Phytopathology/ Original Article

# 2N^vS translocation presence and inheritance of resistance to head blast in wheat

**Abstract** – The objective of this work was to evaluate the effect of the presence of 2N^vS translocation on the inheritance of wheat resistance to head blast, in $F_2$ segregating lines from two biparental crosses. The IPF 86766 and 'Santa Fe' genotypes were used as female parents (both 2N^vS carriers), and 'BRS 404', as the common male parent (non 2N^vS carrier). Using a mixture of three isolates of *Pyricularia oryzae Triticum*, wheat spikes and rachises were evaluated for bleaching severity at five (%BS_5dpi) and seven days post-inoculation (%BS_7dpi), as well as for the number of infection points (IPR) and pathogen sporulation (SPO). Chi-square analyses showed that segregation follows a 3:1 Mendelian hypothesis, with a major locus with a dominant allele: %BS_7dpi for IPF 86766 x 'BRS 404' and IPR for 'Santa Fe' x 'BRS 404'. Therefore, this is the first known study to show that the 2N^vS translocation responsible for resistance to head blast is a major locus with a dominant allele. In a controlled environment, 'BRS 404' shows bleaching severity and SPO similar with those occurring on IPF 86766 and 'Santa Fe'.

**Index terms**: *Magnaporthe oryzae*, *Triticum aestivum*, genetic resistance, rainfed farming, wheat blast.

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## Presença da translocação 2N^vS e herança da resistência à brusone em trigo

**Resumo** – O objetivo deste trabalho foi avaliar o efeito da presença da translocação 2N^vS sobre a herança da resistência de trigo à brusone, em linhagens segregantes $F_2$ de dois cruzamentos biparentais. Os genótipos IPF 86766 e 'Santa Fe' foram usados como genitores femininos (ambos portadores da 2N^vS), e 'BRS 404', como genitor masculino comum (não portador da 2N^vS). A partir da mistura de três isolados de *Pyricularia oryzae Triticum*, espigas e ráquis de trigo foram avaliadas quanto à severidade de branqueamento aos cinco (%BS_5dpi) e aos 7 dias após inoculação (%BS_7dpi), bem como quanto ao número de pontos de infecção (IPR) e à esporulação do patógeno (SPO). Análises de qui-quadrado mostraram que a segregação segue a hipótese mendeliana 3:1, com um loco de efeito maior, com alelo dominante: %BS_7dpi para IPF 86766 x 'BRS 404' e IPR para 'Santa Fe' x 'BRS 404'. Portanto, este é o primeiro estudo conhecido a mostrar que a translocação 2N^vS responsável pela resistência à brusone de trigo é um loco principal com um alelo dominante. Em condições controladas de ambiente, 'BRS 404' apresenta severidade de branqueamento e SPO similares às que ocorrem em IPF 86766 e 'Santa Fe'.

**Termos para indexação**: *Magnaporthe oryzae*, *Triticum aestivum*, resistência genética, cultivo de sequeiro, brusone.

## Introduction

The fungus *Magnaporthe oryzae* pathotype *Triticum* (MoT) is the causal agent of wheat blast. The first description in the world of this disease occurrence was made in Brazil, in 1985. Afterwards, blast disease was found in neighboring countries of South America (Kohli et al., 2011). The main symptom of wheat blast is the spike discoloration due to the infection of the rachis. Infected rachises show a dark lesion of irregular shape where the hyphae growth of the pathogen blocks the passage of sap in plant vessels. Grain production is severely impaired. Diseased plants produce wrinkled grains with low specific weight (Goulart et al., 2007). In 2016, the first occurrence of wheat blast in Asia is reported in Bangladesh causing grain yield losses reaching 51% (Islam et al., 2016). In the 2017–2018 season, wheat blast was also reported in Zambia (Tembo et al., 2020).

Although blast symptoms can occur in all parts of the wheat plant, spike infection is the most destructive form of the disease. Due to the spike / rachis architecture, the chemical control of blast using fungicides is harder and less efficient (Rocha et al., 2014; Boaretto et al., 2017). Thus, host genetic resistance is the most effective method for controlling the disease. It causes neither damage to the environment, nor higher production costs (Torres et al., 2022).

