



Isolation and molecular identification of fungi associated with root rot and head blight of wheat (*Triticum aestivum* L.) in Ain Al-Tamur district, Karbala province, Iraq

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Abstract

This study was conducted in the Graduate Studies Laboratory of the College of Agriculture, University of Kerbala, with the aim of isolating and identifying fungi associated with root rot and head blight in wheat plants using Polymerase Chain Reaction (PCR) technology. PCR analysis results showed that the most pathogenic isolates were *Alternaria infectoria* (B9), *Fusarium equiseti* (K1), *Fusarium oxysporum* (A4), *Fusarium culmorum* (SH2), *Fusarium equiseti* (K6), and *Fusarium pseudograminearum* (F2). The fungal isolates were registered at the National Center for Biotechnology Information (NCBI) under the accession numbers: PV769992.1, PV769993.1, PV769994.1, PV769995.1, PV769996.1, PV770002.1, respectively. The results also showed the frequent occurrence of several species of *Fusarium* in the studied samples.

Keywords: Fungi, *Triticum aestivum* L., Fusarium Head Blight, Field survey, wheat

Introduction

Wheat (*Triticum aestivum* L.), a member of the Poaceae family, is the staple crop for the majority of the world's population. It is grown locally and worldwide. It is rich in proteins, including gluten, minerals (copper, magnesium, zinc, phosphorus, and iron), vitamins (B and E groups), riboflavin, niacin, thiamin, and dietary fiber. The proteins stored in wheat seeds represent an important source of nutrition and energy and play a key role in determining the quality of bread [1]. Wheat is one of the oldest field crops cultivated in the world as a primary source of food. It is cultivated in Iraq in large areas, especially in the northern provinces, as the total cultivated area reached 8,420 thousand dunums for the winter season of 2023, with a total production of 4 million and 248 thousand tons [2]. Wheat is affected by many pathogens and pests, causing significant losses. The most threatening pathogens are fungi, which cause serious diseases on roots, leaves, and spikes, as one of the most threatening plant pathogens in wheat-growing areas [3]. Fusarium Head Blight (FHB), caused by several fungal pathogens, primarily *F. graminearum*, is a major threat to global wheat production. It is a soil-dwelling fungus and is one of the most serious challenges facing wheat cultivation in Iraq and the world, as it leads to

significant yield losses and mycotoxin contamination [4]. which can have acute and chronic toxic effects on humans and animals that consume infected plant parts [5]. Sarowar et al. [6]. reported that the fungus infects the floral tissues of small grains such as wheat and barley, resulting in the production of small, light, shrivelled grains contaminated with mycotoxins, especially deoxynivalenol, which contaminates the grains and acts as a virulence factor to promote the spread of head blight (FHB) in infected spikes, which can interfere with various plant functions to promote disease development [7]. Other toxins produced by the fungus include zearalenone (ZEA) and nivalenol (NIV) [8,9]. Root rot diseases are specific to agriculture worldwide, causing economic losses that vary depending on the causative agent, plant susceptibility, and environmental conditions. Among the various pathogens, root rot fungi such as *M. phaseolina*, *F. oxysporum*, *F. moniliforme*, and *R. solani* are among the most destructive pathogens of cereals including wheat [10]. Pathogens invade root hairs early in the germination stage and cause infection to the true roots, which then colonize the crown region of the plant, causing wilting and early death, resulting in severe economic losses to the crop [11]

Therefore, the study aimed to isolate and diagnose the fungi associated with root rot and ear blight in wheat plants using PCR technology.

Materials and Methods

Field Survey of the Disease

A field survey was conducted during the 2023/2024 agricultural season across several wheat fields in the Ain Al-Tamr district of Karbala to estimate the percentage of infection caused by root rot and spike blight. The fields were selected based on the sampling areas outlined in the aforementioned sample collection table, with two fields chosen from each of the eight designated regions, resulting in a total of 14 fields .

The survey followed the methodology described by Bock et al. [12]. employing a diagonal walking pattern. Five evenly distributed survey points were established within each field, and a standardized area of 1 m² was examined at each point. Within this area, 20 randomly selected plants were assessed for disease symptoms. The infection percentage was then calculated using the following formula:

$$\text{Infection \%} = \frac{\text{Number of Infected Plants}}{\text{Total Number of Examined Plants}} \times 100$$

Isolation and identification of fungi associated with wheat *Triticum aestivum*

The fungi were isolated from the root, crown and spike area of wheat (*Triticum aestivum* L.), which showed symptoms of infection such as stunted growth, root rot, crown rot, and light pink discoloration of the spike sheaths, which later turned brown. The grains produced by the infected spikes were shrunken and lightweight, The infected samples were collected from some farms located in the Ain Al-Tamr district,

west of Karbala province, Table (1), and brought to the postgraduate studies laboratory at the College of Agriculture, University of Kerbala to conduct the process of isolating the pathogens. The plant parts were superficially sterilized using a 1% sodium hypochlorite solution, then five pieces were transferred to each Petri dish containing Potato Dextrose Agar medium using an autoclave at a temperature of 121°C and a pressure of 4 pounds per inch² for 20 minutes and incubated at a temperature of 25 ± 2°C for three days. Then the fungi were purified by transferring a part of the tip of the fungal colony to another plate containing the PDA medium. Then the fungal isolates obtained from the isolation process were stored in test tubes containing the PDA slant medium (Agar slants), and all the tubes were stored in the refrigerator at a temperature of (4°C).

