Determination of *Fusarium* Species Associated with Onion Plants (*Allium cepa*) in Field in Burkina Faso Causing Damping-Off and Bulb Rots

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**Abstract**

Onion (*Allium cepa* L.) is the second most important vegetable crop in Burkina Faso and provides an important source of income for those involved in the sector. However, producers are facing significant pre-harvest and post-harvest losses. To date, knowledge of major diseases of the crop is limited, limiting the development of effective control strategies. The objective of this study was to test the pathogenicity of some species of *Fusarium*. To this end, 33 fungal isolates collected from onion plants in 17 localities and belonging to five *Fusarium* species were used to inoculate onion seeds and bulbs to determine the pathogenic species responsible for damping-off on seedlings and basal bulb rot in Burkina Faso. The virulence of pathogenic isolates was determined according to the percentages of seedling damping-off evaluated 28 Days After Sowing, and the extent of rot in millimetres on inoculated bulbs. The evaluation of isolates on seedlings revealed that the most pathogenic isolates belong to the species *F. proliferatum* (I29, I21, I37, I33, I31), *F. thapsinum* (I35) and *F. solani* (I38) which resulted in 58.33% - 70.83% of seedling damping-off. The most pathogenic isolates on bulbs belong to the species *F. proliferatum* (I4, I29, I32) and *F. oxysporum* (I52, I50, I16) which caused 21.67 to 25 mm of rot on bulbs. Isolate I29 was very virulent on both seedlings and bulbs. The isolates of *F. fujikuroi* species were all low pathogenic on seedlings but one of them, (I27), expressed average pathogenicity on bulbs.

**Keywords**

Fungal Pathogen, *Violet de Galmi*, *Fusarium*, Seedling Damping-Off, Bulb Rots
1. Introduction

In Burkina Faso, agriculture is the main source of income for the poorest populations and the basis for the country’s food security. It employs more than 80% of the population and contributes about 33% to the Gross Domestic Product (GDP) [1]. The State of Burkina Faso, through the Ministry of Agriculture and Hydro-Agricultural Facilities (Ministère de l’Agriculture et des Aménagements Hydro-agricoles) and its partners, are building dams and water reservoirs to promote off-season crops. These crops make a substantial contribution to the fight against food insecurity. Onion (Allium cepa L.) is one of the main vegetable crops in West Africa and more particularly in Burkina Faso where they were introduced around 1915-1920 [2]. In Africa, onion production is very low and does not cover demand [3]. This production deficit, particularly in West Africa, makes this sub-region an import zone for fresh onion from developed countries such as the Netherlands and France. The crop yield varies from country to country, ranging from 20 T/ha in Burkina Faso, Togo and Senegal to 35 T/ha in Niger [4]. The average onion yield in this area is estimated at less than 20 T/ha compared to 40 to 60 T/ha in the United States and the Netherlands [5]. This low yield recorded in Africa is mainly due to biotic constraints [6] [7]. Fungal diseases occupy a prominent place among bio-aggressors [8]. Of these diseases, onion basal rot or Fusarium head blight is recognized as one of the most harmful onion diseases with a damage rate of more than 50% [9] [10] [11] [12]. The disease begins in the field and presents symptoms such as delayed emergence, pre-emergence and post-emergence damping-off, late growth, chlorotic leaves, necrosis, root and bulb discoloration, rot and death of the plant [13]. These effects continue in the post-harvest stage and cause losses in the quantity and quality of bulbs in storage [13] [14] [15]. Among the different Fusarium species recognized as basal onion rot agents in the world, F. oxysporum, F. solani and F. proliferatum are the most frequent species, among which F. proliferatum [12] [16] [17] and F. solani have the capacity to produce mycotoxins [18]. The extent of damage caused by Fusarium species varies not only with the cultivar but also with the type of isolate [10] [19] [20]. In Burkina Faso, several cases of fungal attacks causing significant damage to onion cultivation have previously been reported; however, formal identification of these pathogens has not been done [21] [22]. The findings of Dabiré revealed that seedling damping-off, Fusarium rot, black mould, leaf spot disease, purple spot disease, pink root disease and white rot are the main fungal diseases of onion in Burkina Faso [23]. In view of damage caused by these diseases, a better understanding of the most pathogenic species is needed; and the objective of this study was to test the pathogenicity of 33 Fusarium isolates belonging to five species (F. oxysporum; F. thapsinum; F. proliferatum; F. solani and F. fujikuroi) isolated from onion plants in Burkina Faso, in order to determine the species involved in seedling damping-off and onion bulb rot for the development of an appropriate and sustainable management method for these pathogens.
2. Material and Methods

