup>	28.09 <sup>aA</sup>	96.0 <sup>aA</sup>	82.3 <sup>bA</sup>	53.33 <sup>cB</sup>	61.43 <sup>aA</sup>
Gaivota	26.40 <sup>aA</sup>	23.43 <sup>bA</sup>	90.7 <sup>bA</sup>	91.0 <sup>aA</sup>	63.43 <sup>aA</sup>	63.00 <sup>aA</sup>
Gralha-Azul	23.26 <sup>bA</sup>	22.44 <sup>bA</sup>	85.0 <sup>bA</sup>	90.7 <sup>aA</sup>	59.83 <sup>bA</sup>	60.37 <sup>aA</sup>
Sinuelo	28.61 <sup>aA</sup>	18.43 <sup>bB</sup>	86.0 <sup>bB</sup>	102 <sup>aA</sup>	60.72 <sup>bA</sup>	59.57 <sup>aA</sup>
Mestre	27.36 <sup>aA</sup>	26.54 <sup>aA</sup>	88.7 <sup>bA</sup>	102 <sup>aA</sup>	66.50 <sup>aA</sup>	63.77 <sup>aA</sup>
Sossego	20.84 <sup>bA</sup>	21.72 <sup>bA</sup>	71.7 <sup>bA</sup>	68.7 <sup>bA</sup>	64.27 <sup>aA</sup>	60.77 <sup>aA</sup>
Sintonia	24.12 <sup>bA</sup>	24.50 <sup>aA</sup>	105 <sup>aA</sup>	99.0 <sup>aA</sup>	66.40 <sup>aA</sup>	66.93 <sup>aA</sup>
Toruk	21.14 <sup>bA</sup>	21.43 <sup>bA</sup>	98.3 <sup>aA</sup>	80.0 <sup>bB</sup>	54.53 <sup>cB</sup>	61.36 <sup>aA</sup>
Quartzo	21.64 <sup>bA</sup>	20.64 <sup>bA</sup>	94.7 <sup>aA</sup>	87.3 <sup>aA</sup>	58.67 <sup>cA</sup>	62.63 <sup>aA</sup>

Means followed by the same lowercase letters in the column and uppercase letters in the line (individually for each environment) belong to the same cluster by the Scott-Knott's test ( $P \leq 0.05$ ) or by the Student-Newman-Keuls' test ( $P \leq 0.05$ ), respectively.

The dough elasticity index averaged 61.1%, 58.4%, and 56.7% in PG<sub>rainfed</sub>, L<sub>rainfed</sub>, and L<sub>irrig</sub> (Table 3). It was influenced by the genotype × N rate interaction in PG<sub>rainfed</sub> (Table 3). In this environment, increasing N rates from 40

to 120 kg ha<sup>-1</sup> increased the elasticity index of the genotypes Graúna, Toruk and Sanhaço. In PG<sub>rainfed</sub>, within 40 kg ha<sup>-1</sup> N, the genotypes Gaivota, Mestre, Sossego and Sintonia had lower elasticity index (Table 4). However, within 120 kg



ha<sup>-1</sup> N there was no difference among the genotypes. In L<sub>irrig</sub>, the genotype factor influenced the elasticity index, which achieved the highest value for Sintonia, and the lowest one for Quartzo and Sossego (Table 3).

There were positive correlations (*r*), and above 0.5, between dough tenacity and the falling number (*r* = 0.50\*\*\*), dough tenacity and gluten strength (*r* = 0.68\*\*\*), concentrations of grain protein and wet gluten (*r* = 0.73\*\*\*), and P/L ratio and dough tenacity (*r* = 0.80\*\*\*) (Table 5). Furthermore, negative correlations were also observed, among which are highlighted those between dough tenacity and experimental flour extraction (*r* = -0.52\*\*\*), and P/L ratio and dough extensibility (*r* = -0.78\*\*\*).

