SIMILARITY BETWEEN CORN HYBRIDS USING SANITARY AND PHYSIOLOGICAL SEED PARAMETERS

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ABSTRACT: One of the elements capable of explaining the epidemics of diseases in the fields of corn production is the amount of initial inoculum present in seeds. The objective of this paper was to study the health and physiological quality of maize hybrids grown in the 2019 crop. The experiment was carried out using 22 hybrids represented by: 13K21PW Morgan®, 14K185PW Morgan®, 230 PRO 3 DKB®, 290 PRO 3 DKB®, 3040 VIP3 LG®, 3055 VIP3 LG®, 30F35 Pioneer®, 310 PRO 3 DKB®, 345 PRO 3 DKB®, 8882 Coded DKB, CBS15C385PW Morgan®, CBS15D089PW Morgan®, CBS15SE137PW Morgan®, Coded x40K205 (3898) Pioneer, K9606 VIP3 KWS®, MG580PW Morgan®, MG652PW Morgan®, SH 7930 PRO 2 Sta Helena®, SH 7990 PRO 2 Sta Helena®, SX5885 VIP3 Syngenta®, SX71715 VIP3 Syngenta® and SX8934 VIP3 Syngenta® that were grown in the municipality of Ipameri, GO. Analyzed the percentage of primary root emission (ERP),% of shoot emission (EPA),% of microorganism incidence, % of fungus genera (% GF). Sixteen fungi were detected representing the microflora detected in the seeds of the evaluated hybrids recognized as Penicillium sp., Aspergillus sp., Curvularia sp., Fusarium verticillioides, Gliomastix sp., Cladosporium sp., Gliocladium sp., Bacillus sp., Geotrichum sp., Trichoderma sp., Mucor sp., Stenocarpella maydis, Pythium sp., Chaetomium sp., Sporotrix sp. and Scopulariopsis sp. The multivariate analysis jointed the cultivars with highest physiological activity represented by SX 8934 Vip3 SYN®, SX 71715 Vip 3®, 3040 Vip3 LG®, SHS 7990 PRO2, SX 5885 Vip3, MG 652 PW®, K9606 Vip3®, DKB 345 PRO3. The lowest incidence of microorganism was in hybrids DKB 345 PRO3 and 8882 cod.

Key words: Corn. Hybrids. Plant pathogen.

SIMILARIDADE ENTRE HÍBRIDOS DE MILHO UTILIZANDO PARÂMETROS SANITÁRIOS E FISIOLÓGICOS DAS SEMENTES

RESUMO: Um dos elementos capazes de explicar a epidemia de doenças nos campos de produção de milho é a quantidade de inoculo inicial introduzido a longas distâncias através das sementes. O objetivo deste trabalho foi estudar a qualidade sanitária e fisiológica de


INTRODUCTION

Approximately 90% of crops that are of economic importance to the world are multiplied by seeds, among these, maize (Zea mays L. - Poaceae) is considered the second most important crop of the world agriculture, and is the first most cultivated cereal in the world. According to the 2019 grain bulletin, the corn maize production estimate will be around 234.1 million tons, this year an increase of 6.4 million tons above the previous harvest. The maize presents a timely production to serve the internal demand, serving for animal feed, being the focus of cultivation in the Brazilian fields in the summer is for soybean production (CARVALHO et al., 2016; COMPANHIA NACIONAL DE ABASTECIMENTO - CONAB, 2019).

Among the many physiological potentials of the seeds, germination, characterized as the return of the embryo growth after the physiological rest period, which results in the rupture of the seed cover and consequently the emergence of the seedling above the ground, can be influenced by several factors such as vitality, viability, longevity, degree of maturity, dormancy, genotype, sanity and environmental factors such as water, temperature and light (MARCOS FILHO, 2015).

The sanity factor involves the association of maize seed with aggressive pathogens, with fungi, bacteria, viruses and nematodes being less frequent (GOMES JUNIOR et al., 2009; MINISTÉRIO DA AGRICULTURA, PECUÁRIA E ABASTECIMENTO - MAPA, 2009). Classically the clustering of cultivars, the adaptability and stability of hybrids have used multivariate analysis as a strategy of recognition and separation (SANTOS et al., 2011). Using multivariate analyzes of fungi and yeasts were used to study the relationship between
genders (ARÍNGOLI et al., 2008). In order to understand the impact of the cotton leaf curl disease in the field, multivariate analysis strategies were used (RAZA et al., 2016).