2.1. Pathogenicity Assessment of 33 Isolates of Different Fusarium Species on Onion Seedlings

The pathogenicity of 33 monospore isolates of Fusarium belonging to five species: *Fusarium oxysporum* (13 isolates); *Fusarium proliferatum* (15 isolates); *Fusarium solani* (2 isolates); *Fusarium fujikuroi* (2 isolates); *Fusarium thapsinum* (1 isolate), was assessed on onion seedlings. These isolates were collected from onion plants from 17 production sites in Burkina Faso: Yako, Ouahigouya, Titao, Korsimoro, Kongoussi, Mogtédo, Loumbila, Donsin, Kokologho, Kou-dougou, Réo, Gnassan, Di, Tougan, Dano, Soumasso and Bama (Figure 1) and identified using molecular technology [24].

Seeds of the “Violet de Galmi” (Safari) variety, originating from Niger and widely grown in Burkina Faso, were purchased commercially and used in the study. Pathogenic isolates were determined under greenhouse conditions (between 25˚C and 28˚C), based on the percentages of pre-emergence and post-emergence seeding damping-off evaluated at 12 and 28 days after sowing. The seeds were disinfected using 70% alcohol for 30 seconds, then with sodium hypochlorite at 3% for 3 minutes. For each isolate, a conidial suspension of $10^6$ spores/ml concentration was prepared from a 10-day-old culture and 25 µl of this suspension was placed on each onion seed at planting, just before closing the hole. Twenty-four onion seeds were used per isolate. Seeds inoculated with sterile distilled water were used as a control. The study was conducted following a randomized complete block design consisting of 34 treatments (33 isolates + control) and 6 replications using one bulb per replication. Seeding was carried out in plastic blister packs containing Jarditropic soil, (multi-purpose soil composed of black peat, blond peat, pine bark and fertilizer) previously sterilized. The data were analyzed using the Statistical Analysis System (SAS) software, version 8; 2001. Isolate means separation was performed according to Duncan’s test, at the 5% significance level. Based on the percentages of seedling damping-off (pre-emergence and post-emergence damping-off), the isolates were grouped into three classes using the grouping method proposed by Sassaki [25] with a slight modification: highly pathogenic (seeding rate > 50%), moderately pathogenic (rate between 20% and 50%) and low pathogenic (rate < 20%).

2.2. Pathogenicity Assessment of 33 Isolates of Different Fusarium Species on Onion Bulbs

All the 33 isolates previously tested on seedlings were also used to assess the pathogenicity on onion bulbs using the methodology described by Bayraktar and Dolar [26]. The bulbs of the "Violet de Galmi" variety, bought at the market, were previously cleaned of external scales and roots, and then disinfected with 70% alcohol for 30 seconds. A hole of 4 mm in diameter and approximately 5 mm deep was made on the basal stem of each bulb using a punch. Thereafter, for each isolate, 10 µl of conidial suspension of $10^6$ spores/ml concentration was
placed in each hole. The hole was then closed with a water-proof adhesive (scotch tape). The control bulbs were inoculated with sterile distilled water. After inoculation, the bulbs were placed in an incubation greenhouse at a temperature between 25˚C and 28˚C for 14 days. At the end of the incubation, each bulb was cut longitudinally into two parts and the length of rot in the tissue was measured. The degree of pathogenicity of each isolate was determined by measuring the extent of rot in millimeters on the infected bulbs. The study was conducted following a randomized complete block design consisting of 34 treatments (33 isolates + control) and 6 replications using one bulb per replication. An analysis of variance was performed on the percentages of seedling damping-off and bulb rot lengths caused by the different isolates using Statistical Analysis System (SAS) software, version 8; 2001.

