Pathogenicity of *Fusarium* species associated with Bakanae disease of rice in Burkina Faso

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Short title: Evaluation of the pathogenicity of *Fusarium* species associated with Bakanae disease of rice in Burkina Faso.

Abstract

Contamination of rice seeds by certain *Fusarium* species is a major problem in achieving food self-sufficiency. The aim of this study was to determine the pathogenicity of *Fusarium* species associated with Bakanae disease in Burkina Faso. To this end, a pathogenicity test of 17 single-spore isolates belonging to *Fusarium chlamydosporum*, *Fusarium equiseti*, *Fusarium oxysporum* and *Fusarium thapsinum* species was carried out using the inoculation method by soaking rice seeds of variety FKR 19 in a conidial suspension of 10^6 conidia/ml. The seeds were then sown in trays containing sterilized potting soil and watered daily. Seedling height and the number of infected seedlings were assessed at day 21 after inoculation (DAI), and incidence rate and severity were determined on the basis of the disease scale. Results showed that the *F thapsinum* isolate (012-HB-1) caused the greatest elongation of plants with leaf symptoms typical of rice gigantism disease. Seed inoculation with the isolates (*F. chlamydosporum*, *F. equiseti*, *F. oxysporum*) recorded very low incidence rates (0 to 11.82%), reflecting a low capacity to induce disease symptoms, while the *F. thapsinum* isolate (012-HB-1) showed the highest incidence (88.69%) and severity index (34.35%). Control plants (sterile distilled water) were free from attack, with zero severity. Determining the pathogenicity of the *Fusarium* species responsible for rice gigantism is useful for developing disease management strategies.

Keywords: Rice, *Fusarium*, pathogenicity, gigantism, Bakanae, Burkina Faso

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Pathogénicité des espèces de *Fusarium* associées à la maladie du Bakanae du riz au Burkina Faso

Résumé

La contamination des semences de riz par certaines espèces de *Fusarium* constitue un problème majeur pour l'atteinte de l'autosuffisance alimentaire. L'objectif de cette étude est de déterminer le pouvoir pathogène des espèces de *Fusarium* associées à la maladie du gigantisme ou Bakanae du riz au Burkina Faso. Pour ce faire, un test de pathogénicité de 17 isolats monospores appartenant aux espèces de *Fusarium chlamydosporum*, *Fusarium equiseti*, *Fusarium oxysporum* et *Fusarium thapsinum* a été réalisé par la méthode d'inoculation par trempage des semences de riz de la variété FKR 19 dans une suspension conidienne de 10^6 conidies/ml. Les semences ont ensuite été semées dans des alvéoles contenant du terreau stérilisé et soumis à un arrosage quotidien. La hauteur des plantules et le nombre de plantules infectées ont été évalués au 21e jour après inocuation (JAI), et le taux d'incidence et la sévérité ont été déterminés sur la base de l'échelle des maladies. Les résultats révèlent que l'isolat de *F. thapsinum* (012-HB-1) entraine les plus fortes élongations des plantes avec des symptômes foliaires typiques de la maladie du gigantisme du riz. L'inoculation des semences avec les isolats (*F. chlamydosporum*, *F. equiseti*, *F. oxysporum*) enregistrent des taux d'incidence très faibles (0 à 11.82 %) traduisant une faible capacité à induire les symptômes de la maladie pendant que l'isolat *F. thapsinum* (012-HB-1) affiche la plus forte incidence (88,69 %) et l'indice de sévérité le plus élevé (34,35%). Les plantes témoins (Eau distillée stérile) sont indemnes d'attaque avec une sévérité nulle. La détermination de la pathogénicité des espèces de *Fusarium* responsables du gigantisme du riz est utile pour le développement de stratégies de gestion de la maladie.

Mots clés : Riz ; *Fusarium* ; pathogénicité ; gigantisme ; Bakanae ; Burkina Faso

Introduction

Rice is essentially a foodstuff and remains an important staple worldwide. World rice production is expected to increase by 58 million tonnes to 567 million tonnes (Mt) in 2030 (OCDE/FAO, 2021).

In Burkina Faso, rice ranks 4th among cereals (45,1013 tonnes), after maize (1,853,509 tonnes), sorghum (1,643,722 tonnes) and millet (705,344 tonnes) (DGSS, 2022). The country's rice production does not cover food needs, and in response to the growing demand for rice, the country imports this commodity. Indeed, husked rice imports from 2017 to 2022 varied from 8427.91 to 60008.9 tonnes in Burkina Faso (FAOSTAT, 2023). Despite the increase in production, mainly due to the expansion of lowland and irrigated areas, rice growing in Burkina Faso continues to face major challenges linked to the effects of abiotic

