

## PERFORMANCE OF UPLAND RICE STRAINS IN REACTION TO FUNGAL DISEASES

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**ABSTRACT** - The control of the impact of diseases on plants can be carried out via breeding, through the development of cultivars less susceptible to pathogens. Therefore, this study aimed to assess the severity of fungal diseases in upland rice strains from the Cultivation and Use Value test, belonging to the Upland Rice Breeding Program” of Federal University of Lavras (UFLA), in partnership with “EMBRAPA Rice and Beans” and the “Minas Gerais Agricultural Research Company” (EPAMIG). The experiment was installed in the municipalities of Lambari and Lavras, both in Minas Gerais, Brasil, in the 2015/2016 and 2016/2017 seasons, in a randomized block design with three replications. Were evaluated 14 lines, 11 lines of the program and 3 commercial witnesses, regarding the intensity of diseases (leaf and neck blast, brown spot, grain spot and scalding). There was a significant difference between lineages for all traits, except brown spot, indicating the presence of genetic variability for the characters in question. The study of the interaction was performed, decomposing the statistically significant ones in simple and complex interactions. The experiment presented a coefficient of variation from 29% to 56% and accuracy from 56% to 95.5%, indicating that the estimates were obtained with good to medium experimental precision. It is essential to evaluate diseases in breeding programs in order to select cultivars that are increasingly resistant, as these affect not only productivity, but the quality and final yield of grains. The genotypes CMG 2119 (5), CMG 2162 (1), CMG 1896 (13) and CMG 2168 (2) were shown to be more tolerant to all diseases simultaneously, being, therefore, the most suitable for the launch, considering the character disease resistance. The Upland Rice Breeding Program of UFLA has lines with variability for resistance, strains with potential for release.

**Keywords:** *Oryza sativa* L., genetic breeding, resistance, severity.

### DESEMPENHO DE LINHAGENS DE ARROZ DE TERRAS ALTAS QUANTO A REAÇÃO ÀS DOENÇAS FÚNGICAS

**RESUMO** - O controle do impacto de doenças nas plantas pode ser realizado via melhoramento, através do desenvolvimento cultivares menos suscetíveis aos patógenos. Portanto, este estudo objetivou avaliar a severidade de doenças fúngicas em linhagens de arroz de terras altas do ensaio de Valor de Cultivo e Uso, pertencentes ao Programa de Melhoramento Genético de Arroz de Terras Altas da UFLA, em parceria com a Embrapa e Epamig. O experimento foi instalado nos municípios de Lambari e Lavras, ambos em Minas Gerais, nas safras 2015/2016 e 2016/2017, em delineamento em blocos casualizados com três repetições. Foram avaliadas 14 linhagens, sendo 11 linhagens do programa e 3 testemunhas comerciais, quanto a intensidade de doenças (brusone foliar e de pescoço, mancha parda, mancha-de-grãos e escaldadura). Houve diferença significativa entre as linhagens para todos os caracteres, exceto mancha parda, indicando a presença de variabilidade genética para os caracteres em questão. Foi realizado o estudo da interação, decompondo-se aquelas estatisticamente significativas em interações simples e complexa. O experimento apresentou coeficiente de variação de 29% a 56% e acurácia de 56% a 95,5%, indicando que as estimativas foram obtidas com boa média precisão experimental. É fundamental a avaliação de doenças nos programas de melhoramento visando selecionar cultivares cada vez mais resistentes, já que estas afetam não só a produtividade, mas a qualidade e rendimento final dos grãos. Os genótipos CMG 2119 (5), CMG 2162 (1), CMG 1896 (13) e CMG 2168 (2) mostraram-se mais tolerantes a todas as doenças simultaneamente, sendo, portanto, os mais indicados para o lançamento, considerando o caráter resistência a doenças. O Programa de Melhoramento de Arroz de Terras Altas da UFLA possui linhagens com variabilidade para o caráter resistência, linhagens estas com potencial para o lançamento no mercado.

