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SANITARY QUALITY OF CORN GRAIN SELLED AT CONAB DE PARNAÍBA

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Abstract: This work aimed to analyze the sanitary quality of stored corn grains, through the "Blottertest" and use of the water restriction technique. The experiment was carried out using a completely randomized design (D.I.C.) with 10 treatments and 24 repetitions, in which each repetition consisted of a Petri dish with 8 seeds/plate. The treatments were submitted to NaCl and KCl solution at restriction levels (-0.6; -0.7; -0.8 and -0.9 Mpa), freezing and control. The seeds were distributed in plastic Petri dishes (a total of eight/plate), measuring 90x15mm in diameter, containing three layers of filter paper soaked in the aforementioned solutions. These remained at a temperature of 25°C±2, for a period of 7 days, and were later analyzed for percentage of germination, radicle length and associated fungi, under a stereoscopic and optical microscope. The incidence of the fungal genera Colletotrichum sp., Aspergillus sp., Penicillium sp., Rhyzopus sp., Cladosporium sp., Bipolaris sp., Curvularia sp. and Fusarium sp. The genera with the highest incidence were Colletotrichum sp. (88.6%), Penicillium sp. (4.7%) and Aspergillus sp. (2.5%). The control treatment had the lowest average incidence rate in relation to the water restriction treatment. The lowest percentage of germination (1%) was obtained in the freezing treatment, which provided 9.5% of incidence of fungal genera. The greatest diversity of fungi was observed in the NaCL and KCl treatments (-0.9 Mpa), the same treatments provided the greatest inhibition of germination and radicle emission.
Keywords: Water restriction.Incidence.Fungi. Germination.

INTRODUCTION

Grains in storage can be affected by several microorganisms, including storage fungi such as Fusarium sp., Aspergillus sp. and Penicillium sp., which can also produce mycotoxins that are harmful to human and animal health (Bento et al. 2012; Prestes et al. 2019).

Seed deterioration is a major problem for agriculture, being responsible for serious losses worldwide, especially in the tropics where, in general, high temperatures and relative air humidity prevail during product maturation and storage (Bilia, 1994). Therefore, storage is an important step in the agricultural production chain, as it directly contributes to the quality of the product that reaches the consumer (Costa et al., 2010).

Among the main causes of losses in grain storage is the attack of insects and fungi. Insects, when perforating the grains, facilitate the access of fungi to the tegument, where they initiate the process of deterioration of the same and produce mycotoxins (Souza, 2012). As a consequence, contaminated seeds can reduce the plant population, decrease their productivity and even serve as a vehicle for the dissemination of pathogens, since microorganisms, such as fungi, can be transmitted from seeds to seedlings (Goulart and Fialho, 1998).

Seeds are vulnerable to microbial invasion, once stored, they can be attacked by another group of fungi, called "storage fungi". These fungi are common in nature and can grow on any organic matter that has moisture content and balance with ambient relative humidity of 68 to 90%. (Dhingra, 1985). Thus, the sanitary and physiological quality of corn seeds influence the productivity of this crop due to the role they play both in the establishment of the crop and in the spread of diseases, thus interfering with productivity rates.

Seed analysis aimed at assessing the sanitary quality of a sample is a fundamental procedure with regard to plant defense, seed certification and genetic improvement. The choice of method used to perform such analyzes depends on the type of pathogens,

available conditions and test objectives, among others (Celano et al., 2012). This work aimed to analyze the sanitary quality of stored corn grains, through the Blottertest and use of the water restriction technique.

MATERIALS AND METHODS

OBTAINING THE SEEDS

Hybrid corn seeds were used, obtained from the National Supply Company - CONAB, Parnaíba unit, state of Piauí.

TEST WITHOUT WATER RESTRICTION

The experiment was carried out in a completely randomized design (DIC) with 2 treatments, T1 (untreated corn seed) and T2 (treated corn seed) and 4 replications, each replication consisting of 50 seeds, making a total of 400 seeds.

The seeds were disinfected in 3% sodium hypochlorite for 3 minutes, then washed in distilled water and sterilized (ADE) for 1 minute. Subsequently, the seeds were arranged in plastic Petri dishes measuring 90 x 15mm containing two sheets of sterilized filter paper soaked in ADE. 8 seeds were used per plate.