factors (unfavorable temperatures, floods, drought, etc.) and biotic factors (diseases, insects, birds, weeds) (MARAH, 2022). Rice is attacked by a large number of fungal diseases, including gigantism or "Bakanae" disease, which means "seedling madness" in Japanese. This disease, caused by one or more *Fusarium* species, presents a complex of symptoms including seedling blight, root and crown rot, stunting and the classic symptoms of wilting and abnormal elongation (KVAS et *al*., 2009). In addition to *Fusarium fujikuroi* identified and reported as the fungus responsible for Bakanae disease (AMATULLI et *al*., 2010), several other species of the *Gibberella fujikuroi* complex such as *Fusarium moniliforme*, *Fusarium proliferatum*, *Fusarium andiyazi*, *Fusarium verticillioides*, *Fusarium sacchari* and *Fusarium subglutinans* have been found to cause the disease in various countries (QUAZI et *al*., 2013). Rice gigantism causes high yield losses ranging from 3% to 95% (JING and SUGA, 2021) and is an emerging disease in several rice-producing countries (JEON et *al*., 2013; BASHYAL et *al*., 2016). Gigantism is mainly seed-borne, and high levels of seed infection by *Fusarium* species have been found in rice seed samples produced in Burkina Faso (*Fusarium semitectum* (45%), *Fusarium moniliforme* (33, 1%), *Fusarium equiseti* (17.9%)) (OUEDRAOGO et *al*., 2016). Similarly, work by NIKIEMA et *al*. (2020) revealed the presence of *Fusarium* spp. in 81.35% of rice samples, with infection rates ranging from 1 to 41%. *Fusarium* spp. was found in all agro-ecological zones of Burkina Faso, with prevalence rates ranging from 78.2% to 83.87%. Although precise knowledge of the identity of a pathogen responsible for a given disease remains the first step towards the implementation of adequate disease control measures (O'DONNELL and CIGELNIK, 1997), knowledge of the exact identity of the *Fusarium* species associated with rice gigantism in Burkina Faso has been little investigated. To this day, current research does not provide precise information on the etiology of rice gigantism in Burkina Faso. In order to overcome these shortcomings and contribute to a better understanding of the disease, this study was undertaken. The aim of this study is to determine the pathogenicity of *Fusarium* species associated with Bakanae disease in Burkina Faso.

I. Materials and methods

I.1 Fungal material

Seventeen single-spore isolates from four *Fusarium* species (*F. chlamydosporum*, *F. equiseti*, *F. oxysporum* and *F. thapsinum*), collected from rice seeds produced during the 2018 season in Burkina Faso and previously molecularly identified (NIKIEMA et *al*., 2023) were used for pathogenicity tests to identify the species(es) involved in rice gigantism. These 17 single-spore isolates were selected from 118 single-spore isolates on the basis of their morphological and biometric characteristics (mycelial color, conidial size, number of cells per conidium) (Table I). Thus, for each *Fusarium* species, single-spore isolates with different characteristics were selected for study.

Table I: List of single-spore isolates of *Fusarium* species used for pathogenicity testing

Order number	**Single-spore isolates**	**Species of origin**	**Localities**	**Agroecological zones**
1	085-S-1	*F. chlamydosporum*	Ouinpoulie	Sa
2	082-S-1	*F. chlamydosporum*	Ouinpoulie	Sa
3	087-S-2	*F. chlamydosporum*	Sergoussouma	Sa
4	010-C-2	*F. equiseti*	Komsilga	S-S
5	036-CO-2	*F. equiseti*	Mouna2	S-S
6	098-PC-2	*F. equiseti*	Kolgondiessé	S-S
7	097-PC-2	*F. equiseti*	Kolgondiessé	S-S
8	108-BM-2	*F. oxysporum*	Moundasso1	S-S
9	110-BM-2	*F. oxysporum*	Moundasso1	S-S
10	001-C-1	*F. thapsinum*	Thanghin-Dassouri	S-S
11	002-C-1	*F. thapsinum*	Thanghin-Dassouri	S-S
12	011-HB-1	*F. thapsinum*	Handié-Diehoun	So
13	012-HB-1	*F. thapsinum*	Handié-Diehoun	So
14	015-HB-1	*F. thapsinum*	Handié- Diehoun	So
15	048-N-2	*F. thapsinum*	Basgouema	S-S
16	079-CE-2	*F. thapsinum*	Bassedo	S-S
17	078-CE-2	*F. thapsinum*	Bassedo	S-S

S-S: Soudano-Sahelian; So: Soudanian; Sa: Sahelian

I.2 Plant material

Basic seeds from the FKR19 rice variety supplied by the rice and rice growing program were used for the experiment. FKR19 has a vegetative cycle of 115 days, and is grown in a lowland/pluvial ecology. Basic seed production gives an average yield of 3 to 4 tonnes per hectare. For certified seed, used directly by rice growers, yields can reach 5 to 6 tonnes per hectare. This variety was chosen because it is the most widely used by rice farmers.