Table (1): Areas from which wheat plants (*Triticum aestivum* L.) infected with root rot and ear blight were collected

Location	collection location	Type	Symbol	Collection date
Karbala/ Ain al-Tamr	Fayda Al-Bu Hwa	Jad	F	2024 / 3 / 7
	Abu Qabr	Wafih	K	2024 / 3 / 7
	Al-Shakhat	Ibaa 99	SH	2024 / 3 / 9
	Khadhira	Bora	W	2024 / 4 / 20
	Sadah	Nizar	A	2024 / 3 / 23
	Khashiba	Arabia	C	2024 / 3 / 28
	Wadi Al-Shih	Adna 99	B	2024 / 3 / 30

Testing the pathogenicity of isolated fungi on wheat seed germination

The pathogenicity of the isolated fungi was tested, which were 7 isolates of *Fusarium sp.*, 4 isolates of *Rhizctonia sp.*, and 2 isolates each of *Alternaria sp.*, *Stemphiliium sp.*, and *Macrophomena sp.* on WA medium, using the plate method followed by [13]. A 0.5 cm diameter disc was taken from the edge of pure fungal colonies of 7 days old growing on PDA medium and placed in the middle of a plastic Petri dish containing WA agar medium. Then the inoculated dishes were incubated for 3 days at a temperature of 25 ± 2 °C. After that, the superficially sterilized local wheat seeds were planted on the edges of the fungal colonies growing in the dish at a rate of 10 seeds in each dish. Each fungal isolate was repeated three times, in addition to the control treatment by planting sterilized wheat seeds without fungus and with the same number of replicates . Then, all dishes were incubated at a temperature of 25 ± 2 °C until all seeds germinated in the control treatment, and the germination percentage was then calculated [14] as follows:

$$\% \text{ germination} = \frac{\text{treatment number of sprouted seeds}}{\text{compared to sprouted seeds number}} \times 100$$

Abott's equation [15] was also used to calculate the percentage of inhibition as follow:

$$\% \text{ inhibition} = \frac{\text{by treatment sprouted seeds number} - \text{comparison in sprouted seeds number}}{\text{by comparison sprouted seeds number}} \times 100$$

Testing the pathogenicity of the fungal isolates under study on germination and infection of wheat seeds (*Triticum aestivum* L.) in plastic pots under field conditions

This experiment was carried out by mixing a mixture of soil with peat moss (1:2) and sterilized by a steam sterilizer at a temperature of 121°C and a pressure of 15 pounds per inch² for 60 minutes twice for two consecutive days. Then, the soil was inoculated with the fungus carried on millet seeds at a rate of 1% after mixing the inoculum in polyethylene bags with the soil and peat moss to homogeneity. Then, the soil was placed in 1 kg plastic pots. Then, the soil was moistened by adding water to it and covered with perforated polyethylene bags (to maintain moisture) for 48 hours. Then, local wheat seeds were planted at a rate of 10 seeds/pot and watered carefully whenever necessary. After the wheat seeds in the comparison had completed germination, the percentage of germination and inhibition was calculated.

Molecular identification

To study the genetic and hereditary composition of the selected fungal isolates and compare it with the genome of global isolates, 7 fungal isolates that are the most pathogenic to wheat plants were selected by DNA sequence analysis, where the commercial DNeasy plant kits were used for the purpose of extracting and purifying genomic DNA from pathogenic fungi, following the instructions of the manufacturer. The kit used Ready-To-Go PCR Beads in addition to the primers (ITS4 (5'-TCCTCCGCTTATTGATATGC-3' and ITS5 (5'-GGAAGTAAAAGTCGTAACAAGG-3')), which amplify the Internal transcribed spacer (ITS) duplicated regions located within the small and large subunit genes that make up the ribosomes in the fungal chromosome [16] The PCR amplicons were sent to Macrogen, South Korea. After receiving the data, the data were evaluated, cleaned, and analyzed using Chromas and ApE plasmid Editor. The Basic Local Alignment Search Tool (BLAST) was then used to determine the similarity between the sequences of the studied fungi and those identified globally and stored in the GenBank repository of the National Center for Biotechnology Information. The results of this analysis process were also used to build a genetic tree that shows the degree of similarity between local and global fungal isolates, using the Sequence Alignment Editor BioEdit and Molecular Evolutionary Genetics Analysis programs. The sequences of the nitrogenous bases of the local fungal isolates were then deposited in the GenBank repository after assigning their own symbols [17].

