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Occurrence of Charcoal Rot in Globe Artichoke and Assessment of Inoculation Techniques for Pathogenicity and Management

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HIGHLIGHTS

- Macrophomina phaseolina was for the first time detected on globe artichoke in the world.
- Polyphagous nature of *M. phaseolina* was confirmed on nine plant species using pathogenicity

tests.

• Toothpick inoculation technique could be used for screening resistance to *M. phaseolina*.

Abstract: *Macrophomina phaseolina* is a polyphagous fungus causing substantial yield losses in many plant species. In 2017, *M. phaseolina* was found to be causal agent of wilting and stunting symptoms of globe artichoke (*Cynara scolymus*) in the Mediterranean region of Turkey. There is no knowledge about *M. phaseolina* in globe artichoke and applicable management practice in cultivation of the crop. In the present study, the causal agent was characterized *in vitro* and *in vivo* studies. Pathogenicity tests were carried out using seedlings of globe artichoke and nine plant species (sunflower, chickpea, soybean, sesame, peanut, wheat, maize, cotton and sorghum) in a greenhouse. In addition, five inoculation techniques were assessed to determine the most suitable method for screening resistance to *M. phaseolina* in globe artichoke. Significant (P<0.01) variations were found among the inoculation techniques. Depending on each inoculation technique, death of lateral roots and distinct lesions up to 5.38 cm occurred on primary roots and crowns of globe artichoke. *M. phaseolina* also caused lesions ranging from 1.43 to 9.63 cm on primary roots including crown and stems of tested plant species. *M. phaseolina* was pathogenic to globe artichoke and all the tested plant species, confirming its polyphagous nature. This is the first record of *M. phaseolina* causing root and crown rot in globe artichoke in the world. Moreover, the present study suggested that toothpick inoculation technique could be used for screening resistance to *M. phaseolina* in globe artichoke.

Keywords: Cynara scolymus; Macrophomina phaseolina; characterization.

INTRODUCTION

Macrophomina phaseolina (Tassi) Goid. is a necrotrophic fungus causing considerable losses in many agricultural crops. Yield losses vary according to host plant and environmental conditions. For example, yield loss might be 30% in sesame production [1], while it could be 80% in soybean [2]. Disease incidence may range from 26 to 95% in cowpea and sunflower, respectively [3,4].

M. phaseolina overwinters in soil or plant debris in infected fields. Under favorable conditions, it penetrates roots and crown and produces microsclerotia in root and stem tissues of plants. Typical symptom of the fungus is characterized with occurrence of small black microsclerotia in vascular tissues and stems of plants. Microsclerotia survive in soil up to 15 years, which ensures survival of the fungus. This characteristic might be closely associated with its polyphagous nature [2,5] as the fungus has a wide range of hosts over 500 plant species [6,7,8]. However, main and economically important hosts are soybean, sunflower, maize, sesame, chickpea, sorghum, strawberry and peanut [9,10,11,12,13]. Apart from these, in recent years, the fungus has been reported in chir-pine [14], jute [8], castor bean [15], dogwood [16] and mung bean [17]. These imply that *M. phaseolina* is increasing its host range. Likewise, in the present study, the fungus was found for the first time on globe artichoke in the Mediterranean region of Turkey. Globe artichoke (*Cynara scolymus*), belongs to *Asteraceae* family, is cultivated more than 30 countries in the world [18].

In management of *M. phaseolina*, seed treatments with some fungicides reduce infections, but most of fungicides effective against *M. phaseolina* are not currently used due to environmental health [12]. In addition, survival of microsclerotia in soil up to 15 years makes its management difficult [2,17]. Thus, determining resistant/tolerant hosts and using them in crop rotations are few plausible options for management of the fungus. However, there is no knowledge regarding *M. phaseolina* in globe artichoke and applicable management practice against the fungus in globe artichoke cultivation. The aims of the present study were i) to characterize *M. phaseolina* causing infections in globe artichoke, ii) to confirm pathogenicity of *M. phaseolina* in globe artichoke and other plant species and iii) to determine the most suitable method for screening resistance to *M. phaseolina* in globe artichoke by comparing five inoculation techniques.

