

Identification of seed-borne fungi of onion (*Allium cepa* L.) in Burkina Faso

Tobdem Gaston DABIRE^{1,2}, Schémaeza BONZI¹, Irénée SOMDA¹, and Anne LEGREVE²

¹Université Polytechnique de Bobo-Dioulasso,
Institut du Développement Rural,
BP 1091, Bobo-Dioulasso, Burkina Faso

²Université Catholique de Louvain-la-Neuve,
Faculté des Bio ingénieurs,
Earth and Life Institute,
Croix du Sud 2 box L7.05.03,
1348 Louvain-la-Neuve, Belgium

Copyright © 2016 ISSR Journals. This is an open access article distributed under the **Creative Commons Attribution License**, which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited.

ABSTRACT: This study was carried out to assess the seed-borne fungi of onion in Burkina Faso. Eighteen onion seed samples were collected from local farmers and wholesalers of vegetable seeds in the country and were investigated for fungi. The investigation was done using the “blotter method” on dry seeds and on seedlings. Fungal contamination was detected in all 18 tested samples. Seventeen fungal species belonging to 11 fungal genera were identified in the seed samples: *Aspergillus* was detected in 17 samples, *Fusarium* and *Rhizopus* in 15 samples, *Cladosporium* in 14 samples and *Penicillium* in 13 samples. *Aspergillus niger* and *Fusarium oxysporum*, known to be the causal agents of black mould and basal rot diseases, were detected in 17 and 11 samples, respectively, by seed analysis and in 10 and 9 samples, respectively, by seedling analysis. The infection rates by the fungal species varied from 0 to 90.3% for *A. niger* and from 0 to 13.5% for *F. oxysporum*. *Alternaria porri*, the causal agent of purple blotch disease was recorded lowly on two seed samples at infection rates of 0,5 and 1%. Exotic seed samples showed better health quality compared to local seed samples. These results indicated that the seeds locally produced by farmers in Burkina Faso are for low health quality and needs strong treatments before use to avoid diseases appearance in fields.

KEYWORDS: Onion, Seed Health, infection rate, *Aspergillus niger*, *Fusarium oxysporum*, Burkina Faso.

1 INTRODUCTION

Onion production and commercialization has increased greatly in the past 5 years in Burkina Faso. From 17.126 tons in 1997, the national onion bulb production reached 406.760 tons in 2012 [1], [2]. The main reason for this increase is that onion is an attractive crop for farmers, given the level of income estimated at more than US\$ 5,000 per hectare [1], [3]. Biotic and abiotic constraints, however, still limit onion production in Burkina Faso. The greatest constraint is plant disease, which results in considerable damage and in yield losses ranging from 25 to 80% [4]. Some diseases that occur in onion fields in Burkina Faso have caused such significant losses that farmers are abandoning these areas. Symptoms such as seedling damping-off, foliar lesions and bulb rot have been observed, showing characteristics that are typical of fungal pathogen attack (e.g., appearance and distribution modes in fields). The causal pathogens, however, are not well known because they have been little studied. Many fungal pathogens responsible for onion diseases have been reported in different climatic regions of the world and most of them are seed transmitted. They include *Alternaria alternata*, *A. porri*, *Aspergillus niger*, *A. flavus*, *A. fumigatus*, *A. alutaceus*, *Beauveria bassiana*, *Botrytis allii*, *B. byssoidea*, *B. cinerea*, *B. squamosa*, *Cladosporium cladosporioides*, *C. alliicepae*, *Colletotrichum circinans*, *C. dematium*, *C. gleosporioides*, *Curvularia lunata*, *Drechslera (Pseudocochliobolus) australiensis*, *Fusarium oxysporum*, *F. solani*, *F. moniliforme*, *F. avenaceum*, *F. moniliforme* var.

subglutinans, *Geotrichum* sp., *Humicola fuscoatra*, *Mucor* sp., *Penicillium* spp., *P. cyclopium*, *Pythium* spp., *Rhizoctonia solani*, *Rhizopus nigricans*, *R. stolonifer*, *Stemphylium botryosum*, *S. vesicarium*, *Trichoderma harzianum*, *T. pseudokoningii* and *Trichothecium roseum* [5], [6], [7], [8]. It is through seeds, primarily, that fungal pathogens are conserved and transmitted [9], [7], [8]. Though apparently fungi-free, seed lots can carry various fungi that are responsible for diseases in the field [10]. In Burkina Faso, where the main system of planting onion includes the use of dry seeds, little information on onion seed quality is available. The main onion seed supply sources are wholesalers of exotic seeds and local farm seeds multiplied from exotic seeds. Most of the seed lots used are exotic and often of good quality. In recent years, due to their high cost, some farmers produce and commercialize their own seeds. These seed lots, locally produced and increasingly used, are not assessed in terms of their health quality. This represents a high risk of the propagation of seed-borne diseases in the onion producing areas. The first requirement of an integrated disease management strategy aimed at preventing yield loss is to use healthy seeds. This study sought to assess fungal populations associated with onion seeds in Burkina Faso in order to evaluate seed health quality and identify the origin of fungal diseases, as a basis for the development of appropriate control strategies.

2 MATERIAL AND METHODS

2.1 SEED SAMPLES AND SAMPLING PROCESS

A total of 18 seed samples, representing the seeds lots available throughout the country (Table 1) were collected and tested in this study. These seed samples originated from two main sources. Exotic seed samples were provided by Technisem, King-Agro and Agrobusiness, the three main onion seed wholesalers in Burkina Faso. Local seed samples were mainly provided by farmers from villages in the Sourou Valley area (Upper West region), the main onion seed producing area in the country. All the exotic samples were treated with Topsin WG (70.4% of thiophanate-methyl) and one of local samples was treated with Calthio C (25% of thirame and 25% of chlorpyrifos-ethyl).

Table 1. Origins and characteristics of onion seed samples tested in this study

Sample code	Variety name	Location	Wholesaler/ Farmer/institutions	Origin	Treatment
EXO _{S1}	Violet de Damani	Bobo-Dioulasso	Technisem	Exotic	Topsin
EXO _{S2}	Violet de Galmi	Bobo-Dioulasso	King-Agro	Exotic	Topsin
LOC _{S1}	Violet de Galmi	Ouahigouya	Ilboudo Paul	Local	Untreated
LOC _{S2}	Violet de Galmi	Sourou/Oué	Ouedraogo Salam	Local	Untreated
LOC _{S3}	Violet de Galmi	Ouagadougou	INERA	Local	Untreated
LOC _{S4}	Violet de Galmi	Sourou/ Di	So Honoré	Local	Untreated
LOC _{S5}	Violet de Galmi	Sourou/ Benkadi	Gorou T. Abel	Local	Untreated
LOC _{S6}	Violet de Galmi	Sourou/ Oué/ Niger	Ouédraogo Salam	Local	Untreated
EXO _{S3}	Violet de Damani	Sourou/ Di	Technisem	Exotic	Topsin
LOC _{S7}	Violet de Galmi	Sourou/ Di	Ouédraogo Sita	Local	Calthio C
LOC _{S8}	Violet de Galmi	Sourou/Sababougnouma	Gansoré Moustapha	Local	Untreated
LOC _{S9}	Violet de Galmi	Sourou/ Oué	Gorou T. Abel	Local	Untreated
LOC _{S10}	Violet de Galmi	Sourou/Sokadi	Koné Hubert	Local	Untreated
EXO _{S4}	Violet de Damani	Sourou/ Oué	Technisem	Exotic	Topsin
EXO _{S5}	Prema 178	Korsimoro	UDGPM/K	Exotic	Topsin
LOC _{S11}	Violet de Galmi	Kongoussi	ND	Local	Untreated
LOC _{S12}	Violet de Galmi	Sourou/ Di	Kaboré Amadou	Local	Untreated
LOC _{S13}	Violet de Galmi	Bobo-dioulasso	INERA	Local	Untreated

2.2 DRY SEED HEALTH TESTING

The 'blotter method' was used to test the dry seed, as described by [11]. With this method, 400 seeds per sample were tested in 16 replications of 25 seeds following the international rules for seed testing [11]. For each replication, 25 seeds were placed equidistant on three layers of moistened blotter papers in a sterile Petri dish. They were incubated at 25°C under a light-dark cycle (12/12 h) for 7 days. The seeds were then individually observed under a stereomicroscope at 16x and 25x magnifications in order to detect the presence of fungal colonies and identify the causal species based on morphological characteristics. Where identification was dubious, slides of the fungal colony were prepared and observed under a

microscope at 10x and 40x magnifications. Most of the associated pathogens were detected by observing their growth characteristics on the incubated seeds following the procedure described by [11]. When necessary, some fungi were transferred to another Petri dish filled with Potato Dextrose Agar medium and incubated at 25°C under a light-dark cycle (12/12 h) for 7 days in order to finalize the identification. The frequency of fungal species present in the seed samples and the rates of infection by each fungal species in each seed sample were recorded.