The Petri dishes containing the seeds were incubated at a temperature of 25°C±2 for 7 days, with a 12-hour photoperiod (12h light/12h dark).

TEST WITH WATER RESTRICTION

The design adopted was (DIC) with 10 treatments and 20 repetitions, each repetition consisting of a Petri dish with 8 seeds each, distributed equidistantly. The treatments were: T1: NaCl (-0.6MPa); T2: NaCl (-0.7MPa); T3: NaCl (-0.8MPa); T4: NaCl (0.9MPa); T5: KCl (-0.6MPa); T6: KCl (-0.7MPa); T7: KCl (-0.8MPa); T8: KCl (-0.9MPa); T9: Freezing; T10: Control (without water restriction or 0.0MPa).

The seeds were disinfected in 1% sodium hypochlorite for 3 minutes, and then washed three times in distilled and sterilized water (ADE) for 1 minute. These were arranged in plastic Petri dishes measuring 90x15 mm with three sheets of filter paper soaked in the aforementioned solutions. Incubation on filter paper substrate with freezing was done by soaking the filter paper in ADE and placing it in the Petri dish, after 24 hours of incubation at a temperature of 25ºC±2. Seeds from this treatment were incubated for 24 hours in a freezer and then returned to a temperature of 25ºC±2. The other treatments consisted of soaking the filter paper in solutions with four levels of osmotic potential (-0.6; -0.7; -0.8; -0.9 MPa) induced by two osmotic solutes (KCl and NaCl) (Table 1).

Osmotic potential (Mpa)	gl-1 of distilled water	
	NaCl	KCl
-0.6	7.71	9.94
-0.7	9.02	11.65
-0.8	10.33	13.36
-0.9	11.64	15.08

Table 1. Amount of product used to prepare NaCl and KCl solutions, at the different osmotic potential levels tested.

The Petri dishes containing the seeds were incubated at a temperature of 25ºC±2 for seven days, with a photoperiod of 12 hours of light (daylight fluorescent lamps) and 12 hours of darkness.

SEED ANALYSIS

Seeds were individually analyzed for percentage of germination, radicle length, incidence of fungus under stereoscopic microscopy. The detection and identification of fungi was performed with the aid of a stereoscopic microscope (magnifying glass) and, when necessary, microscopic preparation was performed using an optical microscope.

STATISTICAL ANALYSIS

The results were submitted to the ANOVA Scott-Knott F test (analysis of variance) and the means compared by the Tukey test at a 5% probability level with the help of the Assistat Software, beta version 7.7 (Silva, 2009). The results that showed zero in the statistics were transformed to X+1, where X is the value of zero in the results.

RESULTS AND DISCUSSIONS

The first test (without water restriction) was not feasible due to the early germination of the seeds, reducing the necessary humidity for the development of the fungus (data not shown). In the test under water restriction, incidences of the genera Colletotrichum sp., Aspergillus sp., Penicillium sp., Rhyzopus sp., Cladosporium sp., Bipolaris sp., Curvularia sp. and Fusarium sp. The highest incidence was of Colletotrichum sp. (88.6%), followed by Penicillium sp. (4.7%), Aspergillus sp (2.5%), Cladosporium sp. (2.08%), Rhyzopus sp. (1.6%), Curvularia sp. (0.08%) and Fusarium sp. (0.08%). Among treatments, the highest percentage of incidence by Colletotrichum sp. occurred in water restriction by KCl (-0.8 Mpa), Penicillium sp. by KCl (-0.6 Mpa) while Aspergillus sp.

The efficiency of the treatments in terms of inhibiting seed germination indicated that the freezing method provided the lowest percentage of germination (1%), while the control provided the highest percentage (93.5%), followed by NaCl (-0.7 Mpa) and KCl (-0.6 and -0.7 Mpa), with germination percentages ranging from 83.5 to 87.9% (Table 2). The results obtained corroborate with Rey et al. (2005) who found that freezing wheat and corn seeds reduced germination levels to zero.

Treatments using NaCl (-0.9Mpa) and KCl (-0.9Mpa) showed the lowest average root growth, being 1.58cm and 1.89cm respectively (Table 2). The inhibition of seed germination by the use of KCl and NaCl solutes has already been reported by Coutinho et al., (2001). Therefore, the results shown here reinforce those in the literature also demonstrated by Araújo et al. (2012).