## DISCUSSION

### Weather conditions during the growing season and their implications on technological quality of grains and flour

The means of maximum and minimum temperatures in the three environments indicate that no frost occurred (temperatures <0 °C), nor thermal stress by heat (i.e. temperatures >32 °C) during wheat grain development. Therefore, it is an indication that there was no negative influence of the temperature on wheat technological quality of grains and flour.

Guarienti et al. (2004), while studying the effects of maximum and minimum temperatures on industrial traits of wheat grains, found that the occurrence of mean maximum temperature around 20–25 °C during the grain-filling stage (as observed in the present study) favored some characteristics related to flour quality, such as falling number, TKW, gluten strength, among others. They attributed such results to the fact that the mean maximum temperature extended the persistence of green leaves in the

plants. Thus, it favored the photosynthesis process and the accumulation of photoassimilates in the plant, which were posteriorly translocated to the reproductive organs for grain development, benefitting wheat industrial characteristics.

The mean minimum temperature observed in the present study is within the optimum temperature range for the stage of grain development and filling (Franceschi et al., 2009). Guarienti et al. (2004) observed a negative influence of the mean minimum temperature between 12.5 and 14.4 °C on HW, falling number, and experimental flour extraction. They attributed this negative influence to the activation of some enzymes responsible for the degradation of reserve substances (e.g. carbohydrates and proteins), which occurred combined with the soaking of the grains caused by large water availability in this period. On the other hand, this temperature range throughout the grain-filling period positively influenced the dough gluten strength and P/L ratio.

The accumulated water deficit throughout the wheat-growing season was higher and more significant in L<sub>rainfed</sub> as compared to the other environments. An adequate water availability preceding the stage of grain development and filling improves the nutritional composition of the plants and the subsequent translocation of photoassimilates to the reproductive organs, positively influencing the technological quality of wheat grains (Haberle et al., 2008; Zhao et al., 2009). However, water deficit after the anthesis stage reduces the length of the grain-filling period because it provides earlier senescence of plant leaves and tillers (Garrido-Lestache et al., 2004; Franceschi et al., 2009; Zhao et al., 2009; Brunel et al., 2013). In this context, sparse rainfalls and lower water availability in L<sub>rainfed</sub> reduced the accumulation of reserve substances in the plant tissues and, consequently, decreased the technological quality of grains and flour harvested in this environment.

**Table 5: Pearson correlation coefficient (*r*) between the technological quality traits of grains and flour of ten wheat genotypes cultivated with two nitrogen (N) rates in three environments: Ponta Grossa in rainfed (PG<sub>rainfed</sub>), and Londrina in rainfed (L<sub>rainfed</sub>) and irrigated (L<sub>irrig</sub>) conditions**

Variable	HW	TKW	EFE	FN	GPC	WGC	P	L	P/L	W
TKW	-0.04 <sup>ns</sup>									
EFE	0.07 <sup>ns</sup>	-0.28***								
FN	0.12 <sup>ns</sup>	0.21**	-0.44***							
GPC	-0.02 <sup>ns</sup>	-0.15*	0.06 <sup>ns</sup>	-0.32***						
WGC	0.25***	-0.35***	0.11 <sup>ns</sup>	-0.24***	0.73***					
P	0.09 <sup>ns</sup>	0.13*	-0.52***	0.50***	0.01 <sup>ns</sup>	0.06 <sup>ns</sup>				
L	0.28***	-0.21**	0.18**	-0.24***	0.27***	0.45***	-0.36***			
P/L	-0.16*	0.21**	-0.39***	0.41***	-0.17**	-0.26***	0.80***	-0.78***		
W	0.25***	0.05 <sup>ns</sup>	-0.41***	0.34***	0.21**	0.31***	0.68***	0.34***	0.18**	
EI	-0.04 <sup>ns</sup>	0.29***	-0.12*	0.09 <sup>ns</sup>	0.08 <sup>ns</sup>	-0.13*	-0.02 <sup>ns</sup>	0.20**	-0.12 <sup>ns</sup>	0.34***