2.3 SEEDLING HEALTH TESTING

Seedling health was tested in order to record fungal infection using moistened blotter method as described above. Seeds were sown in trays containing fine sand that has been sterilized twice at 120°C for 30 min over a 24 h period. After filling, the trays were watered with sterilized water. Seeds were individually placed in holes made in concentric circles 3 cm apart. The seedbed was watered with sterilized water and the trays were maintained at room temperature of 25-30°C to obtain seedlings. A total of 400 seeds per sample were sown using 20 seeds per tray. Each tray was considered as a replicate. The watering was repeated once a day, using 100 ml per tray throughout the experimentation period. Two weeks after sowing, the number of seedlings emerged was recorded. Ten seedlings were then taken in random from each tray, rinsed and dried at 30°C for one hour. Each seedling was then aseptically cut into three parts. The 30 fragments obtained from each replication were incubated on three layers of moistened blotter papers in three Petri dishes. They were incubated at 25°C under a light-dark cycle (12/12 h) for 7 days. The fungal species associated with the seedlings were recorded and identified according to their morphological traits using a stereomicroscope. The infection rates of each fungal species were assessed for each seed sample.

2.4 EXPERIMENTAL DESIGN AND DATA ANALYSIS

Each seed lot was assessed in completely randomized block design with 16 replications of 25 seeds during dry seed health testing and with 20 replications of 20 seeds during seedling health testing. The means of infection rates by fungi and seedling emergence rates were calculated using EXCEL 2007 software. These means were then compared by one way analysis of variance using Duncan range test performed with IBM SPSS Stat.23 software.

3 RESULTS

3.1 SEED HEALTH TESTING

Fungal contamination was detected in each of the 18 tested samples. Sixteen fungal species belonging to 10 genera were identified in the tested samples. The five most frequent genera recorded were: *Aspergillus* (Link) Micheli (contaminated 17 out of the 18 samples); *Fusarium* Link (15/18); *Rhizopus* (Corda) Ehrenb (15/18); *Cladosporium* Link (14/18); and *Penicillium* Link (13/18). The 16 species recorded were *Alternaria alternata*, *A. porri*, *Aspergillus flavus*, *A. niger*, *Botryodiplodia theobromae*, *Cladosporium* sp., *Curvularia lunata*, *C. pallescens*, *Exserohilum rostratum*, *Fusarium equiseti*, *F. moniliforme*, *F. oxysporum*, *F. solani*, *Penicillium* sp., *Phoma* sp. and *Rhizopus* sp. (Table 2). The three most common species were *A. niger* with 17 contaminated samples out of 18, *A. flavus* (16/18) and *Rhizopus* sp. (15/18) (Table 2).

Table 2. Prevalence of seed-borne fungi of onion seed samples in Burkina Faso assessed using dry seed analysis

Fungal species	Frequency of seed samples infected
<i>Alternaria alternata</i> (Fr.) Keissler	05/18 (27.77%)
<i>A. porri</i> (Ellis) Cif.	01/18 (05.55%)
<i>Aspergillus flavus</i> Link ex Fries	16/18 (88.88%)
<i>A. niger</i> Van Tieghem	17/18 (94.44%)
<i>Botryodiplodia theobromae</i> Pat.	01/18 (05.55%)
<i>Cladosporium sp.</i> Link.	14/18 (77.77%)
<i>Curvularia lunata</i> (Wakk.) Boejin	06/18 (33.33%)
<i>C. pallescens</i> Boejin	04/18 (22.22%)
<i>Exserohilum rostratum</i> (Drechsler) Leonard & Suggs.	04/18 (22.22%)
<i>Fusarium equiseti</i> (Corda) Sacc.	01/18 (05.55%)
<i>F. moniliforme</i> Sheldon	07/18 (38.88%)
<i>F. oxysporum</i> Schlecht. emend. Snyder & Hansen	11/18 (61.11%)
<i>F. solani</i> (Mart.) Appel & Wollenw	07/18 (38.88%)
<i>Penicillium sp.</i> Link.	13/18 (72.22%)
<i>Phoma sp.</i> Sacc.	07/18 (38.88%)
<i>Rhizopus sp.</i> Ehrenb.	15/18 (83.33%)

The infection rates of the samples varied among species from 0 to 90.3%. The saprophytic species were prevalent: *Aspergillus niger* up to 90.3% of the seeds contaminated; *A. flavus* (0.5-87.5%); *Penicillium sp.* (0.3-88%); and *Rhizopus sp.* (0.8-85.3) (Table 3, 4 and 5). Despite the high frequency of infection by *Fusarium* species, the infection rates by these species were lower than in the case of the saprophytic species: up to 34% for *F. solani* and up to 12.5% for *F. oxysporum*. The infection rates were generally higher for local than exotic seed samples, apart from sample Loc₅₇ which was treated with Calthio DS (Table 3, 4 and 5).

Table 3. The infection rates of exotic onion seed samples by fungal species detected by seed analysis

Fungal species	Sample codes				
	EXO _{S1}	EXO _{S2}	EXO _{S3}	EXO _{S4}	EXO _{S5}
<i>Alternaria alternata</i>	0.0 ± 0.0A				
<i>Alternaria porri</i>	0.0 ± 0.0A				
<i>Aspergillus flavus</i>	0.5 ± 1.4A	2.3 ± 2.9 B	0.0 ± 0.0A	0.0 ± 0.0A	3.3 ± 3.6 B
<i>Aspergillus niger</i>	0.3 ± 1.0A	8.3 ± 5.0 D	0.0 ± 0.0A	0.3 ± 1.0A	24.3 ± 7.9 C
<i>Botryodiplodia theobromae</i>	0.0 ± 0.0A				
<i>Cladosporium sp.</i>	0.3 ± 1.0A	0.0 ± 0.0A	0.0 ± 0.0A	0.3 ± 1.0A	4.0 ± 3.6 B
<i>Curvularia lunata</i>	0.0 ± 0.0A				
<i>Curvularia pallescens</i>	0.0 ± 0.0A	0.0 ± 0.0A	0.3 ± 1.0A	0.0 ± 0.0A	0.0 ± 0.0A
<i>Exserohilum rostratum</i>	0.0 ± 0.0A				
<i>Fusarium equiseti</i>	0.0 ± 0.0A				
<i>Fusarium moniliforme</i>	0.0 ± 0.0A	0.0 ± 0.0A	0.3 ± 1.0A	0.0 ± 0.0A	0.0 ± 0.0A
<i>Fusarium oxysporum</i>	10.3 ± 5.2B	0.0 ± 0.0A	0.0 ± 0.0A	0.5 ± 2.0A	0.0 ± 0.0A
<i>Fusarium solani</i>	0.0 ± 0.0A				
<i>Penicillium sp.</i>	0.3 ± 1.0A	0.0 ± 0.0A	52.5 ± 9.4 B	1.5 ± 2.0A	0.0 ± 0.0A
<i>Phoma sp.</i>	0.0 ± 0.0A				
<i>Rhizopus sp.</i>	59.0 ± 9.5C	4.5 ± 3.6 C	0.0 ± 0.0A	35.8 ± 11.7 B	0.0 ± 0.0A
F. values	453.8	29.8	482.3	137.6	106.1
P. values	0.000	0.000	0.000	0.000	0.000

Means within each column followed by the same letter are not significantly different at the 0.05 level using Duncan range test.