I.3 Preparation of inocula

Each single-spore isolate was grown on PDA medium at 20-25°C under alternating illumination of 12 h of near-ultraviolet light and 12 h of darkness. After 10 days incubation, the surface of the sporulating colony was aseptically scraped off with a metal spatula and transferred to a test tube containing sterile distilled water. After vortexing the mixture (mycelium + water) for one minute, the resulting suspension was filtered through muslin to separate conidia from mycelial fragments. Conidial suspension concentrations were adjusted to around 10^6 conidia/ml using a Malassez cell (ZAINUDIN et *al*., 2008a).

I.4 Seed inoculation and seeding

Grains of rice variety FKR 19 disinfected with 15% sodium hypochlorite for 10 minutes (BAHOUS et *al*., 2010) were soaked in 20 ml of freshly prepared conidial suspension for 24 hours (FIYAZ et *al*., 2014). Control grains (non-inoculated control) were disinfected in the same way and then soaked in 20 ml sterile distilled water for 24 hours. Inoculated and control grains were sown in honeycomb trays containing sterilized potting soil and subjected to daily watering. The potting soil consisted of a mixture of soil taken from the Kamboinsé station to a depth of 20 cm, sand and organic manure (ox dung) in the proportions of 2 parts soil to 1 part sand and 1-part organic manure. The blister packs were placed in a greenhouse at a temperature of 25-30°C ± 3°C and a relative humidity of 60-80%. At the end of the evaluations, Koch's postulate was realized and consisted in re-isolating the fungi from diseased plant organs and then re-identifying them.

I.5 Experimental device

The experimental design was a randomized Fischer block with three replications and 18 treatments (17 isolates + sterile distilled water). Seeding was carried out at a rate of one seed per tray, for a total of 32

seeds per tray per treatment. In all, 54 blister packs were used for the trial. The plates were spaced 50 cm apart, both within and between replicates and treatments.

I.6 Data collection and statistical analysis

Data on the number of emerged seedlings 14 days after inoculation (DAI), seedling height (measured from the base to the tip of the last leaf), number of dead seedlings and incidence were collected on day 21 after inoculation (DAI). Seedling emergence rate, seedling mortality rate and infected seedling rate (including elongated and dead seedlings) (Fiyaz et al., 2014) and attack severity (Zainudin et *al*., 2008a) were calculated as follows:

Emergence rate (%) = (Number of emerged seedlings/Total number of seedlings sown) x 100

Mortality rate (%) = (Number of dead seedlings / Total number of emerged seedlings) x 10

Incidence (%) = (Number of infected seedlings / Total number of emerged seedlings) x 100

Severity index (%) = $\sum$ (Xi -1) ni / [E (xi) - 1] N $*$ 100

Where Xi = disease score for each plant, ni = number of plants in category Xi, N = total number of plants observed, E(Xi)= scale range.

Disease severity was assessed using the 0 to 4 disease scale proposed by ZAINUDIN et *al*. (2008a) (Table II).

Table II: Disease Assessment Scale (ZAINUDIN et *al*., 2008a)

Disease scale	Disease symptoms
0	Healthy, uninfected plants (no external symptoms)
1	Normal growth, but leaves begin to show signs of yellowish-green color.
2	Abnormal growth, leaves elongated, thin and yellowish-green; seedlings also shorter or taller than normal
3	Abnormal growth, elongated; leaves chlorotic, thin and brownish; seedlings also shorter or taller than normal.
4	Infected seedlings with fungal mass on surface or dead seedlings

Data were subjected to analysis of variance (ANOVA) using SAS 9.2 software. Means were compared using Duncan's test, with a confidence level of 5%.

II. Results

II.1.1 Effect of *Fusarium* isolates on seedling emergence and mortality

The results for emergence of inoculated rice seeds and mortality of rice seedlings are presented in Table III. Analysis of variance showed a highly significant difference ($p<0.001$) between isolates for rice seedling emergence and mortality. Emergence and mortality rates ranged from 34.72 to 81.94% and 0 to 30.23% respectively. The highest emergence rate (81.94%) was recorded by the control (seeds inoculated with distilled water). All isolates showed a significant reduction in seedling emergence following seed inoculation compared with the sterile distilled water control. *Fusarium thapsinum* isolate 012-HB-1 recorded the lowest emergence rate (34.72%) and the highest mortality rate (30.23%). The zero-mortality rate of the control (distilled water) was identical to those of isolates (085-S-1, 082-S-1, 087-S-2, 010-C-2, 097-PC-2, 108-BM-2) which did not cause seedling mortality after seed inoculation.