MATERIAL AND METHODS

Sampling

A total of four samples of globe artichoke plants (5-year-old, cv. Bayrampaşa) showing wilting and stunting symptoms were taken from a farmer field (7 ha) in Manavgat district of Antalya province on 24 April 2017. The samples consisted of stems including root and crown parts of globe artichoke and they were obtained from the symptomatic adult plants at different points in the field. Distance from row to row and distance between plants in a row was 125 cm. Climatic conditions of the area was the Mediterranean climate (mean yearly temperature: 18.8 °C), which may provide a suitable environment for thermophilic pathogens including *M. phaseolina* [2].

Isolation

Stems of the symptomatic plants were cut into three main parts (lower stem, crown and roots). These parts were washed under the running tap water and cut into small pieces (1 cm). They were dipped into sodium hypochlorite (NaOCI 2%) solution for 2 min and then rinsed three times with sterile distilled water. After drying on sterile filter papers, 8 pieces were transferred to potato dextrose agar (PDA) containing streptomycin (50 mg/L) per plate (9 cm). The petri plates were kept at 25 °C for 5 days and developing fungal colonies were sub-cultured. As a result, one isolate was obtained from the samples.

Identification

Morphological identification

Micro morphological traits (shape, size and color of hyphae and microsclerotia) of the fungal colonies of the isolate were examined using an Olympus BX43 microscope with SC100 digital color camera. Fungal structures (hyphae and microsclerotia; n=50) of the isolate were examined in detail.

Molecular identification

The isolate was designated as MacCev and mycelia mass of three-day-old culture growing on PDA were taken into eppendorf tubes (5 mL). DNA of the isolate was extracted using DNA purification kit of Promega (Promega Corporation, USA). Ensuing DNA extraction, rDNA fragments were amplified with ITS-1 (5' TCC GTA GGT GAA CCT GCGG 3') and ITS-4 (5' TCC TCC GCT TAT TGA TATGC 3') [16,19] using a SimpliAmp Thermocycler (Applied Bio systems, USA). The amplification was as follows: 1 cycle of 94°C for 3 min, 35 cycles of 94°C for 30 s, at 58.5°C for 1 min, at 72°C for 1 min and at 72°C for 7 min. PCR products were separated in 2% agarose gels, stained with safe DNA dye and visualized under UV light. Sequence analysis was done by GENOKS (Çankaya-Ankara, Turkey). The sequence size of the isolate was 510 bp and deposited at GenBank (http://www.ncbi.nlm.nih.gov) with the accession number of MH593871.

A phylogenetic tree was also constructed to compare relatedness of sequence of the isolate with other sequences of *M. phaseolina* isolates in the GenBank using neighbor-joining method in MEGA version 10.0 program.

Chlorate utilization

Minimal medium containing 120 mM potassium chlorate was prepared as described by Pearson and coauthors [20]. The same medium without potassium chlorate was also prepared to compare chlorate sensitivity of the isolate. Mycelial plugs (5-mm) of the isolate were placed in the center of both media and incubated at 25 °C for 5 days in darkness. Colony radial growth of the isolate on each medium was measured using a caliper. The experiment was set up according to completely randomized design with four replications.

Temperature assay

Mycelial plugs (5 mm) of the isolate were place in the center of PDA and incubated separately at 15, 20, 25, 30 and 35 °C for 48 h. Colony radial growth of the isolate in each temperature was measured using a caliper. The experiment was conducted according to completely randomized design with four replications.

Arrangement of experiments for inoculation techniques

In these experiments, 6-week-old seedlings of globe artichoke (cv. Bayrampaşa) were tested at 30 $^{\circ}$ C in a greenhouse. Each experimental unit was consisted of one pot (25 × 35 cm) containing autoclaved soil and vermiculite (1:1). The experiments were conducted according to completely randomized design with three replications.

Soil inoculation technique

Sorghum seeds were autoclaved at 121 °C for 30 min in two consecutive days. Ensuing cooling, the seeds were inoculated with 5 mycelial plugs (5 mm) of the isolate growing on PDA and kept at 25 °C for 20 days [21]. The autoclaved mixtures of soil and vermiculite (1:1) were put into the pots. Inoculated sorghum seeds were thoroughly mixed into the pots (50 g kg⁻¹). One seedling of globe artichoke was planted per pot. In controls, only sterile sorghum seeds were mixed [22].

Toothpick inoculation technique

Tips of toothpicks (2 cm) were autoclaved at 121 °C for 30 min. They were put on margins of colonies of the isolate growing on PDA. After a 7 day-incubation period, one infested piece of toothpick was inserted into primary root just below crown of each seedling. This process was done by removing soils around crown of seedlings. Afterwards, the removed soil was replaced. In controls, only sterile pieces of toothpicks were used [23].