Genetic resistance can be classified as complete or partial. Complete resistance or vertical resistance is accomplished with one or a few genes. The resistance is isolate-specific, and its segregation takes place in discontinuous classes. Partial resistance, also known as horizontal or quantitative resistance, is accomplished with a set of genes with a minor effect. The resistance may be in different loci (QTL) and contribute to the reduction of the development of the pathogen. It is a non-race-specific form, and it has been described as a source of durable and broad-spectrum resistance in the case of rice blast (Jiang et al., 2020).

The high genetic variability of MoT imposes restrictions on the long-term use of cultivars carrying major resistance genes. In the case of rice, the resistance of these cultivars remained efficient for up to three years, when the virulence / aggressiveness of the pathogen overcomes it (Wang & Valent, 2017). In this context, the search for sources of resistance genes is permanent. The gene or QTL pyramiding are promising strategies to promote durable and broad-spectrum resistance.

Against MoT isolates, five specific resistance genes are described: *Rmg2*, *Rmg3*, *Rmg7*, *Rmg8*, and *RmgGR119* (Zhan et al., 2008; Anh et al., 2015; Tagle et al., 2015; Wang et al., 2018). In addition to these genes, a chromosomal translocation from *Aegilops ventricosa* was associated with higher levels of wheat resistance to MoT isolates (Cruz et al., 2016). Wheat cultivars carrying the $2N^{v}S$ translocation have been used in breeding programs, in South American countries and Bangladesh, aiming at the obtention of blast resistance (Cruppe et al., 2020; Cardozo Téllez et al., 2022).

The $2N^{v}S$ translocation was initially introgressed from *A. ventricosa* ($2n = 4x = 28$) into the interspecific hybrid 'VPM1' (Maia, 1967). It has a cluster of wheat resistance genes to stripe rust (*Yr17*, *Puccinia striiformis* f. sp. *tritici*), to leaf rust (*Lr37*, *Puccinia triticina*), and to stem rust (*Sr38*, *Puccinia graminis* f. sp. *tritici*). Subsequently, the presence of nematodes resistance genes was also identified in this translocation (Jahier et al., 2001; Williamson et al., 2013). Helguera et al. (2003) developed two molecular markers for the identification of the $2N^{v}S$ translocation in wheat accessions. One is a dominant marker (VENTRIUP / LN2), and the other is a codominant marker (URIC / LN2). Recombinant lines derived from the Caninde#1 / Alondra cross were evaluated in field experiments conducted in Bolivia and Bangladesh, regarding blast resistance. Among the seven QTL identified, only one, situated on $2N^{v}S$, was consistently significant across the different environments, explaining from 22.4% to 50.1% of the phenotypic variation (He et al., 2020). A large-effect QTL was also identified in this region as associated with $2N^{v}S$, using an independent doubled-haploid population in controlled conditions studies (Ferreira et al., 2021).

Despite the reduction of the disease symptoms, not all wheat genotypes carrying the $2N^{v}S$ translocation showed a significant reduction in head blast severity, which suggests that the genetic background could influence the expression of resistance conferred by this translocation (Cruz et al., 2016). Ferreira et al. (2021) reported the presence of recombination between the distal region of chromosome 2AS and the translocation marker VENTRIUP / LN2. Such result would explain why some strains amplifying the $2N^{v}S$ marker show variable reactions to the disease.

The objective of this work was to evaluate the effect of the presence of 2N$^V$S translocation on the inheritance of wheat resistance to head blast, in F$_2$ segregating lines from two biparental crosses.

## Materials and Methods

The experiments were carried out at Embrapa Trigo, in the municipality of Passo Fundo, in the state of Rio Grande do Sul, Brazil. The F$_2$ generation of two biparental crosses, IPF 86766 x 'BRS 404' and 'Santa Fe' x 'BRS 404' (Table 1), was evaluated for its response to pathogen inoculation in a controlled environment.