Treatments	Restriction Levels (Mpa)	Germination (%)	Root Length
NaCl	-0.6	87.2 b	2.0d
	-0.7	92.2 to	2.9c
	-0.8	83.5b	2.3c
	-0.9	83.5b	1.58 d
KCl	-0.6	91.0 to	3.8b
	-0.7	93.5 to	2.9c
	-0.8	87.9 b	2.5c
	-0.9	83.5b	1.89 d
freezing	0	1.0c	1.0 and
Witness	0	93.5 to	7.1 to

1MPa = 10 bar; 1 bar = 0.987 atm. NaCl = Sodium chloride. KCl = potassium chloride. Percentages of germination followed by the same letter do not differ by the Tukey statistical test at the 5% probability level.

Table 2. Germination percentage and radicle length of corn grain submitted to water restriction treatment with sodium chloride (NaCl) and potassium chloride (KCl).

As for the freezing treatment, the seeds submitted to this treatment did not germinate, because, according to Farias et al. (2003), freezing causes cell rupture and, consequently, seed death. In contrast, the effect of water restriction, it only inhibits or reduces root protrusion, without seed death. This way, once the seed is physiologically active, the growth of fungi contained in these seeds may suffer the action of certain metabolites released by the seed. Thus, the values observed in the water-restricted method seem to portray more naturally what happens in this interaction.

The incidence and diversity of fungi for each treatment are shown in Table 3. The greatest diversity of fungi was observed in the NaCl treatment (-0.9MPa), with six fungal genera in this treatment (Colletotrichum sp., Aspergillus sp., Penicilum sp, Rhyzopus sp., Cladosporium sp., and Bipolaris sp.). This same treatment showed germination of 83.5% and the lowest radicle growth (1.58 cm). This result is compatible with those presented by Farias et al. (2003) who, when testing the water restriction technique with different solutes in wheat and corn seeds, found that the most negative potentials were more efficient to control germination.

The results shown here also reinforce those obtained by Machado et al. (2004) who report that water potential in the range of –0.8 to –1.0 MPa, were more efficient to temporarily prevent seed germination and promoted a higher rate of seed infection by the fungi Colletotrichumgossypii, C. gossypii var. cephalosporioides, Botryodiplodiatheobromae and Fusariumoxysporum f. sp. vasinfectum.

CONCLUSION

The use of water restriction is a viable alternative for the analysis of corn seed health, with the potential (-0.9 Mpa) of the NaCl and KCl solutes being the most suitable for the health test because it induced the lowest growth of the radicle and a greater diversity of fungal genera, while the freezing treatment, despite inhibiting the germination of the seeds, did not allow the growth of the microorganisms, preventing the evaluation of the health of the seeds.

TREATMENT	Restriction Levels (Mpa)	Incidence of fungi (%)							
		*Colletotrichum*sp.	*Aspergillus*sp.	*Penicillum*sp.	*rhyzopus*sp.	*Cladosporium*sp.	*Bipolaris*sp.	*Curvaria*sp.	*Fusarium*sp.
NaCl	-0.6	73.1	3.1	3.7	-	0.6	-	-	-
	-0.7	79.4	2.5	3.1	-	-	-	-	-
	-0.8	73.1	1.9	3.1	0.6	2.5	-	-	-
	-0.9	74.4	1.2	3.1	0.6	0.6	1.9	-	-
KCl	-0.6	75.0	-	6.2	3.7	1.9	-	-	-
	-0.7	75.0	3.1	2.5	3.1	1.9	-	-	-
	-0.8	83.7	3.1	5.0	3.1	0.6	-	-	-
	-0.9	73.7	2.5	5.0	1.2	3.1	-	-	-
freezing	0	65.0	-	1.9	-	3.1	-	0.6	0.6
Witness	0	75.0	1.2	1.9	-	1.2	-	-	-

1MPa = 10 bar; 1 bar = 0.987 atm. NaCl = Sodium chloride. KCl = potassium chloride.

Table 3. Diversity and incidence of fungi associated with corn grains submitted to the filter paper test with water restriction.