Field epidemics often start with pathogens associated with seeds, which have high transferability to part of the plants so it is imperative that the producer acquires seed lots with excellent quality and exempt of pathogenic agents, through the use of seeds treated with phytosanitary products and the use of resistant hybrids (CASA et al., 2007). Pathogens can associate seeds directly in the field, where phytopathogenic species that cause diseases in the part of the plant that contaminate seeds predominate (MAPA, 2009). When the seeds are stored, known as storage pathogens (MAPA, 2009), the main fungi are known as molds (Aspergillus sp. and Penicillium sp.). Thus, seeds are considered the main source of dissemination and transmission of pathogens to exempt areas (AGUIAR et al., 2001). For Ferrari et al. (2015) the infected seed is the main source of primary inoculum responsible for determinant curves of epidemic progress.

Therefore, seeds to remain free of pathogens and to prevail their proper physiological quality need to be stored correctly, otherwise seed quality will be impaired, being more prone to deterioration by microorganisms (NAMETH, 1998).

The objective of this paper was to study the health and physiological quality of maize hybrids grown in the 2019 crop.

**MATERIAL AND METHODS**

The experiment started by planting 22 corn hybrids represented by: 13K21PW Morgan®, 14K185PW Morgan®, 230 PRO 3 DKB®, 290 PRO 3 DKB®, 3040 VIP3 LG®, 3055 VIP3 LG®, 30F35 Pioneer®, 310 PRO 3 DKB®, 345 PRO 3 DKB®, 8882 Encoded DKB, CBS15C385PW Morgan®, CBS15D089PW Morgan®, CBS15SE137PW Morgan®, Encoded x40K205(3898) Pioneer, K9606 VIP3 KWS®, MG580PW Morgan®, MG652PW Morgan®, SH 7930 PRO 2 Sta Helena®, SH 7990 PRO 2 Sta Helena®, SX5885 VIP3 Syngenta®, SX71715 VIP3 Syngenta® and SX8934 VIP3 Syngenta®, which were cultivated at the RC Cross Experiment Station, Esmeralda Farm, highway BR 050, latitude: 17° 29'31.35", longitude: 48° 12'56.93", altitude: 908 m, located in the city of Ipameri, GO. The soil has already been characterized as dystrophic yellow red latosol.

The seeds were treated before planting with the active ingredients cytokinin + gibberellin + indol acetic (Stimulate®) at the dosage of 300 mL ha⁻¹. The control of pests, weeds, as well as, the cultural treatments were carried out following the recommended prescriptions for the corn crop.

The hybrids were distributed in five blocks randomly distributed in the experimental area that presented the dimensions of 4 x 10 m (block area), with eight crop lines, spacing of 0.5 m and 0.2 m between plants. Excluding the 0.5 m border in all dimensions, the plots presented a useful area of 27 m².
At 100 days after planting the seeds were collected from 20 ears of the hybrids of corn per block composing sub samples that were mixed, representing a single sample. Seed analysis was performed using the “Blotter Test” method.

Initially the seeds were not disinfested for plating. Individually on a layer of moistened filter paper (2 sheets of blotting paper), remaining spaced 1-2 cm apart, depending on the size of their largest size, inside containers such as Gerboxes or equivalents, characterizing the so-called "Blotter Test".

It was used 250 seeds per hybrid will be taken in the test, composing 10 replicates per hybrid which after plating, will remain under incubation in growth chambers with 12-hour photoperiod for a period of 7-8 days at a temperature of ± 22 °C.

The seeds were examined individually with the help of a stereomicroscope at the resolution of 30-80X, due to the occurrence of typical signal of fungi growth; to prepare semi-permanent slides for observation under the optical microscope and subsequent identification of fungi genus.

