CROP BREEDING AND APPLIED BIOTECHNOLOGY

ARTICLE

Identification of RILs for agronomic and grain quality traits in rice through Intraspecific crosses

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Abstract: This study aimed to select recombinant lines and explore phenotypic and genotypic correlations using BLUP. It was conducted in Capão do Leão/RS during two years, in an incomplete block design with intercalary controls, with four replications. 131 and 128 RILs were tested in the F_6 in the F_7 generations, respectively. Plant height; days to flowering; panicle length; number of panicles per plant; number of fertile and sterile spikelets per panicle; one hundred grains weight; yield per plant; broken, chalky, white-belly and red-streaked grains; vitreous whiteness; gelatinization temperature; and apparent amylose content were obtained. According to the study, line F105 is an elite line for improving grain quality, exhibiting high amylose content (27.041%). Canonical (r=0.817), phenotypic (0.541) and genotypic (0.808) correlations inferred that groups of grain quality and agronomic traits were not independent and there was a tendency for the amylose content to be associated with grain yield.

Keywords: Oryza sativa *L., biometric models, BLUP analysis, cooking quality, industrial quality*

INTRODUCTION

Rice is one of the most important staple foods for humankind, being cultivated in several regions and consumed by more than half of the world's population, supplying 20% of carbohydrate source in the world's diet (Mohidem et al. 2022). The American continent is the world's second-largest producer of rice, with Brazil leading the production rankings. Among the states, Rio Grande do Sul stands out as the largest producer (mostly flood irrigated), reaching 7.2 million tons in the 2022/2023 harvest (CONAB 2023).

With the growing population worldwide, an increase in rice yields is needed to match the demand. Yield increases can be achieved by crop management and/or genetic improvement, being the later responsible for lifting people out of poverty and increasing global food supply in the last century (Lenaerts et al. 2019).

In addition to higher yielding genotypes, with better responses to biotic and abiotic stresses, aiming to ensure food security, rice breeding programs in recent decades have also considered the improvement of traits related to grain quality





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³ Universidade Federal de Pelotas, Campus Capão do Leão, 5HXM+QV, s/n, Campus Universitário, 96160-000, Capão do Leão, RS, Brazil (Rahman and Zang 2022). Breeding programs use several strategies to release superior genotypes. The development of rice segregating populations, such as recombinant inbred lines (RILs), through the crossing of divergent genotypes, such as *indica* and *japonica*, can generate progenies with broader genetic variability (Li et al. 2022). Furthermore, they are distinct from each other, both in agronomic and grain quality traits, such as the size and shape of grains, chalky or vitreous grains, as well as a range in amylose contents (Uyeh et al. 2021).

A population of RILs is composed of lines that show homogeneity within and heterogeneity between them. This homogeneity allows for the study of the genetic basis of various quantitative traits and facilitates conducting field experiments with repetitions (Falconer 1987). Also, it can be used for studying genetic loci underlying phenotypic traits (Priyadarshan 2019). They can be used to identify traits contrasting between parents, and mapping without the need to deal with population structure (Rini et al. 2023, Sabran et al. 2023). RILs have been used in studies to understand the genetic loci of yield and grain quality in rice (Sharma et al. 2021, Jin et al. 2023).

To assist the identification of transgressive individuals from a segregating population, mixed models become a valuable statistical tool. BLUP (Best linear unbiased prediction) analysis has the potential to enable a higher success rate in genotype selection since it has properties that allow predicting the genetic value of observed individuals, given that the genetic covariance between genotypes is proportional to their genotypic similarity (Meher et al. 2022). BLUP analyses have been used in rice (Ndikuryayo et al. 2023), maize (Yue et al. 2022), and wheat (Casagrande et al. 2020).

It is common in breeding programs to measure groups of traits related to morphology, quality and grain yield, being most of the morphological traits evaluated in the field, and most of the yield and grain quality traits evaluated in the laboratory. Linear correlation analysis allows to study of the behavior of variable pairs. However, when aiming at the indirect selection of traits of interest between groups, to plan a selection strategy and phenotypically associate several traits in different units, the canonical correlations are shown as an alternative to the analyses (Haghshenas et al. 2019).