Table III: Emergence of inoculated seeds and mortality of rice seedlings

Isolate codes	Conidial suspension and control	Emergence rate 14 DAI (%)	Mortality rate 21 DAI (%)
Distilled sterile water	Distilled sterile water (Control)	81,94 a ± 3,34	0 d
085-S-1	*F. chlamydosporum*	48,61 bcd ± 0,01	0 d
082-S-1	*F. chlamydosporum*	40,27 cd ± 0,81	0 d
087-S-2	*F. chlamydosporum*	55,55 bc ± 0,71	0 d
010-C-2	*F. equiseti*	43,05 bcd ± 0,54	0 d
036-CO-2	*F. equiseti*	55,55 bc ± 0,71	5,62 cd ± 1,11
098-PC-2	*F. equiseti*	51,38 bcd ± 0,29	3,12 d ± 1,39
097-PC-2	*F. equiseti*	38,88 cd ±0,95	0 d
108-BM-2	*F. oxysporum*	43,05 bcd ±0,54	0 d
110-BM-2	*F. oxysporum*	58,33 b ± 0,98	2,08 d ± 2,43
001-C-1	*F. thapsinum*	45,83 bcd ± 0,26	1 d ± 3,51

002-C-1	*F. thapsinum*	40,27 cd ± 0,81	1 d ± 3,51
011-HB-1	*F. thapsinum*	52,77 bc ± 0,43	8,34 bcd ± 5,83
012-HB-1	*F. thapsinum*	34,72 d ± 1,37	30,23 a ± 25,72
015-HB-1	*F. thapsinum*	47,22 bcd ± 0,12	1 d ± 3,51
048-N-2	*F. thapsinum*	47,22 bcd ± 0,12	3,12 d ± 1,39
079-CE-2	*F. thapsinum*	45,83 bcd ± 0,26	13,57 bc ± 9,06
078-CE-2	*F. thapsinum*	41,66 bcd ± 0,67	15,06 b ± 10,55
Probability		<0,0001 ***	<0,0001***
Mean		48.45	4.51

Mean values in the same column followed by the same letter are not significantly different at the 5% level according to Duncan's test. DAI: Days After Inoculation; ***: Highly Significant

II.1.2 Height of rice seedlings assessed at 21 days after inoculation

Results on the height of rice seedlings 21 days after inoculation (DAI) are presented in Table IV. Analysis of variance for height revealed a highly significant difference ($p<0.0001$) between isolates. Plantlet height ranged from 19.47 to 38.58 cm, with a mean of 27.41 cm. Seedlings from seeds inoculated with *F. thapsinum* were significantly taller (27.83 to 38.58 cm) than those from seeds inoculated with the other isolates (19.47 to 24.34 cm) and those from control seeds (21.70 cm) (Figure 1B). *F. thapsinum* isolate caused plant and leaf elongation (Figure 1A), and leaf chlorosis (Figure 1C). However, isolate 012-HB-1 (*F. thapsinum*) showed the highest incidence of elongation and chlorotic leaves (88.69%). Isolate 012-HB-1 (*F. thapsinum*) caused the greatest plant elongation, with leaf symptoms typical of rice gigantism.

Table IV: Height of rice seedlings at 21 days after seeding (DAI)

Isolate codes	**Conidial suspension and control**	**Seedling height (cm) 21 DAI**
Distilled sterile water	Distilled sterile water (Control)	21,70 e ± 0,57
085-S-1	*F. chlamydosporum*	23,90 de ± 0,35
082-S-1	*F. chlamydosporum*	23,93 de ± 0,34
087-S-2	*F. chlamydosporum*	23,46 de ± 0,39
010-C-2	*F. equiseti*	22,51 e ± 0,49
036-CO-2	*F. equiseti*	21,59 e ± 0,38
098-PC-2	*F. equiseti*	23,46 de ± 0,39
097-PC-2	*F. equiseti*	24,34 de ± 0,40

108-BM-2	*F. oxysporum*	23,18 de ± 0,42
110-BM-2	*F. oxysporum*	19,47 e ±0,79
001-C-1	*F. thapsinum*	35,09 ab ± 0,76
002-C-1	*F. thapsinum*	33,55 b ± 0,61
011-HB-1	*F. thapsinum*	27,83 cd ± 0,04
012-HB-1	*F. thapsinum*	38,58 a ±1,11
015-HB-1	*F. thapsinum*	34,45 b ± 0,70
048-N-2	*F. thapsinum*	30,94 bc ± 0,35
079-CE-2	*F. thapsinum*	32,87 b ± 0,54
078-CE-2	*F. thapsinum*	32,58 b ± 0,51
Probability		< 0,0001 ***
Mean		27,41

Mean values in the same column followed by the same letter are not significantly different at the 5% level according to Duncan's test. DAI: Day After Inoculation; ***: Highly Significant