Statistical Analysis

All experiments were conducted according to a completely randomized design (C.R.D.) as single-factor experiments. Means were compared using the least significant difference (LSD) method at a probability level of 0.05 [18].

Results and discussion

Survey Findings on Wheat Fields in Karbala / Ain Al-Tamur

The comprehensive field survey conducted in Karbala Governorate (Ain Al-Tamr district) revealed that all examined wheat fields were infected with root rot and spike blight diseases. The widespread incidence of these diseases is likely the result of several interrelated factors. Continuous wheat monoculture, or the repeated cultivation of other Poaceae crops in the same fields, facilitates the buildup and long-term persistence of pathogenic *Fusarium* inoculum in the soil. Similar findings have been reported globally, where monoculture systems significantly increase pathogen inoculum levels compared with diverse crop rotations that disrupt disease cycles [19,20]. Favorable environmental conditions—particularly warm and humid temperature regimes—further enhance disease development. Such conditions are known to favor the proliferation of soil-borne pathogens responsible for crown and root rot, as well as *Fusarium* head blight, which can also contaminate grains with mycotoxins such as deoxynivalenol (DON) [21,5]. Excessive and recurrent use of chemical pesticides, which may adversely affect both the environment and plant health [22,23,24], Table (2) shows the infection rate and Infection % in the field survey.

Table (2): The Infection % and the Infection Rate in the field survey

Location	collection location	Field No.	Infection %	Infection Rate
Karbala / Ain al-Tamr	Faidat Al-Bu Hwa	1	38.3	36.9
		2	33.5	
	Abu Qabr	1	45	50
		2	55	
	Al-Shakhat	1	38.9	39.6
		2	40.3	
	Khadhira	1	55.9	54.6
		2	53.3	
	Sadah	1	44	42.4
		2	40.8	
	Khashiba	1	23.5	21.9
		2	20.3	
	Wadi Al-Shih	1	13.3	15.9
		2	18.5	

Isolation and identification of fungi associated with wheat plant

The results of the sample collection and isolation of the associated fungi showed that the disease of root rot of spikes was present in all the fields included in the study in Karbala province during the growing season of 2024. The reason for the spread of the disease in these areas may be attributed to the repeated cultivation of wheat or to the cultivation of other crops belonging to the Poaceae family, which led to the accumulation of pathogenic fungal inoculum that remains in the soil. The suitability of environmental conditions, especially temperatures, or the widespread and repeated use of chemical pesticides may have led to the development of resistance to pathogens. Recurrent infection with *Fusarium* spp. has been observed. The results are consistent with what has been reported by several studies [25,26,27,28]. and *Rhizctonia* spp. The results are consistent with what has been reported by several studies [29,30,31].

Testing the pathogenicity of fungal isolates on Water Agar medium

The results (Table 3) showed that all tested fungal isolates led to a significant reduction in the germination percentage compared to the control treatment, in which the seed germination percentage reached 100%. The isolates [K6, SH2, F2, A4] outperformed the rest of the isolates in reducing the germination percentage, as the average germination percentage reached 0.0% (Figure 1), and the inhibition percentage reached 100%. This was followed by isolate (C2), in which the germination percentage reached 3.33% and the inhibition percentage reached 96.67%, while the germination percentage for the rest of the isolates ranged between 10.00 - 86.67% and the inhibition percentage reached 90% 13.33%. The differences in pathogenicity of isolates may be attributed to genetic differences among them, as well as isolates of the same species collected from different regions, or to differences in the ability of isolates to secrete pectin and cellulose-degrading enzymes, including pectinase, phosphatase, cellulase, methylesterase, pectinase, and pectinmethylhydrolase, which have a significant impact on fungal pathogenicity, in addition to the ability of these fungi to produce some toxins of a phenolic and glycosidic nature. These results are consistent with many studies [32,33].