Hectoliter weight (HW), thousand-kernel weight (TKW), experimental flour extraction (EFE), falling number (FN), grain protein concentration (GPC), wet gluten concentration (WGC), dough tenacity (P) and extensibility (L), tenacity/extensibility (P/L) ratio, dough gluten strength (W) and elasticity index (EI). ns, \*, \*\*, and \*\*\*: not significant, significant at *P* ≤ 0.05, *P* ≤ 0.01, and *P* ≤ 0.001 by the *t* test, respectively (*n* = 180 pairs of records for each correlation).

It is worth mentioning that there was no water excess during the final period of the grain-filling stage, i.e. there were no intense and frequent rainfalls close to the stage of grain physiological ripening in the three environments. This weather condition favored the maintenance of the grain quality because it prevented the activation of the  $\alpha$ -amylase enzyme, which usually happens during the grain soaking stage, even though the grain is still in the spike. This enzyme is responsible for the degradation of starch (Whan et al., 2014), which is an essential protein concerning wheat technological quality.

### Technological quality of grains and flour as influenced by nitrogen fertilization under different environmental conditions

Hectoliter weight is defined by the different characteristics of the grains, including their uniformity, form, density and size. For this reason, the milling industry considers the HW as an indicator of grain quality, being used as a parameter for wheat trade (Karaoglu et al., 2010). The grains of the wheat genotypes had HW between 72.5 and 80 kg hL<sup>-1</sup>, which is within the range established by the Brazilian legislation for wheat commercialization (Brasil, 2010). However, the Brazilian commercial typification (i.e. I, II e III, according to RCBPTT, 2016) for the harvested grains differed depending on the response of each genotype to the weather conditions of each environment.

The water availability during the final period of the grain-filling stage indicated no water excess in this period in all environments, which explains the environmental adaptability of HW. According to Franceschi et al. (2009), the HW of wheat grains can undergo severe reductions due to the excess rainfall during the grain ripening stage. In addition, Garrido-Lestache et al. (2004) found that high temperatures and water deficit at the end of the grain-filling stage promoted a negative effect on HW due to the shortening of this period.

The TKW is also a quality indicator for wheat grains. This trait is genetically controlled, but it can be influenced by agronomic crop practices (Tavares et al., 2014; Rozbicki et al., 2015). Prando et al. (2012) and Marinho et al. (2022b) verified that the TKW varied according to the wheat genotype, corroborating the results obtained in the present study. The TKW in PG<sub>rainfed</sub> was, on average, higher than that in L<sub>rainfed</sub> and L<sub>irrig</sub>. This outcome may be attributed to the colder temperature in PG<sub>rainfed</sub> (Fig. 5a), which reduces the plant metabolic rate and increases the length of the grain-filling stage, resulting in a higher TKW (Gaju et al., 2009; Trautmann et al., 2017; Savill et al., 2018). Mamrutha et al. (2020) found that lower night temperature during the wheat grain-filling stage favors the increase of TKW, which can be attributed to the lower plant respiration rate that reduces

the carbon losses, allowing higher starch accumulation in the developing grains (Bahuguna et al., 2017).

Regarding the falling number, the environments with increased water supply (PG<sub>rainfed</sub> and L<sub>irrig</sub>) yielded wheat with higher values for this trait. A suitable water supply is essential for wheat growth and development and directly affects grain yield and quality (Rezaei et al., 2010). All wheat genotypes presented falling number values above 250 s that is the minimum value established by the Brazilian legislation to consider some wheat as a flour improver, which is the best quality classification for bakery (Brasil, 2010). This finding indicates that these wheat genotypes combined with suitable weather conditions during the harvest favored the falling number in the studied environments.