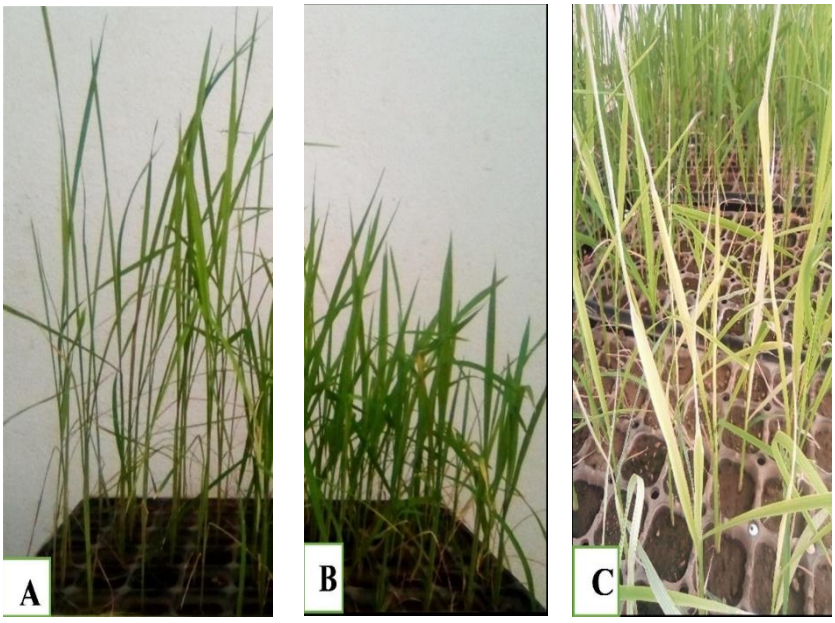

Figure 1: Symptoms of rice gigantism or Bakanae

A: Infected plants with abnormal elongation;
B: Healthy plants with normal size.
C: Infected plants with leaf chlorosis.

II.1.3 Incidence and severity of rice gigantism 21 days after seeding

The results concerning the incidence and severity of the disease recorded 21 days after inoculation (DAI) are presented in Table V. Analyses of variance for incidence and severity revealed a highly significant difference ($p<0.0001$) and a significant difference ($p<0.0067$) between isolates, respectively. Disease incidence ranged from 0 to 88.69%, with an average of 31.71%. Seed inoculation with isolate 010-C-2 (*F. equiseti*) caused no infection on seedlings (0% incidence), similar to the control (sterile distilled water), while isolate 012-HB-1 (*F. thapsinum*) caused the highest incidence (88.69%). Among the isolates studied, only those belonging to the *F. thapsinum* species (001-C-1, 002-C-1, 011-HB-1, 012-HB-1, 015-HB-1, 048-N-2, 079-CE-2, 078-CE-2) produced higher incidence rates than the sterile distilled water control. All other isolates recorded very low incidences (0 to 11.82%), reflecting their low capacity to induce disease symptoms. Control plants (sterile distilled water) were free of attack with zero severity (0%). Isolate 012-HB-1 (*F. thapsinum*) showed the highest severity index (34.35%), followed by 010-C-2 (*F. equiseti*) (32.78%), 078-CE-2 (*F. thapsinum*) (32.28%), 048-N-2 (*F. thapsinum*) (31.11%) and 085-S-1 (*F. chlamydosporum*), (31.15%), indicating that isolate causing severe infections belonged to the species *F. thapsinum*, *F equiseti* and *F. chlamydosporum*, with a high proportion of *F. thapsinum*. Isolate 097-PC-2 (*F. equiseti*), 087-S-2 (*F. chlamydosporum*) and 110-BM-2 (*F. oxysporum*), with severity scores of 18.33%, 15.35% and 12.77% respectively, were not very pathogenic. The other isolates were moderately pathogenic, with 21.41 to 29.01% severity index.

Table V: Incidence and severity of rice gigantism assessed 21 days after inoculation

Isolate codes	Conidial suspension and control	Incidence (%) 21 DAI	Disease severity (%) 21 DAI
Distilled sterile water	Distilled sterile water (control)	0 d	0 e
085-S-1	*F. chlamydosporum*	2,78 d ± 2,89	31,15 b ± 6,93
082-S-1	*F. chlamydosporum*	2,27 d ± 2,94	21,41 bc ± 2,81
087-S-2	*F. chlamydosporum*	4,20 d ± 2,75	15,35 d ± 8,87
010-C-2	*F. equiseti*	0 d	32,78 b ± 8,56
036-CO-2	*F. equiseti*	9,28 d ± 2,24	22,02 bc ± 2,17

098-PC-2	*F. equiseti*	11,82 d ± 1,98	24,44 bc ± 0,22
097-PC-2	*F. equiseti*	2,50 d ± 2,92	18,33 d ± 5,89
108-BM-2	*F. oxysporum*	3,13 d ± 2,85	24,53 bc ± 0,31
110-BM-2	*F. oxysporum*	4,36 d ± 2,73	12,77 de ± 11,45
001-C-1	*F. thapsinum*	49,19 c ± 1,74	24,55 bc ± 0,33
002-C-1	*F. thapsinum*	85,63 ab ± 5,39	28,33 bc ± 4,11
011-HB-1	*F. thapsinum*	61,82 abc ± 3,01	29,01 bc ± 4,79
012-HB-1	*F. thapsinum*	88,69 a ± 5,69	34,35 a ± 10,13
015-HB-1	*F. thapsinum*	67,71 abc ± 3,6	23,81 bc ± 0,41
048-N-2	*F. thapsinum*	47,38 c ± 1,56	31,11 b ± 6,89
079-CE-2	*F. thapsinum*	58,75 bc ± 2,70	28,69 bc ± 4,47
078-CE-2	*F. thapsinum*	71,39 abc ± 3,96	32,28 b ± 8,06
Probability		< 0,0001 ***	<0,0067 **
Mean		31,71	24,22

Mean values in the same column followed by the same letter are not significantly different at the 5% level according to Duncan's test. DAI : Day After Inoculation ; ***: Very Highly Significant.