The IPF 86766 genotype is a wheat line that is part of a collection, received in 2013 from the International Maize and Wheat Improvement Center (CIMMYT). It has been characterized for its reaction to wheat blast and contains the 2N$^V$S translocation. Santa Fe is a North-American wheat cultivar of the WestBred / AGSECO, released in 2003 (Cruz et al., 2012); this cultivar carries the 2N$^V$S translocation. Embrapa developed BRS 404, a cultivar of early maturation that is a non-2N$^V$S carrier. BRS 404 is indicated for rainfed farming in the homogeneous region of adaptation 4 (RHA 4), which encompasses the Brazilian Savanna (Só e Silva et al., 2016). The IPF 86766 line and Santa Fe cultivar were used as female parents in the crosses with cultivar BRS 404. The Anahuac 75 cultivar is highly susceptible to wheat blast, and it was used as control treatment.

The experiment consisted of 25 buckets, with F$_2$ lines (10 seed per bucket) of each population obtained from the crosses of IPF 86766 x 'BRS 404' and 'Santa Fe' x 'BRS 404'. Seed from parental genotypes (IPF 86766, 'Santa Fe', and 'BRS 404') and 'Anahuac 75' were also sowed.

The experimental design was completely randomized. Each bucket contained a mixture of soil, substrate, and vermiculite at the ratio 1:1:1, and they were subjected to greenhouse conditions. Fifteen days after sowing, the plants were thinned, in order to remain five plants per bucket. Wheat plants were kept in greenhouse conditions until the heading time. Then, they were transferred to growth chambers, in a controlled environment for temperature and humidity. There, the plants were subjected to inoculation.

The fungal isolates Py 13.1.018, Py 13.1.023, and Py 13.1.012 were obtained from the Laboratório de Fitopatologia of Embrapa Trigo (SISGEN AC50A60). Monosporic cultures of the three isolates were obtained in 2013 from spikes of the CD 104 wheat cultivar, collected in the municipality of Jandaia do Sul, in the state of Paraná, Brazil. The isolates were grown in oat medium (60 g oat, 12 g agar, qs 1 L water) from 10 to 14 days, at 24±2°C, and received 12 hours of white fluorescent light. The conidial suspension was prepared using two drops of Tween 80 diluted in 1 L water. The concentration of each suspension was adjusted to 10$^5$ conidia per mL and, then, it was mixed in equal proportions. The mixture of these three isolate suspensions was inoculated in the wheat plants.

At the heading stage, wheat plants between 58 and 68 Zadoks growth stages (Zadoks et al., 1974) were subjected to inoculation. The spore mixture suspension was sprayed on both sides of the spikes through a manual sprayer. Wheat plants were covered with plastic bags which were kept in the dark for 24 hours at 26±2°C and relative humidity above 90%. After the initial 24 hours, the photoperiod was adjusted to 12-hour light. The conditions of temperature and relative humidity were maintained. Plants were kept in growth chambers until 14 days post-inoculation (dpi).

For the IPF 86766 x 'BRS 404' and 'Santa Fe' x 'BRS 404' populations, 115 and 91 plants were evaluated, respectively. The repetition was one spike per plant.

**Table 1.** Pedigree, year of release, location of origin, reaction to wheat blast, and 2N$^V$S translocation carrier status of three wheat (*Triticum aestivum*) parents. Embrapa Trigo, RS, Brazil, 2019.

| Parent | Pedigree | Year of release | Origin | Reaction to blast[1] | 2N$^V$S translocation |
|---|---|---|---|---|---|
| IPF 86766 | SRN/AE. SQUARROSA (358)//FILIN/MILAN/3/GONDO | - | Cimmyt, Mexico | R | Present |
| Santa Fe[2] | JAGGER/G1878 | 2003 | Kansas, USA | R | Present |
| BRS 404[3] | MGS 1-ALIANÇA/WT 99172 | 2015 | Embrapa, Brazil | MS | Absent |

[1]R, resistant; MS, moderately susceptible. [2]Source: Cruz et al. (2012) and Gris... (2017). [3]Source: Só e Silva et al. (2016) and Reunião... (2022).