Table (3): The pathogenicity of isolated fungi on wheat seed germination

Seq	Treatments	% germination	% inhibition
1	Control	100	0.00
2	<i>Fusarium equiseti</i> (K1)	0.00	100
3	<i>Rhizctonia</i> sp. (K2)	33.33	66.66
4	<i>Fusarium equiseti</i> (K6)	0.00	100
5	<i>Rhizctonia</i> sp. (C2)	3.33	96.67
6	<i>Stemphiliium</i> sp. (C6)	13.33	86.67

7	<i>Fusarium sp. (sh1)</i>	30.00	70.00
8	<i>Fusarium culmorum (Sh2)</i>	0.00	100
9	<i>Rhizctonia sp. (F1)</i>	10.00	100
10	<i>Fusarium pseudograminearum (F2)</i>	0.00	100
11	<i>Fusarium sp. (F4)</i>	46.67	53.33
12	<i>Fusarium oxysporum (A4)</i>	0.00	100
13	<i>Rhizctonia sp. (A5)</i>	33.33	66.67
14	<i>Stemphiliium sp. (B5)</i>	30.00	70.00
15	<i>Macrophomena sp. (B10)</i>	80.00	20.00
16	<i>Alternaria infectoria (B9)</i>	86.67	13.33
17	<i>Macrophomena sp. (W1)</i>	46.67	53.33
18	<i>Alternaria sp. (W3)</i>	70.00	30.00
L.S.D		5.665	5.962

*Note that each number is the result of three repetitions.

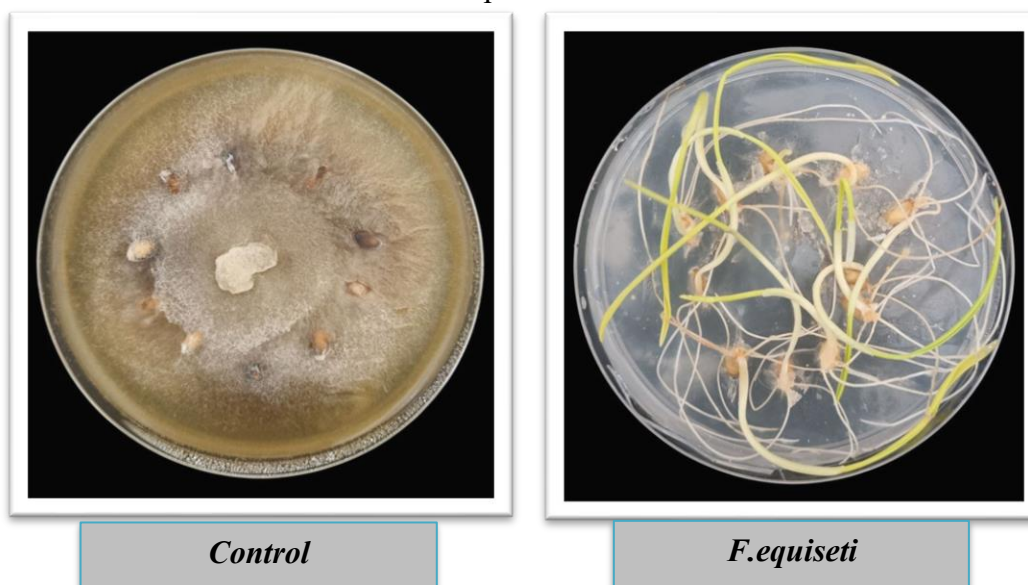


Figure (1): The pathogenicity of the fungal isolates under study using wheat seeds on (WA) medium

Testing the pathogenicity of the fungal isolates under study on germination and infection of wheat seeds (*T. aestivum* L.) in plastic pots under field conditions

The results of the pathogenicity in plastic pots (Table 4) showed that all the fungal isolates under study were able to infect wheat seeds and seedlings, with the germination percentage ranging between 0.00 - 56.67% and inhibition 100 - 43.33%.



These treatments differed significantly from the healthy control treatment, which had an infection percentage of 0.00% and a germination percentage of 100%. The isolates (Sh2, F2, K1, and A4) were superior in their pathogenicity, with a germination percentage of 0.00% and an inhibition percentage of 100% (Figure 2), while the isolate (F2) had the lowest pathogenicity, with a germination percentage of 43.33% and an inhibition percentage of 56.67%. Other isolates varied in their pathogenicity on wheat seeds and plants, with some isolates causing pre-germination seed rot or seedling death. Many *Fusarium spp.* species cause seed and root rot and seedling death on many plant hosts. This is due to their parasitic nature and the abundant growth of fungal mycelium within vascular tissues, which obstructs the flow of water and salts to the leaves. Furthermore, the enzymes and toxins produced by the fungi play a major role in decomposing plant cell walls and allowing the fungus to penetrate plant tissues such as roots, seeds, and stems, potentially preventing germination or killing embryos [33,34,27]. Based on these results, the isolates that were most inhibitory to wheat growth were selected for molecular diagnosis.