In view of the above, and regarding that improving rice grain quality and yield are indispensable for safeguarding food security, the objective of this work was to select promising lines from a cross BRS Querência (*indica*) x Nipponbare (*japonica*), using a mixed model (BLUP). Also, to screen for agronomic and grain quality traits, evaluate the phenotypic and genotypic linear correlations, and the canonical correlations between grain quality and agronomic traits.

MATERIAL AND METHODS

Plant material

The research started in 2010 with the cross BRS Querência x Nipponbare, originating a segregating population of recombinant inbred lines (RILs) where each line constitutes a family. The SSD (Single Seed Descent) method was used to advance generations, obtaining 131 lines in the F_6 generation and 128 in the F_7 generation.

Field experimental details

Field and laboratory phenotyping was carried out in 2014/2015 and 2015/2016, at Terras Baixas Station – Embrapa Clima Temperado, Capão do Leão – RS. The population of RILs and parents (BRS Querência and Nipponbare) were sowed following an incomplete balanced block design, being the parents intercalated as controls, arranged in four replicates (Ramalho et al. 2012).

The experimental plot consisted of one 1.5 m long row, spaced by 0.30 m, being each RIL or parent composed by 15 plants. The basic fertilization was 300 kg ha⁻¹ of NPK (05-20-20), and 60 kg ha⁻¹ of nitrogen as urea, applied in coverage at the start of tillering. The soil was covered with water layer at 30 days after the seedling emergence. The water layer height ranged between 7.5 and 10 cm. Crop management practices, such as weeds, disease and insect control were performed according to the recommendations for irrigated rice in Southern Brazil (SOSBAI 2016).

Measurement of agronomic, yield and quality traits

In the field, plant height (PH, in cm), and days to flowering (DF, in days) were evaluated from 10 plants, randomly. At the end of the reproductive cycle, ten plants from each line were harvested individually. The traits measured were panicle length (PL, in cm), number of panicles per plant (NPP, in units), number of fertile spikelets per panicle (NFS,

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in units), number of sterile spikelets per panicle (NSS, in units), one hundred grains weight (HGW, in g), and yield per plant (YP, in g).

Subsequently, 40 higher yielding lines were selected, also evaluating the parents. The grains of 10 plants from each line were processed in a mini-MT 2012 Suzuki test rig (Máquinas Suzuki S/A) for peeling and polishing. The intrinsic physical quality attributes of the grains were evaluated by a S21 rice grain statistical analyzer (iSuzuki Software) based on the analysis of digital images of each sample. The following parameters were determined: broken grains (BG, in %), chalky grains (CG, in %), white-belly grains (WG, in %), red-streaked grains (RG, in %), vitreous whiteness (VW, in %).

For attributes related to cooking quality, the gelatinization temperature (GT) was determined using an indirect methodology adapted from Martinéz and Cuevas (1989). Three replicates were performed for each line, evaluating six grains (whole, healthy and polished) from each sample, distributing them evenly in a glass petri dish containing 10 mL of 1.7% potassium hydroxide (KOH) solution. The plates were covered and incubated in an oven at 30°C for 23 hours. After this period, the degree of disintegration of rice grains (alkali spreading) was visually observed and classified in three categories: high (1, 2 and 3), intermediate (4 and 5) and low (6 and 7) (Martinéz and Cuevas 1989).

The apparent amylose content (AAC, in %) was determined by the colorimetric method with iodine, according to the protocol by McGrane et al. (1998) with modifications suggested by Hoover and Ratnayake (2001). To prepare the rice flour, 10g of whole grains without defects were selected and ground in a Willey-type mill (Marconi, Piracicaba, Brazil), with a 0.5 mm sieve. The apparent amylose content was determined through a calibration curve prepared with potato amylose. The interpretation of the results was based on the following classes: high amylose (amylose content between 25 and 33%), intermediate amylose (amylose content between 20 and 25%) and low amylose (amylose content between 9 and 20%), according to Coffman and Juliano (1987).