The percentage of primary root emission (PRE) was obtained by the ratio of the number of seeds that showed in Gerbox primary roots by the total number of seeds evaluated by Gerbox (25 seeds). The percentage of air part (PAP) emission was obtained by the ratio of the number of seeds that presented Gerbox aerial part by the total number of seeds evaluated by Gerbox (25 seeds). These represented the two main physiological variables of the experiment.

The percentage of microorganism incidence in seeds (MI) was obtained by the ratio of the number of seeds that presented in the germination powder or symptoms of associated microorganisms by the total number of seeds evaluated by Gerbox (25 seeds). The incidence of genera of fungi (IGF) associated with seeds was obtained by the ratio of the number of seeds presenting the genus identified in the Gerbox by the total number of seeds evaluated by Gerbox (25 seeds). It is possible to recognize in a single seed more than one genus of fungus. At the end of the study, a genus of fungi associated with the seeds of the hybrids was described, representing the microflora incident by maize hybrids in the evaluated germplasm. These represented the main health variables evaluated in the experiment. As regras para análise de sementes seguiram as instruções contidas em Brasil (2009).

Statistical analysis of the physiological and health variables were submitted to parametric and non-parametric tests, Tukey averages comparison test at 5% of probability, multivariate analysis of main components, canonical correlations and grouping using as a measure of Malahanobis similarity, all these analyzes used the R program platform.

RESULTS AND DISCUSSION

Sixteen fungi were detected representing the microflora detected in the seeds of the evaluated hybrids recognized as *Penicillium* sp., *Aspergillus* sp., *Curvularia* sp., *Fusarium verticillioides*, *Gliomastix* sp., *Cladosporium* sp., *Gliocladium* sp., *Bacillus* sp., *Geotrichum* sp., *Trichoderma* sp., *Mucor* sp., *Stenocarpela maydis*, *Pythium* sp., *Chaetomium* sp., *Sporotrix* sp. and *Scopulariopsis* sp.
The PRE is a variable that measures the physiological activity of seeds, and the hybrids that stood out with percentages varying from 89.2 to 100% were SX 8934 Vip3 SYN®, SX 71715 Vip 3®, 3040 Vip3 LG®, SHS 7990 PRO2®, SX 5885 Vip3, MG 652 PW®, K9606 Vip3®, DKB 345 PRO3 (Table 1).

Another important physiological variable PRE was the hybrids SX 8934 Vip3 SYN®, SX 71715 Vip 3®, 3040 Vip3 LG® and SHS 7990 PRO2 because they presented the highest average percentages, ranging from 57.2 to 84.8%, differing statistically of the other hybrids (Table 1).

The hybrids that presented the lowest MI averages were DKB 345 PRO3 and 8882 cod, that is, they were genetic materials that had lesser host ability and consequent less ability to transmission plant pathogens to the field (Table 1).

**Table 1.** Averages of primary root emission (PRE), percentage of aerial party (PAP) and microorganisms incidence (MI) in seeds of different corn hybrids.

