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Fungi associated with maize seed discolouration and abnormalities in South-western Nigeria

B.F.Owolade[1], B. Fawole[2] and Y. O. K. Osikanlu[1]
[1]Institute of Agricultural Research and Training, Moor Plantation, Ibadan, Nigeria.
[2]Department of Crop Protection and Environmental Biology, University of Ibadan, Ibadan, Nigeria.

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ABSTRACT

Three seed samples of maize from Ibadan in South-western Nigeria showing different forms of discolouration and abnormalities were screened for associated fungi. *Fusarium moniliforme* and *Aspergillus flavus* were isolated from all the categories of seed tested. The percentage incidence of *F. moniliforme* was significantly higher on seeds which showed white streaks, purple/pink discolouration, discoloured germ end and wrinkling, than that of any other fungus. *Cephalosporium acremonium* and *Nigrospora oryzae* were also associated with seeds that showed white streaks. *Fusarium graminearum* was associated with purple/pink discolouration while *Drechslera maydis, Fusarium semitectum, Curvularia lunata* and *Colletotrichum graminicola* were observed on seeds with brown spots. *Botryodiplodia theobromae* was more predominant on blackened seeds. *Cephalosporium acremonium, B. theobromae, D. maydis* in addition to *F. moniliforme* were mainly associated with seeds showing discoloured germ ends while wrinkled seeds were observed to harbour *F. moniliforme, D. maydis* and *C. graminicola.*

Key words: Diseases, seeds, *Zea mays*

INTRODUCTION

Maize (*Zea mays* L.) is the main cereal crop grown in South-western Nigeria. It is used primarily as a staple food for human consumption, animal feeds and as raw material for industrial purposes.

The maize seeds are known to be attacked by various types of pathogens. Of these, fungi account for over 75% of reported cases (Cassini and Cotti 1979). These fungi may damage seeds by causing seed abortion, shrinking the seed, reducing seed size, seed necrosis, seed rot and physiological alteration in seeds (Neergaard 1979; Umechuruba 1986; Shetty 1988). These affect maize seeds either in storage or in the field causing seed discolouration, seed rotting and caking, mycotoxin contamination and loss of viability (Ullstrup 1974; Oyeniran 1977; Pattern 1981). Infected seeds act as media for survival of these fungi as well as their dispersal to disease-free areas (Agarwal and Sinclair 1997). The objective of this study therefore was to isolate, identify and obtain information on the incidence of various seed-borne fungi associated with different types of discolouration and abnormalities on seeds of three cultivars of maize common among farmers in South-western Nigeria.

MATERIALS AND METHODS

Seeds of three cultivars of maize viz: TZSR-W, DMRESR-W and DMRLSR-W (Tagged samples 1, 2 and 3, respectively), were obtained from the Institute of Agricultural Research and Training (IAR&T), Ibadan and International Institute of Tropical Agriculture (IITA), Ibadan. The three maize cultivars were grown in Ibadan, Nigeria. The seeds were allowed to dry on the field, and samples were collected immediately after harvest on 10th and 12th of August 1997 and 1998. These seeds were subjected to visual observation and examination under stereoscopic microscope. Seeds that showed distinct symptoms were selected and categorized into three groups viz: discoloured seeds, wrinkled seeds and seeds with discoloured embryo end. Discoloured seeds were further sub-grouped into seeds with white streaks, seeds with brown spots, seeds with pink/purple discolouration and blackened seeds.

Isolation of fungi from seeds

Infected seeds of each cultivar were surface-sterilized in 2% available chlorine in NaOCl for 15min and then rinsed for 2min each in three changes of sterile distilled water prior to plating. Five seeds were plated in each Petri dish containing 10ml of potato dextrose agar (PDA). In each of the categories and subgroupings, a total of 400 seeds were plated in four replicates of 100 seeds per replicate for each cultivar. These Petri dishes were

arranged in a Gallenkamp illuminated growth chamber (model 3B5202B) at a temperature of 28 °C under alternating cycles of 12h light and 12h darkness. On the 8th day, incubated seeds were observed for fungal growth and identification under stereoscopic and compound microscopes. Identification was on the basis of the presence and the characteristics of typical fungal structures such as conidia and hyphae (Barnett and Hunter 1972; Benoit and Mathur 1970; Chidambaram *et al.* 1973).