Statistical analyses

The data obtained were subjected to analysis of variance by the F test ($p \le 0.05$). Subsequently, a phenotypic linear correlation analysis was carried out, using Pearson's, and genotypic correlation, using the Mantel's method, respectively. The aim of these were to study the trends of associations between groups of characters. From the phenotypic correlation matrix, the analysis of canonical correlations was performed. Canonical correlations were used to estimate the maximum correlation between linear combinations of characters distributed in two groups: (1) grain quality traits – VW, BG, CG, WG, RG, AAC and GT; (2) agronomic traits – PL, NFS, NSS, PH, NPP, WHG, YP and DF. Statistical analyses were performed with the statistical software Genes (Cruz 2013).

The model, $y = Xrep + Zg + \varepsilon$, was used in the statistical analyses, where, y: is the data vector phenotypic; rep: is the vector of the repetition effects (assumed to be fixed) added to the general mean; g: is the vector of genotypic effects of lines (assumed to be random); ε : is the vector of errors/residuals (random). For the random effect of lines, the probability distribution was assumed as $IID \sim N$ (0, S^2_{g}), for the residuals the probability distribution was assumed as $IID \sim N$ (0, S^2_{g}). The capital letters X and Z represent the incidence matrices for the effects above mentioned. Data were subjected to mixed model analysis using the REML/BLUP set. The analyses were performed using the SAS software (SAS, 2016).

RESULTS AND DISCUSSION

Plant height is controlled by multiple genes that can be manipulated through breeding strategies to increase yield, since plant height affects plant architecture, apical dominance, biomass, and resistance to lodging (Liu et al. 2018). The population from this cross had lines with higher and lower predicted genotypic means for plant height than the parents in both generations evaluated. However, plant height should not be considered by itself in plant selection, but in conjunction with yield or grain quality traits, especially in the range of means observed in the lines presented (Figure 1A and 1B).

The results showed, for all evaluated traits, a higher amplitude of the confidence intervals in F_6 generation (Figures 1 and 2), and genetic variability according to descriptive statistics (Table 1). It suggests that the decrease in the amplitude of the confidence intervals, the minimum, mean and maximum statistics in the F_7 generation is because the heterozygosity tends to decrease along each cycle of self-pollination, increasing the homogeneity between plants within each line. In addition, the evaluated population is composed of recombinant inbred lines (RILs), which means that the phenotypic



Figure 1. Predicted genotypic means and interval of confidence for the traits plant height, panicle length, number of panicles per plant and number of fertile spikelets per plants, of RILs from the cross BRS Querência (red bar) x Nipponbare (blue bar) in the F_6 (A, C, E and G) and F_7 (B, D, F and H) generations.



Figure 2. Predicted genotypic means and interval of confidence for the traits number of sterile spikelets per plants and yield per plant of RILs from the cross BRS Querência (red bar) x Nipponbare (blue bar) in the F_6 (A and C) and F_7 (B and D) generations.

variations, mainly in F_7 generation, are related to residual heterozygosity, environmental effects, or even eventual mutations (Singh and Singh 2015). No line, comparing the F_6 and F_7 generations, had predicted genotypic means above the genitor BRS Querência for panicle length, number of panicles per plant, number of fertile spikelets and yield per plant (Figure 1C, 1D, 1E, 1F, 1G, 1H, 2C and 2D). However, 36 lines were common in both generations for the number of sterile spikelets, inferring improved performance for the trait (Figure 2A and 2B).

Yield per plant presents quantitative inheritance and consequently are highly influenced by environmental conditions (Hasan et al. 2022). In addition, the State of Rio Grande do Sul had a 10.2% reduction in yield in the 2015/16 harvest, compared to 2014/15, due to the extension of the sowing calendar imposed by the occurrence of heavy rains. The lack