<table>
<thead>
<tr>
<th>Corn hybrids</th>
<th>PRE</th>
<th>PAP</th>
<th>MI</th>
</tr>
</thead>
<tbody>
<tr>
<td>1. SX 8934 Vip3 SYN®</td>
<td>100.0</td>
<td>84.8</td>
<td>87.6</td>
</tr>
<tr>
<td>2. SX 71715 Vip 3®</td>
<td>99.6</td>
<td>80.0</td>
<td>85.6</td>
</tr>
<tr>
<td>3. 3040 Vip3 LG®</td>
<td>98.0</td>
<td>66.8</td>
<td>88.0</td>
</tr>
<tr>
<td>4. SHS 7990 PRO2®</td>
<td>95.2</td>
<td>57.2</td>
<td>86.8</td>
</tr>
<tr>
<td>5. SX 5885 Vip3®</td>
<td>94.8</td>
<td>71.2</td>
<td>88.0</td>
</tr>
<tr>
<td>6. MG 652 PW®</td>
<td>93.6</td>
<td>36.0</td>
<td>100.0</td>
</tr>
<tr>
<td>7. K9606 Vip3®</td>
<td>92.8</td>
<td>52.4</td>
<td>92.8</td>
</tr>
<tr>
<td>8. DKB 345 PRO3®</td>
<td>89.2</td>
<td>43.2</td>
<td>69.2</td>
</tr>
<tr>
<td>9. SH 7930 Pro 2®</td>
<td>87.2</td>
<td>39.2</td>
<td>100.0</td>
</tr>
<tr>
<td>10. 8882 encoded</td>
<td>86.0</td>
<td>49.6</td>
<td>68.0</td>
</tr>
<tr>
<td>11. 30F35®</td>
<td>82.4</td>
<td>51.2</td>
<td>96.4</td>
</tr>
<tr>
<td>12. Cod X40K205 (3898)</td>
<td>79.6</td>
<td>45.6</td>
<td>86.0</td>
</tr>
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<td>13. 13K21PW MG®</td>
<td>74.0</td>
<td>40.4</td>
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<td>14. DKB 290 Pro 3®</td>
<td>64.8</td>
<td>30.0</td>
<td>100.0</td>
</tr>
<tr>
<td>15. MG 580 PW MG®</td>
<td>62.8</td>
<td>42.8</td>
<td>100.0</td>
</tr>
<tr>
<td>16. CBS 15D089 PW MG®</td>
<td>60.8</td>
<td>14.4</td>
<td>100.0</td>
</tr>
<tr>
<td>17. 14K 185 PW MG®</td>
<td>54.4</td>
<td>12.8</td>
<td>100.0</td>
</tr>
<tr>
<td>18. CBS 15SE137 PW®</td>
<td>46.8</td>
<td>13.6</td>
<td>100.0</td>
</tr>
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<td>19. 3055 Vip3®</td>
<td>38.8</td>
<td>12.8</td>
<td>99.2</td>
</tr>
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<td>20. 310 Pro3 DKB®</td>
<td>30.4</td>
<td>4.4</td>
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</tr>
<tr>
<td>21. CBS 15C385 PW MG®</td>
<td>26.0</td>
<td>7.6</td>
<td>98.4</td>
</tr>
</tbody>
</table>

* Means followed by the same vertical letter do not differ from each other to the Skott-Knott test at P<0.05.
Source: Own authorship.

**Figure 1.** Principal components of corn germplasm using sanitary parameters (incidence of microorganisms - IM, incidence of *Fusarium* sp. - FUS) and physiological (primary root emission, ERP) with all variables.

Source: Own authorship.

The variables that most explained the differences between maize cultivars were the incidence of *Fusarium* sp., Incidence of microorganisms (MI) and primary root emission. The hybrids with the highest incidence of *Fusarium verticillioides* were 16, 9, 6, 8, 15, 1, 18, 14 and 1. And, consequently, they presented lower% ERP and % IM. On the other hand, hybrids with higher MI and ERP were 20, 12, 23, 21, 10 and 4 (Figure 1). When evaluating three maize hybrids, the damage caused by *F. graminearum* and *F. vertillioides* was observed.

Figure 2. Principal components of corn germplasm using sanitary parameters (incidence of microorganisms - IM, incidence of Fusarium sp. - FUS) and physiological (primary root emission, ERP=PRE) selecting more dissimilar variables.

Source: Own authorship.

From the quantitative and binary variables selected that explained the physiological and sanitary variation of maize hybrids were PAP and PRE (both physiological variables) and the incidence of F. verticillioides. The hybrids that presented the greatest physiological activities were 310 PRO3®, 14K 185 PW MG® and S47990 Pro2 S4® (Figure 2). The hybrids with the highest incidence of F. verticillioides (FUS) were X40K205 COD and 290 PRO 3®. The cultivar that presented lower PAP, PRE and FUS was CB 515D089® (Figure 2).
The variables that most explained the small differences among maize hybrids were *Fusarium* sp., incidence of *Cladosporium* sp., incidence of *Penicillium* sp.,% primary root emission and % of percentage of air part (PRE). All hybrids showed little difference since they were very close to the axis (Figure 3).

