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Seedborne Infection of *Eleusine coracana* by *Bipolaris nodulosa* and *Pyricularia grisea* in Uganda and Kenya

S. PANDE, Plant Pathologist, International Crops Research Institute for the Semi-Arid Tropics (ICRISAT), Patancheru P.O., Andhra Pradesh 502 324, India; S. Z. MUKURU, Principal Cereals Breeder, ICRISAT Eastern Africa Regional Cereals and Legumes Program, P.O. Box 39063, Nairobi, Kenya; R. O. ODHIAMBO, Research Officer, Department of Plant Pathology, Kenya Agricultural Research Institute, National Research Centre, P.O. Box 57811, Muguga, Nairobi; and R. I. KARUNAKAR, Senior Research Associate, ICRISAT, Patancheru P.O., Andhra Pradesh 502 324, India

ABSTRACT

Pande, S., Mukuru, S. Z., Odhiambo, R. O., and Karunakar, R. I. 1994. Seedborne infection of *Eleusine coracana* by *Bipolaris nodulosa* and *Pyricularia grisea* in Uganda and Kenya. Plant Dis. 78:60-63.

Seed samples of finger millet (*Eleusine coracana*) collected from Uganda and Kenya had mixed infections with *Bipolaris nodulosa* (4-49%) and *Pyricularia grisea* (7-55%). Most samples showed poor germination and were unfit for sowing because of a high percentage of seedborne infection. We obtained similar seed infection counts with the standard blotter, deep-freezing blotter, and agar plate methods, which suggests that any of these methods could be used for routine seed health testing. However, sporulation was better on frozen seeds. Gray or black discolored seeds had higher levels of infection by *P. grisea* than apparently healthy, normal reddish brown seeds. Plating of seed components showed that both fungi were present in the pericarp and endosperm but not in the embryo. Seed transmission tests demonstrated the ability of these fungi to kill young seedlings.

Finger millet (*Eleusine coracana* (L.) Gaertn.), locally known as *wimbi* (in Kiswahili), is widely cultivated as a subsistence rain-fed crop in Kenya and Uganda. The grain is used exclusively for human consumption. Grain yields are low in these countries (8,14), primarily because of two important diseases: blast, caused by *Pyricularia grisea* (Cooke) Sacc.; and blight, caused by *Bipolaris nodulosa* (Berk. & M. A. Curtis) Shoemaker. Both pathogens are seedborne in finger millets (3,10-13) and may cause heavy losses. McRae (9) and Govindu (2) reported more than 50% reduction in grain yield caused by blast.

Several fields in Kenya and Uganda were surveyed during late 1991 and early 1992. Most of the fields had severe incidence of blast and blight. Relatively little is known about the seedborne phase of blast and blight diseases of finger millets grown in Uganda and Kenya. We sampled seeds of finger millet grown in these two countries for seedborne infection by *B. nodulosa* and *P. grisea* and attempted to identify an effective method for routine seed health testing for these two pathogenic fungi.

MATERIALS AND METHODS

Twenty seed lots were collected from fields, village markets, and farmers' storage bins and analyzed for infection by *B. nodulosa* and *P. grisea*. Five randomly selected samples were later assessed by three methods to establish a routine testing procedure for these pathogens. A component plating technique was used to locate these fungi in the seed parts. Disease transmission from seed to plant was studied by the water agar seedling symptom test.

Seed samples were collected during November and December 1991 and January and February 1992. Samples were stored in a refrigerator at 5 ± 1 C. Seeds were tested at the plant pathology laboratory of the Kenya Agricultural Research Institute at Muguga, following standard procedures of the International Seed Testing Association (5). Three methods were used: the blotter method, deep-freezing blotter method, and agar plate method.

