International Journal of Agriculture Innovations and Research
Volume 5, Issue 3, ISSN (Online) 2319-1473
Manuscript Processing Details (dd/mm/yyyy) :
Received : 07/12/2016 | Accepted on : 12/12/2016 | Published : 23/12/2016

Survey of Fungal Seed-Borne Diseases of Rice in Burkina Faso

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***Abstract* – The seed is of particular interest because of its importance as planting materiel and its potential as a vehicle for transmission of fungi. This study was conducted to update the information on the most important seed-borne fungal diseases of rice in Burkina Faso through survey and identification. A set of 151 seed samples was made up from different rice genotypes and from twenty-eight rice growing locations in Burkina Faso. The seed samples were tested by the mean of the blotter and seed washing tests. Thirty-seven fungal species belonging to nineteen genera seed-borne fungi were isolated from different cultivar of *Oryza sativa L.* The most frequent genera isolated were in the order of frequency: *Phoma sorghina* (92%), *Bipolaris oryzae* (84.1%), *Alternaria padwickii* (66.2%), *Curvularia lunata* (51%), *Fusarium moniliforme* (33.1%), *Microdochium oryzae* (30.5%) and *Pyricularia oryzae* (13.5%). However, their incidence varied from 20% to 85% infected seeds. Some fungal species were found in all the rice growing ecologies and most of the released rice varieties in Burkina Faso were found infected with these fungal pathogens.**

***Keywords* – Burkina Faso, Fungi, Rice, Seed-born.**

I. INTRODUCTION

Rice (*Oryza sativa L.*) is the staple food for more than three billion people, over half the world's population. In Africa, where rice is the most rapidly growing food source, about 30 million tons more rice will be needed by 2035, representing an increase of 130% in rice consumption from 2010 [19]. In Burkina Faso, rice is the fourth most cultivated cereal after sorghum, millet and maize [6]. The average yearly paddy production is estimated to 360. 000 metric tons and covers only one third of the demand of the population [6]. Concomitantly, rice productivity is prone to many biotic and a biotic factors affecting yield including diseases. Rice seed is known to be colonised by various types of fungi, among which many are identified as plant pathogens. It is known to be infected by 56 fungal pathogens [15], of which 41 are reported to be seed-borne [17], [18]. Seed-borne fungi are then one of the most important biotic constraints in seed production worldwide. Extensive and deep studies on seed-borne fungi of rice have been carried out all over the world. They cause serious damage to rice production with yield losses ranging from 3 to 100 %. In India, yield losses due to brown spot caused by *Bipolaris* oryzae ranged from 50 to 90% [8]. In Nigeria, 40% of yield reduction was attributable to blast incited by *Pyricularia oryzae* [3] and brown spot has emerged as a major constraint [1]. Germination was reduced from 29% and up to 80% in seed lots infected by *Alternari padwickii*, *Bipolaris oryzae* and *Curvularia lunata* [20], [5] and yield losses due to sheath blight (*Sarocladium oryzae*) were considered higher than 50% in India [12]. In Morocco, *Bipolaris oryzae* was found in 31% rice seeds; and from 94% to 100% rice seeds were infected with *Fusarium moniliforme* and 6% with *Pyricularia oryzae* [9]. In Uganda, the incidence of seed infection with *B. oryzae* varied from 3 to 94% [4].

Seed-borne fungi are responsible for both pre- and post-emergence death of grains, affect seedling viability, vigour, and thus cause some reduction in germination capability and also variation in plant morphology [16], [14]. The promotion of crop yield and quality greatly depends on the quality of seeds and often seed determines the yield. Moreover, there is an important movement of planting material across the borders with neighbouring countries. In this context, it appears clearly that importance of disease-free, viable seeds in crop production cannot be overemphasized. One of the basic strategies to produce certificated and non-infected seeds is the identification of seed-borne pathogenic agents in rice-growing fields. Unfortunately, only scarce data on seed borne fungal diseases occurring on rice in the country are available. This information is necessary for local producers to understand the seed-borne fungal pathogens status infecting rice and their frequency to effectively apply control strategies. Thus, it is urgent to settle sound databases that will accompany rice diseases management in Burkina Faso and the neighbouring rice growing countries. The aim of our study was to update the information on the most important seed-borne fungal diseases of rice in Burkina Faso through survey and identification. The importance, management challenges and the suggested way forward of rice seed-borne diseases management was discussed.