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Trait/ F ₆	Sample size	Minimum	Mean	Maximum	CV (%)	Variance	SD	LCI	UCI
PL	1121	12.82	21.57	77.56	19.91	18.44	4.29	21.30	21.82
NFS	1121	1.20	93.81	363.40	38.03	1273.09	35.68	91.58	95.94
NSS	1121	1.00	25.72	175.00	85.12	479.20	21.89	24.35	27.02
PH	1121	55.00	96.50	152.00	17.27	277.69	16.66	95.46	97.50
NPP	1121	2.00	13.91	66.00	47.26	43.20	6.57	13.50	14.30
HGW	1121	1.34	2.78	4.74	13.25	0.14	0.37	2.76	2.81
YP	1121	1.05	29.15	144.53	57.56	281.57	16.78	28.10	30.15
DF	1121	71.00	88.48	117.00	10.31	83.19	9.12	87.91	89.03
Trait/ F ₇	Sample size	Minimum	Mean	Maximum	CV (%)	Variance	SD	LCI	UCI
PL	1387	12.25	19.92	29.88	15.68	9.76	3.12	19.75	20.09
NFS	1387	0.00	55.11	210.40	44.20	593.30	24.36	53.74	56.42
NSS	1387	2.00	29.05	164.40	74.37	466.62	21.60	27.83	30.21
PH	1417	43.00	91.20	140.00	16.78	234.04	15.30	90.35	92.01
NPP	1386	1.00	12.67	147.00	47.81	36.68	6.06	12.33	12.99
HGW	1384	0.05	2.70	5.22	18.16	0.24	0.49	2.67	2.72
YP	1384	0.05	17.15	74.37	58.31	100.03	10.00	16.59	17.69
Trait/ F ₆	Sample size	Minimum	Mean	Maximum	CV (%)	Variance	SD	LCI	UCI
VW	415	108.31	120.27	128.61	3.06	13.52	3.68	119.89	120.63
BG	415	0.00	3.72	26.76	114.23	18.10	4.25	4.14	3.22
CG	415	0.00	0.02	0.47	247.40	0.00	0.06	0.02	0.03
WG	415	0.00	0.25	4.77	203.36	0.26	0.51	0.20	0.30
RG	415	0.00	0.08	2.25	254.54	0.04	0.21	0.06	0.10
ACC	126	10.26	17.99	30.24	26.61	22.93	4.79	17.09	18.85
GT	126	1.00	5.60	7.00	33.24	3.46	1.86	5.25	5.93

Table 1. Summary of descriptive statistics

CV (%): coefficient of variation, SD: standard deviation, LCI: lower confidence interval and UCI: upper confidence interval; Panicle length (PL), number of fertile spikelets per panicle (NFS), number of sterile spikelets per panicle (NSS), plant height (PH), number of panicles per plant (NPP), one hundred grain weight (HGW), yield per plant (YP), days to flowering (DF), vitreous whiteness (VW), broken grains (BG), chalky grains (CG), white-belly grains (WG), red streaked grains (RG), apparent amylose content (AAC), gelatinization temperature (GT).

of luminosity as the main effect, added by the effects of the decrease in nutrient availability due to fertilizer leaching by water excess, delay in crop cycle, low tillering, and lower crop stand establishment were the reasons for the reduction in yield (CONAB 2016). These data agree with the differences observed between the F_6 (2014/2015) and F_7 (2015/2016) generations (Figure 2C and 2D). In this way, one could suggest an additional year of evaluations before selecting any line of this study for agronomic traits.

The traits related to grain quality, such as apparent amylose content, vitreous whiteness, and broken grains, exhibited higher genetic variability compared to others, as they displayed a wider range of results according to descriptive statistics (Table 1). According to Normative Instruction No. 6, of February 16, 2009, rice intended for sale as grain for consumption falls into several types (BRASIL 2009). Types are expressed numerically from one to five. For it to be considered type one, in a sample of 1000 g, the limit of chalkiness is 2%, 1% red-streaked grains, and 7.5% broken grains. Considering in this work the analysis of 10 g of grains from each line, it was observed that most lines fit into type one for chalkiness, red-streaked kernels, and broken grains (Figure 3). Furthermore, line F105 revealed a higher apparent amylose content than the BRS Querência genotype (Figure 3C) with intermediate amylose content (Teixeira et al. 2021). In addition, it showed low predicted genotypic means for white belly grains, broken grains, and high vitreous whiteness (Figure 3A, 3D, 3F). However, F105 line can be considered an elite genotype with high amylose content and high-quality grain.