3. Results and Discussion

3.1. Results

- Effect of *Fusarium* isolates on young onion plants

The results presented in Table 1 indicated that seed inoculation with isolates resulted in very significant decreases in onion seedling emergence rates (p = 0.0016). Except sterile distilled water used as a control, all isolates produced pre-emergence damping-off ranging from 4.17% to 62.50%. The results also revealed significant variations between the combined percentages of pre-emergence and post-emergence damping-off (p = 0.0044). However, no significant effects were noted on post-emergence damping-off (p = 0.4334). Analysis of pre-emergence damping-off data allowed isolates to be classified into seven different groups (Table 1). The first two groups consisting of six isolates, five of which belong to the species *F. proliferatum* (I21, I29, I31, I33, I37) and one to the species *F. thapsinum* (I35), induced strong pre-emergence damping-off.
(>50%). The next three classes were composed of 20 isolates that caused moderate pre-emergence damping-off ranging from 20% to 50%, and the last two groups with seven isolates, six of which belonged to the species *F. oxysporum*, caused relatively low pre-emergence damping-off ranging from 4.17% to 16.67%.

Post-emergence damping-off ranging from 4.16% to 16.67% was also induced by the isolates (Table 1). Only six out of the 33 isolates (I5, I27, I31, I49, I52, I58) did not cause mortality on plants. However, the statistical analyses did not reveal any significant difference (p = 0.4334) between the isolates themselves or between the isolates and the control for post-emergence damping-off.

By considering pre-emergence and post-emergence damping-off, statistical analysis allowed to classify the isolates into eight pathogenic groups (Table 1). Based on the rate of pre-emergence and post-emergence damping-off caused and on the basis of the grouping proposed by Sassaki [25], the isolates were divided into 3 classes (Table 2):

- The class of highly pathogenic isolates (HPAlS), including seven isolates that caused more than 50% of damping-off on seedlings. Of these isolates, five belong to the species *F. proliferatum* (I21, I29, I31, I33, I37), one to the species *F. solani* (I38) and one to the species *F. thapsinum* (I35).
- The class of moderately pathogenic (MP) isolates includes 22 isolates, 10 of which belong to *F. proliferatum*, nine to *F. oxysporum*, two to *F. fujikuroi* and one to *F. solani*, with seeding damping-off rates between 20% and 50%.
- The class of low pathogenic isolates (PP) composed of 4 isolates all belonging to the species *F. oxysporum*, and the control (sterile distilled water), which caused percentages of seedling damping-off less than 20%.

By grouping Fusarium isolates by species of membership, the analytical results (Table 3) revealed highly significant differences between Fusarium species with regard to pre-emergence and pre- and post-emergence damping-off. All the species studied generally caused significant damping-off on onion seedlings. *F. thapsinum*, represented by a single isolate, induced the highest rates of pre-emergence damping-off (54.17%) which was statistically similar to those caused by *F. solani*, *F. proliferatum* and *F. fujikuroi* but strictly higher than those noted with *F. oxysporum* (22.44%).

For post-emergence damping-off, plant mortality rates were statistically equal for all *Fusarium* species used and no difference was noted between these rates and that of the control (sterile distilled water) (Table 3).

Table 1. Percentages of pre-emergence, post-emergence damping-off and combined percentages of pre- and post-emergence damping-off recorded after inoculation of onion seeds with different *Fusarium* isolates.

| *Fusarium* species | Insulation No. | Pre-emergence seeding cast irons (%) | Post-emergence seeding cast irons (%) | Pre-emergence and post-emergence seeding wheels (%) |
|--------------------|----------------|-------------------------------------|---------------------------------------|---------------------------------------------------|
| *F. oxysporum*     | I5             | 4.17 de                             | 0.00 a                                | 4.17 e                                            |
|                    | I7             | 33.33 abcd                          | 8.33 a                                | 41.67 abcd                                        |
Continued