The number of seeds infected by each kind of fungus was counted and when more than one fungus grew on the same seeds, it was regarded as multiple infection. The data collected were transformed prior to analysis using square root transformation method. Analysis of variance and mean separation were performed using Statistical Analysis Software.

RESULTS

Fusarium moniliforme, Cephalosporium acremonium, and *Nigrospora oryzae* were associated with white streaks radiating from the embryonic ends of the maize seeds. However, the incidence of *F. moniliforme* was significantly higher than other fungi in all the three cultivars tested (Table 1). On cultivar TZSR-W, *F. moniliforme* was identified from 98% of seeds that showed white streaks. The incidence of *F. moniliforme* on seeds that showed purple/pink discolouration was significantly higher than any other fungus isolated on cultivars TZSR-W (89.5%) and DMRLSR-W (52.0%). The percentage incidence of *F. graminearum* was also significantly high on cultivars DMRESR-W (49.0%) and DMRLSR-W(49.5%). *F. semitectum* was also isolated although in traces (0.5%) in seeds exhibiting purple/pink discolouration in cultivar DMRLSR-W.

Fusarium moniliforme, Drechslera maydis, Curvularia lunata and *Colletotrichum graminicola* were observed on seeds with brown spots. The incidence of *D. maydis* (45%) on cultivar TZSR-W was significantly higher than that of other fungi isolated from brown spotted seeds.

Fusarium moniliforme, D. maydis and *Botryodiplodia theobromae* were mostly associated with blackened seeds of the three cultivars tested. The percentage incidence of *B. theobromae* on the blackened seeds was significantly higher than any other observed fungi (Table 1). Table 1 also shows that the incidence of *Aspergillus* spp was significantly high on the seeds of all the cultivars regardless of the type of discolouration.

Table 2. Percentage incidence of fungi in seeds with discoloured embryonic end and wrinkled seeds of three maize cultivars.

	Seeds with discoloured embryonic end			Wrinkled seeds		
Fungus	1*	2	3	1	2	3
Fusarium moniliforme	88.5a**	69.8a	74.0a	72.8a	56.0a	48.5a
Cephalosporium acremonium	9.0b	2.5c	0.5c	0.0d	0.0c	0.0c
Drechslera maydis	11.5b	13.5b	8.0bc	11.3bc	2.5c	9.5b
Botryodiplodia theobromae	8.0b	9.5bc	6.5bc	10.0bc	0.0c	0.0c
Colletotrichum graminicola	0.0c	0.5c	2.5bc	7.5c	9.8b	0.5c
Curvularia lunata	0.0c	0.0c	10.5b	2.0c	C.5c	2.0c
Aspergillus spp	16 0b	21.5b	18.0b	21.0b	43.0a	46.0a
Penicillium spp	0.0c	0 0c	0.0c	0.0c	6.8b	12.5b

*1=Cultivar TZSR-W, 2=DMRESR-W, 3=DMRLSR-W
**Each value is a mean of four replicates (100 seeds/replicate/abnormality).
Means followed by same letter(s) in the same column are not significantly different at P 0.05 (Duncan's Multiple Range Test).

Table 2 shows that the incidence of *F. moniliforme* was significantly higher than that of *C. acremonium, D. maydis, C. graminicola* and *Aspergillus* spp on seeds showing distinct discoloured embryo end in all the three maize cultivars used. Of all the fungi isolated from wrinkled seeds, the percentage incidence of *F. moniliforme* was the highest. *D. maydis, B. theobromae, C. graminicola* and *Curvularia lunata* were other fungi associated with wrinkled seeds. The incidence of *Aspergillus* spp was however, significantly higher in the varieties 2 and 3.

DISCUSSION

The seed-borne fungi isolated include both field and storage fungi. *F. moniliforme, D. maydis, B. theobromae, C. acremonium,* and *Colletotrichum graminicola* were the most important field fungi

Table 1: Seed-borne fungi associated with four different types of discolouration on seeds of three maize cultivars (% incidence)