Spike bleaching severity expressed as percentage (%BS) was estimated at 5 dpi (%BS_5dpi) and 7 dpi (%BS_7dpi). For this evaluation, it was used the diagrammatic scale of Maciel et al. (2013). At 14 dpi, the inoculated spikes were collected and frozen.

Gradually, the spikes were individually threshed, and the number of infection points in the rachis (IPR) was counted. Then, the rachises were weighed and evaluated for pathogen sporulation. Asepsis was performed using a commercial solution of sodium hypochlorite (2.5%) diluted in water at 1:1 (v/v). The material was kept in the solution for one min. For the extraction of the disinfectant product, a double rinse was performed with distilled and sterilized water. The rachises were incubated on Petri dishes with filter paper moistened with distilled water. The Petri dishes were kept in an incubation room for four days in 12-hour photoperiod, at 25°C. After 96 hours, the segments were placed in plastic capped Falcon tubes containing 5 mL sterile water. In order to release the MoT conidia, they were agitated for 40 s at maximum level in the MA 162 tube shaker (Marconi Equipamentos para Laboratórios, Piracicaba, SP, Brazil). The sporulation of each $F_2$ line was quantified using the Eclipse E200MV R optical microscope (Nikon, Tokyo, Japan). The conidia counting was carried out in the Bright-Line Neubauer improved hematocytometer, with 0.100 mm depth, $0.0025$ mm$^2$ (KASVI: Produtos Laboratoriais, Curitiba, PR, Brazil). Three spore counting per infected rachis were performed, to estimate the number of spores per gram of rachis (SPO).

Considering the phenotypic data of both $F_2$ populations, two hypotheses of segregation ratio were tested: the 3:1 ratio, by which a major gene with a dominant effect would control the resistance to blast; and the 1:2:1 ratio, by which a major gene with an additive effect would control the resistance to MoT. The chi-square test ($\chi^2$) was used. The null hypothesis is accepted, when the calculated probability level is equal to or greater than 5%, with n-1 degrees of freedom, by the following formula:

$$\chi^2 = \sum \left(observed - \exp ected\right)^2 \Big/ \exp ected$$

where: $\Sigma$ summation of the data; "observed" is the observed frequency; "expected" is the expected frequency, in accordance with the Mendelian segregation model.

For all evaluated traits, except %BS_5dpi, phenotypic classes were defined according to the limit values of cultivar Anahuac 75, used as the susceptible control. For the %BS_7dpi variable, two classes were considered in 3:1 segregation, as follows: resistant (R), when severity value was between 0 and 30%; and susceptible (S), when severity value was higher than 30%. For the 1:2:1 segregation, three classes were defined for the following severity values in percentage: resistant (R), between 0 and 20%; intermediate (I), between 21 and 30%; and susceptible (S), higher than 30%. For IPR, in 3:1 segregation, two classes were defined based on the following scores: susceptible (S), between 0 and 5; and resistant (R), greater than 5. For 1:2:1 segregation, three classes (S, I, and R) were defined, as follows: susceptible (S), values between 0 and 5 IPR; intermediate (I), values between 6 and 10 IPR; and resistant (R), values larger than 10 IPR. Using the SPO variable values obtained with Anahuac 75 cultivar, two phenotypic classes were defined to test the 3:1 segregation: resistant (R), for values between 0 and $4x10^6$ spores per gram of rachis; susceptible (S), when values were larger than $4x10^6$ spores per gram of rachis.

Fifteen days after sowing, leaf tissues were collected from wheat plants. DNA was extracted according to Saghai-Maroof et al. (1984). The DNA concentration was estimated using the Quant-iT PicoGreen dsDNA Reagent P7581 (Thermo Fisher Scientific, Waltham, MA, USA). Fluorescence was measured using the FLUOstar Omega microplate reader (BMG Labtech, Ortenberg, Germany). The presence of the $2N^VS$ translocation, in the three parental genotypes, and the $F_2$ lines of both populations were identified using the following primers: URIC (5'GGTCGCCCTGGCTTGCACCT3') and LN2 (5'TGCAGCTACAGCAGTATGTACACAAAA3') (Helguera et al., 2003).