|   |   |   |   |
|---|---|---|---|
| I9 | 12.50 cde | 12.50 a | 25.00 bcde |
| I16 | 16.67 cde | 16.67 a | 33.33 abcde |
| I42 | 12.50 cde | 4.16 a | 16.67 cde |
| I49 | 8.33 cde | 0.00 a | 8.33 de |
| I50 | 37.50 abc | 4.16 a | 41.67 abcd |
| I52 | 33.33 abc | 0.00 a | 33.33 abcd |
| I58 | 20.83 bcde | 0.00 a | 20.83 bcde |
| I59 | 12.50 cde | 4.16 a | 16.67 cde |
| I63 | 25.00 abcd | 4.16 a | 29.17 abcd |
| I66 | 37.50 abc | 12.50 a | 50.00 abc |
| I67 | 37.50 abc | 12.50 a | 50.00 abc |

**F. proliferatum**

|   |   |   |   |
|---|---|---|---|
| I1 | 20.83 cbcd | 8.33 a | 29.17 bcde |
| I3 | 4.17 de | 16.67 a | 20.83 bcde |
| I4 | 33.33 abc | 4.16 a | 37.50 abcd |
| I11 | 37.50 abc | 8.33 a | 45.83 abcd |
| I14 | 20.83 bcde | 8.33 a | 29.17 bcde |
| I15 | 29.17 abcd | 12.50 a | 41.67 abcd |
| I17 | 25.00 abcd | 8.33 a | 33.33 abcd |
| I18 | 37.50 abc | 4.16 a | 41.67 abcd |
| I19 | 37.50 abc | 4.16 a | 41.67 abcd |
| I21 | 58.33 ab | 4.16 a | 62.50 ab |
| I29 | 58.33 ab | 12.50 a | 70.83 a |
| I31 | 62.50 a | 0.00 a | 62.50 ab |
| I32 | 20.83 abcd | 8.33 a | 29.17 abcd |
| I33 | 54.17 ab | 8.33 a | 62.50 ab |
| I37 | 58.33 ab | 4.16 a | 62.50 ab |

**F. solani**

|   |   |   |   |
|---|---|---|---|
| I24 | 41.67 abc | 8.33 a | 50.00 abc |
| I38 | 45.83 abc | 16.67 a | 62.50 ab |

**F. fujikuroi**

|   |   |   |   |
|---|---|---|---|
| I12 | 37.50 abc | 8.33 a | 25.83 abcd |
| I27 | 41.67 abc | 0.00 a | 41.67 abcd |

**F. thapsinum**

|   |   |   |   |
|---|---|---|---|
| I35 | 54.17 ab | 4.16 a | 58.33 abc |

**Witnesses**

|   |   |   |   |
|---|---|---|---|
| Water | 0.00 e | 4.16 a | 4.17 e |

**Average**

|   |   |   |   |
|---|---|---|---|
| 31.49 | 6.86 | 38.35 |

**P**

|   |   |   |   |
|---|---|---|---|
| 0.0016 | 0.4334 | 0.0044 |

**CV**

|   |   |   |   |
|---|---|---|---|
| 33.36 | 62.08 | 28.94 |

\(a\) The pre-emergence percentages of damping-off and the percentages of combined pre-emergence and post-emergence damping-off were transformed into Arc sinus before performing the analysis of variance in order to normalize the data and stabilize the variance over the entire data range. (b) The values in the same column assigned with the same alphabetical letter(s) are not significantly different at the 5% threshold, according to the Duncan test.
Table 2. Distribution of isolates according to their pathogenicity on seedlings.