Table (4): Detection of pathogenic isolates using wheat seeds in plastic pots under greenhouse conditions

Seq	Treatments	% germination	% inhibition
1	Control	100	0.00
2	<i>Fusarium equiseti</i> (K1)	0.00	100
3	<i>Rhizctonia sp.</i> (K2)	6.67	93.33
4	<i>Fusarium equiseti</i> (K6)	6.67	93.33
5	<i>Rhizctonia sp.</i> (C2)	23.33	76.67
6	<i>Stemphilium sp.</i> (C6)	26.67	73.33
7	<i>Fusarium sp.</i> (sh1)	20.00	80.00
8	<i>Fusarium culmorum</i> (Sh2)	0.00	100
9	<i>Rhizctonia sp.</i> (F1)	6.67	93.33
10	<i>Fusarium pseudograminearum</i> (F2)	0.00	100
11	<i>Fusarium sp.</i> (F4)	56.67	43.33
12	<i>Fusarium oxysporum</i> (A4)	0.00	100
13	<i>Rhizctonia sp.</i> (A5)	23.33	76.67
14	<i>Stemphilium sp.</i> (B5)	30.00	70.00
15	<i>Macrophomena sp.</i> (B10)	33.33	66.67
16	<i>Alternaria infectoria</i> (B9)	10.00	90.00

17	<i>Macrophomena sp. (W1)</i>	53.33	46.67
18	<i>Alternaria sp. (W3)</i>	20.00	80.00
L.S.D		12.098	7.126

*Note that each number is the result of three repetitions.

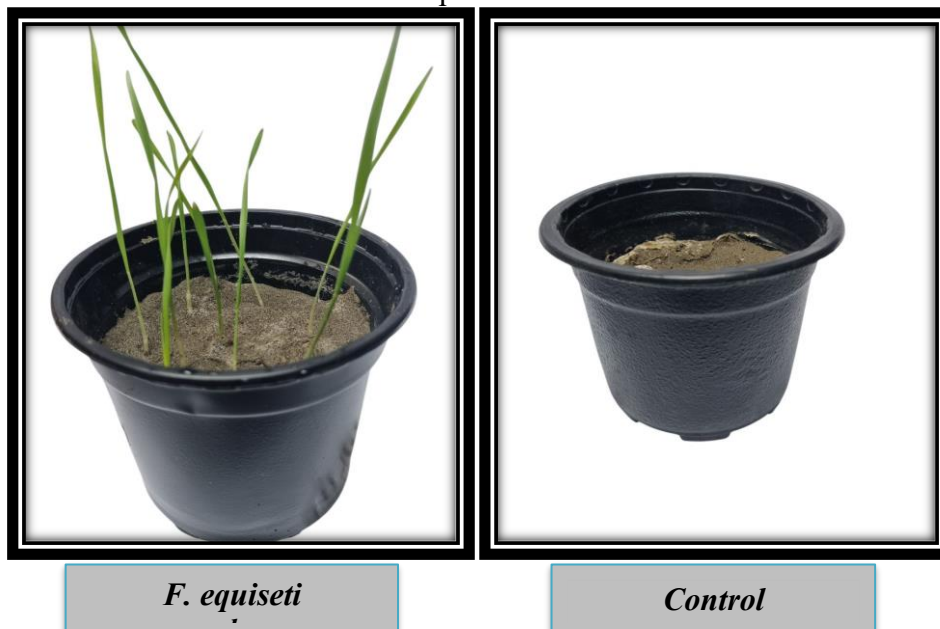


Figure (2): The pathogenicity of a number of fungal isolates under study in plastic pots.

Molecular identification

The results showed that the DNA was amplified by polymerase chain reaction (PCR) with a size of 600 bp using the primers ITS1-ITS4. The analysis of the sequence of the amplified DNA by polymerase chain reaction (PCR) showed that the fungal isolates isolated in this study belong to the fungi *A. infectoria* (B9), *F. equiseti* (k1), *F. oxysporum* (A4), *F. culmorum* (SH2), *F. equiseti* (k6), *F. pseudograminearum* (f2), *F. pseudograminearum* (F2). The results also demonstrated the presence of a percentage of variation between the fungal isolates diagnosed in this study and the isolates previously diagnosed and registered at the National Center for Biotechnology Information (NCBI). Therefore, these isolates were registered at the aforementioned center under the accession numbers (PV769992.1 , PV769993.1 , PV769994.1 , PV769995.1 , PV769996.1 , PV770002.1) , respectively.