URIC / LN2 is codominant and allows of the distinction between homozygous and heterozygous genotypes. PCR conditions were applied according to the descriptions by Helguera et al. (2003). The amplification was carried out using the GeneAmp 9700 PCR System thermocycler (Applied Biosystems, Waltham, MA, USA).

The Shapiro-Wilk's test was used to check the normality assumption, which was not met. Four nonparametric tests were chosen. The Kruskal-Wallis'

H-test was used to look for differences among the classes obtained using the URIC / LN2 marker. The classes with significant results were compared using the Dunn's test. The traits %BS_5dpi, %BS_7dpi, IPR, and SPO were subjected to the Mann-Whitney's U-test, to compare the parents' means. The Spearman's rank correlation was used to estimate the correlation among these traits. All data analyses were performed using the R version 4.2.1 (R Core Team, 2022).

## Results and Discussion

The means of IPR differed significantly between IPF 86766 and 'BRS 404', and those of %BS_5dpi and IPR between 'Santa Fe' and 'BRS 404' (Table 2). These results can be explained by the interaction between these genotypes and the inoculation environment or by the used inoculum suspensions.

At 5 dpi, 'BRS 404' showed a lower bleaching severity than the female parents (Table 2). 'BRS 404' mean is statistically different from that of 'Santa Fe', whose wheat blast resistance showed a high level, as reported by Cruz et al. (2012). For bleaching severity at 7 dpi, 'BRS 404' was not significantly different from the IPF 86766 lineage or Santa Fe cultivar. These results indicate that 'BRS 404' has a good level of blast resistance, differently from what was previously reported (Reunião..., 2022). In 2016, 'BRS 404' was presented as resistant to blast at levels comparable to those of 'BR 18 (Terena)' (Só e Silva et al., 2016). 'BR 18' is the genotype with the longest resistance to wheat blast in Brazil (Torres et al., 2022).

Both resistant parents (IPF 86766 and 'Santa Fe') showed a higher IPR than 'BRS 404'. IPR is an inheritable trait (Ferreira et al., 2021), and its association to wheat blast resistance was first reported by Ferreira et al. (2020). The more intense is the hypersensitivity response associated to the IPR, the more restricted is the growth of the pathogen hyphae. Upon inoculation, the resistance mechanism of 'BRS 404' did not seem to be associated to a strong hypersensitivity reaction.

For both crosses, there was a significant positive correlation between %BS_5dpi and %BS_7dpi (Table 3). IPR did not show any correlation with other traits. This result contrasts with that by Ferreira et al. (2021) which was obtained using a doubled-haploid population inoculated with two isolates separately. Differences for the inoculation method and genetic background of the populations may explain the lack of correlation observed in the present study.

**Table 2.** Mann-Whitney's U test results of the trait means of each wheat (*Triticum aestivum*) genotype[1].

| Wheat genotypes | %BS_5dpi | p-value | %BS_7dpi | p-value | IPR | p-value | SPO | p-value |
|---|---|---|---|---|---|---|---|---|
| IPF 86766 | 2.60 | 0.1435 | 13.20 | 1.0000 | 17.00 | 0.0007 | 4.76 | 0.0081 |
| Santa Fe | 4.97 | 0.0018 | 16.72 | 0.4172 | 16.05 | 0.0000 | 2.79 | 0.3717 |
| BRS 404 | 1.95 | | 14.44 | | 2.00 | | 2.49 | |

[1]BS, bleaching severity; %BS_5dpi, percentage at five days post-inoculation; %BS_7dpi, percentage at seven days post-inoculation; IPR, total number of infection points in the rachis; and SPO, number (x10^6) of spores of *Magnaporthe oryzae Triticum* pathotype per gram of wheat rachis.