| Fusarium species | Isolates | Classes defined according to the level of pre-emergence and post-emergence fonts | TP (>50% of cast iron) | MP (20% - 50% of fonts) | PP (<20% of fonts) |
|------------------|----------|---------------------------------------------------------------------------------|------------------------|-------------------------|-------------------|
| *F. oxysporum*   | I5       | X                                                                               |                        |                         |                   |
|                  | I7       | X                                                                               |                        |                         |                   |
|                  | I9       | X                                                                               |                        |                         |                   |
|                  | I16      | X                                                                               |                        |                         |                   |
|                  | I42      | X                                                                               |                        |                         |                   |
|                  | I49      | X                                                                               |                        |                         |                   |
|                  | I50      | X                                                                               |                        |                         |                   |
|                  | I52      | X                                                                               |                        |                         |                   |
|                  | I58      | X                                                                               |                        |                         |                   |
|                  | I59      | X                                                                               |                        |                         |                   |
|                  | I63      | X                                                                               |                        |                         |                   |
|                  | I66      | X                                                                               |                        |                         |                   |
|                  | I67      | X                                                                               |                        |                         |                   |
| *F. proliferatum*| I1       | X                                                                               |                        |                         |                   |
|                  | I3       | X                                                                               |                        |                         |                   |
|                  | I4       | X                                                                               |                        |                         |                   |
|                  | I11      | X                                                                               |                        |                         |                   |
|                  | I14      | X                                                                               |                        |                         |                   |
|                  | I15      | X                                                                               |                        |                         |                   |
|                  | I17      | X                                                                               |                        |                         |                   |
|                  | I18      | X                                                                               |                        |                         |                   |
|                  | I19      | X                                                                               |                        |                         |                   |
|                  | I21      | X                                                                               |                        |                         |                   |
|                  | I29      | X                                                                               |                        |                         |                   |
|                  | I31      | X                                                                               |                        |                         |                   |
|                  | I32      | X                                                                               |                        |                         |                   |
|                  | I33      | X                                                                               |                        |                         |                   |
|                  | I37      | X                                                                               |                        |                         |                   |
| *F. solani*      | I24      | X                                                                               |                        |                         |                   |
|                  | I38      | X                                                                               |                        |                         |                   |
| *F. fujikuroi*   | I2       | X                                                                               |                        |                         |                   |
|                  | I27      | X                                                                               |                        |                         |                   |
| *F. thapsinum*   | I35      | X                                                                               |                        |                         |                   |

a TP = Very Pathogenic; b MP = Moderately Pathogenic; c PP = Less Pathogenic; X = pathogenicity of species.
Table 3. Importance of seedling damping-off (pre-, post- and pre- + post-emergence) caused by different *Fusarium* species on onion seedlings.

| *Fusarium* species | Number of isolates evaluated | Pre-emergence damping-offa (%) | post-emergence damping-offb (%) | Pre- and post-emergence damping-off (%) |
|--------------------|------------------------------|--------------------------------|--------------------------------|------------------------------------------|
| *F. oxysporum*     | 13                           | (22.44) 26.36 b                | (6.09) 2.04 a                   | (28.53) 30.70 a                         |
| *F. proliferatum*  | 15                           | (37.22) 36.21 ab               | (7.50) 2.52 a                   | (44.72) 41.38 a                         |
| *F. solani*        | 2                            | (43.75) 41.42 ab               | (12.50) 3.21 a                  | (56.25) 48.65 a                         |
| *F. fujikuroi*     | 2                            | (39.58) 38.87 ab               | (4.16) 1.87 a                   | (43.75) 41.68 a                         |
| *F. thapsinum*     | 1                            | (54.17) 47.65 a                | (4.16) 1.84 a                   | (58.33) 50.12 a                         |
| Witnesses          | (0.00) 0.82 c                 | (4.16) 1.84 a                  | (4.17) 8.39 b                   |                                          |
| Average            | (31.49)                      | (6.86)                         | (38.35)                        |                                          |
| P                  | 0.0016                       | 0.5742                         | 0.0002                         |                                          |
| CV                 | 40.64                        | 63.57                          | 33.13                          |                                          |

a The percentages of pre-emergence damping-off and the percentages of combined pre- and post-emergence damping-off were transformed into Arc sinus before proceeding with the analysis of variance in order to normalize the data and stabilize the variance over the entire data range. b The percentages of post-emergence damping-off were transformed into Square Root before proceeding with the analysis of variance in order to normalize the data and stabilize the variance over the entire data range. c The values in brackets represent data expressed in percentages. d The values of the same column assigned the same alphabetical letter(s) are not significantly 5% threshold, according to the Duncan test.

*F. thapsinum* and *F. solani* represented by one and two isolates, respectively, caused the highest rates of pre and post-emergence damping-off on seedlings (56.25% - 58.33%). However, statistical results revealed that the different *Fusarium* species caused the same rate on seedlings but that these rates were significantly higher (28.53% - 58.33%) than those recorded by the control treatment (4.17%).

