Global Journal of Agricultural Sciences. ISSN 1596-2903. Vol. 2, No 2, 2003.90 - 92

FUNGAL DISEASES OF RICE IN NURSERY FARMS IN BAYELSA STATE OF NIGERIA

G.L. FRANCIS-OTOKITO and C. I. UMECHURUBA

(Recieved 15 June 2002; Revision accepted 7April 2003)

ABSTRACT

A survey of fungi associated with seed, seedling and straw samples of three rice varieties (Faro12, Faro 15, and Maliong) obtained from 18 nursery farms in Okuokpoti-Ogbia, a major rice producing community in Ogbia Local Government area of Bayelsa State, Nigeria was investigated. Soil samples from the farms were also tested for fungi. A total of 36 samples were tested per study parameter per rice variety. The standard blotter method was used to test the seed, seedling, and straw samples and the soil samples were tested using the serial dilution plate technique on acidified potato dextrose agar (APDA) medium in Petri dishes. *Botryodiplodia theobromae, Fusarium moniliforme, Fusarium oxysporum, Glomerella cingulata and Penicillium oxalicum* were detected in seed and seedling samples of the three rice varieties at varying percentages. *Fusarium oxysporum* gave the highest percentage occurrence in seeds (48.7%) and seedlings (44.4%) while *F. moniliforme* gave the lowest percentage occurrence in seeds (19.1%) and seedlings (16. 7%). *Botryodiplodia theobromae* (44.4%), *F. oxysporum* (61.1%) and *G. cingulata* (11.1%) were isolated from rice straws while *B. theobromae* (48. 4%), *F. moniliforme* (5.6%), *F. oxysporum* (33.5%) and *P. oxalicum* (10.7%) were isolated from soil samples. All the fungi isolated were found to be seed-borne, seed transmissible and pathogenic except *P. oxalicum* which was not pathogenic. *Botryodiplodia theobromae* and *G. cingulata* which are important pathogens of legumes, root and tuber crops in the tropics and subtropics are reported to be pathogenic on rice for the first time in Nigeria. This is probably due to mixed cropping system. The significance of seed health testing in agricultural production and economy is discussed.

KEY WORDS: Fungal disease, rice, Bayelsa State, nursery farms, Nigeria

INTRODUCTION

Otuokpoti-Ogbia is a major rice producing community in Ogbia local government areas of Bayelsa States of Nigeria. It is estimated that this community produces about 700 tones of milled rice annually for commercial purposes and the people depend on it as a source of starch, protein and fats and for livestock feed. (Personal Communication, Farm Manager, Primabiri Rice Station, Bayelsa). Unfortunately, rice production in the area is adversely affected by many rice diseases such as seed, seedling, foliar, stem and sheath, root, inflorescence and storage diseases. A preliminary visual inspection survey of the rice nursery farms followed by oral interview of the farmers in the area showed that the rice farms had the following symptoms: stunting of seedling, leaf spots, chlorosis of foliage, leaf necrosis, leaf curling, chlorosis of leaf apex, grains discoloration, grains without filling, root distortion and stem rot. Myriad of fungi causing serious devastation in rice nurseries, fields and storage have been reported in major rice growing countries of the world (Mathur and Neergaard, 1970; Neergaard, 1970; Ou, 1985; Dennis, 1994) including some parts of Nigeria (Awoderu, 1972) but none has been reported in Bayelsa State, Nigeria. This study was therefore undertaken to isolate and identify fungi associated with rice seeds, seedlings, straws and soil in 18 nursery farms in Otuokpoti-Ogbia in Bayelsa State. In addition, the pathogenic potential of fungi isolated was determined.