Agar plug inoculation technique

Seedlings of globe artichoke were planted into the pots. Some amount of soil was removed around the seedlings. One mycelial plug (5 mm) of 7 day-old colony of the isolate was attached to just below crown of each seedling. In controls, only agar plug was used. The removed soil was replaced in each pot.

Immersion inoculation technique

CaCO3 (10.3 g) was put into 750 mL V8 juice and centrifuged at 3000 rpm for 20 min. Supernatant of the mixture was diluted (1:4) with distilled water. 60 mL of this suspension was put into a 250 mL Erlenmeyer flask and autoclaved at 121 °C for 30 min. Three mycelial plugs (5 mm) of the isolate were put into the flask and incubated at 25 °C for 7 days. Mycelia of the isolate were collected and macerated in sterile distilled water in a 500 mL beaker. Seedlings of globe artichoke were immersed into the suspension (500 mg mycelia/20 mL) for 30 min and then planted into pots. In controls, seedlings were immersed into distilled water [24].

Corn sand inoculation technique

A mixture of 250 g corn flour, 250 g sand and 375 mL distilled water was put into 2 L- Erlenmeyer flask and autoclaved at 121 °C for 30 min in two consecutive days. 50 mL of the mixture was separated in a 250 mL Erlenmeyer flask. Five mycelial plugs of 5-day-old colonies of the isolate were transferred into the flask and kept at 30 °C for 21 days. Seedlings of globe artichoke were planted into the pots. 15 to 20 mL-inoculum was put around primary root and crown of each seedling. In controls, only sterile mixture was used [25].

Assessment of the inoculation techniques

Two months after inoculation, inoculated and non-inoculated seedlings of globe artichoke were uprooted. Lesion lengths on primary roots were measured using a caliper. Based on these measurements, each inoculation technique was compared with the others.

Arrangement and assessment of experiments for pathogenicity of *M. phaseolina* in some plant species

Nine plant species (sunflower, chickpea, soybean, sesame, peanut, wheat, maize, cotton and sorghum) were tested for pathogenicity of the isolate of *M. phaseolina*. The experiments were set up according to completely randomized design with three replications. Soil inoculation technique mentioned above was used in these experiments. Autoclaved mixtures of soil and vermiculite (1:1) were put into the pots. Inoculated sorghum seeds were thoroughly mixed into the pots (50 g kg⁻¹). Three-week-old seedlings of each plant species were planted into inoculated pots. One seedling of each plant species was used per pot. In controls, seedlings of each plant species were planted in non-inoculated pots.

Two months after inoculation, inoculated and non-inoculated seedlings of each plant species were uprooted. Lesion lengths on primary roots including crown and stems were measured using a caliper. Based on measurement of lesion lengths, pathogenicity assessments were performed for each plant species.

Statistical analysis

SAS 9.1 software program (SAS Institute Inc., Cary, NC, USA) was used for analysis of variance (ANOVA). Means of lesion lengths in pathogenicity and inoculation technique experiments and colony radial growths in temperature and chlorate utilization experiments were categorized using Fisher's least significant difference test (LSD_{0.01}).

RESULTS

Disease symptoms on infected plants in the field

Disease symptoms were observed in a 7 ha farmer field in Manavgat district of Antalya province in the Mediterranean Region of Turkey in 2017. Globe artichoke plants showed wilting, stunting and chlorosis symptoms in the field. Nearly 10 to 15% of the plants displayed these symptoms. When the symptomatic plants were uprooted, reddish-black discoloration and rot in the center of primary roots were observed (Figure 1).

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Figure 1. Disease symptoms, A: stunting, wilting and chlorosis symptoms caused by *Macrophomina phaseolina* in globe artichoke in the field (Antalya province, Turkey), B: rot in central part of the primary root of globe artichoke, caused by *M. phaseolina*.

In addition, as a result of the infections, more than 80% of the lateral roots left in the soil due to their death from the rot.

In vitro characteristics of the causal agent

Hyphae were hyaline brown to dark brown in color, with septa and branched at right angles. Color of microsclerotia was initially light brown and then turned to dark brown and finally black. Shapes of the microsclerotia were irregular but usually spheroid and oblong with 42.70 × 118.27 μ m - 51.32 × 188.75 μ m (average: 80.86 × 121.26 μ m) in diameter. Colony color was initially light but later turned to black with aerial mycelium (over 2 mm) (Figure 2).