Table 4. The infection rates of local seed samples by fungal species detected by seed analysis

Fungal species	Sample codes					
	Loc _{S1}	Loc _{S2}	Loc _{S3}	Loc _{S4}	Loc _{S5}	Loc _{S6}
<i>Alternaria alternata</i>	0.0 ± 0.0A	0.0 ± 0.0A	0.3 ± 1.0A	0.8 ± 1.6A	0.0 ± 0.0A	0.0 ± 0.0A
<i>Alternaria porri</i>	0.0 ± 0.0A					
<i>Aspergillus flavus</i>	7.5 ± 5.0B	23.5 ± 2.9D	57.3 ± 6.8C	10.8 ± 6.8C	5.5 ± 5.9B	57.3 ± 7.3D
<i>Aspergillus niger</i>	43.8 ± 15.1C	70.3 ± 11.8E	38.0 ± 12.8B	90.3 ± 5.1E	0.8 ± 1.6A	81.3 ± 12.9E
<i>Botryodiplodia theobromae</i>	0.5 ± 2.0A	0.0 ± 0.0A				
<i>Cladosporium sp.</i>	8.0 ± 5.0B	2.3 ± 2.5AB	0.0 ± 0.0A	3.8 ± 2.3B	12.3 ± 7.1C	8.0 ± 6.2B
<i>Curvularia lunata</i>	0.0 ± 0.0A	0.0 ± 0.0A	0.0 ± 0.0A	0.0 ± 0.0A	1.0 ± 2.3A	0.0 ± 0.0A
<i>Curvularia pallescens</i>	0.0 ± 0.0A	0.0 ± 0.0A	0.0 ± 0.0A	0.3 ± 1.0A	0.0 ± 0.0A	0.8 ± 2.2A
<i>Exserohilum rostratum</i>	0.0 ± 0.0A	0.3 ± 1.0A				
<i>Fusarium equiseti</i>	0.3 ± 1.0A	0.0 ± 0.0A				
<i>Fusarium moniliforme</i>	0.0 ± 0.0A	3.5 ± 3.6B	0.8 ± 1.6A	0.0 ± 0.0A	0.3 ± 1.0A	0.0 ± 0.0A
<i>Fusarium oxysporum</i>	0.0 ± 0.0A	0.0 ± 0.0A	0.5 ± 1.4A	0.0 ± 0.0A	0.5 ± 1.4A	0.5 ± 1.4A
<i>Fusarium solani</i>	0.0 ± 0.0A	0.0 ± 0.0A	0.5 ± 1.4A	0.0 ± 0.0A	0.0 ± 0.0A	0.0 ± 0.0A
<i>Penicillium sp.</i>	1.8 ± 2.9A	16.5 ± 8.8C	88.0 ± 3.9D	35.8 ± 10.5D	11.5 ± 7.8C	1.0 ± 2.3A
<i>Phoma sp.</i>	0.0 ± 0.0A	0.0 ± 0.0A	0.3 ± 1.0A	0.0 ± 0.0A	0.5 ± 2.0A	0.5 ± 1.4A
<i>Rhizopus sp.</i>	5.3 ± 3.8B	1.5 ± 2.5AB	85.8 ± 6.0D	4.0 ± 3.3B	18.8 ± 6.6D	35.8 ± 10.3C
F. values	99.6	337.8	961.9	699.5	42.0	410.7
P. values	0.000	0.000	0.000	0.000	0.000	0.000

Means within each column followed by the same letter are not significantly different at the 0.05 level using Duncan range test

Table 5. The infection rates of local seed samples by fungal species detected by seed analysis(continued)

Fungal species	Sample codes						
	Loc _{S7}	Loc _{S8}	Loc _{S9}	Loc _{S10}	Loc _{S11}	Loc _{S12}	Loc _{S13}
<i>Alternaria alternata</i>	0.0 ± 0.0A	0.5 ± 1.4A	0.0 ± 0.0A	0.0 ± 0.0A	0.5 ± 1.4A	7.5 ± 5.0B	0.0 ± 0.0A
<i>Alternaria porri</i>	0.0 ± 0.0A	0.0 ± 0.0A	0.0 ± 0.0A	0.0 ± 0.0A	0.0 ± 0.0A	0.0 ± 0.0A	0.5 ± 1.4A
<i>Aspergillus flavus</i>	1.5 ± 2.5A	18.0 ± 5.3D	6.5 ± 5.2C	87.5 ± 4.1F	29.8 ± 5.8D	18.3 ± 6.4C	5.8 ± 6.5B
<i>Aspergillus niger</i>	10.0 ± 10.2C	6.8 ± 4.8B	15.8 ± 9.0E	82.0 ± 4.6E	83.5 ± 7.9E	58.8 ± 13.8D	13.8 ± 8.4C
<i>Botryodiplodia theobromae</i>	0.0 ± 0.0A	0.0 ± 0.0A	0.0 ± 0.0A	0.0 ± 0.0A	0.0 ± 0.0A	0.0 ± 0.0A	0.0 ± 0.0A
<i>Cladosporium sp.</i>	0.0 ± 0.0A	0.8 ± 1.6A	2.0 ± 2.5AB	1.5 ± 2.0AB	1.8 ± 2.9A	10.8 ± 7.4B	1.3 ± 2.4A
<i>Curvularia lunata</i>	0.0 ± 0.0A	0.0 ± 0.0A	0.5 ± 1.4AB	0.3 ± 1.0A	0.3 ± 1.0A	2.0 ± 2.5A	0.5 ± 2.0A
<i>Curvularia pallescens</i>	0.0 ± 0.0A	0.0 ± 0.0A	0.0 ± 0.0A	0.0 ± 0.0A	0.0 ± 0.0A	0.5 ± 1.4A	0.0 ± 0.0A
<i>Exserohilum rostratum</i>	0.0 ± 0.0A	0.3 ± 1.0A	0.0 ± 0.0A	0.0 ± 0.0A	0.0 ± 0.0A	1.0 ± 1.8A	0.8 ± 2.2A
<i>Fusarium equiseti</i>	0.0 ± 0.0A	0.0 ± 0.0A	0.0 ± 0.0A	0.0 ± 0.0A	0.0 ± 0.0A	0.0 ± 0.0A	0.0 ± 0.0A
<i>Fusarium moniliforme</i>	6.3 ± 4.6B	0.0 ± 0.0A	0.0 ± 0.0A	1.8 ± 2.5AB	0.0 ± 0.0A	0.0 ± 0.0A	1.0 ± 2.7A
<i>Fusarium oxysporum</i>	0.0 ± 0.0A	12.5 ± 6.4C	10.8 ± 5.4D	4.5 ± 4.0B	8.8 ± 6.2B	3.3 ± 2.2A	7.0 ± 5.9B
<i>Fusarium solani</i>	1.0 ± 2.3A	34.0 ± 8.9E	0.0 ± 0.0A	0.5 ± 0.5A	0.3 ± 1.0A	0.3 ± 1.0A	4.5 ± 3.2B
<i>Penicillium sp.</i>	0.0 ± 0.0A	0.0 ± 0.0A	3.3 ± 3.9B	9.8 ± 5.2C	0.3 ± 1.0A	0.0 ± 0.0A	7.3 ± 5.7B
<i>Phoma sp.</i>	0.0 ± 0.0A	0.0 ± 0.0A	0.5 ± 2.0AB	0.0 ± 0.0A	0.3 ± 1.0A	0.3 ± 1.0A	0.5 ± 2.0A
<i>Rhizopus sp.</i>	0.0 ± 0.0A	1.5 ± 2.0A	29.3 ± 7.7F	56.3 ± 17.0D	14.3 ± 7.0C	9.0 ± 5.0B	0.8 ± 2.2A
F. values	15.0	127.6	74.4	615.1	597.2	158.6	17.6
P. values	0.000	0.000	0.000	0.000	0.000	0.000	0.000