**Table 3.** Spearman's correlation results of evaluated traits of wheat (*Triticum aestivum*) $F_2$ lines.

| Population | Traits | %BS_5dpi | %BS_7dpi | IPR | SPO |
|---|---|---|---|---|---|
| | %BS_5dpi | 1.00 | | | |
| IPF 86766 x BRS 404 | %BS_7dpi | 0.89** | 1.00 | | |
| | IPR | -0.01 | -0.03 | 1.00 | |
| | SPO | 0.33* | 0.47** | 0.11 | 1.00 |
| | %BS_5dpi | 1.00 | | | |
| Santa Fe x BRS 404 | %BS_7dpi | 0.84** | 1.00 | | |
| | IPR | -0.03 | -0.00 | 1.00 | |
| | SPO | 0.29 | 0.27 | -0.01 | 1.00 |

[1]BS, bleaching severity; %BS_5dpi, percentage at five days post-inoculation; %BS_7dpi, percentage at seven days post-inoculation; IPR, total number of infection points in the rachis; and SPO, number (x 10^6) of spores of *Magnaporthe oryzae Triticum* pathotype per gram of wheat rachis. * and **Significant correlations at 5% and 1% probability, respectively.

For lines derived from the IPF 86766 x 'BRS 404' cross, both evaluations of bleaching severity were positively correlated to SPO (Table 3). This was not observed for the 'Santa Fe' x 'BRS 404' cross. The pathogen sporulation can be used to quantify the disease resistance, according to Leclerc et al. (2019). Considering three wheat cultivars and 13 wheat lines subjected to inoculation of MoT, some authors have not found possible to verify any relationship of dependence between bleaching severity and MoT sporulation (Maciel et al., 2021).

Chi-square analyses ($\chi 2$) were applied for testing the deviation of the observed segregation from the theoretical segregation of phenotypic traits %BS_7dpi, IPR, and SPO. For IPF 86766 x 'BRS404', the chi-square test validated the goodness of fit to the expected population ratio of 3:1 (resistant:susceptible). The hypothesis of a gene with the dominant allele conferring resistance was accepted for %BS_7dpi (Table 4). This result was not observed for the population of $F_2$ lines from 'Santa Fe' x 'BRS 404'. However, the trait IPR for these lines showed that a dominant major gene regulates the inheritance of resistance (Table 4). These apparently conflicting results can be explained by the interaction between wheat genotypes and MoT isolates, which is reported in the literature (Cruz et al., 2012, 2016; Maciel et al., 2014).

In order to confirm the effect of the $2N^VS$, the $F_2$ plants were genotyped with the codominant molecular marker URIC / LN2. The results show the dominant effect of this locus (Table 5). For the population of IPF 86766 x 'BRS 404', it confirms the genetic linkage between the presence of the translocation and the traits %BS_7dpi and IPR. For the population of 'Santa Fe' x 'BRS 404', the linkage is with the trait IPR. The type of effect of this locus is not clear for the trait SPO.

Several works showed the involvement of the $2N^VS$ translocation in the reduction of blast severity indices (Kohli et al., 2011; Cruz et al., 2016; Ferreira et al., 2021; Juliana et al., 2022). The International Center

**Table 4.** Chi-square analyses results for resistance of wheat (*Triticum aestivum*) $F_2$ lines to *Magnaporthe oryzae Triticum* pathotype.

| Ratio hypothesis | Probability (%) | Population | Number of $F_2$ lines | Traits | Reaction classes[1] | | | $\chi^2$ value |
|---|---|---|---|---|---|---|---|---|
| | | | | | R | I | S | |
| 3:1 | 3.84 | IPF 86766 x 'BRS 404' | 115 | %BS_7dpi | 82 | - | 33 | 0.84[ns] |
| | | | | IPR | 58 | - | 57 | 39.68* |
| | | | | SPO | 52 | - | 63 | 54.40* |
| | | 'Santa Fe' x 'BRS 404' | 91 | %BS_7dpi | 37 | - | 54 | 57.23* |
| | | | | IPR | 61 | - | 30 | 3.08[ns] |
| | | | | SPO | 26 | - | 65 | 104.61* |
| 1:2:1 | 5.99 | IPF 86766 x 'BRS 404' | 115 | %BS_7dpi | 60 | 22 | 33 | 56.51* |
| | | | | IPR | 57 | 38 | 20 | 37.03* |
| | | 'Santa Fe' x 'BRS 404' | 91 | %BS_7dpi | 26 | 11 | 54 | 69.55* |
| | | | | IPR | 30 | 27 | 34 | 15.39* |

[1]R, resistant; I, intermediate; and S, susceptible. *Significant at 5% probability. [ns]Nonsignificant.