**Palavras-chave:** *Oryza sativa* L., melhoramento genético, resistência, severidade.

#### INTRODUCTION

The genetic improvement in the culture of rice (*Oryza sativa* L.) for many years has been responsible for the development of cultivars with an ever higher productive

potential. In addition, the various breeding programs aim to obtain plants with greater resistance to diseases, higher grain quality, flowering season suitable for the most diverse regions, as well as cultivars tolerant to lodging and water

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deficit, always aiming to serve the consumer market and the requirements and needs of the producer (SOARES, 2012).

Not unlike, the Upland Rice Genetic Improvement Program of the Federal University of Lavras (UFLA) has been developing, for more than ten years, competitive strains within the rice crop market. One of the objectives of the program is aimed at resistance to the various fungal diseases that attack the rice culture. Among the main ones are: leaf and neck blast (*Pyricularia grisea*) (SANTOS et al., 2011), brown spot (*Drechslera oryzae*) (DALLAGANOL et al., 2014), grain spot (NARAYANASAMY, 2011) and scalding (*Monographella albescens*) (CELMER et al., 2007). Disease assessments are carried out annually, in all harvests and environments, in order to select plants that are increasingly resistant to the greatest number of pathogens possible.

It is a fact that diseases directly influence the quality of grains and greatly interfere with final productivity (EMBRAPA, 2006). In these circumstances, it is necessary to establish an association between fungal diseases with the greatest impact on rice culture with the agronomic traits evaluated, in order to even establish the negative effects of diseases on the final productivity of each cultivar or strain.

The quantification of plant diseases is the basis for the selection of more resistant cultivars. The assessment of disease severity and the elaboration of disease progress curves over the years allow an assessment that is less affected by the time of analysis and environmental variations (CAMPBELL; MADDEM, 1990).

Therefore, the present work aims to evaluate the severity of fungal diseases in upland rice lines from the Cultivation and Use Value (CUV) test belonging to the Upland Rice Genetic Improvement Program of UFLA, in partnership with the “Empresa Brasileira de Pesquisa Agropecuária/Centro Nacional de Pesquisa Arroz e Feijão” (EMBRAPA/CNPAF) and the “Empresa de Pesquisa Agropecuária de Minas Gerais” (EPAMIG).

## MATERIAL AND METHODS

Were used 14 strains from the CUV assays of the Upland Rice Breeding Program of Federal University of Lavras in partnership with “Empresa Brasileira de Pesquisa Agropecuária/Centro Nacional de Pesquisa Arroz e Feijão” and the “Empresa de Pesquisa Agropecuária de Minas Gerais” (EPAMIG). The experiments were implemented in two locations, namely, the municipality of Lavras (MG), at an altitude of 954 m, 21°12’11” south latitude and 44°58’47” west longitude and Lambari (MG), located at altitude of 832 m, 46°31’05” south latitude and 18°34’44” west longitude, in two harvests or agricultural years, 2015/2016 and 2016/2017. The experimental design used was randomized blocks, with three replications. The plots consisted of five lines of 4 m, with a spacing of 35 cm between lines.

The quantifications of leaf blast severity, brown spot and scalding were carried out on the plants of the useful area of the plots, without the application of fungicides. In the evaluation of neck blast, two readings were made in the phase of pasty grain and grain maturation, periods of greater

susceptibility of the crop to the disease. For grain spot, two readings were taken close to the harvest period.

A scale of grades proposed by the International Rice Research Institute (IRRI, 1996) was used, taking into account the severity of leaf diseases. The grades were assigned according to the visual analysis of the affected leaf tissue, being: grade 1 - less than 1% of the affected leaf area; note 3 - from 1% to 5% of the diseased folia area; note 5 - from 6% to 25% of the diseased leaf area; note 7 - from 26% to 50% of the diseased leaf area and note 9 - above 50% of the diseased leaf area. Two to three readings were taken in the stages of greatest infection of each disease in the field, with an interval of 7 days between one evaluation and another. The leaf blast evaluations were carried out approximately 45 days after sowing and those of the brown spot, close to the flowering phase, together with the scalding evaluation. For all diseases, genotypes with a severity score less than or equal to 3 were considered to be of satisfactory resistance for selection.