Means within each column followed by the same letter are not significantly different at the 0.05 level using Duncan range test

3.2 EMERGENCE RATES OF SEEDLINGS

Seedling emergence was assessed using 17 seeds samples because the quantity of seeds of the remaining sample, Loc_{S13}, insufficient. Seedlings emergences rates of exotic seed samples varied from 73.3 to 89.8%. Sample Exo_{S3} provided by TECHNISEM had the highest emergence rate, whereas sample Exo_{S5}, bought from a farmers' organization, UDGPMK, had the lowest emergence rate (Figure 1). Among the local seed samples, the emergence rates varied from 7.5 to 90.3%. Sample Loc_{S12} had the highest emergence rate and sample Loc_{S3} the lowest (Figure 2). Of the 17 analyzed samples, five had

emergence rates above 80%, seven had rates above 50%, one had a rate between 25 and 50% and four had rates below 25%. The highest emergence rate was recorded among the local samples, but all the exotic samples had emergence rates of up to 70% (Figure 1 and 2).

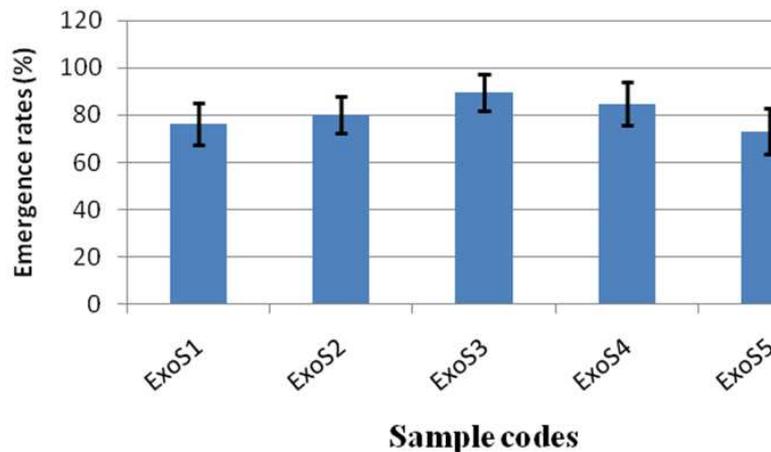


Fig. 1. Seedling emergence rates of exotic seed samples, 14 days after sowing

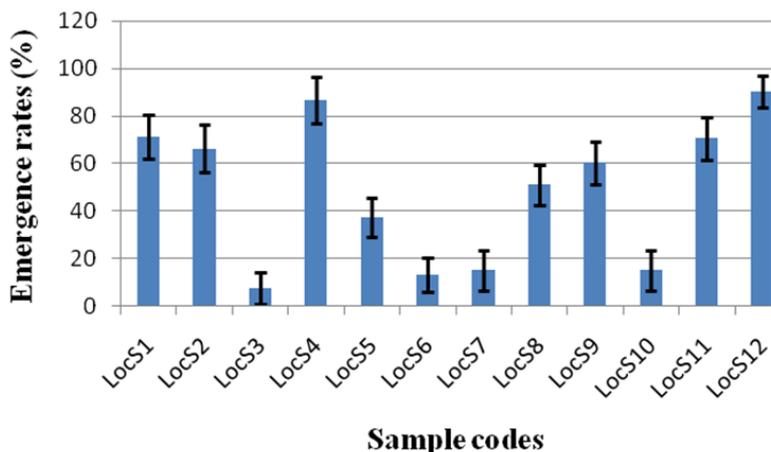


Fig. 2. Seedling emergence rates of local seed samples, 14 days after sowing

3.3 CORRELATION BETWEEN SEED INFECTION AND EMERGENCE RATES

In order to underline a possible relationship between the seedling emergence and the seed infection by fungal pathogens, data of emergence rates and those of infection rates of the seed samples were compared. The infection rates of seed samples by *Aspergillus niger* and *Fusarium oxysporum*, thought to be responsible for seedling damping-off in onions, were compared with percentage of non-germinated seeds per sample. A significant relationship was not underlined between the infection rates of seeds by *A. niger* and *F. oxysporum* and the percentage of non-germinated seeds according to this study. The infection rates of seeds by *F. oxysporum* and the percentage of non-germinated seeds exhibited a lowly perceptible correlation than those with *A. niger* (Figure 3 and 4).

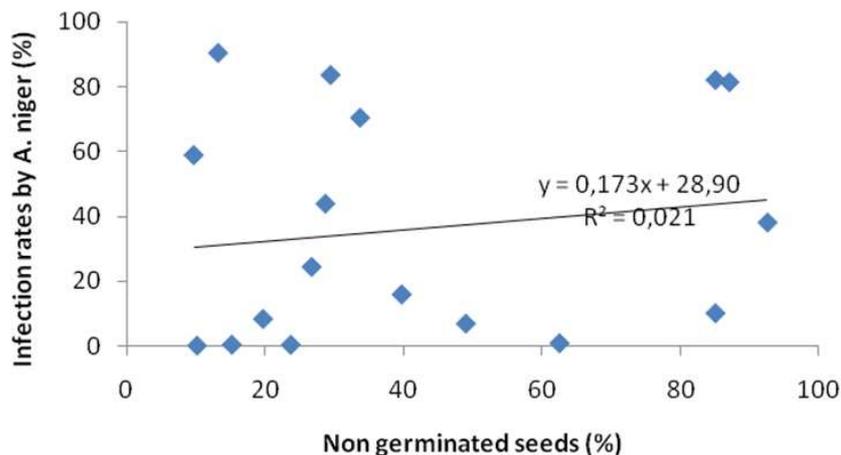


Fig. 3. Correlation between non-germinated seeds and infection rates by *A. niger*

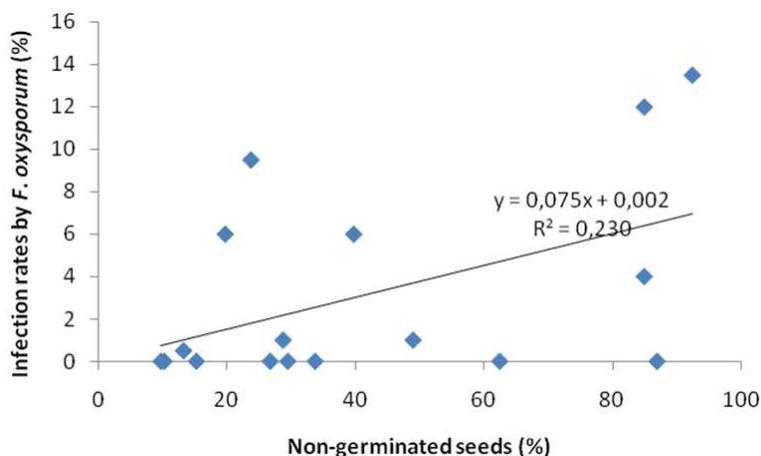


Fig. 4. Correlation between non-germinated seeds and infection rates by *F. oxysporum*

3.4 SEEDLING HEALTH TESTING

Fifteen species of fungi belonging to 10 genera were identified in the 17 onion seed samples tested : *Alternaria alternata*, *A. porri*, *Aspergillus flavus*, *A. niger*, *Bipolaris* sp., *Cladosporium* sp., *Curvularia lunata*, *C. pallescens*, *Exseherohilum rostratum*, *Fusarium moniliforme*, *F. oxysporum*, *F. solani*, *Penicillium* sp., *Phoma* sp. and *Rhizopus* sp. (Table 6). The three species most frequently recorded were: *Cladosporium* sp. (contaminated 16 out of the 17 samples); *Curvularia lunata* (11/17) and *Aspergillus niger* (10/17).