**Effect of *Fusarium* isolates on onion bulbs**

Results of the pathogenicity assessment of isolates on bulbs revealed that inoculation of bulbs with *Fusarium* isolates resulted in internal rotting of bulbs with a length ranging from 0.83 mm to 25 mm with an average of 13.18 mm. The results of the data analysis revealed a highly significant difference between isolates and allowed the isolates to be divided into 15 groups (*Figure 2*). Depending on the degree of pathogenicity (or extent of rot on the bulb), the isolates were classified into four groups: (*Table 4*).

The group of highly pathogenic isolates (TP), composed of seven isolates, three of which belong to the species *F. proliferatum* (129, 14, 132), and four to *F. oxysporum* (116, 150, 152, 166), which caused rots longer than 20 mm (*Figure 2*).

The group of moderately pathogenic isolates (MP) represented by 13 isolates including two belonging to *F. oxysporum* (19, 158), nine to *F. proliferatum* (11, 13, 14, 115, 117, 118, 121, 131, 137) one to *F. fujikuroi* (127) and one to *F. thapsinum* (135) with a rot length between 10 and 20 mm.
Table 4. Classification of isolates according to their degree of pathogenicity to bulbs.

| Fusarium species | Isolates | Classes defined according to the extent of bulb rot |
|------------------|----------|-----------------------------------------------------|
|                  |          | TP (rot > 20 mm) | MP (10 - 20 mm of rot) | PP (2 - 10 mm of rot) | LOC (<2 mm of rot) |
| *F. oxysporum*    |          | X              | X                      | X                      | X                   |
| I5               |          |                |                        |                        |                     |
| I7               |          |                | X                      | X                      |                     |
| I9               |          |                |                        |                        |                     |
| I16              |          |                | X                      |                        |                     |
| I42              |          |                |                        | X                      |                      |
| I49              |          |                |                        | X                      |                      |
| I50              |          |                | X                      |                        |                     |
| I52              |          |                | X                      |                        |                     |
| I58              |          |                | X                      |                        |                     |
| I59              |          |                |                        | X                      |                     |
| I63              |          |                | X                      |                        |                     |
| I66              |          |                | X                      |                        |                     |
| I67              |          |                | X                      |                        |                     |
| *F. proliferatum*|          | X              |                        | X                      |                     |
| I11              |          |                | X                      |                        |                     |
| I13              |          |                | X                      |                        |                     |
| I41              |          |                | X                      |                        |                     |
| I11              |          |                | X                      |                        |                     |
| I14              |          |                | X                      |                        |                     |
| I15              |          |                | X                      |                        |                     |
| I17              |          |                | X                      |                        |                     |
| I18              |          |                | X                      |                        |                     |
| I19              |          |                | X                      |                        |                     |
| I21              |          |                | X                      |                        |                     |
| I29              |          |                | X                      |                        |                     |
| I31              |          |                | X                      |                        |                     |
| I32              |          |                | X                      |                        |                     |
| I33              |          |                | X                      |                        |                     |
| I37              |          |                | X                      |                        |                     |
| *F. solani*      |          | X              |                        | X                      |                     |
| I24              |          |                | X                      |                        |                     |
| I38              |          |                | X                      |                        |                     |
| *F. fujikuroi*   |          | X              |                        | X                      |                     |
| I2               |          |                | X                      |                        |                     |
| I27              |          |                | X                      |                        |                     |
| *F. thapsinum*   |          | X              |                        | X                      |                     |
| I35              |          |                | X                      |                        |                     |
The group of low pathogenic isolates (PP) comprising ten isolates, five of which belong to *F. oxysporum* (I7, I42, I49, I63, I67), two to *F. proliferatum* (I11, I33), two to *F. solani* (I24, I38) and one to *F. fujikuroi* (I2) and which caused rots of lower lengths between 2 and 10 mm.

The group of non-pathogenic isolates comprising three isolates including two of the species *F. oxysporum* (I19, I5) and one of *F. proliferatum* (I19) which caused rots of lengths statistically similar to that recorded by the control (<2 mm).