Denčić et al. (2011), while evaluating the flour quality traits of 140 wheat genotypes, found that the falling number varied due to the genotype factor. This finding was attributed to the dominant genetic effect on this trait as compared to the environmental effect. On the other hand, Yong et al. (2004) verified that the falling number was mainly influenced by environmental factors, considering 39 wheat genotypes cultivated in four agroecological zones from China. Therefore, both factors – genotype and environmental conditions – can change the falling number, in addition to influencing other technological quality parameters of wheat flour and grain (Rozbicki et al., 2015; Souza et al., 2021; Faria et al., 2022; Martins et al., 2023), as observed in the present paper.

The experimental flour extraction changed in the three environments, mainly influenced by the genotype factor. Such trait is related to the hardness of the grains used as a feedstock for wheat flour production. As hardness is a trait that varies among genotypes (Pasha et al., 2010), the flour extraction was also influenced by the genotypes. Furthermore, the positive response of flour extraction to increased N rates applied to wheat crop in L<sub>rainfed</sub> may be attributed to the higher amount of N taken up by plants and mobilized to the grain-filling physiological process. According to Bazzo et al. (2021), the higher N supply through fertilization favors grain development, especially concerning the endosperm – a tissue rich in starch – from which is extracted and produced wheat flour.

The alveography parameters of dough tenacity, extensibility, and P/L ratio varied mainly due to the influence of the genotype factor, which showed an interaction with the environmental conditions. This outcome is attributed to the different adaptability of each genetic material to the specific edaphoclimatic conditions (Gonçalves et al., 2020; Souza et al., 2021). Módenes et al. (2009), while evaluating the rheological properties of different flours from Brazilian

wheat genotypes, verified that both dough tenacity and extensibility varied according to the genotype used, but these alveography parameters were also influenced by the storage period of the wheat grains before milling, indicating that the environmental conditions can modify the alveography parameters before and after harvesting the grains.

The increased water supply in  $PG_{rainfed}$  and  $L_{irrig}$  favored the dough tenacity and, consequently, the P/L ratio. In contrast, the moderate water deficit in  $L_{rainfed}$  provided a higher dough extensibility than that of the other environments. According to Park et al. (2014), the gliadins [proteins that contribute to the viscosity and extensibility of the wheat dough (Bagulho et al., 2016)] are formed and accumulated at the beginning of the grain-filling stage, i.e. before the glutenins [proteins responsible for dough strength, elasticity and tenacity (Barak et al., 2013)]. Moreover, these authors reported that environmental conditions that shorten the grain-filling period, such as water deficit, can increase the relative concentration of gliadins and reduce the concentration of glutenins. Furthermore, variations in these proteins directly affect gluten functionality and wheat rheological properties (Vancini et al., 2019). Thus, this information brings light to the outcomes obtained for dough tenacity, extensibility, and P/L ratio in the environments of the present study.

The moderate water deficit in  $L_{rainfed}$  promoted a higher grain protein concentration as compared to that of the other environments ( $PG_{rainfed}$  and  $L_{irrig}$ ), where the water availability was higher. Other studies have also found that moderate water deficit provides greater grain protein concentration, which was attributed to increased grain N concentration in drought conditions (Haberle et al., 2008; Casagrande et al., 2009; Zhao et al., 2009). In wheat crops, the flowering and grain-filling stages are particularly sensitive to water deficit and high temperatures, which usually shortens the grain-filling period leading to lower grain yield, besides altering the grain quality (Garrido-Lestache et al., 2004; Rekowski et al., 2021). In the condition of drought stress, the protein/starch ratio changes in the direction of protein, resulting in smaller wheat grains with a higher protein concentration (Blumenthal et al., 1993).