Table 6. Prevalence of fungi on onion seedlings

Fungal species	Frequency of seed samples infected
<i>Alternaria alternata</i> (Fr.) Keissler	05/17 (29.4%)
<i>A. porri</i> (Ellis) Cif.	01/17 (05.9%)
<i>Aspergillus flavus</i> Link ex Fries	07/17 (41.2%)
<i>A. niger</i> Van Tieghem	10/17 (58.8%)
<i>Bipolaris</i> sp.	06/17 (35.3%)
<i>Cladosporium</i> sp.	16/17 (94.1%)
<i>Curvularia lunata</i> (Wakk.) Boejin	11/17 (64.7%)
<i>C. pallences</i> Boejin	01/17 (05.9%)
<i>Exserohilum rostratum</i> Drechsler	07/17 (41.2%)
<i>Fusarium moniliforme</i> Sheldon	06/17 (35.3%)
<i>F. oxysporum</i>	09/17 (52.9%)
<i>F. solani</i> (Mart.) Appel & Wollenw	05/17 (29.4%)
<i>Penicillium</i> sp.	01/17 (05.9%)
<i>Phoma</i> sp.	07/17 (41.2%)
<i>Rhizopus</i> sp.	09/17 (52.9%)

The infection rates by the various fungal species ranged from 0.3 to 46.7%. The saprophytic species again had the highest infection rates: *Rhizopus* sp. (0.5-46.7%); *Cladosporium* sp. (3.3-30%) and *Aspergillus flavus* (0.5-13.3%). *Fusarium oxysporum* and *Phoma* sp. also had fairly high infection rates (0.5-13.3%)(Table 7, 8 and 9).

Table 7. The infection rates of exotic seed samples by fungal species detected by seedling analysis

Fungal species	Sample codes				
	Exo _{s1}	Exo _{s2}	Exo _{s3}	Exo _{s4}	Exo _{s5}
<i>Alternaria alternata</i>	0.0 ± 0.0A	0.0 ± 0.0A	0.5 ± 2.2A	1.0 ± 3.1A	0.0 ± 0.0A
<i>Alternaria porri</i>	0.0 ± 0.0A	0.0 ± 0.0A	0.0 ± 0.0A	1.0 ± 3.1A	0.0 ± 0.0A
<i>Aspergillus flavus</i>	0.0 ± 0.0A	7.0 ± 8.6BC	0.0 ± 0.0A	0.0 ± 0.0A	0.0 ± 0.0A
<i>Aspergillus niger</i>	0.0 ± 0.0A	8.0 ± 8.9C	0.0 ± 0.0A	0.0 ± 0.0A	0.5 ± 2.2A
<i>Bipolaris</i> sp.	0.0 ± 0.0A	0.0 ± 0.0A	0.0 ± 0.0A	1.0 ± 4.5A	1.0 ± 3.1A
<i>Cladosporium</i> sp.	30.0 ± 14.1E	17.5 ± 10.2D	3.5 ± 5.9C	3.5 ± 4.9B	10.5 ± 6.0B
<i>Curvularia lunata</i>	0.0 ± 0.0A	0.0 ± 0.0A	2.5 ± 5.5BC	6.5 ± 7.5C	14.5 ± 8.3C
<i>Curvularia pallescens</i>	0.0 ± 0.0A				
<i>Exserohilum rostratum</i>	0.0 ± 0.0A	0.0 ± 0.0A	1.0 ± 3.1AB	0.0 ± 0.0A	0.5 ± 2.2A
<i>Fusarium moniliforme</i>	14.0 ± 15.7C	3.5 ± 5.9AB	1.0 ± 3.1AB	0.0 ± 0.0A	0.0 ± 0.0A
<i>Fusarium oxysporum</i>	9.5 ± 10.0BC	6.0 ± 7.5BC	0.0 ± 0.0A	0.0 ± 0.0A	0.0 ± 0.0A
<i>Fusarium solani</i>	21.0 ± 16.8D	1.5 ± 4.9A	0.0 ± 0.0A	0.0 ± 0.0A	0.0 ± 0.0A
<i>Penicillium</i> sp.	0.0 ± 0.0A	6.0 ± 6.8BC	0.0 ± 0.0A	0.0 ± 0.0A	0.0 ± 0.0A
<i>Phoma</i> sp.	0.0 ± 0.0A	0.0 ± 0.0A	0.5 ± 2.2A	4.5 ± 6.0BC	0.0 ± 0.0A
<i>Rhizopus</i> sp.	8.5 ± 11.8B	0.0 ± 0.0A	3.5 ± 4.9C	5.0 ± 6.9BC	0.0 ± 0.0A
F. values	27.4	17.5	4.2	7.3	47.2
P. values	0.000	0.000	0.000	0.000	0.000

Means within each column followed by the same letter are not significantly different at the 0.05 level using Duncan range test

Table 8. The infection rates of local seed samples by fungal species detected by seedling analysis

Fungal species	Sample codes					
	Loc _{S1}	Loc _{S2}	Loc _{S3}	Loc _{S4}	Loc _{S5}	Loc _{S6}
<i>Alternaria alternata</i>	0.0 ± 0.0A					
<i>Alternaria porri</i>	0.0 ± 0.0A					
<i>Aspergillus flavus</i>	13.0 ± 10.8C	1.5 ± 3.7A	13.5 ± 12.3B	0.0 ± 0.0A	0.0 ± 0.0A	0.0 ± 0.0A
<i>Aspergillus niger</i>	8.0 ± 9.5B	6.5 ± 8.1C	10.0 ± 11.2B	0.5 ± 2.2A	0.0 ± 0.0A	0.0 ± 0.0A
<i>Bipolaris sp.</i>	0.0 ± 0.0A					
<i>Cladosporium sp.</i>	11.5 ± 9.9C	3.5 ± 5.9B	3.5 ± 4.9A	1.0 ± 3.1A	4.0 ± 6.0B	4.0 ± 6.0C
<i>Curvularia lunata</i>	0.5 ± 2.2A	1.0 ± 4.5A	0.0 ± 0.0A	0.0 ± 0.0A	1.5 ± 3.7A	2.0 ± 4.1B
<i>Curvularia pallescens</i>	0.0 ± 0.0A	0.0 ± 0.0A	0.0 ± 0.0A	0.0 ± 0.0A	0.5 ± 2.2A	0.0 ± 0.0A
<i>Exserohilum rostratum</i>	0.0 ± 0.0A	0.0 ± 0.0A	0.0 ± 0.0A	0.0 ± 0.0A	1.5 ± 4.9A	0.0 ± 0.0A
<i>Fusarium moniliforme</i>	0.0 ± 0.0A	0.5 ± 2.2A	0.0 ± 0.0A	0.0 ± 0.0A	0.0 ± 0.0A	0.0 ± 0.0A
<i>Fusarium oxysporum</i>	1.0 ± 3.1A	0.0 ± 0.0A	13.5 ± 9.9B	0.5 ± 2.2A	0.0 ± 0.0A	0.0 ± 0.0A
<i>Fusarium solani</i>	0.0 ± 0.0A	1.0 ± 3.1A	0.0 ± 0.0A	1.0 ± 4.5A	0.0 ± 0.0A	0.0 ± 0.0A
<i>Penicillium sp.</i>	0.0 ± 0.0A					
<i>Phoma sp.</i>	0.0 ± 0.0A	0.0 ± 0.0A	0.0 ± 0.0A	0.0 ± 0.0A	0.5 ± 2.2A	0.0 ± 0.0A
<i>Rhizopus sp.</i>	0.0 ± 0.0A	0.0 ± 0.0A	46.5 ± 27.2C	3.0 ± 4.7B	1.5 ± 4.9A	0.0 ± 0.0A
F. values	19.4	6.6	40.2	3.2	3.5	7.2
P. values	0.000	0.000	0.000	0.000	0.000	0.000