The data were analyzed comparatively based on three common witnesses (RAMALHO et al., 2012), and the averages obtained grouped by the Scott-Knott test (1974) at 5% probability through the statistical program R (2003). Individual analyzes of variance were performed with data on disease severity by location. Subsequently, joint analyzes were used (according to the joint analysis model, Equation 1) for each disease, using the statistical package of the program R (2003).

$$Y_{ij(k)} = m + T_i + A_k + (TA)_{ik} + B_{j(k)} + E_{ij(k)} \quad (\text{Equation 1})$$

Where:

$Y_{ij(k)}$  = observation of treatment i, in block j, within environment k,

m = overall average,

$T_i$  = effect of treatment i, where i = 1, 2, 3, 4, 5 ...14,

$A_k$  = effect of environment k, where k = 1,2,3,4.

This source of variation includes the effect of years and locations,

$(TA)_{ik}$  = effect of the interaction of treatment i with the environment k,

$B_{j(k)}$  = effect of block j within environment k, j = 1,2,3 and

$E_{ij(k)}$  = experimental error independently and identically distributed, following normal distribution, with zero mean and common variance.

It was also performed, with the aid of the statistical program RStudio, the breakdown of significant interactions in order to evaluate the percentage of simple and complex interactions that make up the genotype x environment interaction. The graphs were plotted with the average severity scores adjusted on the x-axis and the identification of each strain on the y-axis. Each line corresponds to an environment. Thus it is possible to visually analyze the behavior of each strain, in each environment considering each disease individually.

## RESULTS AND DISCUSSION

In order to estimate the experimental precision, the values of the coefficient of variation (CV%) and accuracy (Table 1) were obtained. The calculated coefficients of variation were between 29% and 56%, which, according to Pimentel Gomes (2000), are considered high or very high. This supposedly indicates that the experiment obtained poor experimental accuracy. However, when considering experiments with disease assessment, it is acceptable that the magnitude of the variation coefficient is greater, considering the visual assessment and not always carried out by the same assessor, which favors a greater dispersion of data around the mean (COSTA et al., 2002; SARI et al., 2016; NASCIMENTO, et al., 2018).

Costa et al. (2002) proposed a new range of variation coefficient for disease assessments such as leaf and neck blast and scalding. The authors also concluded that variables associated with diseases and lodging, in experiments with a large number of treatments and in randomized blocks, have high coefficients of variation. In this case, the experiment showed accuracy from good to median.

Accuracy (rgg) was also calculated, as it is another estimate that can measure the reliability and quality of the experiment. The accuracy value found for the scalding (ESC) evaluation, 56.8%, indicates a median precision. However, the estimates for LB (leaf blast), NB (neck blast) and GS (grain spot), respectively 95.47%, 95.5% and 75.57%, are considered high, which indicates good accuracy experimental. According to Resende and Duarte (2007) for precision and quality control in CUV experiments, these estimates were considered to be highly accurate.

According to the results of the joint analysis for the evaluation of brown spot (BS) (Table 1), there was no significant difference ( $p < 0.01$ ) in the average performance of treatments. This shows that the strains showed a statistically coincident behavior regarding tolerance to brown spot. The F test showed the presence of variability between environments, which indicates that they differed for this character. The interaction between genotypes per environment (G x E) was not significant, indicating that there was no difference in the average performance of treatments between environments for the average severity of BS.

**TABLE 1** - Summary of the joint analysis for the evaluations of brown spot (BS), scalding (ESC), leaf blast (LB), neck blast (NB) and grain spot (GS), considering the municipalities evaluated.