The grain protein formation requires suitable soil N availability for plant uptake, even during the grain-filling stage, because the N accumulated in the plant biomass is preferentially remobilized for the starch formation and, posteriorly, rises the grain protein concentration (Silva et al., 2019). In this context, Mosanaei et al. (2017) and Ma et al. (2019), studying the effects of N fertilization on wheat crops, verified that increasing N rates increased the grain protein concentration. Such studies highlight

the importance of N for protein composition in wheat grains, corroborating the outcomes obtained in the present study. However, in spite of the higher N rate having increased the grain protein concentration in the three environments, these increases were of low magnitude. This finding indicates that the application of 40 kg ha<sup>-1</sup> N was enough for the plants to achieve a suitable grain protein concentration to meet the industrial requirements of mills and food companies that use wheat flour. Corassa et al. (2018) found that late N fertilization at the ear emergence stage increased the grain protein concentration, but this increase did not significantly contribute to improving the yield and quality of the wheat produced.

Increasing N rates also raised the wet gluten concentration of the harvested grains. According to Stefen et al. (2015), a higher N availability increases the grain protein concentration and, as gluten is formed particularly by the gliadins and glutenins proteins, its concentration also increases. We highlight that the effects of N fertilization on grain quality varied among the environments and were more notable in Londrina ( $L_{irrig}$  and  $L_{rainfed}$ ). Moreover, the varied plant responses to N fertilization in  $PG_{rainfed}$  is evidence that the wheat genotypes have distinct efficiencies for N uptake, assimilation and utilization (Benin et al., 2012; Hawkesford, 2017; Marinho et al., 2022a), which influences the quality of the produced grains. In this context, the difference in wet gluten concentration among the studied genotypes indicates that this grain trait is genetically controlled, although it is influenced by environmental conditions and agronomic crop practices (Denčić et al., 2011; Lidon et al., 2019).

Wet gluten concentration was higher in conditions of lower water availability ( $L_{rainfed}$ ), as verified for grain protein concentration, which can be attributed to the increase of protein/starch ratio in conditions of drought stress (Blumenthal et al., 1993). Furthermore, the lowest values of gluten concentration were observed in  $PG_{rainfed}$  environment where the lower minimum temperatures (below 12 °C) were recorded during grain formation. According to Guarienti et al. (2004), the occurrence of low temperatures when the grains are unripe provides a higher formation of starch than proteins, resulting in grains with lower protein and gluten concentrations, which supports our findings.

Although N fertilization increased the concentrations of grain protein and wet gluten, the N rates did not influence the dough gluten strength of the genotypes in the three environments. This outcome was already expected as N increases the total amount of grain proteins (i.e. albumins, globulins, glutenins and gliadins) but not influences the relative proportion of the proteins that form the gluten, i.e. glutenins and gliadins (Schmidt et al., 2009; Stefen et al.,



2015; Souza et al., 2019b). Schmidt et al. (2009), studying N fertilization in Brazilian wheat genotypes, found that the application of N, although increasing the grain protein concentration, did not significantly influence the dough gluten strength, corroborating the present outcomes. In contrast, Pinnow et al. (2013) verified that N fertilization, besides favoring the wet gluten concentration in wheat grains, also increased dough gluten strength, which may be related to an increase in gluten-formation proteins. Similarly, Guerrini et al. (2020) verified that the grain protein concentration and dough gluten strength increased in response to N rate increase.

The values of dough gluten strength observed in the three environments indicate that this alveography parameter presented high environmental adaptability. The variations observed among genotypes show that dough gluten strength depends mainly on the genotype factor, which was also verified by Penckowski et al. (2010). The dough gluten strength of the genotypes ranged from 115 to  $293 \times 10^{-4}$  J and, although within the range of the Brazilian commercial classification of wheat flour [which varies from  $100 \times 10^{-4}$  J (basic wheat) to  $300 \times 10^{-4}$  J (improver wheat) (Brasil, 2010)], such results are below the reference values reported by the wheat breeders responsible for such genotypes. This outcome is likely related to the weather conditions near the grain ripening stage that influenced grain composition and quality.