The phenotypic, as well as the genotypic correlation, showed a significant positive association with a high magnitude between yield per plant and number of panicles per plant (Table 2), agreeing with previous data (Huang et al. 2020). This suggests it can be a useful trait to be used in the indirect selection of high yielding genotypes. For the other yield components, a positive phenotypic and genotypic correlation was observed between the number of sterile spikelets,



Figure 3. Predicted genotypic means and interval of confidence for the traits vitreous whiteness (A), chalky grains (B), apparent amylose content (C), white-belly grains (D), red-streaked grains (E), and broken grains (F) of RILs from the cross BRS Querência (red line) x Nipponbare (blue line) in the F₆ generation.

panicle length, and the number of fertile spikelets, while the trait number of fertile spikelets revealed a negative correlation with one hundred grains weight (Table 2). Previous reports have also found these associations (Dey et al. 2019). Long panicles with a large number of grains, can affect the translocation of photo-assimilates synthesized by the plant to the reproductive structures, resulting in unevenness in the quality and weight of the grains produced (Ferrari et al. 2022). The trait yield per plant and one hundred grains weight showed only phenotypic correlation (Table 2), suggesting it is due only to environmental effects.

Negative correlations of phenotypic and genotypic origin were found between vitreous whiteness and white-belly grains, apparent amylose content, and gelatinization temperature (Table 2). In general, the milled rice appearance quality (especially grain translucency) is negatively correlated with amylose content in the rice endosperm starch (Li et al. 2018). The rice grain translucency is interrupted by opaque areas in the endosperm (Tao et al. 2022a), creating white-belly grains or white centers. Furthermore, amylose is reduced with the increase in the chalky area and the abundance of long-chain amylopectin, which influences the gelatinization temperature, justifying the results found (Edwards et al. 2017, Tao et al. 2022a).

Correlations between groups of agronomic traits and quality traits are of interest to breeders. In the canonical correlation analysis, it was observed that the two groups, i.e., grain quality and agronomic traits, are not independent, revealing five canonical pairs using the Chi-square test. Therefore, there is a relationship between them (Table 3). A positive phenotypic and genotypic correlation between amylose and yield per plant were found in this study (Table 2). This correlation was also observed in restorer lines of hybrid rice, which suggests amylose content might be used as a reliable trait to improve grain yield (Hasan et al. 2020). Starch is the main component of rice endosperm, and it is mainly consisted of amylose and amylopectin. In the analysis of canonical correlations, it was also possible to verify that the amylose content was related to yield per plant and its yield components, in the first canonical pair (r=0.817) (Table 3). Therefore, it is necessary to emphasize the significance of canonical correlations once it allows higher complexities and accuracy in the results because simple correlations cannot always adequately reflect the cause-and-effect relationships between these traits, as revealed by studies regarding the relationship between phenological and agronomic traits in rice (Haghshenas et al. 2019).

The phenotypic and genotypic correlations revealed a significant positive association between yield per plant and chalked grains, and its negative association with vitreous whiteness (Table 2). Those traits were also associated through canonical correlations, in the first pair (Table 3). When genetically contrasting genotypes for chalkiness, as in the case of this work, are crossed, there is a potential for the presence and eventually an increase in the chalky area and reduction of vitreous whiteness in the progeny (Edwards et al. 2017). Studies related to chemical analysis of the chalky and translucent part of the rice endosperm have shown that the chalky part had a significant change in composition and in starch molecular structures. Therefore, the difference in molecular structures with different molecular size can possibly cause variation in grain weight (Tao et al. 2022a).