SV	DF	MS				
		BS	ESC	LB	NB	GS
Treatments (T)	13	0.300 <sup>ns</sup>	0.957 <sup>**</sup>	8.824 <sup>**</sup>	19.713 <sup>**</sup>	1.7597 <sup>**</sup>
Municipalities (M)	3	43.500 <sup>**</sup>	62.958 <sup>**</sup>	34.665 <sup>**</sup>	41.895 <sup>**</sup>	24.1482 <sup>**</sup>
T x M	39	0.504 <sup>ns</sup>	0.475 <sup>**</sup>	2.828 <sup>**</sup>	4.809 <sup>**</sup>	1.9193 <sup>*</sup>
Repetition	8	0.417 <sup>ns</sup>	0.911 <sup>ns</sup>	1.459 <sup>ns</sup>	5.619 <sup>**</sup>	1.734 <sup>**</sup>
Mistake	104	0.404	0.648	0.782	1.734	0.7547
CSI (%)	---	---	50.95	85.23	74.98	58.86
CCI (%)	---	---	49.05	14.77	25.02	43.14
CV (%)		32,75	39.20	29.11	55.72	36.76
Accuracy (%)		-	56.8	95.47	95.50	75.57
Averages		1.94	2.05	3.04	2.36	2.61

\*, \*\* significant F test at 5% and 1% probability, respectively. ns = not significant, SV = sources of variation, DF = degrees of freedom, MS = mean square, CSI = contribution of simple interaction, CCI = contribution of complex interaction, CV = coefficient of variation.

Differently from what was observed for BS, the effect of treatments was significant for all other diseases evaluated. This shows that the strains of the CUV assay differ significantly from each other when evaluated for scalding severity (ESC), leaf blast (LB), neck blast (NB) and grain spot (GS). This is an important fact, as it is an indication of the existence of genotypes with different levels of resistance in the breeding program. The F test showed the presence of variability between environments when considering these diseases, which indicates that they differed from each other for these characters. The interaction between genotypes by environment (G x E) was highly significant, indicating that the performance of the strains was not coincident in the different environments, that is, there was a difference in the average performance of treatments between the environments for the average severity of ESC, LB, NB and GS.

With the detection of the significance of the lineage x environment interactions, the decomposition of the G x E interaction was carried out. This breakdown aims to detect which percentage of the interaction is due to simple and complex interactions. The complex interactions indicate the lack of coincidence in the ranking of genotypes in relation to environmental variation, that is, there are strains that present altered behavior with the change of the environment, being better in one place than in another. The complex G x E interaction is a complicating factor for breeding programs, as it does not allow the perfect correlation between the genotype and the phenotype in different environments. This hinders both the recommendation of cultivars and the selection of the best genotypes by breeders (CRUZ; CASTOLDI, 1991; CRUZ et al., 2012). Of the diseases evaluated, ESC and GS were

those that presented most of the complex type interaction, with 49.05% and 43.14%, respectively.

By the Scott-Knott test (1974), the strains evaluated for the average severity of brown spot and scalding formed only one group (Table 2). For grain spot, neck blast and leaf blast, the lines were divided into two and three groups, respectively. The separation into groups shows the difference of the strains in terms of resistance to disease. The control BRSMG Caravera, considered moderately resistant to leaf blast, but moderately susceptible to neck blast and moderately resistant to brown

spot, grain spot and scalding (SOARES et al., 2008), has always been associated with the three largest averages and contained in maximum groups, b and c, therefore, together with the CMG 1511 strain, the materials of the CUV assay are less tolerant to most of the diseases under study. The control BRSMG Caçula, considered moderately susceptible to neck blast and scalding and moderately resistant to brown spot, grain spot and leaf blast (REIS et al., 2012), showed susceptibility mainly to leaf blast and grain spot, cases where allocated to group b.

**TABLE 2** - Average scores given for each cultivar, referring to each disease: brown spot (BS), scalding (ESC), leaf blast (LB), neck blast (NB) and grain spot (GS).