**Table 5.** Dunn's test results of genotypic classes of wheat (*Triticum aestivum*) $F_2$ lines obtained using the URIC / LN2 marker[1].

| Population | Genotype | %BS_5dpi | %BS_7dpi | IPR | SPO |
|---|---|---|---|---|---|
| IPF 86766 x 'BRS 404' | Homozygous susceptible | 10.2[2)ns] | 36.4a | 4.0a | 6.9a |
| | Heterozygous | 7.2[ns] | 20.3b | 7.5b | 4.8ab |
| | Homozygous resistant | 7.6[ns] | 19.0b | 7.4b | 3.6b |
| 'Santa Fe' x 'BRS 404' | Homozygous susceptible | 16.0[ns] | 43.7[ns] | 4.6a | 14.5a |
| | Heterozygous | 13.9[ns] | 39.3[ns] | 10.2b | 6.3ab |
| | Homozygous resistant | 9.9[ns] | 31.5[ns] | 11.3b | 4.8b |

[1]BS, bleaching severity; %BS_5dpi, percentage at five days post-inoculation; %BS_7dpi, percentage at seven days post-inoculation; IPR, total number of infection points in the rachis; and SPO, number (x$10^6$) of spores of *Magnaporthe oryzae Triticum* pathotype per gram of wheat rachis. [2]Means followed by equal letters of each population, do not differ by Dunn's test at 5% probability. [ns]Nonsignificant.

for Maize and Wheat Improvement (CIMMYT) published most of these works. CIMMYT develops and evaluates international germplasm collections in phenotyping platform, and is focused on improving wheat grain yield and other traits of interest. Reduced levels of blast severity were observed in IPF 86766, a CIMMYT's 2N$^V$S carrier genotype.

The study of Ferreira et al. (2021) was the only one that showed that 2N$^V$S locus and two minor QTL affect the trait IPR. The QTL at 5B and 7B are both involved when inoculating the isolate Py6025 in doubled-haploid lines derived from 'BRS 209' x 'CBFusarium ENT014'. Exception for the study by Ferreira et al. (2020, 2021), IPR counting has not been considered for phenotyping the head blast resistance because it is a laborious and time-consuming task. However, it was demonstrated that IPR is negatively correlated with blast bleaching severity (Ferreira et al., 2020). It can be hypothesized a strong relationship between IPR and traits of grain production. However, little is known about this trait for wheat blast resistance. In the present study, the variables %BS_7dpi and IPR segregated independently for both evaluated populations.

Using the ratio hypothesis 1:2:1 segregation, the result of the chi-square test was significant for both traits (%BS_7dpi and IPR) and both populations. It indicates the absence of a major effect locus with alleles showing codominance. Genotyping the F$_2$ populations with the codominant molecular marker URIC / LN2 allowed estimating the dose effect of the alleles. The genotyping confirmed the results of the chi-square. This is the first report to find out that the 2N$^V$S translocation responsible for the resistance to head blast is a major locus with a dominant allele.

## Conclusions

1. The 2N$^V$S translocation responsible for wheat blast resistance segregates following the presence of a major dominant allele: %BS_7dpi for IPR 86766 x 'BRS 404' and IPR for 'Santa Fe' x 'BRS 404'.

2. 'BRS 404' is moderately resistant to wheat blast.

3. 'BRS 404', a non-2N$^V$S carrier, shows bleaching severity and sporulation comparable to those of IPF 86766 and 'Santa Fe', when these genotypes are inoculated with Py 13.1.018, Py.13.1.023, and Py 13.1.012.

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