Figure 2. Hyphae and microsclerotia of the isolate of *M. phaseolina* (10x).

These micro morphological traits of the isolate pertained to *Macrophomina phaseolina* (Tassi) Goid [26]. In addition, relatedness of the isolate (MH593871) was compared with other sequences (e.g. MT032391, MN166020, FJ395243 and GU046857) of *M. phaseolina* isolates in the genbank (Figure 3).



Figure 3. Phylogenetic tree showing relatedness of the isolate of *M. phaseolina* (MH593871) with other isolates of *M. phaseolina* in the genbank.

These confirmed identity of the causal agent that had caused wilt and stunting symptoms in globe artichoke plants in the Mediterranean region of Turkey.

In vitro experiments, the isolate of *M. phaseolina* showed restricted chlorate phenotype.

There was significant (P<0.01) difference in colony radial growth of the isolate on both media [with potassium chlorate (+) and without potassium chlorate (-)]. Mean colony growth of the isolate on minimal medium containing potassium chlorate was 7.25 mm, while it was 38.25 mm on minimal medium without potassium chlorate (Figure 4).



Figure 4. Comparison of colony radial growth of *M. phaseolina* in two minimal media (t test).

Thus, the isolate was categorized as chlorate-sensitive.

Colony radial growth of the isolate on PDA was also determined in the temperature assay. As a result, temperature had a significant (P<0.01) effect on colony radial growth of the isolate *in vitro* (Table 1).

Source	DF	Mean of	Source	DF	Mean of	Source	DF	Mean of
		squares			squares			squares
Temperature	4	1057.262**	Inoculation techniques	2	16.955**	Plant species	8	26.805**
Error	15	0.645	Error	6	0.114	Error	18	0.020
Total	19		Total	8		Total	26	
CV(%)	2.14		CV(%)	12.28		CV(%)	3.73	

Table 1. Variance analysis of colony radial growth of *M. phaseolina* in five temperature ranges (15, 20, 25, 30 and 35 °C) and lesion lengths occurred in each inoculation technique and plant species tested.

**Significant at P<0.01.

Colony radial growth significantly (P<0.01) varied in each temperature. As temperature increased colony radial growth of the isolate increased. For example, colony radial growth of the isolate was 39 mm at 25 °C, while it was 55.25 mm at 35 °C (Figure 5), revealing thermophilic feature of the isolate.



Figure 5. Means of colony radial growth of *M. phaseolina* in different temperature ranges (15, 20, 25, 30 and 35 °C).

Assessment of inoculation techniques for screening resistance to M. phaseolina in globe artichoke

All seedlings lost their viability due to severe infection of the isolate of *M. phaseolina* in the soil inoculation technique. However, in the toothpick inoculation technique, distinct lesions on primary roots developed from the point in which toothpicks had been inserted (Figure 6).



Figure 6. Comparison of inoculation techniques, A: seedling death of globe artichoke caused by *M. phaseolina* in soil inoculation technique (left) and healthy seedling of globe artichoke in control (right), B: distinct lesions caused by *M. phaseolina* on primary root (left) of globe artichoke in toothpick inoculation technique and healthy primary roots of globe artichoke in control (right).

In the agar plug inoculation technique, distinct lesions were observed on primary roots. However, these lesions did not extend much further like the other inoculation techniques (toothpick and corn sand). In the immersion inoculation technique, death of lateral roots from severe infection of the isolate of *M. phaseolina* was detected. Thus, no distinct lesions were observed on primary roots. In the corn sand inoculation technique, nearly 70% of the lateral roots died from infection of the isolate but distinct lesions were seen on primary roots.

When the results of all the inoculation techniques were assessed. Lesion lengths on primary roots of globe artichoke seedlings were measured in three inoculation techniques (toothpick, agar plug and corn sand). However, no distinct/measurable lesions occurred on primary roots of the seedlings of globe artichoke except for death of lateral roots in the other inoculation techniques (soil and immersion inoculation). Thus, any measurement could not be done in these inoculation techniques. As a result, three inoculation techniques aforementioned were analyzed statistically based on the measurements of lesion lengths on primary roots.

Differences in lesion lengths on primary roots were significant (P<0.01) in each inoculation technique (Table 1).

Compared to three inoculation techniques, the highest lesion lengths (average 5.38 cm) on primary roots were found in toothpick inoculation technique. However, in corn sand and agar plug inoculation techniques, mean lesion lengths on primary roots were 2.09 and 0.77 cm respectively (Table 2).