MATERIALS METHODS

Sources of Materials Used

Rice varieties used for the study were obtained from Otuokpoti- Ogbia Local Government Area, which was divided into three rice zones for convenience namely: North-East Otuokpoti zone, which covers Okarabholabhu, Owuru, Ewogbo and the Amori-Obele swamps; the south Otuokpoti zone which includes the Iyoduali, Eyadege and Ogbodo swamps and the west Otuokpoti zone, which is made up of Olabhu and Oku swamps. Three varieties of rice namely: Faro 12, Faro 15 and Maliong are commonly grown in these areas. Rice seed samples used in the studies were obtained from farmers of 18 rice farms during the 1996 and 1997 planting seasons. Two samples per study parameter per rice variety were obtained in each of the 18 farms. Rice seedlings were obtained from nursery farms where the seed samples were collected. Seedlings of 2 to 3 leaf stage were used. Soil samples were randomly collected at the depth of 3.0cm from the same nursery farms with a soil auger. In each farm a composite soil sample was obtained and from the composite samples, submission samples of 1kg per nursery farm was taken to the laboratory for analysis. Rice straws that had stayed on the farm for over one farming season were collected from the various farm locations. All the samples (seeds, seedlings, straws and soil) were kept in the refrigerator until when used.

Isolation and Identification of Fungi from Rice Seeds, Seedlings, Straws and Soil

The Standard Blotter Method as recommended by ISTA (1999) was used to isolate seed-borne fungi associate with rice varieties in all the samples tested. After surface sterilization of the seeds with 1% sodium hypochlorite for 5 mins, the seeds were plated equidistantly in sterile Petri dishes containing three layers of sterile filter papers soaked in sterile distilled water. Twenty-five seeds were plated per Petri dish.

The seeds were incubated in an incubator at 25°C 2°C for 7 days. A total of 400 seeds were tested per seed sample. At the end of incubation period, each seed was examined under a stereobinocular microscope (5-50X) for fungal growth and

G.L. FRANCIS-OTOKITO AND C. I. UMECHURUBA: Dept. of Plant Science and Biotechnology, University of Port Harcourt, Nigeria.

identification. Identification was done based on spores characteristics as described by Barnett and Hunter (1992), Burgess *et. al.* (1994) and Streets (1975). Whenever necessary, temporary slides were made and examined under a research compound microscope to confirm identification.

Rice Seedlings

Shoots and roots of rice seedlings from nursery farms of the three zones were tested for presence of fungi. The plant parts were cut into small pieces of 6mm in length, surface sterilized with 1% sodium hypochlorite for 3mins and plated on three layers of sterile filter paper soaked in sterile distilled water and placed in Petri dishes. The tissues were incubator at 25°C 2°C for 7 days. At the end of the incubation period, the tissues were examined for fungal growth, and observed fungi were identified and recorded.

Rice Straws

The same procedure used for the seedlings tests were used for the over-seasoned rice straws collected from the rice zones.

Soil

The dilution plate technique as described by (Fox, 1993) was used in assaying the soil. A working sample of 1g was taken from each submission sample and diluted with 9ml of sterile distilled water in test tubes with caps. The test tubes were tagged 10^0. After mixing the solution thoroughly with a 10ml soil pipettes, 1ml of each solution was taken from its test tube and transferred into another test tube tagged 10^{-1} containing 9ml of sterile distilled water. The process of dilution of soil solution with 9ml of sterile distilled water was carried out up to 10^{-5}. After stirring 10^{-5} solution thoroughly, the solution was plated on acidified potato dextrose agar (APDA) medium at the rate of 1ml per petri dish. Ten dishes were used for each soil sample. A total of 18 soil samples were tested. The petri dishes were incubated, examined and fungi identified and recorded as previously described.

Pathogenicity Test

Seven-day old pure cultures of *B. theobromae, F. moniliforme, F. oxysporum* and *G. cingulata* were used for determining the pathogenic potential of each of them. Conidial suspension for inoculation was standardized at 10,000 conidia per ml in gelatin-water. Gelatin (0.1%) was used as a sticker. Rice seedlings of the three varieties (Faro 12, Faro 15 and Maliong) raised to 2-3 leaf stage in sterile humus soil in plastic containers were sprayed with spore suspension of the different fungi. Ten seedlings of each variety were grown per plastic container and at the time of inoculation trimmed down to five. Twenty-five seedlings of each variety were used for each fungal treatment. After inoculation, plants were kept for 2 days in humid plastic bags and then transferred to glass house (25°C 2°C). Disease assessment was carried out 14 days after inoculation. Control seedlings were sprayed with distilled water only. *Penicillium oxalicum* was not tested because it is considered as a weak field pathogen but a major storage pathogen of grains.