Means within each column followed by the same letter are not significantly different at the 0.05 level using Duncan range test

Table 9. The infection rates of local seed samples by fungal species detected by seedling analysis (continued)

Fungal species	Sample codes					
	Loc _{S7}	Loc _{S8}	Loc _{S9}	Loc _{S10}	Loc _{S11}	Loc _{S12}
<i>Alternaria alternata</i>	0.0 ± 0.0A	0.5 ± 2.2A	0.5 ± 2.2A	3.5 ± 6.7B	0.0 ± 0.0A	0.0 ± 0.0A
<i>Alternaria porri</i>	0.0 ± 0.0A	0.0 ± 0.0A	0.0 ± 0.0A	0.0 ± 0.0A	0.0 ± 0.0A	0.0 ± 0.0A
<i>Aspergillus flavus</i>	0.0 ± 0.0A	0.5 ± 2.2A	0.5 ± 2.2A	0.0 ± 0.0A	0.0 ± 0.0A	0.5 ± 2.2A
<i>Aspergillus niger</i>	0.0 ± 0.0A	2.5 ± 4.4AB	0.0 ± 0.0A	3.5 ± 4.9B	0.5 ± 2.2A	1.0 ± 3.1A
<i>Bipolaris sp.</i>	0.0 ± 0.0A	1.5 ± 3.7A	1.0 ± 4.5A	0.0 ± 0.0A	0.5 ± 2.2A	1.0 ± 4.5A
<i>Cladosporium sp.</i>	0.0 ± 0.0A	7.7 ± 9.2C	9.5 ± 8.3C	11.5 ± 5.9C	8.5 ± 7.5C	20.0 ± 10.3C
<i>Curvularia lunata</i>	0.0 ± 0.0A	2.5 ± 5.5AB	1.0 ± 3.1A	0.0 ± 0.0A	1.0 ± 3.1AB	0.5 ± 2.2A
<i>Curvularia pallescens</i>	0.0 ± 0.0A	0.0 ± 0.0A	0.0 ± 0.0A	0.0 ± 0.0A	0.0 ± 0.0A	0.0 ± 0.0A
<i>Exserohilum rostratum</i>	0.0 ± 0.0A	0.5 ± 2.2A	0.5 ± 2.2A	0.0 ± 0.0A	0.5 ± 2.2A	1.0 ± 3.1A
<i>Fusarium moniliforme</i>	5.0 ± 6.9B	2.0 ± 6.2A	0.0 ± 0.0A	0.0 ± 0.0A	0.0 ± 0.0A	0.0 ± 0.0A
<i>Fusarium oxysporum</i>	12.0 ± 10.1C	1.0 ± 3.1A	6.0 ± 7.5B	4.0 ± 6.0B	0.0 ± 0.0A	0.0 ± 0.0A
<i>Fusarium solani</i>	3.0 ± 4.7B	0.0 ± 0.0A	0.0 ± 0.0A	0.0 ± 0.0A	0.0 ± 0.0A	0.0 ± 0.0A
<i>Penicillium sp.</i>	0.0 ± 0.0A	0.0 ± 0.0A	0.0 ± 0.0A	0.0 ± 0.0A	0.0 ± 0.0A	0.0 ± 0.0A
<i>Phoma sp.</i>	0.0 ± 0.0A	5.0 ± 8.9BC	4.5 ± 5.1B	14.0 ± 10.5C	1.0 ± 3.1AB	0.0 ± 0.0A
<i>Rhizopus sp.</i>	0.0 ± 0.0A	0.0 ± 0.0A	9.0 ± 7.2C	0.0 ± 0.0A	2.5 ± 4.4B	3.5 ± 4.9B
F. values	19.0	4.4	13.9	24.1	13.2	43.9
P. values	0.000	0.000	0.000	0.000	0.000	0.000

Means within each column followed by the same letter are not significantly different at the 0.05 level using Duncan range test

3.5 COMPARATIVE ANALYSIS OF THE FUNGAL SPECIES DETECTED ON SEEDS AND SEEDLINGS

The combined analysis of the two health testing methods was based on the occurrence of 17 fungal species on onion seeds belonging to 11 genera. Some seed-borne fungi can be identified only by seed or seedling analysis. This was the case with *Botryodiplodia theobromae* and *Fusarium equiseti*, which were identified only by seed analysis, and *Bipolaris sp.*, identified by seedlings analysis (Figures 5, 6 and 7). For the species identified by both methods, the results also showed that the frequency of infected samples and the rate of infection differed depending on the method. Among the 17 species identified, six were more frequently detected on seedlings than on seeds. With regard to frequencies and infection rates, saprophytic species were detected more often on seeds than on seedlings, apart from *Cladosporium sp.* which was most

often detected on seedlings. *Aspergillus niger* and *A. flavus* were detected at frequencies of 17/18 and 16/18 by seed analysis and 11/17 and 6/17 by seedling analysis, respectively (Figure 5). Conversely, *Exserohilum rostratum*, *Curvularia lunata* and *Cladosporium sp.* were detected more often on seedlings. *Curvularia pallescens* was detected more often by seed analysis, unlike *C. lunata*, although they belong to the same genus (Figure 7). The comparative results of the *Fusarium* species showed that they were more often detected on seeds than seedlings. For example, *F. oxysporum* was detected at frequencies of 11/18 on seeds and 7/17 on seedlings (Figure 6). With *Alternaria* species, the results did not vary significantly between the methods used (Figure 7).

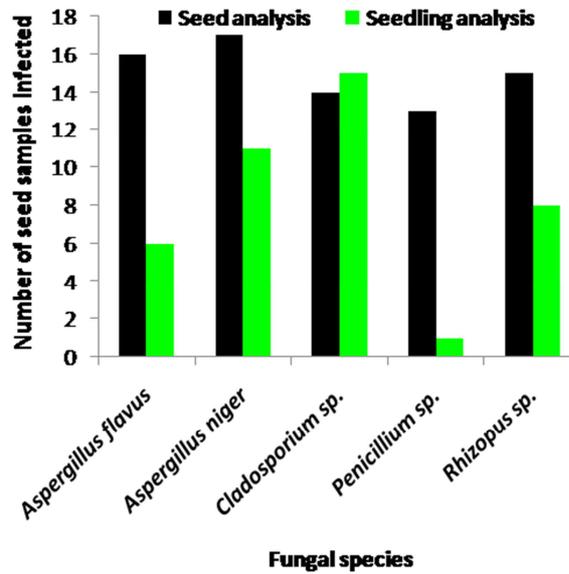


Fig. 5. Comparative occurrence of saprophytic species in seed samples according to the detection method used

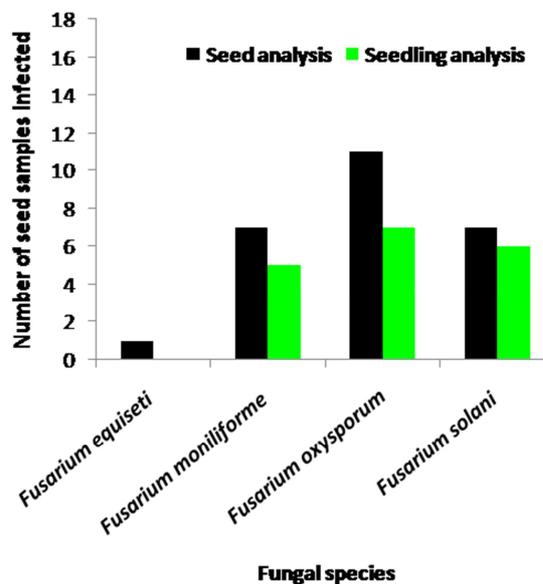


Fig. 6. Comparative occurrence of *Fusarium* species in seed samples according to the detection method used