II. MATERIAL AND METHODS

II. 1. Collection of Seed Samples

Seed samples (151) were collected from various rice genotypes in twenty-eight (28) locations across rice growing regions in Burkina Faso (Fig. 1) according to the International Rules for Seed Testing recommendations [10]. The seed samples were collected in 2013 and 2014. Upland rain fed, lowland rain fed and irrigated rice growing ecologies were concerned by the survey. The seed samples were taken from the most recent seed lots and kept at 5°C throughout the period of the study.

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International Journal of Agriculture Innovations and Research
Volume 5, Issue 3, ISSN (Online) 2319-1473

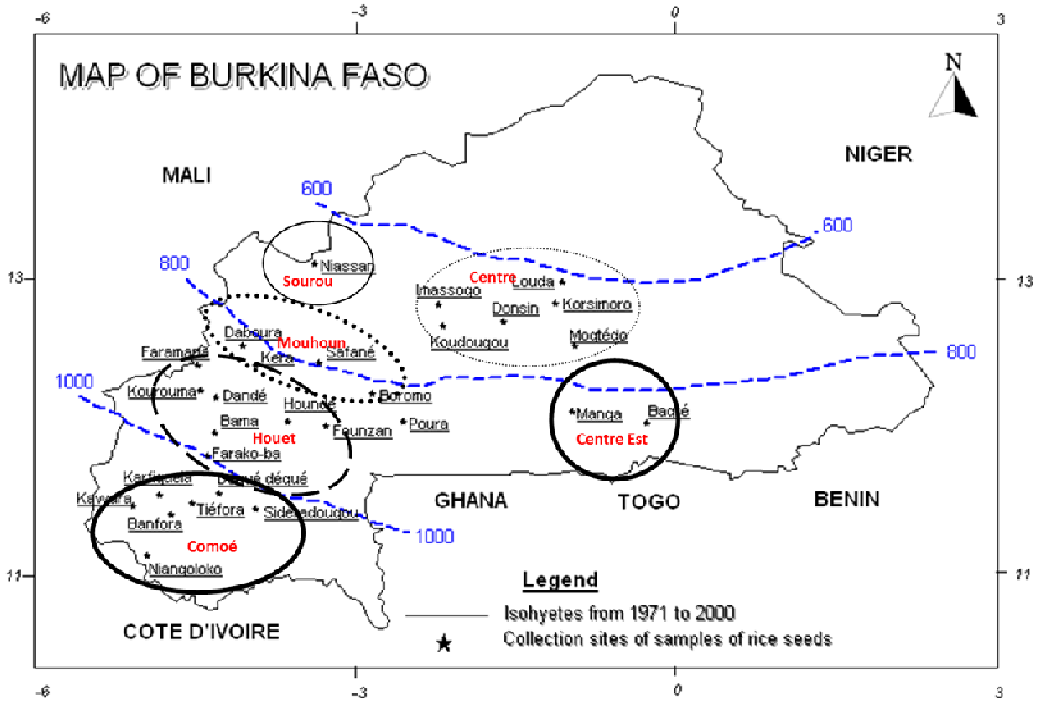

Figure 1. Rice seed sampling locations in Burkina Faso

II. 2. Isolation and Identification of Fungi

Samples of different rice cultivars were used for the isolation and identification of seed-borne fungi using the standard methods as described by Mathur and Kondgsdal [11], notably the seed washing and deep freezing blotter methods.

Seed washing method. The detection of fungal species such as Tilletia barclayana and Ustlaginoidea virens was performed by washing seed samples in sterile water with 1-2 drops of Tween 20then shaked vigorously for 5-10 minutes. The water was collected and filtered through a piece of cheese cloth and transferred in 1. 5 ml tubes. The suspension was centrifuged at 1500-3000 rpm for 2-10 mn. The sediment is collected and re-suspended into 1 ml of Shear' solution (19. 212 g citric acid, 28. 392 g disodium phosphate, 6 g potassium acetate, 120 ml glycerol, 180 ml ethyl alcohol). The collected spores are examined under the compound microscope.