The sanitary and physiological parameters will allow the separation of the hybrids into 3 groups, the former being represented by CBS 15C385 PW MG®, CB 515D089®, MG 580 PW®, SH7930 PRO 2®, 3040 VIP LG®, 30F35®, SX 5885 Pro2 SH® and S47990 Pro2 S4®. The second group formed by 305S VIP3®, K9606 VIP3®, 310 PRO DKB®, 5X71715 VIP 3®, X40K205 COD, CB5PSE 137 PW®, 230 PRO3® and 290 PRO 3®. Finally, the third group formed SX 8934 VIP SYN®, 882 Cod, MG 652 PW®, SH 49900 PRO2®, SH 345 PRO3®, 310 PRO3®, 14K 185 PW MG® and 290 Pro 3® (Figure 4).

**Figure 4.** Grouping using Malahanobis measure of corn germplasm using sanitary and physiological parameters.

Source: Own authorship.

Plant pathogens of the corn crop (CARVALHO *et al*., 2016), *Fusarium verticillioides* causing ear rot, *Stenocarpela maydis* causing leaf spot in the vegetative stages of the crop and *Pythium* sp. which causes seedling rot. The incidence of *Fusarium* spp. With incidence of 0-9% and transmission rates of 0-100% (COSTA JUNIOR *et al*., 2016). The corn starch grains are often the result of *Stenocarpella macrospora* and *S. maydis*, causing damage to quality and yield when quantified (CASA *et al*., 2007). For two centuries after the original description of the species *S. macrospora* is already causing damage in corn (EARLE, 1897). Using DNA barcoding 46 genera of fungi were detected in corn kernels with symptoms of rot (ABE *et al*., 2015) demonstrating different techniques for the detection and promotion of management practices for use in grain or seed forms.

The reduction of the osmotic potential causes a reduction in the performance of seeds of popcorn, and the hybrids behave differently regarding the tolerance to stress caused by KCl, and the cultivar BRS-Angela presented better germination and seedling growth in relation to the others, when submitted to the same levels of osmotic potential of KCl (MOTERLE *et al*., 2007).

The low availability of water causes a reduction in growth, caused by the decrease of the expansion and the cellular elongation due to the decrease of the turgescence (YASSEEN; ALOMARY, 1994), since the excess moisture causes a decrease in the growth of corn seedlings, since it prevents the penetration of oxygen reducing the active metabolism (BORGES; RENA, 1993).
The microorganisms associated with seed years have been neglected their applications and effects on plant stimulation (MORAIS et al., 2016). On the other hand, these plant pathogen associated organisms associated with imbalance associations in the seed-borne population amplify epidemics, serving as initial inoculum and introduction at long distances in the production fields (DANELLI et al., 2011).

After 16 hours of contact with the signals of the both species, they were sufficient for infection, however, they did not interfere in the germination of maize (RAMOS et al., 2014). In the present study, it was observed a reduction in the performance of seed lots.

The multivariate analysis allows a view of relationships that are often evaluated individually, allowing to verify variables of greater influence and degrees of relationship between variables (HANLEY, 1983).

In the case of grain consumption, the risk of contamination by fumigants from F. verticillioides contaminations in corn seeds may interfere with consumption because many mycotoxins are not thermally sensitive (FIGUEIRA et al., 2003). It was also detected in corn seeds using molecular techniques Penicillium sp., Cladosporium sp. and Fusarium sp., being outstanding for presenting the best enzymatic activity of cellulase (ABE et al., 2015).

CONCLUSION

Sixteen fungi were detected representing the microflora detected in the seeds of the evaluated hybrids recognized as Penicillium sp., Aspergillus sp., Curvularia sp., Fusarium verticillioides, Gliomastix sp., Cladosporium sp., Gliocladium sp., Bacillus sp., Geotrichum sp., Trichoderma sp., Mucor sp., Stenocarpela maydis, Pythium sp., Chaetomium sp., Sporotrix sp. and Scopulariopsis sp.

The cultivars that presented the greatest physiological activities were SX 8934 Vip3 SYN® , SX 71715 Vip 3®, 3040 Vip3 LG®, SHS 7990 PRO2®, SX 5885 Vip3®, MG 652 PW®, K9606 Vip3® and DKB 345 PRO3®.

The lowest incidence of microorganism was in hybrids DKB 345 PRO3 and 8882 cod.

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