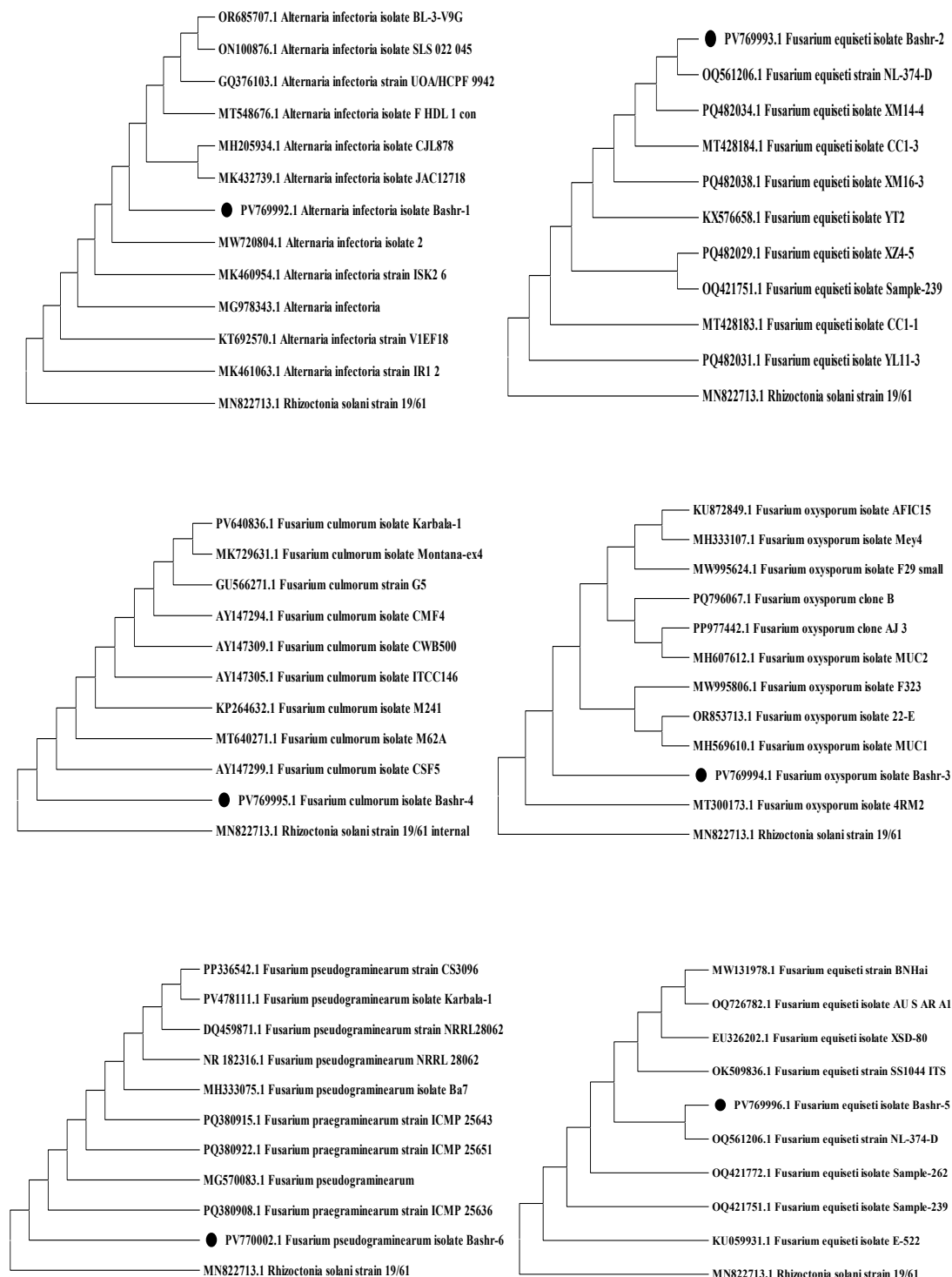


Figure (3): The phylogenetic trees of the Studied Fungi

References

- 1) Khalid, A., Hameed, A., & Tahir, M. F. (2023). Wheat quality: A review on chemical composition, nutritional attributes, grain anatomy, types, classification,