The blotter test: For each sample, four replicates of 100 seeds were tested for health by the blotter method: 25 seeds were placed in 9cm diameter Petri dishes on top of three blotters moistened with sterile water. The seeds were incubated at 25 °C for seven days under alternative cycles of 12 hours of darkness and near-ultra violet light. Kongsdal and Mathur [11] identification key was used to characterize fungi.

Data collection: To assess the health status of the seeds, two data were collected: (i) the frequency (F) of the different fungal species expressed as the percentage of infected seeds and within a single sample (ii) the incidence (I) of the different fungal species expressed as the percentage of seed samples infected. Species such as *Tilletia barclayana* and *Ustlaginoidea virens* were recorded as present or absent.

The frequency (F) and incidence (I)of fungi were estimated according to the following formula:

F (%) = (Number of seeds in which a species occurred)/Total number of seeds

I (%) = (Number of seed samples in which a species occurred)/Total number of seed samples

Three classes were defined to appreciate the frequency (F) and incidence (I) of fungal species: Low= $0 \leq I/F \leq 10$, Moderate= $10 < I/F \leq 50$ and High= I/F> 50%.

III. RESULTS

III. 1. Fungi Frequency and Incidence

The survey revealed the presence of thirty-seven fungal species (Table 1). An important variation in the frequency and the incidence of the different fungi was observed. Out of 37 identified fungal species on rice seeds, we reported that (i) Four had high frequency: *Phoma sorghina* (92%), *Bipolaris oryzae* (84. 1%), *Alternaria padwickii* (66. 2%), *Curvularia lunata* (51%). Belonging these fungi, *Phoma sorghina* and *Bipolaris oryzae* have 85% and 76% of incidence respectively; (ii) Three had moderate frequency: *Fusarium moniliforme* (33. 1%), *Fusariumsemitectum* (45%) and *Microdochium oryzae* (30. 5%);and (iii) Thirteen had low frequency: *Acremoniumspp.*, *Exserohilum rostratum*, *Fusarium equiseti*, *Pyricularia oryzae*, *Nigrospora oryzae*, *Bipolaris bicolor*, *Curvularia oryzae*, *Cercospora janseana*, *Fusarium solani*, *Fusarium oxysporum*, *Sarocladium oryzae*, *Tilletia barclayana* and *Ustilaginoidea virens*. Also, several pathogens were encountered on a single rice seed. The most common

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combination included *Bipolaris oryzae* and *Alternaria padwickii*. The table 2 presented the frequency and incidence of some important fungal species detected in different sampling provinces.

Table 1. Incidence of fungal species on all samples

No	Fungal species	Frequency (%)	Incidence range (%)
1	*Acremonium sp.*	18. 1	0. 5 – 2. 0
2	*Alternaria alternata*	5. 3	2. 0 – 15. 0
3	*A. longissima*	46. 6	5. 0 – 46. 0
4	*A. padwickii*	66. 2	4. 0 - 39. 5
5	*Bipolaris bicolor*	0. 7	0. 5 – 20. 0
6	*B. hawaiensis*	0. 7	0. 5 – 1. 0
7	*B. longirostrata*	0. 7	0. 5 – 1. 0
8	*B. oryzae*	84. 1	2. 5 – 76. 0
9	*B. sorokiniana*	2. 6	0. 5 – 2. 0
10	*Cercospora janseana*	2. 0	0. 5 – 1. 0
11	*Cladosporium herbarum*	7. 9	3. 5 – 15. 0
12	*Curvularia cymbopogoni*	6. 0	0. 5 – 4. 0
13	*C. eragrostidis*	7. 6	1. 5 - 6. 5
14	*C. geniculata*	4. 3	0. 5 - 2. 0
15	*C. lunata*	51. 0	2. 5 – 26. 0
16	*C. oryzae*	2. 0	0. 5 – 1. 0
17	*C. pallescens*	9. 2	0. 5 - 14. 7
18	*Exserohilum rostratum*	17. 9	0. 5 – 6. 0
19	*E. monocera*	0. 7	0. 5 – 1. 0
20	*Fusarium equiseti*	17. 9	1. 5 - 5. 8
21	*F. moniliforme*	33. 1	3. 0 – 20. 0
22	*F. oxysporum*	0. 7	0. 0 – 1. 0
23	*F. semitectum*	45. 0	0. 5 - 11. 0
24	*F. solani*	0. 7	0. 0 – 1. 0
25	*Microdochium oryzae*	30. 5	3. 0 – 35. 0
26	*Myrothecium gramineum*	1. 3	0. 0 – 1. 0
27	*M. roridum*	0. 7	0. 0 – 1. 0
28	*M. verrucaria*	12. 0	1. 0 - 2. 5
29	*Nigrospora oryzae*	12. 0	1. 0 – 5. 0
30	*Phaeotrichoconis crotalaria*	4. 6	0. 5 – 5. 0
31	*Phoma sorghina*	92. 0	18. 0 – 85. 0
32	*Pyricularia oryzae*	13. 3	2. 0 – 55. 0
33	*Sarocladium oryzae*	1. 3	1. 0- 2. 0
34	*Tilletia barcayana*	0. 7	0. 0 – 1. 0
35	*Tricothecium sp.*	1. 3	1. 5 - 3. 0
36	*Ustilaginoidea virens*	2. 0	1. 5 – 2. 0
37	*Verticillium cinnabarinum*	7. 3	0. 5 – 3. 0