Strains	BS	ESC	LB	NB	GS
BRS Esmeralda	1.750 a*	1.583 a	2.056 a	1.500 a	2.056 a
CMG 2119	1.750 a	1.750 a	2.168 a	1.500 a	2.112 a
CMG 2162	1.833 a	1.833 a	2.333 a	1.500 a	2.278 a
CMG 1896	1.833 a	1.833 a	2.444 a	1.667 a	2.333 a
CMG 2168	1.833 a	1.833 a	2.583 a	1.667 a	2.389 a
CMG 2187	1.917 a	1.917 a	2.750 b	1.833 a	2.389 a
CMG 2188	1.917 a	2.000 a	2.833 b	2.000 a	2.500 a
CMG 2170	1.917 a	2.083 a	3.028 b	2.000 a	2.528 a
CMG 1509	1.917 a	2.083 a	3.028 b	2.000 a	2.639 a
CMG 2085	2.000 a	2.250 a	3.139 b	2.167 a	2.888 b
CMG 2185	2.000 a	2.250 a	3.167 b	2.333 a	2.944 b
BRSMG Caçula	2.000 a	2.333 a	3.333 b	2.333 a	3.028 b
CMG 1511	2.167 a	2.500 a	4.611 c	4.917 b	3.167 b
BRSMG Caravera	2.333 a	2.500 a	5.055 c	5.667 b	3.222 b
Averages	1.94	2.05	3.04	2.36	2.61

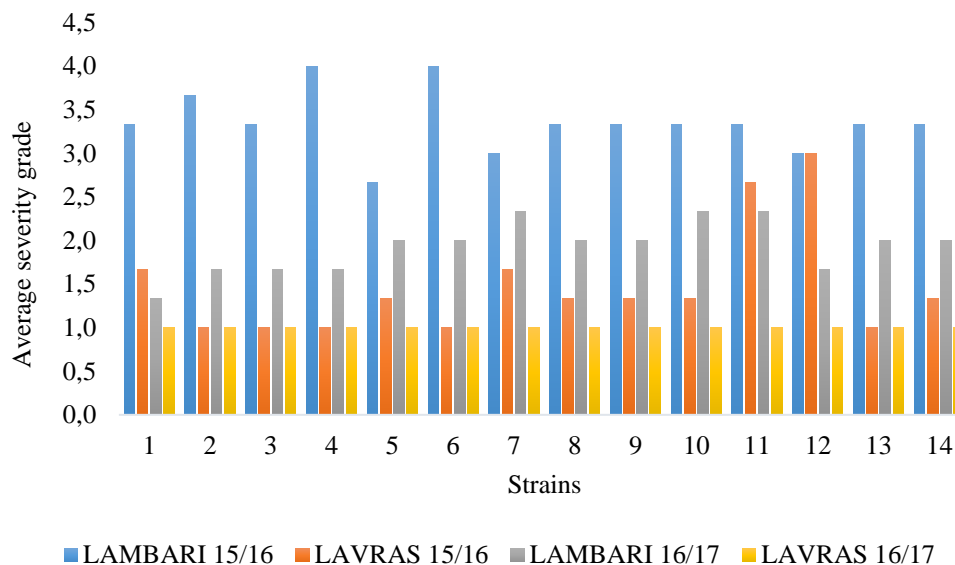
\*Averages followed by the same letter belong to the same group, by the Scott-Knott test ( $P \leq 0.05$ ).

The control BRS Esmeralda, considered resistant to neck blast and with good resistance to brown spot, scalding and grain spot (CASTRO et al., 2014), and the lines CMG 2119, CMG 2162, CMG 1896 and CMG 2168 were the ones that present the lowest averages, being always classified in the group by the Scott-Knott test. Thus, these were the most tolerant genotypes, considering all the diseases in question.

The percentage of disease at each location was statistically different. As shown in Figures 1 to 5, Lambari has a greater tendency for disease to occur when compared to Lavras, regardless of the agricultural year. Thus, strains tend to receive higher scores on the disease scale when evaluated at this location. This fact can be explained by the mild climate, with high levels of rain and humidity and

moderately low temperature. As the diseases evaluated are caused by fungi, this becomes a very favorable climate for their development.