III. Discussion

This study, carried out on 17 single-spore isolates from four *Fusarium* species (*F. chlamydosporum*, *F. equiseti*, *F. oxysporum* and *F. thapsinum*), enabled us to evaluate the pathogenicity of these isolates on rice plants of variety FKR 19 in the greenhouse. According to QUAZI et *al*. (2013), seed inoculation is the most appropriate method for assessing the development of rice gigantic disease, compared with soil inoculation. Pathogenicity determination reveals that all *Fusarium* species affect rice plants. Typical symptoms of rice gigantism were observed 14 days after inoculation only in seedlings inoculated with isolates of *F. thapsinum*. All isolates belonging to this species show some typical disease symptoms to varying degrees, and the most severely infected plants died 5 days after seeding. No symptoms of the disease were observed on the control plants, which were all healthy. The pathogenic power of *Fusarium* species belonging to the *G. fujikuroi* species complex on rice is observed through the inhibition of seed germination and the appearance of symptoms on seedlings. Differents isolates of *F. thapsinum* inhibit seed germination in different ways, and cause seedling mortality. In the present study, the *F. thapsinum* strain corresponding to isolate 012-HB-1 was the most pathogenic on rice

compared with the other *Fusarium* spp. species tested. Our results are similar to those of KHOSRAVI et *al*. (2019) who identified a virulent strain of *Fusarium thapsinum* with an incidence ranging from 21 to 50%. Infected seedlings are morphologically different from others due to their abnormal elongation. According to AHMAD and RAZA (1991), the disease can be transmitted by both seed and soil. The types of symptoms produced on the infected plant may depend on the strain of fungus and the nutritional conditions of the plant. Decay symptoms with powdery growth of conidiophores on the lower parts were not observed on the plants studied. In this experiment, the isolation of *F. thapsinum* from diseased rice plants enabled us to consider its possible role in the development of Bakanae symptoms. The realization of Koch's postulate confirmed that the fungus is pathogenic to rice plants and produces symptoms characteristic of Bakanae. Typical symptoms are thin, elongated, chlorotic seedlings that are often several centimetres taller than healthy seedlings. The increase in plant height is due to the ability of the inoculated species, either genetically or under mutation, to produce gibberellin (GA3), a plant hormone regulating plant development. GA3 is a simple growth hormone that promotes elongation of plant cells (JOHNSON and COOLBAUGH, 1990). Infected seedlings also show stunted growth, but do not produce adventitious or thread-like roots at the lower nodes. In fact, the pathogen also produces certain secondary metabolites that affect the growth of rice plants, such as carotenoids, fusarin, fumonisin, moniliformin and fusaric acid, as various studies have shown (ZAINUDIN et *al*., 2008b). Biological studies of these two substances have shown that fusaric acid causes stunting and gibberellin elongation in rice plants (NYVALL, 1999). Many traits associated with pathogenicity can be found in pathogenic variants, as well as in non-pathogenic variants of a species. The properties conferring pathogenicity are no longer considered as a specific property of the pathogen, but as the result of a complex and dynamic interaction with the host. The complexity of rice gigantism is due to the fact that it is a monocyclic disease whose pathogen is transmitted by seed and soil. According to OU (1985) seed-borne inoculum is a more important source, as soil-borne inoculum is rapidly reduced over time. Under conditions of favorable temperature, relative humidity and sunlight, *Fusarium* spores can survive in the absence of a host (AWAD et *al*., 2010). Survival of the pathogen is the first event leading to pathogenesis. The pathogen hibernates in perithecia and infects seeds

that are not pre-infected from the soil. Infected seeds are also a source of inoculum, and conidia germinate after seeding.