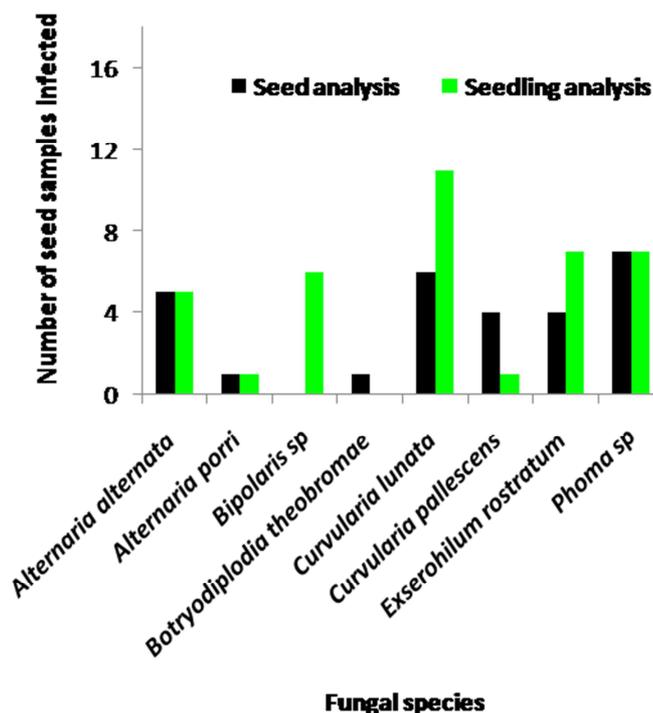


Fig. 7. Comparative occurrence of other pathogenic species in seed samples according to the detection method used

4 DISCUSSION

The overall aim of this study was to evaluate the health quality of onion seeds in Burkina Faso, particularly with regard to the incidence of seed-borne fungi, in order to define appropriate treatments and preventive protection measures that will limit the occurrence of seed-transmitted diseases.

The study involved analyses of seed samples of both exotic and local origin contaminated by pathogenic and/or saprophytic fungi belonging mainly to seven genera (*Alternaria*, *Aspergillus*, *Curvularia*, *Fusarium*, *Penicillium*, *Rhizopus* and *Phoma*). The results accord with those reported by [8] who detected *Alternaria*, *Aspergillus*, *Curvularia*, *Fusarium*, *Penicillium*, *Rhizopus* and *Dreschlera* species in four onion seed samples in Nagpur, India, using the blotter method. With the exception of *Phoma*, the same genera were recorded in onion seeds in Turkey [6], [7].

Among the fungi detected in seeds, the rates of infection by *Aspergillus niger* reached 90% in some samples. Ref. [12], [7] and [8] all reported that *A. niger* was the most frequent species detected in onion seeds. Several authors, including [13], [14], [5], [6] and [7] reported that *A. niger* could significantly reduce seed germination, with roots and shoots being unable to develop in severe cases because of the pre-emergence damping-off of seeds. In addition, *A. niger* had a negative effect on seedling development at 30-35°C [5], temperatures that are common in Burkina Faso. This species is also widely thought to be responsible for black mould in onion, a disease that occurs during storage and causes bulb rot. The fungus grows as small black masses under the bulb scales and in severe infections, these spore masses can cover the whole bulb surface [15], [7]. In many onion producing areas in Burkina Faso, bulbs are stored in straw huts that have a tendency to leak during the rainy season. The hot and moist conditions favor the growth and infectivity of *A. niger* [10]. The presence of this fungus in onion seeds used in Burkina Faso could partly explain the occurrence of seedling damping-off and black rot in the country. As this fungus is ubiquitous in nature and can colonize a wide variety of substrates [16], it is important to use preventive measures or appropriate disinfection to prevent seed contamination by *A. niger*, particularly because fungicide treatments are seldom efficient against this species [17], [13]. In addition, as in the case of *Penicillium* spp. [18], *A. niger* can produce ochratoxin, which is nephrotoxic for humans.

Among the pathogenic fungi, *Fusarium* species showed high infection rates in the onion seeds used in Burkina Faso. The most common species were *F. oxysporum* and *F. solani*, both of which have been reported as causal agents of onion rot in the field [19], [20]. The former is the main causal agent of basal rot in onion [21] and the latter is more harmful at early growth stages [19]. Ref. [22] and [7] reported that *F. oxysporum* is transmitted mainly by seeds and can cause significant reduction in seed germination. In onions, *F. oxysporum* is seed-borne, but it is not always identified when onion seeds are

screened for it [7]. The comparison between the non-emergent rates and the infection rates by *F. oxysporum* showed a significant correlation, suggesting the possible involvement of this fungus in seedling damping-off in onion. With regard to basal rot, symptoms generally appeared during the development of seedlings and bulbs in storage and their occurrence was closely linked to the period, environmental conditions, cultivars and inoculum level [23], [24]. *Fusarium* species, in particular, are known to be responsible for plant damage in warm areas [25], [26], [21]. The optimum soil temperature for the development of *F. oxysporum* is between 28 and 32°C [7]. Yield losses due to this fungus were estimated to be between 2.9 and 89% in some areas of Turkey [7].

Although found in low rates in onion seeds, *Alternaria porri* was detected in some seed samples. This species is responsible for purple blotch on onion leaves. In Burkina Faso, damage to onion leaves caused by *A. porri* can significantly affect farmers' incomes because these leaves are widely used in cooking. Bulb weight is also affected by *A. porri* [27]. This fungus presents problems for Burkina Faso's farmers because it is more virulent in warm regions [28]. A low incidence of *A. alternata* was recorded in some seed samples. This fungus is the causal agent of foliar blight in onion in India [29]. In our study in Burkina Faso it was associated with blotch symptoms on onion leaves. Recent pathogenicity tests have shown that some isolates are pathogenic on onion (unpublished data).

Relatively high infection rates of seeds by *Phoma* sp. were observed in our study. The species was not identified, but *P. terrestris* has been reported in Senegal and causes severe pink root in onion [30]. If it is seed-borne in the case of onion, the risk of introducing or propagating pink root disease in Burkina Faso could be high because among the onion sources suppliers, a major supplier is installed in Senegal.

The role of some seed-borne fungi detected in this study, such as *Aspergillus flavus*, *Cladosporium* sp., *Penicillium* sp., *Rhizopus* sp., *Curvularia lunata*, *C. pallescens* and *Exserohilum rostratum*, in the development of onion diseases is not yet well known.

The assessment of seedling emergence revealed that none of the analyzed samples achieved 100% emergence. Depending on several factors (e.g., agronomic, genetic), the emergence rate of seed samples can also be linked to infection by some fungal species. Severe infection of seeds by saprophytic species led to poor and abnormal seedling development [9]. The exotic samples had higher emergence rates than the local samples, probably because exotic seeds are usually produced in good agronomic conditions and treated with fungicides. Poor storage conditions of local seeds could maybe explained their low quality.

Seed and seedling analyses are complementary tests that produce different results in terms of fungal occurrence and infection rates. The location of the fungi on or within the seed, its virulence and pathogenicity and the prevailing moisture conditions can affect these rates. The method chosen to detect fungi in seed depends on the location of the fungi on or in the seed and the biology of the fungus [9], [11].

5 CONCLUSION

Our study results suggest that onion farmers in Burkina Faso need to disinfect seeds (even exotic ones alleged to have already been treated) before sowing and/or develop control strategies that will improve the quality of the seeds and thereby reduce losses caused by fungal pathogens. Local seed producers need to follow good phytosanitary practices in the field (plant certification) in order to improve the health quality of onion seeds in Burkina Faso.

ACKNOWLEDGMENTS

The authors would like to thank everyone in the Plant Health unit at the SY.NA.I.E Laboratory (Université Polytechnique de Bobo-dioulasso, Burkina Faso) and in the laboratory of Phytopathology (Université Catholique de Louvain), Belgium. They are also grateful to ARES-Programmes PIC, in Belgium for financial support.