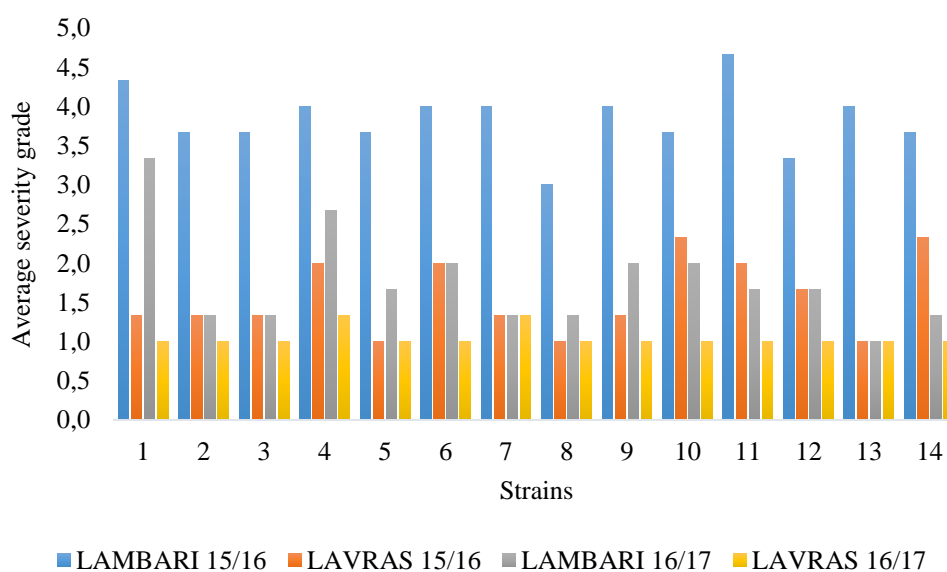
Looking at Figure 1, it can be seen that in the 2015/16 and 2016/17 seasons, the brown spot severity scores were, on average, equal to or less than 4. This shows that this disease has not been as severe in the last two agricultural years. In addition, the large number of banknotes 1, mainly in Lavras - 2016/17 harvest, shows that in many plots, there was no incidence. This disease has a major negative impact on the quality of rice grains as well as on productivity. It can result in reduced germination and death of seedlings formed from infected seeds, reduced photosynthetically active leaf area in young and mature plants (DALLAGNOL et al., 2013).



**FIGURE 1** - Average brown spot (BS) score by location and by cultivar.

The scalding (Figure 2), showed notes that showed greater average severity when compared to BS, mainly in Lambari - 15/16. The averages of the notes presented for the 16/17 harvest were not as high. The symptoms of scalding can be severe and reflect the sharp drop in production of a cultivar (FILIPPI, 2005). An increase in the intensity of the

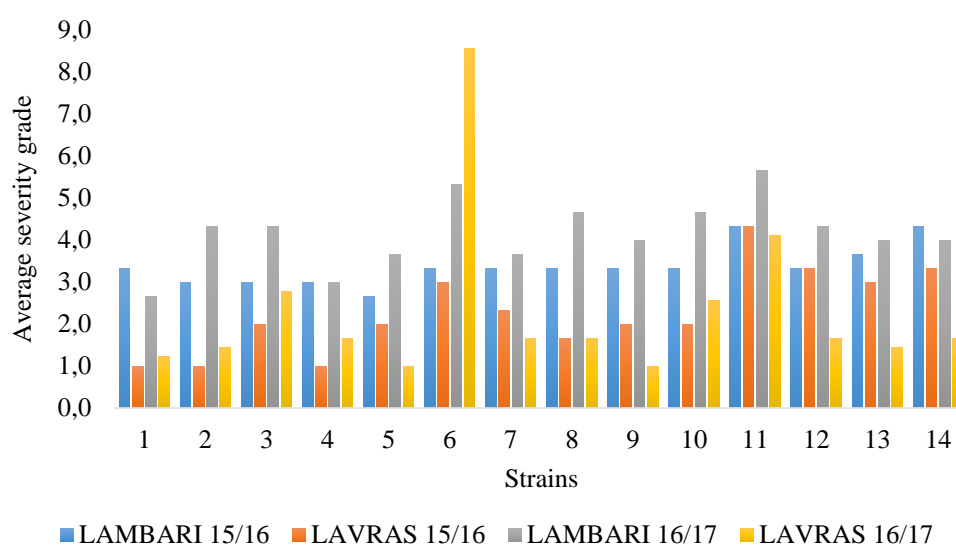
disease has been observed over the years. This places scalding as a disease of great importance in rice culture as well as a warning to breeders and producers, as it can lead to losses of around 30% in rice culture (TATAGIBA et al., 2014).