Depending on the species of isolates, the estimated average rot lengths were between 3.33 and 16.10 mm and that observed with sterile distilled water was 0.83 mm (Figure 3). Statistical results revealed that the species *F. proliferatum*, *F. thapsinum*, *F. oxysporum* and *F. fujikuroi* were pathogenic on bulbs causing rots of significantly longer lengths (11.66 - 16.10 mm) than those caused by sterile distilled water (0.83 mm) (Figure 3). Based on these results, *F. solani* was not pathogenic on bulbs because the length of the rot it caused (3.33 mm) was not control treatment (0.83 mm).

### 3.2. Discussion

Out of 33 *Fusarium* isolates used in the study, 29 isolates representing 87.87% of them were found pathogenic to young onion plants. These pathogenic isolates included seven highly pathogenic isolates causing more than 50% of damping-off on seedlings, and the other 22 moderately pathogenic isolates with 20% to 50% of damping-off rates. The action of isolates was more remarkable on pre-emergence damping-off with 20% - 62.50% damping-off caused than on post-emergence damping-off with rates ranging from 4.17% to 16.67%.

Among the isolates that caused damping-off on young plants, all 15 isolates belonging to *F. proliferatum* were identified as highly pathogenic for 5 isolates and moderately pathogenic for 10 isolates. These results indicated that the species *F. proliferatum* is strongly involved in pre-emergence and post-emergence damping-off of onions in Burkina Faso. For *F. oxysporum*, among the 13 isolates...
Figure 3. Mean lengths of rot induced 14 days after inoculation of onion bulbs by five *Fusarium* species, in the presence of controls (sterile distilled water).

tested, eight were moderately pathogenic, four were identified as low pathogenic and one (I5) was non-pathogenic on seedlings. From these results, *F. oxysporum* also appears to play an important role in onion seedling damping-off in Burkina Faso, however, with a relatively lower involvement compared to *F. proliferatum*.

The species *F. solani*, *F. fujikuroi* and *F. thapsinum* were poorly represented in the study with two, two and one isolates respectively, but all these isolates were pathogenic causing significant damage to young plants, which implies their effective involvement in onion seedling damping-off in Burkina Faso. Indeed, of the two isolates of *F. solani* used in the study, one was highly pathogenic and the other moderately pathogenic on young plants. The only one *F. thapsinum* isolate tested was highly pathogenic by causing severe damping-off, and both isolates of *F. fujikuroi* were moderately pathogenic. The results of this study showed that the *Fusarium* isolates responsible of damping-off on onion seedlings in Burkina Faso belong to *F. proliferatum*, *F. solani*, *F. thapsinum*, *F. fujikuroi* and *F. oxysporum*, with greater aggressive-ness of the *F. proliferatum* and *F. thapsinum* isolates.

Of the 33 isolates used in the study, 30 isolates representing 90.90%, were pathogenic to bulbs. The isolates that caused the most significant rots (rotting length > 20 mm) belong to the species *F. oxysporum* and *F. proliferatum*. All isolates of *F. proliferatum* species used in the study were identified as highly pathogenic (for 3 isolates), moderately pathogenic (9 isolates) and low pathogenic (2 isolates), except isolate I19 which was found to be non-pathogenic. Inoculation of seeds and onion bulbs with isolate I29 resulted in high rates of damping-off on seedlings and significant rotting on bulbs, reflecting the high virulence of this isolate on both seedlings and bulbs. These results also suggest a strong involvement of *F. proliferatum* in onion bulb rot in Burkina Faso.