Disease incidence and severity are higher, particularly in *F. thapsinum* isolates. This is due to the genetic component of the pathogens. Overt damage to the host is a property of host-pathogen interactions. Disease development is a complex biological process in which the parasite depends entirely on its host for survival. Thus, despite their incidence, most isolates do not cause significant abnormal elongation of the seedling. Further studies are needed to clarify whether the variation in aggressiveness among isolates is caused by physiological characteristics of the isolates or by environmental factors, such as temperature and humidity, during the experiment itself. According to SIOU (2013), humidity and temperature play a vital role in *Fusarium* development, conditioning germination and infection. Each *Fusarium* species has a temperature and humidity optimum for which disease expression is maximal (ROSSI et *al*., 2001). For a host plant to be infected, it must be receptive to the pathogen, i.e. in a state of development or growth that allows the parasite to infect it. Depending on the case, this state may correspond to the entire lifespan of the plant, or to a more or less short period, sometimes just a few days (RAPILLY, 1992). As Bakanae is a rice disease that is favored by high temperatures, humidity and CO2 concentration, it is quite possible that it will increase with the hot, humid conditions that prevail in the context of climate change (MATIC et *al*., 2021). It would be advisable to implement an integrated strategy for managing a possible epidemic by monitoring the pathogen in the different rice-growing zones.

Conclusion

This study demonstrated the pathogenicity of *F. thapsinum*, *Fusarium equiseti*, *F. chlamydosporum* and *F. oxysporum* species in the development of rice gigantism in Burkina Faso. All *F. thapsinum* isolates showed typical disease symptoms to varying degrees, and the most severely infected plants died. Isolate 012-HB-1, belonging to *F. thapsinum*, was the most pathogenic on rice, with the highest incidence (88.69%) and a severity of 34.35%. As rice gigantism is an emerging disease, these results indicate the need to develop disease management strategies for sustainable rice production. Future work will involve screening different rice genotypes for resistance to Bakanae disease, with a view to proposing a varietal control method.

Conflict of interest

"All authors declare no conflict of interest".

Author contributions

NWF initiated this research, ZPE supervised the research work, CS and TIG corrected the manuscript, KK and SM supervised the research work.

References

AHMAD R. I. & RAZA T., 1991. Association of *Fusarium moniliforme* Sheld with the rice seeds and subsequent infection in Pakastan. *International Rice Research Newsletter*, 16 (4) :19-20.

AMATULLI M. T., SPADARO D., GULLINO M. L. & GARIBALDI A., 2010. Molecular identification of *Fusarium* spp. associated with Bakanae disease of rice in Italy and assessment of their pathogenicity. *Pl. Pathol*. 59 (5), 839-844. doi :10.1111/j.1365-3059.2010. 02319.x

AWAD W. A., GHAREEB, K., BOHM, J. & ZENTEK J., 2010. Decontamination and detoxification strategies for the *Fusarium* mycotoxin deoxynivalenol in animal feed and the effectiveness of microbial biodegradation. Food additives & contaminants. Part A, Chemistry, analysis, control, exposure & risk assessment, 27(4): 510–520. doi: 10.1080/19440040903571747

BAHOUS M., OUAZZANI TOUHAMI A., BENKIRANE R. & DOUIRA, A., 2010. Influence de l'âge de la plante sur la réceptivité des feuilles du riz à la pyriculariose, l'helminthosporiose et la curvulariose. *Revue Marocaine de Protection des Plantes*, 1 : 73-84.

BASHYAL, B. M., AGGARWAL, R., SHARMAL, S., GUGPTA, S., & SINGH, U. B., 2016**.** Single and combined effects of three *Fusarium* species associated with rice seeds on the severity of Bakanae disease of rice. *Journal of Plant Pathology*, *98* (3) : 405-412.

DGESS., 2022. *Enquête permanente agricole/Résultats définitifs 2021/2022. Direction Générale des Etudes et des Statistiques Sectorielles*. Ministère de l'agriculture des ressources animales et halieutiques, Burkina Faso. 15p

DESJARDINS A. E., MANANDHAR H. K., PLATTNER R. D., MANANDHAR G. G., POLING S. M. & MARAGOS C. M., 2000. *Fusarium* species from Nepalese rice and production of mycotoxins and gibberellic acid by selected species. *Applied and environmental microbiology*, 66(3), 1020–1025. doi :10.1128/AEM.66.3.1020-1025.

FAOstat., 2023. Production du riz au Burkina Faso. https://www.fao.org/faostat/fr/#data/QCL.

FIYAZ R. A., KRISHAN S. G., RAJASHEKARA H., YADAV A. K., BASHYAL B. M., BHOWMICK P. K., SINGH N. K., PRABHU K. V. & SINGH A. K., 2014. Development of high throughput screening protocol and identification of novel sources of resistance against Bakanae disease in rice (*Oryza sativa L.*). *Ind. J. Genet,* 74(4): 414-422.

JEON, Y. A., YU, S. H., LEE, Y. Y., PARK, H. J, LEE, S., SUNG, J. S., KIM, Y. G., LEE, H. S., 2013. Incidence, Molecular Characteristics and Pathogenicity of *Gibberella fujikuroi* Species Complex Associated with Rice Seeds from Asian Countries. *Mycobiology*, *41*(4) : 225-233.