**FIGURE 2** - Average scalding (ESC) score by location and by cultivar.

Leaf blast (Figure 3) is a very severe disease in rice culture (SANTOS et al., 2011). It is considered the most important disease and despite the great attention of breeders in the selection of resistant cultivars. The tolerant cultivars, present in the CUV test, present very varied average grades, ranging from 1 (almost total absence of symptom on the

leaves) to 5 (up to 25% of the leaf area with the symptom). The control BRSMG Caçula (6) is an exception since, in the last harvest, he was severely attacked by the disease, reaching 100% of the leaf area with symptoms, which resulted in a great loss of productivity in this treatment.



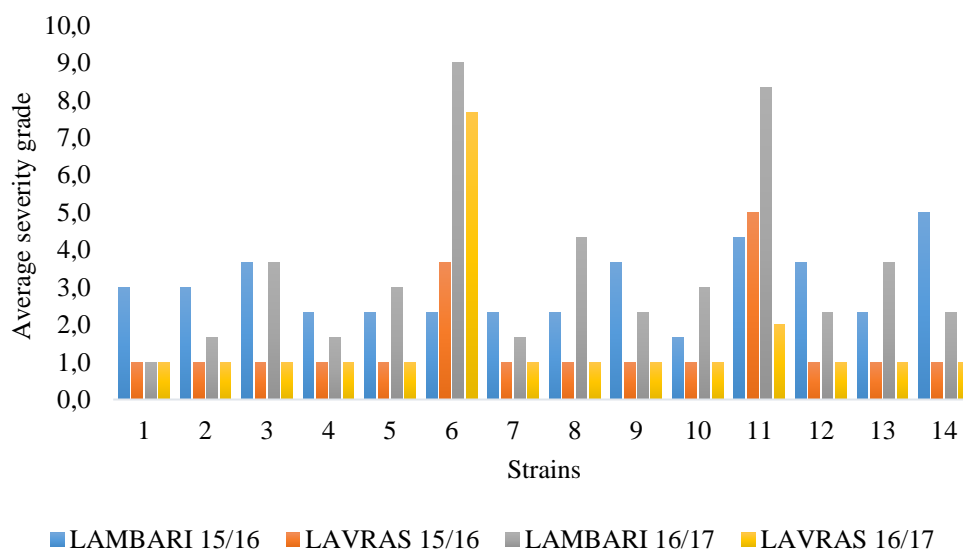
**FIGURE 3** - Average grade of leaf blast (LB) by location and by cultivar.

Neck blast, as well as leaf blast, has a great impact on the final yield of the cultivar. It is the disease considered to be of greatest importance in the culture because it can affect the plant in all its stages of development, that is, from seedling to mature plant (SANTOS et al., 2009). It causes significant losses in susceptible cultivars or when environmental conditions favor their development.

The direct and indirect losses caused by blast, in leaves and panicles, are greater in upland rice grown in the Brazilian Midwest Region, and can reach, in certain situations, 100% loss (EMBRAPA, 2006). Although the

CUV tests have cultivars with very promising grades, a totally resistant cultivar has not yet been selected.