RESULTS

Fungi Isolated From Seeds

Five fungi namely: *B. theobromae, F. moniliforme, F. oxysporum, G. cingulata and Penicillium oxalicum* were isolated from the rice seed samples obtained from farmers of 18 rice farms. All the five fungi were isolated from the three rice varieties. The fungi were isolated more from Faro 15 than from Faro 12 and Maliong. *Fusarium oxysporum* was the most isolated field fungus while *P. oxalicum* was the only isolated storage fungus (Table 1).

Fungi Isolated From Rice Seedlings

Botryodiplodia theobromae, F. moniliforme, F. oxysporum, G. cingulata and P. oxalicum were isolated from the seedlings at mean percentage occurrence of 33.3, 16.7, 44.4, 22.2 and 16.7, respectively. *Fusarium oxysporum* was the most isolated fungus followed by B. *theobromae* (Table 1). *Botryodiplodia theobromae, F. moniliforme, F. oxysporum and P. oxalicum* were isolated from the three rice zones while *G. cingulata* was isolated only from North-East and South zones. All the fungi were isolated from the three rice varieties except *G. cingulata*, which was isolated from seedlings of Faro 15 and Maliong only.

Fungi Isolated From Rice Straws

Botryodiplodia theobromae, F. moniliforme and *G. cingulata* were the only three fungi detected in the over-seasoned rice straws. All but *G. cingulata* were isolated in the three zones and varieties. *Glomerella cingulata* was isolated only in Faro 15 variety from South zone. *Fusarium oxysporum* gave the highest percentage incidence (61.1%), while *G. cingulata* gave the lowest percentage incidence (11.1%). *Fusarium moniliforme* and *P. oxalicum* were absent from the rice straws.

Fungi Isolated From Soil

Four fungi namely: *B. theobromae, F.* moniliforme, *F. oxysporum and P. oxalicum* were isolated from soils from the 18 nursery farms in the three rice zones. *Botryodiplodia theobromae* gave the highest percentage incidence (48.4%), followed by *F. oxysporum* (33.3%) while *F. moniliforme* gave the lowest percentage incidence (5.6%). *Glomerella cingulata* was absent from soils in the three rice zones (Table 1).

Table. 1. Mean percentage occurrence of fungi isolated from rice seed, seedling, straw and soil samples collected from 18 rice nursery farms in Okuokpoti-Ogbia Communities in Bayelsa State.

Fungi Isolated	Seed	Seedling	Straw	Soil	Mean Percentage Occurrence
B. theobromae	38.0	33.3	44.4	48.4	41.0± 3.6
F. moniliforme	19.1	16.7	0.0	5.6	10.4± 0.9
F. oxysporum	48.7	44.4	61.1	33.3	46.9± 3.8
G. cingulata	28.4	22.2	11.1	0.0	15.4± 1.5
P. oxalicum	44.4	16.7	10.7	10.7	18.0± 1.9

Pathogenicity Test

All the fungi tested were found to be pathogenic on the seedlings. *Botryodiplodia theobromae* caused leaf chlorosis, root decay and very weak stems. *Fusarium moniliforme* caused chlorosis of leaf apex, leaf chlorosis and root distortion. *Fusarium oxysporum* caused leaf chlorosis, necroses of shoot base lacks of root hairs and leaf spots while *G. cingulata* caused leaf necrosis, necrosis of leaf apex and very short roots.

DISCUSSION

Seeds are both vectors and victims of plant diseases

(Sheppard, 1999). Results of this study showed that the rice seeds tested are vectors and victims of *B. theobromae, F. moniliforme, F. oxysporum, G. cingulata and P. oxalicum* isolated from tested seed samples. The fungi were isolated from all the rice zones and they caused diseases. Large scale distribution of rice seeds is going on domestically and internationally and rice seeds are often produced, processed, packaged, sold and planted in the same location or another or in the same country or another. The movement of these seeds poses increasing danger of the spread of seed-borne diseases. Losses due to seed-borne fungi include: reduction in seed germination, poor seedling vigour, abnormal seedlings and other damages to the crop at any stage of growth from seedling to harvest and storage (Neergaard, 1979; Ou, 1985; Dennis 1994).