Table 2. Frequency and incidence of some important fungal species detected in different provinces

N°	Fungal species	Comoe		Houet		Mouhoun		Sourou	
		F	I	F	I	F	I	F	I
1	*Acremonium sp*	10	1. 5	10	1	8	0. 5	43	1. 5
2	*A. longissima*	27	9. 5	67	15	38. 5	8	38. 5	6. 5
3	*A. padwickii*	64	31	63	23	38. 5	4	86	38. 5
4	*Bipolaris oryzae*	77. 5	41	97	54	77	2. 5	86	76
5	*Curvularia sp*	31. 5	14	73. 5	7. 5	31	3	71	9. 5
6	*F. moniliforme*	20	6	40	20	54	3	86	3
7	*F. semitectum*	40	11	37	2	38. 5	0. 5	43	1
8	*M. oryzae*	44	6	53	35	8	3	0	0
9	*Phoma sorghina*	84	38	100	85	77	26	86	18
10	*Pyricularia oryzae*	11	3. 5	40	55	10	2	8	15

F= Frequency, I: Incidence

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III. 2. Geographical Distribution of the Fungal Pathogens

The collected data revealed the presence of some important fungal pathogens in all the rice growing regions: *Acremonium* sp., *Altrenaria padwickii, Bipolarisoryzae, Fusarium semitectum, Myrothecium verrucaria, Curvularia spp., Phoma sorghina* and *Exserohilum rostratum. Phoma sp., Alternaria padwickii, Bipolaris oryzae, Fusarium moniliforme, Pyricularia oryzae, Curvularia lunata* occurred in more than 65% of the locations. *Fusarium equiseti, Microdochium oryzae, Nigrospora oryzae and Ustilaginoidea virens* were found in 50% of the locations. Several fungi were found in only two or three locations: *Tilletia barclayana, Cercospora janseana, Curvularia oryzae* and *Sarocladium oryzae*. The prevalence of the fungi was different according to the rice growing areas: *A. padwickii* was detected in 38. 5% and 86% of the tested seed samples collected in the Mouhoun and Sourou provinces respectively. *Pyricularia oryzae* was found more frequently in the Houet province (40%) than the other districts (2 - 11%). The inciting pathogen of the false smut (*U. virens*) was detected mostly in samples from the Center region (17%). Almost all the fungi were present in Cascades and Hauts Bassins regions. A very high prevalence (up to 100%) was observed for *A. padwickii, Bipolaris oryzae, Curvularialunata, Fusarium semitectum, Phoma sorghina* in all the rice growing sites.

Significant differences were observed in the incidence of some pathogens according to the rice growing regions. For instance, the highest frequencies of *P. oryzae* and *Microdochium oryzae* occurred in the more humid regions (over 800 mm rainfall) while those pathogens were not found in samples from the locality of Kaya located in a semi-wetland.