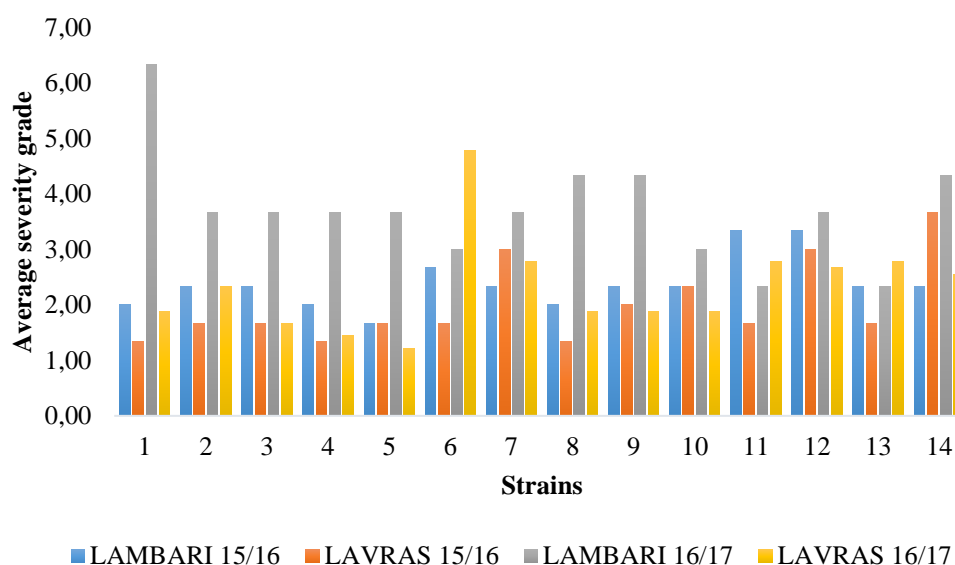
As can be seen in Figure 4, the control BRSMG Caçula and BRSMG Caravera were totally susceptible to neck blast reaching the highest average scores of severity, even when the disease did not show a high incidence. In some cases, they showed almost 100% loss in productivity. This loss occurs because the disease attacks the pedicle of the panicle and can prevent the passage of sap destined for the filling of grains (SANTOS et al., 2011).



**FIGURE 4** - Average grade of neck blast (NB) by location and by cultivar.

The grain spot is also prominent in the culture of rice. It causes great losses in the final yield. Unlike the others, this disease is associated with a complex of pathogens, ranging from fungi to bacteria (YAN et al., 2015). The grains suffer greater breakdown in the processing plant and change in color, which reflects in the

devaluation of the product as well as in the rejection by the consumer, respectively. According to Figure 5, it is possible to observe that the average severity scores were intermediate. The cultivars were not resistant or very tolerant, but in no case was there a severity close to 100%.



**FIGURE 5** - Average grade of grain spot (GS) by location and by cultivar.

The explanations for resistance instability found in this work can be grouped into two topics. The first is the inadequate exposure of materials to the population diversity of the pathogen during breeding programs, where pathogenic diversity is generally high in experimental fields. The second is the high variability of the fungus that causes this disease, which is composed of physiological breeds that have distinct virulence characteristics (FILIPPI et al., 1999). The main control measure, currently, is the use of cultivars with vertical resistance. This type of resistance is governed by one or a few genes that can easily be broken down by the pathogen. The pressure of the inoculum on resistant cultivars in the region shows a rapid drop in the resistance of cultivars, generally from two to three years (SANTOS et al., 2003b). Thus, the planting of larger numbers of resistant cultivars would be an efficient measure to reduce the risks and losses due to these diseases (SANTOS et al., 2002). Thus, it is important to identify the physiological breeds and their frequency to assist genetic improvement programs in the development of cultivars with durable resistance.

## CONCLUSIONS

It is essential to evaluate diseases in breeding programs in order to select increasingly resistant cultivars, as these affect not only productivity, but the quality and final yield of the grains.

The genotypes CMG 2119 (5), CMG 2162 (1), CMG 1896 (13) and CMG 2168 (2) were shown to be more tolerant to all diseases simultaneously, being, therefore, the most suitable for the launch, considering the character disease resistance.

The Upland Rice Breeding Program of UFLA has strains with variability for resistance, strains with the potential to be launched on the market.

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