REFERENCES

- [1] Direction Générale des Prévisions et des Statistiques Agricoles (DGPSA). *Analyse de la filière maraichage au Burkina Faso*. Rapport d'étude MAHRH, Ouagadougou, Burkina Faso, 2008.
- [2] Tarpaga W.V. 2012. *Contribution à l'étude de la montaison prématurée des variétés tropicales d'oignon (Allium cepa L.) : Cas du Violet of Galmi cultivé au Nord du Burkina Faso*. Thèse de doctorat, Université de Ouagadougou, Ouagadougou, Burkina Faso, 2012.
- [3] S. D'Alessandro, S. Alseny. *Evaluation sous-régionale de la chaîne de valeurs oignon / échalote en Afrique de l'ouest*. Bethesda, MD: projet ATP, Abt Associates Inc. Décembre 2008.
- [4] L. Ouedraogo, A. Rouamba. "Identification de deux bactéries responsables de la pourriture des bulbes d'oignon en stockage au Burkina Faso", *Annales de l'Université de Ouagadougou, série B*, pp. 198-204, 1997.
- [5] N.J. Hayden and R.B. Maude, "The role of seedborne *Aspergillus niger* in transmission of black mould of onion. *Plant Pathology*", vol. 41, pp. 573-581, 1992.
- [6] Özer N., Köycü N.D. 1997. *The pathogenicity of Aspergillus niger and some Fusarium species on onion seeds and seedlings*. In: Proceedings of the 10th Congress of the Mediterranean Phytopathological Union, Montpellier, France, 1997.
- [7] Özer N., Köycü N.D. 2004. *Seed-borne fungal diseases of onion and their control* In: Mukerji K.G. (ed.). *Disease Management of Fruits and Vegetables*, Vol. 1, Kluwer Academic Publishers, Dordrecht, The Netherlands, pp. 281-306, 2004.
- [8] S. Dumbre, D. Guldekar, R. S. Potdukhe, "Survey of seed-borne fungi of onion (*Allium cepa* L.) from various locations of Maharashtra". *Journal of Soil Crop*, vol. 21, pp. 221-224, 2011.
- [9] R. Champion, *Identifier les champignons transmis par les semences*. Collection « Techniques et pratiques ». Inra (Eds), Paris, France, 1997.
- [10] N.J. Hayden, R.B. Maude, F. J. Proctor, "Studies on the biology of black mold (*Aspergillus niger*) on temperate and tropical onions. A comparison of sources of the disease in temperate and tropical field crops", *Plant Pathology*, vol 43, pp. 562-569, 1994.
- [11] Mathur S.B., Kongsdal O. 2003, *Common laboratory seed health testing methods for detecting fungi*, 1st Ed. Kandrups Bogtrkkeri Publication, Denmark.
- [12] S.A. F. Nagerabi and R. M. Abdalla 2004, "Survey of seed borne fungi of Sudanese cultivars of onion, with new records", *Phytoparasitica*, vol. 32 no 4, pp. 413-416, 2004.
- [13] K. Tanaka, "Studies on the black mould disease of onion bulbs caused by *Aspergillus niger* van Tieghem", *Review of Plant Pathology*, vol. 70, pp. 82-99, 1991.
- [14] S. A. F. Nagerabi and A. H. M. Ahmed, "The effect of black mould (*Aspergillus niger*) on two Sudanese cultivars of onion", *Tropical Science*, vol. 41, pp. 95-99, 2001.
- [15] Sumner D.R., 1995, *Black mold*, In: Schwartz H.F., Mohan S.K. (Eds.). *Compendium of Onion and Garlic Diseases*. APS Press, St Paul, MN, USA, pp.10-11, 1995.
- [16] S. Rushi, "Pathogenicity of *Aspergillus niger* in plants", *Cibtech Journal of Microbiology*, vol. 1, no 1, pp. 47-51, 2012.
- [17] N. Özer, M. Koç, B. Der, "The sensitivity of *Aspergillus niger* and *Fusarium oxysporum* f. s. *cepae* to fungistasis in onion-growing soils", *Journal of Plant Pathology*, vol. 91, no 2, pp. 401-410, 2009.
- [18] N.M. Tri, *Identification des espèces de moisissures potentiellement productrices de mycotoxines dans le riz commercial dans cinq provinces de la région centrale du vietnam-étude des conditions pouvant réduire la production des mycotoxines*. Thèse de doctorat, Université de Toulouse. Toulouse, France.
- [19] D. Zlata, T. Jelena, N. Stevan, M. Jelica, A. Mijana, R.Svetlana, "Fusarium rot of onion and possible use of bioproduct" *Proceedings of Natural Science*, vol. 114, pp. 135-148, 2008.
- [20] U. Smolinska, W. Kowalczyk, "The impact of the Brassicaceae plant materials added to the soil on the population of *Fusarium solani* (Mart.) and *Fusarium oxysporum* Schlecht", *Journal of Horticultural Research*, vol. 22, no 1, pp. 123-129, 2014.
- [21] Havey M.J. 1995. *Fusarium basal plate rot*, In: Schwartz H.F., Mohan S.K. (Eds.). *Compendium of Onion and Garlic Diseases*. APS Press, St Paul, MN, USA, pp.10-11, 1995.
- [22] A. El Zawahry, H.M. El Aref, N.G. Ahmed, A.A. Aly, "Protein patterns of certain isolates of *Fusarium oxysporum* and *F. moniliforme* and their relation to virulence", *Asian Journal of Agricultural science*, vol. 31, pp. 59-78, 2000.
- [23] M.J. Stadnik and O.D. Dhingra, "Response of onion genotypes to *Fusarium oxysporum* f. sp. *cepae* during the growth phase and in storage", *Fitopatologia Brasileira*, vol. 21, pp. 431-435, 1996.
- [24] K. E. Conn, J. S. Lutton, S. A. Rosenberger, *Onion Disease Guide. A practical guide for seedmen, growers and agricultural advisors*. Seminis vegetable seeds, Inc. Woodland, CA, USA. 2012.

- [25] D.M. Naik and O.L. Burden 1981, "Chemical control of basal rot of onion in Zambia", *Tropical Pest Management*, vol.27, pp. 455-460, 1981.
- [26] F. Kodama, "Studies on basal rot of onion caused by *Fusarium oxysporum* f. sp. *cepae* and its control", *Review of Plant Pathology*, vol. 62, pp. 45-62, 1983.
- [27] Madhavi A., Kavitha, Vijayalakshmi M, "Studies on *Alternaria porri* (Ellis) Ciferri pathogenic to onion (*Allium cepa* L.)", *Archives of Applied Science Research*, vol. 4, no 1, pp. 1-9, 2012.
- [28] K. Shehu, H.A. Suberu, M.D. Magaji, "Amelioration of Purple Blotch Disease in Onion (*Allium cepa* L.) Seedlings with Organic Soil Amendments", *Nigerian's Journal of Basic Applied Science*, vol. 16, no 2, pp. 203-206, 2008.
- [29] E. Shahnaz, V. K. Razdan S. E. H. Rizvi, T. R. Rather, S. Sachin Gupta, A. Muneeb, "Integrated Disease Management of Foliar Blight Disease of Onion: A Case Study of Application of Confounded Factorials", *Journal of Agricultural science*, vol. 5, no 1, pp. 1916-9752, 2013.
- [30] Kane A. 1997. *Effets des fongicides (Basamid, Cryptonol, Enzone) et des endomycorhizes sur la croissance et le développement de deux variétés d'oignon (Allium cepa L.) cultivées sur un sol infesté par Pyrenochaeta terrestris au Nord-Ouest du Sénégal*, Thèse de Doctorat, Université Cheick Anta Diop, Dakar, Sénégal, 1997.