REFERENCES

ARAÚJO, A. E.; MENTEN, J. O. M.; DIAS, C.T.S; CZERMAINSKI, A.B.C.; SANTOS, J.W.; MORAES, M. H. D.**Efeito de restritores hídricos sobre a germinação, comprimento da radícula e níveis de detecção de Colletotrichumgossypii var. cephalosporioides em sementes de algodão**. Summa Phytopathologica, v. 38, n. 1, p.79-83, 2012.

BENTO, L. F.;CANEPPELE, M. A. B., FIGUEIREDO, M. C.;KOBAYASTI, L.;CANEPPELE, C.;ANDRADE, J. P. **Ocorrência de fungos e aflatoxinas em grãos de milho**. Revista do Instituto Adolfo Lutz, v. 71, n.1, p. 44-49, 2012.

BILIA, D.A.C.; FANCELLI, A.l.; MARCOS FILHO, J. **Comportamento de sementes de milho híbrido durante o armazenamento sob condições variáveis de temperatura e umidade relativa do ar**. Scientia Agrícola, Piracicaba, v. 1, n. 51, p.153-157, 1994.

CELANO, M. M.; MACHADO, J. C.; JACCOUD FILHO, D. S.; GUIMARÃES, R. M.**Avaliação do potencial de uso da restrição hídrica em teste de sanidade de sementes de trigo visando à detecção de fungos**. Revista Brasileira de Sementes, v. 34, n. 4, p.613-618, 2012.

COSTA, A. R.; FARONI, L. R. D.; ALENCAR, E.R.; CARVALHO, M. C. S. FERREIRA, L. G. **Qualidade de grãos de milho armazenados em silos bolsa**. Revista Ciência Agronômica, v. 41, n. 2, p.200-207, 2010.

COUTINHO, W. M.; MACHADO, J. C.; VIEIRA, M. G. G. C.; GUIMARÃES, R. M.; FERREIRA, D. F. **Uso da restrição hídrica na inibição ou retardamento da germinação de sementes de arroz e feijão submetidas ao teste de sanidade em meio agar-água**. Revista Brasileira de Sementes, v.23, n. 2, p. 127-135, 2001.

DHINGRA, O. O. **Prejuízos causados por microorganismos durante o armazenamento de sementes**. Revista Brasileira de Sementes, v. 7, n. 1, p.139-146, 1985.

FARIAS, C. A.; DEL PONTE, E.; DAL MAGRO, T.; PIEROBOM, C. **Inibição de germinação de sementes de trigo e milho em teste de sanidade em substrato de papel**. Revista Brasileira Agrociência, v. 9, n. 2, p.141-144, 2003.

GOULART, A. C. P.; FIALHO, W. F. B. **Eficiência de fungicidas no controle de fungos em sementes de milho e seus efeitos na emergência de plântulas em casa de vegetação**. EMBRAPA-CPAO, 19p. 1998.

MACHADO, J. D. C.; OLIVEIRA, J. A. D.; VIEIRA, M. D. G. G.; ALVES, M. D. C.**Uso da restrição hídrica na inoculação de fungos em sementes de algodoeiro (*Gossypiumhirsutum*)**. Revista Brasileira de Sementes, v. 26, n. 1, p.62-67, 2004.

PRESTES, I. D.;ROCHA, L. O.;NUÑEZ, K. V.;SILVA, N. C. **Principais fungos e micotoxinas em grãos de milho e suas consequências**. ScientiaAgropecuaria, v.10, n. 4, p. 559-570, 2019.

REY, M.S.; LIMA, N.B.; SANTOS, J.; FARIAS, C.R.J.; PIEROBOM, C.R. **Crescimento micelial de raças de Colletotrichumlindemuthianum sob diferentes potenciais hídricos**. Revista Brasileira de Agrociência, Pelotas, v. 11, n. 3, p. 309-313, 2005.

SILVA, F. A. S.; AZEVEDO, C. A. V. Principal ComponentsAnalysis in the Software Assistat-StatisticalAttendance. In: **world congress on computers in agriculture**, 7, Reno-NV-USA: American Society of AgriculturalandBiologicalEngineers, 2009.

SOUZA, N. O. S.; PEREIRA, L. F.; MORAIS, F. B.; NETO, F. A.; FAGIOLI, M. Desempenho Agronômico de Genótipos de Milho no Sul do Piauí. In: **congresso nacional de milho e sorgo**, Goiânia, Associação Brasileira de Milho e Sorgo, p. 2845 – 2851, 2010.