For *F. oxysporum*, only two of the 13 isolates tested were found to be non-pathogenic on bulbs. Indeed, four isolates of the species were highly pathogenic, two moderately pathogenic, five low pathogenic and two non-pathogenic, including isolate I5, which also did not cause damping-off on seedlings. Based
on these results, the species *F. oxysporum* also appears to play an important role in onion bulb rot in Burkina Faso. Rushi, [27] had shown that *F. oxysporum* is a fungus that causes damage to several vegetable crops including onions. The only isolate of *F. thapsinum* used in the study caused 10 to 20 mm of rot on the bulbs, indicating its importance in this disease. On the other hand, the two isolates of the species *F. solani* were all low pathogenic on bulbs, thus showing the low involvement of this species in bulb rots. This result contrasts with that obtained by Dabiré [23] who identified *F. solani* as one of the major pathogens responsible for onion bulb rot in Burkina Faso. This suggests the existence of several strains of *F. solani* on onion in Burkina Faso. *F. fujikuroi* also showed moderate pathogenicity to bulb rot because of the two isolates tested, one was moderately pathogenic and the other low pathogenic. This study reports for the first time the pathogenicity of *F. thapsinum, F. proliferatum* and *F. fujikuroi* on seedlings and onion bulbs in Burkina Faso. Jeon et al. [28] noted a predominance of *F. fujikuroi* in rice seeds in Asia. They also demonstrated that this species reduced the germination of rice seeds and caused typical symptoms of Balkanoe disease, leaf elongation and chlorosis on rice seeds or plants. *F. thapsinum* has previously been identified on sorghum seeds [29] [30]. According to Stokhom [30] *F. thapsinum* is one of the most common sorghum seed-borne ascomycetes in Burkina Faso. Pathogenicity tests carried out revealed that *F. thapsinum* caused strong growth inhibition in young plants with wilting symptoms on the leaves. Pedrozo and Little [31] also identified *F. thapsinum* on soybean seeds. Pathogenicity test results showed a reduction in emergence rates and increased mortality of young plants. Several other studies have previously been conducted on most *Fusarium* species worldwide [17] [23] [27] [32]. Indeed, the work of Stankovic [16] revealed that *F. proliferatum* was an important pathogen on onion and garlic (*Allium sativum*) in Europe and that there was a potential risk of mycotoxin accumulation in contaminated plants of these two crops. *F. proliferatum* has also been found responsible for onion and garlic bulb rots in Argentina [33] and India [34]. The pathogenicity of *F. oxysporum, F. proliferatum* and *F. solani* on onions had also been reported by several authors [8] [16] [27]. In Iran, Ghanbarzadeh [8] identified *F. oxysporum* as the most virulent species on both young plants and onion bulbs, *F. proliferatum* as the most destructive on bulbs in storage. Similar to the results of this study, these same authors also recognized *F. solani*, responsible for pre and post-emergence damping-off on seedling but weakly involved in onion bulb rot.

*Fusarium* species live in the soil as chlamydospores and attack the basal parts of the onion in the field [35]. According to Özer and Köyçü [36], the main source of implements can contribute to the spread of inoculum. Care should be taken during harvesting operations to avoid causing injury to bulbs as these injuries are routes of entry for the fungus. *Fusarium* species are associated with seeds. Adequate disinfection of the seeds before planting is necessary to effectively control the diseases they cause.
4. Conclusion

The pathogenicity of the 33 isolates belonging to five *Fusarium* species (*F. proliferatum*, *F. thapsinum*, *F. solani*, *F. oxysporum*, and *F. fujikuroi*) tested, on *Violet de Galmi* variety, induced percentages of seedling damping-off ranging from 4.17% - 70.83% with an average of 38.60%, and lengths of internal bulb rot ranging from 0.83 - 25 cm, with an average of 13.18 cm. *F. thapsinum* (1) and *F. proliferatum* (15) isolates were the most pathogenic on both seedlings (15/15) and bulbs (13/15). The isolates of *F. fujikuroi* were moderately pathogenic on both seedlings and bulbs. All isolates of *F. solani* were only pathogenic on seedlings but very few on bulbs. *F. oxysporum* had a broad spectrum of aggressiveness consisting of isolates with high or low virulence, or even avirulence in all stages of plant development. Indeed, among the 13 isolates of *F. oxysporum*, eight were pathogenic on seedlings and four non-pathogenic. On the bulbs 11 over 13 isolates were pathogenic and two non-pathogenic. Screening of these different isolates over a wider range of onion varieties would confirm the pathogenicity of these isolates on onion and determine susceptible and resistant varieties. This knowledge is necessary for the development of appropriate and sustainable management methods against these diseases.

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Conflicts of Interest

The authors declare no conflicts of interest regarding the publication of this paper.

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