High prevalence of the fungi in the tested samples from the different localities was not correlated with high incidence of infected seeds in those tested samples as shown in Table 2. *Fusarium* spp. showed low incidence (under 5%) in all the localities excepted for *F. moniliforme* in Houet (20%) and *F. semitectum* in Comoé (11%). In contrast, *A. padwickii, B. oryzae, and Phoma sorghina* had a high prevalence and incidence in all the localities.

III. 3. Incidence of the Fungi According to Rice Growing Ecologies

Data in Table 3 show variations in the incidence of some fungi according to the rice growing system. For instance, *A. padwickii, Bipolaris oryzae* and *Curvulariaspp.* showed higher incidence under irrigated than rain fed conditions. The incidence of *B. oryzae*, was very high in Sourou (76. 5%) as well as in Vallée du Kou (54. 5%). On the contrary, *F. moniliforme, Microdochium oryzae, Phoma sorghina* and *P. oryzae* were more prevalent under upland rain fed conditions. Under the upland rained conditions on Farako-Bâ site, the incidence of *P. oryzae* and *Microdochium oryzae* was high as 55% and 35% respectively.

Table 3. Incidence (%) of fungal pathogens according to the rice growing ecology

Fungal species	**Upland rained**		**Lowland irrigated**	
	*Farako-Bâ	*Faramana	*Vallée du Kou	Sourou
Acremonium sp.	1. 0	1. 0	1. 0	1. 0
A. padwickii	20. 0	-	19. 5	39. 5
A. longissima	46. 5	17. 8	15. 5	28. 0
B. oryzae	5. 6	9. 0	54. 5	76. 5
Curvularia spp.	8. 5	6. 5	17. 5	9. 4
Fusarium equiseti	1. 0	3. 0	1. 0	1. 5
F. moniliforme	6. 0	37. 0	4. 0	1. 0
F. semitectum	3. 0	2. 0	1. 0	1. 0
Microdochium oryzae	35. 0	-	-	1. 0
Myrothecium sp.	5. 0	-	-	1. 0
Nigrospora oryzae	-	0. 5	-	-
Phoma sorghina	85. 0	10. 0	14. 0	18. 0
Pyricularia oryzae	55. 0	-	-	15. 0
Sarocladium oryzae	1. 0	-	-	-
Ustilaginoidea virens	-	-	3. 0	-

***:** locations of the Houet province

IV. DISCUSSION

The results of this study confirm the cosmopolistic character of some pathogenic fungi like *P. oryzae, B. oryzae, Phoma sorghina, Curvularia lunata, A. padwickii, Fusarium moniliforme* [13], [15]. These fungi were observed in the sub region of West Africa: Ghana, Sierra Leone [7] and Nigeria [3]. It is admitted that the flow of planting material throughout the Western African countries borders is a big concern because this common practice is an efficient way of the spread of several major fungal diseases. High incidences of *Bipolarisoryzae* in seed samples cause considerable losses in seed germination because of the high rate (80%) of transmission of the pathogen from seed to seedling [2]. Considering these results, it is obvious that the overall status of rice health in Burkina Faso is critical: several important seed-borne fungi are present in the major rice growing sites of the country. These are sites where rice is being cultivated traditionally what may explain the endemic character of some important pathogenic fungi. The

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lowland irrigated ecosystems, which require high inputs for rice production, seem to be more favorable to some important diseases (brown spot, stack burn). On the contrary, blast was more severe under upland rained conditions; this is probably due to the water stress caused by irregular rainfall during the vinter season. High incidences of fungi such as *Bipolaris oryzae* may indicate the unfavorable status of the soil: lack of potassium, manganese or silicium, salinity, etc. This is the case of the Sourou valley where deficiencies in minerals elements has been reported. Upland rained conditions found in the Farako-Bâ locality are more favorable to the development of blast and leaf scald.

V. Conclusion

Our study provide some indications on the seed quality and so help to identify the localities suitable for seed production. In view of the prevalence of pathogenic fungi on sites prospected, it is urgent to set up an efficient seed production system that will provide farmers with quality seeds. Also, tolerance levels of seed infection by fungi like *P. oryzae*, *B. oryzae*, *A. padwickii*, *Fusarium moniliforme* and *Phoma sorghina* should be established by further studies as far as these fungi show high prevalence and high incidence in almost all the tested samples.

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