All the fungi isolated from seeds were isolated also from the seedlings. A relationship therefore, seems to exist between the occurrence of fungi in seeds and their occurrence in seedlings as reported by Mathur and Neergaard (1970) and Meshram *et. al.* (1993). There were no appreciable differences in the percentage occurrences of these fungi in the seeds and seedlings (Table 1). Since seeds are potential vectors of mycoflora, these fungi might have been transmitted through germinating seedlings (Neergaard, 1979).

The role of rice straw in constituting inoculum source has been reported by several workers (Ou, 1987; Miller *et. al.* 1993). *Botryodiplodia theobromae, F. oxysporum* and *P. oxalicum* were isolated from the over-seasoned straws. This means that these fungi can serve as inocula in the next planting season and the circle of infection and re-infection every planting season will continue if the straws are not eliminated. This vividly explains the basic principles of plant to seed (P S) transfer of pathogens as stated by Neergaard (1979) and Ou (1985).

With the exception of *G. cingulata* all the other fungi are to be soil-borne; thereby constituting inocula vectors of the pathogens (Campbell, 1994). When these fungal pathogens are carried by rice seeds, seedlings, straws and soil into uninfected areas, the high level of rice production and economy in Bayelsa State will be adversely affected. Losses will not be restricted to the fields where the seeds are sown but also in storage. The planting and market values of the crop will reduce drastically (Neergaard, 1979).

Results of this study therefore stress the importance of seed health testing in agricultural production. *Botryodiplodia theobromae* and *G. cingulata* which are important pathogens of legumes, root and tuber crops in the tropics and subtropics are reported to be pathogenic on rice for the first time in Nigeria. This is probably due to mixed cropping system practiced in Nigeria.

REFERENCES

Awoderu, V. A., 1972. Studies on the rice blast disease and its causal organisms. PANS. *20*: 416-424.

Barnett, H. L. and Hunter, B. B. 1992. *Illustrated Genera of* Imperfect Fungi. 4th Edition. Burgess Publishing Company, Minnespolis.

Burgess, L. W., Summerell, B. A., Bullock, S., Gott, K. P. and Backhouse, D., 1994 Laboratory Manual for Fusarium Research 3rd Edition. University of Sydney Press. Sydney. 133p.

Campbell, R., 1994. Biological control of soil-borne diseases: Some present problems and different approaches. Crop Protection. 13(1) : 4-13.

Dennis, J. J., 1994. A survey of rice seed-borne fungi in Taiwan. Plant Disease. 78 (3): 316.

Fox, R. T. V., 1993. Principles of Diagnostic Techniques in Plant Pathology. Cambridge, University Press, Cambridge England. 213p.

ISTA. 1999. International Seed Testing Association. International Rules for Seed Testing. Seed Sci. and Technol. Vol. 27 Supplement. 333p.

Mathur, S. B. and Neergaard, P. 1970. Seed-borne fungi of rice in Philippines, India, Portugal and Egypt: Investigations on *Trichoconis padwickii*. Proc. Of First Inter. Symp. on Plant Pathology, New Delhi. Pages 61-81.

Meshram, S. U.; Gondane, H. G.; Pande, S. S. and Gaikwad, S. J. 1993. Response of seed-borne pathogens of cereal crops to *Azotobacter chocolum* strains. Journal of Biological Control. 7 (2) : 87-92.

Miller, B. C.; Foin, T. C. and Hill, J. E. 1993. A rice model for Scheduling and evaluating management actions. Agronomy Journal 85: 938-947.

Neergaard, P., 1970. Seed Pathology of Rice. In Proc First Inter. Symp. On Plant Pathology. New Delhi. Pages 57-68.

Neergaard, P., 1979. Seed Pathology. Vols, 1 & 11. The Macmillan Press Ltd. London. 1191p.

Ou, S. H., 1985. Rice Diseases. 2nd Edition. Commonwealth Mycological Institute, Kew, Surrey, England. 380p.

Sheppard, Jim, 1999. Importance of Seed Health within Seed Testing. ISTA News Bulletin. 120. October 1 9 9 9 . pages 13-17.

Streets, Sr. R. B., 1975. The Diagnosis of Plant Diseases: A field and laboratory manual emphasizing the most practical methods for rapid identification. The University of Arizona Press, Tuscon, Arizona. 11.11p.