Blotter method. Twenty-five seeds were placed on two layers of moistened filter paper and one layer of folded tissue paper in a plastic petri dish. Seeded petri dishes were incubated in a chamber at 22 ± 2 C with alternating 12-hr cycles of near-ultraviolet light (NUV) and darkness. After 6 days, seeds were examined for fungal infection under a stereoscopic binocular microscope. One hundred seeds of each of the 20 samples were tested. Another 200 seeds of each of the five randomly selected samples were tested to compare the efficacy of the blotter method with that of the other two methods.

Deep-freezing blotter method. Seeds of the five selected samples were plated on blotters and tissue paper as in the standard blotter method. After incubation in the dark at 22 ± 2 C for 24 hr, petri dishes were placed in a freezer at −4 C for the second and third days, followed by 5-6 days at 22 ± 2 C with 12-hr alternating cycles of NUV and darkness. Two hundred seeds of each sample were tested. Fungal infection counts were determined as in the standard blotter method.

Agar plate method. Fifteen seeds per plate were sown on potato-dextrose agar (PDA) (Difco Laboratories, Detroit, MI) after treatment with an NaOCl solution (2% available chlorine) for 8-10 min. Incubation conditions were similar to those used in the blotter method. After 6 days, colonies were examined for mycelial growth, color, pigmentation, and sporulation. Two hundred seeds of each of the five selected samples were tested by this method.

Component plating. Component plating (7) was used to study the location of the pathogens in seed tissues. Twenty seeds of each of the five selected samples were soaked individually overnight (18-20 hr) in sterilized distilled water and then dissected aseptically with sterilized needles and forceps under a stereoscopic binocular microscope. The pericarp, endosperm, and embryo from each seed were surface-sterilized with 1% NaOCl for 1-2 min, rinsed four or five times in sterilized distilled water, and then plated either on sterilized wet blotters or on PDA in petri dishes equidistant from each other. Incubation conditions were the same as described for the blotter method.

Discolored versus apparently healthy seeds. The blotter method was used to examine 200 gray to black discolored seeds and 200 normal reddish brown seeds of each of two seed lots for the associated fungal pathogens.

Seed transmission. We used the water agar seedling symptom test suggested for the detection of *Septoria nodorum* in wheat grains (6) for our seed transmission studies. The test was conducted in test tubes, with one seed per tube on 20 ml of 1% water agar. Symptoms were recorded after incubation for 10-12 days. To confirm transmission of disease from seeds to seedlings, isolations on PDA were made from the root, attached seed,

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infected with *B. nodulosa*, but we did not observe such spots on diseased grains of *E. coracana*.

Our comparison of three seed health testing techniques indicated that these two pathogens can be successfully detected by any of the methods. However, the standard blotter method is economical and less cumbersome than the other two methods. Furthermore, it restricts the growth of *B. nodulosa* and *P. grisea* to the seed surface, which helps in identification. Sporulation of the two fungi was greater on incubated seeds tested by the deep-freezing blotter method than by the blotter method and the agar plate method, as has been reported previously (12), and this facilitated their detection and identification (12). Furthermore, in both blotter methods, roots and shoots from germinating seeds did not complicate the observations. In addition, developing symptoms on the seedlings infected by *B. nodulosa* and *P. grisea* were more prominent in the blotter method than in the other methods.

Plating of seed components suggested that most of the infection by these two fungi is in the pericarp and endosperm. Embryos were free of infection. However, we did not observe 100% infection of pericarps by these fungi, as reported by Ranganathaiah and Mathur (12). Our results on embryo infection support their findings.

Transmission tests indicated that both *B. nodulosa* and *P. grisea* are seed-transmitted. Both pathogens sporulated on ungerminated, infected seeds and on rotten or necrotic tissues of seedlings and were able to kill the seedlings. Mitra and Mehta (10) and Ranganathaiah and Mathur (12) also reported seedling mortality.

Our results show that *B. nodulosa* and *P. grisea* are seedborne, are confined mostly to the pericarp, and can cause seedling mortality. Seed treatment with effective fungicides will reduce the initial seedborne inoculum and eventually help in obtaining a healthy crop (1,14). The standard blotter test is effective for routine seed health testing to detect these two fungi.

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