JING, L. F., & SUGA, H., 2021. Various Methods for Controlling the Bakanae Disease in Rice. *Reviews in Agricultural Science*, 9 :195-205. doi: 10.7831/ras.9.0_195

JOHNSON S. W. & COOLGAUGH R. C., 1990. Light-Stimulated Gibberellin Biosynthesis in *Gibberella fujikuroi*. *Plant physiology*, 94(4) : 1696–1701. doi :10.1104/pp.94.4.1696

KHOSRAVI V., JAVAN-NIKKHAH M., SAREMI H. & NAEIMI S., 2019. Evaluation of the pathogenicity of several *Fusarium* species isolated from rice seed in laboratory and tracing of seeds transmission of rice Bakanae disease. *Iranian Journal of Plant Pathology*, 55(3): 193-208.

KVAS, M., MARASAS, W. F. O., WINGFIELD, B. D., WINGFIELD, M. J., & STEENKAMP E. T., 2009. Diversity and evolution of *Fusarium* species in the *Gibberella fujikuroi* complex. *Fungal Diversity*, *34*: 1–21.

MARAH., 2022. *Rapport bilan annuel 2021 « production végétale ».* Ministère de l'agriculture des ressources animales et halieutiques, Burkina Faso, 150 p.

MATIC S., GARIBALDI A. & GULLINO M. L., 2021. Combined and single effects of elevated CO2 and temperatures on rice Bakanae disease under controlled conditions in phytotrons. *Plant Pathol*, 70 :815-826.

NIKIEMA W. F., ZIDA, P. E., KEITA I., KOITA O., THIO I. G., NITIEMA L., KOITA K. & SAWADOGO M., 2023. Molecular Identification of *Fusarium* Species Associated with Rice Seeds Produced in Burkina Faso. *Journal of Agriculture and Environmental Sciences,*12. 6-17. doi: 10.15640/jaes. v12n12a2.

NIKIEMA, W. F., ZIDA, P. E., THIO, I. G., NITIÉMA, W. L., KOITA, K., & SAWADOGO, M. (2020). Incidence de *Fusarium* spp. associé aux semences de riz (*Oryza sativa* L.) au Burkina Faso. *Int. J. Biol. Chem. Sci, 14* (6) : 2160-2171. doi : 10.4314/ijbcs.14i6.18

NYVALL R. F., 1999. Field Crop Diseases. Iowa State University Press, USA. 1021 p.

OU S. H., 1985. Rice Diseases. 2nd ed. Commonwealth Mycological Institute, Kew, Surrey, England, UK. 380p.

O'DONNELL, K., & CIGELNIK, E., 1997. Two divergent intragenomic rDNA ITS2 types within a monophyletic lineage of the fungus *fusarium* are nonorthologous. *Molecular phylogenetics and evolution, 7*(1) : 103-116.

OCDE/FAO., 2021. *Perspectives agricoles de l'OCDE et de la FAO 2021-2030*. Éditions OCDE/FAO, Rome. p 139-154.

OUEDRAOGO, I., WONNI, I., SEREME, D., & KABORE, K. B., 2016. Survey of Fungal Seed-Borne Diseases of Rice in Burkina Faso. *International Journal of Agriculture Innovations and Research*, *5*(3) : 476-480.

QUAZI S. A. J., MEON S., JAAFAR H. & AHMAD Z. A. B. M., 2013. Characterization of *Fusarium proliferatum* through species specific primers and its virulence on rice seeds. *Int. J. Agric. Biol*., 15: 649-656.

RAPILLY F., 1992. Apports de l'épidémiologie à la connaissance de la résistance des végétaux aux agressions parasitaires. *Cahiers Agricultures*, 1(4): 239–248 (1).

ROSSI V., RAVANETTI A., PATTORI E. & GIOSUE S., 2001. Influence of temperature and humidity on the infection of wheat spikes

by some fungi causing *Fusarium* head blight. Journal of Plant Pathology, 83(3), 189-198.

SIOU D., 2013. Développement épidémique de la fusariose des épis de blé et conséquences des interactions entre espèces du complexe fusarien. Thèse de Doctorat, Université Paris-Sud11, 198 p.

SUN S. K. & SNYDER W. C., 1981. The Bakanae disease of the rice plant. In: *Fusarium*: Diseases, Biology and Taxonomy, (Nelson, P.E., T.A. Toussoun and R.J. Cook, Eds). The Pennsylvania State University Press, University Park, PA., 104-113.

ZAINUDIN N. A. I. M., RAZAK A. A. & SALLEH B., 2008a. Bakanae disease of rice in Malaysia and Indonesia: Etiology of the causal agent based on morphological, physiological and pathogenicity characteristics. *J. Pl. Prot. Res,* 48: 475-485.

ZAINUDIN N. A. I. M., RAZAK A. A. & SALLEH B., 2008b. Secondary metabolite profiles and mating populations of *Fusarium* species in section Liseola associated with Bakanae disease of rice. *Malay. J. Microbiol*, 4: 6-13.