Inoculation techniques	Observation	Mean lesion length (cm)
Soil inoculation	Death of entire lateral roots on primary root	No distinct lesion development on primary root
Toothpick inoculation	Distinct lesions around toothpicks inserted on primary roots	5.38 ± 0.35 a
Agar plug inoculation	Partially lesions around agar plugs attached to primary roots	0.77 ± 0.24 c
Immersion inoculation	Death of entire lateral roots	No visible lesion development on primary root
Cornsand inoculation	Distinct lesions on primary roots	2.09 ± 0.39 b

Table 2. Comparison of inoculation techniques for pathogenicity of *M. phaseolina* in globe artichoke seedlings.

LSD (0.01):1.02, ±: standard deviation.

In all the inoculation techniques tested, the isolate was re-isolated from those infected tissues of the globe artichoke seedlings, which confirmed the pathogenicity of the isolate.

Assessment of pathogenicity of *M. phaseolina* in some plant species

Differences in lesion lengths caused by the isolate of *M. phaseolina* on primary roots were significant (P<0.01) among plant species (sunflower, chickpea, soybean, sesame, peanut, wheat, maize, cotton and sorghum) tested (Table 1).

M. phaseolina caused death of most of lateral roots of all the tested plant species. However, visible lesions occurred on primary roots of all the plant species. *M. phaseolina* was re-isolated from those symptomatic plant tissues of all the tested plant species, which fulfilled Koch's postulates. Variations in mean lesion lengths of each plant species were mostly significant (P<0.01) from one another. The highest mean lesion lengths, 9.63 and 8.23 cm, were found in sunflower and chickpea respectively, while the lowest ones, 1.96 and 1.43 cm, were detected in cotton and sorghum respectively (Table 3).

Table 3. Means of lesion lengths on primary roots including crown (collar) and stem parts of plant species tested.						
Plant species and cultivars	Root type	Mean lesion length (cm)				
Sunflower (<i>Helianthus annuus</i>) cv. Turay	Taproot	9.63 ± 0.11 a				
Chickpea (Cicer arietinum) cv. Gökçe	Taproot	8.23 ± 0.15 b				
Soybean (Glycine max.) cv. ATAEM-7	Taproot	3.40 ± 0.10 c				
Sesame (Sesamum indicum) cv. Muganlı	Taproot	2.63 ± 0.05 d				
Peanut (<i>Arachis hypogaea</i>) cv. NC-7	Taproot	2.43 ± 0.20 de				
Wheat (<i>Triticum aestivum</i>) cv. Koç-2015	Fibrous	2.20 ± 0.10 ef				
Maize (Zea mays) cv. Gözdem	Fibrous	2.10 ± 0.10 f				
Cotton (Gossypium hirsutum) cv. ST373	Taproot	1.96 ± 0.15 f				
Sorghum (Sorghum bicolor) cv. Rox	Fibrous	1.43 ± 0.20 g				

LSD (0.01): 0.33, ±: standard deviation.

DISCUSSION

Infection phenomenon of *M. phaseolina* as follows: *M. phaseolina* usually penetrates into roots and then lower internodes in stem. It hampers water and nutrient transport from soil to upper parts of plants. As a result, infected plants show wilting, stunting and chlorosis symptoms or plants may die in severe infections [7]. Root rot pathogens cause damage to primary root, whereas they kill small lateral roots of plants such as bean, peanut, soybean and asparagus. As a consequence, growth of infected plants is retarded and chlorosis appears [27]. Globe artichoke has a taproot system as the plants mentioned above. Similar symptoms aforementioned were observed on globe artichoke plants infected by *M. phaseolina* in the field in the Mediterranean region of Turkey. However, there were no pycnidia on infected plant tissues in the present study. In fact, pycnidia of *M. phaseolina* are rarely observed in different host plants [26,28]. Moreover, the role of pycnidia in epidemiology of the fungus is not well known [12].

In the present study, comparing five different temperature ranges, maximum colony growth of *M. phaseolina* was at 35 °C, implying thermophilic feature and adaptation of the isolate to drought conditions of the Mediterranean climate of Turkey. Sergeeva and coauthors [29] reported that maximum colony growth of isolates of *M. phaseolina* from olive plants was 32 °C. Sánchez and coauthors [13] stated that optimum temperature was 30 °C for mycelia growth of Chilean isolates of *M. phaseolina* from strawberry, while it was 35 °C in Spanish isolates. These may be associated with geographic origin of the isolates of *M. phaseolina*.