	PL	NFS	NSS	PH	NPP	HGW	YP	DF	VW	BG	CG	WG	RG	ACC	GT
PL	-	0.292	0.429*	0.472*	-0.102	0.176	0.176	0.136	-0.147	-0.149	-0.062	-0.087	-0.035	0.201	-0.119
NFS	0.279	-	0.572*	0.141	-0.128	-0.445*	0.285	-0.177	-0.385*	-0.041	0.128	0.113	0.047	0.472*	0.334*
NSS	0.498+	0.685+	-	0.377*	-0.175	-0.286	0.092	0.027	-0.427*	-0.038	0.003	0.003	-0.06	0.345*	0.178
PH	0.521+	0.130	0.413+	-	-0.086	0.180	0.156	0.194	-0.098	0.053	-0.220	-0.339*	-0.127	0.028	0.058
NPP	-0.186	-0.293	-0.253	-0.161	-	-0.001	0.715*	-0.132	-0.015	0.083	0.247	0.084	0.194	0.264	-0.103
HGW	0.214	-0.491*	-0.300	0.194	0.015	-	0.377*	0.164	-0.010	0.080	0.064	0.133	0.008	-0.041	-0.196
YP	0.173	0.221	0.110	0.147	0.635+	0.228	-	-0.047	-0.352*	0.179	0.406*	0.259	0.411*	0.541*	0.075
DF	0.171	-0.192	0.023	0.206	-0.167	0.354+	-0.054	-	-0.199	-0.355*	0.008	0.343*	-0.132	-0.090	0.160
VW	-0.172	-0.437*	-0.467*	-0.108	-0.037	-0.013	-0.447*	-0.213	-	-0.152	-0.221	-0.397*	-0.303*	-0.329*	-0.418*
BG	-0.169	-0.040	-0.039	0.060	0.126	0.087	0.243	-0.375+	-0.145	-	0.234	-0.146	0.163	0.086	-0.010
CG	-0.073	0.147	0.007	-0.242	0.357+	0.075	0.531+	0.010	-0.191	0.238	-	0.538*	0.269	0.306*	0.009
WG	-0.101	0.133	0.004	-0.367+	0.117	0.142	0.323	0.370+	-0.398+	-0.166	0.547+	-	0.379*	0.124	0.009
RG	-0.034	0.063	-0.079	-0.135	0.275	0.008	0.553⁺	-0.133	-0.321	0.177	0.297	0.386+	-	0.135	-0.004
ACC	0.248	0.643+	0.431+	0.018	0.419+	-0.057	0.808+	-0.100	-0.408+	0.119	0.393+	0.159	0.166	-	0.205
GT	-0.134	0.449+	0.237	0.056	-0.142	-0.225	0.106	0.192	-0.498+	-0.015	0.003	0.010	0.010	0.207	-

Table 2. Coefficients of phenotypic correlations (upper diagonal) and genotypic correlations (lower diagonal) between agronomic traits and grain quality traits, evaluated in F_6 RILs from the cross BRS Querência x Nipponbare

*, ** significant (p≤0.01) and (p≤0.05) by t-test; + significant (p≤0.05) by the Mantel test by the bootstrap method with 5000 simulations, respectively. Panicle length (PL), number of fertile spikelets per panicle (NFS), number of sterile spikelets per panicle (NSS), plant height (PH), number of panicles per plant (NPP), one hundred grain weight (HGW), yield per plant (YP), days to flowering (DF), vitreous whiteness (VW), broken grains (BG), chalky grains (CG), white-belly grains (WG), red streaked grains (RG), apparent amylose content (AAC), gelatinization temperature (GT).

The percentage of broken kernels was directly related to days to flowering, in the second canonical pair (r= 0.714) (Table 3), as well as phenotypic and genotypic correlations have shown (Table 2). The increase in the vegetative stage of rice allows the plant to produce more biomass, which contributes to an increase in its storage components, leading to grain filling (Streck et al. 2006). On the other hand, late harvests lead to higher percentages of broken grains, as they are more exposed to adverse weather conditions, increasing the number of malformed grains during the grain filling period, which affects rice processing (Londero et al. 2015).

Through the third canonical pair (r= 0.561), it was possible to observe that the increase in the percentage of white-belly grains had a high association with the decrease in plant height (Table 3), agreeing with phenotypic and genotypic correlation (Table 2). Taller plants become more competitive for light and, consequently, can be more efficient in photo-assimilate synthesis (Liu et al. 2018). Within the rice endosperm, the formation of chalky tissues is related to the insufficient supply of metabolites produced in the vegetative stage of rice development. This causes a disturbance in the growth of starch granules, leading to the appearance of chalky areas (Tao et al. 2022b), which might therefore be related to the lower plant height. It is also important to notice that chalky grain is a complex trait governed by multiple genes and their interactions with the variable environments (Tao et al. 2022b).