- and function of seed storage proteins in bread making quality. *Frontiers in Nutrition*, 10, 1053196. <https://doi.org/10.3389/fnut.2023.1053196>
- 2) Central Statistical Organization, Iraqi Ministry of Planning. (2023). *[Title of the report if available]*. [Publisher if different].
 - 3) Abass, M. H., Madhi, Q. H., & Matrood, A. A. A. (2021). Identity and prevalence of wheat damping-off fungal pathogens in different fields of Basrah and Maysan provinces. *Bulletin of the National Research Centre*, 45, 48–54. <https://doi.org/10.18805/ag.R-1770>
 - 4) Kaur, G., Rana, S. K., Masurkar, P., Anand, S., Sahoo, M., Singh, S., & Sinha, P. (2024). Bio efficacy of fungicides against the wheat head blight causing fungus, *Fusarium graminearum*. *[Journal Name]*. [Volume(Issue)], pages. [DOI if available]
 - 5) Özdemir, F. (2022). Host susceptibility of CIMMYT's international spring wheat lines to crown and root rot caused by *Fusarium culmorum* and *Fusarium pseudograminearum*. *Agronomy*, 12(12), 3038. <https://doi.org/10.3390/agronomy12123038>
 - 6) Sarowar, S., Alam, S. T., Makandar, R., Lee, H., Trick, H. N., Dong, Y., & Shah, J. (2019). Targeting the pattern-triggered immunity pathway to enhance resistance to *Fusarium graminearum*. *Molecular Plant Pathology*, 20(5), 626–640. <https://doi.org/10.1111/mpp.12773>
 - 7) Hao, G., McCormick, S., Usgaard, T., Tiley, H., & Vaughan, M. M. (2020). Characterization of three *Fusarium graminearum* effectors and their roles during *Fusarium* head blight. *Frontiers in Plant Science*, 11, 579553. <https://doi.org/10.3389/fpls.2020.579553>
 - 8) Perincherry, L., Lalak-Kanczugowska, J., & Stępien, L. (2019). *Fusarium* produced mycotoxins in plant-pathogen interactions. *Toxins*, 11(11), 664. <https://doi.org/10.3390/toxins11110664>
 - 9) Abd Oun, A. G. (2023). *Phenotypic and genotypic identification of seed-borne fungi on some wheat cultivars with the determination of chemical compounds efficiency in reducing fungi and their products* (Doctoral dissertation, University of Basrah). <https://doi.org/xxxxxx> [if available]
 - 10) Islam, M. R., Sultana, T., Joe, M. M., Yim, W., Cho, J. C., & Sa, T. (2013). Nitrogen-fixing bacteria with multiple plant growth-promoting activities enhance growth of tomato and red pepper. *Journal of Basic Microbiology*, 53(12), 1004–1015. <https://doi.org/10.1002/jobm.201200141>
 - 11) Moya-Elizondo, E. A., & Jacobsen, B. J. (2016). Integrated management of *Fusarium* crown rot of wheat using fungicide seed treatment, cultivar resistance,

- and induction of systemic acquired resistance (SAR). *Biological Control*, 92, 153–163. <https://doi.org/10.1016/j.biocontrol.2015.10.006>
- 12) Bock, C. H., Poole, G. H., Parker, P. E., & Gottwald, T. R. (2010). Plant disease severity estimated visually, by digital photography and image analysis, and by hyperspectral imaging. *Critical Reviews in Plant Sciences*, 29(2), 59–107. <https://doi.org/10.1080/07352681003604292>
- 13) Christensen, M. J., Falloon, R. E., & Skipp, R. A. (1988). A Petri plate technique for testing pathogenicity of fungi to seedlings and inducing fungal sporulation. *Australasian Plant Pathology*, 17(2), 45–47. <https://doi.org/10.1071/APP9880045>
- 14) Khudier, N. H., & Abdalmoohsin, R. G. (2023, December). First report of *Acrophialophora jodhpurensis* causing root rot and seedling damping-off of pepper (*Capsicum annuum* L.) in Karbala Province, Iraq. In *IOP Conference Series: Earth and Environmental Science* (Vol. 1262, No. 3, p. 032009). IOP Publishing. <https://doi.org/10.1088/1755-1315/1262/3/032009>
- 15) Abbott, W. S. (1925). A method of computing the effectiveness of an insecticide. *Journal of Economic Entomology*, 18(2), 265–267. <https://doi.org/10.1093/jee/18.2.265a>
- 16) White, T. J., Bruns, T., Lee, S. H., & Taylor, J. W. (1990). *PCR protocols: A guide to methods and applications*. Academic Press.
- 17) Jaber, M. H. (2020). *Isolation and identification of fungi causing seed rot and seedling death of wheat (Triticum aestivum) in Karbala province and their control using the integration of some varieties, nano-compounds and the biological agent Trichoderma harzianum* (Master's thesis, College of Agriculture, University of Karbala).
- 18) Al-Rawi, K. M., & Khalaf Allah, A. A. M. (2000). *Design and analysis of agricultural experiments* (2nd ed.). College of Agriculture and Forestry, University of Mosul; Ministry of Higher Education and Scientific Research.
- 19) Bankina, B., Bimšteine, G., Arhipova, I., & Priekule, I. (2013). Winter wheat crown and root rot are affected by soil tillage and crop rotation in Latvia. *Plant Protection Science*, 49(1), 28–34. <https://doi.org/10.2478/plp-2013-0004>
- 20) Tilmann, H., Sommerfeldt, N., & Miedaner, T. (2016). Influence of crop rotation and soil management on soil borne cereal pathogens. *Crop & Pasture Science*, 67(12), 1206–1214. <https://doi.org/10.1071/CP16310>
- 21) Sakr, N. (2022). Fusarium head blight: Biology, epidemiology and control. *The Open Agriculture Journal*, 17, e187433152211150. <https://doi.org/10.2174/1874331502217010150>