The elasticity index of wheat dough was above 50% in all situations. Therefore, all genotypes produced suitable flour for bakery, confirming their commercial classification, i.e. 'bread wheat' or 'bread/improver wheat' (Basso and Foloni 2015a, b; Basso and Foloni, 2016; RCBPTT, 2016; Basso et al., 2017).

Overall, there was a wide frequency of significant correlations between the technological quality parameters of grains and flour (Table 5). Nevertheless, most correlations were of low magnitude, i.e. with a correlation coefficient below 0.5. This finding may be attributed to the high number of record pairs ( $n = 180$ ) in the dataset, which produced a highly significant correlation, even for the cases of the lower correlation coefficients (Cohen, 1988).

Tenacity is a feature that indicates dough stability and resistance to deformation, which justifies its high correlation ( $r = 0.68^{***}$ ) with gluten strength, which is also associated with dough resistance to deformation (Módenes et al., 2009).

The high positive correlation ( $r = 0.73^{***}$ ) between the concentrations of grain protein and wet gluten – a type of grain protein – is justified because both are related to

quantitative analyses, i.e. they only evaluate the 'quantity' and not the 'quality' of the respective traits. The analysis of grain protein concentration (measured in the whole flour) refers to the total protein content in the grain, including albumins and globulins, besides gluten-formation proteins (i.e. gliadins and glutenins) (Antunes et al., 2020). In turn, the analysis of the wet gluten (measured in the white flour) considers only the concentrations of gliadins and glutenins. Other studies also obtained a high correlation ( $r = 0.75$ – $0.95$ ) between grain protein and wet gluten (Rozbicki et al., 2015; Stefen et al., 2015), corroborating the current outcomes.

The negative correlation between the alveography parameters P/L ratio and dough extensibility was also expected, as well as the positive correlation between P/L ratio and dough tenacity, because P/L is a ratio derived from the two other variables (i.e. tenacity/extensibility). Stefen et al. (2015) also obtained a negative correlation with a high coefficient ( $r = -0.94$ ) between P/L ratio and dough extensibility, reinforcing the consistency of this correlation.

Finally, increasing N rates increased the concentrations of grain protein and wet gluten, although these increases did not alter the Brazilian commercial classification of the flour produced by the wheat genotypes. Therefore, N fertilization can be managed by focusing on grain yield at a suitable cost-benefit ratio, instead of trying to improve the quality of grains and flour.

Based on the obtained outcomes, there is evidence that the traits related to the technological quality of grains and flour vary mainly due to genotype and environmental conditions than N fertilization, as observed by Kadar et al. (2019). This finding may be attributed to the distinct adaptability of each genetic material to specific environmental conditions, which directly influence the quality of wheat grains (Xue et al., 2016).

## CONCLUSIONS

A higher supply of N increases the concentrations of grain protein and wet gluten. However, it does not influence the dough gluten strength and the Brazilian commercial classification of the flour produced by the wheat genotypes. Nitrogen fertilization also influences the experimental flour extraction (i.e. flour yield), dough tenacity and elasticity index, whose effects depend on the genotype  $\times$  environment interaction. Environments with higher temperatures favor flour extraction and wet gluten concentration; while lower temperatures increase the TKW and the falling number. Water deficit increases the dough extensibility and grain protein concentration; while higher

water availability favors the falling number, dough tenacity and P/L ratio. Therefore, these outcomes are important drivers for farmers when choosing specific wheat genotypes for the environmental conditions of their farms, when they intend to meet the industrial requirements of mills and food companies that use wheat flour.

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## Authors' contributions

Jéssica Marinho: investigation, analysis and interpretation, writing – original draft, writing – review & editing. Sérgio Silva: conceptualization, study design, methodology, data acquisition, data curation, analysis and interpretation, supervision, validation, writing – original draft, writing – review & editing, graphic design and photographs. Inês Fonseca: analysis and interpretation, writing – review & editing. Martha Miranda: writing – review & editing, photographs. Eliana Guarienti: writing – review & editing. Claudemir Zucareli: writing – review & editing.

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