In the fourth canonical pair (r= 0.432) the increase in

Table 3. Canonical loads between grain quality traits (group 1) and agronomic traits (group 2), analysed in F₆ RILs from the cross BRS Querência x Nipponbare

Troite	Canonical Pairs										
Iraits	1°	2°	3°	4°	5°						
Group 1											
VW ¹	-0.717	-0.190	0.286	-0.283	-0.136						
BG	-0.032	-0.623	0.107	0.325	0.690						
CG	0.558	-0.222	0.353	-0.192	0.085						
WG	0.696	0.298	0.533	-0.289	0.135						
RG	0.319	-0.573	0.453	0.010	-0.322						
AAC	0.716	-0.312	-0.360	-0.017	-0.003						
GT	0.346	0.250	-0.008	0.875	-0.187						
Group 2											
PL	0.129	-0.066	-0.658	-0.418	-0.338						
NFS	0.566	0.014	-0.444	0.404	-0.192						
NSS	0.405	0.042	-0.776	0.170	0.462						
PH	-0.153	-0.121	-0.663	0.330	-0.069						
NPP	0.237	-0.368	0.052	-0.342	-0.136						
HGW	0.020	-0.007	0.107	-0.412	0.564						
YP	0.676	-0.529	-0.382	-0.092	-0.105						
DF	0.248	0.708	0.072	-0.800	-0.121						
r	0.817	0.714	0.561	0.431	0.326						
Chi-Square	1070.361	617.152	323.698	168.253	83.506						
df	56	42	30	20	12						
р	<0.01	<0.01	<0.01	<0.01	<0.01						

¹Vitreous whiteness (VW), broken grains (BG), chalky grains (CG), white-belly grains (WG), red streaked grains (RG), apparent amylose content (AAC), gelatinization temperature (GT), panicle length (PL), number of fertile spikelets per panicle (NFS), number of sterile spikelets per panicle (NSS), plant height (PH), number of panicles per plant (NPP), one hundred grain weight (HGW), yield per plant (YP), days to flowering (DF); r = correlation coefficient; df = degree of freedom; p = significance level.

the gelatinization temperature was related to the decrease in the panicle length, days to flowering, and one hundred grains weight (Table 3). The increase in photo-assimilates in the earlier period of rice development helps to stabilize the grain-filling process. The poor carbohydrate supply and its association with panicle size can influence the yield and quality of rice. Some reports have shown that compact-panicle varieties had worse grain quality (amylose content variation), i.e., lighter grains than loose-panicle ones (Bian et al. 2020). It is due to the competition among the spikelets for carbohydrates. Rice starch with low amylose content is easier to gelatinize than rice starch with high-amylose content (Teixeira et al. 2021).

The fifth canonical pair revealed a correlation r= 0.326 between the groups, and it was possible to verify that the increase in broken grains was associated to the number of fertile spikelets and one hundred grain weight (Table 3). The broke grain is associated with the dynamics of moisture in the hydration and dehydration process, grain moisture at harvest, post-harvest drying process, grain maturation, chalkiness (immature and chalky grains are more susceptible to breakage during the milling process), and genetic influence (Londero et al. 2015). Plants that have more carbohydrate supply, especially more productive ones, usually have a longer growth duration in field, what means more exposure to adverse weather conditions, and tending to have broken grains.

With the establishment of rice quality standards over the years, mainly based on the physical and chemical properties of the grains, there is a need to develop improvement strategies. Therefore, through the strategy and analyses used in this study, it was possible to infer that grain quality traits, mainly those associated to amylose content, influence grain yield and its components. However, hybridization between contrasting parents and the exploration of genetic variability mediated by the selection of promising genotypes, as line F105, continue to be strategies used in rice genetic Identification of RILs for agronomic and grain quality traits in rice through Intraspecific crosses

improvement programs, due to the considerable success in the development of high yielding genotypes with grain quality, thus satisfying the demands of the consumer.

CONCLUSIONS

The F105 line can be considered an elite genotype with high amylose content and high-quality grain from a cross BRS Querência x Nipponbare.

A strong genotypic and phenotypic correlation between yield per plant and apparent amylose content showed that indirect selection can be performed to obtain genetic gain.

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