In the present study, in the experiments of inoculation techniques tested, occurrence of lateral root deaths of seedlings in inoculated plots indicated vulnerability of globe artichoke at the seedling stage. In this context, Beas-Fernández and coauthors [30], Khan [7], Alghuthaymi and coauthors [31] reported that infections of *M. phaseolina* in early plant growth period of bean, sunflower and cotton could cause seedling blight, pre- and post- damping off, root rot, basal stem rot and early maturing of plants. These findings corroborated our results. In addition, considering all of these findings, it may be inferred that impact of *M. phaseolina* on plant growth at the seedling stage could be severe.

Soil inoculation technique was used for pathogenicity tests of *M. phaseolina* in various agricultural crops. For example, Sergeeva and coauthors [29], Khanzada and coauthors [32], Kaur and coauthors [33], Alghuthaymi and coauthors [30], Yeşil and Baştaş [34] used soil inoculation technique for pathogenicity of *M. phaseolina* in olive, okra, pigeon pea, cotton and dry bean, respectively. However, in the present study, in the soil inoculation technique, death of lateral roots of globe artichoke seedlings occurred due to infection of *M. phaseolina*. End of primary roots connecting lateral roots displayed partial infection but no distinct lesion development appeared on the primary roots of the seedlings. Thus, there was no possibility to compare this inoculation technique with the others although re-isolations were performed well. The similar case occurred in the immersion inoculation technique. From these findings, these inoculation techniques did not seem to be suitable for screening resistance to *M. phaseolina* in globe artichoke.

With regard to other inoculation techniques tested, partial death of lateral roots was also observed in the other inoculation techniques (toothpick, agar plug and corn sand). But distinct lesions on primary roots in these inoculation techniques provided measurement and comparison. Comparing these inoculation techniques, in addition to pathogenicity, toothpick inoculation technique could be used for screening resistance to *M. phaseolina* in globe artichoke genotypes. Because, this technique could provide accurate comparison of disease reactions of globe artichoke genotypes with measurement of distinct lesions on primary roots. In this context, Shekhar and coauthors [5] evaluated responses of maize lines against *M. phaseolina* using toothpick inoculation method. In addition, Hajlaoui and coauthors [35] used toothpick inoculation technique for pathogenicity of *M. phaseolina* in strawberry.

In the present study, soil inoculation technique used for pathogenicity of *M. phaseolina* in other plant species (sunflower, chickpea, soybean, sesame, peanut, wheat, maize, cotton and sorghum) caused also death of lateral roots in most of the plant species. However, distinct lesions occurred on primary root including crown/collar and stem, which provided measurement and consequently comparison of pathogenicity of *M. phaseolina* in the plant species. As a result, the isolate of *M. phaseolina* was pathogenic to all the plant species tested in the present study. Mihail and Taylor [28] stated that majority of isolates of *M. phaseolina* from *Asteraceae* family were pathogenic to sorghum, sunflower and bean. Claudino and Soares [15] found that 27 isolates of *M. phaseolina* from various hosts (cotton, sesame, sunflower, peanut and bellyache bush) were pathogenic to castor (*Ricinus communis*) with varying degree of aggressiveness of each isolate. As for Purkayastha and coauthors [6], they reported that irrespective of origin of host, all isolates of *M. phaseolina* were pathogenic to cluster bean. Likewise, Khan and coauthors [36] emphasized that isolates of *M. phaseolina* and confirmed polyphagous nature of *M. phaseolina*. However, Burkhardt and coauthors [37] stated that one genotype of *M. phaseolina* had host specificity in strawberry.

CONCLUSION

Increasing global temperatures may enhance survival and spread of thermophilic/drought pathogens [38, 39]. In this regard, *M. phaseolina* may increase its host range and prevalence in agricultural areas. Likewise, in the present study, *M. phaseolina* was detected for the first time in globe artichoke in the world.

To mitigate influence of this emerging fungus, sustainable, applicable, cost-effective and environmentally friendly management practices should be suggested. In this context, determining resistant/tolerant hosts and using them in crop rotations are few applicable options in the management of *M. phaseolina*. In addition to reporting occurrence of *M. phaseolina* in globe artichoke, the present study suggested that toothpick inoculation technique could be used for screening resistance to *M. phaseolina* in globe artichoke genotypes.

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