- 22) Liu, R., Li, J., Zhang, L., Feng, T., Zhang, Z., & Zhang, B. (2021). Fungicide difenoconazole induced biochemical and developmental toxicity in wheat (*Triticum aestivum* L.). *Plants*, 10(11), 2304. <https://doi.org/10.3390/plants10112304>
- 23) Rebouh, N. Y., Aliat, T., Polityko, P. M., Kherchouche, D., Boulelouah, N., Temirbekova, S. K., ... & Gadzhikurbanov, A. S. (2022). Environmentally friendly wheat farming: Biological and economic efficiency of three treatments to control fungal diseases in winter wheat (*Triticum aestivum* L.) under field conditions. *Plants*, 11(12), 1566. <https://doi.org/10.3390/plants11121566>
- 24) Muhammed, S., & Hussein, H. Z. (2022). Survey and morphological study of wheat root rot disease in some Iraqi governorates. *Indian Journal of Ecology*, 49, 109–113.
- 25) El-Saadony, M. T., Saad, A. M., Najjar, A. A., Alzahrani, S. O., Alkhatib, F. M., Shafi, M. E., ... & Hassan, M. A. (2021). The use of biological selenium nanoparticles to suppress *Triticum aestivum* L. crown and root rot diseases induced by *Fusarium* species and improve yield under drought and heat stress. *Saudi Journal of Biological Sciences*, 28(8), 4461–4471. <https://doi.org/10.1016/j.sjbs.2021.04.027>
- 26) Salamon, S., Żok, J., Gromadzka, K., & Błaszczyk, L. (2021). Expression patterns of miR398, miR167, and miR159 in the interaction between bread wheat (*Triticum aestivum* L.) and pathogenic *Fusarium culmorum* and beneficial *Trichoderma* fungi. *Pathogens*, 10(11), 1461. <https://doi.org/10.3390/pathogens10111461>
- 27) Funnell-Harris, D. L., Sattler, S. E., Dill-Macky, R., Wegulo, S. N., Duray, Z. T., O'Neill, P. M., ... & Mitchell, R. B. (2024). Responses of wheat (*Triticum aestivum*) constitutively expressing four different monolignol biosynthetic genes to Fusarium head blight caused by *Fusarium graminearum*. *Phytopathology*®, 114(9), 2096–2112. <https://doi.org/10.1094/PHYTO-09-23-0234-R>
- 28) Trávníčková, M., Chrpová, J., Palicová, J., Kozová, J., Martinek, P., & Hnilička, F. (2024). Association between Fusarium head blight resistance and grain colour in wheat (*Triticum aestivum* L.). *Cereal Research Communications*, 52(4), 1599–1611. <https://doi.org/10.1007/s42976-024-00379-9>
- 29) Yin, C., Hulbert, S. H., Schroeder, K. L., Mavrodi, O., Mavrodi, D., Dhingra, A., ... & Paulitz, T. C. (2013). Role of bacterial communities in the natural suppression of *Rhizoctonia solani* bare patch disease of wheat (*Triticum aestivum* L.). *Applied and Environmental Microbiology*, 79(23), 7428–7438. <https://doi.org/10.1128/AEM.02838-13>



- 30) Guo, F., Wu, T., Shen, F., Xu, G., Qi, H., & Zhang, Z. (2021). The cysteine-rich receptor-like kinase TaCRK3 contributes to defense against *Rhizoctonia cerealis* in wheat. *Journal of Experimental Botany*, 72(20), 6904–6919. <https://doi.org/10.1093/jxb/erab335>
- 31) Lisiecki, K., Lemańczyk, G., Piesik, D., & Mayhew, C. A. (2022). Screening winter wheat genotypes for resistance traits against *Rhizoctonia cerealis* and *Rhizoctonia solani* infection. *Agriculture*, 12(12), 1981. <https://doi.org/10.3390/agriculture12121981>
- 32) Kwon, H. W., Yoon, J. H., Kim, S. H., Hong, S. B., Cheon, Y., & Ko, S. J. (2007). Detection of extracellular enzyme activities in various *Fusarium* spp. *Mycobiology*, 35(3), 162–165. <https://doi.org/10.4489/MYCO.2007.35.3.162>
- 33) Jaroszuć-Ścisł, J., Kurek, E., Słomka, A., Janczarek, M., & Rodzik, B. (2011). Activities of cell wall degrading enzymes in autolyzing cultures of three *Fusarium culmorum* isolates: Growth-promoting, deleterious and pathogenic to rye (*Secale cereale*). *Mycologia*, 103(5), 929–945. <https://doi.org/10.3852/10-322>
- 34) Abaya, A., Serajazari, M., & Hsiang, T. (2021). Control of Fusarium head blight using the endophytic fungus, *Simplicillium lamellicola*, and its effect on the growth of *Triticum aestivum*. *Biological Control*, 160, 104684. <https://doi.org/10.1016/j.biocontrol.2021.104684>