	White streaks			Pink/purple			Brown spots			Blackened seeds		
Fungus	1*	2	3	1	2	3	1	2	3	1	2	3
Fusarium moniliforme	98.0a	83 0a	87 0a	89 5a	38.5b	52.0a	0 0d	18.5b	26.3a	8.5c	26.5b	20 5b
Fusarium graminearum	0.0c	6.5bc	0.5c	14.0b	49.0a	49.5a	0 0d	0.0d	0.0c	0.0d	0.0c	0.0d
Fusarium semitectum	0.0c	0.0c	0.0c	0 0c	0.0d	0 5c	11 0c	6.5c	2.5c	0.0d	0.0c	0.0d
Drechslera maydis	0.0c	0.0c	0.0c	0 0c	0 0d	0 0c	45.8a	10.8bc	6.0bc	15.0c	21.0b	28.3b
Curvularia lunata	0.0c	0.0c	0.0c	0 0c	0.0d	0 0c	10 0c	9.5bc	13.8b	0.0d	0.0c	0.0d
Colletotrichum graminicola	0.0c	0.0c	0 0c	0 0c	0 0d	0 0c	0.0d	15.8b	18.5ab	17.8c	0.0c	0.0d
Cephalosporium acremonium	5.5c	12 5b	10 8b	0 0c	0 0d	0 0c	0 0d	0 0d	0.0c	0.0d	0.0c	0.0d
Diplodia maydis	0.0c	0.0c	0.0c	0 0c	0 0d	0 0c	19 0b	0.0d	0.0c	0.0d	0.0c	0.0d
Nigrospora oryzae	0.0c	6.0bc	1.5c	0.0c	0.0d	0.0c	0.0d	0.0d	0.0c	0.0d	0.0c	0.0d
Botryodiplodia theobromae	0.0c	0.0c	0.0c	0.0c	0.0d	0.0c	0.0d	0.0d	0.0c	63.0a	49.5a	53.5a
Aspergillus spp	21.0b	14.5b	10.3b	18.5b	13.3c	16 5b	28 5b	35.0a	26.0a	31.0b	17.5b	13.5c
Penicillium spp	0.5c	1.8c	3.5bc	0 0c	0.0d	0 0c	0.0d	0.0d	0.0c	0.0d	0.0c	0.0d

* 1= cultivar TZSR-W; 2=DMRESR-W, 3=DMRLSR-W
** Each value is a mean of four replicates (100 seeds/replicate/cultivar/abnormality)
Means followed by the same letter in the same coloumn are not significantly different at P 0.05 according to Duncan's Multiple Range Test.

isolated in the study. Other field fungi included *Curvularia lunata*, *F. semitectum* and *Nigrospora Oryzaae*. The storage fungi were *Aspergillus* spp and *Penicillium* spp.

The association of *F. moniliforme* with all the different types of seed discolouration and abnormalities, and its high incidence may be due to the susceptibility of the maize crop to attack by this fungus. *Fusarium moniliforme* is known to cause seed and ear rots, stalk rot and leaf spots (Headrick and Pataky 1989). Thomas and Buddenhagen (1980) and Zummo and Scott (1992) have also observed a higher incidence of *F. moniliforme* from maize seeds. In the present study also, this fungus was predominant in the discoloured and wrinkled seeds.

Maize white streaks, often radiating from embryonal end of the seeds, are due to infection by *F. moniliforme*, *C. acremonium* and *N. oryzae*. Although Kumar (1986) observed the association of *F. moniliforme* and *C. acremonoium* with white streaks in maize seeds, *Nigrospora oryzae* is reported eed for the first time. *Fusarium moniliforme* and *F. graminearum* was associated with pinkish to purple seeds. A similar observation was reported by Neergaard (1979). Brown spots on maize seeds were associated with *F. moniliforme*, *F. graminearum*, *D. maydis*, *C. lunata* and *F. semitectum*. Blackened maize seeds were observed to be infected with *B. theobromae*, *D. maydis*, *F. moniliforme* and *C. graminicola*. However, Kumar and Shetty (1983), Neergaard (1979;1981) and Singh and Singh (1981) observed that blackening of seeds of maize is usually caused by *B. theobromae*. Infection levels ranging from 0.5% to 71.5% have been observed on blackened seeds by Singh and Singh (1981). Similar results were obtained in the present study.

Aspergillus and *Penicillium* spp. were the storage fungi isolated from the different forms of dicolourations and abnormalities. *Aspergillus* spp were constantly observed in all the different categories of seeds found in all the three cultivars tested. The influence of these fungi on germination and growth of maize plants is, however, negligible (Kumar and Shetty 1983). Some *Aspergillus* spp. are, however, destructive on stored products and their presence is indicative of poor storage conditions. The findings in this investigation suggest that maize seeds with various forms of discolouration and abnormalities have the potential to cause seed deterioration in storage and epiphytotics in the field.

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